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12th International Congress on Aerobiology
5th International Ragweed Conference

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European Aerobiology Society

International Association for Aerobiology

International Ragweed Society

Vilnius University

European Aerobiological Society

The European Aerobiological Society (EAS) has been established in 2008 as a non-profit association with the aim to promote and foster aerobiology; to facilitate collaboration, research, education, information, technical development, and practical application in the field of aerobiology and to encourage collaboration with other areas of science.

Along these years, EAS has promoted the standardization and quality control of the aerobiological sampling and analysing of pollen and fungal spores in the air, with the aim to make a large pan-European (and worldwide!) network of sites producing homogenised data. Also, there is the willingness to integrate all these datasets in a common database, to facilitate larger and global studies. As outputs there are papers with guides for sampling and analysing and quality control (1,2) and proposing unified nomenclatures (3) and the European standard EN16868 (4). Being involved in the EUMETNET AutoPollen program enables EAS members to get timely information and provides possibilities for sharing expertise and creates collaboration opportunities, the most exciting being to contribute to the evolution towards automatic and real-time detection and analysis methods.

Education is one of the pillars of EAS activities. In this sense, EAS is organizing an EAS Basic Course in Aerobiology, every two years, in collaboration with local research groups and contributes with a certain number of grants offered to participants. The next EAS Basic Course in Aerobiology will be held in Poznan (Poland) in 2025.

An EAS Symposium has been celebrated every 4 years, each time in a different country, gathering not only the fellow aerobiologists but also other related disciplines. In 2024, the recent global health events have led to the 8th European Symposium on Aerobiology (8th ESA) where for the first time ever space, time, participants, science, and knowledge will be shared with the regular meetings of two other sister associations, the International Association for Aerobiology (IAA) and the International Ragweed Society (IRS). This big and genuine event has been named World Aerobiology 2024.

EAS is funded by membership fees and possible surpluses from the ESA or the Basic Courses. Most of the EAS income is devoted to offer grants for participating in the EAS Basic courses, the ESA and other congresses or courses related to Aerobiology. The aim of the grants is, at times, to support young researchers to be introduced in Aerobiology, and, in other occasions, to help young aerobiologists and researchers from areas with difficulties to travel and participate in the scientific events.

The latest information on EAS and advances in aerobiology is available at EAS website: http://www.eas-aerobiology.eu/.

EAS is an active family open to new members, who may be from different scientific fields. We encourage you to become a member and contribute actively to its aims!

(1) Galán et al. 2014. Aerobiologia, 30: 385-395. DOI 10.1007/s10453-014-9335-5
(2) Galán et al. 2021. Aerobiologia, 37: 351-361. DOI 10.1007/s10453-021-09698-4
(3) Galán et al. 2017. Aerobiologia, 33: 293-295. DOI 10.1007/s10453-017-9496-0
(4) CSN EN 16868 - Ambient air - Sampling and analysis of airborne pollen grains and fungal spores for networks related to allergy - Volumetric Hirst method.

International Association for Aerobiology

Aerobiology is the study of bioaerosols (airborne particles of biological origin that are passively transported in the air), and includes their sources, liberation, dispersal, deposition and impact.

The International Association for Aerobiology (IAA) is a worldwide scientific community that aims to promote the development of aerobiology and to facilitate international co-operation in this important field (https://iaaerobiology.wordpress.com).

Aerobiology became a theme in 1964 when the International Biological Program (IBP) was established. An International Aerobiology Working Group was constituted in 1968 to coordinate all national aerobiological programs, and the IAA was subsequently founded on the 11th September, 1974, in the Hague, the Netherlands.

Today the IAA has a membership of about 800 persons representing multidisciplinary fields of basic and applied sciences, such as allergology, bioclimatology, biological contaminants, biological weathering, cultural heritage, indoor air quality, industrial aerobiology, microbiology, palynology, phenology and plant pathology.

As an international association, the IAA is closely associated with national, regional, and continental aerobiological associations and societies around the world, including those in Europe, Asia, Oceania, and the Americas.

The IAA arranges, among others, quadrennial international congresses of aerobiology, and cooperates with international, inter-governmental, governmental or non-governmental organisations in related fields (e.g., is affiliated with the International Union of Biological Sciences (IUBS) and the International Federation of Palynological Societies (IFPS)).

The IAA is associated with the journal Aerobiologia which was established in 1985 and is published by Springer Nature. The IAA's Newsletter is published twice a year and provides the Association's members with unique insights on the latest developments, advances, and activities in aerobiology around the world.

Since 2018 there has been a special IAA Working Group for Young Aerobiologists, established to encourage more young scientists to become active in the field. The IAA is also involved in education and organises an international Advanced Course in Aerobiology. In addition, the Association actively supports Early Career Researchers by providing grants so they can participate in summer schools and conferences.

The International Association of Aerobiology is a unique and collegial community of scholars which actively promotes equity, diversity, and inclusion. Membership of the Association provides the perfect platform for productive collaborations and life-long friendships. We warmly welcome new members (https://iaaerobiology.wordpress.com/home/membership/).

It is a pleasure to welcome you to World Aerobiology 2024. It is particularly pleasing to see so many new colleagues, especially those from the parts of the world where aerobiological studies have only recently started to be carried out. I hope you have an enjoyable congress and make many new friends and collaborators and help to grow the discipline further.

Dorota Myszkowska, President IAA

International Ragweed Society

Recent activities of the International Ragweed Society (IRS): 2022-2024.

IRS conferences: IRS holds biennial conferences, either stand alone or in connection with other international symposia (latest IRS Conference: 2022: Budapest, 51 participants, 17 countries, 3 continents, 32 lectures, and 11 posters).

“Let’s talk about Ambrosia!” From 2023 onwards, IRS is organizing regular online meetings entitled “Let’s talk about Ambrosia!”. These are short online conferences that aim to share all aspects of vital ragweed functions, its impact and means of control with interested persons.

The procedure is simple: a speaker presents his/her project, work or research outcomes concerning Ambrosia species for 15 min., followed by 15 minutes questions to the speaker and discussions of his/her presentation.

The presentations are recorded, and the video is uploaded to the IRS website, where it is available to all IRS members.

Presentations so far:
2023, September 7, 13.00-14.00 (Paris local time): Citizen science to tackle Ambrosia artemisiifolia populations and prevent future invasion? By Professor Arnaud Monty, Liège University, Liège (Belgium).

2023, October 26, 13.00-14.00 (Paris local time): Pollen-food syndrome caused by Ambrosia. By Professora Victoria Rodinkova, Vinnytsia Medical University, Vinnytsia (Ukraine).

2023, December 12, 13.00-14.00 (Paris local time): Ophraella communa in Europe: an update of its occurrence and potential further spread. By Prof. Yan Sun, College of Resource & Environment, Huazhong Agricultural University, Wuhan (China).

Planned presentation:
Ophraella communa project in France: INRAE and CSIRO testing panel of vegetal hosts.

Further activities planned by the IRS Committee

Activities at international conferences and online seminars, and through our website to increase the visibility of the IRS and to increase the number of IRS members. In addition, we made a database containing email addresses of researchers involved in ragweed studies and sent them emails highlighting the benefits of becoming an IRS member. The list was also used to spread information about IRS activities.

Co-organise the World Aerobiology 2024 Conference in Vilnius, Lithuania on July 1-5, 2024.
Updating the website of IRS in particular with the results of research on ragweed and with specifically prepared flyers.

Continuously raise awareness among the public about ragweed's impact, e.g. each year, we hold the Ragweed day, where participating countries organize specific activities.

Find organizations, institutions and companies that financially support our society.

We invite those researching ragweed and its pollen to join the International Ragweed Society. Membership brings many advantages that facilitate international cooperation, participation in joint projects, and joint work. See more: https://internationalragweedsociety.org/

An active link for joining is as follows: https://internationalragweedsociety.org/2023-2024-irs-membership/

ORAL PRESENTATIONS

6

"Pollen Resilience Index": Combined Impact of Air Pollution, Pollen Load, and Heat on Allergy Symptoms

Laura Šukienė, Ingrida Šaulienė, Gintautas Daunys, Rūta Dubakienė, Odilija Rudzevičienė

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Abstract

During the vegetation season, when allergenic pollen is abundant, air pollution and extreme meteorological conditions, especially associated with heat waves, can cause additional health burdens. Allergic or sensitive to pollen individuals may be affected by complex atmospheric factors. In the study, we used data from 2018-2022: (1) allergenic pollen data, (2) depersonalised Pollen Diary data regarding the symptoms of pollen-sensitive individuals, (3) meteorological, and (4) air quality data.

For calculations, we applied open-access data from the Copernicus Climate Data Store ERA5 and the CAMS. Using datasets, a Humidex index was calculated to evaluate heat extremes. CAMS forecast of regulated air pollutants (PM_{10}, CO, SO_2, O_3, NO_2) were used in the study, as well as pollen forecast (*Alnus*, *Corylus*, *Betula*, Poaceae, and *Artemisia*). "Pollen Resilience Index" for pollen allergen-sensitive individuals was calculated using daily average pollutant concentrations, pollen data and Humidex index values. Air pollution has been assessed using short-term limit values for concentrations of air pollutants in accordance with EU and Lithuanian legislation. The pollen thresholds were based on the pollen concentrations relevant to Lithuania and described in the published studies. Pollen sensitive persons used mobile PASYFO application for allergy symptoms as well as web-based Pollen Diary. The depersonalized Pollen Diary data were used in this study. The "Pollen Resilience Index" was validated using 2022 data of air quality and meteorological data from the Copernicus CAMS and ERA5 databases, real-time airborne pollen dispersion data collected by the Rapid-E automated particle detector, and the Pollen Diary's input data on the allergic rhinitis symptoms of individuals sensitive to pollen allergens. For data analysis and statistical evaluation, the R, RStudio and Pytorch software packages were used.

By testing the "Pollen Resilience Index", which includes air quality parameters, Humidex and pollen load, we found that the Index is associated with overall allergic rhinitis symptoms, i.e. increasing value of the Index leads to higher average symptom reports. The findings revealed that the number of Pollen Diary entries reporting symptoms increases with increasing values of the Synergistic Air Quality Index, as the higher index values indicate the worsening atmospheric condition and air quality for human health, which led to the higher frequency of symptom records. Comparing the 2018-2021 and 2022 results, there is a shift in the average symptom scores in 2022. The shift allows us to assume that the symptom severity could be associated with the "Pollen Resilience Index" results.

In addition to air quality, extreme weather conditions need to be considered when developing indexes appropriate for providing information to pollen-sensitive individuals. Developing a "Pollen Resilience Index" that brings together the combined effects of pollen and weather on

pollen-sensitive individuals can be the backbone for developing useful services for public awareness applications.

This research has received funding from the Research Council of Lithuania (LMTLT), agreement No S-MIP-19-53, and supported by the EO4EU (AI-augmented ecosystem for Earth Observation data accessibility with Extended reality User Interfaces for Service and data exploitation) project, funded by the EU's Horizon Europe Research and Innovation Programme under Grant Agreement No. 101060784.

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Investigation of Pollen Dispersion at Various Atmospheric Altitudes Using Unmanned Aerial Vehicles

Ingrida Šaulienė[1], Gintautas Stankūnavičius[1], Ričardas Skorupskas[1], Antanas Gedvilas[2], Ilona Kerienė[1], Laura Šukienė[1], Edvinas Stonevičius[1]

[1]Vilnius University, Vilnius, Lithuania. [2]Lithuanian Association of Unmanned Aircraft Users, Vilnius, Lithuania

Abstract

The transport of pollen in the atmosphere involves both horizontal and vertical dimensions. While horizontal pollen transport has been extensively studied, the upward and downward movement of pollen has not received thorough characterization. This highlights the importance of considering all dimensions of pollen transport to gain a comprehensive understanding of the phenomenon. It is crucial to note that information regarding pollen dispersion at various altitudes can be obtained from stationary devices situated at different elevations above ground. Although these samplers operate in diverse vegetation environments, they still offer limited insights into the vertical characterization of the atmosphere concerning pollen. Our objective was to examine how local vegetation influences pollen diversity and counts at different altitudes within the atmospheric boundary layer. We hypothesized that the impact of local vegetation on both pollen diversity and abundance decreases with altitude, albeit not uniformly.

The fixed-wing UAVs were equipped with two types of collectors designed specifically for our research. The first method allowed airborne particles to be captured through impingement with a frontal force, while the second utilized passive suction. Samples were collected at various altitudes (30, 50, 90, 110, 150, and 250 m above the ground) during 2021-2022, with simultaneous sampling by the Coriolis device at the surface. Each sample was taken for 10 minutes. The experiment was conducted under specific meteorological conditions favorable for normal pollen dispersal, and plant phenophases were observed on each sampling day. Data from the Vilnius aerobiological station on pollen, recorded by the Hirst-type device during the study,

were used. Microscopic analysis of pollen collected by fixed-wing UAVs was performed, and both pollen diversity and the vertical profile of pollen counts were assessed.

An extensive collection of pollen samples was obtained over the study period, including pollen from 24 genera or plant families collected using a UAV and 18 pollen morphotypes collected using a Hirst trap. Pollen diversity was dominated by *Betula*, *Picea*, *Pinus* and Poaceae, regardless of the altitude of pollen collection. Notably, pollen diversity exhibited significant variations with both altitude and the year of sampling. Pollen diversity was found to be significantly different between samples taken at ground level and at 50, 70, 130, and 250 meters. Although neither the Wilcoxon Rank-Sum Test nor the Welch Two Sample Test revealed significant differences in pollen diversity at different atmospheric heights, the data on pollen variability were significantly correlated with the pollen diversity found in samples collected at various atmospheric heights. Pollen diversity was significantly lower at the height levels of 150 and 250 meters compared to the levels between 30 and 110 meters. A more detailed understanding of the vertical dispersion of pollen has been achieved by gathering information from different layers of the atmosphere using UAVs and a specially designed particle collector.

The study was partly implemented in the frame of contract Nr. (1.57) 15600-INS-138

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Association of Ragweed Pollen Episodes in Lithuania with Atmospheric Circulation Patterns

Ingrida Šaulienė[1], Gintautas Stankūnavičius[2], Laura Šukienė[1]

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Abstract

Ragweed (*Ambrosia artemisiifolia* L.) is a harmful invader for native plants, an economic threat to agriculture and a serious health hazard due to pollen allergy. In Lithuania every year, ragweed pollen is detected in the air, with concentrations commonly reaching up to 10 pollen/m^3 in late August and early September. In some cases, airborne pollen concentrations have exceeded 50 pollen/m^3 over the 15 years of observation. The study aimed to determine the relationship between synoptic conditions and ragweed pollen concentrations and reveal if the regional weather regime and the large-scale atmospheric circulation patterns significantly influence airborne Ambrosia pollen levels.

Airborne ragweed pollen count data were collected from a database of aerobiological observations conducted in three Lithuanian cities, Klaipėda, Šiauliai and Vilnius, between 2005 and 2019. Volumetric Hirst pollen traps were used to collect airborne pollen, and samples were

processed following the standardized method EN 16868:2019. To associate pollen concentrations with large-scale weather regimes, we selected atmospheric circulation types in Europe based on the Hess and Brezowsky large scale circulation (Grosswetterlagen - GWL) classification, which is a well-established weather regime set. The statistical analysis was performed using R and RStudio with confidence in the accuracy of the results.

Over the 15-year monitoring period, Vilnius had a higher frequency of ragweed pollen detections and a higher number of cases with high pollen concentrations than other monitoring sites in Lithuania. The common median pollen count for ragweed between 2005 and 2019 is 8-10 pollen/m^3. The main load was found to be in the morning and afternoon hours (6 a.m. to 4 p.m.), with pollen concentrations > 20 pollen/m^3. In 2008 and 2011, ragweed pollen concentrations were significantly higher, with individual days exceeding 100 pollen/m3. Low, often occurring, pollen concentrations are recorded in the West Anticyclonic (WA) and Southwest Anticyclonic (SWA) circulation types. Atmospheric circulation types such as Central European Trough (TRM), Western European Trough (TRW), Central European Low (TM) and Central European Ridge (BM) proved to be suitable for the advection of ragweed pollen to Lithuania, as it is detected more frequently than in other conditions. The highest pollen concentrations over the observation period were found under the synoptic circulation, so-called atypical Transition (Ü). This latter form is usually found when the pressure and geopotential height fields over Europe are changeable or not persistent at all. In our cases, there were few short episodes when the strong warm advection from the south took place.

A better understanding of the properties of large-scale air current structures allows the identification of patterns in ragweed pollen occurrence and contributes to the forecasting of high-risk ragweed pollen episodes.

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Estimating the Uncertainty in Airborne Birch Pollen Modelling

Willem W. Verstraeten[1], Rostislav Kouznetsov[2], Nicolas Bruffaerts[3], Mikhail Sofiev[2], Andy W. Delcloo[1]

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Abstract

Background

More than 25% of the adults in Europe suffer from pollinosis, although the variability across countries might be quite large. In Belgium, at least ~10% of the people develop allergies due to birch pollen. Thus, many people may benefit from the birch pollen forecasting system that was

established for the Belgian territory in 2023 based on the SILAM model (System for Integrated modeLling of Atmospheric composition). The key question, however, is which uncertainty can be expected when modelling and forecasting airborne pollen levels?

Materials & Methods

The uncertainty in modelling airborne birch pollen levels near the surface using SILAM is quantified based on a Monte-Carlo error approach for the season of 2018 in Belgium using varying major model input data and ECMWF ERA5 meteorological data. The relative Coefficient of Variation (CV%) is used as measure for uncertainty. The studied key model input datasets that drive the birch pollen model are the:

- map with the amount (areal fraction) and location of birch trees on a native 0.1° x 0.1° grid,

- map with the start and end of the birch pollen season on a native 1° x 1° grid, and

- ripening temperature of birch catkins.

For each input dataset 100 randomly sampled data layers were prepared for running SILAM 100 times. For the maps, in each 1° by 1° block containing 100 grid cells (0.1° x 0.1° native grid), we randomly redistribute the birch pollen emission sources 100 times. From the resulting 100 SILAM model runs 100 spatial-temporal datasets on surface birch pollen levels were produced and its variation was summarized by the CV%.

Results

From the analysis we find that the uncertainty in the amount and locations of birch pollen emissions sources in SILAM on resulting modelled airborne birch pollen levels near the surface in Belgium is substantially high, with CV% values ranging between ~15% and ~35%. The parameters indicating the start and end of the season, however, are at least equally important. CV% values up to 50% are found in the southeastern parts of Belgium. By adding up all the model input uncertainties including the impact of the catkins-ripening temperature, we obtain CV% values of 50% and more. If we assume an accumulated error of 20-40% from all meteorological data, a CV% value near 60% can be expected. These error values in modelled pollen levels are in the same order of magnitude than the reported errors in monitored pollen levels, based on the reference Hirst method.

Conclusions

The uncertainty in airborne birch pollen levels near the surface modelled using SILAM and quantified as the CV% is more than 50%. It is the same order of magnitude than the reported errors from observed pollen counts Hirst type devices at monitoring stations.

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Analyzing the Airborne Birch and Grass Pollen Monitoring Network in Belgium

Willem W. Verstraeten[1], Nicolas Bruffaerts[2], Rostislav Kouznetsov[3], Mikhail Sofiev[3], Andy W. Delcloo[1]

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Abstract

Background

In Belgium, ~10% of the people is sensitive to birch pollen and ~15% to grass pollen affecting the quality of life badly. Mitigation measures can ease the pollen induced symptoms, but this requires early warning systems that can produce timely information on forthcoming pollen episodes. The performance of such systems can be improved when pollen observations from strategically well-chosen pollen monitoring stations are assimilated. Prior to assimilating observations into forecasting models, the network quality and coverage of the pollen monitoring stations should be evaluated.

Materials & Methods

Here we explore the network quality (i) and network coverage (ii) of the current five pollen monitoring stations in Belgium based on the methodology developed by Sofiev et al. (STOTEN, 2023).

To analyze the network quality (i), we first spatially interpolate the daily observations of grass and birch pollen by applying the radial-based function on the five monitoring sites of the Belgian network. We compare these interpolated values with a reference dataset that includes spatio-temporal distributions of daily surface airborne birch and grass pollen levels. The ability of the network to reproduce the concentration field over the region of interest is then quantified by the Root-Mean-Square-Error (RMSE). The reference dataset for both birch as well as grass pollen is taken from the operational early warning system on the website of the Royal Meteorological Institute of Belgium. This system implements the SILAM model (System for Integrated modeLling of Atmospheric coMposition).

For evaluating the network coverage (ii) of the five current monitoring stations we perform a footprint-based analysis. At each of the five locations of the monitoring stations we run SILAM in the backward mode (three days), resulting into the travelling trajectory of the captured birch and grass pollen in the air observed at the network stations. Nine pollen seasons (2013-2021) were analyzed using ECMWF ERA5 meteorology.

Results

The network performs well with respect to the spatio-temporal reference dataset for birch pollen derived from SILAM since we find that more than 78% of the daily RMSE values are less

than their mean value. For the 2015 birch pollen season more than 99% is reached. For 2020 this is only ~44%. For grass pollen we obtain 80% for the network quality, with values ranging between 63% and 97% for 2021 and 2017 respectively.

The footprint analysis shows that on average the coverage of the monitoring stations for birch pollen is quite good. There are, however, large differences during the 2013-2021 seasons which might be due to the typical large inter-seasonal variation in birch pollen production. For grass pollen, the average coverage is better, and the inter-seasonal variation much lower.

Conclusions

First results show that for the observations of airborne birch and grass pollen the monitoring network in Belgium is of good quality and its coverage is sufficient. All depends, however, on the applied time scale. At smaller time scales such as days or hours, the network configurations may degrade faster than on larger time steps such as weeks, months, or seasons.

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What Drives the Long-term Changes in Airborne Birch and Grass Pollen Levels in Belgium?

Willem W. Verstraeten[1], Nicolas Bruffaerts[2], Letty de Weger[3], Rostislav Kouznetsov[4], Mikhail Sofiev[4], Andy W. Delcloo[1]

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Abstract

Background

Airborne pollen may contribute to increased respiratory allergies. In Belgium, ~10% of the people is sensitive to birch pollen and ~15% to grass pollen. In the future, even more people might be affected since changes in climate and land-use tend to increase the amount of allergenic pollen in the air and prolong the pollen seasons. We aim at disentangling the effect of changing meteorology and changing vegetation dynamics on the estimated long-term trends of birch and grass pollen levels near the surface for Belgium.

Materials & Methods

We apply the pollen transport model SILAM (System for Integrated modeLling of Atmospheric coMposition) using ECMWF ERA5 meteorological data in a bottom-up emission approach for

the period 1982-2019 for two model scenarios. In the reference scenario (i), SILAM was run during 38 seasons with one fixed emission map of birch and grass pollen sources. Changes in airborne surface birch and grass pollen levels are then due to the varying meteorological data only. In the updated scenario (ii), SILAM was run with 38 different pollen emission maps (one for every pollen season), which were derived from merging multi-decadal datasets of spaceborne NDVI with forest inventory data and grass distribution maps for the period 1982-2019. This run combines the effect of both the changing emissions as well as the varying meteorology. The difference of both scenarios might be a good indication for the effect of vegetation dynamics on the trends. Temporal trends (Theil-Sen) are computed for daily pollen levels and for the daily meteorological model input for the period 1982-2019. For each model gridcell we estimate the association between trends in pollen and meteorology using the Kendall correlation.

Results

From the trend analysis, the strong increase in birch pollen concentrations is associated with increasing radiation, decreasing precipitation and decreasing horizontal wind speed near the surface. Inter-seasonal variation in birch pollen production intensifies the climate induced increase of airborne birch pollen concentrations with ~6%.

The resulting strong reduction in grass pollen concentrations over time is mainly driven by a decreasing trend in grass pollen sources. This decrease is ~10 times stronger that the impact of the changing meteorology. When only considering meteorology, a large increase in grass pollen emissions over time should be expected. The overall trend reduction is mainly positively associated with changes in precipitation and wind, and is negatively associated with radiation.

Conclusions

During the period 1982-2019, airborne birch pollen levels near the surface in Belgium increased substantially due to changes in both meteorology as well as in vegetation dynamics. For the same period, airborne grass pollen levels decreased which could be attributed due to changes in vegetation dynamics which overcompensated the increasing impact of meteorology on the trend.

13

Influence of Vegetation Spatial Resolution on the Prediction of Pollen Concentrations Over Northern Italy

Sofia Tagliaferro[1], Mario Adani[2], Nicola Pepe[3], Gino Briganti[2], Massimo D'Isidoro[2], Maira Bonini[4], Antonio Piersanti[2], Sandro Finardi[3], Pierpaolo Marchetti[1], Francesco Domenichini[5], Mihaela Mircea[2], Maria Gabriella Villani[2], Alessandro Marcon[1], Camillo Silibello[3]

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Abstract

Introduction: in a worldwide increasing trend of allergic diseases, accurate forecasting models will be useful to inform allergic populations about pollen concentrations. High-resolution models could also supplement pollen observational data, especially where dense monitoring networks are lacking.

Aim: to introduce and validate, for the first time, a pollen modelling system over the Veneto Region (Northern Italy) at 3 km spatial resolution. We analysed the influence of the spatial resolution of vegetation coverage (VC) maps on predicted pollen concentrations for alder, birch, olive, grass, and ragweed.

Methods: using the Flexible Air quality Regional Model (FARM), the model simulated pollen dispersion, diffusion and deposition for the year 2019, using VC maps, phenological pollen emission algorithms (used in the Copernicus Atmospheric Monitoring Service (CAMS) European air quality ensemble), meteorological forecasting (by the prognostic non-hydrostatic model WRF), and the pollen boundary conditions provided by a similar modelling system operating at national scale. Two VC datasets were used: a) the European CAMS dataset, available at ~10 km horizontal spatial resolution (CAMS VC); b) a high-resolution dataset (detailed VC) obtained from the combination of CORINAIR data (250 m horizontal resolution) for olive and grass, European Forest Institute (1 km horizontal resolution) for alder and birch, a national inventory (1 km horizontal resolution) for ragweed. Predicted daily averaged concentrations obtained with CAMS (a) and detailed VC (b) were compared to the observations collected at 15 monitoring stations, using model performance indicators (RMSE) and pollen seasonal-derived parameters. A stratified analysis was performed to assess differences in model performance between lowland and mountain environments.

Results: an important reduction of mean RMSE was obtained for alder and birch pollen running the model with detailed VC (detailed VC vs. CAMS VC: 15.69 vs. 133.58; 17.83 vs. 52.54 p/m^3, respectively), while the mean RMSE resulted slightly higher for grass (24.49 vs. 20.68 p/m^3). Similar mean RMSEs were obtained for olive (3.82 vs. 3.98 p/m^3) and ragweed pollen (3.90 vs. 3.90 p/m^3). Results from the comparison of predicted and observed Seasonal Pollen Integrals (SPIn) were consistent with the RMSE patterns. Overall, the start date predictions were more accurate than the end date, with better results using CAMS VC for some pollens. The improvement in prediction of daily concentrations obtained using detailed VC was particularly evident in the mountain environment.

Conclusions: our findings indicate that incorporating detailed VC maps into pollen dispersion models improves prediction accuracy, particularly in complex areas where details are crucial (mountain environment). Enhancing pollen forecasts in response to climate-induced variations would improve the well-being of the allergic population.

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Crossroads of Aerobiology and Astrobiology – Aerobiological Research Opportunities and Experimental Setup in the Stratosphere

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Abstract

Since the discovery of possible bacterial particles in the stratosphere in the 1930s, bioaerosol particles in the stratosphere have been studied to understand how far the biosphere extends, what types of bioaerosol particles and how they exist in the extreme stratospheric environment and how these bioaerosol particles can reach above the troposphere. Although little is known about the ecology in the stratosphere, not much work has been done in the stratosphere due to its limited accessibility. The physical and biological dynamics of bioaerosol particles in the stratosphere are closely related to aerobiological and astrobiological research topics such as global-scale long-range transport of bioaerosol particles, planetary protection, and the emerging and evolution of life. Thus, the development of the new experimental methodologies in the stratosphere will lead to the acquisition of the new high-altitude aerobiological research opportunities.

In the presentation, we will present the details of our experimental platforms for aerobiological research projects in the stratosphere that we have established using scientific balloons, and the research objectives of each research project.

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Guide to Determine the Allergenic Potential of Tree Pollen in the Netherlands

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Abstract

Allergic rhinitis, triggered by airborne pollen, is a prevalent condition significantly impacting the quality of life for patients and incurring substantial societal costs. Mitigating high concentrations of pollen in the air is crucial for establishing a safe environment for individuals with allergies. Due to climate change, the heat in cities during the summer is a recurring problem. The local climate can be improved by using the cooling properties of trees, providing shade and cooling by evapotranspiration. When selecting tree species for planting, it is relevant to consider the allergenic nature of the pollen they produce. Existing (global) guides classifying the allergenicity of tree pollen exhibit considerable variation in content and interpretation, rendering them unsuitable for a local context. Therefore we decided to develop a allergenicity guide for tree pollen relevant for the Netherlands.

Initially, a compilation of the prevalent tree species in the Netherlands was created. Following that, three information sources were employed in the development of the guide (1) A systematic literature search was conducted to compile information on the allergenicity of pollen from the various tree species. (2) An assessment was made of local pollen production based on the seasonal pollen index (SPI). and (3) The sensitization patterns in a group of Dutch patients with potential inhalation allergies were examined. Based on the data from these three sources the allergenicity of the pollen from the most common trees in the Netherlands was categorized into five classes. Out of the 61 tree species in the Netherlands, three species/genera were identified as possessing a (very) high allergenic potential, five species as moderate, 14 tree species as weak and 39 species as low. For the 14 species with a weak allergenic potential (minor) allergenic properties have been described for other parts of the world, but sensitization to these pollen in the Netherlands is still low. However, planting large numbers of these tree species should be avoided since their allergenic potential may change due to climate change or overplanting.

The approach outlined in this study can be applied to create allergenicity guides for tree pollen in different geographical areas. It is highly recommended to develop region-specific guides, as they furnish individuals responsible for tree planting decisions with pertinent information tailored to their specific locality.

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The Partitioning Processes of Sea Ice Associated Ice Nucleation Particles Impacting the Arctic Clouds

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Abstract

The Arctic is a region that is particularly vulnerable to climate change, as it is warming at a much faster rate than the rest of the globe. This warming causes a decline in multiyear sea ice cover, which results in an increase in ice-free waters with a much lower albedo compared to ice, therefore leading to a positive feedback and enhanced warming. Another factor that plays a role in regulating the temperature in the Arctic is the type and extent of cloud cover. Aerosols, that can serve as cloud condensation nuclei or ice nucleating particles (INPs), are key for cloud formation and aggregation state of water; liquid or ice. Some microorganisms produce INPs, but it is not well understood which microorganisms are most relevant, which environments they inhabit, and how active they are.

In this study, we investigated the partitioning of INPs between the Arctic marine and atmospheric environment by combining in situ measurements with laboratory experiments, deploying cold-stage INP measurements and amplicon sequencing of the bacterial and eukaryotic communities.

First, we determined if sea ice acts as a reservoir for INPs and, whether the INPs are partitioned into the sea ice during its formation or whether they are produced by microorganisms within the sea ice. We used a modified ice-finger to grow sea ice using sea water collected in West Greenland. We found that INPs are not enriched in the ice, but they stochastically incorporate into the ice fraction during sea ice formation. Next, we studied the temporal and spatial dynamics of INPs in Arctic sea ice cores that were collected before and during the spring sea ice phytoplankton bloom. We observed a higher concentration of INPs active at -10 C (INP_{-10}) present towards the bottom of the sea ice core with concentrations 10-100-fold higher than those in the under-ice water. Finally, we determined the potential of sea-ice as a source of atmospheric INP. In separate experiments, we filled bulk water and water from melted sea ice from Nuuk and Station Nord into a temperature-controlled sea spray simulation chamber and quantified and characterised the microorganisms and INPs present in the bulk water and air before and after the water bubbling. We observed that the highly active INPs are efficiently aerosolized by bubble-bursting together with specific bacterial and eukaryotic taxa. In light of these findings, this study provides new insights into the role of Arctic sea ice as a reservoir for INPs and the microorganisms that potentially produce them. Additionally, it explores the mechanisms by which INPs are released into the Arctic atmosphere. Last, these results can be used to improve cloud and climate model predictions in the Arctic region.

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Challenges for the Continuation of Long-term Pollen Datasets With New Automatic Pollen Networks

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Abstract

Introduction

New automatic pollen monitors have been developed in recent years and are increasingly used across Europe. Starting in 2021, the stations of the Swiss pollen network were gradually equipped with automatic Swisens Poleno monitors, being made the official devices in January 2023. Besides for the numerous advantages of automatic pollen measurements, the new method also implies changes in measurement characteristics and data. It is therefore necessary to define procedures for how the long, valuable historical pollen data can be combined with these new observations and continued into the future. They are the basis for climate monitoring and analysing climate change impacts. In an ongoing study, parallel Poleno and Hirst measurements are compared and differences and regularities characterized. The final goal is building transfer models between Hirst and Poleno data and homogenizing the historical data.

Methods

For comparison and quality management, all Hirst type traps were run in parallel with the Poleno devices until at least the end of 2023, with some Hirst measurements being continued at certain sites thereafter as well. Daily mean pollen concentrations of Hirst and Poleno data from the 15 Swiss monitoring sites were compared for 2023 and of 10 sites for 2022. The Poleno raw data were multiplied with a scaling factor for each pollen taxa, identical for all sites in both years, to generate concentrations of similar intensity to the Hirst data. The data were analysed and visualized with correlation analysis, different plots and the Wilcoxon test for the differences of the Seasonal pollen integral SPIn.

Results

The correlations between the daily pollen concentrations of the Poleno and Hirst were high, frequently above 0.8, showing that the general seasonal pattern of both measurement devices was very similar. However, the mean daily pollen concentrations differed between the Hirst and Poleno by more than the measurement accuracy of Hirst. The mean ratio of the SPIn Poleno/Hirst for the years 2022/2023 was for *Corylus* 1.00/0.53, *Alnus* 1.10/1.15, *Fraxinus* 1.31/1.56, *Betula* 1.15/2.08, *Fagus* 1.31/0.62, *Quercus* 0.52/0.75 and Poaceae 0.91/1.02. The significant difference in the intensity ratios of some of the taxa in the two years is difficult to interpret. A common feature was that the Poleno data were mostly higher at low pollen

concentrations and similar or lower at high concentrations than the Hirst. The comparison study provided several indications of incorrectly identified pollen by the Poleno.

Conclusion

In general, the results from the parallel measurement phase show that with one or two years of parallel measurements, little can be said about the regularity of the differences between Poleno and Hirst concentrations for most pollen types. However, only these initial comparative analyses have been carried out so far and more precise analyses of the influence of wind or temperature on the measurements are still pending. The systematic differences at lower and higher concentrations are a promising feature for future models for data homogenization. The ongoing improvement of the Poleno identification models, and the future calibration of the Poleno devices will reduce the unknown parts in this comparison.

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Evaluation of applying spatial interpolation of current airborne pollen concentrations where no monitoring exists to improve pollen monitoring networks

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Abstract

Allergy to airborne pollen impacts daily life of about 20% of the population, especially during spring, when the main pollen season occurs. Allergy symptoms can be reduced or avoided when restricting exposure to the allergens. Pollen monitoring programs help in this way by identifying and quantifying the pollen loads in the atmosphere. In addition, automatic monitoring traps facilitate this information in near real-time. However, due to budget limitations or lack of experienced personnel, it is not possible to have a monitoring trap in every town or city. Therefore, other solutions are needed to know the pollen content in the atmosphere.

Mathematic interpolation models have been largely used in many fields to calculate the distribution of a variable of interest along a spatial area. One of such interpolation methods is "Kriging", which has variants, i.e., co-Kriging, that include in the calculation other variables (co-variables) known to affect the presence/absence of the variable of interest.

Thanks to the knowledge acquired from decades of pollen monitoring, temperature (modulated by altitude) and rainfall emerged as the two most important weather variables to impact pollen levels in the air. With the co-Kriging method, it can be calculated how much pollen is in the air between the locations with a pollen monitoring station, taking into account the rainfall and temperature from the area of interest.

In our work, we used co-Kriging to calculate pollen concentration of birch and grass pollen, with altitude and rain as co-variables, in Bavaria (south of Germany). The project was funded by the Bavarian State Ministry of Health, Care and Prevention in collaboration with the Bavarian Health and Food Safety Authority (LGL). The pollen data, provided by the LGL, which implements the Electronic Pollen Information Network Bavaria (ePIN), and online-weather information from the "Deutscher Wetterdienst" (DWD) were used as input values. The accuracy of the interpolation was tested for 3 hours, 12 hours and daily values.

We did an internal validation of the predicted interpolations with the leave-one-out method, and also an external validation, *i.e.*, checking the predictions with four independent pollen measurements at selected locations using conventional Hirst-type pollen traps. The internal and external validations both showed that the best results were obtained at 12- and 24-hours resolution, with more than 0.6 correlation between observations and interpolations (minimum value considered as a good interpolation). The accuracy of the interpolation at 3 hours decreases with wrongly predicted high pollen peaks. When outliers are removed, the correlation between observations and interpolations overpasses 0.6 in three of the four validating sampling sites.

Providing the best possible interpolation between stations will avoid that patients need to select themselves which pollen monitoring station suits for their symptoms best, and will expand the use of automatic devices. This practice is already common in other fields, like local weather parameters where no stations exist. For pollen this was missing. Our system will assist allergic individual via the Bavarian ePIN to better react to local pollen exposure *i.e.*, enabling targeted avoidance if needed. Thus improving the monitoring service.

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The Impact of Climate Change on the Spread of Airborne Pollen in Northern Italy - the Results of 27 Years of Monitoring in Parma

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Abstract

Background: Pollen grains play an important role in the etiopathogenesis of seasonal respiratory allergies, which are increasing in prevalence and severity worldwide. Climate change is one of the possible explanations for the increase of pollen allergies. Warmer springs and autumns can cause some plants to produce pollen earlier or to extend the pollen season. The increased CO_2 concentrations also enable plants to produce more allergenic pollen, in larger quantities. Pollen count data could be considered the proxy for aeroallergen exposure. Long pollen data sets allow to investigate the trends in seasonal characteristics over time. This study examines temporal variations in seasonality of airborne pollen and meteorological data recorded over 27 years (1997–2023) in Parma (Italy).

Methods: The study was performed collecting pollen by a Hirst spore trap placed at 18.2 m above ground level, latitude 44°48'N, longitude 10°16'E. We have considered pollen data from the following taxa: *Betula, Corylus*, Cupressaceae (Taxaceae), *Platanus*, *Ambrosia*, Poaceae, total pollens and *Alternaria* spores. Start and end date, duration, date of peak, peak value, and Seasonal Pollen Index (SPI) were examined. Temporal variations in pollen seasons were displayed as the number of days from January 1 (DOY, day of the year). Daily average pollen concentrations were expressed as the number of grains/m^3, and the SPI were expressed as grains. Meteorological data were obtained from the Meteorological Station at the University of Parma and daily averages temperature, relative humidity (%) and total rainfall (mm) were considered. Linear regression analysis was carried out by using Microsoft Excel to investigate trends in meteorological data over time.

Results: A weak positive trend was observed for SPI of *Corylus* ($r = 0.45$) and Cupressaceae ($r = 0.30$). The start date resulted precocious for *Corylus* and Poaceae ($r = 0.4$), but late for *Betula* ($r = 0.57$). The end date and the duration of pollen seasons were longer for *Corylus* ($r = 0.4$), Poaceae ($r = 0.38$) and total pollen ($r = 0.44$), the duration become shorter for *Betula* ($r = 0.40$). The peak date was anticipated for Poaceae ($r = 0.58$), while the peak values were reduced for Poaceae ($r = 0.45$) and total pollen ($r = 0.33$). Regarding *Ambrosia* the duration were shorter ($r = 0.36$), and the peak date postponed ($r = 0.48$). No significant differences were observed for *Platanus* and *Alternaria* spores. A significant decrease of the relative humidity ($r = 0.63$) and a significant increase of annual average temperature ($r = 0.67$) were observed. No significant variation of total rainfalls was observed.

Discussion: The observed changes in pollen season parameters were associated with characteristics that can be related to exposure (i.e., duration, peak value, and SPI). The results

provided useful information to determine exposure of the allergic population to pollen in a One Health perspective. This highlights gaps in current knowledge and the need to quantify the impact of the climate change and land use.

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The Effectiveness of the Copper Electrostatic Filtration System "Aerok 1.0" for Air Disinfection

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Abstract

Background: Bioaerosol represents a risk for human health and the integrity of materials. Considering that people spend 90% of their time indoors, maintaining high indoor air quality is crucial. Air quality is essential for health protection and for preserving indoor environmental conditions. The aim of this study was to evaluate the effectiveness of the air treatment device "AEROK 1.0" (hereinafter AEROK) in reducing microbial, particle and pollen airborne contamination in working indoor environments, during the current activity.

Methods: AEROK, equipped with cupper electrostatic filtration system, was placed in a room used for administrative purposes at the Laboratory of Hygiene and Public Health, of the Department of Medicine and Surgery (University of Parma). The concentration of airborne microorganisms was measured using active sampling and microbial sedimentation rate was measured by passive sampling. *Triptic Soy Agar* medium (TSA) was used to measure bacteria and *Sabouraud Dextrose Agar* medium (SDA) was used to measure fungi. TSA plates were incubated for 48 hours at 36±1 °C and SDA plates were incubated for 120 hours at 25±1 °C. Microbial monitoring was performed for five working days. In the same days the microclimatic parameters were measured, using a device equipped with probes for temperature, relative humidity, and CO_2. A particle counter was used to evaluate particles concentration ≥ 0.5; 1.0; 2.0; 5.0; 10.0; 25.0 μm of diameter over a period of 200 minutes. The sampling of airborne pollen was carried out, continuously for 7 days by a 7-day volumetric Hirst-type pollen trap; the windows were kept open to allow the aerospora entry from outside. Statistical analysis was performed by using the SPSS version 28.0; p values <0.05 were considered statistically significant.

Results: The decrease in airborne bacterial contamination assessed by active sampling was 56.72% and 69.13%, with MD8 and DUOSAS active samplers, respectively. The decrease in

airborne bacterial contamination assessed by passive sampling was 56.49%. For fungi, the reduction was assessed at 38.7 by active sampling. Moreover, a significant reduction of airborne particles with diameters ≥0.5, 1.0, 2.0 µm ($p<0.001$) was observed. AEROK significantly reduced the ratio indoor and outdoor concentrations of total pollen ($p<0.001$) and spores of *Alternaria* spp ($p<0.05$).

Discussion: This study provided an assessment of the effectiveness of the AEROK for air disinfection (microbial, particle, pollen contamination) in a working environment under field operating conditions. These results are particularly relevant, since their obtained during the current working activity and, in the case of pollen and fungal spores monitoring, with windows kept open. The approach used also represents a contribution to the definition of a standardized model for the evaluation of the effectiveness of these devices.

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Co-exposure Analysis to Allergenic Airborne Pollen and Fungal Spores in Italy

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Abstract

Background

Seasonal allergic respiratory diseases are major public health issues, with high prevalence and significant burden. An overall assessment of the epidemiological and clinical trial data suggests that between 50% and 80% of patients consulting allergists are poly- sensitised. Co-exposure to several allergens in poly-sensitised patients is associated with more severe nasal and asthma-like symptoms, with respect to mono-sensitised one. Pollen co-exposure is also becoming more prolonged and more intense, as climate change is modifying pollen season, pollen production and allergenicity. The results of a recent study, comparing data from several European countries, indicate that the phenomenon of the simultaneous presence of multiple airborne allergens is common, especially in regions with a temperate climate. Italy is considered a country with a temperate climate, where the climates can be very different as can the complexity of the vegetation composition. This study shows preliminary results of co-exposure to high concentrations of airborne pollen and fungal spores of *Alternaria* in some Italian regions, referring to 2019.

Methods

The chosen monitoring stations (Bari, Napoli, Florence, Parma and Aosta) belong to five Italian regions (Apulia, Campania, Tuscany, Emilia-Romagna, and Valle d'Aosta, respectively). The data on pollen concentration were obtained by the OpenData web services of the Italian Aerobiological Monitoring Network (POLLnet). To analyse the variations in environmental concentrations the following pollen taxa (*Alnus* sp, *Betula* sp, *Ambrosia* sp, *Artemisia* sp, *Corylus* sp, *Cupressaceae-Taxaceae*, *Quercus* sp, Poaceae, *Olea* sp, Pinaceae, Plantaginaceae, Platanaceae, Urticaceae) and fungal genus *Alternaria* were chosen. The threshold values were chosen according to the literature. The study analysed the values higher the threshold values.

Results

The days with pollen concentrations above the threshold values differ considerably from one region to another. In the regions of southern Italy, such as Campania and Apulia, the phenomenon of co-exposure is marginal; in others, especially those of centre and northern Italy (Tuscany, Emilia-Romagna and Valle D'Aosta), it is particularly evident. The region with a higher number of days with concentration of pollen over the threshold values was Emilia-Romagna, with 216 days, 149 days with co-exposure values for two pollen types, 59 days with three pollens, 6 days with four pollens and 1 day even five different types of pollen. Regarding *Alternaria* spores the results showed 105 days with concentration above the threshold values, with 75 days of co-exposure with pollen (*Ambrosia*, Plantaginaceae, Urticaceae). The region with low pollen concentration was Campania with 49 days above the threshold values and only 1 day with co-exposure values (*Olea* and Plantaginaceae). The concentration of Alternaria spores never exceeded the threshold values.

Discussion

Co-exposures and poly-sensitization to several pollen types and fungal spores are very common in real-life practice. The present study shows the first preliminary results in any Italian region. In some of them, the co-exposure is particularly evident in terms of number of days and number of pollen types involved. Our aim is to extend the observations to a higher number of Italian regions, comparing the trends over time and considering the climatic zones.

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Microscale Pollen Release and Dispersal Patterns in Flowering Grass Populations: Exploring eDNA and Isolated Festuca rubra Flowering in a Rural Landscape Setting

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Abstract

Background and Aims: Grass pollen is the main contributor to seasonal allergies and is thus a major threat to public health. However, substantial knowledge is lacking regarding their release and dispersion processes, leading to a reduced understanding of these ecological factors, their predictability and efficiency. Furthermore, the variability of grass pollen atmospheric diversity has not been fully explored, contributing to uncertainty in their seasonal distribution. These are relevant issues, as grasses vary in their species-specific allergenicity and have highly heterogeneously distributed source areas. In this study we aim to investigate local level heterogeneity of airborne grass pollen release and dispersion processes, with a focus on characterizing the taxonomic diversity using eDNA and molecular ecological methods.

Methods: Three established pollen sampling stations located in close proximity (< 300 m apart) within the Lakeside campus in Worcestershire, United Kingdom were utilized to investigate microscale grass pollen release and dispersion factors. Bi-hourly grass pollen concentrations were sampled for 54 days (14.05 to 06.07 in 2019) and modelling in a MANOVA framework with local meteorology and wind conditions (incl. turbulent kinetic energy) to investigate relevant environmental factors for the release and dispersion processes. Simultaneously, grass pollen eDNA was collected using multicyclone equipment, sequenced using Illumina MiSeq for metabarcoding (ITS1 and ITS2), referenced against a custom bioinformatics database containing all UK grasses and analysed using Shannon's Diversity Index (α-diversity) to identify the airborne grass species diversity. Furthermore, the flowering phenology of an isolated red fescue (*Festuca rubra*) population was concurrently observed to identify its contribution to atmospheric pollen levels.

Results: We found that grass pollen varied at the microscale level. This was likely attributed to the dispersion of grass pollen from local source areas and local topography. The metabarcoding analysis revealed six genera that dominated the seasonal atmospheric profiles (*Agrostis, Alopecurus, Arrhenatherum, Holcus, Lolium* and *Poa*), accounting for, on average, 77% (40 – 92%) of the relative abundance of grass species reads. The grass genera and relative abundances varied significantly between the three microscale sites. Relative humidity, solar radiation, temperature, wind speed and mean TKE were all relevant for the release and dispersal processes of grass pollen. The *Festuca rubra* population contributed almost 40% of the relative

pollen abundance to the adjacent sampling station, but only contributed 1% to the sampling stations located 300 m away.

Conclusion: Our results show that most grass pollen has limited dispersal distance as there was substantial variation in grass pollen concentrations and species composition over short geographical scales. Multiple local meteorological factors, including TKE, were relevant for the release and dispersion processes of grass pollen.

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Fungal Non-pollen Palynomorphs in a Temperate European Lowland Forest: Insights and Aerobiological Implications

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Abstract

Fungal Non-Pollen Palynomorphs (fNPPs) can serve as important paleoecological indicators within forest ecosystems. This study represents the first exploration of fNPPs in a temperate European lowland forest. I examined fNPPs in moss samples collected from the Białowieża Forest, known for its rich fungal biodiversity. The analysis revealed 98 morphological types, predominantly saprotrophs thriving on diverse substrates, and highlighted the interconnections among forest characteristics, herbaceous plant diversity, and dead wood components, influencing fNPP composition, richness, and proportion.

Two primary gradients emerged, delineating samples based on taxonomic richness and composition of fNPPs, showcasing the complex dynamics within the forest ecosystem. Environmental predictors, including forest type and herb pollen richness, significantly impacted fNPP taxonomic richness and diversity. Notably, deciduous forests exhibited higher fNPP richness, correlated with increased herb pollen richness. Conversely, fNPPs decreased with higher volumes of stumps, coarse woody debris, and canopy openness, underscoring the complexities of forest management's impact on fungal diversity.

These findings not only illuminate the dynamics of local ecosystems but also provide a solid framework for aerobiological research in forest environments. Consequently, we initiated aerobiological monitoring in the Białowieża Forest - a biodiverse natural environment known for its rich fungal species diversity, providing an ideal setting to explore the composition of airborne fungal particles. Its unique features, including a natural level of dead wood, offer a valuable opportunity to investigate the fungal species composition and concentrations in the atmosphere.

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Exploring the Influence of Spatiotemporal and Meteorological Variation on Norwegian Atmospheric Pollen: A Multiregional Bioaerosol Modelling Study

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Abstract

Background and Aims: Being able to predict plant responses to their environment is essential in many ecological, economical and medicinal disciplines. This is especially relevant in terms of climate change, which is expected to affect temperature, precipitation and all derivate meteorological patterns via changing atmospheric and oceanic circulation. Plant phenology has been shown to be strongly dependent on environmental variation, both short-term via weather fluctuations and long-term via climate change, with consequences to food-webs, production potential and pollen seasonality. This is relevant for many species, including but not limited to birches, oaks and grasses, all of which produce highly allergenic pollen to many human populations. However, to what degree different types of environmental variation is expected to alter pollen seasonality has not been fully explored for all pollen types, and many research findings remain contradictory. The primarily aim of our study is to explore how temporal, spatial and meteorological variation contributes to pollen seasonality within the Nordic country of Norway, and if possible identify ecological relationships influencing the variation.

Methods: We modelled the daily concentrations of seven common Norwegian pollen types: alder (*Alnus*), hazel (*Corylus*), willow (*Salix*), birch (*Betula*), pine (*Pinus*), grass (Poaceae) and mugwort (*Artemisia*) using generalized linear models with negative binomial distributions. The data was obtained from twelve regional sampling stations and for up-to 28 years. The individual models contained three temporal (DOY, month and year), four spatial (latitude, longitude, altitude and sampling height) and six meteorological variables (air temperature, precipitation, relative humidity, atmospheric pressure, wind speed and solar radiation). The meteorology was obtained from the MET Nordic dataset with full cover. The variables were compared thematically and individually in a reduced-model framework using chi-squared anova tests supplemented by AIC and Nagelkerke Pseudo-R^2 to assess the variation contribution.

Results: The full models were able to explain the most variation in comparison to reduced models, with R^2 up-to 49.5%, 51.7% and 59.5% for pine, birch and grass pollen respectively. Temporal variables were able to explain more variation than spatial or meteorological variables on average for most pollen types, with the exceptions being hazel and grass, for which spatial variables had higher explanatory power. While the contribution of individual variables varied based on the pollen type, on average, month, altitude and maximum temperature were the variables in each thematic category with the highest contribution.

Conclusion: Temperature cues and the natural timing of phenological development were identified as the main pools of variation for the pollen seasonality. While our models could

explain a substantial proportion of the variation, model understanding could likely be enhanced by including source maps and regional atmospheric transport modelling.

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Test of a New Rapid-e+ Air Flow Cytometer for Automatic Monitoring in Aerobiological Network

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Abstract

The research in the frame of SYLVA project (GA no. 101086109) investigated the performance of a new version of the Plair SA air flow cytometer, known as Rapid-E+, for monitoring airborne pollen in operational networks. In this study we created a machine learning algorithm to classify 27 pollen types commonly suspended in the air of Pannonian plain. The evaluation of pollen identification has been performed both on reference dataset and by side-by-side comparison to standard Hirst type measurements (CEN EN 16868) in real life monitoring from April to September 2023.

The ability to identify different pollen classes was similar to Rapid-E (precision, recall and F1 score at 83 %, 85 %, and 0.84, respectively). However, the agreement with manual Hirst-type observations was lower, except for total-pollen measurements (Spearman Rho = 0.7). Notable differences in fluorescence measurements were observed among three tested Rapid-E+ tested, leading to significant errors when applying recognition algorithms across devices.

The study confirmed the potential of combining fluorescence and laser scattering for distinguishing pollen. However, the major uncertainty is seen for fluorescence spectrum and fluorescence lifetime measurements that both are important for distinguishing pollen from other aerosols and between different pollen classes. Additional endeavours are necessary from the manufacturer to boost stability and the signal-to-noise ratio of fluorescence measurements. This is expected to improve classification for more bioaerosols of interest, especially for regions where various classes of pollen and fungal spores are concurrently present in the atmosphere.

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Association Between Google Trends Data Flows and Pollen Exposures in Italy

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Abstract

Background: Over the past decades, the prevalence of seasonal allergic respiratory diseases has increased in Western countries. Airborne pollen monitoring by Hirst-type samplers is time-consuming and requires continuous expert involvement. Recent studies investigate allergic diseases and pollen concentrations using internet search platforms in search of simpler and more cost-effective perspectives. This approach provides a more accessible means of monitoring environmental and medical events related to allergies, potentially improving public health responses.

This study was carried out to explore the use of Google Trends (GTs) as a proxy of epidemiological surveillance for pollen respiratory allergies.

Methods: The study analysed search data about pollen allergy from 2004 to 2019 in Italy. We searched in GTs the keywords allergy-related search (ARS) as: "rhinitis", "asthma", "allergy", "antihistamine" "pollen", and pollen-type search (PTS) (Poaceae, hazelnut, birch, olive, Cupressus, ragweed, and plan tree) from some Italian regions. For testing the correlation between ARS and PTS google trend search or PTS, ARS and pollen concentrations, Kruskal-Wallis's test and Person's test were used, respectively. Five Italian regions (Piedmont, Emilia-Romagna, Tuscany, Apulia, and Campania) were analysed. Pollen concentrations were obtained by the OpenData web services of the Italian Aerobiological Monitoring Network (POLLnet) available online. The statistical analysis was performed by SPSS version 28.

Results: The GTs data analysis shows that Emilia-Romagna had the highest median searches for ARS (125), followed by Veneto (98), and Friuli-Venezia-Giulia (93). The highest PTS of Poaceae (98), ragweed (100), hazelnut (100.5), plan tree (92.5), and olive (97) were observed in Piedmont. In Emilia-Romagna (p =0.0003), Campania (p =0.0014), and Apulia (p =0.0001), significant correlations between ARS and PTS were found for Poaceae. Hazelnuts correlate in Emilia-Romagna (p =0.0003), Tuscany (p =0.004), and Apulia (p =0.0001). Birch in Emilia-Romagna (p =0.0004), Tuscany (p =0.0022), Apulia (p =0.0000), and Campania (p =0.0000). The correlation was significant for olive in Emilia-Romagna (p =0.0013), Tuscany (p =0.012), Campania (p =0.0001), and surprisingly not in Apulia. Regarding the plan tree in Emilia-Romagna (p =0.0003), Tuscany (P = 0.018), Campania (p =0.0000), and Apulia (p =0.0001), we found a significant correlation. Regarding pollen concentration, a significant correlation was found as follows: ragweed with ARS (p =0.043) and Poaceae with PTS (p =0.021) in Piedmont; Poaceae in Emilia-Romagna with ARS (p = 0.026) and PTS (P =0.050); in Campania with ARS (p =0.028) and PTS (p =0.015); and in Apulia and Tuscany only with PTS (p =0.015, p =0.018, respectively).

Discussion: Internet search data by GTs could be a useful tool to implement information obtained from the analysis of pollen data. Further investigations are needed to better understand the meaning of the results obtained. In the future, this approach could improve forecasting in a system that includes, in addition to what is used today, data on the flows of searches carried out on the internet.

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Immigrants and Seasonal Respiratory Allergies: a Look Observing Internet Search Flows - the Situation in Italy

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Abstract

Background: In Italy, immigrants from low-income countries initially exhibit lower allergy prevalence than natives, but over time, seasonal respiratory allergies increase with different patterns between first- and second-generation immigrants, with Sub-Saharan populations more susceptible to asthma. In 2021, Italy had 5,193,669 foreign residents (8.8% of the total population). Most of them living in Central-Northern Regions (Lombardy, Lazio, and Emilia-Romagna). Romanians, Albanians, Moroccans, Chinese, and Ukrainians are among the top five communities. The study was carried out to explore the use of Google Trends (GTs) as a proxy of seasonal respiratory allergies in immigrants living in Italy related to airborne pollen.

Methods: By GTs we analysed flow searched terms from Italy related to allergy and performed in English, French, Romanian, Albanian, Chinese, Ukrainian and Arabic languages, from 2009 to 2019. The search terms flow as such as “allergy”, “rhinitis”, “asthma”, “antihistamine” and “pollen” were analysed. Descriptive statistics were conducted using Kendall's Tau-b correlations. Values of $p < 0.05$ and < 0.01 were considered significant as 0.01 = ** and 0.05 = *. The analysis was carried out with SPSS version 28.

Results: GTs search data has showed distinct trends in pollen-related allergy queries from Italy, categorized by language. In Italian, April recorded the highest percentage of searches (12.13%), followed by May (11.84%) and March (9.40%), while the search rate from July to September remained around 7%. French and Chinese searches reached the peak in April (12.17% and 23.85%, respectively), while Albanian and Romanian searches peaked in June. Arabic searches exhibited two peaks in April (11.93%) and in August (13.68%). We observed a positive strong correlation between Italian and English**, Albanian**, French**, Romanian**, and Chinese**. There was no

correlation between Arabic and Italian or Chinese, with correlations observed with Albanian* and Romanian*. We observed also strong correlation between Chinese *vs* English**, or French**. This approach has raised the problem of translating GTs search terms into Cyrillic; unfortunately, in this case we did not find any matches.

Discussion: The variation in search patterns across languages highlights different dynamics of awareness and concerns related to pollen-related allergies within different linguistic communities of immigrants, considering that French and English are also common in some African countries. The tourism population movement could also have some influence considering that in Italy e.g., in 2019, there were 65 million arrivals (ISTAT data) of which 71.7% coming from European Union countries and 18.3% from non-European area, especially from the United Kingdom. Another important variable it might have been the rate of computer literacy interfering with online searches skill. In any case, the analysis of the flows of allergy-related internet search terms carried out from Italy in some of the languages used by the different ethnic groups could give useful information considering the not negligible number of immigrants who, for various reasons, may have a different attitude towards the disease and low access to care.

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Evaluating Automatic Pollen Identification Algorithms Against the Mean of Four Hirst-Type Traps: The Renewed Eumetnet Autopollen-Cost Adopt Campaign App

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Abstract

Automatic pollen monitoring is currently being adapted in Europe and elsewhere. Due to the dynamic developments in the field of artificial intelligence (AI), which is used for identifying pollen, new algorithms are constantly under development. Each new algorithm needs to be evaluated. However, how to achieve this is a challenge. Currently, most algorithms for automatic pollen identification are compared to data from Hirst-type traps even if it is known that these instruments suffer from large errors and high variability. Nevertheless, currently there is no other way to check if data from an automatic pollen monitor is correct.

One solution is to compare the data against the mean of a few Hirst-type pollen traps. This way the variability that exists between Hirst-type pollen traps is reduced, although systematic errors

remain. Few studies have placed four Hirst-type traps at the same location except for the EUMETNET AutoPollen – COST ADOPT campaign held over the 2021 pollen season in Munich, Germany.

In this campaign, four Hirst-type traps were situated on the same roof within 5 m of each other (1-3). Their clocks were adjusted and checked with a Witschi chronomaster (www.witschi.com, a mechanical clock controller), followed by manual test runs. The flow was adjusted to 10 l/min with a Burkard hand held rotameter, but corrected to the exact flow with an Easyfluxâ resistance free anemometer. Pollen per m^3 were calculated using the anemometer flow, as described in (3).

Although not perfect, this is the best we have using the current methods (4).

This dataset is publicly available at: https://autopollen.shinyapps.io/APP_AUTOPOLLEN_compare/. This is not the same app as used for the publication of the AutoPollen campaign (1), but has a few optimisations making it suitable for checking new algorithms, also those that were not available at the time of the campaign. The raw data from the automatic monitors has been stored, and can be re-analyzed with any new algorithm, with that algorithm then added to the database and connected to the app. In the new version of the app, any algorithm can be selected in the "Scatter plot" view, allowing comparison not just of automatic vs manual observations, but also various automatic instruments against one another. In addition, if a point in the Scatter plot or time series needs closer inspection, the date and value is displayed, so the original data can be easily consulted.

While this is no longer the same "blind" check of the algorithms as carried out for the intercomparison (i.e. the Hirst-type data are now public, during the campaign they were held closed to the algorithms would be tested without knowing what the "baseline" manual data showed), the app still allows an internal validation and assessment against previous versions of algorithms. An external validation using unknown manual observations is additionally suggested.

New algorithms can be submitted at any time. Please contact the app administrator (see the app: https://autopollen.shinyapps.io/APP_AUTOPOLLEN_compare/).

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Spatiotemporal Modelling of Grass and Birch Pollen Concentration Across Switzerland: a Comparison of Statistical, Machine Learning and Ensemble Methods

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Abstract

Statistical and machine learning are commonly used to model spatial and temporal variability in environmental stressors, to support the estimation of exposure in epidemiological studies. We aimed to compare the performances, strengths and limitations of six different algorithms in the retrospective spatiotemporal modeling of daily birch and grass pollen concentrations at a spatial resolution of 1 × 1 km across Switzerland.

Daily birch and grass pollen concentrations were available from fourteen measurement sites across Switzerland for the years 2000-2019. We considered several temporal, spatial and spatiotemporal predictors including meteorological factors, land-use, elevation, species distribution and Normalized Difference Vegetation Index to develop spatiotemporal models using six different statistical and machine learning algorithms: LASSO, Ridge, Elastic net, Random forest, XGBoost and ANNs. We then combined these six models through multiple linear regression to develop an ensemble hybrid model. We optimized model structures through feature selection and grid search techniques to obtain the best predictive performance. We used train-test split and cross-validation as validation strategies to avoid overfitting and over-optimistic models.

The 5^{th}–99^{th} percentiles for concentrations of hazel, alder, birch, ash, and grass pollen at the pollen monitoring stations were 0 – 1153 and 0 – 290 pollen grains/m^3, respectively. The hybrid ensemble model achieved the best RMSE on the external dataset for both grass and birch pollen with 19.67 and 94.35 pollen grains/m^3, respectively. Nonlinear models (Random forest, XGBoost and ANNs) achieved the lowest test RMSE for both pollen types ranging from 19.97 to 25.36 pollen grains/m^3 and from 105.86 to 140.47 pollen grains/m^3 for grass and birch pollen respectively, compared to linear models. (LASSO, Ridge, Elastic net). Among the linear models, Ridge regression performed poorly for both pollen types.

The random forest algorithm yielded the best performance among the six evaluated modelling methods and in general, except for XGBoost other machine learning algorithms performed better than statistical models. They are able to explain complex, non-linear relationships between environmental factors and measured concentrations. The ensemble hybrid model outperformed the six statistical and machine learning algorithms.

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Influence of Meteorological Variables and Air Pollutants on Measurements From Automatic Pollen and Spore Monitors

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Abstract

This study examines the influence of meteorological variables and air pollutants on the performance of automatic pollen and spores monitoring devices, as part of the EUMETNET Autopollen COST ADOPT-intercomparison campaign held in Munich, Germany, during the 2021 pollen season.

The campaign offered a unique opportunity to compare all automatic monitors commercially available at the time, a Plair Rapid-E, a Hund-Wetzlar BAA500, an OPC Alphasense, a KH-3000 Yamatronics, three Swisens Polenos, a PollenSense APS, an IBAC2, a WIBS, a Sextant to the average of four manual Hirst traps. The investigation aimed to elucidate how meteorological variables and air pollution impact on particle capture and identification efficiency. Therefore, instruments were placed at the same location to be under the same environmental conditions.

The analysis showed coherent results for most devices regarding the correlation between environmental conditions and pollen concentrations. This reflects on one hand, a significant correlation between weather and airborne pollen concentration, and on the other hand the capability of devices to provide meaningful data under the conditions under which measurements were taken.

However, the correlation strength varied among devices, reflecting differences in design, algorithms, or sensors used. Also, results from some monitors were affected to a minor degree by specific weather variables.

Additionally, it was observed that different algorithms applied to the same dataset resulted in different concentration outputs, highlighting the role of algorithm design in these systems (monitor + algorithm).

Notably, no significant influence from air pollutants on the pollen concentrations was observed, suggesting that any potential difference in effect on the systems might require higher air pollution concentrations or more complex interactions.

Our findings suggest that the application of real-time devices in urban environments should focus on the associated algorithm that classifies pollen taxa, which should include possible biases derived from specific weather variables. The impact of air pollution, although not to be excluded, is of secondary concern as long as the pollution levels are similar to a large European city like Munich.

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Association Between Short-term Pollen Exposure and Blood Pressure in Adults: a Repeated-measures Study

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Abstract

Background: Recent studies have related high pollen concentrations to increased cardiovascular mortality and morbidity, yet very little research concerns pre-clinical cardiovascular health, including effects on blood pressure (BP). The EPOCHAL panel study investigated the exposure-response relationship between ambient pollen exposure and systolic and diastolic BP in adults.

Methods: BP was measured in 302 adults with and in 94 without pollen allergy during the pollen season, on approximately 16 days per person (6305 observations). Experienced individually-

relevant pollen exposure (IPE) in the 96 hours prior to each BP measurement was calculated considering sensitization towards seven highly allergenic pollen and ambient pollen concentrations. Generalized additive mixed models were used to study the association between mean IPE in the last 96 hours and BP, adjusting for individual and environmental time-varying covariates. Effect modification by pollen allergy status, sex and BMI was evaluated.

Results: Positive non-linear associations between IPE and both systolic and diastolic BP were found in the allergic but not in the non-allergic group. BP increased sharply for exposures from zero to 60/80 pollen/m^3 (diastolic/systolic BP), followed by a tempered further increase at higher concentrations. Increases of 1.79 mmHg [95% CI: 0.65– 2.93] in systolic and 1.34 mmHg [0.45 – 2.23] in diastolic BP were associated with 96-hour average pollen exposure of 400 grains/m^3, compared to no exposure. Obesity (4.91 mmHg) and female sex (2.18 mmHg) were associated with larger BP increases.

Conclusions: The finding that short-term pollen concentration is associated with increased systolic and diastolic BP in persons with pollen allergy strengthens the evidence that pollen may cause systemic health effects and trigger cardiovascular events.

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Profiling the Tropical Fungal Aerobiome and Airborne Plant Pathogens of Ghana via Minion Metagenomic Sequencing of Air Collected by Drone-mounted Samplers

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Abstract

Hypotheses & background

To date, there have been no DNA-based metagenomic studies into fungal community structure and diversity in tropical Sub-Saharan African air. We hypothesised that the microbiome of West African air would be dominated by tropical species and that we could sample fungal spores by impaction onto Vaseline-coated rods mounted on a UAV, which was imaging forest and crop canopies.

Methods

13 air samples were collected onto Vaseline-coated acrylic rods mounted on drones flown at heights of 15-50 metres above ground for 10–15 minutes at three sites in Ghana. An additional

four air samples from a contrasting ecological region (temperate, UK) were included for comparison. Purified DNA was extracted from the Ghanaian air samples and the internal transcribed spacer (ITS) region was amplified using fungal-specific primers. Subsequent minION third-generation amplicon sequencing was under-taken with downstream bioinformatics analyses utilizing GAIA cloud-based software (at genus taxonomic level).

Results

Principal Co-Ordinate analyses based on Bray-Curtis beta diversity dissimilarity values resolved fungal assemblages from Ghanaian and UK air (>5000 km apart) distinctly but did not distinguish between individual Ghanaian sites (25–120 km apart) that were largely homogenous. In Ghanaian air samples, despite the very short flight times, a mean of ~133 operational taxonomic units (OTUs) were identified in each sample, demonstrating considerable alpha diversity. In Ghanaian air, fungal assemblages were heavily skewed at the phylum taxonomic level towards the ascomycetes (73.1%) as opposed to basidiomycetes (7.3%); at the class level, the Dothideomyectes were predominant (44.2%) followed by the Sordariomycetes (15%). The most common fungal genus in Ghanaian air was cosmopolitan and globally ubiquitous Cladosporium (~20% of reads). Interestingly, many fungal genera containing economically important phytopathogens of tropical crops were also identified in Ghanaian air, including Corynespora, Fusarium and Lasiodiplodia.

Conclusions & Perspectives

The drone based sampling at heights of 10-20m allowed sample collection at the same time as optical imagery data were collected for other purposes and provides a snapshot of spores present in a region as previous work shows double the species diversity at 10m compared to 1m. The heterogeneity of the Ghanaian samples is not surprising due to the sampling height above ground level of 10-50 m resulting in well-mixed air representing a regional scale being sampled. Combining spore collection with metagenomic sequence-based technologies (e.g. minION sequencing) will in future enable rapid real-time monitoring of the entire aerobiome to profile abundance and genetic traits (e.g. fungicide resistance) of many pathogens simultaneously.

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High Volume Cyclones in plant pathology – development and use cases

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Abstract

Background & hypotheses: Cyclones have been used since the 1950s to collect airborne spores. The Burkard miniature cyclone was used by many groups but only samples 16.5 litres of air per minute. Recent developments used to sample spores of crop pathogens include the Agri Samplers High Volume cyclone, which samples 200-270 Litres (* depending on version used) into 2ml screw-top tubes, and the Root Applied Sciences HVC, which samples 370 liters per minute into 1.5 ml Eppendorf tubes. To facilitate rapid DNA-based detection of airborne spores, it is helps to sample spores directly into a tube for ease of downstream processing steps. We hypothesise that sampling a high volume of air allows collection of a larger sample than collected by standard impaction traps, which will improve sensitivity of molecular detection methods. Various recently developed high volume cyclones (HVCs), such as the Agri Samplers High Volume Cyclone (AS HVC) and the Root Applied Sciences HVC (RAS HVC) were assessed.

Methods: Direct comparisons of devices were not made but various case studies in which the two HVCs were used successfully for pathogen monitoring are summarised here. The first used the AS HVC sampling 270 L/min in the UK (on a range of pathogens) and Brazil (on wheat blast). A modified version of the AS HVC sampling 200L/min in Australia (on a range of pathogens) and the RAS HVC in vineyards to detect grape powdery mildew in California.

Results: In the UK, ascospores of Sclerotinia (size 9 x 5 µm) and sporangia of Phytophthora infestans (Potato blight) (size 30 x 15 µm) were detected using the AS HVC and compared well to samples collected using a Burkard seven day spore trap. In Brazil, spores of the wheat blast pathogen (Pyricularia oryzae Triticum lineage) (conidia size 20 x 10 µm) were frequently quantified in air samples throughout the year. In Australia, Botrytis cinerea (conidia size 10 x 7 µm) and Myrtle Rust (spore size 13.75 x 12.5 µm) were collected using the same device modified to sample 200 L/min and the performance of the device was far better than the standard Burkard seven day spore trap. The RAS HVC has been used to monitor Grape Powdery mildew (Uncinula necator; conidia size 25-30 x 14-15 µm) in California with great success. In wind tunnel studies, it was found to collect more spores than a Burkard seven day trap, enabling greater sensitivity of detection by qPCR.

Conclusions: HVCs are effective sampling devices and have been used successfully to monitor a wide range of plant pathogens. Collection efficiency is often lower than most impaction traps but by sampling a much greater volume of air, the ability to detect rare spores or low concentrations of spores early in the spore season is enhanced. Further work is in progress to

optimise their use, including preventing remobilisation and loss of spores that have been deposited in the collection tube.

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Using Air Sampling to Monitor Fungicide Resistance in Plant Pathogens

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Abstract

Purpose & hypotheses: For over 20 years we have detected spores of plant pathogens using DNA-based diagnostic assays such as qPCR. Usually these target the detection and quantification of individual species. This study tested whether specific primers to detect and quantify the proportion of Pyricularia oryzae Triticum lineage (PoTl) with resistance to quinone outside inhibitors (QoI, FRAC group 11) fungicides could be used with DNA extracted from spore traps to monitor pathogen populations and advise farmers to switch to more effective fungicides.

Background: A previous study (Fraaije et al 2005) used Burkard seven day spore traps in wheat fields at Rothamsted Research (UK) in 2003 to detect the G143A mutation conferring resistance to QoI (Strobilurin, FRAC group 11) fungicides in populations of Zymoseptoria tritic, which is the most important fungal disease of wheat in Europe. A nested qPCR was used to quantify the concentration of airborne ascospores of Z. tritici and estimated the proportion with the G143A mutation of the cytochrome b gene. In 2003, the proportion of spores with the mutation started at around 30% but this increased to nearly 100% in fields that received further QoI sprays but increased only slightly in unsprayed fields. QoI fungicides stopped being used against Z. tritici and were always used with other modes of action when used against other pathogens.

Methods: The new study used Agri Samplers High Volume Cyclone Spore Traps at Londrina in Paraná State, a major wheat cropping region in Brazil. The HVC spore traps sampled 270 Litres per minute, depositing spores into 2ml screw-top tubes which were pre-loaded in a carousel to allow automated collection onto a new tube each day during a week. Samples were sent for lab-based DNA extraction and existing quantitative PCR (qPCR) assays used to estimate spore concentrations of Pyricularia oryzae Triticum lineage (PoTl), which causes wheat blast (brusone) in Brazil. In addition, on 10 days with high PoTl spore concentrations, further qPCR tests combined with SNP detection pyrosequencing found the proportion of PoTl with resistance to QoI fungicides (QoI-R) based on the G143A mutation of cytochrome b. The impact of weather on the dynamics of wheat blast aerosol populations was also investigated.

Results: Airborne spores were collected each day. PoTl inoculum was consistently detected in aerosols during the wheat cropping seasons from 2019 to 2021 but amounts varied significantly between seasons with greatest amounts in 2019 and during the non-crop season in 2020 and 2021. The occurrence of QoI resistance varied between 10 and 91%, which was related to wind direction due to nearby grassland that had not been sprayed with fungicides but is also affected by the same pathogen acting as an unselected reservoir of the pathogen population. In addition, statistically significant but low correlations were found between the airborne spore concentrations of the pathogen and various weather variables.

Conclusions: For wheat blast, this system provided prior detection of airborne spores ahead of symptom appearance and provided early indication of the prevalence of fungicide resistance alleles.

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Development of a New Dispersion-model Based Uk Pollen Modelling System.

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Abstract

Met Office pollen forecasts have traditionally been based on expert judgement by the National Pollen and Aerobiology Research Unit (NPARU), coupled with an adjustment for the forecast meteorology by the Met Office (NPARU-MO). These provide a five-day total pollen forecast for each of 16 regions over the UK. We have developed a prediction system based on the Met Office NAME dispersion model which allows the provision of automated, species-specific, gridded forecasts at 0.05° and hourly resolutions.

Within NAME we have implemented a new pollen emission scheme. The seasonal cycle is parametrised based on a heat sum method, while short-term variations are dependent on the vapour pressure deficit, precipitation, wind and an additional diurnal cycle. Six species are currently included in the model: alder, hazel, birch, oak, grass and nettle.

The UK pollen observation network is run by the Met Office, with observations recorded using Burkard volumetric spore traps of the Hirst design at around 11 sites. Pollen grains are counted bi-hourly and recorded along with the total daily sum. Use of the bi-hourly data has been critical for developing parametrisations for NAME. In addition we have developed a model verification

system which allows us to compare hourly NAME predictions to the bi-hourly data, providing a deeper understanding of the results compared to using daily resolution outputs.

Hindcast simulations have been carried out for 2022 and 2023 and results compared against observations. Metrics quantifying the prediction skill have been computed and compared to those for the NPARU-MO and the CAMS ensemble pollen forecasts. The main variable to verify against is the UK Daily Pollen Index (DPI), which converts each species concentration using health-based thresholds into an integer, representing Low, Moderate, High or Very High pollen levels, with the maximum value across all species giving the overall DPI.

The results demonstrate that for each year simulated, the NAME predictions for DPI have improved correlation, bias and RMSE compared to both NPARU-MO and the CAMS ensemble. If the group of species considered is limited to those six currently included in the model, the improvement in skill is greater still. This indicates that the current six species model captures the majority of the pollen types influencing the UK forecast but that there remains some scope for improvement by the addition of other species. The inclusion of oak in NAME gives a significant improvement in performance for DPI compared to the CAMS ensemble, which currently does not include this species. Results vary between the species, with daily mean grass pollen correlations exceeding 0.7, while for tree species such as birch, there is a significant inter-annual variation in the magnitude of the pollen season which is harder to predict for forecasting but can be corrected for with historical seasons.

This verification has provided us with enough confidence in our model that we are now developing a UK pollen air concentration reanalysis. This will cover the period 2000-2023 and be bias corrected using observations: this will be particularly useful for pollen health impacts studies.

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Using Lfd Assays to Assess the Occurrence of Potato Blight Sporangia in an Automated Real Time System

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Abstract

Lateral flow assays, which detect sporangia of the potato blight pathogen *P. infestans* were developed based on recombinant IgG antibody fragments. The specificity of these lateral flow devices (LFD) were tested under laboratory and field conditions using a range of pathogens commonly found in the air in potato crops. LFD devices were used within an auto analyser trap specifically designed for early detection of crop disease pathogens within cropping environments. The autoanalyser uses a high volume cyclone sampler to trap particulates and micro-organisms in the air. One analyser can monitor a wide geographic area of around 100 Ha (dependant on local environmental conditions). The system has been tested using potato blight (*Phytophthora infestans*) as the target organism. The automated function enables samples to be routinely analysed for potato blight sporangial presence as detected using a specific LFD device. The analyser transmits 4G mobile data for storage in the cloud. Local pathogen data is analysed alongside local weather data within disease risk models to determine risks of crop disease infection. Analysis outputs at a local, regional or even national level can be viewed on multiple display devices with automatic alerts set at predetermined levels.

The daily LFD estimates of potato blight sporangia were compared to molecular estimates using similar daily samples obtained by PCR techniques. There was a high correlation between LFD and molecular estimates of the presence of *P. infestans* sporangia in air samples over a 2 month sampling period. Fragmentation of samples using beads increased the sensitivity of the assay. This could be used in the early stages of crop grow to identify initial development the *P. infestans* in the crop.

The project brings together necessary components of a disease management system and delivers user-friendly, in-field tests to growers and agronomists. Estimates of disease within the crop could be obtained in the absence of potato blight symptoms. A capability for both automated LFD assays and molecular assay is possible using the autoanalyser estimated disease levels in air samples. Further development of the project will use multiplex tests within the system for several pathogens.

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The Role of Ultraviolet Light and Hydrogen Peroxide in Airborne Virus Deactivation: Insights from a 19 m^3 Atmospheric Chamber Study

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Abstract

Amid the COVID-19 pandemic's challenges, the development of effective airborne virus disinfection strategies has become a critical public health priority. This study assesses the efficacy of UVA and UVC ultraviolet light in conjunction with hydrogen peroxide (H_2O_2) for disinfecting aerosolized viruses within a controlled indoor environment. A 19 m^3 atmospheric simulation chamber was used to evaluate these disinfection methods using bacteriophage T4 in a pig mucin medium, closely mimicking the aerosolized particles produced during human respiration.

By constructing an aerosol generation system based on saturator/condenser mechanisms and utilizing pig mucin as an aerosol carrier, the original size of the aerosolized particle (120 nm) was increased to 550 nm, resulting in a complex structure consisting of the virus enveloped by a mucin solution, similar to exhale of aerosol during breathing.

The study investigated the capability of UVC to deactivate viruses at different doses. The study demonstrated that UVC light could damage 90%, 99.8%, and 99.9% of viruses at respective UVC doses of 7, 8.93, and 17.83 mJ/cm^2. Comparing these results with previous studies on mucin-enveloped viruses on respiratory filters revealed that higher UVC doses necessary for virus inactivation, which emphasizes the protective role of mucin against UVC. The effects of UV lights and H_2O_2 on the T4 bacteriophage were evaluated by measuring DNA copies using polymerase chain reaction (PCR).

The research also explored the effectiveness of UVA light in virus deactivation. Although UVA was found to be less efficient than UVC, it still played a contributory role in reducing viral loads, especially at higher exposure doses. Reduction of 80% detected virus DNA at a UVA dose of 135.5 J/cm^2 underlines the potential utility of UVA light as a supplementary method for air disinfection, particularly in scenarios where UVC application may be limited.

The investigation into hydrogen peroxide (H_2O_2) efficacy in virus deactivation yielded significant insights, especially when used in conjunction with ultraviolet light sources. The results showed that a concentration of 32 ppb resulted in 78% virus DNA destruction in dark conditions. When combined with UVA radiation at a dose of 135.5 J/cm^2, the addition of H_2O_2 elevated the virus destruction completely. Similarly, the presence of UVC light at a dose of 7 mJ/cm^2, in combination with H_2O_2, resulted in DNA destruction over 99.999%.

In conclusion, the integration of UVA or UVC irradiation with hydrogen peroxide in the gas phase represents a promising advancement in air disinfection technology. The study's innovative approach to simulating human respiratory emissions through aerosolized bacteriophage T4 and pig mucin medium sets a new standard for evaluating disinfection methods, with significant implications for public health and indoor air quality management.

Acknowledgment

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Evolution of the Ragweed Pollen Load in the North-west Milan Area

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Abstract

Background: Previous studies showed a reduction of ragweed pollen levels in the area North-Westof Milan up from 2013 due to the spread of *Ophraella communa* LeSage *(o. communa)*, an insect that preferably feeds on *A. artemisiifolia,* thus preventing pollen production. The aim of this study was to monitor the evolution of the ragweed pollen load in that area in parallel to the presence of *O. communa*, and to assess the possible impact on health of such variations.

Methods: Pollen grains were sampled by three Hirst volumetric traps located in Legnano (L), Magenta (M) and Rho (R) from 1995 to 2023. The seasonal pollen parameters were assessed. The areas infested by *o. Communa* were checked by visual inspection up from 2014. Clinical data were collected by three allergy clinics in the surroundings of the pollen monitoring stations, from 2005 to 2022. Pollen and clinical data trends were assessed by nonparametric means.

Results: Seasonal Pollen Integral (SPI) and peak value (C max) showed a significant down trend in all three monitoring stations ($p<0.05$), as did the number of days showing ≥ 100 p/m^3, ≥ 50 p/m^3 and 10-99 p/m^3($p<0.05$). Further, a significant drop in the number of days with 10-50 p/m3 ($p=0.019$) was observed in M and R. Nevertheless, since 2018 a fluctuating SPI, was observed in all stations, more evidently in L and M, and an increase of C max was also observed mainly in L.

A marked drop in the number of areas infested by *0. Communa* was detected, varying from 181 in 2014 to 28 in 2022 and 35 in 2023, in parallel to the general decrease of the ragweed infested areas. However, an increased ratio between areas infested by *O. Communa* and areas infested by ragweed was observed.

A significantly reduced rate of ragweed pollinosis ($p=0.002$) was observed in one allergy clinic located where ragweed infestation was less intense. The reduced rate previously observed in another allergy clinic was confirmed, albeit no longer statistically significant, although if a punctual increase of that rate occurred since 2018.

Conclusions: Over time, ragweed pollen load in the area of Milan confirms a reduction. A positive impact on health seemed to be confirmed only where the infestation was less intense. However, SPI and C max fluctuated since 2018 in coincidence with a decreased presence of *O. communa*. Nevertheless, a sporadic increase of the ragweed allergic subjects was detected, possibly reflecting an increase of pollen levels associated with the decrease of *O. communa* in some areas.

Keywords: Ragweed, *Ophraella communa*, Pollen exposure

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Indoor Air Quality of Public Closed Spaces: Beyond Co2 to Chemical and Microbial Pollutants

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Abstract

Indoor air contains a mixture of chemical and microbial compounds. Poor indoor air quality (IAQ)can affect the health of exposed people. The COVID-19 pandemic highlighted the need to prioritize design strategies to improve IAQ in post-pandemic times. To this end, the Belgian law of 6 November 2022 aiming at improving indoor air quality in enclosed public spaces went into

effect and an Indoor Air Quality Platform was created by the Federal Public Services Health, Food Chain Safety and Environment (FPS Health) .

To support the activities of FPS Health and of this Platform, Sciensano, the national public health institute of Belgium, plays a supportive role by offering scientific input and by conducting research on the indoor air quality of public spaces. One of the first tasks of Sciensano is doing a chemical and microbial characterization of indoor air quality in those public enclosed spaces.

Firstly, based on the available literature, an extensive list of pollutants was created, including more than 250 chemical and microbial pollutants. From this list, the most important pollutants were retained by national and international experts. Secondly, methods and sensors were selected to measure the shortlisted pollutants. Suppliers and manufacturers were contacted and sensors and (reference) instruments were purchased.

For the chemical characterization, these instruments are currently being tested in public spaces, starting with sports centers and cultural centers, as well indoors as outdoors. Long-term continuous measurements of chemical pollutants and microclimate parameters are being conducted using sensors indoors and outdoors. Later, MS-based methods will be used for a more in-depth analysis of the chemical content of these pollutants.

For the microbial characterization, a range of devices were purchased to sample and analyze indoor air quality, each employing distinct methods to gain a comprehensive understanding of the microbial composition of indoor air. Over the next few months, we will evaluate the accuracy and efficiency of these devices and determine the most suitable methods for microbial air quality assessment.

Besides different approaches, also different public locations will be tested during monitoring campaigns. Through thorough testing and analysis, we aspire to fine-tune our methodologies based on the insights gleaned from the results and to validate them in order to use them in future indoor air studies and by third parties.

The outcome of these monitoring campaigns will allow us to have a first view of the microbial and chemical pollutants present in public spaces and to investigate the influence of outdoor air on IAQ. In time, this knowledge could also contribute to the establishment of an "indoor barometer" as a quality indicator, but also to an inventory and (better) standards for the permissible concentration of biological and chemical pollutants in various public spaces as well as recommendations for improvements of the indoor air quality.

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Bacterial Viability and Air Quality: Experimental Approach and Results at the Atmospheric Simulation Chamber Chambre.

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Abstract

Bioaerosols consist of airborne particles of biological origin, including living and dead microorganisms such as bacteria, viruses, fungi, microalgae, and not viable particles such as pollen, spores, and animal dander. Their presence in the atmosphere influences the spread of diseases and allergies and, according to some studies, a high concentration of these microorganisms is related to the cloud formation process, atmospheric chemistry, and can influence the precipitation processes. For these reasons, the interest in bioaerosols is increasing rapidly to broaden the knowledge of their characterization. The experiments conducted in Atmospheric simulation chambers (ASCs) can provide valuable information on bioaerosol thanks to a confined artificial environment. ChAMBRe (Chamber for Aerosol Modelling and Bio-aerosol Research) is the first ASC in Europe designed for bioaerosol studies; it's managed by INFN at the Physics Department of the University of Genoa, Italy and the research is focused on the investigation of the airborne bacteria behaviour in different atmospheric conditions [2].

Our experiments were performed with *Escherichia coli*, *Bacillus subtilis* and *Pseudomonas fluorescens*: the experimental protocol includes bacteria cultivation, bacteria injection in the chamber, exposure to different gas pollutants, and verification of cultivability [1];[3]. The bacteria survival rate is evaluated in a first set of experiments (baseline experiments) in "clean air" conditions by comparing the bacteria total concentrations (measures by WIBS-NEO counter) with the Colonies Forming Units (CFU) collected on Petri dishes by Andersen impactor at certain time intervals. Then the experiments were repeated with the addition of polluting gases, in different concentrations, in the chamber to highlight the differences in bacterial viability.

In other experiments, bacteria were exposed to NO_2 and NO concentrations of 900 and 1200 ppb for both pollutants. The average lifetime in the baseline experiments was about 35 minutes for both bacterial strains. For NO, a concentration of 900 ppb does not affect the viability of either *E. coli* or *B. subtilis*, while a concentration of 1200 ppb reduces the lifetime to about 25 and 21 minutes, respectively. For NO_2, the same reduction in viability was measured for *B. subtilis* for both concentrations, while for *E. coli* a higher NO_2 concentration resulted in a greater reduction in viability.

Experiments with *P. fluorescens* are now underway; in the future experiments will be repeated with other bacterial strains, with other polluting gases and with the addition of impinger sampling to better understand the mechanisms of bacterial viability and cultivability in bioaerosol conditions.

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Comparison of Airborne Cupressaceae Quercus and Poaceae Pollen Between Tulsa and Cordoba

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Abstract

This study aims to investigate the relationship between meteorological parameters and airborne pollen concentrations for Cupressaceae, *Quercus,* and Poaceae in Tulsa (Oklahoma) and Cordoba (Spain). Understanding this relationship is crucial for predicting pollen exposure, which can significantly impact allergic individuals. This study provides insights for helping sufferers manage their symptoms better and reduce exposure during high pollen periods. This research highlights the importance of regional weather variation on pollen dispersal and the need for tailored allergy management strategies in different geographical locations.

The study presents a comprehensive analysis of pollen seasonality, its intensity, and the influence of weather conditions on airborne pollen concentration in two different biogeographical areas: Cordoba (Spain) and Tulsa (USA). The study investigated how meteorological parameters, like temperature, humidity, wind speed, and rainfall affected the pollen season for these pollen types from 2010 to 2014. The study delves into the main pollen season (MPS) of Cupressaceae, *Quercus*, and Poaceae, dividing the MPS into pre-peak and post-peak periods and employing statistical analyses to draw correlations between pollen concentration and weather conditions, and emphasizes the ability to forecast the pollen concentrations behavior according to meteorological parameters from the same day and from the previous day. The Cupressaceae MPS starts in Cordoba before that in Tulsa, while the start of the MPS for *Quercus* and Poaceae was similar at both sites. Higher pollen concentrations characterize the pre-peak period when compared with post-peak. In Tulsa, pollen concentrations from all pollen types were more affected by weather conditions on the same day. In contrast, in Cordoba, the weather conditions from the previous day had a stronger

influence on Cupressaceae and *Quercus* concentration, but for Poaceae pollen, the weather conditions from both the previous day and the same day had an equal impact. Nevertheless, the pollen concentrations for the next day could be forecasted considering the meteorological parameters predicted for the present day.

Results highlight the need for adopting prediction strategies that can meaningfully improve the quality of life for people affected by pollen-induced allergies. In addition, the differences observed between the two locations at the MPS and the different effects of weather conditions on pollen concentrations highlight the importance of regional approaches for predicting the behavior of pollen concentrations under predictive weather conditions.

This research builds upon findings reported in a recent study published in the Grana journal (Helfman-Hertzog et al. 2023, https://doi.org/10.1080/00173134.2023.2222123)

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The Effect of Preseason Precipitation and Temperature on Quercus and Cupressaceae Pollen Seasons

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Abstract

Meteorological conditions affect both the main pollen season (MPS) and the main pollen index (MPIn). The temperature and rainfall influence the MPS start and intensity, respectively.

The current study was undertaken for *Quercus* and Cupressaceae in Cordoba (Spain) and Tulsa (Oklahoma, USA): 1) to investigate the impact of rainfall before pollen release to predict MPS using a novel method for pollen analysis: the Dry Days Since the Last Rain (DDSLR); 2) to predict the main pollen season start (MPSS). The study employed two distinct approaches for temperature analysis. The first approach involved calculating chilling and forcing units using a specific threshold, with the forcing units determined through the Growing Degree Days (GDD°) method. The second approach focused on computing the Accumulative Chilling Temperature (ACT) across various levels and defining forcing temperatures in terms of GDD°.

The findings indicated that the MPSS for *Quercus* typically occurs from mid to late March at both sites. However, there were notable differences in the MPS of Cupressaceae between the sites in all examined parameters because different species are represented in the pollen type. In Cordoba, the Main Pollen Season End (MPSE) for both pollen types can be predicted using

accumulative rainfall data from late September and alternatively the DDSLR method. For Tulsa, the DDSLR method also effectively predicts the intensity of Cupressaceae pollen SPIn, but not the MPSS and not the MPSE.

The study established a chilling temperature threshold of 0°C for both *Quercus* and Cupressaceae pollen at both sites. For *Quercus*, the forcing temperature threshold varied between the sites, although the MPSS was the same, with Tulsa showing a lower average temperature from December to February compared to Cordoba. However, the forcing temperature threshold for Cupressaceae was consistent across both sites. The GDD° method yielded more accurate predictions for *Quercus* when chilling units started from January 1, rather than December 1, and from November 1 for Cupressaceae in Cordoba. In Tulsa, the calculation month did not impact accuracy. The ACT levels in Tulsa were higher than in Cordoba, and while the ACT method indicated similar forcing temperatures threshold for Cupressaceae in both locations, there was a notable 2°C difference for *Quercus*.

The DDSLR method exhibits potential for prediction of the MPSE and SPIn. For *Quercus* and Cupressaceae at both sites, the MPSS is more accurately forecast using the ACT method compared to the GDD° method. When considering the predictions' success alongside the duration of the forcing heat period, it emerges as the most effective for forecasting MPSS, particularly for *Quercus* in Tulsa and Cupressaceae in both locations.

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Real-time Observation of Fluorescent Biological Aerosol in the Finnish Sub-arctic

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Abstract

Primary Biological Aerosol Particles (PBAPs) are widely distributed in the atmosphere and can be transported thousands of kilometers. In addition to their fundamental role in human health, PBAPs are considered highly efficient ice nucleating particles (INPs). These are particles or macromolecules that, by their presence inside a supercooled cloud droplet, induce freezing at much higher sub-zero temperatures than without. Thus, affecting lifetime and radiative budget of clouds and consequently climate. PBAPs are particularly interesting in arctic and boreal regions. Since these areas contain less anthropogenic aerosol, the relative influence of PBAPs

on cloud formation may be enhanced. INPs, active at high sub-zero temperatures for example, have been linked to biological aerosol in a boreal forest [1] and in the Arctic [2].

This study investigates PBAPs at the Pallas supersite, a pristine site in Finnish Lapland 170 km north of the Arctic circle, at the edge of a boreal and sub-Arctic climate. We use a Wideband Integrating Bioaerosol Sensor (WIBS 5/NEO – Droplet Measurement Technologies) to measure the concentration and size distribution of fluorescent aerosol particles (FAPs) in near real-time. The WIBS detects and sizes particles from 0.5 to 30 µm and measures the autofluorescence of single particles using two excitation wavelengths (280 and 370 nm) and two emission bands (310 – 400 nm and 420 – 650 nm). In parallel, air samples have been collected on filters and will be quantified concerning INPs using the ice nucleation spectrometer of the KIT (INSEKT) [1]. Traditional Hirst trap measurements and pollen counts from a SwisensPoleno Jupiter online automatic cytometer [3] will be compared to the WIBS data to assign particle types to the FAPs. The high time resolution of the instrument enables us to link FAP concentrations to airmass origins and meteorology.

During the campaign (September to December 2022, April to September 2023), the average total HFAP concentration (Highly Fluorescent Aerosol Particles, considered to be mostly of biological origin [4]) is about 0.032 cm^{-3}, a contribution of 4 % to the total aerosol concentration measured with the WIBS. HFAP concentration undergoes a strong seasonal cycle and increases exponentially with ambient temperature (r: 0.81 for a subset of HFAPs), with a maximum in August. Snow coverage has a strong influence: The average HFAP concentration during snow coverage is reduced rapidly by 70 %. Once snow melt starts in May, HFAP concentrations increase again. Backward trajectories of airmasses (HySplit) suggest that most HFAPs are emitted from local vegetation. Comparing our results with HIRST data and INP concentration will give more insights into the contribution of biological particles to primary ice formation in the sub-Arctic.

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Evaluating Automatic Pollen Identification Algorithms Against the Mean of Four Hirst-type Traps: the Renewed Eumetnet Autopollen-cost Adopt Campaign App

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Abstract

Automatic pollen monitoring is currently being adapted in Europe and elsewhere. Due to the dynamic developments in the field of artificial intelligence (AI), which is used for identifying pollen, new algorithms are constantly under development. Each new algorithm needs to be evaluated. However, how this should be achieved is a challenge. Currently, most algorithms for automatic pollen identification are compared to data from Hirst-type traps even if it is known that these instruments suffer from large errors and high variability. Nevertheless, currently there is no other means to check if data from an automatic pollen monitor is correct.

One solution is to compare the data against the mean of a few Hirst-type pollen traps. This way the variability that exists between Hirst-type pollen traps is reduced, although systematic errors remain.

Few studies have placed four Hirst-type traps at the same location except for the EUMETNET AutoPollen – COST ADOPT campaign held over the 2021 pollen season in Munich, Germany.

In this campaign, four Hirst-type traps were situated on the same roof within 5 m of each other (1-3). Their clocks were adjusted and checked with a Witschi chronomaster (www.witschi.com, a mechanical clock controller), followed by manual test runs. The flow was adjusted to 10 l/min with a Burkard hand held rotameter, but corrected to the exact flow with an Easyfluxâ resistance free anemometer. Pollen per m3 were calculated using the anemometer flow, as described in (3).

Although not perfect, this is the best we have using the current methods (4).

This dataset is publicly available. This is not the same app as used for the publication of the AutoPollen campaign (1), but has a few optimisations making it suitable for checking new algorithms, also those that were not available at the time of the campaign. The raw data from the automatic monitors has been stored, and can be re-analyzed with any new algorithm, with that algorithm then added to the database and connected to the app. In the new version of the app, any algorithm can be selected in the “Scatter plot” view, allowing comparison not just of automatic vs manual observations, but also various automatic instruments against one another. In addition, the meta data of any point of the Scatter plot is displayed, so the original data can be easily consulted.

While this is no longer the same "blind" check of the algorithms as carried out for the intercomparison (i.e. the Hirst-type data are now public, during the campaign they were held closed to the algorithms would be tested without knowing what the "baseline" manual data showed, except for the scaling factor for Poleno), the app still allows an internal validation and assessment against previous versions of algorithms. An external validation using unknown manual observations is additionally suggested.

New algorithms can be submitted at any time. Please contact the app administrator (see the app: https://autopollen.shinyapps.io/APP_AUTOPOLLEN_compare/).

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Effects of Ash Dieback on Pollen Viability of Fraxinus Excelsior L. And Investigations on the Potential Effect of Long-range Transport on Pollen Viability

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Abstract

European ash trees are currently severely threatened by ash dieback, and it is expected that ash populations will be further fragmented. The viability of ash pollen plays a major role as long-distance pollen transport can only be successful if pollen remain viable under the prevailing environmental conditions until pollination.

In this study, we tested and compared four different methods for assessing pollen viability: TTC test (2,3,5-Triphenyltetrazoliumchlorid), Alexander's stain, Acetocarmine stain and pollen germination. We developed an optimized germination medium for assessing pollen germination using a multi-stage experimental setup and tested different concentrations of sucrose, boric acid, calcium nitrate and agar. Additionally, we tested three different storage temperatures (4 °C, -20 °C and -80 °C) in their ability to maintain pollen viability. In a subsequent step, experiments in a climate chamber were conducted, testing different possible environmental conditions and their effect on pollen viability. Pollen were exposed to three different climate scenarios over a period of four weeks and their viability was regularly evaluated. We assessed pollen viability of ash trees in ash seed plantations and forest stands and tested if trees affected by ash dieback in varying degrees are characterized by differences in pollen viability.

Our findings indicated that both, the TTC test and pollen germination, provided the most reliable results. Due to its fast application, the TTC test was chosen for subsequent investigations. Storage temperatures of -20 °C and -80 °C were found to be suitable; a temperature of 4 °C cannot be recommended as this temperature was linked to a fast decline of viable pollen. Our results demonstrated that temperature and UV radiation can have a high impact on the viability of pollen, with severe declines in viability as temperature increases. However, under average environmental conditions prevailing during the typical flowering period, viable pollen could still be detected after 28 days. The pollen viability assessment in the field revealed a slight tendency towards a decrease in viability in more severely damaged ash trees, however this trend was not significant.

Hence, the disease has no significant impact on pollen viability and thus on the fertilization capability of male ash pollen. We conclude that prevailing weather conditions during long-range transport can affect successful pollination. Thus, a further fragmentation of ash stands and the potential increase in spring temperatures in the context of climate change might be linked to an impoverishment of the gene pool.

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Indoor Air Fungi in a Changing World - Insights From a Twenty-year Study in Greek Environments

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Abstract

Fungal propagules are an omnipresent constituent of the atmosphere outdoors and indoors. Fungi affect both the biosphere and the abiotic elements of every environment, positively and negatively. The presence of fungi in enclosed environments, as influenced by a potential global change, is at the forefront of interest in recent years, because of the importance of fungi in human affairs.

The diversity, abundance and spatio-temporal variations of airborne fungi have been studied in Athens, Greece for over twenty years, in diverse indoor environments in comparison to fungi in the ambient air. Industries, schools, museums, libraries, art galleries and residences have been studied by a culture-dependent volumetric method on various nutrient media. For the genera *Aspergillus* and *Penicillium* morphological analysis was supplemented with molecular tools. The influence of the outdoor atmosphere and biotic and abiotic factors indoors were assessed.

The comparative analysis in all environments, revealed a core mycobiome comprising the prevalent genera *Penicillium*, *Cladosporium*, *Aspergillus* and *Alternaria* in descending order. Yeasts were also abundant indoors. The fungal spectrum also included about ten genera (*Aureobasidium*, *Botrytis, Fusarium*, *Mucor*, *Paecilomyces, Trichoderma*, etc) registered frequently in low concentrations and several genera (*Arthrinium*, *Beauveria*, *Chaetomium*, *Curvularia*, *Rhizopus, Stemphylium, Trichothecium*, etc) found rarely. Fungal concentration ranged significantly among the diverse built environments with values lower than 500 CFU m^{-3} in schools and museums, whereas numbers exceeding 1000 CFU m^{-3} registered mostly in residences. In the absence of indoor sources, the outdoor air was the major influence of indoor fungal aerosol. Human presence and activity exhibited an aggravating effect on the fungal air content. The implementation of preventive measures and practices indoors reduced considerably fungal concentration. The diversity and abundance of airborne fungi were very low in buildings with a green roof system. Temperature was a determining factor indoors and, it is the best predictor of fungal concentration outdoors based on statistical analysis. Because of climate change, the impact of temperature may be more crucial in the future, on the abundance and diversity of airborne fungi globally and in local scale.

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Airborne Algae Spectrum and Concentration of Inland Temperate Climate Region

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Abstract

Background: Airborne microalgae and cyanobacteria (hereafter referred to as airborne algae) are a permanent constituent of the atmospheric microbiome, especially in tropical but also temperate climates. Despite their relatively high concentrations in the air and the allergenicity of some genera being known for a long time, these components of airborne microbiome are often overlooked and underestimated due to the challenging identification.

Method: Our study site Bratislava (48°1492' N, 17° 0734' E) is located in the temperate zone in inland central Europe, but the sources (both aquatic and terrestrial) from which algae could be aerosolised are abundant. Using a gravimetric method with open plates at the same time as the Hirst-type volumetric sampler, we were able to cultivate airborne algae and use it as a reference for correct retrospective identification of those captured by the volumetric trap. In this way, we obtained reliable data on their presence in the air of the given area over three years (2018, 2020–

2021). The data were used for an analysis of their temporal variation and influence of environmental factors.

Results: The mean annual concentration recorded during the study period was 19,182 cells*day/m^3, with the greatest abundance in 2020. Unlike pollen grains, airborne algae were present in the samples throughout the year, with the highest abundance in February and April. However, the peak daily concentration reached the highest value on October 21, 2021 (1,011 cells/m^3). The mean daily concentrations ranged from 44 cells/m^3 in 2018 to 69 cells/m^3 in 2020. From gravimetric samples, we identified ten genera of airborne algae belonging to two taxonomic groups: Chlorophyta (7) and Cyanobacteria (3). Six of them, namely *Bracteacoccus* sp., *Chlorella* sp., *Desmococcus* sp., *Stichococcus* sp., *Klebsormidium* sp. and *Geminella* sp. represented nearly all identified airborne algae from volumetric samples. The most abundant genus was *Bracteacoccus* (57%) and the least abundant was *Geminela* (<2%). The analysis of intra-diurnal periodicity was performed only for the six abundantly occurring genera. A relatively constant prevalence of airborne algae was observed throughout the day, with a peak between 2 and 4 p.m. for most genera. However, notable deviations from this trend were found in the cases of *Klebsormidium* and *Geminella*, which peaked at 8 a.m. and 8 p.m. respectively. The environmental conditions differed in all years of our study, with 2020 as the rainiest and 2018 as the warmest and most polluted. Spearman's correlation coefficients showed the wind speed and relative humidity as the most influential. The wind speed was a significantly positive factor for all the taxa except *Klebsormidium*, while relative humidity was significantly negative for all taxa, but *Geminella*.

Conclusion: The prevailing genus *Bracteacoccus* is a known causative agent responsible for respiratory allergies, alongside other prevalent genera like *Stichococcus* and *Chlorella*. Considering that airborne algae in Bratislava have reached values compared to some genera of pollen grains, it can be inferred that these microorganisms have the potential to pose a threat to human health.

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Investigation of the Ability of Automated Fluorometry and Holography Sensors to Discern Airborne Pollen of Pooideae, Panicoideae, and Chloridoideae Grasses.

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Abstract

Poaceae (grass) pollen is the most common outdoor allergen source globally, inducing allergic rhinitis in susceptible people throughout the spring and summer seasons. In Australia, the prevalence of allergic rhinitis (19%) and asthma (11%) is considerable creating a major health and economic burden. Chloridoideae and Panicoideae grass species that occupy subtropical regions have distinct ecological and phylogenetic variations compared to the temperate Pooideae grass species, and differ in pollen morphology, allergen composition and immune recognition. However, with traditional microscopy techniques, grass pollen cannot be distinguished beyond family level. We hypothesize that automated sensors can discern between grass subfamilies or/and genera using holography and/or fluorometry techniques. Thus, this project is a pioneering test of automatic holography and fluorometry in a subtropical region of the Southern Hemisphere. We have employed the SwisensAtomizer and SwisensPoleno Jupiter to aerosolize and test five dried grass pollen samples (Stallergenes Greer, Lenoir USA) from three subfamilies; *Paspalum notatum* (Bahia) and *Sorghum halepense* (Johnson) from Panicoideae, *C. dactylon* (Bermuda) from Chloridoideae, *and Lolium perenne* (Ryegrass) and *Phleum pratense* (Timothy) from Pooideae. In addition, fresh *P. notatum* pollen was collected in vicinity of our main pollen monitoring site at Rocklea, Brisbane; Lat -27.54, Lon 153.00, Queensland, Australia) and compared with dry *P. notatum* pollen. Datasets were examined for differences in distributions of holographic features (e.g. equivalent area, axis lengths, solidity, eccentricity) and fluorescence spectra (e.g. relative intensity across five channels measuring emissions from three excitation wavelengths) of different grass pollens by applying a non-parametric Kruskal-Wallis test. In three repeated measurement campaigns of the same dry *L. perenne* pollen sample, distributions of equivalent area, eccentricity, solidity, and axes length appeared to be similar, and relative fluorescence intensity values also appeared to be consistent, which indicated the reliability and reproducibility of the automated sensor in the field. We conducted measurement campaigns with five grass pollen types, and noted that all these five grasses, even from same subfamily, showed differences in equivalent area maximums, major and minor axis lengths, and to some extent eccentricity and solidity. At an excitation of 280 nm and emission channel at 435 nm, there was a significant difference detected between each of the dry grass pollen samples from three subfamilies. Comparison of dried and freshly collected local *P. notatum* pollen revealed that the equivalent area, sphericity, as well as the major and minor axis lengths of fresh pollen appeared larger than dried pollen. There were also noticeable differences in fluorometry parameters between dried and freshly collected local *P. notatum* pollen. This is the first in field aerobiological application of automated holography and fluorometry in the Southern Hemisphere, and the first study to show that automated sensors can distinguish between airborne pollen to subfamily and/or genus level for common subtropical and temperate grasses. This study shows potential for identifying and monitoring

different and locally relevant grass pollens in near real time to help patients allergic to pollen of separate grass families to manage allergy symptoms more effectively.

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Large Regional Variation in Airborne Alternaria Spores

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Abstract

Background: Alternaria spores are routinely measured in many countries due to their allergenic potential. In Denmark, spores are measured in the capital Copenhagen, however, there could be high regional differences across the country, and measurements from neighboring countries may even represent some regions better than the national monitoring site. Especially as the most important source areas for Alternaria, grasslands and crops, are unevenly distributed across the country, and differences in distances to such areas, may lead to great local variation.

Methods: Alternaria spores were collected with Burkard Volumetric Hirst type spore traps from a total of seven sites in Denmark (Copenhagen, Viborg, Odense, Aarhus, Aalborg, Esbjerg) and Northern Germany (Borstel). Except Borstel, these are all urban settings. Placement and collections follow the standard recommendations in aerobiology. Spores were counted from June 1st to September 1st, 2020, covering the main Alternaria spore season in Denmark and Northern Germany.

Daily and 2-hourly concentrations of Alternaria spores were calculated. Correlations between sites and test for significance were computed in R-studio for all data and for days with concentrations above 100 spores/m^3. Season duration were calculated using the 95%-method.

Results: Variation in season duration ranged from 46 to 61 days, and season start was found to occur 17 days later in Aalborg than in Copenhagen. The spore integral varied from 10.506 – 26.547 and peak 2-hour concentrations from 738 to 3320 at the 7 sites. The time of peak was found to occur from the 5^{th} to 11^{th} of August. Copenhagen was found to have the lowest peak, lowest pollen integral and longest season.

The diurnal pattern for all stations peaked around 18:00 and had minimum around 06:00, except for Esbjerg with no clear peak. Esbjerg deviate from the other sites, as it is a coastal location, about 2-5 km from the sea from SW to NW, with predominantly western winds. Borstel had the strongest diurnal pattern.

All stations were significantly correlated for daily counts (R^2 0.38-0.94). For 2-hour values all but Esbjerg with Borstel and Viborg were significantly correlated (0.57-0.91). For the days with spore counts above 100 spores/m^3, all combinations of sites were significantly correlated, except Esbjerg with Copenhagen, Viborg and Borstel (0.29-0.93). For the daily counts however, only two site combinations showed statistically significant correlations, Aalborg-Copenhagen (0.72) and Bortel-Viborg (0.90).

Discussion and conclusions: Borstel has the most pronounced diurnal profile, and Esbjerg the least. This indicates strong local sources near Borstel, and the absence of nearby sources for Esbjerg.

The environmental variables in the region appear to impact seasonal and diurnal patterns synchronously causing an autocorrelation between the data. This is reflected in correlations, and most prominent for the 2-hourly data. However, when restricting data to moderate to high concentrations, little correlation is observed between sites. Consequently, one station is insufficient to estimate high exposure periods of spores. No clear pattern was found in correlation and distance between the sites. This suggest that additional stations is needed to detect the variation in exposure within a rural-urban region like Denmark and Northern Germany.

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Evolution of the Urban Forests of the City of Granada Through the Analysis of Pollen Emissions Throughout a Climatological Series

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Abstract

Changing climate conditions are having a significant impact both on the adaptive response of species growing in urban forests (UF) and on their evolutionary dynamics. The pollen emissions carried out by plants during their reproductive period have proven to be a precise indicator of the response that the different species of urban trees to increasingly frequent and extreme climatic events, exacerbated by urban microclimate conditions. This study analyzes the pollen dynamics of the most common anemophilous tree species in the urban forest of the city of Granada, south-eastern Spain, over a climatological series (30 years), so that resilience and its potential response to water stress can be identified. The urban forest of Granada is made up of about 45,000 specimens, the most abundant species being *Platanus* x *hispanica* (>5000 specimens), *Ulmus* spp (2879 specimens), *Acer* spp. (2541 specimens), *Cupressus* spp (2474 specimens), *Citrus aurantium* (2305 specimens), *Populus* spp. (2069 specimens), *Phoenix* spp. (1152 specimens), *Ligustrum* spp. (700 specimens) and *Quercus* spp. and *Pinus* spp. with about 500 specimens each. Pollen data have been collected using a Hirst-type volumetric sampler located at the Faculty of Sciences of the University of Granada over the last 32 years. The Annual Pollen Integral (APIn) of the total pollen of the city of Granada is 61,352 pollen*day/m3, with values that range from 31,283 of year 1999 to 107,145 of year 2022. Regarding the trends followed by the APIn of the main types of pollen of anemophilous species, they are significant and positive for *Cupressus, Pinus, Platanus* and *Quercus*, all of them of Mediterranean origin. Although some of them are important sources of emisión of allergenic bioaerosol, they have continued to be used as preferred in the new green spaces built in the city to accommodate the increase in population and urban growth. However, pollen types of tree species with a higher dependence on water resources such as *Ulmus, Acer, Populus* and *Salix* showed a decreasing APIn trend. All of these species have a greater dependence on increasingly scarce water resources in the form of natural precipitation and a greater vulnerability to prolonged periods of drought and increasingly high winter temperatures. It has also been possible to detect how new species have been incorporated into the city's urban forest probably to replace those that are having greater vulnerability to other effects of climate change such as the increase in pests and diseases. These results highlight the urgency of rethinking the species hither to selected as preferential in urban forests in the Mediterranean region, due to the accelerated decline and premature species death that some of these are suffering as a consequence of their mesophilic character. The expansion of the catalog of species that must first accompany and then replace the native species that still endure the increasingly extreme environmental conditions of the Mediterranean region must be planned.

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Effects of Climate Change on the Onset of Pollination in Barcelona (NE Spain)

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Abstract

Background

The effects of global warming are numerous and recent studies reveal that temperature is the meteorological variable that presents a clearer relationship with the start of the pollination season of most of the observed airborne pollen taxa.

Barcelona recorded an increase of 0.17ºC/decade in the period 1914-2021. It is therefore expected that in the future these increases will influence the pollination timing and specifically the start date.

Study's purpose

To predict the start pollination date for the taxa: *Olea*, *Pinus*, *Pistacia*, *Plantago*, *Platanus* and Deciduous *Quercus* over the 21st Century in Barcelona following the projected future temperatures estimated by the European RESCCUE (RESilience to cope with Climate Change in Urban arEas) project.

Methods used

Airborne pollen data in Barcelona (41º23'37.42'' N, 2º09' 53.72'' E, 93 m a.s.l.) for the period 2000-2019 obtained with the standardized European methods for the six pollen taxa studied and the corresponding daily mean temperature provided by the Servei Meteorològic de Catalunya and recorded in the Fabra Observatory (41º25'N, 2º07'E, 415 m a.s.l.) were used to establish the Forecasted Start Main Pollination Season (SPSF). To this end, the START forecasting model was used. The model uses statistical methods based on the cumulative sum of average daily temperatures for a set of test dates and above a set of thermal thresholds. The START model was applied to the forecasted daily mean temperature at the Fabra Observatory obtained from the RESCCUE project for the global climate models BCC-CSM1.1, CNRM-CM5, MIROC-ESM-CHEM, MRI-CGCM3 and NorESM1 and the scenarios RCP2.6, RCP4.5, RCP6.0 and RCP8.5 to calculate the SPSF.

Results obtained

The initial parameters obtained to forecast the SPSF differed from one taxon to another. *Olea* was the taxon with the highest sum of temperatures (1350°C), while *Platanus* the lowest (505°C). *Platanus* was the taxon that required a later beginning date (January 21) while for *Olea*, *Pinus*, *Plantago* and Deciduous *Quercus* the counting day was 1st January. *Pistacia* and *Plantago* showed the highest temperature threshold (5°C), *Pinus* the median (2°C) and *Olea*, *Plantago* and Deciduous *Quercus* did not show a threshold (0°C).

Focusing on 20 years of observed flowering start dates (2000-2019), we have obtained an already existing advance for *Pinus*, *Pistacia* and *Platanus* and a delay in *Olea*, *Plantago* and Deciduous *Quercus*. The START model applied to the period 2006-2100 has shown negative trends (72% significant) for the six taxa and all climatic models and scenarios, indicating an advancement in the dates of the start of pollination. In the most emissive scenario, RCP8.5, advances in flowering at the end of the century ranged, on average for the six pollen types, between 15 days (BCC-CSM1.1) and 27 days (MIROC-ESM), while for the stabilization scenario, RCP4.5, the advances ranged between 7 days (CNRM-CM5) and 12 days (MIROC-ESM). All taxa were sensitive to these increases in temperature, the most being *Olea*, and the least *Platanus*.

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Aerobiological Study of the Concentrations of Cupressaceae Pollen and Cup a 1 Allergen in Bellaterra (Barcelona)

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Abstract

Background

The allergenic importance of the Cupressaceae pollen in the Mediterranean area has been demonstrated in recent decades by the significant increase in allergic manifestations during winter. Main species contributing to Cupressaceae pollen type are *Cupressus sempervirens* and *C. arizonica*, together with several ornamental species and, in the natural landscapes, *Juniperus* species. Cup a 1 is the major allergen in these species.

Study's purpose

The behavior of airborne Cupressaceae pollen is well known in Catalonia, due to a long tradition (beginning 1983) of measuring it using palynological methods. But, with the aim of helping to better assess the potential risk of this allergy, the Respiratory Allergy Committee (CAR) of the Catalan Society of Allergy and Clinical Immunology (SCAIC), carried out a comparative study of the concentrations of the Cup a 1 allergen and Cupressaceae pollen in the atmosphere of Bellaterra (Barcelona).

Methods used

The samples were collected throughout the years 2020 to 2022 using a High-Volume Sampler (CAV-A /mb, MCV S.A.) to obtain Cup a 1 samples every two days, and a Hirst collector (to obtain the daily pollen samples), both located on the roof of the C building at the Universitat Autònoma de Barcelona in Bellaterra (41º30'20" N, 02º06'28" E, 245 m.a.s.l. and 23 m.a.g.l). Allergen analyses were carried out using the ELISA technique and pollen analyses following the methodology of the Spanish Network of Aerobiology (REA). Spearman rank test has been applied to calculate correlations.

Results obtained

As expected, Cupressaceae pollen was present in the atmosphere mainly from January to April and, with much lower concentrations, in autumn. In contrast, Cup a 1 allergen was detected from January to June and in autumn, showing a dynamic more similar to the taxon Total Pollen than to Cupressaceae pollen. It has been observed that the year with higher pollen concentrations is not the year with higher allergen concentration.

Spearman test showed a positive and significant correlation between Cup a 1 and Cupressaceae pollen (0.244 in 2020 and 0.412 in 2021 both with $p < 0.01$ and 0.194 in 2022 $p < 0.05$) and with the Total Pollen (0.535, 0.232 and 0.646 $p < 0.01$, respectively).

This study reveals that there are cross-reactions between the Cup a 1 allergen and other pollen types than Cupressaceae, therefore, the allergic symptomatology can be extended to periods in which Cupressaceae pollen is not detected in the air.

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Betula and Pinus Pollen: Phenolic Compounds Content and Composition

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Abstract

Pollen dispersed during plant flowering contains phenolic compounds that are essential for cell development. Knowledge of these compounds depends on the research objectives and methods. We aimed to determine the suitability of methods for extracting phenolic compounds from *Betula* and *Pinus* pollen. To investigate the total phenolic and flavonoid content, bioactivity, and composition of individual phenolic compounds in pollen extracts.

Catkins of *Betula* (16 samples) and mature strobiles of *Pinus* (20 samples) were collected in Lithuania in 2022. Pollen was dried and sieved. Different methods were used to extract free phenolic compounds (free PC) from *Betula* and *Pinus* pollen: treated with liquid nitrogen before extraction (1) and a variant without nitrogen (2); ground and poured with 70% methanol (ratio 1: 10) and incubated in an ultrasonic shaker (3) with different shaking times (10, 20 and 30 min, at 40±1 ˚C temperature); shaken in an orbital device (4) with constant duration (300 rpm, 16 h, 18±2 ˚C). Alkali hydrolysis was used to prepare cell wall-bound phenolic extracts (bound PC) pollen extracts. Two extracts each of *Betula* and *Pinus* were prepared by treating the pollen by enzymatic hydrolysis and maceration methods. The spectrophotometric method was used for the analysis of total phenolics, flavonoid content and antioxidant activity (according to DPPH and ABTS radicals). High-performance thin-layer and liquid chromatography were used for the analysis of individual phenolic compounds in pollen extracts.

According to the results of the different extractions, it is sufficient to isolate phenolic compounds from *Betula* pollen by the ultrasonic shaking method, shaking the samples for 10 min at 40±1 ˚C with a shaker power of 113 W. For *Pinus* pollen, orbital shaking resulted in better extraction. Other extraction methods did not increase the content of phenolic compounds in the extracts. All extraction methods confirmed that *Betula* pollen released 2.5-5.0 times more bound PC and free PC into the methanol-water environment than *Pinus* pollen. *Betula* pollen extracts contained 20% more (~40 mg/g) free PC than bound PC, whereas in *Pinus* pollen it was the same (~7 mg/g). *Betula* pollen extracts showed a high biological activity which positively correlated with the total flavonoid content (bound PC $r = 0.95$, $p < 0.01$; free PC $r = 0.62$, $p < 0.05$). One of the most dominant phenolic acids in both plants' pollen extracts was trans-ferulic acid. Bound PC extracts of *Betula* pollen accounted for 70-94% phenolic acids, with a strong signal of bioactive chlorogenic acid. Free PC extracts contained bioactive rutin, which accounted for 2-3% of the total *Betulla* pollen individual phenolic compounds. *Pinus* pollen extracts show weak signals of individual phenolic compounds. Ferulic acid accounted for 57% and gallic acid was identified as 13%. Knowledge of

the composition and bioactivity of phenolic compounds in the pollen of most pollinating plants can be useful for modelling ecosystem food chains.

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Interdisciplinary Approach to Research on the Optical and Microphysical Properties of Allergenic Pollen

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Abstract

The objective of the study was to characterize pollen particles through examination of pollen type and abundance near the surface and within the atmospheric boundary layer using a unique combination of remote (Mie-Raman polarization lidar) and in-situ (Hirst-type spore trap) measurements. We investigate the spread of pollen within the boundary layer and identify conditions that facilitate the dispersion.

Lidar observations at the Warsaw National Facility of Aerosol, Clouds, and Trace Gases Research Infrastructure (ACTRIS ERIC) were evaluated. Manual analysis of pollen type and concentration at the ground level on a daily temporal resolution was done via microscopic examination. In-situ measurement data gave us a unique ability for verification of the pollination events identification. Our methodology is based on bell-like shape pollination signatures in lidar-derived volume depolarization ratio profiles backed up by favorable meteorological conditions.

A comparison of lidar characteristics and in-situ measurements was conducted using in-situ daily pollen concentration and a determined set of conditions that match the boundaries of the typical pollination case, including relative humidity, temperature, wind speed, maximum of boundary layer top, and pollination time.

A combination of in-situ and remote detection methods allowed the characterization of pollen type and its vertical extent. In the future, we will assess to what extent the synergy of this combined approach can help in faster pollination detection and if it can be used for monitoring/prediction purposes.

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The Composition and Sources of Airborne Bacteria and Proteinaceous Ice Nucleating Particles in the High Arctic Marine Region During Spring

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Abstract

The Arctic is a particularly vulnerable region on Earth, where climate change takes place at an intense pace. Clouds represent an essential element within the Arctic atmosphere and play a crucial role in the regional radiative balance. The physical properties of clouds are tightly interlinked with the presence of aerosols that can serve as cloud condensation nuclei (CCN) and as ice nucleating particles (INPs), which facilitate the formation of cloud droplets and ice crystals, respectively. Consequently, they affect cloud thickness, lifetime, and albedo.

More studies propose that various biological aerosols e.g., aerosolized microbial cells, proteinaceous compounds and fragments actively contribute to cloud processes serving as INPs active at high subzero temperatures (>-15˚C). However, our understanding of microorganisms responsible for producing compounds serving as INPs, their source environments, and their level of activity, remains highly uncertain.

Given the profound impact of climate change in the Arctic region, understanding the role of biological INPs in the atmosphere becomes particularly critical during Arctic melt season. Here, we present an overview of bioaerosol observations and sources tracking from the recent Arctic expedition "Atmospheric rivers and the onset of Arctic melt" (ARTofMELT 2023).

Biological INPs are thought to originate from the ocean and meltwater sources during the Arctic Spring and Summer. To assess the potential contribution of these sources to INP active aerosols, aerosols were generated from bulk seawater and sea ice melt water with a temperature-controlled sea spray simulation chamber. The presence of microorganisms in the bulk water and aerosol was quantified using flow cytometry and qPCR while the composition of the microbial communities was determined by amplicon sequencing. Additionally, fluorescent bioaerosols generated by the chamber were analyzed using a Multiparameter Bioaerosol Spectrometer (MBS). Simultaneously, ambient air samples were analyzed for the presence of microbial cells, bioaerosols, and the composition of the collected microbial community. The ice nucleating properties of water, sea ice melt, and aerosols from the chamber and ambient aerosol were also measured to determine their relevance for Arctic cloud formation.

Preliminary results from the ambient measurements revealed low concentrations of airborne bacterial cells and highly active INPs. From the sea spray simulations, we found that ice melt,

snow melt and seawater samples generated a high flux of bacterial cells which were accompanied by INPs active predominantly at low freezing temperatures (<-15˚C). Therefore, it seems that the local sea spray is not a likely source of proteinaceous INPs detected in the Arctic spring atmosphere, which will be further explored through bacterial community analysis. Our results will thus provide comprehensive insights into the contribution of local and long-range transported sources of bioaerosols to the Arctic.

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Evaluation of the Decay Rate of Airborne Viruses in an Exposure Chamber During Air Treatment by a Uvc-based Technology

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Abstract

The COVID-19 pandemic has highlighted the importance of airborne transmission of respiratory viruses, and proper control of indoor air quality has become essential. Several new mitigation devices have been developed for this purpose, and evaluation of their performance is critical. Standardized testing of the performance of microbial air purifiers was limited prior to the pandemic, and agencies responsible for standardization protocols have sought to provide new guidelines. However, standard evaluation methods are not uniform. They are based on cultivable bacteria (ASTM E3273-21), bacteriophages (AHAM-AC-5-2022), or bacteria, fungi, and viruses (ASHRAE Standard 185.3P). They also differ in their methods for quantifying treatment efficiency, some using $\log_{10}$ reduction, others percentage reduction, or clean air rate. However, all test protocols agree on the need to evaluate using a well-controlled exposure chamber.

In this study, the decay rates of an airborne infectious virus were determined under the influence of a UVC-based treatment device using a 17 m^3 exposure chamber. MS2 bacteriophages were aerosolized at a concentration ranging between 10^8 and 10^9 PFU/m^3 of air using a 24-jet Collison nebulizer. Bacteriophage sampling was performed on a 25 mm polycarbonate membrane at a flow rate of 2 L/min for a duration of 5 min. Once the aerosol generation was completed, 4 samples were obtained at times (0,15,30,45 minutes) with and without UVC treatment. The bacteriophage concentration was determined according to the lysis plate counting method and the decay rate was subsequently calculated. The natural reduction curves (decay rate without

UVC) allow to precisely measure the intrinsic efficiency of the UVC treatment. Similarly, three to five positions were sampled, depending on the test, and the results were used to evaluate the spatial variability of the air cleaners.

Due to the high MS2 concentrations inside the chamber, monitoring the decay rate was possible for up to 60 minutes with no detection limitation. Results demonstrated that the generation protocol led to a high degree of spatial uniformity of concentrations in the exposure chamber with a standard deviation obtained from the logarithmic data of 0.17. High reproducibility between experiments was also observed. Interestingly, a linear correlation was observed between the volumetric flowrate of the tested devices and their equivalent air change per hour. In certain conditions, the UVC-based device achieved 99.9999% (6 log) reduction in MS2 concentration per hour of treatment. This study not only demonstrated the value of using exposure chambers to evaluate the performance of mitigation tools, but also underlined the added benefits of kinetic analysis protocols. This performance assessment method was optimized to allow the evaluation of both the spatial and temporal performances of air cleaning devices. However, such an approach requires multiple sampling, which becomes challenging when different types of microorganisms (bacteria, fungi, viruses) are required to be tested. Organizations setting standards for the evaluation of these devices should strive for uniformity, consider the cost and workload associated with their protocols, and may tailor their requirements to the specific microbial risk expected in the indoor environment being cleaned.

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Optimizing the Numerical Pollen Forecast Models COSMO-ART and ICON-ART Through Integration of Real-Time Pollen Observations: Insights and Lessons Learned

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Abstract

Numerical pollen forecast models play a key role in providing pollen information to stakeholders such as allergy sufferers. However, uncertainties inherent in such model output remain a critical issue, potentially reducing the value of pollen forecasts based on model output.

In the past years technologies for monitoring pollen concentrations in real-time made substantial improvements. As a consequence, such data became increasingly available. This opened new opportunities for improving numerical pollen forecast models by integrating real-time pollen observations in model runs. This may reduce the gap between modelled and observed pollen concentrations.

We present a method to integrate real-time pollen observations into the pollen forecast models COSMO-ART and ICON-ART. The basic idea is to adapt the phenological parameterizations and the overall tuning factor. We tested this approach by a number of model runs. The model domain covers the greater Alpine Area in Central Europe using a mesh size of 1.1 km. This high resolution ensures adequate representation of the complex Alpine terrain. Modeled species include Corylus, Alnus, Betula and Poaceae. Verification was performed against daily and 3-hourly observations from 13 Swiss pollen stations. The analyses consider both traditional Hirst-type measurements and data from the newly established real-time pollen monitoring network in Switzerland.

Using this model configuration we demonstrate that substantial improvements in pollen forecasts can be achieved by fine-tuning model parameters based on pollen observations. However, the effect strongly depends on the situation and species. While for Poaceae the improvement was large, the high-frequency fluctuations of pollen emission of the other species partially remain a challenge.

In addition, we show that numerical pollen modeling can profit from advances in numerical weather prediction. ICON-ART generally outperforms the older model COSMO-ART. This may be due to more accurate modeling of relevant parameters such as the wind speed.

To conclude, the implemented method for real-time use of pollen observations shows substantial improvements of the pollen forecasts and represents a milestone in numerical pollen modeling. However, the method can be further improved both conceptually and in terms of the chosen parameterization. This will remain a topic in the coming years.

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Unmanaged Grasslands Are a Reservoir of Alternaria and Other Important Fungal Species With Differing Emission Patterns

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Abstract

Background & Aim

Alternaria is a ubiquitous fungal genus that is allergenic to humans and pathogenic to plants and animals. Grasslands are known habitat for *Alternaria.* We hypothesise that grasslands harbour a diversity of fungal species whose spores have varying emission patterns. The purpose of this study was to examine the potential of grasslands for emission, pattern, diversity and composition of *Alternaria* and other fungal species.

Methods

To test the hypothesis, we used Hirst-type and multi-vial cyclone samplers to collect air samples from two grassland sites (unmanaged and managed) and a non-grassland site at Lakeside campus of the University of Worcester, Worcester, United Kingdom. One grassland site is a circular plot (unmanaged) that was originally planted and had been left uncut for three years. The second grassland site (managed) is a site along roadside grass area that is cut once every year, typically after most grasses have flowered. Sampling was done for the period May to September 2019. We used optical microscopy and Illumina MiSeq sequencing to investigate the concentration, diversity and composition of the fungal spores from each site alongside meteorological variables. Kruskal-Wallis rank sum test with post-hoc pairwise and Wilcoxon tests with Bonferroni correction were used to examine differences in the bi-hourly *Alternaria* concentrations between the sites. Shannon's Diversity Index (α-diversity) was used to examine the diversity of *Alternaria* and other fungal spores between the unmanaged and non-grassland sites.

Results

The results showed that the unmanaged (61 days) and managed (62 days) grassland sites were a strong source of *Alternaria* spores with considerably higher number of clinically important days compared with the non-grassland site (47 days). Moreover, the managed grassland site varied in *Alternaria* spore emission pattern from the unmanaged site, probably due to the environmental variables associated with the local ecosystem and the cutting frequency. Furthermore, both the unmanaged grassland and non-grassland sites showed a high diversity of *Alternaria* and other fungi such as *Cladosporium*, *Ascochyta*, *Botrytis* and *Aureobasidium*.

Conclusion

Overall, the study shows that unmanaged grasslands are a strong source of fungal spores with allergenic and pathogenic potential and have varying emission patterns, compared with nearby monitoring stations normally found in urban zones. Allergy patients that are sensitized towards fungal spores are advised to take precautions when they are near unmanaged/uncut grass areas as spore concentrations near these sources can be much higher than reported by urban based monitoring networks. This information is useful for atmospheric modelling of airborne fungal spore sources and has implications for allergy sufferers and those with frequent exposure, such as farmers.

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Infection Prediction Modeling of Vineyard's Fungal Pathogens Integrating Weather Data With Ai-driven Spore Detection Systems

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Abstract

Grapevine fungal diseases constitute a major cause of yield loss in European viticulture. In Switzerland, the main causes of concerns are currently represented by two pathogens: downy and powdery mildews. In order to prevent severe infection outbursts of great economic and agricultural relevance, it is important to promptly protect the vineyard with repeated fungicide applications. The planning of such treatments is increasingly being coordinated through decision support platforms which often make use of specific infection prediction algorithms relying on meteorological data and parameters associated with the evolution of the pathogen's life cycle. Here we present two highly modulable and scalable bioinformatics pipelines and consequent field-validated analyses of the infection cycle stage predictions with 1) a model for the oomycete *Plasmopara viticola* (PV, causal agent of grapevine downy mildew), and 2) a model for the ascomycete *Erysiphe necator* (EN, causal agent of powdery mildew). Agroscope's Vitimiteo-Plasmopara (downy mildew) and Oidium (powdery mildew) prediction models, currently deployed and accessible by Swiss wine-growers trough the Agrometeo online platform, are rethought and redeveloped for research purposes while being upgraded with the integration of real-time spore detection data from SMALA (SMart Agriculture using Lasers and Artificial intelligence). These operate via AI-driven classification of in-field spore holographic imaging. While Agrometeo's models make use of meteorological data including air temperature, rainfall, relative humidity, and leaf wetness, the SMALA systems capture and

analyse holographic images of the airborne fungal spores available within the vineyard. Thus adding a further dimension for a more precise infection prediction modeling. Both infection prediction models have been redeveloped from scratch and allow for a greater customizability of epidemiological parameters and handling of different types of input data, as well as providing an increased code modularity facilitating the addition and modification of the pathogen's life cycle algorithms. Such bioinformatic rework enables large-scale parametric searches and parallel-run simulations. This allows for the optimization of the input epidemiological parameters and life cycle stages, in adaptation to any vineyard environment and study-case application, or even to different agricultural pathosystems. The results of the models' simulations, parameter optimization, and field-validation across a number of Swiss vineyard plots are presented here in comparison with real-time SMALA spore detection data and field observations of the epidemiological development of past and current grapevine growing seasons. Exploring and exploiting such data integration possibilities will hopefully help towards building the necessary technological tools and real-life applications for farmers and advisory services with a desire for a more precise management of plant disease and more sustainable agricultural practices.

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Intersecting Skies: The Development of an Insect Detection Algorithm Using a Snowfall Camera in Hyytiälä

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Abstract

To study trends in insect decline, accurate monitoring of insect populations is crucial. Current monitoring techniques for Pterygota with traps are largely labour intensive and destructive. Here, we show the potential of using a non-invasive camera system combined with computer vision image processing for non-destructive insect monitoring. For this, we use the open source Video In Situ Snowfall Sensor (VISSS), which is an instrument developed for characterizing particle shape and size in snowfall but incidentally also captures insect observations. The VISSS is composed of two cameras with approximately 60 μm resolution which center on a joint observation volume of approximately 6 cm^3, with telecentric lenses that enable accurate measurements of size. The cameras record shadow images of particles within the observation volume illuminated by green LED backlights at a frame rate of 140Hz. Using data recorded in 2022 when the instrument was situated in Hyytiälä, Finland, we developed a proof-of-concept insect identification framework. The algorithm splits images between hydrometeors and possible insects. This is further refined, through masking of the insect, categorizing the relative

size and flight path information. We show how this can be used to monitor Pterygota for daily and seasonal cycles.

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Forecasting Birch, Grass and Hazel Airborne Pollen Concentrations With Machine Learning

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Abstract

High concentrations of airborne pollen cause seasonal allergies and more serious adverse respiratory health events. With the changing climate, the abundance of airborne pollen increases, affecting allergic people. The first method of allergy management is allergen avoidance, which is typically achieved by predicting daily pollen concentrations. Timely alerts become possible through accurate forecasts for the next days, providing significant public benefit. While accurate forecasts are readily available for pollutants like particulate matter, a comparable level of reliability in pollen forecasts is still a challenge.

In this study, our goal was to develop a tool based on machine learning methods for short-term forecasting pollen concentrations of the most allergenic taxa in Central Europe, such as birch, grass, and hazel. We used separate ensemble Random Forest regression models for each taxon to make retrospective daily forecasts for 2006-2022 period. The data used for learning the models come from the aerobiology station in Wroclaw (southwestern Poland). Pollen grains were collected with a Burkard trap and counted according to the recommendations of the International Aerobiological Society. The developed sets of model predictors included meteorological parameters from Weather Research and Forecasting (WRF), lagged pollen concentrations, Growing Degree Days, amount of pollen emitted for previous years, and categorical predictors describing temporal variation. We used time series cross-validation to select models' parameters and evaluate their performance. We also compared the performance of the models for selected taxa in terms of different error metrics and showed which variables were significant for the prediction of each taxon. For birch e.g. the most important variables included lagged pollen concentrations, average and maximum temperature, Growing Degree Days, precipitation, week/month variation, and pollen count for previous years. The developed models show a satisfying ability to detect days with high concentrations and will be used to predict pollen concentrations based on the automatic near

real-time pollen detector newly installed at the station and weather forecasts provided by the WRF model.

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Swisspollen: First Experiences and Challenges Running an Automatic Pollen Monitoring Network

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Abstract

Following a public tender in 2019, MeteoSwiss gradually deployed an operational automatic pollen monitoring network over Switzerland (SwissPollen), consisting of Swisens Polenos. The first year of the deployment (2020) was focused on the development of identification algorithms for the most important allergenic pollen taxa in Switzerland and on ensuring quality and reproducibility of measurements. In 2021, the real-time pollen measurements were integrated operationally into the numerical forecast model and into the diffusion channels for products going to end-users (website and mobile application). Finally, in 2022, the whole end-to-end operational chain was run in a pre-operational mode to establish maintenance and monitoring procedures for the network.

Since January 2023, automatic pollen measurements are the official pollen measurements at MeteoSwiss, delivering data for 15 sites spread throughout the country. Five manual stations (Hirst-type) are kept in the long-term to maintain climatological time series and to monitor new species in different sites relevant for climate studies in Switzerland. Raw data produced by the automatic sensors are systematically archived to be used as material for the continuing development of SwissPollen and to perform reanalysis as improved machine-learning algorithms become available. The amount of produced data requires a dedicated infrastructure, with storage in the petabyte range and GPUs for model training, which we describe for potential future users.

We present the concept for network maintenance and monitoring, highlighting the challenges met while operating the automatic systems. Specifically, the diversity of the environment within the network (e.g. rural vs. urban) and of the particles present in the atmosphere over several seasons (e.g. Saharan dust) largely exceeds what can be met in punctual test campaigns. Based on this experience, we comment on potential improvements on the specifications of the measurement systems and their maintenance as defined in the 2019 SwissPollen public tender.

Finally, we provide indicators quantifying the impact of the new measurements, forecasts and distribution channels in terms of usage statistics by the general population. The operational running costs for the network remained constant when comparing SwissPollen with the manual network (pre-2019), which results in a positive return on investment due to the provision of real-time data about the pollen load and of improved forecasts to the public.

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Lidar and in-situ Detection of Pollen: Experiences From the Pericles Campaign

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Abstract

We present results from the 2023 Pericles campaign that took place at the MeteoSwiss aerological station in Payerne, Switzerland. A combination of aerosol in-situ measurements, remote sensing instruments, weather measurements and pollen instruments were run on the same site for the duration of the campaign (May to September). In addition to the MeteoSwiss operational aerosol-water vapor Raman Lidar and wind lidar, a fluorescence Lidar prototype, the Biolidar, was the centerpiece of the campaign. This instrument recorded the fluorescence signal resolved over 32 channels in addition to the elastic backscattering signal. The recorded fluorescence signal in the Planetary Boundary Layer (PBL) and in the lower free troposphere was assigned to bioaerosol from local and remote sources. Local bioaerosol sources were monitored by various instruments: Plair Rapid-E, WIBS Neo, Hund BAA500, Swisens Poleno and Hirst sampler, as well lipidomics analysis (Coriolis sampler). Poleno and Rapid-E described the Total Pollen and Grass pollen season similarly to standard Hirst measurements. Each of those instruments can assess different aerosol properties (in particular fluorescence) at various size ranges and timescales. Biolidar measurements with strong signal in the chlorophyl domain could be ascribed to grass pollen present in significant concentrations at ground level, as confirmed by the in-situ instruments, and in the air column above the measurement site. Remote sources of fluorescing particles, e.g., biomass burning from forest fires, on the other hand were identified using back-trajectory analysis.

The campaign further exploited the potential of the COSMO-ART operational weather model that provided 3D pollen concentration fields over Switzerland with a resolution of 1 km by 1 km. The COSMO-ART profiles integrate the real-time in situ operational measurements from the SwissPollen network (15 sites equipped with Swisens Polenos). COSMO-ART simulations presenting strong pollen concentrations were compared to the recorded fluorescence signal from the Biolidar. This allowed for the first time a validation of the model against measurements in the vertical by matching the observed and modeled profiles in situations where several layers of aerosols were present (e.g. originating from local or remote sources).

Finally, we discuss challenges related to the use of fluorescence Lidar measurements for qualitative and quantitative understanding of pollen dynamics in the PBL and, to a lesser extent, in the lower free troposphere. Such challenges comprise daytime use of the Biolidar, limited bioaerosol identification from the integrated Lidar signal, and finally technical efforts towards an operational system.

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Seasonal Dynamics of Arctic Marine Ice-Nucleating Particles

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Abstract

There is growing evidence that the oceans constitute an important source of ice-nucleating particles (INPs) in the atmosphere, aerosolized through sea spray. These particles play a crucial role in cloud formation and cloud properties by inducing ice crystal formation. Microorganisms, in particular, can produce ice-nucleating proteins which are efficient catalysts in the formation of ice, triggering heterogeneous freezing between -1˚C and -15˚C. INPs have been measured in sea bulk water and sea surface microlayer, and specifically Arctic waters have been shown to exhibit ice-nucleation activity at high temperatures. In addition, terrestrial environments have long been recognized as substantial reservoirs of INPs. The runoff from these terrestrial environments, facilitated by meltwater and rivers, could have the potential to contribute a substantial influx of INPs to coastal marine environments. Our understanding of the extent of this input, the properties, and concentrations of INPs, and their connection with the microbial community in sea bulk water and sea surface microlayer remains limited. Furthermore, there is a lack of investigation into the temporal and spatial distribution of INPs in sea water. This information, coupled with atmospheric INP measurements, is needed to improve predictions of

INP emissions from the ocean to the atmosphere. Therefore, we conducted a sampling campaign at Disko Island, Greenland, and collected sea bulk water, sea surface microlayer, and air samples from May to September 2023. Freshwater samples were collected from a river in continuation of a marine transect spanning eight km offshore to investigate the impact of terrestrial runoff on the coastal marine microbial community and INPs. Our investigation reveals distinct seasonal variations in INP concentrations, ice-nucleation activity, and microbial community at a regularly visited marine site throughout the sampling campaign. Air samples were collected simultaneously at this marine site, and regularly from a foreland with a specific focus on capturing sea spray emissions and investigating the INP concentrations and the airborne microbial community. Our results, further, demonstrate a pronounced input of INPs originating from terrestrial runoff into the sea surface microlayer within coastal marine waters. However, this was not observed in the bulk water, attributed to stratification resulting from the introduction of freshwater. The study unveils seasonal dynamics of INPs and microbial communities and a prominent impact of terrestrial runoff in Arctic marine waters. Moreover, it emphasizes the importance of considering the marine environment as a major source of atmospheric INPs and, further, contributes valuable insights to improve predictions of INP emissions from the ocean to the atmosphere.

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Scaling Training Efficiency in Pollen Recognition through Human-Free Annotation Initialization

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Abstract

The accurate identification and quantification of pollen types are crucial in aerobiological studies, given the profound impact of pollen on public health, agriculture, medicine, and ecosystem research. Traditional manual counting methods are not only laborious but also introduce limitations on the volume and speed of data analysis required in these fields. Addressing these challenges, our research introduces a practical deep learning based methodology that significantly advances the automation of pollen recognition and quantification.

In the initial phase we employ an innovative one-shot annotation technique that necessitates only a single query image for each type of pollen, and we further refine it with classical image processing techniques. This approach enables the fully automated annotation of pure pollen reference slides, eliminating the need for trained professionals in the annotation process. Although the accuracy of this method is not completely perfect, it facilitates the creation of extensive high-quality training datasets at scale overcoming the traditional barriers faced in pollen analysis. These datasets are instrumental in training state-of-the-art object detection models capable of accurately identifying and quantifying mixed pollen types.

By applying modern object detection models on the generated training data set, including the DETR and YOLOv8 models, our system is capable of identifying pollen grains on a mixture of reference samples with remarkable accuracy (>98% mAP@0.5). We also introduce an iterative annotation refinement loop within our training protocol to enhance the quality of training data and further improve model performance. This process involves the re-annotation of the original dataset using models that have been trained on the previously generated training data.

This advancement represents a significant leap forward in automated pollen analysis, offering a solution that is scalable, accurate, and efficient. Our approach promises to transform the field of aerobiology, contributing to public health and environmental monitoring with unprecedented precision and speed.

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Impact of Environmental Nitrogen Enrichment on Birch Pollen Allergy

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Abstract

Background:

The prevalence of allergy to aeroallergens is rising and attributed to lifestyle- and environmental changes such as nitrogen pollution, although causality is often complex to infer. Despite its known ecological impact, the impact on pollen allergenicity of environmental nitrogen pollution remains poorly circumscribed. We hypothesize that soil nitrogen pollution may directly affect pollen production and/or properties, resulting in pollen collected from different environmental nitrogen deposition conditions exhibiting different allergenicity in sensitized patients.

Methods:

Fresh *Betula* pollen samples were collected throughout Europe (Belgium, Sweden, Ireland, Poland, and Spain). In 2022, 66 samples were collected in a paired design in Belgium. Groups of birch trees located close to each other but with different environmental nitrogen conditions (predicted low vs high nitrogen) were sampled. In 2023, samples from Belgium (n = 35) and Europe (n = 63) were collected over a gradient of environmental nitrogen based on nitrogen deposition map data (Eutrophication caused by atmospheric nitrogen deposition in Europe, European Environment Agency, EEA). Nitrogen deposition values were extracted from the EMEP MSC-W modeled air concentrations and depositions (European Monitoring and Evaluation Program). Nitrogen and carbon content were measured in fresh pollen and leaf samples (CHNS elemental analyzer, Carlo Erba EA1108). Soluble proteins were extracted and normalized for protein concentration (Bradford). A selection of pollen extracts (13/66 for 2022 and 20/98 for 2023) representing the nitrogen gradient, was used to determine their allergenicity in pollen-

allergic adult patients (n = 60 in total) using *ex vivo* basophil activation testing (BAT) and specific IgE (sIgE) determination (in-house streptavidin-biotin-based ImmunoCAP, Thermo Fisher). For BAT, area under the curve (AUC), half maximal effective concentration (EC50), and maximal reactivity were compared.

Results:

For pollen samples collected in 2022 (13/66), 20 patients were tested and no difference in BAT reactivity was observed. Also, no difference in sIgE titer to predicted low vs high nitrogen pollen was seen in these patients (mean 12.51 vs. 12.47 kU_A/mL, paired t-test, $p = 0.9075$). For pollen obtained in 2023 in Belgium (10/35) and Europe (10/63) over a nitrogen deposition gradient, a significantly higher estimated reactivity was observed with lower predicted nitrogen deposition (BAT AUC estimated effect for total N-deposition -539.178 (BE, $p = 0.014$) and -402.631 (EU, $p = 0.041$), linear mixed model analysis). We are awaiting results that correlate sIgE with nitrogen deposition values. However, we did observe a significant positive estimated effect of initial pollen protein concentration on BAT reactivity (AUC 8.213 (BE, $p < 0.001$) and 25.053 (EU, $p < 0.001$)), despite our correction for this in our experimental setup. Initial protein concentrations did not correlate with nitrogen deposition estimates or measurements (total N-deposition, $n = 41$, $\rho = -0.3078$, $p = 0.0503$).

Conclusion:

Betula pollen collected during two consecutive sampling rounds in Belgium and across Europe from sites with varying levels of environmental nitrogen deposition did not consistently impact pollen allergenicity. Further analysis will assess how environmental nitrogen enrichment affects pollen production capacity and its protein characteristics.

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Automatic Pollen Monitoring Using Image Recognition

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Abstract

Automatic pollen monitoring that is correctly able to recognize individual pollen is already a fact, and is finding application especially in Europe. Currently a few instruments are on the market, of which two are currently suitable for Europe as their capacity to recognize pollen at the species level has been proven in several publications.

Networks of these instruments are being built. Here we focus on the Hund POMO BAA502.

Since the installation of POMO in the ePIN (electronic pollen information network) in 2018 however several features of the instrument were improved, besides expansion of the existing network outside Bavaria.

1. **Technical:** new algorithms were developed that are now based on AI. The performance of these algorithms against Hirst or the older Software can be seen on https://autopollen.shinyapps.io/APP_AUTOPOLLEN_compare/. In addition, hourly or 24h samples (wintertime) can now be selected by the user. 3 Monthly consumable magazines are now standard.
2. **Dissemination:** the data for Europe is now displayed at www.pollenscience.eu, using different languages. New methods are added, i.e in the EU-project SYLVA (coordinator Mikhail Sofiev) the data are made publically available in EBAS at https://ebas-data.nilu.no.
3. **Validation:** checking which image the instrument classified as which pollen species is publically available at validation.pollenscience.eu: Birch pollen in Cordoba? Impossible, but checking the images it turned out to be true.

Every image can also be manually labelled by pollen experts for later use in training for a specific location or a neophyte, enabling the introduction of new species to the algorithm using real-life pollen images (no artificial feeding of pollen needed).

Although the technical improvements will continue, the current performance is already equal if not surpassing the manual Hirst-type method, with the advantage of delivering the data fast. Thus using the automatic data as we do now with Hirst-type data is no objection.

Therefore we are developing:

1. A system that interpolates the measured pollen concentrations between the measuring stations (co-Kriging). Instead of guessing what is the best station for you, a mathematical model calculates the expected pollen data at your location
2. Improving the pollen forecast: numerical models can be improved by including the actual measured automatic data. The ePIN-opt project is doing that for SILAM, in cooperation with Mikhail Sofiev.
3. Including the pollen measurements into apps for patients, also including the predictions for individual patients. This project POLARISE, were automatic pollen counts and pollen concentration predictions are combined is under development

The current automatic pollen measurements are of such quality for some instruments (Hirst versus POMO for birch r^2=0.87, for grass 0.74, see website above), that we should at least start diverting a substantial portion of our time to the application of the new measurements, such that allergic individual and the treating doctors already profit now.

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Renewed Map of the World of Pollen and Spore Monitoring Stations

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Abstract

Having an international inventory of who is measuring which types pollen and spores at which locations in the world is of high importance for aerobiologists. Of course, having one website where all pollen data are available free of charge would be even better but this is currently not feasible. Reasons for this include that generating pollen data is time consuming, labor intensive and costly. Providing all data online at no cost would lack the acknowledgement and of course impact points from publications by the owner and thus financing for the data generators.

Thus, a website was created in 2018 where all known pollen monitoring stations are shown on a map. By clicking on a location, the owner and his/her contact data are displayed along with details on the station (device type, location, pollen types, etc.). No pollen data itself is shown. Thus, the data owner can be contacted for his pollen data.

A disadvantage of the original map was the amount of work to keep the map updated. In 2018 many stations were not known to the initiators of the map, but fortunately, new stations were submitted by the users. Unfortunately, the contact information of data owners and care takers of the station often changed, making the map obsolete. Updating the map was time-consuming and needed funding.

To solve this, we designed a self-maintaining, new version of the map of pollen and spore monitoring stations in the world. The owner of every station can add his/her station all by him-/herself. This new station is then immediately displayed on the map, but with the label “unconfirmed”. Afterwards the map administrator must approve the stations, to avoid erroneous stations. Once approved, anybody can make any changes to an existing station, such as editing the email address, telephone number, website for the data, etc. The new data will immediately be displayed on the map along with the label “Edit Requested”. At the same time, an email is then sent to the data owner to notify changes were made on his/her station. If he/she agrees, the “Edit Requested” label will be changed to “Confirmed”. If not, the new data is discarded and the previous data is restored. Also, the station owners are automatically contacted via email when one year has passed after the last update, to ask whether the station’s data are still correct. If the station owner does not update data within one month, a reminder is sent. If the data is not updated after six months, a second reminder is sent. When nobody then replies, direct contact is made.

Due to the expanding number of stations we have automized the process to update the map together with the station owners. This will keep the map of the world of pollen and spore monitoring sites as up to date as possible, and contact between pollen and spore stakeholders optimal.

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The SwissPollen network: Proof-of-Concept and Network Design

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Abstract

The SwissPollen national monitoring network has been fully operational since January 2023, however, it took several years to reach this level of maturity. As more and more such networks are being established across Europe, this presentation will describe the first steps of the process including results from an initial test phase, network design and site choice, as well as a brief overview of the IT infrastructure established to support the Swiss operational network.

After a public World Trade Organisation (WTO) tender process was won by the Swiss-based start-up Swisens, a proof-of-concept (PoC) phase was carried out over the main pollen season in 2020. Three Swisens Poleno instruments were setup in parallel in Payerne, Switzerland, and were evaluated across three axes: sampling and sensitivity, production of training datasets and operational use, performance of identification algorithms. Reproducibility was excellent, with correlations between the three devices ranging between 0.96-0.99, while comparison with manual measurements showed correlations between 0.92-0.98. Over 90 training datasets were produced using fresh pollen samples over the test season and used later to train the identification algorithm. All three devices also showed excellent stability with no interventions or maintenance required over the 4-month test period. In terms of identification, the Swisens Poleno performed well compared to manual observations, identifying the minimal list in the test criteria as well as several additional taxa.

After this evaluation phase and approval of the measurement system for operational purposes, the network rollout was started. The network design process was based on simulations from the COSMO-ART numerical forecast model used in combination with population data to optimise the location of sites. Essentially, the model output was analysed to understand the spatial coverage of each station and, together with the population density maps, areas that were not well-covered by the manual monitoring network were identified. The majority of the automatic sites are located at or very close to where the manual instruments were placed (particularly the Global Climate Observing System (GCOS) sites, which were maintained for climatological purposes), with three additional locations identified to make a total of 18 stations. Currently 15 stations are being run operationally, with the remaining three to be installed within the coming year.

Finally, to store all the data produced by the operational network and to further develop identification algorithms a dedicated IT infrastructure was established. This includes two servers, one used for quality assurance, network surveillance, and data storage, and the other with GPU capacity that is used for development and as data storage backup. Furthermore, a set of redundant tape archives are used to store all raw data for long-term access. This is particularly important so that when new algorithms are developed they can be used to reprocess the entire timeseries and produce homogeneous datasets.

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Pollen Exposure and Paediatric Emergency Room Admissions in Northern Italy (2008-2022)

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Abstract

Pollen exposure has been linked to adverse respiratory outcomes in the allergic population.

We analysed the short-term association between tree pollen exposure and counts of Emergency Room (ER) admissions in the Veneto region, Northern Italy.
For 2008-2022, we retrieved daily pollen concentrations and ER admissions for natural causes in the population residing within 10 km from 12 monitoring stations. We focused on individuals aged 0-17 years since, according to available data, most of ER admissions in children are linked to respiratory diseases. We designed a case-crossover study stratified by area and day of the week/month/year.

We estimated associations using nonlinear distributed lag quasi-Poisson regression restricted to the March-October period and adjusted for holidays, temperature, and precipitation. Pollen concentrations were categorized according to the thresholds defined by the Italian Society of Aerobiology Medicine and Environment (SIAMA): no pollen, low-to-medium concentrations, and high concentrations. In detail: *Alnus*, *Betula* and Corylaceae (≤0.5;0.6-50;>50 p/m^3), Cupressaceae (≤4;4.1-90;>90 p/m^3), Oleaceae (≤0.5;0.6-25;>25 p/m^3).

There were around 1 million paediatric ER admissions in the region during the study period. Across all areas combined, the 99th percentile of daily concentrations ranged from 24 p/m^3 for *Alnus* to 280 p/m^3 for Corylaceae. We observed mainly immediate effects following pollen exposure. At lag 0, relative changes in ER admission counts (with 95%CI) for days with low/medium and high concentrations, as compared to no pollen, were: *Alnus* +3.0% (+1.9,+4.1%) and +9.8% (+5.6,+14.1%); Oleaceae +4.6% (+3.7,+5.6%) and +7.8% (+6.2,+9.4%); Corylaceae +1.9% (+0.7,+3.1%) and +2.9% (+1.1,+4.8%); Cupressaceae, +1.0% (+0.1,+2.0%) and +2.1% (+0.3,+3.9%). Associations for *Betula* pollen were null.
Exposure to *Alnus* and Oleaceae pollen showed the strongest associations with increased paediatric ER admissions in the region over the last two decades. Pollen forecasting alerts could aid susceptible individuals in implementing self-protection strategies.

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Real-time Spore Detection of Downy and Powdery Mildew in Vineyard Using Holographic Imagery

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Abstract

The oomycete *Plasmopara viticola* (PV) and the ascomycete *Erysiphe necator* (EN) represent major issues in grapevine disease control, causing respectively downy and powdery mildew. As polycyclic pathogens, both exhibit fast asexual cycles, leading to prolific spore production and dissemination. To counteract these diseases effectively, growers rely on preventive application of fungicides according to decision support systems tools based solely on meteorological parameters for predicting future infections. However, the absence of spore load data in such models sometimes results in missed infection predictions or, more often, in predicted infections at times when no spores are present, leading to unnecessary fungicide treatments and extra production costs. Adding a biological parameter such as the spore concentration in the air may represent a useful addition for more reliable predictions of infection events, and would allow growers to treat only when necessary. For this purpose, we developed SMALA (SMart Agriculture using Lasers and Artificial intelligence), a spore detector device able to independently capture, classify and count PV and EN spores in real time.

Our innovative device uses digital holography, a cutting-edge technology capable of producing highly accurate 3D spore prints unique to each species with cost-effective materials. When a laser beam hits spores on a thin wafer, it generates holographic images that capture diverse information such as spore size, density, and thickness. These holographic images are then analyzed by two artificial intelligence algorithms specifically trained to differentiate PV and EN spores from potential interferents of comparable sizes such as pollen or spores of closely-related species. Following AI training with pure cultures of spores and pollens in laboratory, correlation matrices presented 97.3% and 95.5% accuracy.

Validation studies conducted in Changins, Switzerland, compared the performances of the spore detection device with traditional passive and active spore trap using multiplex qPCR on environmental samples. The results demonstrated good accuracy and efficiency in identifying and quantifying PV and EN spores throughout the season.

Field assays conducted in 2023 further underscored the efficacy of this novel technology in vineyard disease management. By leveraging spore load data obtained from the detector, we were able to provide informed treatment decisions, resulting in a significant reduction of powdery mildew incidence in the field. In particular, targeting spray application after we observed the first peak of spore release, proved to be highly effective in minimizing disease spread. In total, we observed up to 30% reduction in the number of fungicide application throughout the growing season.

Moreover, the integration of spore load data into existing forecasting models, such as Vitimeteo-Plasmopara, holds tremendous potential for enhancing infection prediction accuracy and optimizing treatment timings. The ability to monitor spore inoculum heterogeneity within vineyards in real-time enables growers to tailor disease management strategies to specific microenvironments, while adopting more sustainable agricultural practices. By understanding the epidemiology of each disease at a finer spatial scale, it is possible for growers to implement interventions precisely where and when they are needed most, thereby minimizing both disease incidence and pesticide usage.

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Prediction of Hazel and Cypress Pollen Concentrations Using the SILAM Model

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Abstract

Corylus avellana (common hazel) and *Cupressus sempervirens* (Mediterranean cypress) are among the early blooming allergenic species in Europe, often flowering in late winter and/or early spring. Their distribution across Europe is rather scattered so that they do not form extensive forests or mixed woodlands. Consequently, their pollen concentrations remain relatively low, even at their peak, compared to species such as birch. An adequate prediction of their levels, especially at the edge of the distribution range, is a challenge due to the low pollen counts.

Hazel often blooms in Central Europe as early as January or February, and even in warmer winters, it can flower as early as December — similarly, cypress typically flowers in January or February. The distribution of cypress is concentrated around the Mediterranean region, with some ornamental plantings found outside this area. Although an exact map of cypress habitats was not available, a customized map was created from ECOCLIMAP data.

Pollen concentration forecasts utilized the Finnish Meteorological Institute's SILAM atmospheric dispersion model, which is an Eulerian chemistry transport model. Generating numerical pollen forecasts requires plant distribution, abundance, and flowering time information. For the early spring pollinating trees, a degree day model is often suitable for the prediction of the flowering onset. In the SILAM model, the accumulation of degree days for these early blooming species begins on January 1st. However, for cypress and hazel, this may be too late, as flowering can commence before the model starts the heat accumulation, particularly in warm winters and in warmer habitats. Some cypress species (e.g., *Cupressus arizonica*) and junipers (e.g., *Juniperus*

oxycedrus) flower even before the turn of the year, but their flowering was not attempted to be modelled in this study.

Pollen concentrations of hazel are counted from most pollen observation sites in the EAN database, allowing for direct comparison with model predictions. However, model comparison with cypress pollen counts should be treated carefully due to the variety of flowering times and distribution patterns within the Cupressaceae family. The genus specifications within the Cupressaceae family are normally not reflected in measurement reports. As model evaluation shows, the SILAM predictions of hazel concentrations in Eastern Central Europe are well-matched, but the forecasts for northern areas are less reliable.

In conclusion, the numerical prediction of pollen concentrations for early blooming species like hazel and cypress remains challenging in Europe, especially in regions where these species are less common. Further research is needed to improve the plants' distribution representation and correction of the pollen release time in the SILAM model with special attention to the areas where phenological variability within the families is most pronounced.

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Influence of Photoselective Nets' Colour in Pollen Quality of 'Matua' and 'Tomuri' Kiwifruit Cultivars

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Abstract

Proper fructification in fruit tree crops is only achieved if an effective transference of pollen from anthers to the stigmas occurs and the pollen grains exhibit germinative capacity to originate pollen tubes able to fertilise the oospheres. So, pollen quality is crucial to obtaining fruits with the required size and weight for the market demand. However, to our knowledge, the influence of covering nets, such as Photoselective nets (PNs), in pollen quality is not characterised.

It is essential to know how pollen can respond to the presence of PNs, given that this agricultural technology is increasingly used and influences the level of shading and the proportion of spectrally modified and unmodified radiations reaching plants under them.

So, this study aimed to investigate the influence of PNs on the pollen quality of 'Matua' and 'Tomuri' kiwifruit cultivars by assessing morphological, fertility and biochemical properties.

Pollen samples were collected from the kiwifruit male pollinators 'Matua' and 'Tomuri' at a 30-year-old commercial orchard in Guimarães, Northwest Portugal. Plants grown in four experimental conditions were tested: uncovered control and grown under yellow, pearl and grey PNs. Male flowers were randomly collected at full flowering from all quadrants at various heights of each canopy and pooled per condition and cultivar (100 flowers in each pool). Pollen was separated by sieving and stored at −20 °C until analysis.

The pollen grain size parameters and exine microperfurations were studied by Light and Scanning Electronic Microscopy, respectively. Fertility was analysed using viability assays by fluorescein diacetate (FDA) staining quantified by flow cytometry and in vitro germination. Total soluble proteins (TSP) and sugar (TSS) content were quantified by biochemical/colourimetric assays. Potential disparities in the molecular structure of the pollen grain wall were analysed through Raman spectroscopy. Data normality and homoscedasticity were verified using the Kolmogorov-Smirnov test or the Shapiro-Wilk (when $n < 30$). A two-way ANOVA followed by a Duncan *post hoc* test was applied except for Raman spectral analysis, where a one-way ANOVA was used per cultivar to test the hypothesis of significant differences in the prominent Raman peak's (W) mean values between experimental conditions.

Results showed that pollen collected from plants under the PNs had a significantly bigger width and area and a lower germination rate. TSP and TSS content variations induced by the PNs were cultivar-dependent but only significant for the pearl PN. Raman spectra of the pollen from plants grown under the nets presented some bands that significantly shifted from their original position, indicating differences in the vibration modes of the molecules but no overall changes at its structure or organisation level. Our study showed that PNs can influence several pollen traits and that the cultivar can affect the outcome.

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Spatiotemporal Variation in the Concentration of *Cladosporium* Sp. And Alternaria Sp. Spores in the Urban-rural Transition Zone

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Abstract

Fungi have a wide influence on ecosystems, as the same species can be both saprotrophs and pathogens. They also affect human health by releasing wind-transported spores, which, when inhaled, may trigger allergy symptoms. Spores of *Alternaria* sp. and *Cladosporium* sp. are typically included into aerobiological monitoring due to their high abundance and allergenicity. The spatial arrangement of the „rooftop" measuring stations means that only averaged information for large areas is available, and mismatches with concentrations at the 'ground level' are often recorded. It is believed that local sources substantially contribute to spores concentrations, and the main source areas of spores are agricultural lands, especially during harvesting. However, it is unclear how proximity of crops affects other areas, where people typically live. Therefore, the aim of this study was to determine the differences in fungal spore concentrations between different land use types and to compare spore concentration data recorded at the rooftop level with that obtained from ground-level stations in the nearest vicinity.

Portable volumetric traps (Burkard Manufacturing Co Ltd) were used to conduct three ground-level measurements campaigns (before, during, and after harvesting) throughout the main sporulation season in 2023. The devices sampled air at 15-minutes intervals, in a synchronized manner. Nine sampling points within a radius of 2 km from the standard 'roof-top' aerobiological monitoring station (based on 7-day Hirst-type trap) on the outskirts of Poznań (western Poland) were established. These points represented the main land use types: agricultural, urban and forest. Simultaneously, wind speed and direction were recorded at each sampling site using portable meteorological stations.

Both *Cladosporium* sp. and *Alternaria* sp. spore concentrations increased during the week of harvesting; however, these changes were more pronounced near the fields and forest sites in the case of *Alternaria* sp., and urban sites for *Cladosporium* sp. After harvesting, a further boost in the concentrations of *Alternaria* sp. spores on all sites and a return to pre-harvesting levels of *Cladosporium* sp. spores was observed. The highest spore concentrations were recorded on agricultural sites: 1401 s/m^3 for *Alternaria* sp. (August 23rd, between 14:50 and 15:05), and for *Cladosporium* sp. 68191 s/m^3 (July 25th, between 8:55 and 9:10). During these time periods, the rooftop concentrations were much lower, i.e., 212 s/m^3 and 1156 s/m^3, respectively. The increases in the spore concentrations in forests and urban areas coincided or were slightly

delayed compared to situation in the fields. Diurnal patterns in spore level was characterized by distinct afternoon peak for *Alternaria* sp. and around noon peak for *Cladosporium* sp.. In general, ground-measured concentrations were higher and had higher variance than roof-top spore data.

Our results shows that the highest concentrations of fungal spores were observed in the direct vicinity of cereal crops. However, on this fine spatial scale, the increase and changes in spore level happened almost simultaneously in other land use types. Notably, there is under-representativeness of spore concentrations measured at the rooftop. The collected data allowed us to assess the real exposure to fungal spores near people's places of residence.

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Predictive Modeling of Oleaceae and Quercus Pollen Seasons in Thessaloniki, Greece

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Abstract

Airborne pollen is set to impact over 50% of the EU's population by 2025, with Thessaloniki, Greece experiencing significant increases, with pollen concentrations doubling every decade. In the Mediterranean region, Oleaceae and Quercus emerge as primary sources of airborne allergenic pollen, presenting challenges in predicting the onset and ending of their main pollen season (MPS), i.e. the period where the majority of the annually produced pollen can be found in the air. To meet this demand, this study explores six methodologies, offering diverse perspectives on predicting the Oleaceae and Quercus pollen season. In the Biology Department of Aristotle University (40°37'N, 22°57'E) in Thessaloniki, a Hirst-type spore trap operates, following European Aerobiology Society guidelines. Utilizing data collected from 2016 to 2022, alongside meteorological data from ECMWF reanalysis, which includes temperature, precipitation, and photoperiod.

Various predictive methodologies discernible for forecasting MPS encompass distinct approaches. The Double Threshold Air Temperature Sum Approach (DT) relies on ontogenetic development stages influenced by air temperature, utilizing two critical thresholds. Thermal Time models (TT) integrate heat accumulation factors like Heat Units, Growing Degree Days, and chilling requirements. Similarly, the Distribution Method (DM) fits a Gaussian distribution

based on observed pollen counts, enhancing prediction accuracy. Moreover, Partial Least-Squares Regression (PLS) is utilized to construct phenometeorological models, alongside Temperature-Photoperiod (TP) models, which integrate temperature and photoperiod. Lastly, employing Machine Learning Techniques (MLT) to forecast the last year of the dataset and the walk-forward method, enhances predictive capabilities. The selected techniques encompass Artificial Neural Network (ANNs), Multi-Layer Perceptron (MLP), Naive Bayes (NB), Random Forests (RF), Linear Discriminant Analysis (LDA), and an Ensemble approach integrating these methodologies.

For Oleaceae, DT, TT, and TP models effectively capture pollen periods, whereas PLS method accurately identifies MPS start, with only a +5 day deviation in one year for the ending. Regarding the MLT, for onset prediction, NB and ensemble methods demonstrate excellent performance with a deviation of approximately ±1 day, while employing the walk-forward method reveals MLP as the most satisfactory performer. Conversely, for ending prediction, LDA with a deviation of merely ±2 days emerges as the most accurate, whereas the combination of ANNs and MLP exhibits a deviation of approximately ±1 day when employing the walk-forward approach. Regarding Quercus, the DT and TT methodologies exhibit minor deviations and satisfactorily capture the MPS with a maximum deviation of ±5 days. Similarly to Oleaceae, the PLS method demonstrates exceptional accuracy in predicting both onset and ending. In MLT, ANNs are highly effective in onset forecasting using the walk-forward method, with a maximum deviation of ±5 days. For ending prediction, MLP achieves a deviation of ±3 days. This study advocates for an optimal prognosis achieved through the synergistic utilization of PLS and MLT methodologies.

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Towards Low-cost Hyperlocal Pollen Detection Using Optical Particle Counters and Artificial Intelligence

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Abstract

Introduction

Low-cost sensors have revolutionised the measurement of air pollution by reducing the cost, size and weight of air pollution measurements. This revolution has allowed for more measurements to be made and it has allowed for measurements to be made in previously inaccessible locations. It has also allowed for multiple measurements to be made simultaneously using numerous sensors in urban arrays leading to so called hyperlocal urban measurements.

In this presentation, we will show how low-cost optical particle counters (OPCs) can be combined with source apportionment algorithms, including machine learning, to measure pollen in the atmosphere. The relatively low number concentration of pollen particles compared to particulate matter (PM) air pollution makes this a ‘needle in the haystack’ type of problem.

Methodology

We use Alphasense OPC-N3 optical particle counters to measure the total PM concentrations in multiple size bins from 0.3 – 40 mm. This size range encompasses many pollen species and their fragments called sub pollen particles (SPPs).

We use various algorithmic approaches to separate the pollen particles from the greater PM background (Mills et al. 2023a). Traditional source identification and apportionment techniques, such as k-means clustering and positive matrix factorization (PMF) are used. We also utilize machine learning approaches, including neural networks and random forests, to construct pollen proxies. Explainable artificial intelligence (XAI) is used via the SHAP approach to provide illumination to the ‘black box’ artificial intelligence models (Mills et al. 2023b).

Results

The different approaches for constructing pollen proxies are evaluated through intercomparison with campaign data using Hirst-type samplers (Maya-Manzano et al. 2023). All the algorithmic approaches show some skill, but the random forest and neural network approaches show particularly promising results, with AI constructed proxies successfully replicating temporal trends in pollen and high pollen events. The models are demonstrated for grass, oak, birch, pine and total pollen species. We will show how neural network hyperparameter tuning can be used for improved model performance. Using the SHAP method, we will show which model input features, including OPC size bin, relate to the target pollen concentration.

Implications

Through synthesis and evaluation of the results, we will highlight how the combination of low cost OPC sensors and advanced algorithmic approaches has great potential for low-cost sensing of pollen, this has the potential to allow for the hyperlocal monitoring of pollen at a significantly lower cost than the current sparse regulatory networks.

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Identification of Hotspots in the Source Regions of Ragweed Pollen Arriving in Türkiye and the Impact of Meteorological Conditions on Hysplit Model Results

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Abstract

Background and Objectives

Ambrosia artemisiifolia (common ragweed) is a wind-pollinating and invasive species in Türkiye. Understanding the long-distance transportation of *Ambrosia* pollen is crucial for assessing its impact on human health and devising effective management strategies. We investigated

whether the highly allergenic *Ambrosia* pollen recorded in two stations in 2023 could be the result of long-distance transport (LDT) and aimed to identify potential sources of *Ambrosia* pollen grains.

Material and Methods

The aerobiological data were recorded at two monitoring sites (Ankara and Zonguldak) situated in Türkiye by using a volumetric Hirst-type pollen trap. Nonparametric statistics were utilized to examine the impact of meteorological variables on daily pollen counts. Employing the R mgcv package, we applied a generalized additive model (GAM) to capture both combined and individual meteorological effects. HYSPLIT analysis and the DBSCAN (Density-Based Spatial Clustering of Applications with Noise) clustering algorithm were also utilized to identify potential sources of long-distance transported pollen to two stations in Türkiye. We analyzed every back trajectory that reached the stations on days with elevated pollen concentrations to pinpoint areas where air masses came close to the ground, within 100 meters. Subsequently, we applied the DBSCAN clustering algorithm to cluster these points, optimizing parameters based on the ratio of the number of clusters to the silhouette value. After noise points were removed, we converted clustering points into polygons and selected centroids as hotspots of potential sources. Additionally, we used these source points to generate forward trajectories, illustrating the conditions under which air masses were transported to the sampling stations

Results

According to GAM results, minimum and mean temperature, rainfall, wind speed and wind direction were identified as significantly related factors on pollen distribution. Our study supports the hypothesis that the long-distance transport of ragweed pollen to Türkiye is primarily influenced by air masses from the north and/or northeast direction. HYSPLIT simulations with elevation data revealed one of the potential sources around the Sea of Azov and parts of Russia facing the Black Sea, where large ragweed populations were observed. Forward trajectory analysis from the potential sources indicated that cyclonic movements in the Black Sea region could significantly affect the trajectory of air masses, either directing them towards inner Anatolia or preventing their arrival.

Conclusion

In regions where *Ambrosia* invasion has not fully occurred, such as in Türkiye, assessing meteorological factors in the regions where hotspots exist alongside those in the station's location may have a positive impact on the quality of the results obtained when evaluating the concentration of airborne *Ambrosia* pollen.

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The Effects of in Vitro Exposure to Ozone and Nitrogen Dioxide in Pollen of Different Tree Species

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Abstract

Atmospheric pollutants are reported to affect pollen; however, results are sometimes contradictory among studies, and comparison is challenging because experimental designs are variable. Also, it is suggested that plant species present different sensitivity and resilience to atmospheric pollution and distinct exposure concentrations. So, we aimed to investigate the influence of O_3 and NO_2 exposure on the pollen viability, protein content, oxidative stress, and wall composition of four tree species: *Betula pendula*, *Corylus avellana*, *Acer negundo* and *Quercus robur*, under the same experimental conditions.

The pollen samples were collected directly from the trees during their peak flowering period and in vitro exposed to O_3 or NO_2 in an environmental chamber at three concentration levels (half, equal and two times EU limit concentration values for human health protection). Pollen viability was assayed using fluorescein diacetate (FDA), and soluble protein content was quantified according to the Bradford method. Stress conditions were evaluated by detection of the reactive oxygen species (ROS) using a fluorescent ROS indicator (DCFH2-DA), through the activity of NADPH oxidase quantified by spectrophotometry based on the nitro blue tetrazolium assay (NBT), and superoxide dismutase (SOD) activity by immunoblotting. Wall composition changes were analyzed by Raman microspectroscopy. A one-way ANOVA was applied to evaluate the quantitative effect of the O_3 and NO_2 pollutant gases, followed by the Turkey post hoc test ($p<0.05$) when significant differences were detected.

A significant statistical reduction in pollen viability induced by O_3 and NO_2 was observed, but the percentage loss varied depending on pollen species, gas tested and their concentrations.

The pollutants' effects on pollen total soluble protein (TSP) were contradictory compared to the blank samples. In *A. negundo* and *Q. robur* pollen, almost all samples exposed to the pollutants presented no change or a significant decrease in TSP content. The other species showed similar behaviour, having equal or higher TSP content than the blank samples.

A stimulation of pollen oxidative defences was observed in response to pollutants' exposure. An overall trend of an increase in ROS percentage after pollen exposure was registered, although it was not always statistically significant for *B. pendula* and *C. avellana*. The NADPH oxidase enzymatic activity was the parameter with the lowest significant change, with only the higher concentration levels inducing an increase in the activity and NO_2 being the most important.

Significant changes with increased SOD expression were found mainly at the limit and two times limit concentration. Small changes in the pollen wall chemical structure after exposure to NO_2 and O_3 were observed in the spectra' intensity and some peaks' flattening into shoulders.

Overall, NO_2 affected all species more than O_3 and *A. negundo* emerged as the species demonstrating greater resilience overall. *B. pendula* and *C. avellana* pollen displayed comparable responses to pollution.

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Effect of Atmospheric Pollen Concentrations on Daily Asthmatic Outpatient Cases in Augsburg, Germany- A Time Series Analysis

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Abstract

Introduction

As a consequence of climate change, air temperatures in Europe are expected to increase. This has a direct and indirect impact on human health. Rising air temperatures have been leading to an earlier onset of the vegetation and flowering period and thus to an earlier start of the pollen season. However, some questions remain open: How do extreme temperatures and pollen concentrations interact? Are there differences among pollen taxa? For which population groups is the health risk particularly high? Is it possible to define threshold values and hence develop or improve warning systems? To answer these questions, we work with the following data sets:

(1) Medical emergency cases requiring no inpatient treatment in Augsburg provided by the Kassenärztliche Vereinigung Bayern (KVB). For this study we considered all cases with ICD-Codes J45.- (Asthma bronchiale) or J46.- (Status asthmaticus).

(2) Daily aerobiological data monitored by an automatic Pollen Monitor Hund BAA500, located at the University Clinic of Augsburg.

(3) Meteorological data including daily temperature values (minimum, maximum and mean) provided by Deutscher Wetterdienst (DWD).

Limited by the available pollen data the study period includes 5 years (2018 – 2022).

Methodology

A (penalized) distributed lag non-linear model (DLNM), which constitutes a start of the art approach for modelling complex relationships in the context of epidemiological time series analysis, is used. Thereby, we control for seasonality, day of week and public holidays in the city of Augsburg. Furthermore, the temperature and the pollen concentrations are included in the model as cross-basis objects in the framework of DLNMs. This specific parametrization allows for the modelling of complex, nonlinear and delayed relations between the exposure variable (e.g. pollen concentrations) and the outcome variable (asthma-related cases). Also, the fraction of the total number of cases attributable to a specific cause can be estimated.

Results

We found that *Betula* and Poaceae pollen had the strongest impact on asthma cases in the study period. The relative risk (RR, relative to 0 pollen concentration) was calculated for the 50^{th}, 90^{th} and 95^{th} percentile of the pollen concentrations. For *Betula* we obtained an increased risk of 1.02 (1.01, 1.03), 1.34 (1.10, 1.63) and 1.70 (1.19, 2.44), while for Poaceae we found 1.03 (1.00, 1.06), 1.29 (1.01, 1.65) and 1.56 (1.02, 2.37).

Further the DLNM model indicated differences in the lag structure, with *Betula* showing a stronger immediate (same day) effect with a faster decay compared to Poaceae. For *Betula* the same day RR (95^{th} perc.) was 1.14 (1.04, 1.25) and 1.06 (1.01, 1.11) after three days, while a slower decay of the RR from 1.09 (0.99, 1.19) to 1.05 (1.00, 1.11) was observed for Poaceae.

Discussion and conclusion

In our study in Augsburg, we found that *Betula* and Poaceae, two of the most allergenic pollen taxa in the area, together with other environmental variables, could be related with asthma-related emergency cases. Revealing differences in the effect's strength and lag patterns among the different pollen taxa can help to improve prevention and the protection of vulnerable groups.

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Long-Distance Transport of Birch Pollen to Iceland: A Pioneering Study

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Abstract

Iceland's geographical position makes it an ideal site for studying long-distance pollen transport from surrounding land masses. The vascular flora of Iceland is young, originating postglacially, and has a low species count. Birch pollen is one of the two main allergy triggers in Iceland, necessitating a comprehensive understanding of its sources for allergy management. Long-distance transport (LDT) events for birch pollen have been documented in the literature. However, these descriptions, compared to other allergenic plant taxa, such as e.g. ragweed, are relatively rare and are usually associated with the presence of pollen grains outside the expected pollen season.

We aimed to investigate the LDT of birch pollen in Iceland and its potential contribution to local airborne birch pollen levels. Birch pollen counts were analysed at two locations: Reykjavik (S Iceland) and Akureyri (N Iceland), with air samples collected from 1998 to 2023, using the volumetric method. We examined a long-term series of pollen and meteorological data and assessed atmospheric circulation patterns. Trajectories of moving air particles were calculated backward for 96 hours at 1-hour intervals and at three different altitudes: 500, 1000, and 1500 meters above ground level (a.g.l.).

The study aimed to explore: 1) the occurrence of birch pollen before local flowering time, and 2) unusually high concentrations of pollen in the air due to a combination of local and long-distance sources. Four years stood out when pollen seasons started significantly earlier, exceeding the mean value over several years: 2003, 2006, 2014, and 2019. In these years, pollen grains were recorded in April well outside the local growing season. Some years saw pollen grain counts surpassing the multi-year average, notably 2005, 2017, and 2022 for Akureyri, and 2010, 2011, 2012, and 2013 for Reykjavik. These years, marked by unusually high pollen counts, were selected for analysing a possible LDT of birch pollen. The study showed that LDT is one of the factors that influenced the course of the pollen seasons both in Reykjavik and Akureyri, and that source areas of pollen transported by LDT are different for Akureyri and Reykjavik. Our results indicate several extra-regional pollen transport episodes to Iceland. The main provenances were: (1) The British Islands (mainly Scotland) (2) The Scandinavian Peninsula (3) Central and Eastern Europe (4) Greenland and (5) Canada (Labrador Peninsula). The number of pollen grains spanning from a few grains up to more than 200 pollen grains per day. Our study

shows that LDT events could appear either in one of both investigated locations or in both places depending on meteorological conditions. The LDS events could be short and last only for a few hours during the day, but could also last for a few days. We showed that LDT of pollen influencing pollen seasons in Iceland occurred over significant distances spanning from 290 km (source areas in S Greenland) to about 2,000 km (Canada). We showed that LDS events need to be taken into account in Iceland as a factor responsible for triggering episodes of pollen allergy.

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Urban Tree Pollens: Improving Allergy Management Through Advanced Pollen Identification and Spatial Modelling.

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Abstract

Urban trees play a crucial role in improving the quality of life in cities, providing numerous ecosystem services. However, they can also contribute to worse air quality through the production of pollen. Exposure to allergenic airborne pollen is a risk factor for respiratory allergies and a major public health concern, especially as climate change is lengthening the pollen season. Pollen concentrations vary spatially and temporally, especially within a city, which makes pollen monitoring a critical tool for research and healthcare improvement. However, in Montreal (Canada), pollen concentrations are currently obtained from a single station assumed to represent exposure over the very large urban area. Therefore, none is known about how pollen levels – for all pollen types, such as grass, weeds, and trees – vary within the city and whether this variation affects the development and exacerbation of allergic reactions. Furthermore, due to negligible morphological differences between related species, pollen grains are rarely identified at the species level. In this project, we focus on pollen from urban trees, which are the first to bloom, collected using 25 gravimetric traps placed throughout the island of Montreal. The results of our study are based on 950 pollen samples collected between 2021 and 2023. To improve the process of pollen identification, we developed an innovative integrated approach that combines flow cytometry with machine learning enabling faster and more accurate identification of specific types of pollen at the species level. Then, we developed spatio-temporal models by combining the collected pollen data with the canopy composition and other environmental factors. Our results revealed that the distribution of airborne pollen in Montreal, in terms of total load and relative abundances, is complex and influenced by much more than nearby trees. These findings, coupled with our innovative approach, represent a

major step towards better-informed, science-based urban greening plans, aiming to minimize allergy impacts.

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Assimilating Real-time Observations for Bioaerosol Forecasting: What Works, What May Work, What Will Not Work

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Abstract

Background

Development of real-time bioaerosol monitors, flow cytometers and impactors, has been largely justified by a necessity to have these measurements on time for combined model-measurement forecasting of bioaerosol concentrations in the air. This idea was based on a tremendous success of data assimilation in the meteorological world about 40 years ago, which was expected to be easily transferable to air quality and pollen/spores forecasting. Reality, however, proved more complicated. Classical data assimilation of the model state (pressure, temperature, etc for meteorological models, concentrations of pollutants for AQ models) failed to provide lasting improvement of AQ forecasts. The same problem was demonstrated for pollen. The origin of the challenge is now well understood, from both theoretical and practical standpoints: external forcing (first of all, emission of pollutants and meteorology) quickly overshadows the effect of assimilation. The assimilation procedure has to adjust this forcing in order to have a meaningful lasting impact. Extrapolation of this knowledge to the bioaerosol forecasting is the current challenge.

Methods

The presentation will be based on modern data assimilation techniques, such as 4-dimensional variational assimilation, as well as Ensemble Kalman Filter. Unlike simpler techniques (3-dimensional variational assimilation or optimal interpolation), these tools allow for assimilation of the model forcing and even its parameters, which is the main pre-requisite for the lasting forecast improvement.

Results

We will demonstrate examples of data assimilation, both simplified artificial cases and from real-life applications, showing the current status of the bioaerosol models and their capabilities in assimilating the real-time data. The considered real-life cases will include retrospective European Pollen Reanalysis as the most-classical example of assimilation of the long-term Hirst-type data, and pollen forecast in Bavaria, which makes use of the ePIN near-real-time data for improving the forecasts.

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Aerotape: real-time detection of ragweed pollens by optical imaging

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Abstract

Due to global warming, ragweed is expected to produce high concentrations of allergenic pollens in the air over lengthened pollen seasons, therefore increasing the risks of associated allergies. Today, ragweed is a major public health issue and the European Union has put in place several prevention measures to "assessing and controlling the spread and the effects of common ragweed in Europe" according to ENV.B.2/ETU/20110/0037.

In this context, there is a need to densify the local monitoring capacity to better account for exposure risks, but also to geolocate the hotspots of ragweed sources. In many areas impacted by ragweed, local fighting efforts require to be better driven and assessed. Automatic middle-cost instruments can fulfill this need as some of them combine in real time a qualified ragweed detection with wind conditions, therefore produce key data for inverse modelling tools.

To date, the conventional method that satisfies the Hirst standard EN 16868-19 consists in collecting pollen grains which then undergo chemical treatment to be identified and quantified under an optical microscope by an analyst. Despite the robustness and fair reproducibility, it has proven through the ages, the Hirst method suffers from the laborious nature of its 2-step operation (collection then analysis) which prevent it to be networked at a local resolution.

The Aerotape approach was designed to overcome these limitations as our compact instrument 40x30x20 cm integrates an automatic collection stage and an optical imaging process allowing direct digitization of all types of particles in the size range of 1 to 200 µm. Aerosol recognition is achieved in real time using aerosol descriptors and/or deep learning. Aerōtape is among the three automatic instruments that will contribute to the metrology research programme BIOAIRMET funded by EURAMET from 2024 to 2027, under the coordination of PTB in Germany.

The Aerotape was installed in the city of Beaurepaire (5,000 inhabitants), between Grenoble and Lyon, in a rural area highly impacted by ragweed, on the roof of the town hall, from the end of July to the beginning of October 2022.

Our results report the temporal monitoring of ragweed pollen concentrations and were compared for validation to those of a collocated Hirst sampler. The research work consisted in comparing different methods to identify ragweed grains among varied other atmospheric microparticles: image treatment, on one hand, involving colorimetric and morphological descriptors, like circularity, solidity, convolution..., and deep learning methods in particular the CNN neural network technique. The Hirst counts and Aerōtape counts are comparable, whether Aerōtape data are obtained by image treatment or by deep learning. Therefore, image treatment seems a sufficient approach to produce ragweed pollen recognition in real time. However, the deep learning method must be spared, as it can offer new features in conditions, like in dusty environments, when descriptors for image treatment get confusing.

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Improving Urban Air Quality and Public Health Through Smart Green Infrastructure Design

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Abstract

Climate change has a major impact on temperatures, especially urban areas retain a lot of heat, known as the urban heat island effect. Green infrastructure offers potential cooling benefits but can also pose health risks if not appropriately designed. Within the Dutch Research Council financed BENIGN project, my objective is to create guidelines for smart urban green infrastructure that promotes public health. This study investigates the impact of different green infrastructure designs on local airborne pollen and fine dust concentrations.

Pollen and fine dust were monitored biweekly over two years before and after the redesign of green infrastructure in ten urban localities in Leiden, The Netherlands. Vegetation was mapped in these localities, including flora in privately owned gardens. Aerial pollen was identified and quantified using DNA metabarcoding and automated image analysis, whereas fine dust was

measured using a particle counter and automated image analysis. Local samples, including pollen from private gardens and flower stalls, were captured using a handheld sniffer. SEM images revealed fine dust adsorption on pollen exposed to car exhaust fumes. Results will be used to calibrate models predicting pollen-fine dust interactions to guide green infrastructure design for improved air quality and public health.

To validate the developed guidelines, volunteers will walk through streets featuring allergenic and non-allergenic plants. Their exhaled breath will be chemically analyzed for biomarkers to quantify hay fever symptoms—a novel, non-invasive method. This study aims to underscore the direct impact of local flora on hay fever symptoms, advocating for street designs with non-allergenic plants in municipal planning.

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Methodologies of Construction and Resulting Configurations of a European Automatic Bioaerosol Monitoring Network

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Abstract

Background

A growing number of real-time bioaerosol monitors in Europe brought up a problem of their optimal distribution around the continent. High initial investments suggest that the number of these monitors will not reach the level of the European Aeroallergen network (~300 active stations, up to 600 installations in total). This financial limitation calls for optimization of the emerging network.

Methodology

Two approaches to construction of an automated pollen monitoring network for Europe are considered: a network for direct representation of concentration fields and a network for model-based data assimilation, inverse problem solution, and forecasting. Strengths and weaknesses of each approach are discussed and related to a declared purpose of the network establishment, criteria of its performance, and the key beneficiaries and stakeholders of the data. The technological side of he network design involves pollen dispersion modelling, footprint computations, and inverse problem solutions of varying levels of complexity. A multi-step design methodology includes the network representativeness and redundancy evaluation and improvement followed by the model-based analysis of its quality.

Results

It is demonstrated that, depending on the criteria and types of data usage, the network configuration can be drastically different. Several configurations are constructed for arguably most-frequently used criteria and ways of the data usage.

Applications of the methodology are demonstrated for real-life and artificial examples, including a recently published example of German pollen monitoring network and potential Europe-wide automated network with limited number of stations. Dependence of the network fidelity on the number of stations is discussed for these examples.

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Trends in Allergic Sensitization and Exposure to Olive Pollen in Alentejo, Portugal: the Impact of Land Use

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Abstract

Olive pollen is among the most allergenic species in the Mediterranean and the second cause of pollen allergy. The growing increase in the cultivation of olive groves as well as its use as an ornamental plant in Alentejo may have an impact on exposure to allergens, potentially increasing the risk of allergic sensitization. This study aimed to comprehensively characterize the prevalence and trends of allergic sensitization in Alentejo, Portugal, in relation to the pollen levels over the last decade.

A retrospective analysis (2006-2021) of the population sensitization data found at Clinidata®XXI at HESE was performed; the levels and type of specific IgE for the different allergen groups, with a special focus on olive tree pollen were collected. The pollen levels were collected from https://lince.di.uevora.pt/polen/index.jsp, University of Évora. Analysis of the association between the two variables was performed.

The extension of cultivar groves increased over the las decade in Alentejo, having almost tripled around the Alqueva basin (from ~20,000ha to >70,000ha). The annual pollen index varied

between 1,441-10,403 pollen/m^3, the highest being 2021 season. The annual pollen index presents a tendency to increase in the last year. Interestingly a tendency to diminish between 2011 (~10,500 pollen/m^3) and 2018 (~4000 pollen/m^3) while it tended to increase between 2018 and 2021 (>15,000 pollen/m^3), not following the steady increase profile of olive cultivar groves.

Among the sensitized population, aged 1 to 96 years, 35.5% corresponded to the 4-12 age group. The highest prevalence was found in the 13-20 age group (47.4%). The IgE levels were also higher in the groups 4-12 and 13-20 years old. The prevalence of positive olive sIgE also followed a biphasic pattern, reaching a nadir in 2017 – 2018, the years with the lower levels of olive pollen, followed by an increase between 2019 and 2021. The highest mean levels of sIgE were registered in 2013, 2018 and 2019 and the lowest in 2014 and 2020 seasons. When the mean levels of sIgE is considered, a 5-fold and a 2-fold increase were observed in 2021 compared to 2018 and to 2011, respectively, in keeping with the high pollen levels. When taken together, the profile of sIgE followed the profile of pollen index with the annual pollen index, evidencing an association between these variables.

In conclusion, these results suggest that the exposure to highest concentration of olive pollen favours the increase of sensitization of the population, particularly children. Other data should be analysed to validate the observations in this case study.

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Exposure to Airborne Alternaria Spores Under Different Bioclimatic Conditions Across Bavaria, Germany

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Abstract

Background: *Alternaria* sp. is a plant pathogen that can infect more than 4,000 plant species, making it responsible for 20 percent of agricultural production losses. In addition, the average sensitisation rate in Europe to *Alternaria* allergens is approximately 9%. A better understanding of the relationships between spore concentration at different altitudes or weather patterns can be applied to a more effective use of fungicides as well as to the improved diagnosis and treatment of spore-related respiratory allergic diseases. This study provides a regional assessment of the spatiotemporal abundance of airborne *Alternaria* spores in a variety of climatic and pollution regimes. We hypothesised that the regional land cover would the main predictor of the abundance of the potential spore sources.

Methods: In 2015, airborne *Alternaria* spores' abundances were examined at 23 locations in Bavaria, South Germany, using volumetric Hirst-type traps in the framework of 'ePIN testnetz' project and counts were made every two hours. Differences between bioclimatic zones in Bavaria were investigated. Full-factorial analysis of variance and covariance were applied (ANOVA, ANCOVA, post-hoc Bonferroni test), and regressive and explorative analyses when testing for additional and continuous co-factors with meteorological parameters, air pollutants and land uses.

Results: The total seasonal fungal index (SFI) of *Alternaria* spores varied considerably between locations. The cluster analysis showed five main groups, defined on the basis of the maximum concentration and monthly distribution of spore count. The first was characterised by the highest value in SFI but no longer season. This group included Bayreuth, Bamberg and Hof, sites located in the north. The diurnal pattern of *Alternaria* spores showed a peak in the middle of the day (10h to 16h), however, the average altered significantly during the day in different sites; e.g. in Augsburg, Bamberg, Hof and Gaissach there is a clear peak during the evening, or at early morning the stations at higher elevations near the Alps. Regarding the environmental drivers mostly influencing Alternaria spore concentrations, these were rather NO_2 and maximum temperature, than the hypothesised land uses.

Conclusion: Daily biological contamination due to *Alternaria* exceeded 100 spore/m^3 air for more than 30 days at most of the sites, what is considered a risk threshold for respiratory allergy, and correlated significantly with maximum temperature and NO_2. In a future world, warmer and more polluted, fungal spore allergy may also be an emerging allergy risk threat. This points out the need to include spores' monitoring in all the existing information networks.

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Species-level Identification of Cupressaceae Pollen-type Grains From Airborne Samples Using Indirect Techniques

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Abstract

Cupressaceae pollen type is increasing its relevance from the allergenic point of view because of the increase of its pollen load in Mediterranean cities. Pollen grains belonging to the Cupressaceae family (also including the Taxaceae and Taxodiaceae families) are morphologically undistinguishable under light microscopy. However, different species are characterised by a specific pollen size range. The diversity of the species of Cupressaceae and Taxaceae families, represented by native and exotic ornamental species, produces long intermittent pollen seasons whose pollen sources are difficult to identify. The pollen size, the pollination timing of the species and the distribution of sources in the city constitute indirect approaches for species attribution. The hypothesis to be tested in this study is that pollen size range significatively changes throughout the Cupressaceae/Taxaceae pollen season, and that the potential source can be indirectly assigned based on pollen size, phenology and distribution. We also intend to study local differences between two different sampling points.

Pollen size was measured for, i) pollen samples collected directly from Cupressaceae/Taxaceae families and, ii) airborne pollen samples collected using a Hirst-type volumetric trap following the standardised aerobiological method. In both cases, we measured at least 100 pollen grains for each sample, and discarde ruptured pollen grains. In the case of Cupressaceae/Taxaceae species, 8 different common native and exotic species in the natural vegetation and green urban spaces of central Spain were sampled. In the case of airborne samples, we selected one sample for each of the eight months in which the percentage of this pollen type is relevant in the air (October to May 2022-2023) using data from two pollen traps in the city of Madrid (central Spain). Polar and equatorial axes were measured, and the mean was used as an estimate of the pollen diameter. Also, high-resolution remotely sensed products provided by the Copernicus Land Monitoring Service at a 10-m resolution combined with the National Forest Inventory were used to characterise the potential sources.

The results showed significant statistical differences between samples throughout the pollen season and between sampling locations. Also, the pollen size measurements showed significant differences for the studied species. We observed changes in pollen size from October 2022 to May 2023, with the minimum size observed in December 2022. This minimum size matches with the size range of the species *Juniperus oxycedrus* L. (average size 26.1 ± 2.5 µm) which is abundant in the Mediterranean forests and shrublands surrounding the city of Madrid. Common ornamental species in Mediterranean cities such as *Cupressus sempervirens* L. and *Hesperocyparis arizonica* (Greene) Bartel (= *Cupressus arizonica* Greene) showed a larger pollen size (30.3 ± 2.1 and 33.2 ± 3.4 µm, respectively). These species would be the responsible sources

of the airborne pollen grains over a longer period during the Cupressaceae/Taxaceae pollen season. Our study based on indirect techniques provides a good estimate to analyse the origin of pollen types from diverse species.

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First Insights From Real-time Bioaerosol Measurements at the Sonnblick Observatory (3106m asl)

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Abstract

In this presentation we will discuss data from real-time measurements with a SwisensPoleno Jupiter during a long-term measurement campaign conducted at the Sonnblick Observatory together with the University of Vienna within the VINAR (Vienna Network on Atmospheric Research) framework. The campaign aimed at characterizing coarse mode particles at higher altitudes with special regard to bioaerosols (GeoSphere Austria), dusts and microplastics (University of Vienna). The analysis presented here is focused on bioaerosols.

Bioaerosols are a very diverse subgroup of atmospheric aerosols and are ubiquitous in our atmosphere. The motivation to study bioaerosols is manifold: they do not only affect human or plant health, but are also known to play a crucial role in cloud forming processes. Nevertheless, our knowledge about the temporal and spatial distribution of bioaerosols in the atmosphere is still clearly limited. Many health related measurements are offline and are prevalently conducted in populated areas. Especially measurements at higher altitudes, distant from local sources and in atmospheric regions where clouds actually form, are scarce.

The Sonnblick Observatory, a global GAW (Global Atmosphere Watch) station and part of the European infrastructure ACTRIS, is located at the main ridge of the Austrian alps at the top of Mt. Hoher Sonnblick and is freely exposed to all cardinal directions. From April to November 2023 the SwisensPoleno Jupiter owned by the University of Vienna was installed at the Sonnblick Observatory and continuous measurements were conducted. The SwisensPoleno Jupiter is a real-time aerosol monitor that obtains scattered light (used as trigger), two holographic images and fluorescence signals of single aerosol particles as they fly by. Fluorescence emissions are induced by three excitation light sources with the wavelengths of 280, 365 and 405nm and a spectrometer captures the emissions within five wavebands

(central wavelengths: 357, 435, 483, 562, 676 nm). The instrument settings were adjusted to detect particles between one and 150µm in size.

In a first step, time series of concentrations and fractions of fluorescent particles (as a proxy for bioaerosols) were analysed to investigate trends and identify periods of particular interest. Time series reveal a clear increase in concentrations and fractions of fluorescent particles as the season proceeds from early spring to summer (fluorescent fraction: 0.2 in April; 0.6 in June). In a second step, we took a deeper look at the fluorescent characteristics of the particles together with the holographic images. In a previous laboratory study we obtained representative fluorescence signals for three classes of bioaerosol particles: pollen, plant debris and fungal spores. We use these data to cluster and interpret the atmospheric measurements. One interesting finding of our analysis is that fungal spores are very frequently present at this altitude throughout the season. Combining these data with meteorological measurements and FLEXPART simulations we also discuss potential sources of bioaerosol and typical atmospheric conditions driving their presence.

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Modelling Grass Pollen Load in Temperate and Mediterranean European Areas Based on Source Spatial Distribution

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Abstract

Airborne grass pollen is one of the most important causes of pollen allergy worldwide. Still, the identification of its specific emission sources is a challenging task because of the ubiquitous nature of this pollen type. Previous studies have explored the contribution of the most abundant grass species and grassland habitats to airborne grass pollen at a local spatial scale using *in situ* phenological samplings and remote sensing data. This background allowed us to hypothesise that wild grasslands are a more important pollen emission source than human-managed areas dominated by grass species. The main aim of this study was to extend this hypothesis by modelling the grass pollen load measured by aerobiological stations in temperate and Mediterranean areas during the last two decades using remotely sensed land cover data at continental scale.

The mean Annual Pollen Integral (APIn) for stations in the Iberian Peninsula and Central Europe was calculated for a minimum of 10 years during the last two decades. Years with APIn values below the 10th percentile and above the 90th percentile for each station were removed to get a more centred mean and to avoid anomalous years. The potential sources considered in this study were derived from the continental-scale remotely sensed products provided by the Copernicus Land Monitoring Service. The combination of different products allowed us to characterise the spatial distribution of different grass-dominated habitats, namely: intensively managed grasslands (seasonally cut and tilled pastures and arable lands), wild grasslands (natural or semi-natural grasslands including Iberian "dehesa" systems). The relationship between airborne pollen and potential emission sources was established using the Concentric Ring Method which permits estimating the influence of emission sources based on the distance from the observation sites.

The results showed a clear dominance of the airborne pollen contribution from wild grasslands compared to human-managed grasslands and crops. Wild grassland distribution explained between 60-70% of the variance of the mean grass APIn, depending on the spatial resolution of the source map and the maximum influence distance considered. A detailed analysis of the emission influence and source distance of wild grasslands provided an influence mathematical function with maximum values of correlation of 0.75 (Spearman's test) between airborne pollen and source abundance. Conversely, intensively human-managed pastures and arable lands only explained about 10-20% of the variance of the mean grass APIn, obtaining a non-coherent influence function. These results constitute valuable evidence of the land-use and vegetation types with the major airborne pollen contribution of grasses at a large spatial scale. Our findings are the basis for comprehensively study the factors involved in the long-term pollen trends of grasses by information of the land use and land cover changes for the last decades in Europe. Also, the influence function for the most important pollen sources can be combined with short-term meteorological data to develop a forecasting model of the interannual APIn of grasses.

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Pollen Production Assessed in International Phenological Gardens During 2019-2023 Reveals Potential Impacts of Climate Change

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Abstract

Allergy is the most common chronic disease in Europe and poses a major risk to human health. Arising from the complex interplay between the environment and genetics, this problem may become even worse: Climate-change-related effects have already been observed in the timing and duration of the pollen season, airborne pollen concentration and pollen production, pollen allergenicity, and plant and pollen distribution.

This study examines how environmental factors impact the pollen production of *Betula pubescens* Ehrh. (downy birch) across Europe. By examining approximately 1,000 male inflorescences of downy birch clones, from 2019 to 2023, in 37 International Phenological Gardens spanning a wide geographic range in Europe, we conducted a thorough examination of reproductive metrics such as inflorescence numbers and pollen quantities. We analyzed the effects of meteorological factors such as average air temperature, precipitation levels, as well as O_3 and CO_2 concentrations. We explored a set of different model specifications using regression techniques and evaluated potential masting effects.

Substantial geographic variations were observed in average pollen production, ranging from 1.3 (2022) to 2.5 million pollen grains per catkin (2021). Regression analyses highlighted the importance of the previous summer's average temperature and ozone concentrations on pollen production per catkin. Whereas warmer conditions in summer aligned with higher pollen production, O_3 levels were related with opposed effects. Moreover, the number of catkins was *inter alia* positively influenced by the average temperature and precipitation of the previous summer.

Our findings imply that changing climatic conditions may increase birch pollen production, which, in turn, will pose a short- and mid-term threat to allergic individuals, with mitigation strategies urgently needed to lower the climate change impact.

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Assessing the Environmental Risk Posed by the Allergenicity of Urban Green Spaces in Temperate Latitudes

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Abstract

Introduction

Airborne pollen significantly impacts respiratory health, and anthropogenic climate change is altering pollen season intensity, timing, and duration. Monitoring pollen dispersal and the spatial distribution of sources is increasingly vital for public health, especially regarding ornamental trees in cities, often selected for aesthetics without considering allergenicity. Species like Betula sp., Alnus sp., Corylus sp., and Fraxinus sp., who are common in European parks pose a risk in urban green areas, potentially triggering severe allergies. In urban green areas, there is a concentrated number of trees and, depending on their composition, they might cause severe symptoms in allergy sufferers.

It is therefore important to evaluate the allergenicity of urban green spaces to identify potential sources of risks for allergy sufferers and to formulate suggestions for urban planning. To determine the allergenicity of an urban park (Westfriedhof in Augsburg, Germany) the urban green zone allergenicity index (IUGZA) was applied and adapted to the characteristics of temperate latitudes.

Methods

The IUGZA, developed in Mediterranean areas, was used to determine the allergenicity of Westfriedhof, Germany.

Tree locations and physical characteristics were provided by two different data sets: (1) Augsburg Official Tree Register and (2) detailed locations of birch trees in the city, carried out in a previous study.

The parameters necessary for the index calculation, allergenic potential (ap), the duration of pollination (dpp), pollination strategy (ps) and maturity height tree (Hi), were obtained through literature or on-site measurements for Augsburg. The area that is occupied by each species in the park (Si) was calculated with a method based on allometric analysis for urban trees.

Subsequently, the index was evaluated with pollen data concentration by Burkard portable spore trap during the main pollination period in the study area.

Results

There are 1427 trees (66 species) with 74.7 trees per hectare in Westfriedhof. The most allergenic trees in the study area (Betula sp., Corylus sp., Fagus sp., Carpinus sp., Fraxinus sp.) accounted for 35% of the trees in the park. The most abundant species were Tilia sp. (17%), Fagus sp. (15%) and Carpinus sp. (11%).

The values for both dpp and ap were categorized into groups. These were adjusted due to different flowering periods and different allergenicities within Germany compared to Mediterranean areas. According to the calculation with the IUGZA, the allergenicity risk of the Westfriedhof assumes a value of 0.44 due to only the arboreal stratum.

Discussion and Conclusion

According to the $I_{UGZA,}$ the allergenic potential seems high (> 0.3), though some parameters were adjusted for southern Germany. High allergenicity was expected due to some highly allergenic trees, yet most have moderate allergy. Other factors, like different pollination periods, shrub layer, grass presence, and long-distance transport, were not considered. Adjusting allergenicity and flowering period is necessary, and tree height should be included in calculations, not just maximum height.

Implementing IUGZA in temperate parks highlights the need for additional parameters to explain allergenicity risks. Consideration of increased pollen potency, population sensitization, and cross-reactivity due to climate change is essential.

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Sylva-DNA-classifier: Taxonomic Classification and Visualization of Large-scale Metagenomic Sequences Produced With Oxford Nanopore Sequencing Technologies

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Abstract

Environmental metagenomic air samples contain large amount of information that can be unraveled with a suitable toolkit. Among numerous objectives, taxonomic identification and relative abundance estimation are one of the key points of interest in a metagenomic DNA analysis. This type of analysis for large sequence datasets requires considerable computing resources with the publicly available bioinformatic software. These programs are optimized for analysis of the next-generation sequencing data and often target only bacteria. The SYLVA-DNA-classifier is a pipeline developed to efficiently and accurately classify large quantities of the 3rd generation sequencing data across all kingdoms, with a special focus on plants and fungi. The pipeline contains three stages. Firstly, the sequenced data go through preprocessing to meet the quality requirements. Secondly, the data undergo taxonomic classification and abundance estimation. Finally, postprocessing steps include production of time series, removal of sequence data identified as human, creation of graphical presentation of the data, and submission of the sequence data to a European sequence database (ENA).

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Pollen and Spore Dispersal in a Complex Terrain Assessed With Experiments and a Small-scale Climate Model

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Abstract

Atmospheric dispersion models play a central role in predicting pollen concentrations, especially at a regional or global scale. However, their effectiveness in complex topographic

and small-scaled areas is limited. The aim of this study was to experimentally investigate the spatial and temporal dispersion of spores and pollen and evaluate their integration into a simulation model.

The KLAM_21 model, typically used to analyse cold air flows in orographically structured terrain, was employed for this purpose. In the field, experiments were conducted in autumn 2020, as well as in summer 2021 and 2022, in a small side valley located near Eichstätt, Germany, to validate the simulated wind speeds and obtain information on dilution effects. On nights favourable for cold air flows, bioaerosols (Lycopodium spores and *Fraxinus excelsior* pollen) were released with a self-built apparatus at a constant flow rate and quantified downslope with portable volumetric air samplers with a temporal resolution ranging from 30 sec to 10 min within one hour. Wind measurements were carried out with ultrasonic anemometers and mobile weather stations.

The simulation results provided spatial and temporal wind information, revealing a clear confluence of cold air outflows on the valley floor with average wind speeds of approx. 1.4 m/s. In contrast, the wind measurements in the field showed slightly lower average wind speeds for each experiment. The analysis revealed a decrease in bioaerosol concentration with increasing distance from the release source, observed both in the field trials and simulation results, although the simulation overestimated the continuous tracer release. The information on wind speed derived from pollen transport was also overestimated by the simulation but matched well with the measured wind speeds in the field.

It can therefore be summarized that the integration of pollen data in KLAM_21 helps characterizing cold air movements in a complex terrain. However, it is crucial to further improve the homogeneity of pollen release. This approach is promising for analysing the impact of cold air flows on pollen transport, particularly relevant for populations in areas prone to cold air flow and accumulation.

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Advancing Dna Extraction Techniques for Atmospheric Bioaerosol Samples: Evaluating Commercial Kits and Purification Strategies

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Abstract

We present a DNA extraction protocol for bioaerosols that produces high-quality molecules suitable for the third generation sequencing and other applications. We developed this protocol for glass-fiber filters, widely used for a range of purposes from atmospheric monitoring to air purifiers. The protocol combines multiple extraction methods coupled with SPRI paramagnetic bead technology AMpure XP developed by Beckman Coulter for rapid and efficient DNA purification. Compared to phenol-chloroform-based protocols, our protocol does not require the use of these hazardous chemicals and has proven to be more efficient than some of the column-based commercial kits recommended by their providers. The protocol has proven to be effective for samples with relatively high biological yield – to the filter mass ratio, as detected with traditional phenol-chloroform-based method, being >70 nanograms of potential DNA on the filter to one milligram of the glass fiber filter. For the ratio lower than 15 nanograms per a milligram of filter, the protocol fails to provide sufficient amount of DNA for sequencing as a result of the extraction. As a result for high-yield-filter sample DNA extraction, we succeeded extracting an acceptable quality of DNA suitable for sequencing with the 3rd generation Oxford Nanopore technologies.

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Can the Characteristics of the *Betula* Pollen Season Be Similarly Represented by Two Nearby Pollen Stations?

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Abstract

Can the characteristics of the ***Betula*** pollen season be similarly represented by two nearby pollen stations?

Jasmin Meixner, Johanna Jetschni & Susanne Jochner-Oette

Background

In the context of climate change, increasing urbanization and the potential exacerbation of allergic diseases, heightened attention on fundamental research regarding aerobiological processes in urban areas is necessary. This study therefore focused on differences and influential factors on *Betula* pollen concentrations at two nearby urban pollen stations.

Material and Methods

Betula pollen were sampled using Hirst-type volumetric pollen traps (Burkard Manufacturing Co Ltd.) during 2019 and 2022 in Eichstätt (EI) and Ingolstadt (IN; distance approx. 25 km), Bavaria, Germany. To determine the characteristics of the pollen season, start and end dates as well as peak dates, peak counts, and the seasonal pollen integral (SPln) of the *Betula* pollen season have been identified. Additionally, hourly meteorological data were collected at ground level. In Ingolstadt, we additionally measured wind speed and wind direction at roof level. The following parameters have been processed for further investigation: daily mean temperature T_{mean}, daily maximum temperature T_{max}, daily mean relative humidity RH_{mean} and daily precipitation sum p_{sum}. The data obtained were used to carry out Spearman correlation analyses and we tested for differences between the two sites using Mann-Whitney U test.

Results

The comparison of the *Betula* pollen season characteristics in Eichstätt showed only minor variations regarding the beginning of the pollen season, whereas the end of the pollen season and thus the length varied between 16 and 35 days. The characteristic of the pollen season in Ingolstadt showed only marginal deviations from those in Eichstätt, but was linked to a considerably higher peak value and SPIn in 2020.

The considered main meteorological parameters varied only slightly between the two sites in spring (MAM). Notably, moderate positive relationships have been found for T_{mean} (EI: $r_s = 0.396$, $p < 0.001$; IN: $r_s = 0.345$, $p = 0.003$) and T_{max} (EI: $r_s = 0.410$, $p < 0.001$; IN: $r_s = 0.390$, $p < 0.001$) and negative relationships for RH_{mean} (EI: $r_s = - 0.424$, $p < 0.001$; IN: $r_s = - 0.307$, $p = 0.010$).

Conclusions

Since pollen traps from monitoring stations are installed at a minimum height of 10 meters above ground level, they should represent the background pollen concentration of a larger region. Our study has shown that differences between the two stations exist only in some cases, primarily concerning the length of the pollen season and the SPln and peak values.

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Bioaerosols in the Atmosphere at Two Sites in Northern Europe in Spring 2021: Results of an Experimental Campaign

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Abstract

During spring 2021, a coordinated observational and modelling campaign targeting biogenic aerosols in the air was performed at two locations in Northern Europe: Helsinki, Finland, and Siauliai, Lithuania. The campaign covered the transition of atmospheric biogenic aerosols profile from winter to summer, from March 1 till mid-May in Siauliai and from 12 March till end of May in Helsinki. The observations included core-program samplers and daily and daytime filter collectors. The core of the program was based on 2- and 2.4--hourly sampling in Helsinki and Siauliai, respectively, with sticky slides (Hirst 24-h trap in Helsinki, Rapid-E slides in Siauliai). The slides were subsequently processed extracting the DNA from the collected aerosols, which was further sequenced using the 3-rd generation sequencing technology, Oxford Nanopore Technology. The hourly aerosol concentrations at the Helsinki monitoring site were obtained with a Poleno flow cytometer, which could recognize some of the aerosol types.

The DNA collection from air by the applied techniques was confirmed feasible: all but one delivered sufficient amount of DNA for the following analysis, in over 40% of the cases the amount of extracted DNA was sufficient for sequencing without the PCR step. The campaign DNA analysis produced the first high-resolution dataset of bioaerosol composition in the North-European spring. It also highlighted the deficiency of generic DNA databases in applications to atmospheric biota: about 40% of samples were not identified with standard bioinformatic tools.

After optimization of the bioinformatic analysis protocol and building of custom sequence databases for plants and fungi, the amount of unidentified sequences decreased down to ~20% in average. However, large variation in unidentified sequences was observed between the samples. The general bioaerosol profiles were determined for the duration of the transition period from winter to spring for both locations, Helsinki and Siauliai. The biogenic background coming from the ventilation of the buildings in a city was observed to be a more significant factor in error source affecting the analysis than estimated in the initial report.

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Ambient Pollen Exposure and Allergy Symptom Severity

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Abstract

Background: Ambient pollen exposure causes nasal, ocular and pulmonary symptoms in allergic individuals, but the shape of the exposure-response association is not well characterized. We evaluated this association and determined (1) whether symptom severity differs between sub-populations; (2) how the association changes over the course of the pollen season; (3) which pollen exposure time lags affect symptoms.

Methods: Adult study participants (n=396) repeatedly scored severity of nasal, ocular and pulmonary allergic symptoms, resulting in three composite symptom scores. Pollen concentration data were provided by the Federal Office of Meteorology and Climatology (MeteoSwiss) for the Basel site, which was centrally located in the study area at a median distance of 2.3 km to participants' homes (interquartile range 1.1-4.8 km). We calculated hourly individually-relevant pollen exposure to seven allergenic plants (alder, ash, birch, hazel, grasses, mugwort, ragweed) considering personal sensitization (based on skin prick tests for the same seven plants) and exposure time lags of up to 96 hours. We fitted generalized additive mixed models, with a random personal intercept, adjusting for weather and air pollution as potential time-varying confounders.

Results: We identified a clear non-linear positive association between pollen exposure and ocular and nasal symptom severity in participants with a self-reported pollen allergy, but not in those without. The exposure-response curve was clearly non-linear: symptom severity increased steeply with increasing exposure initially, but attenuated beyond approximately 80 pollen/m3. We found no evidence of an exposure threshold, below which no symptoms occur. While recent pollen exposure in the last approximately five hours affected symptoms most, associations lingered for up to 60 hours. Grass pollen exposure (compared to tree pollen) and younger age (18-30 years, as opposed to 30-65 years) were both associated with higher nasal and ocular symptom severity.

Conclusions: Our findings are of interest for pollen warning systems. Considering the lack of evidence for a threshold, below which no symptoms are observed in the allergic population, warning systems may reserve the colour green (suggesting a “safe” level) for "no pollen" situations only. In addition, differentiating pollen concentration categories beyond approximately 80 pollen/m^3, where symptom severity increases more slowly with increasing pollen exposures, should be done with caution to ensure the categories reflect the allergy sufferers’ experience. While the greatest health impact of pollen exposure is acute (within ~5 hours after exposure), our findings suggest that effects persist over multiple days. Therefore,

warning systems may consider the inclusion of multi-day (lagged) pollen concentrations in the future.

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Influence of Anthropogenic Air Pollution on the Air Resistome in Central Spitsbergen

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Abstract

The emergence of antimicrobial resistance poses a challenge in infection treatment and represents one of the paramount threats to humanity in the 21st century. Numerous studies encompassing all clinically important antibiotic resistance genes (ARGs) in pathogenic and commensal bacteria in various environments have been conducted. However, detailed investigations concerning outdoor air resistome, particularly in polar regions, remain sparse.

This study aimed to characterize polar air resistome through quantitative analysis of selected ARGs. Our hypothesis posits that ARGs are present in polar air, with their abundance being correlated to anthropogenic air pollution level. The study area was Spitsbergen (Svalbard Archipelago), where increasing anthropopressure is a factor contributing to negative environmental changes. Polar regions, characterized by their remoteness from densely populated areas and non-complex ecosystems, provide a unique setting to explore how human activities affect the resistome gene pool in these environments.

Total DNA was isolated from air samples collected in Pyramiden, a site heavily influenced by human activities such as tourism and abandoned coal mines, as well as near Petunia Bay (similar meteorological conditions as in Pyramiden, but without anthropogenic impact) and the Sven and Ferdinand glaciers. It was followed by screening for molecular determinants of antibiotic resistance, including *intI1*, bla_{TEM}, $bla_{OXA\text{-}1}$, *tetA*, and *tetM* genes, using conventional PCR techniques. Quantitative analysis of class 1 integron integrase (*intI1*) and selected ARGs (bla_{TEM}, $bla_{OXA\text{-}1}$, *tetA*, and *tetM*) was carried out by using digital PCR. Moreover, we measured air pollution indicators: PM1, PM2.5, and PM10 particulate matter as well as formaldehyde (HCHO), total volatile organic compounds (TVOC), temperature, and humidity by using a portable sensor, to assess correlation between air physicochemical parameters and the level of ARGs.

The *intI1* gene and ARGs were detected in all sampling locations, with concentrations ranging from 1.7×10^{1} to 1.7×10^{2} copies/m^3. Principal Component Analysis (PCA) revealed that the primary

variables accounting for variability were related to air pollution and the concentration of *bla*$_{TEM}$ and *intI1* genes, with the highest concentrations observed in samples collected in Pyramiden. Air pollution indicators, such as PM2.5 concentration, were also higher in Pyramiden (12.59 μg/m^3) compared to the vicinity of the glacier and Petunia Bay (10.42 μg/m^3 and 10.39 μg/m^3, respectively). Those differences were statistically significant.

In conclusion, all analyzed genes were detected across the three sampling locations, which suggests the pervasiveness of antibiotic resistance genes in the Arctic atmosphere. Higher concentrations of *bla*$_{TEM}$ and *intI1* genes in Pyramiden, coupled with elevated PM2.5 levels, suggest that anthropogenic activity shapes the air resistome of this remote environment.

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Historical Overview of the Presence of Common Ragweed Airborne Pollen Since 2000 in Northern Italy

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Abstract

The aim of this study is to investigate the spatial and temporal changes of Ambrosia pollen in the geographical area of Northern Italy during the last 23 years. Ambrosia sp., also referred to as ragweed, is considered an invasive alien species in Europe. Originally native to North America,

this annual plant prefers mostly anthropogenic habitats and can tolerate different climatic conditions. This geographical variety, from habitats in the Po Plain to alpine locations, characterizes the regions of Northern Italy, considered in this study. In addition to its broad ecological niche, the common ragweed causes health, environmental, and agronomical problems in many parts of the world. To evaluate the potential risk to human health due to the exposure levels to this highly allergenic pollen, investigating the change in pollen concentrations over time can be relevant. Furthermore, this study gains insight into the trend of the concentration of ragweed pollen during the new century in Italy.

Considering the time interval 2000-2023, Ambrosia pollen data were collected at 34 aerobiological monitoring sites in North Italy; all sites had at least 12 years of daily pollen concentration (P*m-3) data. 31 out of 34 monitoring sites belong to the Italian pollen network POLLnet. All stations performed the aerobiological monitoring using a volumetric Hirst sampler and adopting a common method consistent with UNI EN 16868:2019 EU standard procedure. The dataset has been checked for completeness. Referring to the flowering period of Ambrosia (i.e., July-October), 3% of the yearly data series were excluded since the completeness was lower than 80%.

Ambrosia pollen concentrations were analyzed to verify if changes occurred in the study area regarding pollen amounts (i.e., Seasonal Pollen Integral, SPIn, the sum of daily concentrations in the July-October period) and seasonality. Firstly, considering the SPIn values averaged over the entire 2000-2023 study period, results show a large spatial variability, from the lowest values in Brunico (BZ) (9 P*m-3*day) to the highest values in Magenta (MI) site (3667 P*m-3*day). This confirms previous results, evidencing in the Milan area mean SPIn values above 2000 P*m-3*day, while the seven stations in the Alpine and pre-Alpine area display on average values below 100 P*m-3*day; eastern stations show intermediate values ranging from 100 to 500 P*m-3*day. Referring to the first occurrence in Italy of Ophraella communa verified in 2013, the comparison (Mann-Whitney U Test) of mean APIn values registered before (i.e., in 2000-2012 period) and after (i.e., 2013-2023 period) displays a significant decrease only for Legnano, Magenta, Rho, Piacenza ($p<0.001$), Omegna, Novara ($p<0.01$), and Bolzano ($p<0.05$). Further analyses will include the pollen seasonality and the spatial distribution with thematic map production.

Considering the spatial and temporal variation of Ambrosia pollen amounts, it is of crucial importance to continuously monitor this invasive plant and its pollen, for human health and environment protection, in a large and inclusive One Health perspective. The findings of this study can build the base to improve the current management measures, by adding new ones, which are increasingly comprehensive and can be used as supportive information for decision-makers for future handling steps of this invasive species.

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Bioaerosol Flow Cytometry in the Atmosphere in Finland

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Abstract

Bioaerosol flow cytometry as a technique to analyse biological particles suspended in the air has recently taken a significant leap forward with the release of state-of-the-art automated real-time monitors. The current study presents results acquired with one of these devices, Swisens Poleno Jupiter. There are currently two monitoring stations in Finland equipped with Poleno Jupiters located in Helsinki at the Finnish Meteorological Institute's rooftop and about 200 km north of the Arctic Circle in Lapland in Pallas Atmosphere-Ecosystem Supersite on the top of Sammaltunturi.

The device facilitates in-flight measurements of particle shape, size, and fluorescence, encompassing real-time bioaerosol identification. The working principle of Swisens Poleno is as follows: laser light scattering triggers the measurement, providing an initial estimate of particle size, velocity, and alignment by combining information from two trigger lasers. Following the trigger, two focused images, 90° apart, are reconstructed using digital holography, and UV-induced fluorescence provides information about the particle composition. Finally, the optical polarization characteristics of the particle are measured before it exits the device. Our recognition algorithm utilises a Convolutional Neural Network (CNN) model originated from the one proposed by Sauvageat et al. (2020). It was trained on the dataset generated by providing the collection of pollen typical for Southern Finland to the device in laboratory conditions. The algorithm demonstrated remarkable performance, achieving an overall accuracy of nearly 90%. Most misclassified events belonged to the same family, which is understandable given the close resemblance between certain pollen grains, such as those of Betula and Corylus.

Transitioning to real atmospheric conditions revealed new challenges. The primary challenge was the occurrence of false positives, which was partially mitigated by incorporating fluorescence and polarization signals. The recognition algorithm was assessed in the EUMETNET AutoPollen Intercomparison Campaign, where its performance was on par with two other top-performing algorithms (Maya-Manzano et al., 2023). In this study, we present the comparison results to manually operated Hirst-type pollen trap data for the 2023 season in Finland. The correlation between hourly pollen concentrations measured by Poleno and the Hirst-type trap pushed close to 90% for the main pollen species, thus being consistent with the laboratory scores.

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Pollen Allergenicity Variations Within the City - Potential Effects of Pollution Pockets?

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Abstract

Air pollution aggravates asthma and respiratory allergies evoking higher incidence and/or symptoms worsening in heavily polluted areas. In urban areas, road traffic is one of the major sources of air pollutants, including vehicle exhausts and secondary pollutants formed in the atmosphere. Dispersion of pollutants originated from traffic is directly connected with the spatial configuration of the urban area and traffic conditions, as well as with meteorology.

The *Platanus hispanica* is a very disseminated tree in man-inhabited spaces, both rural and urban, specially creating shade areas on traffic roads and gardens. Pollen from these trees, dispersed in early spring, contacts closely with air gaseous pollutants generated by automobile traffic, such as nitrogen oxides and ozone, whose effects on its allergenic properties are still unclear.

The objective of this work was to evaluate allergenicity in *P. hispanica* pollen from different locations within Madrid city in relation to local air pollutant levels.

Pollen was harvested in 2019, from similar size *P. hispanica* trees in four locations in the city of Madrid, selected based on Air Quality Monitoring Stations (AQMS) proximity (namely *Retiro*, *Pz.Eliptica*, *Ens. Vallecas* and *M.Alvaro stations).* Daily NO, NO_2 and O_3 concentrations were obtained from https://airedemadrid.madrid.es/portal/site/calidadaire and were compared between stations, in the previous month to pollen harvest (21 feb – 21 mar 2019).

For each location the pollen from different trees was pooled, to create a representative sample. Pollen protein extracts were prepared: a) in bicarbonate buffered solution, followed by samples lyophilization for ELISA measurements; b) in phosphate buffered solution for western blot. Total protein was quantified by the BCA method. Pla a 1 allergen was quantified by specific ELISA. Western blot was used to compare pollen Pla a 1 relative content according to location and immunoblot using pooled sera from allergic individuals was used for IgE recognition patterns.

NO and NO_2 levels were higher in *Pz.Elíptica*, reaching a maximum of 111 and 108 μg/m^3, respectively. *Retiro* station registered the lower values for both pollutants (max. 30 and 61 μg/m^3, respectively). Daily mean O_3 concentration varied between 20-80, 20-68 and 16-65 μg/m^3 in *Retiro*, *Ens. Vallecas* and *Pz.Elíptica*, commonly higher in the *Retiro* station during the whole period.

Pla a 1 concentration varied between 1.2±0.2 and 2.9±0.2 μg Pla a 1/mg protein and was significantly different in *Retiro* (higher O_3 levels) and *Pz Eliptica* (higher NOx levels). Semi-quantitative analysis by western-blot revealed an equivalent profile.

IgE-recognition pattern showed several bands differentially detected in the four locations, with band 52.3±0.5 more intense in *Retiro*, 40.8±0.4, 33.8±0.3 and 27.8±0.3 kDa more intense in *Pz.Elíptica*, 49.7±0.5 and 47.6±0.3 kDa more intense in *Ens. Vallecas* and 25.6±0.3 kDa and 61.4±0.8 kDa more intense in *M.Alvaro*.

Taken together, these results show that pollution pockets with distinct characteristics could be found within Madrid city influencing pollen specific allergenic profiles. A better understanding of the effects of environmental pollutants on the allergenic characteristics of pollen is essential to understand the phenomenon of increased respiratory allergy incidence in urban spaces.

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The Allergenic Potential of Green Urban Areas in the Macaronesian Islands: The Case of Funchal City (Madeira)

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Abstract

Background: More than half of the world's population lives in cities and a considerable proportion is exposed to several allergenic pollen types. The plant cover composing the urban ecosystems may comprise allergenic plants because of unsuitable green space planning. Past and recent urban green designs relied mostly on aesthetic and management criteria. Public gardens are elements that constitute the urban green infrastructure. Such units provide ecosystem services that are becoming a key concern in urban planning strategies and policies.

Aim: The study aims to determine the overall allergenic potential of two highly visited public gardens in Funchal and consider potential allergenic risk scenarios with the calculate respective Index of Urban Green Zone Allergenicity.

Methods: The parks were visited between March and August of 2021 and the data on the number of plants, covered area and taxonomic identification in each park was recorded.

Biodiversity and the resilience of ecosystems were assessed by application of Santamour rule. To estimate the potential allergenicity of both urban parks, a quantitative index, which estimate the allergenicity of urban green spaces in general (IUGZA, Index of Urban Green Zone Allergenicity) was calculated.

Results: A total of 173 plant species belonging to 63 different families were recorded in the Municipal Garden, whereas in Santa Catarina Park 210 species were inventoried, corresponding to 68 plant families. About 93% and 86% of the species from Municipal Garden and Santa Catarina Park, respectively, presented null or low allergenic potential. From the inventory carried in the 2 green areas, it was possible to identified 6 additional species that belong to the National List of Invasive Species. 2 herbaceous – *Phytolacca americana* (Phytolaccaceae), *Zantedeschia aethiopica* (Araceae); 3 shrubs – *Agave americana* (Agavaceae), *Crassula ovata* (Crassulaceae), *Lantana camara* (Verbenaceae), and 1 tree – *Pittosporum undulatum* (Pittosporaceae).

Conlusions: An IUGZA is a managing tool for pollen allergen emissions in urban green spaces. Higher species diversity in sub-tropical urban green areas seems to reduce their allergenicity indexes. Species with higher potential allergenic values tended to exhibit low representativeness and occupied surfaces. The meteorological parameters (such as temperature, precipitation, etc.) should be included in future studies given their influence on biological parameters that define the IUGZA value.

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Multi-Comparison of Data From Automatic Pollen Measuring Devices for Winter-Early Spring Flowering Taxa

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Abstract

Background: During the last decades, airborne pollen concentrations have been continuously monitored worldwide by using the Hirst-volumetric methodology. It requires highly skilled staff, and it is a very time-consuming task overall for optical microscopy analysis. Furthermore, the traditional weekly change routine produces a significant temporal delay in having the actual data. It is clear why there is a high necessity for newly validated automatic aerobiological methodologies to solve those problems. The homogenization validation of pollen data from automatic systems with the visual determined from Hirst traps will allow us to compare results and maintain historical time series. This work aims to compare airborne pollen concentrations provided by the Hirst method and three different automatic systems. We aim to calculate the scaling factors needed to produce intercomparable concentrations.

Methods: We compared both daily and three-hourly data of winter-early spring taxa: *Cupressus*, *Fraxinus*, Urticaceae, and total pollen concentration, detected with a Hirst trap with those obtained from three different automatic devices: Swisens Poleno Jupiter, Hund BAA-502, and automated real-time pollen sensor (APS-300, Pollen Sense LLC). The study was carried out from December 2023 to March 2024 in Córdoba (Southern Spain) under Mediterranean conditions. The automatic devices are based on different techniques, such as image recognition, fluorescence spectroscopy, or digital holography. We calculated the ratio among Hirst and automatic measurements and homogenization of scaling factors to compare with Hirst data as the reference method. We also calculated the correlation coefficients (r) between Hirst and automatic devices.

Results: *Cupressus* and *Fraxinus* showed significant intra-seasonal correlation values between the automatic devices and Hirst. The significant correlation coefficient (r) for APS-300 data with r = 0.94 for *Cupressus*, r = 0.63 for *Urtica*, and r = 0.52 for *Fraxinus*; the BAA-502 data revealed r = 0.87 for *Cupressus*, and r = 0.1 for both *Fraxinus* and *Urticaceae*. In the case of SwisensPoleno, the statistics were lower. The average scaling factor for APS-300 was 1.11, and for BAA-502 was 1.02, indicating that both devices provided comparable concentrations. In the case of Swisens Poleno, it was 12.26 on average. A significant correlation was observed in the APS-300 but not in the BAA-502 or SwisensPoleno for *Urtica* pollen.

Conclusion: This research study is the first of its kind in a Mediterranean environment, presenting preliminary findings. Automatic pollen devices identify pollen concentrations in real-time data with stable measurements. As we just compared the pre-load commercial algorithms, this study shows the potential results of the three tested automatic systems. We observe similar concentrations provided by Hirst and both APS-300 and BAA-502, neglecting the use of scaling factors. When comparing samplers, we do not necessarily get similar concentrations due to the different measuring methods, but a similar distribution curve is noted, which is necessary for the application of a homogenization index. The results observed are expected with the commercial global algorithms, but further studies are required to investigate the performance in more detail after local classification algorithms are developed.

Keywords: Aerobiology; Automated monitoring; Validation; Intercomparison campaign; Pollen; Real-time

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Real-Time Sporangia Assessment of *Phytophthora Infestans* and *Alternaria* spp.

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Abstract

Many plant pathogen agents spread by airborne inoculum (i.e. spores, sporangia). This airborne transmission is highly efficient, allowing nearby and even distant host plants to become infected under favourable weather conditions. For example, late blight (*Phytophthora infestans*) and early blight (*Alternaria* spp.) can jeopardize potato yields by this mode of transmission. In Switzerland, efforts to control potato late blight (*P. infestans*) involve up to 10 fungicide applications annually, making potatoes one of the most treated arable crops. With increasing demands for sustainable agriculture and reduced pesticide use, precision agriculture tools, including decision support systems (DSS), are gaining importance. We assume that real-time monitoring of airborne sporangia can significantly enhance infection risk assessments, leading to optimised fungicide usage. In this work, we trained and validated an AI-based classifier for *P. infestans, A. alternata*, and *A. solani* identification, using the SwisensPoleno Jupiter, an airflow cytometer designed by Swisens AG for bioaerosol monitoring. This instrument uses holographic imaging and fluorescence spectroscopy to characterize single aerosol particles. Initial laboratory tests involved artificial infections of potato leaves with late and early blight to generate training data for the algorithm. The SwisensPoleno Jupiter detected and measured sporangia, producing 35 datasets with up to 12'904 particles in total for algorithm training and testing. Our experimental evaluation shows that SwisensPoleno Jupiter combined with the AI algorithm is highly effective in classifying spores, with an accuracy of over 90% for *A. alternata*, *A. solani* and *P. infestans*. Field trials were conducted near potato fields in Zurich-Affoltern and Zollikofen during the seasons 2022 and 2023, complemented by a Burkard® spore trap for comparison. Microscopic analysis identified high sporangia concentrations correlating with late blight outbreaks. Ongoing analysis aims to correlate these data with symptom observations, weather conditions, DSS PhytoPRE risk assessments, and SwisensPoleno data. The project goal is to evaluate real-time sporangia resp. spores monitoring to improve infection risk assessments and enable more targeted plant protection strategies.

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Real-Time Monitoring of Downy and Powdery Mildew Spores for Improvement of Diseases Risk Models in Viticulture

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Abstract

Despite occupying only 3% of Europe's agricultural land, viticulture accounts for 40% of its pesticide use. Fungal and fungal-like diseases such as powdery (*Erysiphe necator*) and downy mildew (*Plasmopara viticola*) can cause drastic yield and quality losses and require frequent fungicide treatments. The timing of fungicide application is crucial for successful disease control. Therefore, vine growers need precise disease pressure forecasting models to optimize and minimize overall fungicide use. These models are currently based on meteorological measurements and estimation of biological data like airborne fungal spores' concentration. The goal of this work is to provide measured ambient spores concentrations in real-time. For this purpose, we use an automatic air-flow cytometer developed by Swisens AG (SwisensPoleno Jupiter) for the detection and quantification of *E. necator* and *P. viticola* spores (fungi reproductive unit) in ambient air. The instrument identifies each aerosol particle using holographic imaging and fluorescence spectroscopy combined with AI-based post processing. The approach was validated by performing an extensive evaluation in both laboratory and in the field. In a first step, we performed laboratory measurements to obtain training data for machine learning, using naturally and artificially downy and powdery mildew infected leaves. Datasets from *E. necator* and *P. viticola* containing 8570 and 4942 potential particles related to downy and powdery mildew could be generated with SwisensPoleno Jupiter. To differentiate and classify fungal spores measured by SwisensPoleno, we trained a multimodal Convolutional Neural Network. It resulted in 90% of correct classifications for *E. necator* and 93% for *P. viticola*. In addition to lab trials, we installed four SwisensPoleno in Changins college's vineyard in Nyon, Switzerland, for the 2023 season. To better understand the spore flying dynamics within a vineyard, the instruments were placed in 2 different positions (stations) with 2 different inlet heights. Additionally, a 7-day recording volumetric spore trap (Burkard®) was used as reference to determine the spore concentration. Manual disease assessment based on visual symptoms was performed two times during the season on vines close to the spore traps. Manual concentration assessment during the first two weeks of August 2023, with the Burkard spore trap showed similar values for spores' concentration of *P. viticola* (33 and 26 spores/m^3/day, respectively). For *E. necator,* the second week resulted in a decrease of spores' concentration (26 versus 13 spores/m^3/day). In the last week of August 2023, we observed that 47% and 21% (station 1) and 29% and 53% (station 2) of the leaves showed downy or powdery mildew symptoms respectively. Analysis and comparison of Burkard spore trap, symptom observations and fungal spore concentrations by SwisensPoleno Jupiter is under investigation. We believe

that by incorporating real-time spore concentrations, forecasting models can be improved, leading to greater confidence and ultimately more targeted and reduced fungicide use in viticulture.

The project is supported by the Swiss Confederation (State Secretariat for Education Research and Innovation SERI and the Federal Office for Agriculture FOAG) and the Chips Joint Undertaking and its members, including the top-up funding by Sweden, Czechia, Finland, Ireland, Italy, Latvia, Netherlands, Norway, Poland, Spain.

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Co-Exposure of Nasal Epithelial Cells to Different Bioaerosols at the Air-Liquid Interface

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Abstract

Background: We had previously reported that pollen pre-exposure increases the susceptibility of respiratory epithelial cells to rhinovirus infection. In a large-scale data analysis study on pollen and COVID-19 cases across the world, we observed that SARS-CoV-2 infections were positively and significantly correlated with weather parameters and airborne pollen concentrations. In view of the constant spread of new viral pathogens, it is of particular importance to study co-exposure to respiratory viruses and pollen in more detail.

Aim of study: To characterize the immune response of nasal epithelial cells in a physiological model of aerosol co-exposure to pollen and coronaviruses.

Methods: Primary nasal epithelial cell (HNEC) air-liquid interface (ALI) cultures were exposed to particle aerosols of birch, grass and ragweed pollen in a VitroCell® Powder X chamber. After 24 and 48h, cytokines and chemokines were measured in apical washes and basolateral supernatants, and the response of the cells to the pollen aerosols was compared to the response after stimulation with pollen extracts and liquid suspensions. In a virus infection model,

HNEC-ALIs were infected with the human coronavirus (hCoV) 229E, and infectious virus particle production (Plaque assay) as well as viral genomes (dPCR) were assessed after 48 and 72h.

Results: The commonly used pollen extracts resulted in the release of cytokines into different compartments than exposure to the more physiological pollen aerosols. The response to birch pollen aerosol was characterized by an early (24h) induction of IL-8, IL-6, IL-1b, IL-18 and IL-25, and a late (48h) release of TNF-a and CCL22. Exposure to grass- and ragweed pollen resulted in a stronger overall cytokine response than to birch pollen, with a significantly stronger release of IL-1 family cytokines and IL-25. hCoV 229E infection at 0.25 MOI led to a rapid (24h) pro-inflammatory cytokine production mainly on the apical side, whereas type I or type III interferons were not produced at any time.

Outlook: In a co-exposure model, pollen pre-exposure will be combined with subsequent virus infection, and cytokine response and virus replication will be compared with virus infection in the absence of pollen pre-exposure. In addition, another infection model with hCoV NL63 will be established to further investigate the immune response under co-exposure to coronaviruses and birch pollen.

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Airborne Environmental DNA and the Unraveling of Terrestrial Plant Biodiversity

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Abstract

Alpine plant communities are crucial hotspots acting as open-field laboratories for studying the effects of climate change on plant composition, productivity, and phenology. Estimating plant biodiversity over large areas, though, can be challenging due to the labor-intensive efforts required for surveys carried out by traditional methodologies.

The ALPoll project aims to investigate the role of bioaerosols in the estimation of alpine biodiversity, including the informative potential of DNA.

In order to design optimized protocols to analyze environmental DNA (eDNA) from gravimetric aerobiological sampling, a preliminary study was conducted in a rural site, on a set of 13 weekly specimens, collected in duplicate, by a Sigma-2 gravitational sampler of airborne particles. eDNA was extracted with Qiagen DNeasy Plant Mini Kit and ITS1 and ITS2 barcodes were

selected for the DNA amplification, targeting the sequences with diverse primer pairs. Different enzymes for the amplification (GoTaq® and KAPA HiFi DNA polymerases) were tested. Illumina sequencing was performed and an efficient bioinformatic pipeline was designed to improve raw data analysis and precision and efficiency of the taxonomic assignments. Starting from Banchi et al. (doi.org/10.1093/database/baz155) and Quaresma et al. (doi.org/10.1038/s41597-024-02962-5) and intersecting the plant species list for the Italian Alps of Trentino (La Flora del Trentino, Prosser et al., 2019), a curated reference database was built. Sample replicates were also analyzed by optical microscopy for the comparison of data.

Results report the taxonomic resolution of both techniques and seasonal changes in the biodiversity of aerobiome, together with quantitative estimates of airborne biological particles for plant taxon. The number of Operational Taxonomic Units (OTUs) produced by the raw sequences, as well as their taxonomic classification referred to the reference database, differs depending on selected barcodes (ITS1 and ITS2), primer pairs and polymerases, used in the DNA amplification process. In particular, ITS1 barcode generates the higher number of OTUs (858 vs 787 of the best ITS2 combination of primer pairs and polymerase), but the higher number of plant taxa is classified after the ITS2 sequencing (45 families, 82 genera and 50 species). The results are consistent with ITS1 being more effective for fungi taxonomic assignment, and ITS2 maximizing plant classification, thus potentially complementing each other. The comparison of microscopy and metabarcoding data shows a high degree of correlation for many taxa (e.g. Poaceae, Pinaceae or Salicaceae). In addition, the accuracy of classification achieved by ITS1 or ITS2 results is in the vast majority of cases more specific than classification by microscopy.

The final contribution of this study will allow a definition of the different informative potential and complementarity of the two techniques and to obtain an optimized protocol for eDNA metabarcoding to be implemented in the ALPoll project.

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Ozone-induced Post-translational Modifications of Bet V 1 Into Intact Birch Pollen Grains: Chemically Impossible ?

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Abstract

Ozone (O_3) exacerbates the severity of allergy symptoms to pollen grains (PGs) of certain species. Post–translational modifications (PTMs) of allergens may partly explain this symptom-pollutant relationship. To induce PTMs, O_3 must pass through outer pollen structure, cross the plasma membrane and then penetrate the cytoplasm to reach inner allergens. In this study, we question the plausibility of the O_3 diffusion through the PG. We compare the quantity of O_3 captured under *in vitro* conditions with the average number of Bet v 1 allergen molecule on the scale of a single birch pollen grain (BPG) while considering the effect of lipids.

The average quantity of ozone captured is 0.2 pg/BPG, equivalent to 2.5 x 10^9 molecules of O_3. A mass of Bet v 1 of 4 pg/BPG corresponds to approximately 140 x 10^6 Bet v 1/BPG. The stoichiometric ratio is therefore approx. 18 molecules of O_3 per Bet v 1 allergen. We also estimated the quantity of O_3 captured by lipids to be approx. 2/3 of the total, leaving only 6 molecules of O_3 per Bet v 1 allergen. Furthermore, considering that Bet v 1 represents at most only 15% of soluble proteins and neglecting other cellular components that can potentially react with ozone (non-soluble proteins, free amino acids, nucleic acids, other antioxidants), the encounter between an O_3 molecule and an allergen present in the cytoplasm of a BPG seems highly unlikely. More complex reactivity mechanisms may be at play for intact BPG, involving diffusion of proteins or reaction products with O_3. BPG rupture followed by exposure to O_3 could be a direct mechanism of PTMs for Bet v 1. We suggest that PTMs of proteins may occur during their diffusion from the cytoplasm to the outer part of PG upon rehydration during contact with epithelial cells. Addressing the issue of atmospheric reactivity at the pollen grain scale allows for proposing new mechanisms for the complex links between pollution and respiratory health.

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Deciphering the First Thunderstorm Asthma Event Reported in the Paris Region (June 11, 2023)

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Abstract

A thunderstorm asthma event (TAE) is characterized by a sudden increase in acute respiratory illnesses in the hours preceding or following a thunderstorm, attributed to intense exposure to aeroallergens. A few dozen TAEs have been recorded worldwide, including one event in France (Nantes, June 2013). On June 11, 2023, a TAE occurred, characterized by an influx of patients admitted to emergency medical services for asthma, leading to an unusual shortage of oxygen kits. Here, we describe the characteristics of this TAE, based on data of the Paris region from hospital emergency departments, emergency medical services (SOS Médecins) for home and office consultations, pollen count data collected by the RNSA (French aerobiological monitoring network) and air pollution data from the ambient air quality monitoring agency. The storm broke out in the late afternoon of Sunday 11 June in the Paris region. The emergency room visits for asthma increased by 260% on Saturday and 900% on Sunday compared to previous days' average. The number of asthma-related consultations conducted by outpatient doctors rose by 75% on Saturday and 430% on Sunday. Emergency room activity began at around 6pm on Saturday 10 June, 24 hours before the storm. The peak in activity was observed between 4 and 5 am on Sunday. The most affected population was young adult males aged 15 to 44 years. Most patients had no previous history of asthma. Asthma attacks were not particularly serious: 7% of patients were admitted to hospital (compared with an average of 10%). Similar to the TAE in Melbourne in 2016, ozone concentrations of around 100 µg/m^3 were measured for several hours on the afternoon of Saturday 10 June. The aerobiological content of the air was characterized by the presence of urticaceae and grass pollen, with maximum concentrations of 75 and 93 pollen grains per m^3, respectively. A significant peak in mould spores occurred on Sunday 11 June for *Ascosporium*, *Cladosporium* and *Helicomyces*. Analyses of pollen samples are underway to better understand the aerobiological conditions in the atmosphere during this episode, and in particular whether the pollen broke up. It is urgent to strengthen research in the field of predicting TAEs, and to improve communication on this topic with patients, especially young allergic adults with no history of asthma.

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Plant-related Biodiversity in the Alpine Air: a Review

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Abstract

Aerobiology can provide answers on the impacts of global change on plant biodiversity. It has been recognized that alpine environments are susceptible to such changes. However, there are only a few studies worldwide addressing plant-related particle biodiversity in air samples in open areas at high elevations or high geographical latitudes. This study reviews aerobiological papers that focus on assessing plant biodiversity in environments that are either part of the alpine biome or are functionally connected to it.

PubMed was searched for "pollen and alpine"; morphological studies, taxonomical studies, honey studies, fossil pollen studies, and non-English studies were excluded from the resulting papers. Further relevant studies were retrieved from bibliographic references of the same articles and from Google Scholar.

Based on 48 articles reviewed, i) the air sampling; ii) the identification method; iii) the bioaerosol biodiversity in relation to alpine vegetation were analyzed.

As for i), deposition sampling is the method of choice to collect the alpine bioaerosol, while only a few studies use volumetric air samplers. As for ii), the current state of the art for the identification of pollen and non-pollen palynomorphs is microscopic analysis. Yet, results from DNA metabarcoding show a higher taxonomic resolution in identifying plant taxa, than microscopic analysis alone can achieve. As for iii), the establishment of relationships between bioaerosol and plant biodiversity implies the assessment of vegetation diversity and abundance at different scales from the receptor site. Back trajectory models are employed to trace the origin of extra local, long-distance sources.

On the whole, the alpine bioaerosol mirrors the vegetation of wind-pollinated taxa from the immediate receptor site, e.g. herbaceous such as Poaceae, Cyperaceae, Juncaceae, and ferns. Entomophilous taxa, in contrast, are underrepresented. The biodiversity from the alpine air, however, does not only originate from local sources but also from extra-local, regional, and often over-regional areas. For the Eurosiberian plant region, the articles reviewed consistently report pollen from woody plants (Pinus, Picea, Corylus, Betula) above the timberline. Microscale air currents (0 - 2 km) cause the influx from around and below the timberline into the alpine air at the receptor site. Besides, mesoscale air masses (2 - 200 km) including topography-driven convections, thunderstorms, nighttime depositions as well as long-distance transport events (200 - 2000 km) add taxa to the bioaerosol.

Knowledge on the composition of the plant bioaerosol in alpine environments facilitates the reconstruction of past climate, models of climate change scenarios, the interpretation of gene

flow, and the genetic makeup of populations. Such is a valuable tool for plant conservation management in alpine environments.

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Improving Forecasting Pollen Concentration Using a Hirst Network and Phenological observations.

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Abstract

Pollen modeling is a crucial field for understanding the impact of pollen on human health and the environment. Deterministic models are the most common type of pollen model. They are generally more accurate than probabilistic models, but they require a good understanding of the physical laws that govern pollen production, release mechanism, and dispersion; further, a precise distribution of plants is needed, and it is not always possible with invasive species or seasonal plants that are continuously spreading.

This study aims to develop a neural network model that can accurately predict pollen concentration and the start of the pollen seasons in France.

We tested different neural network models combining phenological observations, pollen data from a Hirst network, and meteorological data, selecting two locations (Avignon and Brest). Over the 20 years of data, several random sets were selected, excluding different weeks, to test the ability to predict pollen concentration.

The "best" model was able to predict with accuracy above 80% pollen concentrations and the arrival of the grass season with an accuracy of 6 days. This is significantly better than the accuracy of either deterministic or probabilistic models alone. Notably, our findings indicate that pollen concentrations from Hirst samplers facilitate reliable prédictions at one week.

The model is a promising tool for predicting the arrival of the pollen season and improving forecasting pollen concentration. It is more accurate than traditional models and can provide early warnings to people with allergies.

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Analysis of Historical Clinical Symptoms and Pollen Data in France: a New Symptom Model

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Abstract

This study investigated the relationship between allergic symptoms, pollen data, and weather patterns in France. We analyzed an 18-year dataset of clinical symptoms reported weekly by e-mailed questionnaires by over 100 allergologists nationwide, alongside pollen concentration data collected by the French National Aerobiology Network (RNSA). The pollen concentrations are monitored by 70 captors displayed in the French territory (pollens.fr).

Our analysis revealed a concerning increase in number and severity of allergic reactions over the past few years, with a more significant increase in the southern part of France, with more than 50% of the number of patients over the analyzed period.

To understand the factors influencing these trends, we developed a response function model at the regional level. This model incorporates weather data and pollen concentration to predict the likelihood of allergic symptoms. The new model demonstrates good agreement with the observed data, achieving an accuracy of 0.8 on average.

These findings highlight the growing burden of allergic reactions in France and provide valuable insights for improved allergy prediction and management. Our novel symptom model, with its high accuracy, has the potential to be a useful tool for allergologists and patients alike.

Furthermore, continuous monitoring of clinical data and pollen concentration is crucial for understanding the evolving trends of allergic reactions and refining our models for better prediction and future risk management.

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Pollen Light Backscattering Towards Pollen Lidar Remote Sensing

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Abstract

The Hirst trap has been the historically preferred method for pollen monitoring due to its simplicity and reliable data collection. Recent advancements in AI technology have enabled pollen real-time detection sensors. However, since most pollen sensors operate on the ground, the vertical layering of airborne pollen still needs to be revealed. Lidar remote sensing allows a high-frequency time sampling with vertical resolution (Miffre et al., 2020).

Pollens are complex-shaped particles exhibiting an overall spherical shape with small-scale complex morphological features such as spikes, apertures, and sub-micrometric cavities (Halbritter et al., 2018). As a result, no analytical solution to Maxwell's equations exists for pollen, which is beyond the reach of numerical light scattering models due to their large size. The iLM laboratory instrument (Miffre et al., 2020) allows an evaluation of the backscattering lidar particles' depolarization ratio of various pollen such as birch, ragweed, pine, or cypress (Cholleton et al., 2022), hence opening new insights into lidar remote identification of pollen.

Here, we present the published outputs of this laboratory experiment (Cholleton et al., 2022), as well as the first applications in the field when coupling lidar remote sensing experiments with standard pollen counters installed in Lyon, France.

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Methodological Challenges in the Study on the Immunogenicity and the Allergenicity of Birch Pollen Proteins

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Abstract

INTRODUCTION

Birch pollen proteins consist of 20-25 subunits with a molecular weight ranging from 10 kDa to over 100 kDa. Immune response of individuals sensitive to pollen allergens may differ regarding protein fractions and their source of origin. The goal of the study was to recognize the main allergenic birch pollen proteins originating from the specimen growing in Krakow (Poland) and its close vicinity.

METHODS

The 27 birch pollen samples were collected from the birch specimen, in Kraków and its close vicinity. The immunoreactive protein bands were detected by chemiluminescence using ChemiDoc (Bio Rad, USA) and analyzed by Image Lab software calculating molecular weights and volumes. Then immunoreactive properties of individual subunits were analyzed by immunobloting in two groups of patients included into the project (studied and control groups; 20/20 respectively). To recognize the main birch pollen allergen Bet v1 the comparative electrophoregrams with the commercial, recombinant Bet v1 protein (Antibodies.com) and the sample of diagnostic extract for skin prick tests (SPT) (Hal Allergy) were performed. The selected bands of molecular weights: 12 kDa, 14 kDa, 16 kDa, 17 kDa, 18 kDa, 26 kDa, 35 kDa, 37 kDa, 50 kDa and 75 kDa have been cut out from the stained gels. The Basophile Activation Test (BAT) was made with them in the whole blood of five sensitive and five non-sensitive patients. To identify the proteins present in the selected bands, in-gel digestion with trypsin was performed and an ultra-high performance liquid chromatography coupled with electrospray ionization-quadrupole-time of flight-mass spectrometry (UHPLC-ESI-Q-TOF-MS) and nano *liquid chromatography* with trapped ion mobility-quadrupole *time*-of-flight mass spectrometer (nanoLC-TIMS-TOF-MS) were used. The proteins were identified by matching the protein database.

RESULTS

SDS-PAGE with commercial Bet v1 and the diagnostic SPT extracts showed a clear band of MW between 17.8 kDa and 18.9 kDa, and a slight response from the selected patients. In immunobloting with the diagnostic SPT sample the reaction with the proteins of 14, 35, 55 kDa were detected. Immunobloting with 27 birch pollen samples showed a great specificity of immunological response in reaction with patients sera. The most frequently, the protein of 12 kDa was detected in more than 80% of sensitive patients and in more than 60% of the control individuals. Moreover, in sensitive patients, the bands of MW 35, 25, 37 kDa prevailed.

The percent of activated basophiles in allergic patients was significantly higher as compared to control persons for all of analyzed protein subunits, where the molecules of 75, 37, 35 and 26 kDa were the strongest activated. In total, 147 proteins were analyzed by mass spectrometry, including allergenic proteins such as: Bet v1, Bet v2 and Bet v6. The protein of 75 kDa was not treated until now as an allergenic molecule.

CONCLUSIONS

The combination of immunoreactive subunits and fractions of birch pollen proteins makes a highly unique pattern for individual persons and may be accepted as their specific fingerprint of immunological activity of allergenic proteins in reaction with IgE antibodies present in human sera.

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From Land to Sky: Tracing Iceland's Land Use Transformation through Patterns of Pollen Concentrations in the Atmosphere.

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Abstract

Airborne pollen concentrations exhibit a strong correlation with the flowering intensity of wind-pollinated species in proximity to monitoring sites. While the pollen spectrum and its fluctuations are shaped by climatic conditions and resource availability, the impact of land use dynamics remains a significant but understudied factor. Iceland lacks indigenous Pinaceae pollen producers, with all trees from this family introduced during the 20th century and subsequently planted in tree plantations across the country. Particularly, the expansion of conifer cultivation has been steadily rising, causing large-scale land cover transformations.

In our investigation, we analyzed extensive pollen monitoring datasets spanning over three decades from two sites in Iceland: Reykjavik in the southeast and Akureyri in the north. We utilized the Seasonal Pollen Index (SPI), representing the cumulative daily pollen concentrations for each type, alongside metrics for land occupation by conifer plantations.

Our findings show a direct correspondence between burgeoning conifer plantations and escalating total Pinaceae pollen concentrations in the atmosphere, revealing a tangible reflection of ongoing land use changes on airborne pollen dynamics. Moreover, changes in trends of afforestation with Pinaceae are also closely related to trends in pollen concentrations.

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Local Invasion of Common Ragweed (*Ambrosia artemisiifolia* L., Asteraceae) Has No Impact on *Ambrosia* Pollen Seasons in Kraków, Southern Poland

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Abstract

In Europe, common ragweed (*Ambrosia artemisiifolia*), an annual invasive plant of North American origin, is well known as posing a threat to human health by producing highly allergenic pollen. This threat may become even more severe as common ragweed populations are likely to increase due to global warming. In Poland, common ragweed is still relatively sparsely distributed, but the number of persistent populations has increased. Interestingly, an inventory of the only previously known population of common ragweed in Kraków, southern Poland, carried out in 2020, confirmed a significant increase in its abundance. Moreover, in the next three years of floristic surveys in the city, 36 new populations were discovered. Most of them turned out to be ephemeral and disappeared after one season, and their presence was related to contaminated grain in bird feeders. However, some of the populations remained stable or even began to increase successively in each following season, showing the highest abundance on railway areas and roadsides. To verify whether the local invasion of *A. artemisiifolia* affects the ragweed pollen seasons in Kraków, the aerobiological data from the pollen monitoring station were analysed. Despite the increase in the common ragweed range within the city, the ragweed pollen seasons were not significantly affected since 2020 and still mostly rely on the pollen influx from long-distance transport. However, residents became increasingly exposed to ragweed pollen, mainly due to the proximity of residential and recreational areas to the largest populations of common ragweed. Awareness of local authorities and residents should be raised, because, in the absence of appropriate protective measures (mowing, uprooting), common ragweed may start to become a serious problem in urban space. The presence of a larger ragweed population should also be publicised in the medical community, especially given the influx of patients of Ukrainian origin who may have acquired an allergy to ragweed while still living in Ukraine.

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Investigation of the Potential of Using Pollen Isolated From Vehicle Air Filters As Evidence in Forensic Cases

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Abstract

Forensic palynology is a branch of science that enables the use of pollen and other palynomorph evidence obtained from a crime scene as evidence in criminal cases. Studies in the field of forensic palynology show that soil samples obtained from textile products such as clothes, ropes, and fibers, as well as from tires, pedals, and fenders of vehicles contain pollen and are one of the biological evidence used in the capture of criminals. Several preliminary studies are showing that pollen evidence from vehicles is not limited to these and that pollen accumulated in the air filter of vehicles can also have important contributions to solving criminal cases. This study, it is aimed to examine the potential of pollen analyzed from the air filters of motor vehicles to be used in solving incidents such as theft and kidnapping. For this purpose, air filters were taken and analyzed once a month in March, April, May, and June from buses traveling on designated routes. The data obtained as a result of the analysis of the filters were compared with the data of pollen traps. In this study, the vehicle air filters of buses traveling a total of 124,140 km on the routes determined in Kestel, Görükle, Osmangazi, and Mudanya regions of Bursa province in March, April, May, and June 2019, which were changed once a month, were examined palynologically in the laboratory. The acetolysis method was used for pollen analysis from vehicle filters. The obtained pollen data were compared with the data of Hirst-type volumetric pollen traps in Bursa Kestel, Görükle, and Osmangazi regions, which collect the pollen in the atmosphere of these regions. Data from vehicle air filters and volumetric pollen samplers were analyzed, and pollen was examined under a light microscope. The data were statistically suitable for the PCA test and analysis. A total of 43 taxa and 241,227 pollen/m^3 were detected from the volumetric pollen traps located in the specified regions for four months, while 44 taxa and 5,515 pollen/m^3 were detected from the vehicle filters of the municipal buses. The analysis of the data by PCA test shows that pollen is concentrated in the atmosphere in April, May, and June. *Platanus* sp. pollen, which was abundant in atmospheric data in March and April, was observed in very small amounts in air filters. As a result of the statistical analysis, similar to the atmospheric data, pollen belonging to Gramineae and *Pinus* sp. taxa were also seen in the filter in May. As a result of this study, it was determined that the taxa in atmospheric data and filter data were similar to each other. Due to the population density and the barrier effect of buildings, the pollen amounts in the filter and atmosphere are not similar in most of the stations.

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Investigation of Pollen Attachment on Different Fabric Types

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Abstract

Biological aerosol-induced allergic diseases have been increasing with the effect of climate change and environmental pollution in recent years. Pollen grains, which are carried indoors by adhering to the production materials of the clothes and goods used indoors, can trigger IgE-related reactions in susceptible individuals. Another importance of pollen grains lies in understanding their function as secondary trace evidence in the analysis of forensic cases. Palynological studies contribute to the temporal and spatial analysis of forensic events. It has been reported that textiles act as collectors and carriers for pollen and that the capacity and persistence of pollen grains to adhere to textiles varies. Fabrics are used as source material in forensic investigations. In this study, the aim was to determine pollen grain adhesion properties to fabrics. For this purpose, a washing procedure was applied and the effectiveness of the number of washes in the removal of pollen grains attached to fabrics was investigated. Whether the pollen recovered from fabrics can be used as evidence and the adhesion properties of pollen grains with different morphological characteristics to different fabric types were investigated. In this way, data that can help individuals with pollen allergy to choose the right fabric type can be obtained. In addition, the role, validity, and necessity of pollen analysis in forensic cases to accurately match the crime scene with the suspect or victim associated with the incident is emphasized. In this study, 5 types of fabrics commonly used in the textile industry in Turkey (chintz, artificial leather, linen, denim, combed cotton) and pollen from *A. artemisiifolia, C. album*, *C. avellana*, *P. officinalis*, and *P. × acerifolia* plants were selected for testing. The amount of pollen suspended in one milliliter of distilled water was determined with a hemocytometer. A certain amount of pollen was weighed on a precision balance and suspended in aerosol bottles with 30 ml of distilled water. The fabrics were sprayed with pollen suspensions. The fabrics were washed with Tween-20 solution and distilled water until no significant amount of pollen remained. After each wash, the number of pollen recovered from the fabrics and the pollen retention in the fabrics were determined, depending on the number of washes. The experiment consisted of three repetitions for each pollen-fabric combination. According to the findings, all fabrics were found to be passive pollen collectors and all pollen could be retained between the fabric fibers. In all combinations of pollen and fabric, the first wash was the most effective for removing pollen from the fabrics. Pollen could still be recovered from the fabrics after at least four washes. Pollen persistence on fabrics is influenced by the structural attributes of both the pollen and the fabric. It was determined that the effectiveness of the washing protocol varied depending on the weaving type of the fabric, fiber structure, yarn weight, and morphological properties of the pollen. This article emphasizes the importance of pollen as evidence in forensic investigations. This study was supported by the Bursa Uludağ University Scientific Research Council (Project no: FHIZ-2022-1069).

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Optimisation of Airflow Sampling Using an Airplane: a Novel Approach in Defining the 3-d Atmospheric Biodiversity

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Abstract

Background: Bioaerosol identification and quantification are essential for addressing health-related hazards and establishing appropriate exposure thresholds. Most observations have been acquired by sampling with various stationary devices, usually located up to 25 m above ground level, and mostly in highly populated, urban environments. However, more information is needed regarding the biodiversity and abundance of bioaerosols along vertical profiles, which could contribute significantly to more efficient meso- and micro-scale modelling of airborne pollen and fungal spores, in addition to long distance transport modelling. The main goal of this study was to develop a novel method for the 3-dimensional atmospheric detection and monitoring of bioaerosols by use of an aircraft, as well as the standardization and applicability of the techniques acquired at the operational and real-life level.

Methods: Biomonitoring took place by A) the 'gold-standard' techniques (Hirst-type volumetric traps), B) newly developed isokinetic air inflow tubes design for application at up to 160 km/h. Both were conducted simultaneously in the field during the first phase of the project in Roskilde (Denmark). Samples were taken in different timeframes throughout a day, viz. 10 min, 20 min, 30 min and 60 min, on the rooftop of a building, approximately 4 m above ground. Samples were stained and mounted on glass slides and under cover slips and, then, all types of pollen taxa and spores were counted microscopically and extrapolated to particles per m3 of air. Accordingly, during the second phase, the selected optimal techniques were applied from an ultralight aircraft, with the appropriate devices mounted to the outside of the aircraft, and sampling took place outdoors at various vegetation and urbanisation environments.[1] Airborne sampling was done within the planetary boundary layer at 150m, 450m and 900m above ground. Simultaneously, collocated sampling was conducted at ground level using Hirst type and portable volumetric samplers.

Results: Statistical analysis was performed for taxa whose pollen and fungal spores contributed more than 0.5% to the total pollen and spore concentration, respectively, for both ground-based and aircraft sampling. It was found that the isokinetic aircraft samplers had approximately 3.5 times higher inflow rate compared to the Hirst-type samplers. The best timeframe for the isokinetic sampler was found to be the 20min timeframe, for which a satisfactory amount of

pollen grains was counted (more than 100 pollen grains in total). During the test flights, more than 500 pollen grains/m³ were detected at >900m of altitude. Moreover, the diversity of pollen was higher at higher altitude, a likely outcome of longdistance transport.

Conclusion: Pollen may fly as far and as high as the environmental conditions allow them to and at a greater distance than is currently thought. Such incidents have to be monitored regularly, which is possible via novel detection methods within a 3-dimensional biomonitoring concept.

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Evaluation of the Pollen Classification Models Used in the Automatic Pollen Monitoring Network in Switzerland

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Abstract

Pollen allergies and asthma impact up to 40% of the European population. The number of allergic patients has shown a constantly increasing trend over the past three decades with certain groups of the overall population, including children, being particularly strongly affected (Rönmark et al. 2009, Laatikainen et al. 2011.). Pollen allergies represent a financial burden for the Public Health system, estimated up to €150 billion annually. Climate change will likely worsen this scenario, with global warming and increased CO_2 concentrations causing longer flowering periods and more plant activity, that can cause longer and more intense pollen seasons in the future (Ziska et al. 2019). Agriculture and sylviculture can also benefit from pollen monitoring. It is thus of utmost importance to provide up to date and high temporal resolution pollen concentration data. The current measurement networks still rely strongly on the manual method (Hirst-type traps), which provide daily average data with a delay of up to 9 days, burdened with large uncertainties due to sampling related issues.

The EU Horizon Europe SYLVA project is aimed at developing an automated measurement network across Europe, i.e., an infrastructure which consists of a network of automatic samplers coupled to reliable classification algorithms to provide real time or near real time concentrations of relevant pollen taxa. Two important steps towards setting up a unified infrastructure are, i) the homogenization of the manual and the automatic data, ii) the selection of accurate and robust operational models, which cannot be performed without a rigorous comparison of the data.

In this work, we compare three different models developed in the Swiss Automatic Pollen Monitoring Network. Data obtained from the Poleno network in Switzerland between 2021 and

2023 are analyzed and the model predictions are compared with data from the operational manual network using multiple statistical metrics. Comparison of the models for seven known pollen taxa (Alnus, Betula, Corylus, Fagus, Fraxinus, Quercus and Poaceae) show that the current operational automatic model has an excellent performance. It provides daily average pollen concentrations from the automatic measurements that correlate well with the manual data across all measurement sites, with Kendall and Spearman correlation coefficients both superior to 0.7 for nearly all of the taxa. Furthermore, a comparison of the different models for the seven above mentioned taxa showed that models that use only holographic images from the Poleno instrument perform remarkably well on a reference dataset consisting of the above mentioned seven pollen taxa having a balanced accuracy of 0.968 and an F-1 score of 0.964, which can be further improved by exploiting fluorescence data provided by the Poleno instrument (Erb et al. 2023).

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Increase in Airborne Allergenic Pollen in Trentino (North Italy) Over a 30-year Period (1989-2018) is Connected to Temperature Rise.

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Abstract

The aim of this study is to verify if changes occurred in the seasonality and yearly amounts of airborne pollen in San Michele all'Adige - Northern Italy, and to evaluate their relation with climate change-related variables, such as air temperatures and frost days in the period 1989 - 2018.

The study of airborne pollen and its spatio-temporal changes is highly important due to the allergenicity of many pollen taxa. The pollen allergy, in fact, interests 25-40% of population, globally, and displays an increasing trend.

Climate change may impact pollen allergenicity and production, as well as plant distribution, with the potential spread of neophytes that produce allergenic pollen. The main hypothesis of our research is that climate change impacting our study area influences pollen dispersal in the atmosphere, and therefore on human-health related issues.

Airborne pollen was collected using a volumetric Hirst-type aerobiological sampler (Lanzoni VPPS 2000), and the daily concentration of airborne pollen ($P^{*}m^{-3}$) was calculated for a total of 24 arboreal (AP; trees and shrubs) and non-arboreal pollen taxa (NAP; herbaceous) over a 30-year period. The sampling and analysis of airborne pollen have been performed in accordance with the UNI EN 16868:2019 European standard procedure. The main pollen season (MPS) descriptors were calculated for each taxon. The presence of a monotonic upward or downward temporal trend in pollen season descriptors was verified (non-parametric Mann-Kendall test) and changes were analyzed in relation to air temperature, precipitation, and land use; in addition, pollen data were analyzed clustered into three decadic blocks (non-parametric Kruskal-Wallis ANOVA) to minimize interannual fluctuations and maximize relevant change signals.

The major change observed during the study period was the significant increase in annual pollen integral (APIn). This change is strongly emphasized when analyzing the pollen data in three decadic blocks, both for all the considered taxa (+58%) and for Arboreal Pollen (AP; +155%). When considering single taxa, APIn shows a significant positive trend for Cupressaceae/Taxaceae, *Ulmus, Populus, Salix, Ostrya, Quercus, Olea, Plantago*, Cannabaceae, and *Ambrosia*; a significantly earlier start of the MPS is proved for *Rumex* and Poaceae. Cumulated APIn shows a significant positive correlation with annual T min (<0.005) and T mean (<0.001), both of which showing a significant increase, and a negative correlation (<0.025) with the number of frost days.

The research demonstrates an increasingly larger amount of airborne pollen connected to climate change, such as rising temperatures and milder winter conditions in the study area, leading to a major threat to people suffering of pollen allergies.

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Long-term Trends in Airborne Pollen Levels in Thessaloniki, Greece, Over the Period 1987-2023: Higher, Earlier or More Complex?

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Abstract

Long-term trends in airborne pollen concentrations have been documented more and more frequently over the last two decades. The Inter-Governmental Panel of Climate Change (IPCC)

have acknowledged airborne pollen abundances and seasons as bio-indicators of climate change. The ongoing climatic change is known to be more intense in more vulnerable ecosystems like higher latitudes and altitudes and the Mediterranean basin. Likewise, the Balkans have been identified as one of the bioclimatic regions that have been witnessing some of the most robust increasing trends across Europe, with Thessaloniki having been a top changing location in terms of Annual Pollen Index (API). While increasing trends in atmospheric pollen levels have been reported almost worldwide, neither a wide spectrum of taxa has been always investigated, nor a variety of environmental factors has been examined.

The aim of this study was to investigate potential trends in the API of the most abundant plant taxa (>1% relative contribution to the total API) in Thessaloniki, Greece, during the whole biomonitoring period, 1987-2023. We attempted to identify potential causality relationships with various environmental parameters, including major ecosystem changes, namely air temperatures, precipitation, air pollutants, wildfires, land use changes. This was explored for 9 different taxa, viz. *Carpinus*, *Corylus*, Cupressaceae, Oleaceae, Pinaceae, *Platanus*, Poaceae, *Quercus*, Urticaceae. Apart from the APIs, we also tested for trends towards earlier, longer or more highly peaked pollen seasons. Various statistical approaches were adopted, among which General Linear Models and Generalized Non-linear Models, Dynamic Regression Models, and Structural Equation Models.

We found that for seven out of nine taxa, as well as their aggregate, APIs have been increasing, of which six significantly: *Carpinus*, Cupressaceae, Pinaceae, *Platanus*, *Quercus* and Urticaceae. The observed trends were non-linear; they started exponentially, but became linear in the last two decades. The increase was most pronounced in the mid-2000s, when the APIs for the aggregate abruptly increased by approximately 50%. Also, increases throughout the pollen time-series were more intense (higher slope) for the highest APIs per taxon, which signifies more intense masting years rather than simple increases in the averages. For the pollen-season-related attributes (onset, peak, end and duration), the trends were mostly non-significant, however there was a clear tendency towards earlier starting seasons, more highly peaked and sometimes extended later in the year. Land use changes, wildfire events and temperatures (but not precipitation) were consistently the environmental factors that explained most of the variance of the APIs increases for the six taxa.

Our results suggest that the ongoing climate change plays a dramatic role in increasing airborne pollen abundances, nevertheless, changes are not linear; also, and they are particularly pronounced after extreme temperature events and because of land use changes. Only the totality of these mediating factors can explain a considerable part of each taxon's API variance. These results raise issues regarding predictions of climate change effects on plants' reproductive outputs and associated increased pollen allergic responses in the future.

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Higher Temperature Settings in Urban Environments Are Connected With Increased Pollen Production in Anemophilous Woody Taxa

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Abstract

Plants' reproductive outputs have been reported by IPCC as sensitive to environmental change. Nonetheless, the actual pollen amount produced per reproductive unit, be it inflorescence, individual plant, or any other, has been studied only to a limited extent, probably because of the arduous nature of the sampling collection and laboratory processing. Pollen production estimates and their responsive ability are also important because of the associated health risks, as the pollen produced per individual plant and per taxon determines the fraction of the airborne pollen to be captured in the air of an area. The aim of the study was to assess the pollen production of woody taxa, which are common in the urban vegetation of a Mediterranean city and to examine how much this productivity matches the airborne pollen concentrations of the same taxa.

We studied the pollen production of 12 anemophilous woody taxa, in and around the city of Thessaloniki, Greece, for two consecutive years. Selection of taxa was based on their relative abundance in the regional vegetation and on their high allergenic potential. They were *Alnus glutinosa*, *Carpinus orientalis*, *Cupressus arizonica*, *Cupressus sempervirens* subsp. *horizontalis*, *Cupressus sempervirens* subsp. *pyramidalis*, *Juniperus communis*, *Juniperus oxycedrus*, *Platanus orientalis*, *Pinus pinea*, *Platanus* x *hispanica*, *Populus alba* and *Thuja orientalis*. For each taxon, 10 reproductively mature individuals were selected in at least two stations, usually of different elevation and/or with different exposure, so as to investigate the variability of their responses under different environmental stressors. From each sampled individual, three floral units were collected in each side of the canopy (north, south) and counted inside a taxon-specific sized frame, depending on floral unit density on the canopy. Additionally, tree morphological features were measured (trunk perimeter and height, individual height, two perpendicular to each other diameters of the canopy). The content of floral units in pollen was analysed in the lab, counted under microscope and extrapolated into productivity scales of floral unit and pollen.

Pollen production ranged from thousands to millions of pollen grains at the inflorescence level ($7.43^{*}10^{5} \pm 1.21^{*}10^{6}$), and from millions to billions at the individual tree level ($2.31^{*}10^{11} \pm 5.94^{*}10^{11}$). For most taxa, productivity was higher for individuals found in areas with southern direction. Pollen production for ten of the 12 taxa differed between years; the highest productivity was observed during the previous year (2010) for conifers that form their floral units in the previous summer, whereas for 2011 for the later-flowering conifers or for broad-leaved. Year 2010 was warmer by 1.2 oC compared to the following two years of sampling. Airborne pollen concentrations did not always correlate strongly with the respective pollen production amount, when significant at all.

Our findings show that plants produce more pollen in more urban settings, in areas with southern directions, or during warmer years. This productivity did not always match the airborne pollen concentrations, suggesting complex pollen transport. As climate change induces increases of pollen for many taxa, it is expected to pose, an increased threat for allergic individuals.

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Sampling Air and Water in a Wastewater Treatment Plant: 16S rRNA Gene Metabarcoding for the Identification of the Bacterial Communities

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Abstract

The present study is focused on the analysis of the wastewaters and the bioaerosol formed in the aeration tank of the wastewater treatment plant (WWTP) in Trieste (Italy). The aim is to characterize the bacterial community present in the wastewaters using the environmental DNA (eDNA) metabarcoding of the 16S rRNA gene, which is a technique that allows the simultaneous identification of many taxa of bacteria in the same environmental sample. Currently, no standardized methods for bioaerosol sampling and analysis for molecular ecology research exist. Hence, the aim of this study is to develop a procedure for the characterization of microorganisms in the environmental bioaerosol by comparing different sampling devices and defining both a sampling strategy and laboratory protocols for the identification of the species present in the WWTP. Bioaerosol samples were collected as close as possible to the aeration tank emissions with three different sampling devices using three different sampling strategies: filtration, swirling aerosol collection, condensation growth tube. In addition to bioaerosol samples, water samples were collected directly from the sewage in order to obtain a true representation of the microbiological community present in the plant. Samples were transported to laboratory where total DNA was isolated. A quantitative real-time PCR (Polymerase Chain Reaction) was performed using three primers that together target the V3 and V4 subregions of the 16S rRNA gene in order to cover the entire bacterial community. The PCR step allowed to obtain libraries of sequences representative of the microbial diversity, which were sequenced on an NGS (Next Generation Sequencing) platform. This technology permitted to generate millions of reads from the samples, which were then compared to a reference database for the taxonomic assignment and determination of the relative abundance of the various components of the microbiome. In wastewater samples the bacterial groups were all dominated by the following bacteria OTUs: *Acinetobacter* (26,37% of total sequences in all water

samples), *Arcobacter* (17,44%), Arcobacteraceae (13,7%), *Pseudarcobacter* (5,19%), *Bacteroides* (4,94%), *Aeromonas* (4,92%). In the aerosol samples the most abundant genera of bacteria were: *Stenotrophomonas* (68.4%), *Klebsiella* (12.9%), *Enterobacter* (3.9%), *Delftia* (2.9%), *Acinetobacter* (2.8%), *Pseudomonas* (2.3%). Overall, the air samples have a lower number of genera in respect of the wastewater ones in terms of relative abundance and alpha diversity rarefaction curves of the bacterial richness. In addition, air samples are not as homogenous to each other as the wastewaters, and it seems that different sampling systems have different sampling efficiencies, therefore the results obtained from a bioaerosol study may differ according to the air sampler used. In particular, between the air samples, the BioSpot-VIVAS sample stored in DNA/RNA shield™ contains a higher number of genera in respect to the others, which contain instead one dominant genus of bacteria. To implement the study of the biodiversity present both in air and wastewaters a metagenomic survey can be followed, in order to identify a vast number of genomes and perform a functional characterization present in the samples.

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Evaluation of European Pollen Forecasts From CAMS Regional Models for 2022

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Abstract

Background

The quality of everyday life and efficiency in daily operations are often affected by the environmental conditions. The timely information about the expected levels of air pollutants and allergenic pollen in particular, will support the better planning of the outdoor activities of the sensitive individuals. The Copernicus Atmosphere Monitoring Service (CAMS) routinely provides the pollen concentration predictions among the others atmospheric parameters. The 4-days forecasts over the European domain are generated by 11 state-of-the-art chemical transport models and their ensemble. In this study we analyze the performance of the pollen forecasts by comparing them with the data of aerobiological observations.

Methods

To evaluate the model's performance daily timeseries were computed from the first-day hourly forecast of the CAMS models. Twelve model datasets were considered: CHIMERE, DEHM, EMEP, EURAD, GEMAQ, LOTOS-EUROS, MATCH, MOCAGE, MINNI, MONARCH, SILAM, and

ENSEMBLE. The latter was calculated as a median of all original models. The target species were birch, olive, grass, and ragweed during the pollen season in 2022 (1 Jan – 31 Oct). The pollen observations for the same period were available from the 100 European Aeroallergen Network (EAN) stations in Europe (provided within a contract agreement between CAMS and EAN). The statistical characteristics used for estimation of the forecast accuracy of every model were: model mean bias, temporal correlation coefficient, root mean square error, shift of the pollen season start/end for aerobiological and medical season.

Results

The reliability of pollen predictions in 2022 was varying depending on the model, statistical parameter and pollen taxon. The analysis had revealed the underestimation of the birch daily levels (less than 30% of mean concentrations 60 pollen/m3) for the majority of the models, while for ragweed the absolute bias was about 25% (of the mean 21 pollen/m3). The mean temporal correlation was about 0.5 - 0.6 for birch and above 0.6 for grass. The timing and duration of the birch season were predicted quite good by most of the models with errors in the season start/end about ± 2-3 days on average. The mean olive concentrations have negative bias (within about 50% of the mean value 20 pollen/m3) for most of the models while for the grass predictions only 6 models have lower than 50% (mean 21 pollen/m3). Olive season propagation was represented with lower accuracy than for birch that worsened the correlation scores.

Conclusions

Despite the fact that all current CAMS models use the identical formulation of the pollen emission source, their predictions are significantly varying from model to model due to the different model's architecture and possible implementation issues. The analysis didn't reveal any superior model among the 11 CAMS models, however, the ensemble out of them had regularly demonstrated better scores for all forecasted pollen.

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Learning from the Past: Requirements for a Successful Pollen Counting Network

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Abstract

Creating a successful pollen counting network requires several key attributes. We identify four necessary factors: robustness, availability, scalability, and self-perpetuation. First, a successful pollen counting network should be robust, meaning it should not be sensitive to single points of failure. This ensures that the network remains operational even if individual components or stations experience technical or other issues that prevent it from making pollen count contributions. Robustness guarantees consistent and reliable data collection, which is crucial for accurate pollen monitoring. Second, the network should be available. In other words, a pollen network must be capable of providing data at a level of resolution that is useful to its users. Manual counting methods, with their inherent latency of 24-96 hours or more, are unable to sustainably maintain a daily or hourly level of availability without a substantial increase in time commitment and cost. Automated systems that provide real-time data are preferred, as they offer immediate access to pollen information for decision-making and allergy management. Third is scalability. The network should have the ability to expand its coverage quickly and easily without significant additional effort. This ensures that a larger geographical area can be monitored effectively, providing comprehensive pollen data for various regions. Manual counting networks are not scalable. For each additional counting location, someone has to collect the specimens and process them, requiring a nearly linear increase in effort. Fourth, a successful pollen counting network should be self-perpetuating. This means that as pollen counting networks mature, they should continue to grow and operate without requiring substantial investments of time, talent, or capital. Individual contributors to all manual counting networks have shifting priorities, changes in funding situations, retirement, maintenance, or other issues that all result in a dwindling number of stations reporting pollen as networks age. This creates the opposite of a self-perpetuating model, and consequently, pollen networks of the past have all struggled to sustain themselves. Manual counting methods do not align with the criteria we have defined for a successful pollen counting network.

We report on a world-wide network of automated pollen counting devices. The network began in 2016 when the first two Pollen Sense APS devices were provisioned. Two more devices were provisioned in 2018. From 2018 to 2021, growth of the network was linear, adding 34 devices on average each year. Growth of the network in 2022 and 2023 proceeded at a much faster rate, adding 59 devices to the network each year. In 2024, the world-wide Pollen Sense APS network is on track to surpass 300 counting stations. By leveraging automated systems that offer real-time data, expanding coverage efficiently, and engaging participants effectively, we can create a pollen counting network that delivers accurate and comprehensive pollen information, thus improving allergy management and enhancing our understanding of pollen-related phenomena.

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Particle Hunter: Collaborative Correction to Benefit All

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Abstract

Identifying pollen or other airborne particulates via the application of artificial intelligence (AI) vision models is becoming standard practice. But a single model, no matter how well trained, is not able to perform equally well in all situations, applications, or environments. Multiple AI vision models are needed for high accuracy and precision of pollen identification. But managing the creation, training, maintenance, and deployment of trained vision models at enterprise scale present significant challenges for accurate identification of pollen by automated pollen collection devices. We report on Particle Hunter, the model management platform employed by Pollen Sense to continuously improve and continuously deploy AI vision models.

In order to successfully identify pollen types from images AI vision models must do at least two things. First, the AI software must be able to detect pixel groupings within any given image frame that contain items of interest. We refer to the models that accomplish this first level of identification as detector models. The detector models determine which parts of the image frame are part of the background, which parts of the image frame will be identified as a particle, and which high-level class each particle belongs to (i.e. pollen grain, mold spores, plastic particles, or another particulate type). Second, the AI software must be able to classify the pollen grain or mold spore or other particulate of interest into appropriate species/genera/families or other appropriate groupings. We refer to the models that accomplish this second level of identification as classification models. Particle Hunter is used to manage both the detector and classification models. This is important because Pollen Sense has trained hundreds of unique vision models for dozens of geographical areas for each season of the year.

Particle Hunter includes an interface allowing users to see what particles have been detected, classified, as well as the probability of classification. Should a classification be incorrect, users can click on the particle, classify it correctly, and then submit the user-verified particle classification directly to the training library, enabling immediate improvement. Additionally, Particle Hunter enables users to easily add new species or other particulates to the training library. If known pollen species or other particulate matter (e.g. Cannabis sativa or asbestos) are collected via a given APS unit, Particle Hunter can be used to tag images and create a training dataset. Furthermore, Particle Hunter can be used to 'hunt' species from the wild. Hunting involves identifying a small number of target images of the species of interest, pooling images from one or more APS sensors within a specific time frame, then asking for it to 'hunt' for more. Users then review and cull the resulting images. After a few iterations, users can quickly generate datasets sufficiently large to use for training purposes. This iterative training allows for collaborative correction and improved classification of pollen, mold, and other airborne particulates, thus enabling more accurate classification models and making it possible for the community of users to benefit from a worldwide network of sensors.

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Ice Nucleation Activity Is in the Air: Using Hot-Air Balloon to Sample PBAPs in Southern Brazil

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Abstract

Primary Biological Aerosol Particles (PBAPs) are airborne solid and insoluble particles originate from living organisms such as bacteria, fungi, and pollen. They can impact directly and indirectly on Earth's climate as well as on human and animal health. These particles play a crucial role in triggering cloud drops and ice formation as they can act as ice nuclei (IN). In this research project, by collecting air samples with hot air balloons, we aimed to quantify the activity of ice nuclei (INA), examine their influence as cloud condensation nuclei (CCN), and explore links between the global climatology of hail and PBAP-IN in the Southern region of Brazil. Samples were collected at ground level and 150 m above-ground level using a Microbial Air Monitoring System (MASS100) and a Kärcher vacuum cleaner, and tested *in situ* using PORTABINA (Portable Ice Nucleation Activity system, developed in partnership with INRAE France and CNR-IBE Italy). For fungal cultivation, we used a Petri dish containing DRBCm with antibiotics, whilst, for the bacteria cultivation, the TSB with antifungal was used. Sampling was seasonal and with annual replication (2022 and 2023). The samples collected with the Karcher, both summer and winter samplings, showed ice nucleation activity from -3 to -10° C. These results corroborate with the biogenic origin of ice cores present in the air, as these freezing temperatures are only possible in the presence of biological IN. The highest amount of IN was found during winter, with average of 0.13 at 150 m and 0.22 cumulative PBAP-IN per m^3 at ground level, compared to 0.07 per m^3 at 150 m and 0.09 at 0 m, in the summer samples ($p<0.05$ at -10° C). Referring to the sampling of cultivable microorganisms, fungi presented a difference in viable spores between the two seasons ($p=0.005$), winter samples presented on average of 84.85 CFU/m^3 more than summer. A similar pattern was found for bacteria ($p=0.002$), with an average of 64.97 CFU/m^3 more during winter. These results reveal a seasonal variability and a possible relationship between the abundance of these microorganisms and environmental conditions. This research is a precursor in assessing the IN activity of air samples collected from a tropical environment. It contributes to deeper comprehension of the intricacies of this environment and its dynamic interactions with microorganisms. Furthermore, the insights gained will be utilized as input data for numerical modelling in cloud physics as part of our ongoing project.

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Bayesian Hierarchy of Co-sensitization to Pollen in Fungi-sensitive Patients Corresponds With the Pollen Season Timing of These Allergens

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Abstract

Background: Allergenic sensitization to fungi is a significant factor of allergic rhinitis and bronchial asthma. However, there is limited data regarding the nature of comprehensive sensitization in individuals sensitive to fungi. Therefore, the aim of our work was to comprehensively determine the sensitization profile of patients hypersensitive to fungal allergenic components in the Ukrainian population, identifying its regional specifics and features of these patients' co-sensitization to allergens of other groups and establishing potential relationships between causative allergens and their ability to provoke this hypersensitivity.

Method: To address the established aim, we analyzed patterns of sensitization to allergens of different groups in fungi-sensitized individuals from 17 regions of Ukraine, aged from 1 to 89 years, who underwent multiplex allergy text ALEX in 2020-2022 years. To analyze the obtained data, a set of programs was developed using Python and R programming languages, implementing the K-means++ clustering method. Bayesian networks were constructed based on the created clusters, allowing for the assessment of the probabilistic interplay of allergen molecules in the sensitization process of patients.

Results: It was found that patients sensitive to fungi are polysensitized, with 97.04% of them having unique allergological profiles, comprising from 2 to several dozen allergens from different groups. Sensitivity to Alt a 1 dominated and was observed in 79.39% of patients, 62.17% of them were sensitive the solely to Alt a 1. The sensitization rates to fungi in general and specifically to *Alternaria* were lower in western parts of Ukraine, especially in the Carpathian region, situated within the Broad-leaved Forest zone.

Bayesian Analyses suggested that the immune response to Alt a 1 may act as the primary trigger for sensitization to other allergens and may contribute to a high probability of developing sensitivity to grasses (primarily to Phl p 2), ragweed extract, and the major component Amb a 1 (pectate lyase), as well as to pectate lyase Cry j 1 and cat allergen Fel d 1.

Established hierarchy of the sensitization development when Alt a 1 determines sensitivity to Phl p 2 of grasses and this allergen then leads to the sensitivity to *Ambrosia* corresponds well with the timing of the seasons of the *Alternaria* and mentioned plants.

Sensitivity to *Ambrosia* extract may dominate in the development of sensitization to ragweed pollen in fungi-sensitive patients indicating the importance of different allergenic components of this plant's pollen. This hypothesis, along with the assumption that Phl p 2 may be the main trigger for sensitivity to grasses in patients with *Alternaria* allergy, requires further clinical investigation.

Conclusion: Sensitivity to *Alternaria* prevailed among fungi-sensitive patients.

The sensitization rates to fungi were lower in the western parts of Ukraine, especially in the Carpathian region, situated within the Broad-leaved Forest zone.

The immune response to Alt a 1 may act as the primary trigger for sensitization to other allergens and may contribute to a high probability of developing sensitivity to grasses, ragweed extract and pectate lyases Amb a 1 and Cry j 1 as well as to cat's Fel d 1.

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Aerobiological Cooperation in Northern Europe: Fresh Ideas, Modern Devices, New Technologies

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Abstract

After a period of relatively low collaborative activity, aerobiological teams of Northern Europe (Fennoscandia, Baltic countries and Iceland) are again joining the efforts. The key topics of

interest and concern are similar to those in other European regions: continuing paradigm-change due to new opportunities and challenges related to the new technology, interactions with users and potential new areas for contributions, financial sustainability of the monitoring and forecasting activities, etc. The specificity of Northern Europe, apart from comparatively similar climatic conditions and vegetation, is in extensive collaborative experience, active regional associations, and possibilities for common funding. A large sparsely populated territory poses particular challenges in organization of regular monitoring and forecasting: dense observational networks are unfeasible, which also makes model development and evaluation difficult.

During recent years, coordinated developments emerged in several challenging directions.

Automatic monitors.

Transition from manual to automatic networks has started in most of the Nordic countries, with limited (1-3) flow cytometers being installed and tested in regional environments. Hierarchical networks consisting of a few high-end and a larger number of mid- or low-cost automatic monitors are being considered in practically all countries. As cost-saving alternatives, Poleno Mars (mid-range) and PollenSense (low-range) attract most attention in comparison with the high-end models like Poleno Jupiter. There is a general understanding that the data from the monitors should be made publicly available after the installation and operationalization period is over: this is the only way to get the maximum return from the real-time observations. Coordination is emerging in the location of the new devices, in order to maintain homogeneous network across the region. Responding to the infrastructure challenges of the new devices, coordinated efforts have been agreed, with links anticipated to the emerging SYLVA infrastructure.

Climate change and biodiversity monitoring.

Northern Europe is warming much faster than the rest of the continent. Related changes in plant phenology, shifts of flowering seasons and habitats are followed in several countries. In this connection, importance of very long time series in the region (e.g., first observations in Sweden were started in 1960s) is very high, and efforts are undertaken to continue these with as little disturbance as possible.

Pollen modelling.

Nordic countries possess four atmospheric composition models: SILAM, MATCH, EMEP and DEHM. All of them are members of CAMS European ensemble and equipped with pollen production modules. Their application in a joint high-resolution regional ensemble is under discussion.

Users and stakeholders.

New conditions and possibilities allow for more diverse interaction with users of aerobiological information. Apart from traditional public-health applications, connections are being established with the agriculture and forestry industries (primarily fungal spores monitoring) and climate change and biodiversity monitoring.

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Advancing Automated Identification of Airborne Fungal Spores: Guidelines for Cultivation and Reference Dataset Creation

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Abstract

The presence of bioparticles in the air, including fungal spores, is a major concern for human and plant health and requires robust and precise monitoring systems. While a European norm based on the manual volumetric Hirst method exists, there's a growing interest in technologies allowing automated real-time monitoring. Most of them rely on machine learning for the identification of bioaerosols. However, the diverse nature of airborne particles in terms of size, properties and composition presents challenges, among which the availability of well-curated datasets for training algorithms. While collecting reference material for pollen is relatively straightforward, current automatic monitoring methods for fungal spores rely on limited training data, hindering broader applicability. This study, which was conducted in the frame of the SYLVA project (GA no. 101086109) and the COST Action ADOPT (CA18226), aims to address this gap by outlining best practices for collecting reference material from controlled cultivation and creating datasets specifically tailored for training algorithms to classify airborne fungal spores. Critical aspects such as access to reference fungal species, *in vitro* cultivation, sporulation yield, clean spore isolation, dry aerosolization, and dataset cleaning have been explored for a series of 17 fungal species from the Belgian fungi collection BCCM/IHEM, including 5 *Alternaria* species with contrasted morphological profiles. Simple classification models were developed as proof-of-principle to assess recognition capabilities from the holography and/or fluorescence data measured by the SwisensPoleno Jupiter (Swisens AG) and laser-induced scattering and fluorescence data measured by the Plair Rapid-E+ (Plair SA). The models were trained using 80% of reference data, while 10% was used for validation to avoid overfitting during training and the remaining 10% was left aside for testing the identification performance. For Plair Rapid-E+, classification accuracy for 7 genera was shown to vary from 0.43 to 0.75 depending on the taxon (F1 score 0.577), recognizing best *Botrytis cinerea* and *Cladosporium* (class created as a mix of 3 species). For SwisensPoleno Jupiter, the initial performance obtained for classification of 8 genera by using only holographic images (F1 score 0.77) has been significantly improved by complementing them with fluorescence measurements (F1 score 0.83). Classification accuracy varied between 0.55 and 0.95 with the best performance for *Curvularia lunata* and *Alternaria* (class created as a mix of 5 species). Differentiation of species was also shown to be possible for *Cladosporium*, with more difficulty for some *Alternaria* species, while

the F1 score remained good (0.72). Overall, this protocol is paving the way for more efficient, standard and accurate automatic identification of airborne fungal spores.

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Optimisation of Germination Test for Birch Pollen

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Abstract

Plants are subjected to environmental stresses due to pollution and global changes like climate warming and rising atmospheric CO_2 levels. Without systematic studies conducted under similar conditions, it is currently not feasible to evaluate how these abiotic stresses affect pollen quality.

This study focuses on birch (*betula pendula*) pollen, which is highly allergenic and commonly found in northern Europe, using *in vitro* germination as an indicator of its quality and viability. Despite its significance, *in vitro* germination of birch pollen remains poorly documented, and no standardized protocol has been established. However, the protocol by Brewbaker and Kwack (1963) is the most commonly used, employing a germination medium composed of 100 ppm H_3BO_3, 200 ppm $MgSO_4$, 100 ppm KNO_3, 300 ppm $CaNO_3$, and 10% sucrose. In practice, mineral concentrations are adjusted to optimize the germination rate of the pollen taxon of interest. In our study, we maintained the magnesium and potassium concentrations at 100 ppm and 200 ppm, respectively. We experimented with various concentrations of boric acid (50-150 ppm) and calcium nitrate (50-300 ppm), as described by Colas and Mercier (1999) as essential minerals for the *in vitro* germination of birch pollen. We added sucrose (10-15%) to our mixtures and conducted experiments in both liquid and agar-based media using Agar-Agar at concentrations ranging from 0.6% to 1%. However, the agar-based medium was quickly abandoned due to preliminary results showing low *in vitro* germination rates.

Birch pollen was sampled in 2023, dried at room temperature for 24 h in a glass desiccator, and then stored at -80°C until use. Two hours before the experiments, it was thawed at room temperature. During this time, a liquid culture medium was prepared. 40 µL of this medium were deposited on a microscope slide, onto which the pollen was sprinkled before being covered with a cover slip. The slide was then placed in an airtight box containing moistened absorbent paper for incubation at room temperature. The germination rate was determined by counting a total of 200 pollen grains using an optical microscope (Zeiss Axio Imager Z2) after 24 h of

incubation. A pollen grain was considered germinated if its pollen tube measured at least three times its diameter (approximately 22 μm).

The germination rates ranged between 31 and 62% depending on the mineral mixture, the optimal culture medium being the one containing 150 ppm of H_3BO_3, 300 ppm of $CaNO_3$, and 10% sucrose. We observed that a higher concentration of boric acid facilitates faster elongation of pollen tubes, enabling optical examination of microscope slides in less than 6 h, typically within a single workday. Note that the growth of pollen tubes may present challenges for counting due to tube entanglement after 24 h. Parallel viability tests conducted using the Alexander staining method revealed a viability ranging between 96% and 98% for the studied pollen. While birch pollen was mostly viable, its germination capacity could be abnormally reduced due to its sensitivity to temperature and humidity variations, posing challenges in standardizing *in vitro* germination protocols.

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Small-Scale Variability in the Bet v 1 Release During Pollination of Betula Pendula in Poznań, Poland

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Abstract

Small-scale variability in the Bet v 1 release during pollination of *Betula pendula* in Poznań, Poland

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***Key words*:** birch pollen, allergenicity, ELISA, day-to-day variation, Bet v 1

Background: Birch pollen, a prominent global allergen, poses significant health risks by inducing respiratory allergies in susceptible individuals. Despite its widespread impact, understanding the temporal and spatial variations in birch pollen allergenicity remains a challenge. This study aimed to elucidate the small-scale variations in the release of Bet v 1, i.e. the major pollen allergen of *Betula pendula*, and their potential drivers, including internal physiological processes and external environmental factors.

Methods: This small-scale experiment was conducted during 10 consecutive days of pollination of *Betula pendula* in Poznań, Poland. Male birch catkins were collected daily from 7 individual trees located within the radius of 500m. Pollen grains released from catkins were sieved, extracted, and subjected to ELISA to quantify Bet v 1 content per pollen. Collected data were compared with daily meteorological data to determine whether the changes in Bet v 1 content are somehow influenced by the effect of weather conditions.

Results: A distinct day-to-day pattern in the release of Bet v 1 was observed. Specifically, the mean amount of Bet v 1 was very low at the beginning of pollination period, reaching its peak approximately one week after initiation of pollen release. Next, a subsequent decrease in Bet v 1 was noticed. Spatial analysis further revealed significant differences in allergenicity among individual trees within the same population, with some trees exhibiting up to threefold differences in allergenicity (ranging from around 5 to 18 pg Bet v 1/pollen). No significant correlations ($p>0.05$) were observed between Bet v 1 amount and selected meteorological variables, such as daily mean temperature and insolation.

Conclusions: The clear pattern in the release of Bet v 1 from pollen suggests that the temporal production of Bet v 1 per individual tree is governed by internal physiological processes, likely related to enhanced allergen production when the catkins (and pollen) are fully developed. Additionally, the individual production of Bet v 1 is likely more related to the genetic or physiological states of the tree rather than external factors. This is strengthened by the fact that none of the included meteorological factors significantly affected the Bet v 1 level.

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Exposure to Allergenic Pollens in a Private Garden

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Abstract

The risk of allergy for people sensitized to pollen during outdoor exposure is well established. Health authorities recommend limiting outdoor activities and outings in order to reduce pollen exposure during peak pollen periods. This study focuses on people's exposure during simple outings in their garden. The study took place in a private garden in the town of Chéreng in the Hauts-de-France region, from 21 March to 2 May 2023, with weekly monitoring of pollen counts in the air by installing a Sigma 2 Like Trap (SLT) passive proximity in the garden at a height of 1.5 m from the ground. The 8-week campaign took place during the main birch pollen season. The collection slides were replaced each week and then observed using an optical microscope to count and identify the pollen grains. Readings were taken over the entire coated surface (24×50mm). These data were then cross-referenced with meteorological data and pollen counts from the nearest Hirst trap, located in Lille, France.

During this field campaign, a total of 26,294 pollen grains were recorded, representing 15 different species. The overall trend shows that 7 of them have a high pollen allergenic potential (PAP=3), 3 have a moderate allergenic potential (PAP=2), and 5 have a low allergenic potential (PAP=1). Two major families of highly allergenic species predominate: birch pollen (Betula), accounting for 55%. and cypress (Cupressus), for 17%. They were followed by ash (Fraxinus), 9%, hornbeam (Carpinus), 6%, oak (Quercus), 4%, and poplar (Populus), 3%. The other pollen taxa (Alnus, Fagus, Ulmus, Corylus, Poaceae) individually accounted for less than 2% of the remaining grains observed.

The weekly trend in pollen counts by species globally followed the different pollen bulletins from the RNSA. Birch pollen grains (BPG) counts showed a gradual increase with a peak during mid-April (17/04). However, during the week preceding this peak, a significant decrease in BPG counts was observed. By correlating our data with the meteorological data, we noted a significant drop in temperatures (down to 5°C) and rather cloudy, foggy weather. Birch catkins probably closed their stamens to protect themselves from the cold, which would explain the drop in atmospheric concentration of BPG following this cooling. We calculated exposure indices (Ie) to describe the relative apportionment of ambien pollen taxa, weighted by their allergenic potential, as follows:

Index of exposure (Ie) = Pollen Allergenic Potential (PAP) × Number of Pollen Grains collected (NbPG) / 1000

Birch (Ie=44) and cypress (Ie=13) recorded the highest exposure indices, followed by ash (Ie=7) and hornbeam (Ie=5). This is in line with observations made using the SLT trap, reflecting the pollination of ornemental trees and shrub hedges planted in and around the garden, and describing local exposure at human height. In conclusion, a simple outing in one's garden exposes individuals to several types of pollen, some of which having a high allergenic potential. Increased short-distance exposure to highly allergenic tree species, along with background exposure, can exacerbate pollen allergy symptoms.

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Grass Pollen Concentration Across Urban-rural Gradient: Integrating Aerobiological Data With Species-level Botanical Survey, Meteorology and Remote Sensing

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Abstract

Grass pollen is considered a global allergen adversely affecting on allergy sufferers. Depending on the region, the sensitization rates can reach up to 30%. Although lots of research has been done on grass pollen, there are still some gaps in assessing dependencies particularly between pollen concentration at ground level and local vegetation at species level. There are evidences that grass pollen concentration at street/ground level can be higher than at roof level but it can also be lower. This variation may result from local wind conditions, obstacles, vegetation structure, species composition and others. We hypothesized that grass pollen concentration at roof level will reflect the concentrations measured at different sites (urban, forest, agriculture). Moreover, we expected to assess the contribution of particular grass species to the total grass pollen pool.

Burkard portable volumetric traps were used to measure the hourly concentration (HC) of grass (Poaceae) and separately rye (*Secale cereale*) pollen across the urban-rural gradient in Poznan, Poland. During intensive monitoring campaign with simultaneous measurements at 11 sites (10 sites at 1.5 m a.g.l. and 1 trap at roof level) in May and June 2023 we collected 726 hourly samples. Also, we measured wind direction and speed at three sites using Kestrel 5500 weather stations. In the vicinity of the monitoring sites (up to 30 m) we performed also a detailed botanical survey for grass species and assessed the land cover using aerial photographs and laser scanning.

The grass pollen HC was the highest at sites with meadows (average >44 pollen/m^3; absolute maximum: 854 pollen/m^3) where 9-15 grass species were recorded. The lowest HC was noted at sites where grass was mowed before the pollen season, the number of grass species was 2-3 and low vegetation covered < 30% of the area within a 30 m buffer. But there were exceptions such as relatively high HC (up to 227 pollen/m^3) at one urban site with only 16% vegetation cover, caused by *Lolium perenne* dominant flowering in the second half of June. We also observed high HC of rye pollen close to rye field (134-445 pollen/m^3) but only 1-3 rye pollen/m^3 were simultaneously found in samples collected merely 145 m further at ground road between oat (*Avena sativa*) fields.

We can conclude that assessing the real exposure to grass pollen using only the rooftop data may give inconsistent results, because of large differences between close ground sites, even with very similar land cover. Completing aerobiological measurements at ground level with detailed botanical survey gives the information on grass pollen concentration at species level directly in places where people live. As there is a clear link between meadows and rye fields and high grass pollen concentrations in the vicinity, the detection of such areas using remote sensing may be efficient in indicating places of high risk of the exposure to Poaceae pollen.

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Twenty Years of Aerobiological Monitoring of Allergenic Proteins in Poznań, Poland – Achievements and Perspectives

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Abstract

Background: The monitoring of allergens in the air of Poznań, Poland, celebrates its 20th anniversary. This exciting scientific journey began in 2005 with participation in the EU-funded project MONALISA (LIFE05 ENV/F/000068). Over the following years, the Laboratory of Aerobiology (LA) has been involved in several other international and national projects focused on allergen detection, both as a partner, e.g., in HIALINE project (No 20081107, EAHC) and COST Actions (ADOPT CA18226), and as a coordinator (NN404015439, NCN 2011/03/D/NZ7/06224, and NCN 2013/09/D/NZ7/00358). Allergen research is still ongoing, primarily focusing on investigating the allergenicity of Cupressales pollen and determining the impact of various environmental factors on pollen allergen properties (NCN 2020/39/B/ST10/01554). Additionally, the effect of fungicides and fungal infections on allergen production is studied within Doctoral and Master projects.

Methods: The primary method used for allergen collection and quantification involved the integration of state-of-the-art samplers, e.g. cyclones and cascade impactors, along with an immunoenzymatic assay (ELISA). These methods were progressively complemented with mRNA transcript analysis, electrophoresis, and flow cytometry. The investigated allergens included Phl p 5 (a major grass allergen), Bet v 1 (birch), Art v 1 (*Artemisia*), Amb a 1 (ragweed), and the major allergen of *Alternaria alternata* spores (Alt a 1). Airborne monitoring was supplemented with experimental analysis of allergens extracted directly from plants or fungal colonies, extending beyond the standard quantification of aeroallergens.

Results: The effects of the conducted research are impressive. It includes not only 8 published articles (cited almost 500 times) but also extensive transfer of knowledge and new collaboration opportunities with foreign scientific units, leading to the further development of competences, experimental techniques, and overall scientific experiences of LA. The most important results concern the estimation of spatio-temporal variation in the allergen content in the air (e.g., for *Ambrosia* allergens), its relationships with pollen and spore levels, detection of airborne subpollen (and subspores) fragments (in PM10, PM2.5, and PM0.12 air fractions), and determination of population-specific (e.g., for *Betula pendula*) and species-specific differences in pollen allergen levels (e.g., within three *Artemisia* species).

Conclusions: The 20-year journey involving allergen detection and quantification is truly fascinating. It clearly demonstrated that for a thorough assessment of exposure to airborne allergens, aeroallergen monitoring should be an integral part of traditional pollen/spore

monitoring. There is still much to investigate, such as the need to incorporate allergen data into forecasting models, determining the environmental factors affecting pollen/spore allergenicity, and defining the appropriate thresholds of allergens for allergy symptoms. This is not the end of the road…it's just a beginning.

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ExoPLANiTS: The Curated Database for Fungal ITS Metabarcoding in Aerobiological Studies

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Abstract

One of the challenges in the field of environmental DNA analysis is represented by the relatively high rate of mislabeling of sequences in vast, publicly accessible, yet not curated databases such as NCBI. This issue complicates the accurate identification and cataloging of biodiversity, as the presence of incorrect or mislabeled sequences can lead to errors in data interpretation and subsequent analyses.
Building on the foundation of our curated database PLANiTS, we present exoPLANiTS contributing to the innovation and improvement of the resolution in environmental DNA metabarcoding studies. This database incorporates fungal sequences, extending the original plant-only ITS database that is PLANiTS. This expansion is aimed to significantly enhance the comprehensiveness and accuracy of biodiversity assessments, catering to a broader spectrum of environmental taxa by a blend of manual and automatic curation.
The methodology behind exoPLANiTS involves sequence extraction from NCBI, homology-based identity checks to ensure the accuracy of the data, and employment of clustering algorithms to minimize redundancy. The bulk of this process, which optimizes the selection of representative sequences based on the composition of clusters at various taxonomic levels, can be fully automated by leveraging open source software such as blast, vsearch and cd-hit, and python libraries such as ete3. This minimizes the required manual intervention and ensures the feasibility of periodic updates and mantainance.
From an initial dataset of 2.3 million sequences retrieved by querying ncbi, we refined it to 606,203 representatives by clustering at 99% similarity, extracting associated taxonomic information, and computing concordance statistics across seven taxonomic levels. Clusters of

at least 10 sequences showed genus and species concordance below 95%, with about 22,000 clusters having genus concordance under 90%. Using ITSX software, we extracted ITS1, ITS2, and completed ITS subsequences, re-clustered them at 100% identity, and then aligned the representatives against the UNITE database using BLAST. This process identified and removed ~10% of sequences likely from non-fungal sources, resulting in a refined database of 117,531 ITS1, 147,229 ITS2, and 76,136 complete ITS sequences. The taxonomy field was updated for each representative, based on cluster composition and a 90% consensus rule for clusters with 10+ sequences; those with fewer sequences required 100% concordance for assignment. Singleton clusters were flagged. Validation involved comparing our taxonomy assignments against UNITE database entries, enhancing accuracy up to the family level.
To test the precision and sensitivity of exoPLANiTS, we employed it on the reiteration of three studies performed by our group in the past. Briefly, these studies were aimed at exploring the variability of aerial plant and fungi communities through time and space in several region of Italy. Overall, using this database we were able to replicate the previous results, and improved them by increasing the taxonomical resolution of OTUs.
In conclusion, exoPLANiTS represents a significant advancement in the field of environmental DNA analysis. Its development and successful application in diverse studies underscore the importance of integrating comprehensive and accurate metabarcoding techniques in biodiversity research.

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How the Birch Pollen Proteins Affect the Immunological Response of Allergic People?

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Abstract

INTRODUCTION

Pollution, as an abiotic stress factor may affect a different immune response of individuals with birch allergy. The goal of the study is to estimate, whether the observed differentiation in

physico-chemical properties of birch pollen proteins is associated with the variability of their immunoreactive and allergenic properties.

METHODS

Birch pollen material was collected from 27 birch specimen, at selected 9 sites in Kraków and at the less polluted areas, up to 80 km of the city, in 2022. The study group contains 30 individuals with confirmed allergy to birch pollen and manifesting clinical symptoms during the pollen season (22W, 8M; mean age 44.2 yrs, ±14.4), while to the control group, 30 persons without birch allergy were included (25W, 5M; mean age 41.8 yrs, ±11.6). In all patients sIgE concentration against the main seasonal inhalant allergens, including birch extracts and the selected birch pollen components (Bet v1, Bet v2, Bet v4, Bet v6) (Enzyme-Allergo-Sorbent-Test, Euroimmune Polska) was measured. The following analyses were performed in all pollen samples: total protein content (Bradford method); Bet v1 concentration (ELISA Assay); UHPLC-DAD and UHPLC-DAD-QqToF-MS analyses of birch pollen UAE extracts and the total flavonoids content determination (colorimetric assay); and SDS-PAGE using Mini Protean II electrophoretic apparatus (BioRad, USA). The selected samples from all studied sites were used to immunoblotting with patients sera. Friedman's rank ANOVA test was performed and Kendall's coefficient of agreement was calculated to analyze whether is a difference in the number of protein bands among tree specimen and how the patients react similarly to them.

RESULTS

Total protein content ranged from 641.67 µg/ml to 2068.59 µg/ml and was significantly higher in pollen samples in Kraków vs out of Kraków ($p=0{,}04$), while in the case of Bet v1 concentration the differences were not statistically significant. Total flavonoid content expressed as rutin equivalent ranged from 5.04 to 11.83 mgRE/g. The major compound from this group was quercetin-3-*O*-sophoroside (2.1 – 12.4 mg/g), a natural ligand of Bet v1 and kaempferol 3-*O*-hexosyl-deoxyhexosyl-hexoside (0.2 – 5.4 mg/g). Beside flavonoids birch pollen contained phenolamides with N',N''-diferuloylspermidine isomers as main components (7.1 -19.6 mg/g).

In all patients and in both groups, separately, a statistically significant differences in the number of bands between specimens was found ($p<0.05$). The low values of the Kendall coefficient (for the whole group; for allergic patients and for the control individuals we as follows: 0.26; 0.23 and 0.36, respectively), suggest a lack of consistency between patient reactions to individual specimens, what would indicate that a given specimen may induce different reactions in individual patients.

CONCLUSION

The results indicate that environmental conditions may affect the immunological response of allergic persons to the natural pollen allergens.

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Performance of the Swisens Poleno Jupiter Particle Classifier: Comparative Analysis and Local Sample Training in Ireland

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Abstract

The Swisens Poleno Jupiter represents the cutting-edge of real-time flourescent particle detection and measurement, specifically designed for real-time bioaerosol monitoring. The initial classification model was originally designed by MeteoSwiss and was developed in central Europe, based upon 14 types of pollen commonly found there. In order for it to function accurately in other regions, it requires adaptation to suit the local conditions, which leads to substantial increases in data quality post adaption. Swisens developed a model that combines the power and abilities of multiple algorithms while also incorporating fluorescence data, in a novel approach to bioaerosol monitoring and modelling. In Dublin, an initial Hirst comparative assessment was carried out, and subsequent to that, samples of 18 types of locally obtained pollen were atomised into the Poleno device as part of a training procedure for new model development. After training and deployment on the bioaerosol dataset, changes in Hirst-Poleno correlative accuracy, and thus model accuracy changes, were determined. Both processes are described herein, with

The Swisens Poleno Jupiter arrived in Dublin with its dataset based upon pollen types collected in central Europe. From initial operation in the Spring of 2023, until the end of the calendar year, both the Swisens Poleno Jupiter, and a traditional Hirst 7-day volumetric sampler were co-located on the roof of the Irish meteorological headquarters, the Met Éireann pyramid. This campaign found varying levels of agreement between the Hirst, and the Poleno devices, depending upon the pollen type monitored. Little to no correlation was seen for types such as *Corylus* and *Alnus* (*Alnus* $R^2 > 0.1$), while more encouraging values were observed for *Betula* and *Cupressaceae* (*Cupressaceae* $R^2 > 0.6$).

After this a second phase campaign was initiated in 2024. Common pollen types found in Ireland were collected in the immediate surrounding locality and atomized into the Poleno Jupiter, starting with *Corylus* and *Alnus* pollen in January. The primary objective was to train the new Irish-based model, fine-tuning it for local conditions and paving the way for nationwide device rollouts. The Hirst-comparative results of the second model were compared with those from the previous model, to identify any changes in accuracy between the devices. This most recently developed dataset correlated more with the Hirst method data than the previous generalised European dataset, with many pollen types now having R^2 values of greater than 0.8.

As rollout of more Poleno devices commences during the network expansion phase, the training process will be repeated for each device, once installed in its respective location. Regular

updates based on native Irish pollen types will enhance accuracy and ensure reliable monitoring results. These additional sampling locations will follow the same system as this initial campaign, with a co-located Hirst traditional sampler in operation alongside the new Swisens Poleno devices. The establishment of the network of monitoring stations nationwide, will provide a first characterisation of the bioaerosol variations already present throughout the islands' diverse ecological landscape.

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Assessment of the Reproductive Capacity of the Invasive Species Amorpha Fruticosa in the Framework of LIFE PRIMED Project

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Abstract

Climate change causes biodiversity loss and facilitates invasive species proliferation, notably in coastal ecosystems. Ecological restoration efforts in Greek and Italian coastal areas, specifically the 'Nestos Delta' and 'Bosco di Palo Laziale', target habitats like 'Pannonian-Balkanic turkey oak-sessile oak forests' (91M0), 'Alluvial forests with *Alnus glutinosa* and *Fraxinus excelsior*' (91E0*), and 'Mediterranean temporary ponds' (*3170). These habitats face increased vulnerability due to climate change and poor forest and water management practices.

During the implementation of the LIFE PRIMED project (LIFE17 NAT/GR/000511), the invasive species *Amorpha fruticosa L.* was identified in the 'Nestos Delta'. Originating from southwestern North America, *A. fruticosa* was initially introduced to Europe for ornamental purposes but has since evolved into a hazardous invasive species across southeastern Europe. It poses a threat to the 91E0 * priority habitat by suppressing native tree species, altering successional sequences, and reducing biodiversity. Our study focused on *A. fruticosa* reproductive potential across nine 1x1m^2 plots along the western banks of the 'Nestos River'. The high number of inflorescences (176 ±5) and fruits (115,412±6,462) per m^2 underscores its significant reproductive capacity. This was corroborated also by seed germination experiments we conducted, using *A. fruticosa* seeds collected from the Nestos area. Results indicated a germination rate exceeding 50% in both light and dark conditions.

As a result of the above, an invasive control plan is urgently required. Initiatives are underway in the Nestos Delta through the LIFE PRIMED project to limit the spread of *A. fruticosa*, employing seven distinct treatments: (a) vegetation shading, (b) vegetation enhancement with *A. glutinosa*

seedlings, (c) vegetation enhancement with *Populus alba* cuttings, (d) repeated removal of *A. fruticosa*, (e) ground covering with geotextile, (f) grazing and (g) control area. *A. fruticosa* and *Ambrosia artemisifolia*, both Invasive Alien Species (IAS), invade by occupying similar functional spaces to native species. Both species share physiological traits and are expected to respond similarly to invasive control plans. Bioecological studies suggest that applying control measures like mowing during flowering in European forest areas could mitigate IAS regeneration and reproduction rates, based on climatic indicators and traits of *A. artemisiifolia*. Further research and After-Life application are strongly suggested due to their presence around the 'Nestos River' pilot area.

The EU-funded initiative LIFE PRIMED (LIFE17 NAT/GR/000511) operates in the Delta of River Nestos in Greece and the Forest of Palo Laziale in Italy. Initial findings suggest collaborative efforts to develop water harvesting systems and adaptation strategies can enhance water resilience in degraded forest ecosystems. The project has tackled erratic rainfall impacts by installing hydraulic systems in Palo Laziale and a wellpoint-based water network in the Nestos Delta. Given rapid environmental changes, studying, and managing invasive plant species is crucial.

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Evaluation of European Pollen Reanalysis

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Abstract

The European Pollen Reanalysis for alder, birch, and olive airborne pollen was released at the end of 2023, with only technical validation of the procedure presented in the paper accompanying the dataset. The current presentation provides a first glance on the quality of the reanalysis and highlights its strengths and areas of improvement. The analysis is performed at a few individual stations in different parts of Europe: Finland, Latvia, Lithuania, Belgium, and Spain.

The reanalysis was built using only assimilation of the total seasonal pollen production, i.e. the phenological models and their parameters were left completely intact. This allowed an almost-

complete separation of the absolute values of the predicted time series and their temporal behaviour. The first ones were corrected during the assimilation, the second ones were practically the same in both the initial SILAM run and the final reanalysis.

SPIn time series at individual stations.

The Seasonal Pollen Integral at individual stations is the most-affected parameter by the assimilation: with exception of the long-range transport, SPIn is directly proportional to the regional pollen production. Not surprisingly, the reanalysis demonstrated a strong improvement of this parameter practically at all stations and all tree genera. The most significant improvement was for birch pollen, except for Southern Europe, where local pollen production is very small, and the season is almost entirely decided by the long-range transport.

Absolute bias and RMSE.

The reanalysis is generally unbiased by its design. However, predictions in some individual years and at specific stations could manifest noticeable deviations, offset in other years. As a result, RMSE was mostly decided by the temporal correlation: low-correlating stations showed higher RMSE.

Temporal correlation at individual stations.

The intra-seasonal concentration evolution was not affected by the assimilation (except for the changes in the long-range transport), so the model skills remained practically the same for the initial and final runs. The most-challenging genera was alder, which SILAM considers as a single taxon. Such simplification was acceptable in regions with only one dominant species or with close flowering times but led to significant errors if several species flowered at different times in the same region. The birch timeseries are less sensitive to this feature because different taxa almost always have close flowering periods.

Season start and end.

These are the parameters practically not affected by the assimilation. Therefore, the key features of the operational SILAM version have been manifested in all three genera. The model was reproducing the season start with a comparatively high accuracy (majority of the considered stations showed just a few days of an error), whereas the season end was quite significantly delayed.

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Improved Correlations Between Burkard and Automatic Pollen Monitor When Complementing Holographic Images With Fluorescence Spectroscopy

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Abstract

The number of people affected by pollen allergies has increased significantly over the last decades and allergic asthma accounts for a substantial number of deaths annually. Current automatic pollen monitoring includes only a limited range of allergenic pollen, necessitating advancements for more precise information. The SwisensPoleno Jupiter produced by Swisens AG, Switzerland, combines imaging and biochemical information in the form of fluorescence spectroscopy for single particle characterization. These two methods complement each other, allowing for classification and differentiation of more relevant pollen species. This has already been shown in previous work, however only in laboratory conditions, while here, a classification model with many pollen taxa and other aerosol particles has been trained and applied to outdoor measurements. The new classifier was trained on 27 pollen taxa, containing over 300,000 measured particles. The performance increase achieved with holographic images and fluorescence spectroscopy was assessed with data from the EUMETNET AutoPollen intercomparison campaign in Munich, Germany, in 2021, where automatic instruments were operated co-located with _Burkard® traps. Correlations between SwisensPoleno Jupiter and daily averages from four Burkard® increased by 35 to 64%, compared to the previous classifiers based on holographic image data only. For Fraxinus a correlation as high as R^2 0.93 (previously R^2 0.62) was achieved, while for Poaceae it reached R^2 of 0.92 compared to R^2 0.56. Quercus improved from R^2 0.68 to R^2 0.92. These improvements are driven by a significant reduction in off-season misclassifications.

Efforts continue to analyze data from several SwisensPoleno Jupiter and assess results in countries other than Germany.

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Allergenicity of Birch Pollen and Associated Health Effects: in Quest for Responsible Environmental Factors

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Abstract

Environmental stress associated with ongoing climate change and air pollution can impact pollen allergenicity, modify pollen characteristics and raise the frequency and intensity of allergic responses. The aim of the study was to investigate the allergenic potential of pollen from birch trees grown in various climatic conditions but with limited genetic variability from clones in International Phenological Gardens across Europe, and to identify the environmental and pollen-intrinsic factors involved. Along with the molecular composition of the pollen, the skin prick test (SPT) response of allergic individuals was assessed.

Between 2019 to 2022, we collected pollen samples from birch trees across Europe. We collected data on environmental parameters, namely air temperature, relative humidity, precipitation, urbanisation Index, and levels of NO_2, O_3 and CO_2, as well as tree-specific factors (tree age and size, and potential presence of virus infection of pollen). These environmental factors were investigated for relationships with allergenicity-related parameters, such as content of total protein, Bet v 1, lipid mediators, endotoxin, serotonin, and histamine in the pollen extracts, additionally to the SPT reaction of patients to the corresponding pollen extracts.

The severity of the SPT reaction was significantly and positively correlated to Bet v 1, total protein and histamine content of the pollen samples. Bet v 1 was correlated positively with air temperature, NO_2, and CO_2, but with a high inter-annual variability signifying complex interaction effects among all variables.

Our findings highlight the influence of climate-change-related parameters on allergenicity and associated skin responses: warmer locations and/or years, with higher air pollution favour the production of more allergenic birch pollen and increased skin reactions in sensitized individuals.

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Establishing a Methodology for Airborne Corona Viruses Detection in Outdoor Air

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Abstract

Airborne viruses, such as influenza, corona, and rhinovirus, are responsible for many respiratory infections, some causing the spread of severe acute respiratory diseases like the recent pandemic of severe respiratory syndrome coronavirus 2 (SARS-CoV-2). The establishment of monitoring method could determinately contribute for the early detection of respiratory viruses in the air and to anticipate mitigation strategies. It was aimed at developing a methodology to detect viruses in outdoor air.

The sampling was performed using a high-volume cascade impactor (CHEMVOL, Butraco) with 2 stages (PM>10 & PM10). Filters were preserved at -80ºC. Total RNA extraction was performed with the Phenol-Chloroform method using TRItidy GTM reagent according to the manufacturer's instructions. The commercial E.Z.N.A.® Total RNA Kit-I was used to RNA purification. Real-Time Reverse Transcription PCR was executed to detect the N-gene from the Sarbecovirus family and RdRp gene from SARS-CoV-2 using the ViroReal® Kit SARS-CoV-2 Multiplex. A protein-rich fraction was obtained with ammonium bicarbonate buffer extraction followed by lyophilization. Spike protein was assessed by specific SARS-CoV-2 Antigen Test Kit.

The samples from the last week of December 2020, first and second weeks of January 2021, from both PM>10 and PM10, were positive for the N-gene and Cq>33, identifying Sarbecovirus family. The RdRp gene was undetectable, probably due to low virus concentration. The protein extracts from the same periods tested positive for the specific antigen spike protein.

In conclusion, all results combined confirm the detection of airborne corona virus and establish the bases for a molecular-based method for virus monitoring in ambient air thus eventually providing the base for early alert systems allowing the implementation of preventive measures to control outbreaks and mitigate future pandemics.

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New Approaches for Bioaerosol Automatic Identification With the SwisensPoleno

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Abstract

In the last few years, operational measuring networks with automatic bioaerosol monitoring instruments started to emerge. This monitoring is based on different types of data: image, fluorescence or light scattering measurements taken automatically by the instrument and coupled with identification algorithms. Currently, those algorithms mostly rely on classical supervised machine learning methods. Specifically, image inputs are processed through convolutional neural networks and fluorescence/light scattering data through fully-connected neural networks or Support Vector Machines. Currently, the development of new identification models is progressing and follows advances in terms of measurement and algorithms. Here, we present recent research focusing on pollen identification with the SwisensPoleno (Swisens AG). The results and methods are expected to extend to other bioaerosol monitoring instruments as well.

We first show that combining holographic images and fluorescence inputs improves the quality of identification compared to single input models. We compare the performance of three neural network models with different inputs. We observe that information on shape and composition complete each other well in the case of pollen identification. The global balanced accuracy for the combined model (images + fluorescence) reached 99.2% while the holography- and fluorescence-based models respectively reached 96.8 and 87.8%. Those results encourage the use of multiple input models as a standard, especially when considering other types of bioaerosol that have variable shapes/compositions.

Nevertheless, high identification performance with supervised learning methods as above is obtained at the cost of data preparation, i.e., create, clean and label the training data, a workload expensive task. As opposed to supervised learning, self-supervised methods do not require labelled data as they exploit the data's structure to derive their own labels. These methods allow us to consider operational data as a source of information for training the algorithm. In this second study based on images only, we tested the performance of SimCLR, a self-supervised method trained on operational data. Coupled to few-shot learning, the SimCLR output can be calibrated to known classes using only a few labelled images (50 or less). Promising results were obtained with a global balanced accuracy starting at about 50% when a single example is given for few-shot learning and increased to the range of 75-85% when 50 examples per taxon were given. Instruments relying on images, such as the BAA500 (Helmut Hund GmbH) and the SwisensPoleno can greatly benefit from this approach as it is easy to gather a few examples from operational data via visual inspection and/or comparison with Hirst-type parallel manual measurement.

In conclusion, both exploratory studies pave the way for including more diverse particle types in automatic monitoring such as fungal spores and other (non-)biological particles. The first improves the identification performance and the second considerably lowers human effort.

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Pollen and Fungal Spore Monitoring in Germany – More Than 20 Years of the Third Millennium

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Abstract

Introduction:
In Germany, the lifetime prevalence of allergic rhinoconjunctivitis in adults is 14.8%. German adults are most commonly sensitised to *Poaceae* (specifically *Phleum)* pollen, followed by *Betula*, *Poaceae* (specifically *Secale*), *Alnus, Corylus, Fraxinus, Artemisia* and *Ambrosia*. In the management of pollen allergy, prevention represents an important complement to treatment, medication, and/or immunotherapy. To monitor airborne pollen in Germany and inform the public, the non-profit, non-governmental German Pollen Information Service Foundation (PID) was founded in 1983.

Methods:
The PID monitors airborne pollen using volumetric spore traps (Hirst type) with subsequent light microscopy pollen analysis of the collected samples following VDI-4252-Blatt-4:2019-03 (Germany) and DIN-EN-16868:2019-09. The PID monitoring network consists of 35 monitoring stations across the country (as of February 2024). Besides the main pollen taxa (*Alnus, Ambrosia, Artemisia, Betula, Corylus, Fraxinus, Poaceae, Secale*), numerous other pollen taxa are monitored which, albeit in lower proportions, may also induce allergies in the population (e.g. *Fagus*, *Quercus*, *Plantago*) or are of ecological or local importance (e.g. *Pinus*). Annual Pollen Integrals (APIn) of the 8 main pollen taxa and 3 fungal spores (*Alternaria, Cladosporium, Epicoccum*) are presented in graphical form together with the coefficient of determination (R^2) for the years 2001–2023 (*Ambrosia* from 2006). Daily average pollen concentrations were calculated for each pollen taxa based on the concentrations of the monitoring stations active on each day (01.01.–

31.12.), and then summed to the pollen taxa-specific APIn for each year. The same method was applied to fungal spores.

Results:
The APIn graphs indicate an increasing trend in pollen concentration for *Corylus* (R^2 = 0.39), *Alnus* (R^2 = 0.17), *Poaceae* (R^2 = 0.14), *Betula* (R^2 = 0.07), *Ambrosia* (R^2 = 0.05), *Fraxinus* (R^2 = 0.03) and a decreasing trend for *Artemisia* (R^2 = 0.17) and *Secale* (R^2 = 0.12) over the last 23 years in Germany. The trend in fungal spore concentration in Germany is increasing for *Alternaria* (R^2 = 0.14) and *Cladosporium* (R^2 = 0.14]) (R^2 was not calculated for *Epicoccum*). The PID also focuses on pollen of plants that are spreading within Germany, enriching the country's pollen spectrum (e.g. *Ailanthus*, *Acer negundo* [= *Acer*]) or prolonging the pollen season of certain pollen taxa (*Alnus* × *spaethii* [= *Alnus*], ornamental and late flowering *Poaceae* species) and that may have an impact on the health of the German population. The monitored pollen data for up to 50 pollen taxa are processed textually by PID into detailed weekly pollen forecasts (https://www.pollenstiftung.de), and data for 8 pollen taxa are integrated by the German Weather Service into daily graphical pollen load forecasts (https://tinyurl.com/4yzzjux2). As an additional preventive tool, Germany-wide pollen calendars are issued by PID every 5 years, most recently in 2023.

Conclusion:
This presentation highlights the importance of long-term and continuous pollen monitoring i) to gain insight into changes in the pollen spectrum and pollen counts, and ii) as one of the foundations of allergy prevention tools. In addition, allergy prevention needs to know where to find reliable information on the countrywide pollen and fungal spore situation.

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Airborne Pollen and Fungal Spores Six-years Monitoring in a Desert City of Northwestern Mexico: Impacts on Population Health and Relationships With Climate Change.

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Abstract

Aerobiological studies are still scarce in northwestern Mexico where allergenic
pollen have great impacts on health. Current global pollution and climate change
problems are
closely related to many allergic diseases, enhancing the need to continue researching these is
sues and improve life quality. The city of Hermosillo, Sonora has a high rate of allergic disease
s associated with poor air quality. In recent years, the allergic reaction to biological
agents such as pollen and fungal spores has increased, according to allergy experts in the city.

Aerobiology studies agree that poor air quality related to atmospheric particulate matter a
nd the effect of climate change
such as increased ambient temperatures can potentiate the effect of
these aeroallergens on population health. Because warmer air temperatures can bring for
ward the start of plant growth and the start of flowering, leading to an earlier- starting and lon
ger-lasting pollen season. In this work, there are 6 years of data on the concentrations of
pollen and fungal spores in the air of the city, which were monitored following the
standardized methodology with a Hirst-type spore trap proposed for global aerobiological
studies (for example, Red Mexicana of Aerobiology). Climatic variables were also
analyzed for the same six-year period to explore statistical
correlations between climate and airborne pollen concentrations in different seasons. Likewise,
data from the
General Hospital of Social Security for the city was recovered, the results of the records of pati
ents treated for allergic diseases and specific tests of allergic sensitivity to different
types of pollen and fungal spores was obtained for some years. Statistical correlations were
carried out in order to establish the type of relationship that exists between the
concentrations of pollen and fungal spores in the air with the effects on the health of the
population.
The results show that there are two periods of the year where there is very poor biological air
quality: between March-April and from August to November (mean of
six years). Correlating with the largest number of cases of positive allergic sensitivity tests to di
fferent types of pollen and spores. The city's population responds almost immediately to
an increase in the concentration of pollen and/or fungal spores in the air. Among the most
abundant
types of pollen in the air with a high level of allergenicity are Grasses (Poaceae), quelites (Chen
opodiaceae-Amaranthaceae), ragweeds (*Ambrosia*) and some
trees such as Mezquite (*Prosopis*) and Palo Verde (*Parkinsonia*). While the
most abundant fungal spores in the city are

Alternaria, *Cladosporium* and Basidiospores. In these 5 years, the correlation analyses for these two peaks indicate that a link exists between pollen in the air and decreases in precipitation and temperatures, and an increase in relative humidity. An inter-annual variability in pollen concentrations was recorded related to different weather condition s suggesting an effect of climate change. This work provides useful information for the general population about the times of year where we have increasing aeroallergens at this city and for health authorities to know which biological agen ts cause most the allergic reaction in the population of Hermosillo.

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Automatic and Real-time - the Ongoing (R)evolution in Aerobiological Monitoring

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Abstract

In less than a decade, what once seemed a distant goal has become a reality. In situ pollen and fungal spore data can now be obtained automatically in real time and at high temporal resolution. Technological advances are making it possible to gain a much more detailed understanding of the parameters that control aerobiological phenomena, and thus to improve forecasts. Further developments may also make it possible to monitor other large aerosols, such as microplastics. In Europe, the users' community is actively developing validation and standardisation methods, as well as a common infrastructure that will provide easier access to these technologies and to the data produced. With these new developments, aerobiology - the study of bioaerosols - is entering a new dimension, with cutting-edge tools to serve fields as varied as health, agriculture and research.

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World History of Aerobiology

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Abstract

It has long been suspected that atmospheric particles could affect living beings. Hippocrates (460-377 b. C.) maintained that "*Man can be attacked by epidemic fevers when he inhales air infected with pollution hostile to the human race*". Lucretius Caro (98 b. C. - 54 b. C.), presented that "…*When the sun's rays let in pass through the darkness of a shuttered room, you will see a multitude of tiny bodies all mingling in a multitude of ways inside the sunbeam, moving in the void…*". The first references related to pollen appeared during the 15th century, with Monardi (1462-1536) studying the stamens in flowers and Van Helmont (1577-1644) presenting the "*rose catarrh*".

The origin on aerobiology can be traced back during the 17th century with the "*first microscopy*" building by Leeuwenhoek (1632-1723). Micheli (1679-1737), illustrated the "*seeds*" of many fungi, discovering that these particles can be contaminants transported by the air.

In the 18th century, Sprengel (1793) presented the adaptive mechanism for airborne pollination; and Night (1799) added that the wind could transport pollen over great distances.

At the beginning of the 19th century was accepted the fact that pollen from some plant species, and spores from ferns, mosses, and fungi, were usually released into the air and wind transported. As an interesting example, important to remark the great role of Darwin (1809- 1882) and Ehrenberg (1849-1872) in Aerobiology. When Darwing traveling in the "Beagle", on a scientific expedition, he found near the Cape Verde Islands dust transported in the atmosphere from North Africa; Ehrenberg helped in the identifications of organisms, both supporting the importance of long-distance transport. Pasteur (1822-1892) studied airborne particles using a volumetric method; and Miquel (1850-1922) carried out the first long and periodic sampling of the atmosphere with volumetric methods. In application to Medicine, Bostock (1773-1846) presented to the Royal Society of Medicine the "*catarrhus aestivus*", and Blackley (1820-1900) demonstrated this sickness experimentally with himself, introducing the term of "*hay fever*".

The term of "Aerobiology" was introduced with Meier (1893-1938). In 1964, the *International Biological Program* (IBP) supported this discipline, and the NASA funded the Atmospheric Biology Conference. IBP officially ended in 1974, and the *International Aerobiology Association* (IAA) was created in the first International Congress of Ecology, held in Hague in July 1974.

At the middle of the 20th century, and interesting discussion was focused on the quality results obtained with the different spore traps in used; for example, in a symposium on Aerobiology, held by the Linnean Society of London in 1957, Gregory (1907- 1986) criticized that *"…the various kinds of spore trap in use up to 1950 shows that freely exposed traps could never provide unbiased*

estimates of all constituents of the air spore at the frequent intervals necessary to show that their numbers were affected by changing weather". Today, the most used instrument in the world is the Hirst type spore trap, a method counting with the European norm EN16868, 2019.

However, at the beginning of 21st century, special interest arose for automatic pollen and fungal spore counting systems. Today, these studies progress in the frame of the EUMETNET AutoPollen Programme for "*Serving as a Proof-of-Concept for a European automatic pollen monitoring network using high temporal-resolution real-time measurements*".

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Aeromycology: Research Progress and Future Challenges

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Abstract

The fungi are omnipresent, abundant constituents of virtually all ecosystems on Earth. They serve diverse functions exhibiting an unusually high degree of pleiomorphism and have evolved to utilize air as their key dispersal medium. The role of airborne fungi is of much concern for human affairs. The interest for their study was triggered by phytopathogenic and allergenic fungi and nowadays has been expanded to fungi in houses, working environments and schools for well-being of occupants, in hospitals for detecting infectious agents, in industries for safety of products, in libraries, museums and galleries for protection of cultural heritage, in ambient air for air quality and correlation to pollution, in natural processes for biometeorology, etc.

A systematic study of airborne fungi was launched in early 20th century, although their presence in the air has been documented much earlier. The gravitational method on glass slides or petri plates were originally used. It was the mycologist J. Hirst who recognized the need for a volumetric sampler with time resolution and engineered in 1952 the first spore-trap slit impactor that is widely used until today. The development of several samplers were based on trapping of fungal spores either on adhesive surfaces for direct identification or on nutrient media for recovering fungi. The two strategies are complementary, but each one has limitations that even their combination cannot eliminate. The advent of genetic analysis revolutionized aeromycological studies. Molecular tools were employed in order to contribute to accurate identification of common species in the air, in large genera like *Aspergillus*, *Cladosporium*, *Penicillium* or non-sporulating fungi. Furthermore, the high throughput sequencing enabled the direct analyses of environmental DNA originating from the air revealing a wealth of fungal diversity dominating the atmosphere and in much higher concentrations than anticipated. Metagenomics, metatrascriptomics and metabolomics are very promising for aeromycological

studies. In addition, sampling devises have been developed which are able to detect, analyze and identify airborne fungal spores and pollen by light scattering, laser-induced fluorescence or holography, using signal or image processing in order to classify spores in real time or near real time.

A plethora of fungal species has been documented in the air worldwide, both outdoors and in diverse constructed habitats. Nonetheless, our knowledge of their occurrence in air is still incomplete, due to the difficulty in identification and to the dissimilarity of investigation methods. For monitoring and development of a world net on airborne fungi, there is an urgent need for standardization of methodology based on classical and modern methods. This can be achieved by collaboration of research groups moving toward complementary practices. An holistic approach incorporating additional evidence into existing knowledge will advance aeromycology in a new era.

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Embedding Mask-air (Digital Health) and Silam (Aerobiology Modelling) for an Optimal Control of Allergy to Pollen

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Abstract

Plant species vary under different climate conditions and the distribution of pollen can be used to assess the impact of climate change. In 2015, MASK-air® (Mobile Airways Sentinel networK for rhinitis and asthma) was launched as a project of the European Innovation Partnership on Active and Healthy Ageing (EIP-on-AHA, DG Santé and DG CONNECT) to develop an early warning system to inform patients about the beginning of the pollen season. SILAM (System for Integrated modeLling of Atmospheric composition) is a global-to-meso-scale dispersion model developed by the Finnish Meteorological Institute (FMI). It provides information on atmospheric composition and air quality as well as pollen and pollution. POLLAR (Impact of Air Pollution on Asthma and Rhinitis, EIT health) has combined the MASK-air clinical data with SILAM data. A new Horizon Europe grant, CATALYSE (Climate Action to Advance HeaLthY Societies in Europe; grant agreement number 101057131), which started in September 2022, aims at better understanding climate change as well as ways to counteract it. One of the objectives of this project is to develop early warning systems and predictive models to improve the effectiveness of adaptation strategies to climate change. One of the early warning systems to be developed is focused on allergic rhinitis (CATALYSE Task 3.2). It stems from a collaboration between the

FMI (Finland), Porto University (Portugal), MASK-air SAS (France), ISGlobal (Spain), Hertie School (Germany) and the University of Zurich (Switzerland).

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Healthy and Low Allergenicity Green Spaces for the Cities of the Future

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Abstract

Emissions of allergenic pollen by some of the main species that make up urban green spaces constitute one of the main disservices of urban green infrastructure, with a great impact on the health of the population. Some of the causes that have generated this situation have been the massive use of a few plant species, the introduction of exotic species, botanical sexism or inadequate management and maintenance of these spaces, exacerbated by the impact that climate change is having on them. the reproductive phenology of some species. The allergenic characterization of the main species of urban trees and the estimation of the allergenic risk that these spaces may pose to the population are some useful tools for the planning and design of sustainable and healthy green spaces for the percentage of the population affected by adverse reactions to the presence of allergenic pollen.

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Aerobiology in 2050: What We Know and a Vision for Filling in the Gaps

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Abstract

Aerobiological knowledge has increased substantially since the mid-1900s. Building on this solid foundation, the field of aerobiology is currently going through a multifaceted revolution. With regard to pollen aerobiology, this includes the technological transition to automated real-time monitoring, remote sensing of source regions, environmental DNA identification of airborne pollen, the development of sophisticated modelling and forecasting systems, and digital health applications. Some of the benefits and advances of these include more-detailed aerobiological data (finer taxonomic levels and temporal scales), broader geographic coverage, and more immediate, accessible and personalised pollen information for self-management of allergic respiratory diseases. Despite this revolution, there remain major gaps that the international aerobiological community should aim to fill over the coming years. Perhaps the biggest is the

lack of knowledge about the aerobiology of several regions of the world. The establishment of an international network of aerobiological monitoring with coordination, cooperation, and data sharing facilitated perhaps through a United Nations organisation (e.g., WMO, UNEP) would go a long way to rectifying this current inequity.

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Pollen Monitoring and Forecasting Within the Copernicus Atmosphere Monitoring Service (CAMS)

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Abstract

Since 2014 the Copernicus Atmosphere Monitoring service, implemented by ECMWF on behalf of the European Commission, delivers relevant near real time (NRT) information about the atmospheric composition and its impacts on air quality, human health, ecosystems, radiatiative forcing , solar radiation and climate variables. CAMS operates at the global and regional (European) scales. It is built upon an integrated approach which combines observation data and models with the most up-to-date and appropriate methodologies to provide the best ambient air concentrations forecasts and assessments. Pollens monitoring and forecasting is one of the component of the regional portfolio and more details about their implementation and performance are given in this presentation.

Pollen modelling is carried out by a unique set of eleven regional air quality models run by European teams which have developed robust cutting-edge systems. The median average of the outputs provided by the eleven models is used to build up an ensemble model, which is more robust and usually performs better than the individual models. Modelling work is complemented and evaluated by monitoring data gathered from European aeroallergen monitoring networks.

CAMS provides every day of the year forecasts of pollen concentrations[1], expressed in grains/m3, for 6 species considered as among the most allergen: alder, birch, grass, mugwort, olive, ragweed. Collaboration with interested users and experts from the aeroallergen community is essential to tailor developments and propose service evolutions that reflects current state of the art and needs. CAMS has set-up several approaches to work in close collaboration with the user communities and they will be detailed in the presentation: national collaboration programmes (NCPs) to facilitate CAMS data uptake by national experts, contribution to the Copernicus Thematic Hubs, in particular the Copernicus Health Hub that

gathers and displays showcases for targeted applications, contribution to other initiatives like the European Climate and Health Observatory (Clim-ADAPT)[2].

[1] https://atmosphere.copernicus.eu/charts/packages/cams_air_quality/products/europe-air-quality-forecast-pollens?base_time=202406200000&layer_name=composition_europe_pol_grass_forecast_surface&level=key_0&originating_centre=85_2&projection=opencharts_europe&valid_time=202406200000

[2] https://climate-adapt.eea.europa.eu/en/observatory/evidence/projections-and-tools/cams-ground-level-pollen-forecast?size=n_10_n&filters%5B0%5D%5Bfield%5D=issued.date&filters%5B0%5D%5Btype%5D=any&filters%5B0%5D%5Bvalues%5D%5B0%5D=All%20time&filters%5B1%5D%5Bfield%5D=language&filters%5B1%5D%5Btype%5D=any&filters%5B1%5D%5Bvalues%5D%5B0%5D=en

POSTER PRESENTATIONS

5

Diurnal Pattern of Poaceae and Betula Pollen Flight in Central Europe

Mariel Suarez-Suarez[1], Isabel Costa-Gómez[2], Jose M. Maya-Manzano[3], Jesús Rojo[4], François Hentges[5], Ildiko Porcsin[5], Roland Sarda-Estève[6], Dominique Baisnée[6], Carsten Schmidt-Weber[1], Jeroen Buters[1]

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Abstract

Allergies constitute a mayor risk to human health; in Central Europe the most common allergies are provoked by grass or birch pollen allergens. We determined the intra-daily behavior of airborne pollen grains of grasses (Poaceae *ssp.)* and birch (*Betula ssp.)* in Central Europe, based on data obtained from a network of automatic pollen monitors over Europe (www.pollenscience.eu). Our aim was to determine the time of day when the lowest concentrations occur, to provide allergic individuals the optimal time to ventilate their homes. The study was carried out in three Central European capitals, Berlin (Germany), Paris-Saclay (France), and Luxembourg (Luxembourg), as well as in eight stations in Germany (Altötting, Feucht, Garmisch-Partenkirchen, Hof, Marktheidenfeld, Mindelheim, Munich and Viechtach), these eight locations in Bavaria belong to the electronic pollen information network (ePIN). The diurnal rhythm of these eleven locations was analyzed for either the complete, first week, peak week, peak day and last week of the pollen season. The data studied were reported as pollen/m^3 measured in 3 h periods. Stations were classified as city, semi-populated or countryside areas using land-use and population density criteria.

Grass pollen has a more pronounced diurnal rhythm than birch pollen concentrations. The average pollen percentage registered through the daily hours (6–21 h) during the main pollen season was 80% for Poaceae and 69% for *Betula*. A significant difference was observed when comparing day (6–21 h) versus night (21–6 h) for all stations. No difference was detected between city and countryside for both pollen types, although for Poaceae a longer period of maximum concentrations was observed in big cities and higher day/night-time differences were registered in the countryside (6.4) than in cities (3.0). The highest pollen concentrations were observed between 9 and 18 h for grass by representing 58.3% of daily pollen, but the rhythm was less pronounced for birch pollen. Additionally, we detected the intra daily trends for pollen from grasses were clustered according to a longitudinal gradient, meaning that the biological clock drives the pollen release.

After studying the diurnal patter of birch and grass, the results suggest that for allergic individuals who want to bring in fresh air in their homes with low amounts of pollen, the recommendation is to open windows after 21 h, but even better early in the morning between 6 and 9 h before pollinations (re)starts.

Data of the electronic pollen information network (ePIN) were kindly provided by the Bavarian State Office for Health and Food Safety. The ePIN project is part of the Bavarian climate adaptation strategy and is being led by the Bavarian State Office for Health and Food Safety (LGL) as the central specialist authority on behalf of the Bavarian State Ministry of Health and Care (StMGP, https://www.stmgp.bayern.de/). implemented for consumer health protection in Bavaria.

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The Potential Impact of Nearby Greenery on the Presence of Allergenic Pollen in School Buildings

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Abstract

Being indoors is one of several protective measures suggested to help reduce exposure to inhaled seasonal allergens. Nevertheless, there is no guarantee that pollen will not be present indoors or that allergy symptoms will be completely avoided. A wide range of studies have shown that pollen can be found in living, working or school environments. Indoor concentrations of particles are generally lower than those found outdoors. Plants growing naturally or planted for ornamental purposes near buildings may influence the diversity and quantity of pollen indoors. To address the issue of allergenic pollen-producing plants in public green spaces, it is recommended to control the variety and abundance of allergenic plants in urban areas. The study explored the contribution of school greenery to indoor pollen levels.

The research was carried out in schools located in different urban environments: downtown (located in the city centre), centre-periphery (located on the outskirts of the city), suburb (located outside the city). Indoor sampling took place during the autumn-winter period of November-December 2020 and the spring period of May-June 2021. For the analysis of pollen deposited on the premises, 179 samples were collected using the gravitational sedimentation method and 652 samples were collected using dust swabs. The surrounding green areas of the

schools were also surveyed. The trees were identified to genus level, maintaining taxonomic coincidence with the observed indoor pollen.

The school environment was dominated by 5 plant genera: *Acer*, *Betula*, *Populus*, *Thuja*, *Tilia*. Pollen representing 24 plant taxa was found in education classrooms and school corridors. Regardless of how far the schools were from the city centre and the sampling method, *Betula* and *Pinus* pollen dominated the samples. *Aesculus*, *Picea* and *Quercus* pollen were more abundant in suburban schools. *Picea* pollen was found to be abundant in the schools of the city centre in the spring. Pollen from *Betula* trees was found in all schools included in the study, regardless of the quantity of birch trees in the surrounding green areas. *Acer* pollen was detected in air samples where *Acer* and *Thuja* were the dominant plants in the school environment. Although *Populus* was predominant in the vicinity of some schools, it was not detected indoors despite the flowering being at an intensive stage at the time of sampling. *Pinus* pollen was found to be present in low quantities in the swab samples, whereas it was more abundant in samples collected using the gravitational method.

According to the study, it has been observed that *Betula* and *Pinus* pollen, being easily transported in the atmosphere, can also be found indoors in schools without these plants in the nearest surroundings. The amount of indoor pollen of the other morphotypes corresponds to the diversity of plants in the neighboring stands.

This research received funding from the Research Council of Lithuania (LMTLT) with agreements' No S-MIP-19-53 and No S-MIP20-52.

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Temporal Trends of Seasonal Pollen Indexes in the Veneto Region, Northern Italy (2001-2022)

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Abstract

Introduction: human exposure to pollens and the burden of allergic respiratory diseases have ultimately increased. Climate changes may be in part responsible for impacting pollen allergenicity, seasonality, distribution, and load.

Aim: to investigate the temporal trends in seasonal pollen indexes of 9 allergenic pollen families/genera in the Veneto Region, Northern Italy.

Methods: available daily data from 20 monitoring stations covered 2001-2022 for Corylaceae, Cupressaceae, Graminaceae, Oleaceae, and Urticaceae (pollen families), and 2006-2022 for *Alnus*, *Betula*, *Ambrosia*, and *Artemisia* (pollen genera). The pollen dataset was checked using the quality control function of the R "AeRobiology" package and the moving mean method was applied for the imputation of missing data. The 95-percentage method was applied to identify the start/end dates of pollen seasons during each year, and the seasonal cumulative pollen concentration (Seasonal Pollen Integral, SPIn) was calculated. The non-parametric Theil-Sen median slope method was used to examine 10-year trends in the pollen season's start day, duration, and SPIn. The analysis was applied to the Veneto Region and by climatic zones (Alpine, Subcontinental East, Subcontinental West). The statistical analyses were carried out using the statistical software STATA version 17 and RStudio version 4.2.2.

Results: a clear upward trend in SPIn was observed for most pollens, with median slopes ranging from 326.3 (*Alnus*) to 1089.7 p·m^3/10yrs (Corylaceae); exceptions were *Artemisia*, whose SPIn decreased over time (-26.0 p·m^3/10yrs), and *Ambrosia*, *Betula*, and Urticaceae, which did not show a clear trend. For families, pollen seasons showed a trend for anticipation (from 4.7, Urticaceae, to 13.5 days/10yrs, Oleaceae) and extended duration (from 8.3, Cupressaceae, to 13.3 days/10yrs, Oleaceae). The magnitude of increase/decrease in SPIn over time showed geographical heterogeneity, being generally more pronounced in Subcontinental areas compared to the Alpine one.

Conclusion: we observed deep variations in pollen load and seasons in the Veneto Region over the last two decades, suggesting a probable influence of climate change. Primary prevention strategies are needed to mitigate allergen exposure in the population.

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Bioaerosol Particles as Electron Carriers in the Global Electric Circuit

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Abstract

The global electric circuit is a conceptual electric circuit formed between the ionosphere and the earth through the atmosphere. The global electric circuit has been recognised since the early 20th century when the Carnegie curve was observed. Although it has been said that the global electric circuit is dominated by the relationships between lightning, ionospheric disturbances, and the atmospheric charging, little is known about the influence and role of the charging state of the atmospheric particles.

In order to study the details of the role of charged particles, especially charged bioaerosol particles, we have been working on estimating the amount and directions of electrons carried by airborne bioaerosols such as pollen, microorganisms, and fungal spores. In the poster, we will present the possible relationships between the ionosphere and lightning and charged bioaerosol particles. In addition, we will discuss a possible explanation for the electric current in the global electric circuit caused by the fragmentation of a bioaerosol and present the method we have developed to observe electron transport through bioaerosol fragments.

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Self-Management and Information Requirements of Allergic Rhinitis Patients in the Netherlands: A Focus Group Study

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Abstract

Introduction

Allergic rhinitis is a common condition affecting 20-30% of the population in north-western Europe and wind pollinating pollen is one of the main triggers of the symptoms. Patients often do not seek help from their GP, even though the complaints may have major impact on their daily live. This study delved into the self-management practices, preferences, and beliefs of individuals with allergic rhinitis, focusing on their needs for information and related practices.

Methods

Four separate focus groups, consisting of a total of 20 participants, were conducted—two online and two in-person. Focus group recordings were transcribed verbatim by a typist agency, and subsequently analysed using Atlas.ti (version 22). This software package facilitates and systematizes the coding of specific pieces of text (codes) and the analysis of the resulting collective of codes. Finally, a thematical structure was created to organize and categorize recurrent and common themes in the interviews. The purpose was to identify and highlight essential elements within the collected data.

Results and Discussion

Many participants hesitated to label themselves as patients, downplaying their concerns and attempting to minimize direct confrontation with their condition. A prevalent sentiment among patients was a lack of confidence in the efficacy of medications and healthcare's capacity to alleviate their symptoms. Consequently, there was a limited receptiveness to information, such as personalized pollen predictions. The results were consolidated based on three interconnected themes: "Being ill, but not a patient: it's bad, but you learn to live with it", "Individual search for what does or doesn't work" and "Information needs & sources". Our results indicating that self-management practices exhibit significant diversity and are highly individualized. Moreover, these practices tend to be more reactive to the occurrence of (early) symptoms rather than adopting a preventive approach. Furthermore, our research illustrates how entrenched self-management practices diminish patients' receptiveness to information about pollen concentrations. Over the years, patients have developed scepticism regarding the precision and utility of pollen forecasts, as well as doubts about the efficacy of medications and the extent to which healthcare can provide assistance. Being receptive to new information and adopting different self-management practices necessitates patients' willingness to acknowledge their status as patients, which involves taking their condition more seriously—a psychologically burdensome process. Additionally, striving to enhance self-management comes with the potential for disappointment. Therefore, communication efforts aimed at improving information for allergic rhinitis patients and supporting their self-management should be mindful of these psychological challenges and potential setbacks.

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A Simple Mechanism for UAV Bioaerosol Sampling in the Lower Atmosphere

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Abstract

Understanding the movement of bioaerosols, such as spores and pollen, through the atmosphere is important for a broad spectrum of landscape research, including agricultural fungal outbreaks and pollen threats to public health. As spores and pollen can be transported in the air over large distances, the use of aircraft have historically played a role in detecting and mapping their presence in the lower atmosphere. We present a lower-cost alternative to costly and specialized aircraft, and associated equipment, in the form of 3D printable components and common lab supplies mounted on an uncrewed aircraft (UA). Conveniently, this setup does not require additional electronic components to control collection during flight, using the UA landing gear mechanism instead. We demonstrate that this apparatus can collect fungal spores in the atmosphere and describe the potential impacts for the environment and on collection efficiency. Considerations include: 1) competing airflows from the UA rotors, flight trajectories, and wind; 2) flight altitude; and, 3) agar concentration of the collection medium.

Complex biological mechanisms and atmospheric dynamics dictate the release, transport and deposition of bioaerosols. Economical methods to sample bioaerosols in the lower atmosphere can increase the amount and type of data collected and unlock new understanding. The methodology presented here provides an economical method to sample bioaerosols that can help improve landscape-level understanding of the dispersal of bioaerosols.

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The Effects of Changing Climate on Long-term Trends in the Pollen Seasons of Fraxinus, Quercus and Ambrosia Artemisiifolia in Slovakia

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Abstract

Background: The ongoing climate change affects life cycles, including flowering, of some plant taxa. Anemophilous plants with allergenic pollen are also affected by this change, impacting people who suffer from pollen allergic respiratory diseases, such as rhinitis or asthma. The main factors influencing the timing, duration and intensity of pollen seasons of these plants are changing meteorological variables and levels of air pollutants. It is imperative to study these changes, as the prevalence of these allergic diseases is high and still on the rise. This study is looking for trends in the pollen seasons of both woody (*Quercus* and *Fraxinus*) and herbaceous taxa (*Ambrosia artemisiifolia*) during the last two decades.

Method: The changes in their pollen season characteristics were studied at two urban sites in Slovakia with long-term aerobiological data – Bratislava and Banská Bystrica. Hirst-type pollen trap was used to capture pollen grains at both locations and the obtained samples were analysed using standard aerobiological methods. Daily means of pollen concentration for the chosen taxa were calculated as the number of pollen grains per cubic meter of air (pollen/m^3). Trends were observed in the following characteristics of the Main pollen season: start date, end date, duration, peak value, SPIn (Seasonal Pollen Integral), number of High days (days when the pollen concentration was high enough to cause symptoms in all people suffering from polinosis) and the date of the first High day. The threshold for High days was set at 50 pollen/m^3 for *Fraxinus* and *Quercus* and 20 pollen/m^3 for *Ambrosia*. The significant trends in pollen season characteristics were correlated with the significant trends identified in the meteorological (temperature, precipitation and relative air humidity) and air pollution (concentrations of PM_{10}, SO_2, O_3 and CO) parameters.

Results: In woody taxa, we found a trend towards an earlier onset, longer duration, and intensification of their pollen seasons, especially in *Quercus*. They were mostly correlated with rising temperatures at the time of flower bud formation – summer and autumn of the previous year. As for the invasive *A. artemisiifolia*, we found trends towards an earlier start date and longer duration at both sites, as well as later end day and an increased number of High days in Banská Bystrica. The prolongation of its pollen season was correlated with an increase in temperature and relative air humidity during the summer and autumn of the same year. Regarding air pollutants, we found mostly declining trends, except for the rise of CO in the air of Banská Bystrica. The correlation of air pollution with the pollen season intensity was negative for SO_2 and NO_2, and positive for CO.

Conclusion: It is important to study the long-term trends in pollen seasons, as they tell us more about the effects of climate change on plant life cycles. These changes also impact the lives of

people who suffer from allergic diseases and their study enables us to make better predictions of future pollen seasons of allergenic plant taxa.

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Holography-based Aerobiological Monitoring: a 2-year Intercomparison Campaign Versus the Standard Hirst Method in Brussels, Belgium

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Sciensano, Brussels, Belgium

Abstract

Around 10% of the Belgian population is allergic to the pollen emitted by trees of the Betulaceae family (birch, alder, hazel and hornbeam) and an estimated 18% to grass pollen. To prevent and treat respiratory symptoms and reduce the allergy burden, pollen grain concentrations have been continuously monitored using the standard Hirst method with a volumetric spore trap. However, this offline method can only provide results retrospectively, from 1 day to 1 week, and previous evaluation studies have shown certain limitations in terms of sampling efficiency and measurement accuracy.

Recently, several automatic real-time instruments have been developed to allow the identification and quantification of airborne pollen. After a 2-year campaign performed in Brussels (Belgium), daily concentrations of 13 pollen taxa measured by the Swisens Poleno Jupiter were compared to the measurements of the Hirst method. The highest intraseasonal correlation values between both methods were found for *Betula*, *Fraxinus* and *Poaceae*. While some pollen taxa showed similar seasonal kinetics but with scaling discrepancies, other pollen taxa frequently presented out-of-season false positive peaks as results from the automatic monitor. In all, the holography-based monitoring in real conditions appeared to be relatively reliable within the seasons of most allergenic pollen taxa. Further improvements are expected by training the identification algorithm with reference datasets generated from pollen collected in the local environment.

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Exposure Chambers and Detection Methods of Airborne Infectious Viruses to Support Studies in Aerovirology

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Abstract

COVID-19 pandemic has hit the world in 2020 and highlighted the risks of airborne spread of respiratory viruses. At the start of the pandemic, a limited number of aerovirology studies had been conducted, and no standardized methods were available. To assess mitigation techniques and health risks imposed by respiratory viruses in indoor environments, adequate sampling and analytical methods are required. In the current study, exposure chambers were designed to optimize sampling and analytical methods, and to test mitigation methods to limit the spread of airborne viruses.

Two exposure chambers were designed, one of 0.1 m^3 to aerosolize human coronavirus for the optimisation of analyses of virus infectivity from air samples, and another one of 18 m^3, in which high concentrations of MS2 bacteriophages were aerosolized to evaluate the performance of mitigation methods against airborne viruses. Being not pathogenic to humans and more resistant than coronavirus, MS2 bacteriophages can also be easily produced in large quantities with a rapid infection rate in petri dishes, thereby justifying their use as aerosolized viruses in the 18 m^3 chamber. Viral aerosols were generated with the 1 or 24 jets Collisons nebulizer. For safety reasons, negative pressure was maintained in both chambers during generation of viral aerosols. Sampling of the two coronavirus (hCoV-OC43 and229E) and MS2 was carried out on 25 mm polytetrafluoroethylene and polycarbonate membranes, respectively inserted in two-section PVC cassettes (SKC inc., Pennsylvania, USA). Sampling flow rate was adjusted to 2 L/min and based on preliminary tests; the optimized sampling time was set to 5 minutes.

In this study, we used two coronaviruses, which harbours a luciferase reporter gene incorporated into the NS2 gene providing a luminescence-based enzymatic assay for quantification of infectious airborne viruses in cell culture. HRT18 cell lines are used for hCoV-OC43 replication, while Huh-7 cell lines are used for hCoV-229E. The development of a RT-dd-PCR assay, also dependent on intracellular multiplication of viruses in the respective cell line, proved to show an excellent correlation (R^2= 0.998) with the luminescence assay and considerably improved the detection limit required to support the attenuation studies.

To assess the effectiveness of attenuation methods, high, stable concentrations of aerosolized infectious viruses are required in the chambers before applying attenuation treatments. This requirement was validated in both chambers. In the 0.1m^3 chamber, infectious hCoV-OC43 and hCoV-229E reached concentrations of 10E7 and 10E9 ARN copy/m^3 of air, respectively and for MS2, in the large chamber, concentrations of 10E10 PFU/m^3 of air were reached. Natural decay due to mechanisms, such as diffusion, electrostatics, and inertia, led to a decrease in the number

of infectious viruses, which was increased following the implementation of the attenuation method. Only weak natural decrease of airborne viruses was observed in both chambers.

This project has led to the development of two generation chambers well-suited for studies in aerovirology, as well as for the evaluation of tools for the mitigation of airborne infectious viruses. The performance of attenuating device toward airborne viruses were tested in these chambers following their implementation.

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More Pronounced Chemical Distinctions Between Airborne and Flower Pollen Than Between Pollen Collected Along a Pollution Gradient in the Netherlands.

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Abstract

Introduction

The incidence of allergic diseases has significantly risen over recent decades. A crucial factor contributing to allergic rhinitis (hay fever) symptoms is the pollen produced by wind-pollinated plants. This pollen develops within flowers, is subsequently released into the air, and becomes exposed to environmental factors and air pollution. To explore the chemical transformations occurring in pollen after its release, we conducted a study in both a rural (Veluwe) and an urban (Amsterdam) location in the Netherlands, utilizing Fourier Transform Infrared (FTIR) spectroscopy.

Material and Methods

In the spring/summer of 2020, amid the COVID-19 pandemic, airborne pollen was sampled from nine taxa (*Alnus, Betula, Fagus, Fraxinus, Pinus, Plantago, Poaceae, Quercus*, and *Salix*) at street level in Amsterdam and on the Veluwe using portable pollen samplers (called Pollensniffers); Flower (pollen) samples were gathered directly from plants in the identical environment and timeframe during which the Pollensniffer was deployed. FTIR spectra were obtained for numerous individual pollen grains within each taxon. After measurement, all processing and plotting of the spectra was conducted in R (version 4.2.3).

Results and Discussion

No significant differences were observed in the spectra from airborne pollen collected at the rural versus urban sites; this could be attributed to a potential reduction in pollutant concentrations due to COVID-19 lockdown measures in place. Nevertheless, consistent variations in the FTIR spectra between airborne and flower-collected pollen were identified across all pollen taxa. Post-flower release, the chemical composition of the pollen underwent changes: (i) polysaccharides were converted to monosaccharides; (ii) alterations in protein concentration and/or nitration/oxidation levels occurred; (iii) modifications and/or reductions in lipid concentration were observed. These transformations may impact the allergenic properties of the pollen. Our findings raise questions about the validity of the common medical industry practice of conducting allergenic research solely on pollen directly obtained from flowers.

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The Pollenservice Vienna – Launching a New Communication Strategy

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Abstract

Purpose: A regional pollen service was established for Vienna in the end of 2023: the Pollenservice Vienna of the MedUni Vienna. Its purpose is to inform the local population accurately about the special aerobiological situation in a metropolis.

Hypotheses: Vienna is the capital of Austria and a metropolis. As such it is especially subjected to climate change and pollination occurs under special conditions. This includes also the planting strategy in cities and the urban heat island effect. In addition, a new communication strategy was developed to overcome misunderstandings that frequently occurred in our experience when providing pollen forecasts.

Main methods: A literature review pointed to a pioneer work, that demanded a simpler provision of pollen forecasts, ideally as a traffic light system (green, yellow and red). In accordance with this publication the forecast for the next three days was designed.

Results: The proposed color code was adopted and applied to the pollen forecast on the website www.pollenservice.wien. One green filled box was used for "no/hardly any pollination", two yellow filled boxes were used for "pollination" and three red filled boxes were used for "high pollination". Furthermore, the length of the textual pollen forecast was reduced and simplified.

Terminology was changed to “pollination” or “full flower” instead of “burden”, which is a more imprecise term as it refers to a condition that is based on multiple factors (pollination, health, air quality, stress). The last step was to focus more on visuals. Therefore, photos appear large, and videos are integrated on the website. On top of that, persons interested are informed via X on the channel “Pollenpaar Bastl”.

Conclusion: The first post on X reached 1.9k impressions, which is impressive for the small Austrian community on this channel. The visuals improved the first impression of the website and directly relate to the state of the season. The new code for the pollen forecast for the next three days is easy to interpret for everyone and barrier-free as it does not only relate to the color code, but also to a stepwise increase of the displayed boxes. In addition, the reduced textual pollen forecast explains the situation of pollination fast and clear. The communication strategy of the Pollenservice Vienna was thus successfully improved.

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Allergenic Pollen Seasons and Regional Pollen Calendars for Norway: Seven Pollen Types in Twelve Regions

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Abstract

Background and Aims: The environment of the Scandinavian country of Norway is exceptionally diverse. This is primarily the result of its wide-ranging latitude, longitude, altitude and high microscale topographical heterogeneity. This has contributed to complex climatological gradients and allowed for many different habitats and species. While the identification of Norwegian species has long been ongoing, the modern monitoring of Norwegian pollen started in Trondheim in 1980. Since then, the pollen monitoring network has expanded to encompass twelve regions spanning the length of the country. However, relatively little is still known of how airborne pollen in Norway varies, both spatially and temporally. This is especially relevant for allergenic pollen that elicit immunological reactions in the Norwegian public. To address this gap in knowledge we aim to create updated pollen calendars with seasonal statistics for pollen types with allergenic potential for Norway.

Methods: The Norwegian pollen monitoring network monitors all pollen bioaerosols, yet seven main pollen types of allergenic potential were considered for this study: alder (*Alnus*), hazel (*Corylus*), willow (*Salix*), birch (*Betula*), pine (*Pinus*), grass (Poaceae) and mugwort (*Artemisia*). The

pollen season is monitored from the start of the growth season to the 30th of September, generally considered the end of the growth season. For the pollen calendars to be reliable only recent and comparable data were used, with the years between 2007 and 2022 (16 years) being averaged to construct the calendars for the twelve regions. Four aspects of the pollen season were used: First to last observation of the season, main season (Seasonal Pollen Integral (SPIn) 95%) and timing of high and very high pollen concentration thresholds, with the threshold varying between tree and herb types and identified from the literature. The number of days of high concentrations were modelled with SPIn using a linear regression framework to investigate the connection between days of increased risk and the strength of the season for each pollen type.

Results: Regional pollen calendars were created for all pollen types except when SPIn of a type in a region had very low and unreliable average SPIn. The tree pollen season was identified to occur between January and mid-July, with birch and pine being on average the most prominent pollen types in all regions. The herb pollen season was identified to occur between June and mid-August, with the mugwort season being mostly absent. The grass pollen season was mild on average, being more severe in Kristiansand. South-eastern regions had on average the highest pollen loads, e.g., Oslo, Kristiansand and Lillehammer, while northern regions had on average the lowest pollen loads, e.g., Bodø, Tromsø and Kirkenes. SPIn and number of high days had positive significant relationship for all types ($R^2 > 0.85$) except for mugwort.

Conclusion: Regional pollen calendars and seasonal statistics contribute to reliable information that can be used by medical professionals to effectively and timely manage and treat seasonal pollen allergies in Norway.

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The Pollen Calendar, a Useful Tool for Clinical Management of Allergic Patients but Also for Scientists to Highligth Climate Change Impact on Pollen Loads

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Abstract

Background: The pollen calendars are based on the processing of data collected over several years of airborne pollen monitoring and represent the seasonal trends in the concentrations and

time of different types of pollen referred to a specific geographical area. They can be considered a useful tool to evaluate exposition risk for patients, mainly if they are travelling for work or tourism. The aim of this study was to build the pollen calendar in Parma (Italy) based on 27 years of data recording and to compare the first and the last periods.

Methods: Daily average pollen concentrations from 1997 to 2023 were considered and the first and last 5 five years were compared by the Cox-Stuart test. The following taxa were considered: *Betula, Corylus*, Cupressaceae, *Platanus*, Poaceae, total pollen and *Alternaria* spores. The average of temperature, relative humidity and the total rainfalls were also analysed.

Results: A significant difference between the period 1997-2001 *vs* 2019-2023 for *Corylus*, Cupressaceae and *Alternaria* was found with higher concentrations of daily average in the last 5 years. *Corylus* increased from 16.7 to 33.5 pollens/m^3; Cupressaceae from 40.1 to 104,5 pollens/m^3; *Alternaria* from 178 to 276.4 spores/m^3. The daily average of *Platanus*, Poaceae and total pollen have decreased while *Betula* and *Ambrosia* have increased over the last 5 years even though not significantly. During the period 2019-2023, Cupressaceae, total pollen, Poaceae had a significant precocious start date (14, 11 and 6 days, respectively) and a significant late end date than 1997-2011. On the contrary, the *Corylus* pollen season began and ended earlier than 1997-2001 ($p = 0.011$). The pollen season of *Betula* began and ended later ($p = 0{,}011$) and *Alternaria* spores ended later ($p = 0.001$). No significant differences were observed for *Platanus* and *Ambrosia*. The average temperatures increased significantly, in the last 5 years of 1.2°C; humidity decreased significantly in the same period. Total precipitations decreased from 796.9 to 710.7 mm, but not significantly.

Discussion: Pollen calendars are location specific linked to the local distribution of flora and climate. Aerobiological monitoring and pollen calendars are relevant for diagnosis, treatment, prevention of seasonal respiratory allergies and can also help in limiting the exposure, to guide the therapy before high pollen loads. Moreover, pollen calendars can provide useful information for people who need to move for work or tourism giving answers to the following questions: where is it safe for allergic patients to travel? What season? For which allergic patients? This study gives answers for the Parma territory. Thanks to 27 years of pollen data recording, this pollen calendar allowed us to highlight the shift of pollen loads and pollen season for some taxa, related to climate change.

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Is the Flow of Internet Search Terms a Useful Tool for Assessing Trends and Relationships to Obtain New Information on Pollinosis in European Countries?

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Abstract

Background: Pollen exposure is a major cause of respiratory allergies worldwide. However, it is unclear how everyday exposure is related to symptoms and how exactly allergic patients may be affected spatially and temporally. In the recent years, studies have also utilised internet search platforms such as Google Trends (GTs) to deepen environmental and medical events. Some of them has been devoted to investigating internet search terms related to allergic diseases correlated with pollen concentrations. This study aimed at acquiring new information on pollinosis examining if the searched term distribution of symptoms, medication, and pollen differs from a country to another hypothesizing different distributions of pollen allergies.

Methods: From 2004 to October 2019, we examined multiple time series of GTs search data on rhinitis, allergy, pollen, asthma, and antihistamines in several European countries. We looked at data from Austria, Finland, France, Germany, Hungary, Italy, Portugal, Romania, Sweden, and the United Kingdom. GTs data for "allergy," "rhinitis," "asthma," "antihistamine," and "pollen" were extracted. We added the scores for the searched terms and examined the distribution of interest in each country.

Results: In the selected European countries, the highest peaks of search were in Spain, May (15.13%), Austria, April (14.99%), Italy and Romania, May (12.60%), Portugal, April (10.10%), United Kingdom, May (12.54%), France, April (12.22%), Hungary, August (12.19%), and Netherlands, April (9.14%). The Kruskal-Wallis test showed significant differences between the monthly medians of GTs in these countries over the period 2004-2019.

Discussion: The results showed that GTs data are suggestive for different pollen season according to geoclimatic situation of the analysed countries. GTs data can be used as a proxy for the identification of the onset and variation of symptoms and medication score for allergic patients depending on the searched time distribution. Cultural differences, internet habits, and term translations differ among European countries, which may have influenced the results. Further investigations to better understand the potential of the approach used are needed.

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Land-use Patterns and Fungal Bioaerosols in the Brazilian Atlantic Forest Biome

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Abstract

Whilst the importance of fungal primary biological aerosol particles (PBAPs) has been recognised, few studies have empirically assessed how land-use patterns influence them. Here, we show the impacts of different land-use patterns on fungal PBAPs within the Brazilian Atlantic Forest biodiversity hotspot. Spanning a distance of *ca.* 600 km within this biome, we collected fungal bioaerosols in the following land-use patterns: a 25-yr old coffee plantation, a 2.5-yr old Eucalyptus plantation, a 3-months-old maize crop, a 2-yr old and another 7-yr old native forest reforestation area, and a native forest fragment. Using the portable Burkard air sampler, a total of 14 morphotype-species were found. *Cladosporium* sp. comprised as much as about 95% of all fungal spores collected, being present in all samples (frequency of 100%). Forest systems had as much as 5-to-19-fold more fungal spores in the air than maize crops. Sampling height and time (morning *vs.* afternoon) did not influence fungal concentration and number of species. In addition, using data derived from an eddy covariance tower in the Eucalyptus site, we estimated the emission rate to be 6649 spores $m^{-2}\ s^{-1}$. Our study confirms that land-use patterns affect fungal PBAPs, and that replacing large areas of native Atlantic Forest by monospecific stands, a homogenisation of airborne fungi is occurring, with unknown consequences for climate regulation.

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Exposure to Allergenic Airborne Pollen in Three Different Locations of the Po Valley-Milan Area

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Abstract

Background

Respiratory allergies caused by airborne pollen represent an increasing public health problem and involve high healthcare costs.

The main allergenic pollen species present in the Po valley (northern Italy) are: *Alnus*, *Betula*, *Ambrosia*, *Artemisia*, *Corylus*, Cupressaceae, *Carpinus*, *Ostrya*, Graminaceae, *Olea, Pinus, Cedrus and* Urticaceae.

The aim of the study is to evaluate the exposure of the population to these pollen species in three different areas of the Po valley: the Milan city center and two towns located in its surroundings.

Methods

Pollen grains were sampled by three Hirst volumetric traps located in Legnano, Milano and Desio. Daily average pollen concentrations were expressed as particles per cubic meter of air (p/m^3). Observation period ranged between 2017 and 2023.

The Annual Pollen Integral (API) of different pollen species was examined.

A non-parametric statistical approach (Kruskal-Wallis test by rank, and subsequent Pairwise Wilcoxon rank sum tests, and unpaired two-samples Wilcoxon test for two groups comparison) was used to compare the median values of API across locations. The data were considered statistically significant for values of $p < 0.05$.

Some descriptive statistics such as, mean, median, interquartile range and standard deviation of the sample data were also calculated.

Results

The Kruskal-Wallis test indicated significant differences among the median API levels of various pollen types across the three locations, namely *Alnus*, *Ambrosia*, *Artemisia*, *Corylus*, *Carpinus* and *Ostrya*, Cupressaceae, Graminaceae and Urticaceae.

The pairwise comparison, using exact Wilcoxon rank sum test, revealed significant differences between Desio and Milano for most pollen species, except for *Carpinus* and *Ostrya*. Instead, Milano and Legnano were significantly different for *Alnus*, *Ambrosia*, *Corylus*, Cupressaceae and Graminaceae.

Legnano and Desio differed significantly only for *Artemisia*, Cupressaceae and Urticaceae.

Furthermore, the unpaired two-samples Wilcoxon test demonstrated a significant difference between Milano and Legnano for *Pinus*.

No significant differences were observed for *Betula*, *Olea* and *Cedrus*.

Generally, median pollen levels were higher in Legnano and Desio compared to the Milan city center, with the exception of Olea, which exhibited similar levels across all locations.

Conclusions

The findings highlight a significant variation in pollen exposure across the three locations. Specifically, individuals living outside the Milan city center experienced higher pollen concentrations, except for *Olea*. Further investigation is warranted to evaluate the prevalence of various types of pollens in the area and their actual health implications. While pollen load serves as an indirect indicator of health impact, additional studies are necessary to assess the specific rates of pollinosis and their consequences on public health.

Keywords: Pollen exposure; Allergy; Po valley, nonparametric tests

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The Ragweed Finder: a Citizen-science Platform for Monitoring the Distribution of Ambrosia Artemisiifolia

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Abstract

This study aims to assess the distribution of *Ambrosia artemisiifolia* (ragweed, Asteraceae) to gain insights on the overall spread of this weed in Austria from 2017 to 2023 by using a citizen-science platform, the Ragweed Finder. Information about the distribution of ragweed is important for agricultural purposes and especially for allergy sufferers sensitized to its pollen.

Ragweed, a plant classified as alien invasive species by a European Regulation (No. 1143/2014), was introduced to Europe in the 19th century and has since become a significant problem. Approximately 33 million Europeans are sensitized to its pollen. The project focuses on Austria, which partially belongs to the Pannonian Plain, one of the most severely infected regions in Europe and explores the propagation of this species by engaging citizen-scientists.

The Ragweed Finder platform was launched in 2017 as a website and in 2019 as a mobile app with the goal to collect ragweed distribution data in Austria. The reporting process contains a checklist for identifying ragweed to ensure high identification quality. It requires just a few more mandatory details, such as location, date, a photo of the ragweed population and an e-mail address. Additional information on the location, size of the population or experienced allergic symptoms may be provided voluntarily. Each report is reviewed by an expert, to check if the reported plant is identified correctly. For this study, pollen data from Hirst-type pollen traps of Eastern Austria and its neighboring countries were queried from the European Aeroallergen Network database for comparison with the Ragweed Finder reports.

The Ragweed Finder recorded 10,259 reports between 2017 and 2023. 9,476 or 92.4% of them were identified correctly. The platform shows an increasing trend in reports, with an exception for 2020, where the decline is potentially linked to the SARS-CoV-2 pandemic. The Austrian counties contributing most to the area of the Pannonian Plain (Burgenland, Lower Austria, and Vienna) are responsible for 80.2% to 89.6% of the positive ragweed reports. Additionally, the number of daily reports was compared to the ragweed pollen concentration in the Pannonian plain and revealed a shift of the reporting time of ragweed to earlier developmental stages (before the ragweed pollen concentration peak time).

The idea for using citizen-science as an additional data source for providing more detailed information to pollen allergy sufferers turned out to be a well-accepted and frequently used platform. The gathered data is greatly appreciated by most Austrian counties and was essential for enrolling the "Burgenland Ragweed Control Act" in the year 2021. Future assessments will determine the effectiveness of this legislative measure. However, the success of the Ragweed Finder platform demonstrates the potential of citizen-science in providing near real-time and accurate data for documenting the spread of *Ambrosia artemisiifolia*.

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Characteristics of Birch Pollen Seasons During 25 Years in Ljubljana, Slovenia

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Abstract

Background: Birch pollen is one of the most important aeroallergens in Slovenia according to clinical studies. The largest source of pollen is Betula pendula Roth., which mostly grows in forest clearings and edges, in addition also planted as an ornamental tree. The study area of the dynamics to birch pollen exposure was Ljubljana, the capital city of Slovenia. We analysed a 25-year pollen data set (1999-2023) and the impact of meteorological conditions on the daily birch concentrations.

Methods: Aerobiological and meteorological measurements were taken at the same location in the city (46° 3.9198' N, 14° 30.8130' E). Aerobiological sampling and analyses were carried out by the recommended methodology, summarized in the standard EN 16868:2019. Measurements were performed with a Hirst type sampler, 19 m above ground level. Daily average pollen concentrations were given as number of pollen grains per cubic meter of air. Characteristics of the main pollen season (start, end, length, number of high pollen days) were calculated according to the European Academy of Allergy and Clinical Immunology (EAACI) definition, whereby annual pollen integral (APIn) was also analysed. The meteorological data were collected via Slovenian environment agency, the influence of the selected weather parameters on daily pollen concentrations was determined using Spearman's rho correlation.

Results: The birch pollen season lasted 25 days average (from April 1 to April 25), start and end dates varied by about one month. The longest season was recorded in 2022, when it was extended by two weeks. Only a slight trend towards season prolongation was noticed, but not statistically significant. On average the season peak occurred on April 9, with outstanding years

2012, 2014 and 2017, when the highest loads were already in the last week of March. 2013 was unique since grains appeared quite late, in the middle of April. From 1999 to 2011 most APIn values were lower than the average of 3895 pollen × day per cubic meter (except 2006 and 2010). Years from 2012 onwards exceed the average APIn (except 2021 and 2023), indicating a slight increase in APIn throughout the observed period. The highest number of days with pollen concentration above 100 grains per cubic meter was 19 days (2022). Spearman's rho correlation analyses have demonstrated that daily birch pollen concentration had a positive correlation with maximum temperature (rs = 0,393), average temperature (rs = 0,363), and sunlight duration (rs = 0,313). Negative correlations were found with relative humidity (rs = - 0,304) and precipitation (rs = - 0,203).

Conclusions: Birch pollen season in Ljubljana is usually in full swing in April, characteristics of season vary from year to year and are related to weather conditions. Observations of APIn values show a slight increase, but without any cyclical rhythms. According to EAACI definition the pollen season in the last decade on average lasts longer, which has negative effect on pollen allergy sufferers.

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Influence of Atmospheric Boundary Layer Over the Surface Cupressaceae Pollen Levels in an Urban Environment

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Abstract

relation to surface pollen levels. This research aims to fill this gap by examining how ABL dynamics impacts surface pollen levels during periods of notable concentration, using a 6-year dataset (2017-2022) of coincident lidar Doppler and Cupressaceae pollen data.

The study was conducted in the city of Granada (Spain, 37.18° N, 3.61° W, 680 m a.s.l.), utilizing daily airborne Cupressaceae pollen counts recorded by the of the Biological Air Quality Unit of the University of Granada (UCBA-UGR). Aerobiological data were collected using a volumetric

suction Hirst-type sampler Lanzoni VPPS 2000, situated 20 meters above ground level on the Faculty of Sciences building. In parallel, measurements of the 3D wind field and turbulent properties of the ABL were obtained using the Doppler lidar StreamLine (Halo Photonics) at the ACTRIS AGORA facility of the University of Granada.

Spearman's correlations and Generalized Linear Models (GLMs) were used to study the link between Cupressaceae pollen concentrations at surface and the variables related to the ABL dynamics, such as vertical wind speed (w), wind speed (v_h), wind direction (dir), turbulent kinetic energy dissipation rate (ε) and wind shear (sh) for different height ranges (from 90 up to 500 m a.g.l.) and different times of the day. On one hand, Spearman's correlations indicated significant negative correlations (p-values < 0.05) for all variables except w across various height ranges. These results show a decrease of Cupressaceae pollen concentrations at surface when these variables increase individually at altitude ranges well above the surface. On the other hand, GLMs revealed a strong statistically significant relationship for sh, ε, and dir (all of them with p-values < 0.001) in the lower height ranges (from 90 to 250 m), showing a negative relationship for ε and positive for sh. This discrepancy between the sign of its effect or correlation shows a more complex relationship among them. In the higher height range (from 90 to 500 m) only ε and dir are statistically significant (p-values < 0.01). The categorical variable dir shows a positive effect for the directions that coincide with the main emission sources in the city at any height range, especially at nighttime. Finally, it was found that the variables related to the ABL dynamics at any height ranges can explain over 14% of the variability of the Cupressaceae concentrations at surface in this urban environment. Therefore, ABL dynamics have been shown to have a significant effect on surface pollen concentrations.

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Disassembling the Pollen Grain: Its Role as Atmospheric Aerosol Beyond the Reproductive Function of Plants

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Abstract

The pollen grain is the male gametophyte that participates in the reproduction process of Spermatophytes, being transferred during pollination to the female structures of the plants by biotic and abiotic agents. In order not to see its viability altered during this transport, the pollen is covered with the exine or pollen wall, which is a mixture of biological compounds that protect it from different environmental stresses and keep it unaltered and functional. The main biological compounds of this outer shell are essential amino acids and proteins, carbohydrates and saccharides, fatty acids and lipids and organic acids. The role of most of these compounds is crucial in the germination and formation of the pollen tube, and in the dehydration processes that take place during dispersal. Some of these compounds (proteins) are responsible for generating a symptomatic allergic response in the sensitive population, and may also have a relevant role in physical processes in the atmosphere acting as cloud condensation nuclei (CCN) and ice nucleating particles (INP). This work aims to determine the role that organic compounds of different pollen types from the Mediterranean region can have on the processes of cloud formation and allergic response in the sensitized population. To this end, sub-pollen particles (SPP) from different species (*Acer, Cupressus, Dactylis, Fraxinus, Olea, Pinus, Platanus, Populus, Quercus*) were subjected to liquid chromatography-mass spectrometry techniques for protein extraction, determining their content by means of Bradford Assay Method. whereas the organic speciation for these pollen types was determined by gas chromatography coupled to mass spectrometry. The results show that the pollen grains of *Acer, Populus* and *Quercus* are the ones with the highest concentrations (in mg/mL) of protein; However, the highest concentrations of allergenic proteins are presented by *Olea*, *Dactylis* and *Cupressus*, although the latter is the one that contains the lowest concentration of total protein of all the types studied. Regarding the presence of organic compounds, *Olea* and *Pinus* are the ones with the highest concentration, while *Quercus* and *Populus* are those with the lowest concentrations of organic compounds. Organics speciation shows saccharides are the most abundant components in most pollen types, contributing over 87.57% in *Quercus*. Amino acids, mainly Proline, are quite abundant in *Populus* and *Acer,* however, they have not been detected in all species. In contrast fatty acids are present in all pollen types (palmitic acid is the most abundant except for *Dactylis*) although its contribution is very variable among species (almost 40% in the case of *Populus* and less than 5% in *Quercus*). This different composition of soluble and insoluble organic compounds will have an effect on the hygroscopicity parameter and suggests that pollen grains, and above all, the sub-pollen particles (SSP) that are formed during atmospheric dynamics, may have a potential

role to act as CCN (Casans et al., 2023). The results also confirm the role of proteins as the main allergenic component in the pollen types of native Mediterranean species.

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Changes in Trends and Intradiurnal Dynamics of the Platanus Pollen Type

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Abstract

The genus *Platanus* L. is made up of woody trees that are widely used as ornamental trees in urban forests around the world. Their characteristics, such as rapid growth, good ecological adaptability to urban conditions, and tolerance to air pollution, make them one of the most important and relevant urban trees. However, its massive use in urban green infrastructure has some negative effects, such as the emission of allergens that cause severe respiratory symptoms in allergy sufferers.

In recent years, it has been observed important environmental changes related to alterations in the phenological cycle of plants; at the same time, it has detected increasing trends of pollen allergy prevalence. For all this, the knowledge of the diurnal and seasonal patterns of pollen concentrations is important because it contributes to improve the prevention of allergy in urban areas.

This study aims to establish if the annual trend and the hourly dynamic of *Platanus* pollen type have changed over the 31 years, and whether allergy symptoms have been modified.

The study of temporal dynamics was carried out in the city of Granada (south-eastern Spain, 37°11' N, 03°35' W, 685 m.a.s.l. and 23 m.a.g.l) using a Hirst-type volumetric trap. The samples were analysed according to the norms of the Spanish Aerobiological Network, and the annual trend was studied during the period 1992-2022. In this period, the average pollen season was calculated at 90% for the *Platanus* pollen type to carry out the hourly count. The aim was to compare the hourly dynamics of this type of pollen between the years 1993-1994, at the beginning of the series, and to compare it with the hourly dynamics of the end of the series, years 2021-2022.

The Annual Pollen Indices (APIn) of *Platanus* pollen type showed a significant trend increase (Spearman Test $p < 0.001$) that can suppose an annual increase of 59 pollen grains (Theil Shen estimator).

The comparative study between the intradiurnal dynamic pollen of 1993-1994 and 2021-2022 showed a change in the hourly distribution of *Platanus* pollen. While in the period 93-94 the maximum pollen concentration was registered between 13.00 and 18.00h, peaking at 15.00, in the period 21-22 this maximum was concentrated between 13.00 and 22.00h, peaking at 18.00.

The increase in APIn over the 31 years studied and the increase in the number of hours during which pollen remains in the air may be causing a change in allergic symptoms, increasing and extending the hours of the day when symptoms are more severe, so that the impact of these ornamental trees on public health may be worsening.

All this information is achieved thanks to the funding obtained from the following research groups and projects: RNM-110 and RNM-170 Research Group; Spanish Society of Allergy and Clinical Immunology (SEAIC); and Grant C-EXP-167-UGR23 funded by Consejería de Universidad, Investigación e Innovación and by ERDF Andalusia Program 2021-2027.

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Detection of Botrytis Cinerea Spores in Greenhouses With Hirst Type Spore Traps and N3 Optical Particle Counters.

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Abstract

Background: Real-time detection of rising fungal spore concentrations in greenhouses could prevent huge losses in horticulture. Successful use of low-cost optical particle counters has not been reported to date, and previous such studies on the application of these have focused on the measurement of the larger pollen particles, and outdoor measurements, where climatic parameters have greater fluctuations. In this study we wanted to increase the knowledge regarding the magnitude and variation in *Botrytis* spores in greenhouses, and link these to the potential use of low-cost sensors.

Methods: Tests of low-cost optical particle counters from Alphasense (OPC-N3) for quantification of a known fungal spore (*Botrytis cinerea*) were performed in a closed chamber. Aerosols were simultaneously quantified using a Grimm Portable Aerosol Spectrometer (11E). Size distributions were analyzed in R-studio for identification of a "signature signal" from the *Botrytis* spores. A model was run on the N3 data on selected particle size bins for a certain time window with spore release. If the linear coefficient is positive and the quadratic coefficient is negative, the system indicates a BLIPS (Binomial Logic Identification of Pathogenic Spores) for that timestamp. This dichotomous classification allows for a simple yet effective prediction of the potential presence of fungal spores at the analyzed time.

Following this, a study was run in three greenhouses on Funen, Denmark, for a period of three months in 2022. Each station was equipped with an OPC-N3 with a built-in RH-sensor and a Burkard Hirst type spore trap. In each greenhouse, climate variables were also monitored. A total of 1009 three-hour resolution concentrations were calculated at selected times from the Hirst traps, following standard practice in aerobiology.

Results: Results from the chamber study show that the low-cost particle counter, N3, accurately detects the relative particle size distribution of the spores, when compared with the reference instrument, and therefore potentially can be used in environmental detection of fungal aerosols. It was also seen that the BLIPS-model defines the particle size-distribution of the spores in the chamber well. However, when comparing with Hirst data from the greenhouses, we found no apparent correlation, and the BLIPS estimated from N3 readings did not provide a proxy for high concentrations of *Botrytis* spores.

High RH was seen to mainly affect the lowest bins of the N3 sensor, 0.34 and 0.46 µm, which were not included in the BLIPS model. RH inside the greenhouses was kept low.

Botrytis spore concentrations inside the three greenhouses were seen to vary between 0 and a maximum of 671, 2359 and 3165 spores/m^3, respectively, with diurnal peaks in the afternoon.

Discussion and conclusions: Hirst type quantification was useable for quantification of airborne *Botrytis cinerea* spores in horticultural settings. Strong temporal variation was observed inside the greenhouses, and peaks coincided with high horticultural activity and daylight. Utilizing optical particle counters for detection of fungal peak concentrations still requires further refinement of methods for measuring and/or data analysis. Further studies will be made on additional data from 2023.

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Advancing Pollen Monitoring in Ireland: Integrating Real-Time Technology with Traditional Methods.

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Abstract

Background:

Understanding the impact pollen allergens have on public health, is largely dependent on effective pollen monitoring. This study aims to improve pollen monitoring in Ireland using the real-time monitoring devices (the Swisens Poleno Jupiter) whilst also validating these real-time results via the conventional Hirst sampler methodology.

Methods:

The Swisens Poleno Jupiter was commissioned in Ireland for the first time in January 2023. A fundamental step to achieving accurate pollen recognition with the Swisens Poleno includes building a comprehensive database of particles of interest. By aerosolization of fresh known pollen samples into the instrument, the Poleno's classification algorithm can adapt to ambient pollen and therefore ensure its long-term relevance in pollen surveillance. Swisens Poleno's real-time data acquisition capability was evaluated via its comparison with Hirst data (hourly and daily resolution).

Using historical pollen data from a Hirst trap, updated pollen calendars for Ireland have been designed to highlight the common release periods for allergenic pollen. Air dispersion modelling techniques such as HYSPLIT, were used to determine the origin of pollen in Ireland, thereby helping to streamline environmental monitoring efforts.

Results:

This study marks a significant progress in pollen monitoring by comparing the Hirst and Swisens Poleno samplers, good agreement was seen for many species (R^2 >0.8). Equally by applying air dispersion modelling techniques, (HYSPLIT) an enhanced understanding of pollen dispersion patterns in Ireland was determined, aiding in the identification of sources.

The updated pollen calendars generated from historical data and more recent yearly data provide valuable insights for individuals susceptible to hay fever, enabling better preparation and management of symptoms.

Overall, this study showcases the potential of the Swisens Poleno Jupiter as a powerful tool for improving pollen monitoring in Ireland, offering real-time insights into allergen exposure, and contributing to the advancement of environmental and public health initiatives.

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Continuous Detection of Pathogenic Bioaerosol Using Antibody Labeled Magnetic Beads and Flow Cytometry

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Abstract

Biogenic aerosols play a key role in various infectious diseases, like Covid-19 and Influenza, but also Tuberculosis and Aspergillosis. Therefore, detecting and characterizing bioaerosol is crucial to monitor its transmission and its effects on human health.

Traditional detection of pathogenic airborne particles is either done via collection on agar plates, incubation and subsequent analysis, or via collection on filters and subsequent laboratory analysis (e.g. qPCR, ELISA). While using agar plates is simple, it is time consuming, sometimes even lasting several days for incubation. ELISA or qPCR can be faster, but are labor-intensive, also requiring special laboratory infrastructure and trained staff.

Especially in hospital settings, with high risk of infection due to already compromised immune systems of patients, rapid as well as automated identification of common pathogens transmitted via air is of dire need. Our proposed concept offers a significantly reduced detection time, on the order of minutes to hours, thereby substantially enhancing response capabilities, particularly in time-sensitive environments such as hospitals, healthcare facilities, the food industry, or overly crowded indoor settings.

Hence, in this proof of concept study we build and test a bioaerosol sampler to selectively detect bacteria via antibody labeling and a flow cytometer.

The set-up consists of three main parts. In a first step, the bioaerosol is sampled via a particle into liquid sampler (PILS) at a maximum sampling rate of 500 L air mL^{-1} of liquid. Specific bioaerosols are then selectively identified using an immunoassay and finally analyzed in a flow cytometer.

In this proof of principle study, Escherichia Coli bacteria are used as sample bioaerosol. E. coli have successfully been stained using a primary-secondary antibody array as well as a nucleic acid stain, and can be distinguished and sorted from background and other bacteria.

E. coli was chosen for the proof-of-concept of this study as antibodies are commercially available, it is non-pathogenic, easy to cultivate, and can be modified to carry fluorescent protein, making it possible to confirm its presence in each step. Nonetheless, with the modular approach of using antibodies to detect bioaerosols, any pathogen could be analyzed as long as there is a specific antibody available. This is the concept's main strength, as it makes the set-up versatile like a toolbox. Moreover, it would be possible to detect multiple pathogens in parallel or add a second antibody for more robust quantification. Obtaining different antibody(s) allows

the prospective end-user to detect whatever species relevant in their surroundings, making it suitable for all fields concerned about airborne pathogens bearing antigens.

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Improving the Pollen Forecast for Central Europe by Including Automatic Pollen Monitor Data in the Numerical SILAM Model

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Abstract

In Central Europe the most common allergens are due to allergens from pollen, especially from birch and grass. Our aim was to adapt and improve a statistical forecast model in a local scale, to provide allergic individuals with the risk of pollen exposure in the following days. We adjusted the predictions of the European scale SILAM (System for Integrated modeLling of Atmospheric coMposition) model for birch pollen (*Betula ssp.*) by integrating data obtained from the network of automatic pollen monitors over Europe, specifically in Germany (www. Pollenscience.eu). The study was carried out for Germany, with one station located in Berlin, one in Leipzig.

The prediction of the model with the standard setup showed an overestimation of pollen concentration and early prediction in all the stations in central Europe. In order to improve this prediction, some parameters were adjusted. First, as Birch pollen forecasting model is driven by the climatology and source emission, the local weather was used from Deutscher Wetterdienst (DWD), and the vegetation map from European Atlas of Forest Tree Species (https://forest.jrc.ec.europa.eu/en/european-atlas/atlas-data-and-metadata/), because in other studies this map showed a good correlation with the pollen concentration in the study area. Second, as the season timing was not adequate for our study area (the prediction was to early), we fitted the most appropriate start and end of the season for each station by increasing the heat sum to about 34% from the original setup. And third, the predicted seasonal birch productivity what too high. We reduced this for all station locations based on the measured data. As a result, the start and end of the season had a better correlation between the observed data and the forecast, in some stations the correlation increased up to 30%.

Now that we have the model in the proper timing with the proper concentration predictions, the next step is to keep improving the model with the inclusion of the actual online measured data from the automatic pollen monitoring in an operational setup. With this approach we expect to increase the accuracy on the prediction and provide better prediction to allergenic individuals.

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Allergenic Street Tree Diversity in Australian Urban Landscapes

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Abstract

Background

Allergen exposure in urban areas is often linked to bioaerosols, such as pollen and fungal spores, which can cause allergies and respiratory diseases in humans and other animals. Increasing urbanisation is associated with an increase in allergic disease and greater spread of respiratory pathogens, however there is limited consideration for bioaerosol transmission in urban landscape design, likely due to the lack of knowledge on localised bioaerosol emission, which has previously been difficult to achieve using conventional bioaerosol monitoring methods, such as microscopy. Here we hypothesised that the overall allergenic potential of urban trees varies between cities, due to differences in the composition and distributions of street tree plantings by local authorities.

Methods

Four publicly available street tree inventories in Australia (Adelaide, Melbourne, Perth, Sydney) were examined for the presence of trees with moderate to very high allergenic potential. Data were filtered to exclude blanks and examine species. The number of trees per inventory were converted to percentages to account for differences in land area covered prior to analysis. Alpha diversity measures (Shannon and Simpsons' diversity index) were used to examine the diversity between the four urban areas. Potential allergenicity was mapped onto street tree distributions using R, presuming a 50m emission radius to examine allergen distributions in urban areas.

Results

In the four Australian urban environments surveyed, the most abundant allergenic tree species was *Eucalyptus chamaldulensis*, which was notably highly represented in Melbourne. The majority of very high and high potentially allergenic trees were introduced species, such as *Ulmus*, but native species, such as *Callistemom*, were also widely represented as moderately allergenic species. Melbourne showed the highest proportion of trees with moderate or higher allergenic species overall (33%) while Perth had the least (7.7%). Despite this difference, two-way ANOVA showed that there was no significant in the overall potential allergenicity values between the different urban areas, although there was a significant effect was in the type of allergenic tree species present in each city. By mapping potential allergenicity values and emission radii onto street tree maps it was possible to identify allergen hotspots within urban greenspaces.

Conclusions

The incorporation of potentially allergenic trees in urban environments has important implications for human health and future urban design. There was no difference in overall allergenic potential of each city, therefore suggesting health effects will be comparable between cities. Using potential allergenicity values and street tree inventories it was possible to view allergen hotspots for large urban areas and this improved resolution will allow future landscape design policies to incorporate bioaerosol aware design elements while maintaining planting structures that support biodiversity and native/indigenous heritage.

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Deeper Insight Into Bioaerosols Diversity by Using Image Flow Cytometry and eDNA Analysis

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Abstract

This study aimed to test a combination of novel methods in investigating the aerobiome diversity beyond commonly seen when using standard Hirst type approach.

Four 12-hour samples (two night samples from 6 pm to 6am CET and two day samples 6 am and 6 pm) were collected in Novi Sad situated in the Pannonian region of Serbia. Sampling took place on roof level after the main pollen season in October and November 2023 by using high-volume SASS2300 wet wall cyclone sampler including the SASS 4000 concentrator (flow 4000 l/min). Simultaneously, aerosols were measured with Hirst-type Lanzoni VPPS2010 sampler (flow 10 l/min) and Rapid-E+ air-flow cytometer (flow 5 l/min).

Each of collected 12-hour samples were divided in three equal 1 mL aliquots allowing separate analytical procedures. Environmental DNA (eDNA) extraction was performed using the DNeasy PowerSoil Pro Kit and concentrations were quantified using a fluorometric approach and real-time PCR. Microbial cultivation and colony morphological identification was performed after 72 hours incubation at 26°C on malt extract and tryptic soy agar. Finally, 1 mL of sample with 0.1% glutaraldehyde, stained with nucleic acid binding dye acridine orange (5µg/mL) and analyzed using Amnis® ImageStreamX Mark II imaging flow cytometer.

The Hirst-type samples analysis confirmed abundant presence of bioaerosols (October 2229-9600 pollen and spores/m3; November 1180-4859 pollen and spores/m3) with dominance of Cladosporium spp., Agrocybe spp. and hyaline spores. Microbial cultivation identified viable fungi and bacteria, with CFU/ml ranging from 1x103 to 3.8x105. However, concentrations of extracted eDNA were rather low (under 2 ng/ul). Nevertheless, the results of real-time PCR confirmed the presence of bacterial, fungal and plant DNA in all samples. Image flow cytometry analysis showed that between 19-30% of best focused, single cells were positively stained with fluorescent dye, whereby very few (about 15 %) of those are larger than >10 um.

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Using Dual Polarisation Weather Surveillance Radar to Determine Temporal and Spatial Patterns of Flying Ant Emergence in the Uk

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Abstract

Weather Surveillance Radar (WSR) is a meteorological tool which incidentally records volant animals including insects. The UK's "Flying Ant Day" is characterised by the mass emergence of winged ants (predominantly Lasius niger and Lasius flavus species). Although the UK's Met Office highlights this phenomena in its radar each year, Flying Ant Days, verified through a Citizen Science Survey of over 4500 responses in 2021-2022. Flying Ant Days, verified through a Citizen Science Survey of over 4500 responses in 2021-2022. The biomass concentrations varied on both a temporal and spatial scale. Temporally we tended to see two peaks during the day, typically one smaller then one larger. These peaks were recorded at maximum of 100 mg per m3 on the 9th July 2021 and 30 mg per m3 on 7th July 2022. In addition, using a geodesic approach to the radar beam, high fidelity altitude measurements were made, showing ants routinely detected at altitudes of 2 km and a swarm was observed above 5 km in altitude for around 40minutes on the 17th July 2022. This study highlights the untapped potential of using WSR in aeroecology and aeroentomology to shed light on the complex behaviours of flying ants, revealing significant insights into their emergence, biomass distribution, and altitude preferences during the UK's Flying Ant Days. By bridging the gap between meteorological observation and ecological research, we pave the way for future interdisciplinary studies to broaden this approach providing insight into both local ecosystems and global biodiversity.

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Sampling Spores and Microorganisms in the Stratosphere

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Abstract

Spores are the survival and dissemination units of fungi. Many are designed for optimal airborne dispersal while maintaining long-term survival. Depending on the chemical and structural nature of their walls, they are highly resistant to extreme temperatures and UV radiation. It has been shown that *Botrytis cinerea* conidia stored dry at -80°C are still able to germinate after more than 20 years in storage. Given their anemochorous nature and resistance to abiotic factors, it would therefore be possible for spores of pathogenic fungi to be aeroported through the stratosphere, i.e., the atmospheric layer between 10 km and 50 km altitude. The stratosphere receives intense ultraviolet radiation, and temperatures generally ranging from 0°C to -70°C. Little is known about the spread of pathogenic fungi in high-altitude airspace. Our study examines the possibility of the presence of viable fungal spores in the stratosphere and the exploration of the diversity of non-cultivable fungi, bacteria and pollen above 12,000 meters using biomolecular techniques.

For this purpose, we designed a low-cost device capable of sampling particles in the stratosphere. It consists in a sealed polystyrene box with two ports on the top and bottom sides, allowing air circulation. A rotating arm sampler with four sticks coated with petroleum jelly spin in the resulting air stream. The opening of the ports is performed by mobile covers driven by servomotors, controlled by an Arduino Uno microcontroller connected to a pressure sensor. Moreover, an on-board radiosonde continuously transmits GPS position, relative humidity, and temperature data. An internal camera captures the opening, closing, and sampling processes during the desired altitude segment. Additionally, a control box, that never opens during flight, monitors potential contamination below the stratosphere. Both the measurement and control boxes are sterilized under UV-C, sealed and attached to a hydrogen-inflated radiosonding balloon. Upon reaching an altitude of 12,000 meters, the cover's device opens, and airborne particles are collected. Once the balloon bursts (at around 35,000 m; -63°C), a parachute deploys during the descent, and the cover closes at 12,000 meters. This ensures exclusive sampling in the stratosphere. Optionally, the payload can be dropped at a desired altitude.

Two test flights conducted in October 2023 and January 2024 up to 21,000 meters altitude, allowed us to validate the device and the sample recovery method and to confirm the optimal sampling conditions. Half of the collected samples were cultured on fungal medium, while the other half underwent for deep sequencing. Our results revealed that the control box remained sterile, indicating no contamination during sampling. Furthermore, six living species of fungi were identified from the winter sampling, demonstrating the viability of spores despite low pressure and temperature. The next phase involves multiple flights up to 35,000 m throughout the year and various altitude ranges to gain a better understanding of spore localization and diversity, contributing valuable insights to plant/animal diseases management and ecological studies.

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Comparison of Allergy Potency of Poaceae Pollen in Three Monitoring Stations in Türkiye

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Abstract

Background and Objectives

Airborne pollen grains from the Poaceae (Grass) family are among the most significant allergens responsible for allergic diseases affecting the upper and lower respiratory systems. The Poaceae family is widely distributed in Türkiye, with several genera contributing significantly to pollinosis, including *Phleum* sp., *Dactylis* sp., *Lolium* sp., *Festuca* sp., *Poa* sp., and *Cynodon* sp. Allergy to Poaceae pollen is highly prevalent in our country, with its effects intensifying during certain seasons. In this study we aimed to investigate Poaceae pollen and Phl p 5 levels in three stations for a year showing the influence of meteorological factors on their distribution.

Material and Methods

The aerobiological data were recorded at three monitoring sites (Ankara, Zonguldak and Gebze) situated in Türkiye by using a volumetric Hirst-type pollen trap for grass pollen and a volumetric multi-vial cyclone sampler for Phl p 5 allergen during the pollen season in 2022. Phl p 5 allergen concentrations were measured by ELISA assay.

Results

Higher pollen and allergen concentrations were registered at Gebze station, situated in the northwest part of the country, with the Seasonal Pollen Integral 104% higher than Zonguldak station and 227% higher than Ankara station. Pollen potency was also found to be higher at Gebze station. The associations between pollen and allergen concentrations were found to be significantly and positively correlated in all stations. However, during some days of the flowering season, the allergen concentrations did not correspond to the airborne pollen values. Based on the results of Spearman correlation analysis, in Ankara station, Poaceae pollen exhibited an inverse correlation with pressure, while Phl p 5 allergens showed a positive correlation with temperature and a negative correlation with relative humidity. In the Zonguldak station, Poaceae pollen showed a negative correlation with temperature, relative humidity, and total precipitation, whereas Phl p 5 allergens exhibited a positive correlation with temperature and wind speed, and a negative correlation with total precipitation and pressure. In the Gebze station, Poaceae pollen showed a negative correlation with temperature, relative humidity, and

total precipitation, while Phl p 5 allergens exhibited a positive correlation with temperature and a negative correlation with relative humidity.

Conclusion

The variability in Poaceae pollen loads across different monitoring stations may be influenced by the unique characteristics of the local flora in each area. Since existing larger urban green areas in Gebze station could cause the highest allergy potency and affect the people who live there.

Key words: Poaceae, pollen, Phl p 5, allergen, meteorological factors

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Large Intra-urban Variations and Autocorrelations of Grass Pollen Concentrations in the City of Aarhus, Denmark

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Abstract

Background: Grass pollen is the most important outdoor aeroallergen in Europe. Traditionally, public warning systems and epidemiological studies on grass pollen concentrations are based on daily concentrations from a single observational site. This may introduce substantial uncertainties in the exposure studies and the warnings to the public. Previous city scale studies from Aarhus reported seasonal development in the peak time of grass pollen . They also demonstrated dependence of daily grass pollen concentrations on the location of the sources within the urban zone. How this impacts short term variations of grass pollen exposure in the urban environment is unclear. Here we study intra-urban variations in 2-hourly grass pollen concentrations within the city of Aarhus.

Methods: Three Burkard pollen traps were placed on buildings 15m-20m above ground in the city of Aarhus in 2010 and 2011. Pollen from grasses are identified at the family level using 13% of each slide. Daily and 2-hourly grass pollen concentrations are calculated using recommended terminology for aerobiological studies. The daily concentrations are used to determine the pollen season according to the 95% method. The 2-hourly concentrations are compared between the three sites as well as with operational data from the nearby city of Viborg 60km away. The data from the sites are tested for normality, spatial correlations and statistical significance here calculated using either parametric or non-parametric methods. Finally, we analyse the autocorrelation – or temporal correlations - in the 2-hourly data on all data from the four sites. All calculations are performed in Python.

Results: Pollen seasons, defined by Viborg data, comprised 61 and 82 days for 2010 and 2011 accordingly. The season pollen integrals at the four sites vary within 1458-2376 pollen*day/m^3. Each site provided 1482 2-hourly data points covering 2 seasons. The normality test show non-normal distributed data and correlations based on the Kendall-tau test statistic vary from 0.37 to 0.47, all statistical significant. In general, autocorrelations show a gradual decrease with increased lag-time at all sites, modulated with a cyclic variation. Highest autocorrelations, up to 0.4 are found for exactly 24h lag time. Lowest autocorrelations, down to 0.14 with 12h and almost zero with 36h lag time.

Discussion and Conclusion: The 2-hourly concentrations show medium correlations between all sites, hence considerable variation over the urban zone. Likewise, it demonstrates a spatial correlation. The autocorrelation analysis shows the data are strongly temporally dependent. Consequently, any outcomes on statistical significance should be treated with great care. Importantly, the autocorrelations are very strong, where the 24h cycle is apparent several days in advance. Equally important, there is limited autocorrelation only with 12h and 36 lag time, demonstrating a limited connection between daytime and nighttime values. A likely reason is limited long distance transport combined with high settling velocities and considerable emission of grass pollen within the urban zone. This study did not take into account urban meteorology and other environmental variables, but found significant correlations. This suggest that meaningful correlations are achievable for predicted grass pollen concentrations within the urban zone using relevant environmental variables.

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Birch Pollen Modelling Over Central Europe – Model Evaluation and Sensitivity Analysis

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Abstract

Modeling and forecasting of pollen concentration and pollen season properties are among the most important goals of aerobiology and have wide social impact, as the number of affected individuals is increasing. Although Chemical Transport Models (CTMs) have been used for air pollution modelling for more than 40 years, a growing effort to simulate pollen dispersion and concentrations with complex CTMs has started in the last decade. The application of CTMs for pollen modelling gives the ability to estimate the population co-exposure to air pollution and bioaerosols with the same modelling framework.

In this study we investigated the performance of EMEP MSC-W, a state-of-the-art Eulerian chemical transport model, in predicting the birch pollen season for Poland in five chosen years. Furthermore, we performed a 'sensitivity study' for two seasons to test how the model responds when selected emission parameters are modified. The emissions were modelled using a heat-sum-driven deterministic model. We ran the simulations for seasons from 2015 to 2019 (BASE runs). The sensitivity study was performed for years 2018 (moderate emission year) and 2019 (high emission year) and as its part we modified individually either the total amount of birch pollen grains (N_tot_birch), or season length in growing degree-days (delH). The predicted birch pollen concentrations were compared with observational data obtained from 12 sites in Poland. The observations were

gathered using a Burkard 7-day volumetric pollen trap and analysed following the recommendations of the International Association for Aerobiology.

Overall, the model tends to predict both the onset of the season and its end to occur too early when compared to the observations. The mean difference (for all stations), between the model and observations, in the start of the season varies from 3 days in 2016 to 12 days in 2015 and 2019. The seasonal pollen integral (SPIn) and the concentrations tend to be underestimated, especially in high seasons. The modification of delH results in later ends of the season, decreased concentrations in the first part of the season, and higher concentrations toward its end. Modification of N_tot_birch resulted in an increase of concentrations and SPIn in a directly proportional manner. The time series of pollen concentrations for different N_tot_birch values have the same temporal course, with the difference being a multiplication by a constant factor.

In conclusion, changing N_tot_birch can improve the overall magnitude of predicted concentrations, while increasing delH does seem to improve the overall predicted season characteristics and evolution, however with little to no effect on overall pollen concentrations and SPIn. However, premature season start still needs to be addressed in the model.

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Exploration of Microplastic Existence and Diversity in Indoor and Outdoor Air Samples: Morphological and Polymeric Analysis

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Abstract

Background and Objectives

Microplastics, defined as non-biodegradable particles smaller than 5 mm in size and originating from petroleum-based sources, can exhibit various shapes or be shapeless. They accumulate easily in the atmosphere due to their small size and frequently use in areas such as textile, construciton, industry and factory. So it was occur very important issue for our future.Most studies have focused on aquatic environments but in our daily life we exposed to microplastics when we inhaled. Their prevalence in both terrestrial and aquatic environments has led to significant concerns regarding their impact on human health and the environment, prompting numerous studies in recent years. While research on atmospheric microplastics remains a

burgeoning field, their significance in the air we breathe is increasingly acknowledged. This study aims to investigate the presence of microplastics in indoor and outdoor air using active and passive sampling methods.

Material and Methods

Passive sampling was conducted indoors using glass petri dishes in areas such as classroom, laboratory, and herbarium at Ankara University's Beşevler 10th-year campus, while outdoor sampling utilized a High Volume Air Sampler (BGI900 model) on the building roof. In active sampling, we collected air samples in PM10 filters. Glass petri dishes were left for 2 days to collect samples. For the combustion process to remove organic matter, a 30% H_2O_2 solution was utilized for all samples. After incubating at 65˚C for 72 hours, they were filtered using a vacuum filtration set with nylon net filters (pore size: 10µm). Filters were examined under a stereo microscope. Microplastics were then selected and classified based on their shape (fiber, fragman, foam, beads, film), size and color under a stereo microscope. FT-IR analysis was employed to characterize the polymeric structure of microplastics obtained in significant quantities.

Results

Fibers were found to be the most prevalent shape type in both indoor and outdoor airborne microplastics, followed by fragments. ATR-FTIR analysis revealed polyethylene terephthalate (PET), polyamide 66 (PA 66), polypropylene (PP), polyurethane-Reaction Injection Molded-amine (PUR-RIM-AMINE), polyoxymethylene (POM), acrylonitrile butadiene styrene (ABS), polyvinylchloride (PVC) and polyamide 6 (PA 6) as the most prevalent polymer types in both environments.

Conclusion

Given the limited research on atmospheric microplastics, this study contributes to understanding the potential significance of atmospheric microplastic pollution and aims to draw attention to critical perspectives on this issue.

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Potentially Pathogenic and Antibiotic-resistant Bacteria in the Air in Public Buildings

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Abstract

Growing antibiotic resistance among bacteria is one of the main threats to public health in the modern world. In 2017, WHO published a list of 12 "priority pathogens" that pose the greatest threat to human health due to their antibiotic resistance phenotype. The list includes, among others, carbapenem-resistant, extended-spectrum β-lactamase-producing *Enterobacterales* (CRE and ESBL), vancomycin-resistant *Enterococcus faecium* (VRE), and methicillin- and vancomycin-resistant *Staphylococcus aureus* (MRSA and VRSA). These bacteria are particularly dangerous due to the possibility of transmitting resistance genes through horizontal gene transfer. One of the genetic elements that enable bacteria to acquire and incorporate resistance genes into their genome are integrons. They are widespread among bacteria, especially those of anthropogenic origin. The aim of the study was to determine the occurrence of potentially pathogenic and antibiotic-resistant bacteria in the air collected in public buildings in Poznań, Poland. The frequency of class 1 integrons among airborne heterotrophic bacteria was also examined.

Indoor air microbiota samples were taken in two secondary schools, two university buildings, one health care clinic and one pharmacy. The air microbiota was collected by sedimentation method and with the use of Sartorius MD8 Airport microbiological sampler. Bacterial isolates were selected on Mannitol Salt Agar, Brilliance E. coli / Coliform Selective Agar, Cetrimide Agar and Brilliance CampyCount™ Agar. Dedicated chromogenic media were used for selection of ESBL, CRE, MRSA, and VRE strains. In order to detect integron-harboring strains, bacteria were isolated on BHI agar medium supplemented with streptomycin. Class 1 and 2 integrase genes were then identified by PCR. The ESBL and CRE phenotypes were confirmed by the disk-diffusion methods and by using PCR assays to identify the respective antibiotic resistance genes. Identification of potentially pathogenic bacteria was carried out by using Sanger sequencing and API tests (BioMérieux) as well as species-specific PCR assays.

In the indoor bioaerosol, the total number of bacteria ranged from 4.7×10^2 cfu/m^3 to 3.2×10^3 cfu/m^3 and the number of coliforms from 0 to 1.3×10^2 cfu/m^3. Gram-positive bacteria were prevalent in the indoor air and included opportunistically pathogenic species like staphylococci (*Staphylococcus aureus, S. edpidermidis, S. lugdunensis*, and *S. haemolyticus*) and enterococci (*Enterococcus faecalis and E. faecium*). Gram-negative potentially pathogenic bacteria included *Enterobacter cloacae, Escherichia coli, Pantoea agglomerans, Acinetobacter baumannii*, and *Pseudomonas fluorescens*. In the indoor air, there were streptomycin-resistant *intI1*-positive heterotrophic bacteria present. Their number ranged from 1.6×10^2 cfu/m^3 to 4.2×10^2 cfu/m^3, their frequency from 9% to 33% and was the highest in the pharmacy. Moreover G(-) rods resistant to carbapenems and methicillin-resistant staphylococci were found. The presence of methicillin-

resistant bacteria was particularly alarming as their number in the air from one of the university buildings reached 1.8×10^1 cfu/m^3. The results of the study showed that air may be a route of spreading of opportunistically pathogenic and antibiotic-resistant bacteria.

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Evaluation of the OPC-N3 as a Low Cost Pollen and Spore Counter in a Controlled Environment

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Abstract

Background: Real-time instruments for automatic pollen detection is currently revolutionizing aerobiology. However, many instruments are very costly, putting a limit on the number of instruments that can be used within a country. Low cost instruments, with an expected lower accuracy, could here be a supplement to these expensive instruments. Therefore, we explore the real-time particle counter, the OPC-N3 from Alphasense, as a low-cost candidate for pollen detection.

Methods: We used a OPC-N3 optical particle counter to detect pollen in a controlled setting. Pollen were aerosolized by the Swisens atomizer within a Biohazard-II cabinet. This maintains a clean environment and prevents false signals from larger particles originating from external sources. A funnel was used to guide bioaerosols emitted by the atomizer directly into the 5 mm wide inlet of the OPC-N3. Between each experiment, we conducted a thorough cleaning of the area and verified the absence of pollen and spores before a new test run. We investigated eight bioaerosols: *Alnus glutinosa*, *Ambrosia artemisiifolia*, *Corylus avellana*, *Dactylis glomerata*, *Lolium perenne*, *Lycopodium spp*, *Parietaria judaica* and *Urtica dioica*. The pollen and spore samples were acquired from commercial providers. Throughout the experiment, real-time particle counts were monitored on a laptop using Alphasense's dedicated software. Particle emissions were targeted to approximate a rate of one particle per second. The incoming data were recorded as particle counts in each of the 24 size bins (between 0.35-40 µm), with measurements taken every second. The collective particle counts for bioaerosols were then presented as a particle size distribution density curve for each species and it is assessed if the particle counter is able to detect at least 50% of the incoming particles.

Results: Five out of the investigated species exhibit size distributions that closely resemble Gaussian distributions. For these five species, the peak particle size is prominently clustered within specific bins resembling the typical size of the pollen grains. In contrast, the two grass species, *Lolium perenne* and *Dactylis glomerata*, exhibit only partial adherence to a Gaussian distribution, most likely because most of the grass particles are too large to be detected by the OPC-N3. Lycopodium spp, the sole non-spherical particle, presents a distinct pattern with particles distributed across bins 5-12 (1.3-10.0 μm) and a significant number in bins 22-24 (>31 μm). As with the grass species, identifying the most likely detected particle size for Lycopodium spp remains a challenge.

Discussion and Conclusion: The results suggest that the OPC-N3 can directly detect smaller spores, smaller weed pollen and most tree pollen, while it cannot be used to directly detect grass pollen. Importantly, we used pollen from commercial providers which are dehydrated and defatted. Fresh pollen can be expected to be slightly larger. This will mean that the capture rate will be slightly smaller in a field setting. Nevertheless, the OPC-N3 can be a valuable low-cost instrument, depending on the experimental setting for the direct detection of many pollen types.

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Does a Changing Climate Matter to Plants? Results of Phenological Observations From VUŠA Botanical Garden

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Abstract

As the world records climate changes, not only fears about the consequences, but also evidence of clear changes in nature due to climate change continue to emerge. Questions related to changes in plant phenology have arisen and research has been started for at least a couple of decades.

Study's purpose – after reviewing the 17-year monitoring data, to check whether the time of occurrence of phenophases and duration of vegetation in VUŠA Botanic Garden changes significantly due to relatively rapid global climate changes. **Hypothesis** – in times of rapid climate change the results of the phenological observations of the first period of phenological observations (2007-2015) differ significantly from the results collected in the second period (2016-2023).

Methods. In Vilnius University Šiauliai Academy Botanical Garden (23.267° E, 55.917°, alt. 117 m) 18 species are obtained from IPG and observed since 2007. By the IPG requirements, the following 8 phenological phases are observed: UL, MS, JS, BF, FF, RF, CL and LF. Non-parametric Mann–Whitney U test was used to compare periods, and Spearman's correlation for the phase occurrence trends.

Results. Comparing the beginnings, endings and lengths of the vegetation seasons in the first period of phenological observations with the second period for all species in general, it is noted that recently vegetation ends at the same time (N.S.), begins earlier ($Z=-2.56$; $p<0.05$), and therefore the seasons are getting longer (N.S.). For most of the species observed, the data shows trends, but they are statistically unreliable, however, some indicate the following, statistically reliable changes over the observation period. In *Corylus avellana* L. case, the length of the season in second period increases by an average of 25 days ($Z=-2.407$, $p<0.05$), the beginning of the season (flowering) occurs earlier by an average of 22 days ($Z= -2.118$, $p<0.05$), the unfolding of the leaves ($r_s=0.74$, $p<0.01$) and ripening the fruit ($r_s=-0.58$, $p<0.05$) occurs earlier. According to G. Kalvane et al. (2009), during 30 years till 2000, the flowering of European hazel in Lithuania advanced by 3.5 to 27.4 days. Therefore, our result shows the process of change is still under way. Half of our observed *Salix* species shows finish of vegetation season earlier: *Salix × smithiana* Willd. by 7 days ($z=-2{,}66$, $p<0{,}01$) and *Salix viminalis* L. by 8 days ($z=- 2{,}03$, $p<0{,}05$) on average. Previous study shows the onset of phenological autumn in Latvia and Lithuania also started earlier (Kalvane et. al. 2009). The start of the season of *Sorbus aucuparia* L. was brought forward by 8 days ($z=-2{,}12$, $p<0{,}05$), the plant blooms earlier ($r_s=-0{,}59$, $p<0{,}05$), but the fruit ripens later ($r_s=0{,}71$, $p<0{,}01$). According to our results *Larix decidua* Mill. unfolds its leaves also latter ($r_s=0{,}72$, $p<0{,}05$). **Conclusions.** Five plants out of 18 observed under the IPG program in the botanical garden respond to changing climatic conditions in the duration of the vegetation season and changes in the time of phenophases. The species that indicates the most changes is common hazel, this can threaten to the harvest, the associated fauna, but also people suffering allergies.

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Influence of Meteorological Conditions and Climate on Pollen Season Characteristics for the Early-flowering Plant Taxa in Poland

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Abstract

Background: Air pollution poses a significant threat to health, the economy, and the environment. In the context of air quality, there is a growing focus on biological particles, particularly allergenic pollen grains. It is estimated that 40% of allergies are attributed to pollen grains. Factors such as urbanization, anthropogenic air pollution, and especially climate change are believed to contribute to the increasing prevalence of allergic diseases. Therefore, it is crucial to understand the variability of pollen seasons and the key factors affecting them.

Aim: The study aims to determine the relationship between the characteristics of the pollen season and meteorological parameters, and to assess the impact of recent climate change on the variability and trends of pollen seasons.

Methods: In total, five early-flowering plant taxa i.e.: alder (*Alnus* sp.), hazel (*Corylus* sp.), poplar (*Populus* sp.), elm (*Ulmus sp.*) and willow (*Salix* sp.), were considered in the analysis. Pollen data were collected by Hirst-type volumetric traps at 9 stations located in Poland, Central Europe, between 2003 and 2022. Weather data were gathered from nearby meteorological stations. Pollen data from particular stations were compared with each other to indicate their spatial variability. Machine learning algorithms, such as random forest, gradient boosting, and neural network, as well as agrometeorological variables like SAT (sum of active temperature), Growing Degree Days (GDD), and Chilling Hours (CH), were used to assess the influence of meteorological variables on pollen season characteristics. Additionally, trends and changes in pollen seasons over the study period and crucial parameters for indicating the impact of climate change, have been examined. We determined the dynamics of change between two variables - time and pollen season characteristics (the date of the beginning and end of the season and the maximum amount of pollen from the entire season).

Results: Stations located in Central Poland exhibits the highest year-to-year variation in pollen season characteristics, which is related to a transitional influence between Atlantic and continental effects. Mean, maximum, and minimum air temperatures, as well as insolation duration, were the parameters that had the strongest impact on pollen season course. Additionally, air humidity and precipitation were found to be key factors due to their potential influence on the wet deposition of pollen grains. Most of the examined taxa exhibited a trend toward an earlier start to the pollen season. Some taxa, especially in the western part of Poland, revealed longer and more intensive seasons.

Conclusions: The analysis showed a close relationship between meterological conditions and the course of the pollen season of the studied taxa. The observed trends of longer and more intense pollen seasons may have a particularly negative impact on allergic people.

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Health Cost of Airborne Allergenic Pollen – a Case Study for Birch and Grass

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Abstract

Exposure to respirable allergenic materials (aeroallergens) from bioaerosols can stimulate the production of antibodies in the human body and cause allergic airway diseases (AAD), such as asthma and allergic rhinitis. AAD is a serious public health concern worldwide with the most prevalent impacts among children. Asthma in general is estimated to account for about an average of over 1300 deaths per day globally. Moreover, several studies have shown that climate change will worsen the impact of allergy in the next decades.

The aim of this study is to estimate the impact of allergenic pollen concentrations on population morbidity. We have extended the AlphaRiskpoll tool, which is regularly used for anthropogenic pollution haelth impact analysis towards two most allergenic in Central Europe pollen taxons – birch and grass. We have analysed several health endpoints such as e.g. severe allergy symptoms, rhinits or eye irritation, dyspnea, nasal symptoms and related to that emergency department visits and taking allergy medications. The relative risk functions were developed based on recent literature reports from European studies. The AlphaRiskpoll were used for 20 years of pollen observations (years 2003-2022) from south-west Poland (Wrocław station). Airborne pollen counts were gathered using a Burkard 7-day volumetric pollen trap and analysed following the recommendations of the International Association for Aerobiology. Pollen grains are counted under a light microscope with 400 magnifications along four longitudinal transects. The results were expressed as the number of pollen grains per m3 of air as a daily mean value (pollen grains m-3). The airborne pollen concentrations, together with population data were used as input data to the AlphaRiskpoll. The results show that the number

of pollen-related symptoms fluctuates from year to year. During 2003-2022, 11 people died prematurely on average per year due to birch pollen exposure. In addition, the findings indicate a statistically significant increasing trend in e.g. dyspnea, children asthma hospitalization and ocular symphoms in the last 20-years.

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Airborne Basidiospores of Thelephoroid and Tomentelloid Fungi

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Abstract

Thelephoroid and Tomentelloid fungi include mycorrhizal, saprobic, wood decay fungi and belong to Thelephorales. Basidiospores of Thelephoroid and Tomentelloid fungi are present in the air and appear in the spore-trap samples from time to time. However, the airborne basidiospore of these fungi are less studied or overlooked in aeromycology and often identified as unknown spores. These basidiospores from air samples collected in the last 10 years have been morphologically studied and compared with the basidiospores from fruiting bodies collected from forests or voucher specimens to determine their identities. Basidiospores of Thelephoroid and Tomentelloid fungi: *Beoletopsis*, *Hydnellum*, *Sarcodon*, and *Thelephora*, *Tomentella* are described.

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The Expression Level of Allergenic Proteins in Mugwort (*Artemisia Vulgaris*) Pollen Grains in Areas With Varying Degrees of Urbanization.

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Abstract

The continuous increase in the number of people suffering from pollen allergies is particularly exacerbated in large cities. Studies conducted on various taxa often show increased pollen grain production as well as allergenic proteins in plants growing in areas with increased air pollution and urbanization. Allergy to weed pollen proteins is a serious health problem and one of the main causes of respiratory allergies worldwide. In particular, mugwort (*Artemisia vulgaris*) pollen is one of the most important herbaceous allergen sources in Europe, where its pollination season occurs in late summer, causing the majority of allergy symptoms.

The aim of the study was to answer the question of whether *Artemisia vulgaris* pollen grains produce varying amounts of allergens depending on the degree of urbanization, and thus environmental pollution, in which the plant grows. Populations growing in the city of Poznań (Poland) were selected for the experiments, including those in the city center with very busy streets, and on the outskirts of the city in areas characterized by low urbanization and low traffic.

Quantitative determination of the level of protein expression was performed using real-time PCR method. Experiments were conducted for six known allergens of mugwort pollen grains.

A. vulgaris allergens classified into families of pathogenesis-related proteins exhibited higher expression levels in more urbanized areas compared to populations on the outskirts of the city. A similar relationship was observed for pectate lyase. Conversely, allergenic profilin showed higher expression levels in agricultural areas located on the outskirts of the city. According to the adopted criteria for dividing the studied populations, the variability in the expression level of pollen allergenic polcalcin was not correlated with the degree of urbanization. The recorded differences in the expression level of transcripts of the studied proteins in selected populations indicate that the environment in which the plant grows is a factor that can influence the amount of produced mugwort pollen allergens.

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Distribution *Helminthosporium* and *Fusarium* Concentrations in the Wheat Crop and Its Relationship With Meteorological Variables.

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Abstract

Wheat is one of the most important cereal crops in the world and is used as a staple food, as it is a source of calories and plant-based protein in the human diet. This crop is susceptible to attack by different phytopathogenic fungi such as *Helminthosporium* or *Fusarium* and can devastate plantations. *Helminthosporium* is a devastating pathogen that causes foliar and root diseases, *Fusarium* reduces grain yield and quality, which compromises harvest and marketing. Our goal is to understand how environmental conditions can influence the behavior of *Helminthosporium* and *Fusarium*.

The study was carried out in a wheat (*Triticum aestivum*) plantation of the Galician autochthonous variety Caaveiro, located in A Limia, Ourense, Galicia, in Northwestern Spain. The study was conducted during three growing seasons from 2021 to 2023. A Lanzoni VPPS-2010 volumetric sampler is used to monitor spores. The identification and counting of spores in the air have been carried out following the methodology proposed by the Spanish Aerobiology Network. The phenological study was conducted according to the BBCH scale. Meteorological variables have been provided by a meteorological station located near the spore trap.

The concentration of *Helminthosporium* spores was higher in the 2022 season (1173 spores) with a peak concentration ocurring on June 29 with 66 spores/m^3 coinciding with the Inflorescence emergence stage (S-5). On the other hand, the higher *Fusarium* concentration take place in 2021 (2953 spores), the peak concentration was registered on May 20, 2022 with 339 spores/m^3 coinciding with the Germination stage (S-0). The meteorological variables that exert the greatest influence on the concentration of *Helminthosporium* and *Fuarium* were the parameter related to temperatures.

External aerobiological sensors together with knowledge of the phenological and meteorological variables of a certain region can become a valuable tool to know the phytosanitary status of a crop and thus make appropriate decisions regarding the application of different phytosanitary treatments.

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Quercus Pollen: Trends and Allergy Risk in a Climate Change Scenario

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Abstract

The *Quercus* genus, representing an important natural resource, is also described as a high sensitizing pollen type. As the most prevalent pollen type in Alentejo, it potentially impacting respiratory health of susceptible individuals. The aim of this study was to evaluate the impact of *Quercus* pollen on respiratory allergic disease in Alentejo, in a climate change scenario._

Data on antihistamines sales (group-1 histamines antagonists - G1); group-2 SOS drugs - G2) were obtained from the National Pharmacy Association (ANF) (2004-2021). The pollen was monitored (2002-2021) using standard Hirst-type traps and identified by optical microscopy, according to the standard methodology (REA.com). The meteorological variables were obtained from ICT/CGE platform (Portugal) and their impact on the Seasonal Pollen Index (SPIn), Pollen Season Duration (PSD) and Daily Pollen Concentrations (DPC) was investigated by statistical methods.

The results evidenced an impact on allergic disease, by positive associations of the April's SPIn with April's antihistamines sales, both G1 and G1+G2 (R: 0.656*; p=0.028 and R: 0.642*; p=0.033, respectively). Meteorology strongly influenced the SPIn but not the PSD. The meteorological factors on SPIn, a negative correlation with precipitation (P) (R:-0.549*; p=0.022), relative humidity (RH) (R:-0.506*; p=0.045) was observed, while the wind speed (WS) was positively correlated (R: 0.689**; p=0.002). Meteorology also influenced the DPC. Positive correlations between the temperature, global solar radiation and WS and direction and DPC were observed. On the contrary, P and RH were negatively correlated with DPC.

The results suggest that *Quercus* pollen contributes to the worsening allergy symptoms in Alentejo evidenced by the increase in the antihistamines sales. Moreover, it suggests that by influencing the pollen loads, climate affects the risk of exposure to this sensitizing agent thus changing allergic respiratory outcomes over the years.

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Grass Pollen and Aeroallergens and Their Relationship With Meteorological Factors in Ourense City (Nw Spain)

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Abstract

The report proposed by the European Academy of Allergy and Clinical Immunology estimates that 150 million European citizens suffer from chronic allergic diseases. At the current time, imminent climate change influences the onset, duration and intensity of the pollen season for many species, which will lead to an increase in allergenic diseases in the future. In Europe, the prevalence of pollen allergy in the population is around 40%, with grass pollen-induced allergy being the most common. In the present study we detected in the atmosphere grass pollen and the allergen Lol p1, which is a Beta-expansin with a molecular weight of 27 kDa. Our objective is to determine the concentrations of grass pollen and allergens and relate them to meteorological variables to warn allergic people of periods of high concentrations of both, pollen and allergens.

The study was conducted in Ourense, in the North West Spain from 2009 to 2018. Pollen sampling was carried out using a Lanzoni VPPS-2000 volumetric trap, and for the allergen quantification a Multi-Vial Cyclone Sampler was used. Allergen content in the samples were detected using a specific 2-site ELISA and Lol p1 antibodies. Meteorological data were provided by the Agencia Estatal de Metorología (AEMET). The Principal Component Analysis (PCFA) was applied to determine the interrelationship between pollen allergen and weather variables. Finally, we performed a correlation analysis with the objective of knowing which are the climatic variables that exert the most influence on pollen and allergen concentrations.

During the study period grass pollen and allergens were detected in the atmosphere from last days of April to August. The higher total pollen concentration was registered in 2016 with 6658 pollen grains and the pollen peak took place on 2010 with 421 pollen/m^3. The higher total allergen concentration was registered in 2013 with 24.38 ng and the allergen peak was registered in the same year with 1.67 ng/m^3. From the PCA analysis, 4 principal components have been extracted, since they had eigenvalues greater than or equal to 1.0 and the components together represent around 86.6% of the variability of the data set. In the same way, a correlation analysis was carried out, it was noted that the variables that exert the greatest influence on the concentrations of pollen and allergens were temperatures, and a negative influence on the concentrations was exerted by the variables rain and relative humidity.

Studies on the effects of pollen and its aeroallergens on human health are essential, as allergies are likely to increase in the future due to climate change. For this reason, pollen and allergen studies can be a valuable tool to predict periods of risk of grass allergy and thus be able to alert people sensitive to this type of pollen.

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New Regional Pollen Calendars for Sweden and Some Observations of Changes in Seasonality

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Abstract

The current Swedish pollen calendar has for 30 years been a help, both to allergic people and to the professionals, such as medical doctors. With changes in climate and vegetation, there is now a need to update the calendar and to separate it into regional calendars.
Here we show new pollen calendars based on up to 40 years of pollen data from 23 sites in Sweden. Many of these coincide well with the previous calendar but Sweden is a long country with climate zones including oceanic, summer-warm humid continental, subarctic and tundra. We have thus made regional calendars by coalescing the local sites into four regions based on seasonal statistics, such as start and end, periods of high levels and duration.
The seasons were calculated for ten allergenic pollen taxa (Corylus, Alnus, Salix, Ulmus, Betula, Fagus, Quercus, Poaceae, Artemisia and Ambrosia) and four that are considered non-allergenic. (Pinus, Picea, Tilia and Taxus/Juniperus).
The recalculation has also confirmed previous studies that show changes in seasonality towards earlier flowering, especially for the spring flowering species. We can also see that the changes in seasonality are not the same in all places. With the biggest difference being between northern and southern Sweden.

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Biological Characterisation of Hailstones from Two Storms in South Brazil

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Abstract

Although studies focusing on the physicochemical properties of aerosols/clouds have not been performed extensively, even less attention has been given to hailstones and their biological composition. Here, we present the results of the physical and microbiological characterisation of 20 hailstones collected in Southern Brazil originating from two storms. Nearly half of the hailstones (9 out of 20, or 45%) did not contain any cultivable bacteria or fungi. A total of 18 bacterial species were found in hailstones from both storms, and the genus *Bacillus* was found in 5 out of the 11 hailstones, with *Bacillus cereus* being the most frequent bacterial species. Fungi, on the other hand, were only present in four hailstones derived from a single storm, with three fungal species identified and *Epicoccum nigrum* being the most frequent fungal species. HYSPLIT modelling indicated the different flow of air masses from the Amazon and Pacific Ocean that contributed to the loading of microorganisms found in the clouds at the time of the two storms. Our findings suggest that ca. 50% of hailstones have cultivable bacterial or fungal species, which came mainly from the local landscape with intrusions of air masses derived from the Amazon and the Pacific Ocean.

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Exploring a Novel Measurement Method Based on Fluorescence Measurements and Holographic Images and Its Application to the Characterization of Plant Debris – a Laboratory Study

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Abstract

Plant debris is a subgroup of primary biological aerosol particles (PBAP) and comprised of decaying and fragmented plant materials such as leaf litter. Plant debris is considered the largest mass fraction of PBAP and the main source of atmospheric cellulose. Leaf litter might act as atmospheric carrier for bacteria or fungi and cellulose can also act as cloud forming particle. Plant debris is an understudied fraction of biological particles and studies are mainly based on offline techniques involving filter sampling and extensive chemical analyses.

In this laboratory study we use a novel state-of-the-art online bioaerosol monitor, the SwisensPoleno Jupiter, and explore its potential to characterize plant fragments based on fluorescence measurements and holographic images. For each particle (detection size range: 0.5 to 100 µm), the instrument obtains two images from orthogonal views and measures the fluorescence emission within five wavebands (center wavelength: 357/435/483/562/676 nm) triggered by three excitation light sources (280/365/405 nm).

In the first part of the study, we evaluate the performance of the measurement system by analysing pure biological substances with well-known fluorescence properties. We discuss differences and possible ambiguities of the measured fluorescence signals of nine pure substances representing biological fluorophores of different groups (amino acids, coenzymes, and biopolymers). To quantify the performance, we trained a supervised machine learning model and compare the particle class predicted by the model with the true particle class. We find that the instrument can distinguish very well between most fluorophores: for six of nine samples, the prediction accuracy is 99% or higher (others score above 93%).

In the second part of the study, we investigate the fluorescence and shape characteristics of twelve plant fragments from diverse sources: four tree leaves, five seed fibers, and two plant waxes. Respective biological materials were collected in the environment and mechanically fragmented in the laboratory to obtain particle sizes within the micrometer size range. We find that all samples exhibit sufficient fluorescence to be detected by the instrument. Compared to the pure fluorophores, the plant fragments have much less distinct fluorescence properties and show many overlapping features indicating that fluorescence is not caused by a single fluorophore but by a mixture of fluorophores. The prediction accuracy of the trained model ranges from 68% (seed fiber) to 99% (plant wax).

In addition, we analyse particle shape parameters determined from the holographic images. We highlight that, although all materials except two were mechanically fragmented the same way,

some characteristic shape features remain. In fact, including the holographic images in the machine learning model significantly improves the classification performance for some of the samples.

We conclude that the instrument is very well capable of identifying particles with distinct fluorescence features and clearly has the potential to also classify complex particles such as plant debris.

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Dynamics and Behaviour of Erysiphe Necator Atmospheric Spore Content in Vineyards in Central Spain

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Abstract

Erysiphe necator is a biotrophic fungus that parasitizes species of several genera of the *Vitaceae* family. In grapevine (*Vitis vinifera*), it causes the disease known as powdery mildew, which is very problematic in viticulture. For the control and prevention of this disease, it is necessary to monitor the variables that affect the environment-host-pathogen system. The aim of this study was to analyse the dynamics and behaviour of the atmospheric content of *E. necator* spores, as well as their relationship with different meteorological variables and the phenology of the grapevine.

The study was conducted during the year 2023 in a vineyard belonging to the land of the Designation of Origin Uclés, D.O Uclés, located in the west of Cuenca province (Castilla-La Mancha region, central Spain). Aerobiological sampling was carried out using a Hirst volumetric spore trap placed in a vineyard of the Syrah grape cultivar. Samples were prepared and analysed following the methodology established by the Spanish Aerobiology Network. Phenological sampling was carried out weekly on 20 vines, using an adaptation of the BBCH scale. Meteorological data were obtained from the Spanish Meteorological Agency (AEMET). The relationship between spore concentrations and meteorological variables was analysed using Spearman's non-parametric correlation test and Principal Component Analysis (PCA). Finally, an intradiurnal analysis of spore concentration was carried out.

A total of 3389 spores/m^3 of *E. necator* were obtained during the study period. The daily peak spore concentration occurred on 28 June with 121 spores/m^3. The highest daily concentrations (>50 spores/m^3) coincided with the flowering stage and the beginning of fruit development of the vines, while the lowest concentrations occurred during senescence. In periods such as spring and early autumn, where average temperatures range from 14 to 27°C, temperature influences the daily spore concentration in a significant and positive way, while an increase in relative humidity (RH) influences it negatively. However, during summer, with mean temperatures above 27°C and no rainfall, RH and dew point positively influence the daily spore concentration. Intradiurnal analysis showed that the highest spore concentrations occurred between 12:00 and 17:00.

Flowering and fruit development are very susceptible stages for *E. necator* infection. Sporulation development is strongly influenced by weather conditions, being impaired by very high temperatures, but favoured by warm temperatures, low RH and low rainfall, which increases vulnerability in vineyards in Mediterranean climate zones. Progress in the knowledge of the environment-host-pathogen system in these areas will help to decide the most appropriate times for the application of phytosanitary treatments.

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The Impact of Rainfall on Airborne Dynamics of Alternaria Spores and Its Major Allergen Alt a 1: a Preliminary Study

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Abstract

Alternaria spores are commonly found in the atmosphere and have been described as a significant source of allergens. Although up to 17 allergens have been characterized in this fungal genus, the glycoprotein Alt a 1 is the major allergen, reacting with over 90% of IgE serum in patients sensitized to *Alternaria*. Traditionally, the exposure to this allergen has been assessed by aerobiological spore counts, but this method does not always offer an accurate estimation

of airborne allergen load. Previous studies have shown that allergens can be release from spores into the air under specific weather conditions such as thunderstorms, posing a higher risk to the allergic population. However, there is a lack of knowledge regarding the factors that facilitate the release and persistence of allergens in the atmosphere, particularly those related to fungi. This study aims to determine the impact of rainfall on allergen release from the spores and to explain potential discrepancies between the airborne concentration of allergen and *Alternaria* spores. The study was conducted in León (Spain) over a one-month sampling period (May 2017). This month was selected due to the particular meteorological features, being warmer than usual and keeping the normal precipitation values for this period. Airborne spores were sampled using a Hirst-type volumetric sampler, following the methodology proposed by CEN legislation EN 16868:2019. Daily spore concentration was measured following the methodology proposed by Galán et al. (2021). Furthermore, the allergenic fraction was collected by a cyclone low-volume sampler and the major allergen Alt a 1 was quantified by ELISA on daily samples. Additionally, rainfall variables were measured using a disdrometer. The non-parametric Spearman rank correlation was applied to analyze the relationships between rainfall parameters and the airborne concentration of spores and Alt a 1. The results reveal a noticeable delay between the presence of *Alternaria* spores and Alt a 1 due to rainfall events. Precipitation days with raindrop sizes ranged between 0.125 and 4 mm, distributed throughout the day, mainly at midday, had null or very low concentration values of *Alternaria* spores. However, the allergen concentration usually increases following a couple of days of rain, typically associated with light precipitation events (raindrop sizes < 4 mm and low intensity), coinciding with maximum temperatures around 20 °C and an increment in minimum temperatures. On the other hand, punctual rainfall events with raindrop sizes higher than 4 mm could favor the release of *Alternaria* spores. This preliminary study highlights the importance of analyzing different rainfall parameters to better understand the discrepancies between aerobiological counts and allergen concentration.

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Cupressaceae Airborne Pollen and Interaction With Atmospheric Pollutants

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Abstract

Cupressaceae pollen is a relevant cause of respiratory allergies in the Mediterranean, with increasing prevalence. Our study aimed to determine the pattern of Cupressaceae pollen in the atmosphere of Porto, correlate it with meteorological parameters and evaluate the effects of in vitro exposure to atmospheric pollutants in the allergenicity of its pollen.

Airborne pollen was monitored from 2003 to 2023 using a 7-day Hirst-type volumetric spore sampler. Spearman's rank correlation was used to relate airborne pollen concentration and meteorological factors. *C. lusitanica* Mill. pollen was exposed to NO_2, O_3 and both in a controlled environmental chamber at a concentration equal to the limit value for human health protection (2008/50/EC). The effects in pollen allergenicity were analysed through Western blot, using sera of subjects sensitised to Cupressaceae pollen and through ELISA using Cup a 1 antibody. IgE binding was expressed as optic density values of the reactive bands, and a ratio between the pollutant exposed and the blank samples was determined for each band. A paired t-test was applied per IgE reactive band, combining all sera information. An ANOVA was done on ELISA results to ascertain the significance of Cup a 1 content changes due to pollutant exposure.

Cupressaceae main pollen season occurs from January to March, being February the month with the highest concentrations. Airborne pollen was positively correlated with air temperature and negatively with rainfall and relative humidity. O_3 and O_3/NO_2 mixture induced an overall increase in the total IgE reactivity of the patient sera. However, different trends could be observed in analysing band-specific IgE binding depending on the sera and reactive band. A quantitative significant increase of Cup a 1 content was observed in the samples exposed to the pollutant gases.

Our study showed the presence of Cupressaceae pollen in the air for nearly 3 months, which can be significantly affected by meteorological conditions. Also, it was observed that pollen allergens can be affected by air pollution, but the impact on allergy exacerbation might vary depending on the type of pollutant and the patient's sensitisation profile.

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Bi-hourly Airborne Ailanthus Altissima ([Mill.] Swingle) Pollen Concentrations in the City Center of Berlin, Germany, at Rooftop and Street Level

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Abstract

Introduction

The Tree-of-Heaven (*Ailanthus altissima*, AA) is a fast-growing, dioecious tree species native to China and Korea. In 2019, it was classified as an invasive alien species of Union concern in the European Union. Sensitization and allergic reactions to AA pollen have been reported in patients from numerous countries worldwide. And yet, monitoring of airborne AA pollen is still very limited. This study investigates the intra-diurnal and spatial distribution of AA pollen at two different heights in Berlin, Germany, considering local AA vegetation and meteorological data on wind conditions.

Methods

Air was sampled using Hirst-type volumetric spore traps at a central site in Berlin, at both rooftop (CharR; 23 m above ground) and street level (CharS; 3 m above ground), 80 m apart. Sampling spanned from May 25 to August 1 (defined as annual pollen integral [APIn]) for the years 2012–2021 at CharR and 2015–2017 and 2020–2021 at CharS. Bi-hourly and daily pollen concentrations were obtained by light microscopy pollen analysis of daily samples. Pollen season (PS) parameters (95% PS definition) were calculated using R (package AeRobiology). Information on the distribution of AA trees within 1 km of the site was gathered from the Berlin Tree Register and field campaigns in 2019 and 2020. Hourly wind data from the Tempelhof weather station of the German Weather Service were combined with bi-hourly pollen concentrations.

Results

The average APIn for all available years was 1,311 pollen grains/m^3 (p/m^3) at CharS and 145 p/m^3 at CharR. The average APIn ratio between CharR and CharS was 1 to 10.0, ranging from 1 to 4.9 to 1 to 13.1. Pollen season for CharR started earliest on May 31, 2018, and ended latest on July 11, 2020, while for CharS it started earliest on June 5, 2016, and ended latest on July 7, 2015. The average PS duration was 21.2 (CharS) and 22 days (CharR). Peak daily concentrations reached 53 p/m^3 at CharR on June 12, 2017, and 344 p/m^3 at CharS on June 21, 2021. Both stations exhibited similar intra-diurnal patterns with highest pollen concentrations observed between 12:00–14:00 at CharR and between 14:00–16:00 at CharS, and lowest concentrations at night between 02:00–04:00 at both traps. Westerly winds and wind speeds exceeding 5 m/s coincided with higher pollen concentrations at CharR, while at CharS the highest values were reached with winds from the west and northwest, corresponding to the four nearest male AA trees (17 m to 120 m from the trap).

Conclusions

This multi-year study identified clear intra-diurnal patterns in AA pollen flight, highlighting midday/afternoon as peak exposure times in central Berlin. Sampling height and the presence of nearby trees significantly influenced the occurrence of AA pollen. Given the expected spread of AA and the allergenicity of its pollen, it is recommended to conduct further research on the influence of AA vegetation and meteorological conditions on AA pollen dispersal and to include AA pollen in routine pollen monitoring, especially in areas colonized by this species.

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Using Satellite Imagery to Assess the Effects of Urban Heat Island on the Phenology of Betula Pendula Roth.

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Abstract

To properly understand the timing of phenological events, it is important to consider the indirect impacts of human activity on plants. Urban heat island (UHI) is causing shifts in the natural calendar of plant phenological events. One of the major consequences of these changes is a shift in the time of pollination. The main objective of this study is to explore the impact of the urban-rural gradient on the temporal patterns of phenological stages using satellite imagery data. While previous studies utilizing satellite data characterize plant phenology collectively, without differentiation between individual stand types and dominant species, this approach oversimplifies the complex interactions, especially considering distinct reactions among tree

species to the UHI influence. To address this research gap, this study concentrates on stands of a single species - silver birch (*Betula pendula* Roth.), providing a species-specific comprehension of UHI impact. The chosen species of birch tree is recorded to be one of the main aeroallergens in Poland and Europe, causing significant implications for public health and the environment. The widespread presence of the chosen birch tree species in both urban and rural areas provides a unique opportunity to conduct a complex study, allowing for a better understanding of its phenological patterns and the potential impacts of the urban-rural gradient on them. This research focuses on six different study areas around selected larger cities in Poland. Chosen areas cover various parts of the country, including central, northern, western, and southern regions. By categorizing the tree stands based on their proximity to urban areas, it is explored how the distance from the city influences the main phenological stages, specifically the start of the growing season (SOS), the peak of the growing season (POS), and end of the growing season (EOS). Employing the satellite-derived Normalized Difference Vegetation Index data, the Elmore model is utilized to curve-fit the phenological events and analyze their temporal dynamics. Vegetation indices provide information about leaf development but there is evidence that the date of leaf emergence and the onset of flowering, indicating the start of the pollen season, is similar. The dataset comprises information from more than 500 tree stands located in a 0-30 km range from urban areas. Results reveal notable variability in the start of the season dates (earliest SOS - March 17th, latest SOS - April 14th), emphasizing higher fluctuations compared to the more constant end of the season dates. Fluctuations in the peak of the season dates suggest a potential dependence on the start of the season, underscoring the significance of early-season events in influencing the overall phenological season. Analysis of mean land surface temperature maps confirms the existence of a surface UHI effect in overlapping urban areas. Despite challenges, this study contributes as a pioneering work focusing on birch tree phenology within the context of surface UHI analysis. The research provides valuable insights into the effects of urbanization and climate change on plant phenology, showcasing the applicability of satellite imagery in studying these phenomena.

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Effects of Climate Change on Herbaceous Pollen Trend

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Abstract

Background

Different herbaceous species release pollen dangerous for the human health. This study aimed to: assess the pollen load and pollen seasonal parameters trends; assess the possible advance or delay and extension of the pollen season and whether there was an association between these variations and climate change; define the season of that kind of pollen.

Methods

Pollen were sampled by Hirst trap located in Legnano (North-West Milan area, Italy). The pollen considered were: *Artemisia, Cannabaceae, Chenopodiaceae/Amarantaceae, Graminaceae* and *Urticaceae*. Observation period ranged between 1995 and 2022. Since *Ambrosia* pollen is the most important allergenic particle in that area, it was considered in another study. The start of the season was chosen as the day when the cumulated daily counts reached 2.5% – and the end as the day when the 97.5% – of the Seasonal Pollen Integral (95% period). We analyzed pollen trends using linear regression considering several dependent variables: the start date, peak emission date, peak emission value, end date, duration, Annual Pollen Integral, number of days with a concentration value greater than 1 within a season as a function of meteorological features such as the daily average of standard temperature (K) at 2 m above the surface, i.e. the "nose-level" temperature, the relative humidity (%) at a height of 2 m above the surface, convective precipitation (mm) (water level accumulated during last one hour), wind speed (m/s), and surface short-wave solar downwards radiation (J/m^2).

Results

Graminaceae and *Urticaceae* exhibit a statistically significant negative correlation between the start of the pollen season and the temperature. This suggests that a rise in temperature leads to an earlier onset of the season for these species. *Urticaceae* are particularly sensitive to weather conditions, with a statistically significant positive correlation between the duration of the pollen season and the temperature: warmer temperatures tend to prolong the *Urticaceae* pollen season. An increase in solar radiation is associated with an extended pollen season for *Urticaceae* and a rise in temperature is directly linked to an increased number of days with pollen concentrations exceeding 1 p/m^3. Regarding Artemisia, increased precipitation and relative humidity are associated with an earlier end to the pollen season, while higher solar radiation tends to prolong it. Higher wind speed significantly prolongs the pollen season, while relative higher humidity has the opposite effect.

Conclusions

In conclusion, *Urticaceae* pollen appears to be the most sensitive herbaceous pollen to weather conditions, as evidenced by the correlation between its seasonal parameters and various climate factors. *Graminaceae* and *Artemisia* pollen also show sensitivity to climate variables, although to a lesser extent.

These findings are significant as *Urticaceae* and *Graminaceae* pollen are major contributors to allergic disorders. Therefore, changes in the prevalence of these allergies could be associated with climate change.

Further research is warranted to explore the potential correlation between climate change, trends in pollen seasonal parameters and variations in the prevalence of respiratory allergic disorders. Understanding these relationships can support public health strategies and adaptation measures to mitigate the impacts of climate change on allergic diseases.

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Relationship Between the Annual Fluctuation in Pollen Distribution and Meteorological Conditions

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Abstract

The objective of the study is to assess the correlations between meteorological conditions and airborne pollen concentration. Daily data on pollen concentration were obtained from monitoring stations located in three distinct sites across Lithuania: Šiauliai, Vilnius, and Klaipėda. These stations are situated in different regions of the country. The study focused on analyzing the pollen counts of *Alnus* and *Betula*. The investigation period spanned from 2005 to 2019. The information about air temperature and precipitation was obtained from meteorological stations located in these areas.

The pollen seasons start and end times, seasons duration, annual maximum daily pollen concentration (grains/m^3) and its date were analysed. The Annual pollen index integral (APIn) and the Seasonal pollen index integral (SPIn) were calculated. It was also determined what part of the annual pollen count was recorded during 5, 10, 15, 20, 25, and 30 consecutive days with the highest pollen concentration. All these indicators were correlated with meteorological conditions. The relationship between the analysed indicators was determined using the Spearman correlation coefficient. Relationships were considered statistically significant when $p < 0,05$.

On average in the territory of Lithuania the *Alnus* pollen season begins in the second decade of March and ends in mid-April. The average duration of the season varies from 29 days in Vilnius to 35 days in Šiauliai. During the pollen season, approximately 96-97% of the annual pollen count is typically recorded. The maximum daily concentration is observed at the end of March, with an average of 22-27% of the total annual pollen quantity recorded. During the five consecutive days featuring the peak pollen concentration, it commonly accounts for 55% of the total annual pollen count. On average, the corresponding percentages increase to 73% over 10 days, 83% over 15 days, 89% over 20 days, 92% over 25 days, and 96% over 30 days, respectively. Evaluating the relationship between *Alnus* pollen season and air temperature, we can affirm that air

temperature influences the onset of the season (r – -0,83- -0,86). Correlation between pollen season and precipitation is weak and statistically insignificant.

The *Betula* pollen season typically starts in late April and ends in mid-May, with average season duration of 31-32 days. Approximately 94-97% of the total annual pollen count occurs during the *Betula* pollen season, with its peak concentration manifesting during a five-day span accounting for 51-55% of annual pollen count. The correlation between *Betula* pollen season dates and monthly air temperature remains weak and becomes statistically insignificant when considering precipitation.

It is noteworthy that a significant proportion, exceeding 96%, of pollen is observed to fall within a 30-day continuous timeframe. While air temperature plays a role in influencing the start of the *Alnus* pollen season, no such strong relationship is observed for *Betula*. Moreover, the impact of precipitation on pollen distribution is deemed statistically insignificant.

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Two years of using SwisensPoleno Mars in Iceland

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Abstract

There's a rising need for technology that gives detailed and efficient data in real-time without needing an operator. Our understanding of automatic pollen monitoring suggests that it performs optimally in regions with minimal variability in pollen species. By definition, Iceland emerges as an ideal location to implement automatic systems. Iceland, the second-largest island in Europe, possesses distinct biogeographical traits owing to its strategic location bridging Europe and North America and straddling the Arctic and Boreal zones. Its climatic dynamics are shaped by diverse air masses and ocean currents, leading to volatile weather patterns. The vascular flora of Iceland, emerging postglacially, comprises 426 indigenous taxa. Unlike its counterparts in Greenland and Scandinavia, Icelandic flora showcases a unique Atlantic-European component.

Pollen levels in Iceland are monitored from April/May to September annually in two locations: Reykjavík (since 1988) and Akureyri (since 1998). The daily average pollen counts are recorded using the volumetric method employing the Burkard Seven-Day Volumetric Pollen Trap. Additionally, since July 28, 2022, pollen monitoring in Akureyri has been augmented by the Swisens Poleno Mars automatic pollen monitoring system, covering two grass pollen seasons and one birch pollen season. Comparative analysis between the two monitoring

systems is conducted utilizing basic statistical techniques such as the Pearson correlation coefficient.

Long-term pollen monitoring data highlights a limited diversity of airborne pollen in Iceland's aeroplankton, with Betula spp. and Poaceae emerging as the predominant pollen allergens. Iceland's pollen seasons typically experience a delayed commencement compared to continental Europe. Although daily grass pollen concentrations from both instruments exhibit similar trends, a detailed examination reveals instances of false positive results. Following adjustments to the grass pollen identification methodology, satisfactory outcomes are achieved. Birch pollen, alongside grass pollen, constitutes another significant pollen type in Iceland. However, in 2023, testing with the Poleno Mars system uncovered numerous false positive occurrences, underscoring the necessity for further refinement in Betula pollen classification.

The presence of dust particles in the air constitutes another challenge. It is evident that a significant number of dust particles are being misclassified as pollen although they substantially differ morphologically from pollen grains. It should be noted that Icelandic soils are uniquely special on a global scale. Most of Icelandic soils form in volcanic parent materials. That's why Iceland is among the most active dust source areas in the world. Functioning as an active desert, it experiences dust events annually, with the dust capable of traveling long distances.Thus, at least in the case of Iceland, resolving the dust issue requires still improvements.

It can be concluded that the implementation of automated pollen monitoring systems necessitates an adjustment period (spanning more than one pollen season) to tailor the system to local environmental conditions. Fine-tuning each pollen taxon individually is imperative. The deployment of Swisens Poleno Mars in Iceland has yielded invaluable insights into pollen seasons, particularly aiding in grass pollen measurements. Nonetheless, continuous efforts are indispensable to enhance Betula pollen classification.

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Airborne Pollen Trends in Tétouan (NW of Morocco)

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Abstract

Trends of the airborne annual pollen integral (APIn) and pollen season of principal woody and herbaceous plants in Tétouan were analysed over a 10-year monitoring period (2008–2017). Pollen was continuously sampled by means of a 7-day recording volumetric pollen trap by Burkard. Pollen trends were analysed by using Mann–Kendall tests and Sen's slope.

Aerobiological data were correlated with temperature and rainfall. A significant decreasing trend in annual minimum temperature was revealed together with significant decreasing trends in the APIn observed for Cupressaceae, *Cannabis*, *Parietaria*, *Pinus* and *Quercus*, this being highly significant for Cupressaceae and *Pinus*. On the contrary, the seasonal intensity of *Mercurialis*, *Morus* and *Olea* showed nonsignificant trends. Besides this, 77% of the studied pollen types showed a tendency to decreasing the peaks value, these trends being significant for Cupressaceae (-204.67 pollen/ m^3 per year) and *Pinus* (-14.33 pollen/ m3 per year). The end of the *Quercus* pollen season showed a marked tendency to occur earlier across the years (-4.5 days/year) and the start day of *Cannabis*, Cupressaceae, *Pinus* and *Poaceae* to occur later (+ 7.13, 2.33, 1.67 and 2.5 day/year, respectively),respectively), shortening the duration of the respective pollen seasons but not with a significant trend. Regarding the association between the pollen season intensity and meteorological parameters, six pollen types showed at least one statistically significant coefficient correlation. The decreasing and significant trend in the intensity of the APIn diminishes also the exposure to airborne pollen for allergic sufferers, having implications in the field of public health. Decreasing trends in annual minimum temperature and the general lack of significant trends and correlation coefficients between the parameters of the pollen season of different pollen types and monthly mean temperatures and rainfall suggest that interannual variability in the data is due to human interventions, deforestation, fires and the opposite response of some species to warming in Fall/Winter and Spring, and this could be the reasons for the observed behaviour in the pollen season.

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Oak and Beech Pollen Floating in the Air – Tendency or Variability of the Intensity and Course of Pollen Seasons?

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Abstract

It is known that air temperature is the environmental factor that has the greatest impact on plant vegetation, including the course and intensity of pollen seasons. Due to the increase in global temperature, plants react by accelerating the dates of spring phenophases, including the beginning of pollination. Temperature is also the most important parameter in models describing the dynamics of phenomena occurring in the biosphere. The aim of this study was to check whether oak and beech pollen has appeared in the air earlier in the last 27 years and whether the concentrations have changed. An interesting problem is to detect a treshold date in the course of pollen seasons, after which the course of seasons becomes similar and to check whether these dates depend on the weather. Aerobiological research has been conducted

since 1997 using the volumetric method, and 2024 will be the test year. The intensity, dates and patterns of pollen seasons were compared. Beech and oak are characterized by high variability in the intensity of the seasons, with a clearly marked rhythm in the case of beech, which produces huge amounts of pollen once every four years. In the case of oak, the previously recorded 4-year cycle has become unstable, perhaps as a result of more frequent temperature extremes. High concentrations of oak and beech pollen were often recorded in the same years (e.g. 2003, 2006, 2020, 2022). The synchronization of the abundant pollen production of these tree species is strong (r=0.5). No statistically significant trends were found in the case of season dates for both taxa. If the oak season started later, its length and intensity decreased. In the case of beech, a negative relationship was found between the beginning of the season and its length. Predicting years of abundant pollen is difficult, but it seems that in the case of beech, temperatures during the period of inflorescence formation are crucial, and the key date may be the solstice on June 21, after which the days shorten. In the case of oak, the most important are thermal conditions in the pollen year. The obtained results confirm the 'wind pollination hypothesis', which is one of the explanations for the masting phenomenon.

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Botanical Composition of Pollen Collected by Apis Mellifera L. From Uludağ and Its Surroundings, Northwest- Türkiye

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Abstract

In recent years, consumers' interest in bee pollen as a natural bee product, whose value has been recognised more and more, has been increasing due to its nutritive and energizing properties as well as its bioactive substances beneficial to health and therapeutic properties. The composition of bee pollen varies depending on the species. Nutritional value and protein content varies depending on the botanical source. Botanical source variation varies according to climatic conditions and geographical location. The colour of bee pollen varies mostly depending on the botanical source. For bee pollen to find more applications, its characterisation based on floral and geographical origin is required. This study aims to assess the plant

preferences of bees in and around Uludağ, which is a region known for its rich plant diversity. The study also aims to create a pollen calendar that can assist in determining the variations in bee pollen from geographically specific botanical sources. This information can be useful for bee nutrition and hive management. In this study, the botanical origins of pollen samples obtained from beekeeping activities in different regions of Uludağ were determined using palynological methods. Bee pollen was collected from traps in various regions of Uludağ (Cumalıkızık, Çöreler, Kozluören, Oylat, Tahtaköprü, Soğukpınar) during the months between April and August in 2021, with fifteen-day intervals for four months. The samples were analyzed at Bursa Uludag University Aerobiology Research Laboratory. Analyses were carried out with samples containing 10 g of pollen. Pollen granules were placed on blotting paper and classified according to their color using the color scale of Kirk (1994) under a stereo microscope. Following the method of Wodehouse (1935), one preparation was prepared when the bee pollen contained one color, and separate preparations were prepared for each color when it contained two or more two colors. The preparations were photographed using a microscope at x100 magnifications and identified. Various pollen databases and publications were used to identify pollen at the family, genus, and species levels through comparison. In this study, 46 families were identified in the collected samples. The most common family was Rosaceae (13.30%). It was followed by the following families with a frequency above 1%, respectively; Cistaceae (11.52%), Brassicaceae (8.07%), Fabaceae (7.68%), Asteraceae (7.25%), Fagaceae (6.93%), Ranunculaceae (5.99%), Apiaceae (3.73%), Poaceae (3.69%), Plantaginaceae (2.72%), Polygonaceae (2.33%), Scrophulariaceae (2.27%), Lamiaceae (2.17%), Salicaceae (2.05%), Onagraceae (1.89%), Moraceae (1.57%), Solanaceae (1.57%), Caprifoliaceae (1.54%), Resedaceae (1.43%), Euphorbiaceae (1.29%), Clusiaceae (1.17%). The highest diversity occurred in the second period of July (16-31) and 29 different families were identified. In light of the data obtained, the variations of botanical resources providing bee nutrition between April and August in Uludağ and its surroundings were determined and a pollen calendar was prepared. This study highlights the significance of plant characterization and botanical diversity in Uludağ and its surroundings for beekeeping. This study was supported by the Bursa Uludağ University Scientific Research Council (Project no: FBO-2020-188).

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The Effect of Fungicides on the Production of the Major Allergen of Alternaria Alternata Spores (Alt a 1) - Preliminary Study

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Abstract

Introduction: The spores of Alternaria spp. are highly important inhalant allergens with Alt a 1 as the main allergenic protein, which is a glycoprotein associated with the plant infection process, including interactions with plant defence proteins (PR5) and promotion of fungal penetration into the plant. The most effective protection against pathogenic fungi is through fungicides. However, the use of fungicide may have adverse effects, e.g. by the increase production of mycotoxins. Hypothetically, similar phenomenon may occur with relation to allergenic proteins released by spores. Therefore, the aim of the study was to evaluate the impact of fungistatic agents on the Alt a 1 level in A. alternata spores.

Methods: The identification of fungal isolate was confirmed by molecular methods, i.e. the amplification of the ITS fragment of rDNA and sequencing of the obtained product. The effect of six fungicides differing in their active substance in three concentrations: 0.01%, 0.1% and 1.0% have been used in the experiments were used towards six isolates of Alternaria alternata (KPPJ, BPR-1893, KZF32, KZF35, KZF42, KZF47) The minimum inhibitory concentration (MIC) and minimum fungicidal concentrations (MFC) methods, as well as the plate cultures, were applied to determine the degree of inhibition of fungal growth by the tested agents. The concentration of the Alt a 1 in spores was determined using the enzyme-linked immunosorbent assay (ELISA). Spores extracted from the mycelium were counted using a hemocytometer.

Results: The sensitivity of A. alternata isolates to fungicides was differentiated therefore three isolates (KPPJ, BPR-1893 KZF32) and three (3,4,6) fungicides with specific MIC and MFC values were used for further analyses. The highest average increase in allergenicity was observed for the KZF isolate, while the lowest for KPPJ. The highest mean spore number was observed for the BPR-1893 isolate at 3 (0.01% concentration) and 4 (0.1% concentration). The results of the conducted research showed that the impact of individual fungicides is varied and depends not only on the type of active substance, but also on the sensitivity of a given isolate. Depending on the fungicide and its concentration, differences were observed in both the number of spores produced and the content of the Alt a 1 allergen.

Conclusions: It is expected that the results of the experiments will be an explanation of the effect of fungicidal substances on the Alt a 1 content in fungal spores, which may significantly expand the knowledge about the influence of various factors on the allergenicity of fungi.

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Applying Machine Learning and Statistical Models to Low-Cost Aerosol Sensors for Anomaly Detection

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Abstract

Currently, the U.S. is investing in Biological Detection for the 21st Century (BD21) that performs monitoring for airborne releases of biological agents using anomaly detection sensors and data analytics. BD21 sensors collect particle size, concentration, and fluorescence data, which are analyzed using data fusion techniques and machine learning algorithms to identify biological threats from background particles. Despite significant investments, the Government Accountability Office has stated, "Biological aerosol sensors that monitor the air are to provide data on biological material in the environment, but common environmental material such as pollen, soil, and diesel exhaust can emit a signal in the same range as a biological threat agent, thereby increasing false alarm rates… false alarms produced by biological sensor technologies could be reduced by using an anomaly detection algorithm in addition to the sensor" (Government Accountability Office, https://www.gao.gov/assets/720/714434.pdf).

Low-cost aerosol sensors (that measure particle size and concentration) deployed by individuals in populated areas have recently gained popularity. PurpleAir (https://map.purpleair.com/) has a real-time air quality map with sensor data on particle size distribution and concentration, as well as an application programming interface that facilitates data collection. We hypothesize that the greater concentration of PurpleAir sensors may enable development of advanced anomaly detection algorithms that overcome the current challenges with background aerosols. To test our hypothesis, we have identified and developed various statistical and machine learning approaches to identify anomalies within PurpleAir datasets.

We have ingested a year of PurpleAir data from outdoor sensors across the Washington, D.C., metro region. Data consist of particle concentrations across 0.3 μM, 0.5 μM, 1.0 μM, 2.5 μM, 5 μM, and 10 μM size ranges, as well as temperature and relative humidity. Additionally, we have obtained the meteorological data on wind speed and direction for the relevant aerosol sensor measurements.

We've engineered an array of statistical and machine learning methodologies for anomaly detection within temporally aggregated data. Our approach uncovers irregularities and inconsistencies across geographic data by analyzing data patterns, time trends, relational structures, and sequential deviations. We are finalizing development of synthetic data with various meteorological conditions to simulate anomalous events by replicating the particle size distribution and concentration within a biological plume and overlaying the data on existing PurpleAir datasets with matching meteorological conditions.

We will present the different algorithm approaches that we have developed with the benefits and disadvantages of each approach in identifying the synthetic anomalous events that represent a biological attack. This includes the sensitivity, specificity, timeliness, and computational requirements for each approach. We have also identified requirements for sensor density and placement for optimal detection as additional sensors increase sensitivity and specificity but incur an additional cost and require more resources.

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Comparing Grass and Birch Pollen Seasons (1998-2023): Akureyri, Iceland (65 N) vs. Krakow, Poland (50N) – Exploring Weather Contrasts and Climate Change Implications

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Abstract

In Europe, grasses (Poaceae family) are the major cause of allergic reactions due to pollen, given their wide geographical range. Among trees, the most allergenic pollen is produced by birch in northern, central, and eastern Europe. In our study, we compared the pollen seasons of birch and grass in two different locations in Europe: in Akureyri, North Iceland, and in Central Europe in Krakow, Poland.

Akureyri (65 N), the second-largest urban area in Iceland, is nestled in the northeast of Iceland, surrounded by towering mountains. Situated along one of the longest fjords in the country, Akureyri enjoys a mild climate despite its proximity to the Arctic Circle, with summer temperatures reaching up to 25˚C and average winter temperatures hovering around 0˚C.

Krakow (50N), Poland's second-largest city, sits on the banks of the Vistula River within the Lesser Poland Voivodeship. Positioned at the convergence of several physiographic regions, offers diverse landscapes. Krakow experiences a temperate, warm, and transitional climate, serving as a boundary between maritime and continental climates.

For our data we used air samples collected from 1998 to 2023, using the volumetric method. We examined a long-term series of pollen and meteorological data. First, we made the basic statistics to describe the pollen season characteristics (start and end day, length of the pollen

season, seasonal pollen index SPI, peak day, and value). Then linear regression was used to determine trends in pollen season characteristics, while Spearman's correlation analysis was used to identify the relationships between the characteristics of pollen seasons and meteorological data.

Both birch and grass pollen seasons appear in Krakow around one month before Akureyri. Although the amount and dynamics of grass pollen seasons are very similar in both locations, the dynamics and amount of birch pollen are dramatically different, with much higher levels in Krakow.

Regarding the birch pollen season, in Akureyri, we observed a statistically significant negative trend in the end date of the pollen season, resulting in a statistically significant shortening of the season's length. In Krakow, the SPI (Seasonal Pollen Index) increases significantly, along with a notably higher value for the birch pollen peak.

In the case of grass pollen, significant increases in SPI were observed in both towns.

We observed a statistically significant increase in temperature and precipitation in Akureyri, as well as a temperature increase in Krakow. However, there seems to be a lack of correlation or weak correlation between pollen season characteristics and meteorological data, especially in Akureyri. It appears that, although the general trend in climate change is more pronounced in Akureyri than in Krakow, the changes in pollen season characteristics are less apparent. This could be due to the relatively short pollen seasons in Iceland and the fact that weather conditions, although increasing over the years (in particular temperature), are still relatively less conducive for plant growth.

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Study on the Temporal of Airborne Pollen Grains in the City of Bragança (Portugal).

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Abstract

This study aimed at providing initial insights into the temporal variation of airborne pollen concentration in the city of Bragança, Portugal, also exploring its geographical dependence, as different climatic regions exhibit distinct vegetation patterns. This is of great relevance for understanding and preventing allergic diseases in a region where allergic population reaches noneworthy figures.

The city of Bragança is located in the Trás-os-Montes region, in the north-east of Portugal. It has an average annual rainfall of 632mm, distributed irregularly throughout the year, and an average annual temperature of 11.5ºC. Trees, shrubs and ornamental and non-ornamental herbaceous species can be found around the sampling site, several of which produce allergenic pollen. Also, major influence from perennial and annual crops is registered. To analyze the airborne pollen concentration, a 7-day Hirst-type volumetric spore sampler (Lanzoni VPPS 2000), was used. Samples were collected from January to July, in three years (2021 to 2023). This period was selected because it corresponds to the season with higher concentrations and greater influence on the airborne pollen concentrations.

During the study period, 47 different pollen types were identified, 29 of which came from trees, 16 from herbaceous plants. The distribution of the pollen concentration in the air showed several days with high values due to the diversity of taxa present in the atmosphere and their different flowering seasons. In the winter months 26 different types of pollen were observed, the most predominant being Cupressaceae. It was also observed the presence of *Alnus, Corylus, Fraxinus* and Poaceae pollen. Urticaceae was also identified, but at lower levels.

In spring 43 different types of pollen were observed. March and April were the months with higher diversity, the most frequent being *Quercus* pollen together with *Platanus, Populus, Salix, Pinus,* Rosaceae, Ericaceae and Urticaceae. In April, Brassicaceae, Fabaceae, *Juglans, Olea, Plantago*, and *Rumex* pollen were registered. In May, we must emphasize the large quantity of *Quercus* pollen, which stood out from the rest, but also, in smaller quantities, Poaceae, *Olea*, Ericaceae and *Rumex* pollen.

In June and July, 36 pollen types were identified, the most prevalent being *Castanea* and Poaceae, but *Olea, Quercus*, Urticaceae and *Plantago* pollen were found in the urban atmosphere of Bragança.

The spring months, as they contain higher concentrations of pollen in the air, and especially, a greater diversity of pollen, are the most problematic for allergy sufferers.

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Low-Cost Detection of Pollen Using Machine Learning Through a Forest Canopy

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Abstract

Conventional techniques for monitoring pollen currently have significant limitations in terms of labour, cost and the spatiotemporal resolution that can be achieved. Pollen monitoring networks across the world are generally sparse and are not able to fully represent the detailed characteristics of pollen release and its spatial distribution within the biosphere. There are few studies that observe airborne pollen concentrations on a local scale, and even fewer that do so in ecologically-rich rural areas and close to emitting sources. Better understanding of these would be relevant to occupational risk assessments for public health, as well as ecology, biodiversity, and climate.

In this presentation, we will firstly introduce the recently developed method for low-cost detection of pollen bioaerosols using optical particle counters (OPCs) and machine learning to process the data (Mills et al., 2023a; Mills et al., 2023b).

Subsequently, we will present a specific study which applies the previously trained machine learning models on data collected from low-cost sensors in a mature forest environment to estimate local pollen concentrations. We use the OPC sensors to characterise both particulate matter and pollen concentrations, first during a colocation period, then during a period when sensors are placed at different heights through the canopy, on a 40 m observational tower within the forest.

We observed that the pollen concentrations derived from this method follow expected diurnal trends and interactions with meteorological variables. While PM_{10} (particle matter with diameter less than or equal to 10 mm) concentrations were suppressed within the canopy height, Quercus pollen concentrations appeared greatest at this height (20-30 m). Quercus pollen concentrations were lowest at the greatest height that is above the canopy (40 m), which is congruent with previous studies of background pollen in urban environments. The attenuation of pollen concentrations as sources are depleted is also observed across the season and at different heights, with some evidence that the pollen concentrations persist later at the lowest level beneath the canopy (10 m) where catkins mature latest.

We assess the efficacy and usefulness of this method for monitoring pollen and discuss directions for future development. The results so far demonstrate the potential of this method to collect a wealth of information on pollen concentrations, in unique locations and at spatial resolutions that were not previously possible.

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Are There Allergy-friendly Parks in Poznań, Poland?

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Abstract

***Key words*:** pollen allergy, Poznan, Poland, urban green spaces, *Platanus*, IUGZA

Background: Greenery is a crucial element in urban spaces. However, poorly planned and managed green spaces can have a negative impact on health, primarily in the context of allergic diseases caused by pollen grains. Instead of serving a health-promoting function, green areas may become unpleasant for a significant number of inhabitants. Therefore, it is essential to determine the city areas where the exposure to allergenic pollen is potentially the lowest (or the highest). Consequently, the main objective of this study was to evaluate the allergenic potential of green urban spaces in the city center of Poznań (Western Poland), and eventually to select the most allergy-friendly areas.

Methods: The inventory was conducted in 10 parks and the green spaces covering all the streets and alleys of Poznań city center (approximately 2,7 km^2~270ha). The detailed inventory of parks includes such aspects as: tree location, species identification, physical characteristics, e.g. trunk diameter, height, enumeration of tree species. Determination of the allergenic potential of certain species (scale from 0 to 5) has been assesed by reviewing clinical and aerobiological literature. Tree distribution was mapped using dedicated GIS software's (e.g. QGIS, ArcGIS and R). IUGZA index (Cariñanos et al. 2019), supported with airborne LiDAR data, was used to evaluate the allergenic potential of selected urban green spaces.

Results: A total of 5246 trees from 83 species were documented, representing 50 genera and 29 botanical families. The most abundant genus was *Acer*, followed by *Tilia* and *Aesculus*. Among 83 species recorded, there were 46 species with known association to pollen allergy, representing such genera as *Platanus, Betula, Fraxinus, Carpinus, Juniperus, Taxus,*

Cupressus, and *Corylus*. In total, they accounted for 83% of all selected trees. 32.2% of trees possess allergenic potential at levels 3, 4, or 5. The most allergenic trees – level 5 (e.g. *Platanus, Betula, Carpinus*) accounted for 10.7% of all trees. In general, selected urban green spaces differed markedly in their allergenic potential (expressed by IUGZA index).

Conclusions: It is evident that the city center of Poznan has a high number of allergenic species, particularly due to the intensive planting of *Platanus*. Fortunately, there are also places with a much lower allergenic potential, characterized by dominant entomophilous tree genera such as *Acer, Robinia*, and *Prunus*, providing some relief for individuals suffering from pollen allergies. However, since pollen can be transported from one place to another by air currents, additional tasks, such as local pollen monitoring using portable volumetric traps, are currently being conducted to obtain precise information on pollen concentrations in specific urban green areas.

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The First European Airborne Pollen Reanalysis for Major Allergenic Trees: Alder, Birch, and Olive

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Abstract

Background

The prevalence of allergic rhinitis in Europe has been growing during the last several decades. Global change components such as climate change and land-use transformation are directly affecting the main factors driving the airborne pollen levels released by anemophilous plants. A retrospective long-term assessment of pollen abundance in the air is therefore useful for epidemiological studies, to track the biodiversity and ecosystem evolution, and may also help to estimate the species migration tendencies. The existing studies on the pollen long-term changes are usually limited to the individual station or specific regions. In the present reanalysis the combination of in-situ measurements and modern modelling tools are used to reconstruct the year-to-year variations of alder (*Alnus*), birch (*Betula*), and olive (*Olea*) pollen levels across Europe from the 1980s until 2022.

Methods

The SILAM model (System for Integrated modeLling of Atmospheric coMposition, http://silam.fmi.fi) was used as the main working tool for computation of the reanalysis. This off-line chemistry transport model contains the pollen specific phenological and emission source modules. In the simulations the pollen season onset and its following propagation were based on the model internal formulations only, whereas the corrections to the mean pollen production were introduced with the four-dimensional variational (4DVAR) data assimilation independently for every year. The data of in-situ pollen observations of European Aeroallergen Network (EAN) in 34 countries and national pollen databases were used to form non-overlapping assimilation and validation datasets. The 4DVAR assimilation of emission correction technology is embedded in the SILAM model code. The meteorological reanalysis ERA5 was used as an input to drive the SILAM computations.

Results

Assimilation of the airborne pollen concentrations was performed separately for every year and each tree genera. It resulted in a better representation of pollen observations (including those which were not assimilated) than the unconstrained model run. The reanalysis produced the near surface and three-dimensional hourly pollen concentrations, dry and wet deposition, as well as corrected pollen emission fields. Data are provided on the regular lon-lat grid of 0.1° x 0.1° cells for longitude range 25W-45E, and latitude range 30N-72N (700 x 420 grid cells).

Conclusions

The first long-term pollen reanalysis was produced for three allergenic tree genera in Europe (1980-2022) with the use of extended 4-dimensional variational data assimilation of the observational data of the EAN and national pollen databases by the SILAM model. The procedure substantially improved the model representation of the inter-seasonal variations of pollen absolute levels, thus creating the first systematic assessment of airborne pollen abundance in Europe throughout the last four decades.

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An Automated Low-cost Sensor System for Fine-temporal Analysis of Pollen Emissions

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Abstract

Background: Understanding and mitigating the effects of pollen on human health requires information on the temporal and spatial variation in pollen emissions. We are limited in studying that by constraints of the existing pollen sampling devices. Traditional impaction devices are reliant on laborious manual microscope slides preparation and counting. Although robust in pollen identification accuracy, these devices fall short in delivering fine temporal information on pollen concentrations. Emerging automatic measurement systems, on the other hand, provide near real-time measurements but high costs restrict the extent of their use. Studying spatial and temporal variation in pollen emissions requires placing several sensors across a landscape. The ideal sensor would be affordable, compact, with low power-requirements, and able to sample at high temporal frequency.

We assess the suitability of an automated system using low-cost sensors for measurements of pollen emissions at high temporal resolution directly in the pollen source, i.e., within the crown of allergenic trees taxa. We use this system to ask whether daily pollen release dynamics correspond to fluctuations in meteorological parameters.

Methods: We used a commercially manufactured optical particle counter from Alphasense, OPC-N3, which draws in the air with a built-in fan, counts particles and segregate them by size based on light scattering intensity. Designed to sample particles up to 40 µm in size, the instrument is theoretically capable of detecting pollen grains from most anemophilous tree taxa. Five OPCs were placed individually in waterproof enclosures, each equipped with a commercial microprocessor for data logging and batteries. Instruments were fixed on hazel and alder trees at the University of Exeter Penryn Campus in winter 2024 (in progress) and one sensor was placed on the roof of an adjacent building to represent the background particle concentration. Reference manual measurements of pollen concentrations are performed intermittently with Burkard's Personal Volumetric Air Sampler (PVAS). Local meteorological and micrometeorological conditions are recorded by the weather station at Penryn Campus and by temperature and humidity loggers placed alongside the OPCs.

Results and conclusions: Comparison with PVAS indicates that OPC-N3 can detect pollen signal from flowering hazel and alder trees. We demonstrate that validation with PVAS is necessary because of the sensors' sensitivity to high humidity and tendency to overestimate particle concentrations during humid weather or rainfall events. Once the extensive validation period is over and OPCs tested on other tree taxa (birch and oak) the full potential of the sensors

to detect fine scale variations in pollen emission process can be inspected. This study shows the potential of low cost OPCs to measure temporal and spatial variation in pollen concentrations in field conditions, and how this potential can be maximised.

Keywords: Optical Particle Counter (OPC), Low-cost sensors, automated pollen monitoring

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Implementing the Aerobiological Monitoring Network of the Lombardy Region (Italy)

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Abstract

Background

Several studies highlighted the importance of the aerobiological monitoring for the human health: it is an important prevention tool, seeing the negative impact shown by the aerobiological particles on the human health and the increase of the respiratory allergies, often related to the climate change.

Before the COVID-19 pandemic, in Lombardy 12 monitoring stations were active. That stations used the same standardized sampling method, referring to the UNI-EN 16868:2019. They regularly provided the weekly pollen bulletin.

Just shortly before the pandemic, the implementation of a regional aerobiological network was decided in the framework of the General Directorate for Welfare of the Region. A working group was officially approved, with the final objective to implement the regional aerobiological network.

Unfortunately, due to the very severe pandemic, many human resources were necessarily moved to the fight against COVID-19 and some aerobiological traps were consequently stopped. The results was the decreasing of the active monitoring stations, dropping from 12 to 7.

The aim of our work was to analysed the situation of the aerobiological monitoring in Lombardy, in order to restart with the implementation of the network.

Methods

At the end of the COVID emergency, the work to implement the aerobiological network has restarted.

A recognition of the available tools was carried-out: how many active pollen-traps, how many supporting equipment, how many specific data-base and dedicated software to processing data and produce the bulletins. Parallel, the required pollen traps were assesed based on the evaluation of the environmental homogeneous areas (i.e. plain or mountain, urban or rural areas) and some particular situation of the vegetation (i.e. zones highly infested by ragweed). At the same time, the financial support needed and the availability of expert people in this field were also evaluated.

Results

The financial support was found amoung the NRRP (National Recovery and Resilience Plan) and used to buy new traps and equipment.

The locations of the new traps were identified and the future aerobiological network has been drawn, hypothesizing 14 stations.

Only in one case, a dedicated software was available and, based on this one, the characteristics of a common regional software were determined. Parallel, the format of the bullettin was defined.

Few people expert in aerobiology and able to examine the aerobiological still remain.

By the Regional Document which defines the annual planning of the Regional Health Service (RHS), the aerobiological monitoring has been officially included among the activities to be developed by the Departments of Hygiene and Health Prevention of the 8 Agency for Health Protection of the Lombardy Region, collaborating with other entities of the RHS (i.e. Hospitals).

Conclusions

Based on our results, we plan to start with the new aerobiological network during the 2024, covering the whole regional territory, implementing a common way to communicate aerobiological data and creating a regional data base to facilitate the large-scale data processing.

Furthermore, specific training course, periodic meeting and quality control program will be necessary in order to mantein an adeguate level of aerobiological knowledge finalized to protect the human health.

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Bioaerosols in Urban Settings: Pollen and Fungal Spore Concentrations and Diversity at Different Heights, Indoors and Outdoors

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Abstract

Most people spend two thirds of their time indoors, either in their residences or in occupational environments, and hence, it is imperative to know the indoor air quality. The aim of this study was to detect indoor biodiversity as compared to the outdoor one, in terms of pollen and fungal spore taxa, and explore the extent to which this biodiversity affects indoor environments.

We investigated the concentrations of these two types of bioaerosols along an 11-floor building, with sampling being conducted twice every week for a full calendar year. Five sampling heights were selected, from 1.5 to 32 m. For four (4) of them (at 1.5m, 10m, 18m and 25m), two personal Hirst-type volumetric samplers were used, one indoors and one outdoors. Sampling at all four heights was completed within two hours, each lasting for 20 min. During the same period, a stationary Hirst-type volumetric sampler was operating on a continuous basis at the rooftop (32m) of the same building . Pollen grains and fungal spores in the air samples were identified and counted under an optical microscope; results were expressed in particles m^{-3} of air.

Outdoor concentrations of both bioaerosol types were generally decreasing with increasing distance from the ground, with the difference between the ground level measurements compared to the greater heights being statistically significant ($p<0.05$). Patterns of decrease were clearer and steeper for fungal spores. Outdoor concentrations were always higher than those indoors for all taxa. Beta diversity of both bioaerosol types between indoor and outdoor concentrations increases with increasing sampling height.

Evaluation of bioaerosol abundances and diversity, both outdoors and indoors, and quantification of their inter-relationship in urban settings, where people usually reside and work, are very important in our everyday life. Our findings indicate that our indoor bioaerosol exposure is largely defined by outdoor concentrations, regardless of the sampling height. Hence, continuous campaigns in both environments, and in a three-dimensional perspective, are considered necessary to develop appropriate modelling tools, which will aid to effective mitigation measures against allergic diseases.

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Quick but Not Dirty: Leveraging Spreadsheets in Classical Pollen Monitoring

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Abstract

Background

As opposed to automated monitoring now being deployed, classical airborne pollen monitoring (CAPM) still relies on standardized procedures where traps catch particles to be identified and counted under the microscope. Compiled data can then make their way to reports or datasets, used in turn to feed public services such as daily pollen content or forecasts, and research projects. Disseminating the data through different outlets, or routing them to specific pipelines (e.g. feeding regional, national, or European databases or monitoring schemes) often requires filling or reformatting the data files. While the vast majority of labor spent in CAMP goes to the actual, manual counting, a sizable amount also is spent in the data handling procedures, and applications and protocols exist to try to ease the task. As we came in charge of CAMP in Navarra (Spain) as contractors for the LIFE NAdapta project in 2019, we tweaked off-the-shelf spreadsheets to create a simple yet efficient integrated working environment that minimizes data handling and errors in the lab, while maximizing accessibility, reporting, and ease of transfer to external clients.

Methods and Results

NA-PoleOn (v.23) consists of Microsoft Excel and Google Sheet workbooks working in tandem to automatically pipe counts straight from the microscope to the public endpoints. In Excel, either online or offline, a sheet contains an array of definable individual buttons that act through a VBA function as counters/recorders for each pollen type. The technician clicks on the corresponding button as s/he encounters each grain along the prescribed search pattern and the data record accrues into a dataset. Other cells allow selection of counting parameters. Excel functions automatically organize data from the dataset into tables and files as required by various clients (local and regional governments and services, national and European databases) in their respective input formats with little or no human intervention. A Google Sheet workbook links back to the Excel workbook whenever it's online and pulls data as they become available, directly exposing monitoring data (last week's, current, and one-week predictions) to the general public as a simple color-coded array suitable for mobile phones, and to practitioners as a desktop web page with a more complex layout including plots and trend. When compared with traditional (manually-created) data reporting, NA-PoleOn has reduced the operator workload by an estimated 15%.

Conclusions

The system we devised does away with any intermediate human data handling in CAMP e.g. scratchpads or lists to be transferred elsewhere, thereby reducing possible errors to either misidentification or a wrong button hit (although the data can be corrected on the fly, too), while not relying on third-party specific apps that may require updates or versioning. As all coding and data pipes are built into common, general-purpose office automation applications, the system is itself future-proof and requires no particular adaptation or versioning on the user, although new or changed requirements by the clients can be implemented as needed. While NA-PoleOn is currently set for Navarra, it can be configured to any monitoring region using Hirst-type traps.

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The Pollen Sense Network and the Misery Index of Cities in North America

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Abstract

Two of the main goals of building a pollen counting network are first, to know what the pollen load is in a given area and second, to empower people with this knowledge so they can make informed choices so they can feel better. Without data, people experiencing allergies cannot know what allergens trigger their symptoms, find effective ways to reduce exposure, or obtain the right treatments to obtain relief for their condition. They are completely reliant upon healthcare providers. With data, people are given tools to make informed decisions that can have a direct impact on their well-being. Pollen Sense has built a network of 147 devices across North America. Data from the Pollen Sense network are being put to use at a continental scale.

One use of the Pollen Sense network is to identify the cities in the United States that are the most challenging places to live as determined by their pollen counts throughout the year. We do this by computing a Misery Index for tree, grass, and weed growth forms. The Misery Index normalizes the raw pollen counts to a scale of 0 to 100. The higher the number, the greater the likelihood pollen-sensitive people may experience allergy symptoms. The Misery Index is more readily understood by lay persons than raw pollen counts. Although numeric, the Misery Index is similar to the low, moderate, high, and very high designations established by the National Allergy Bureau and defined by pollen counts being at or below the 50th percentile, between the 50th and 75th percentiles, between the 75th and 99th percentiles, or above the 99th percentile, respectively.

During 2023, the cities with the highest Misery Index during the spring were Philadelphia (PA), New York (NY), Bridgeport (CT), Poughkeepsie (NY), and Hartford (CT). The cities with the highest Misery Index during the fall were Madison (WI), Milwaukee (WI), Chicago (IL), Minneapolis (MN), and Des Moines (IA). While pollen-sensitive residents in these cities may experience allergy symptoms when pollen is high, they can decrease their exposure to pollen if they know when during a given day pollen is highest. A useful tool helping to disseminate near real-time pollen data to interested users is Pollen Wise which can be used to get hourly Misery Index estimates for any given location within North America.

Results from this study are significant in multiple ways. First, this is the largest application of an automated pollen counting network to date. Second, pollen data are becoming available at temporal and spatial scales that are relevant to the personal health of individuals and this has never before been possible. And third, data from this network of automated pollen sensors demonstrates it is possible to overcome challenges with cost, space, and maintenance requirements to build a more comprehensive and efficient pollen counting network.

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Balancing Health, Esthetics and Biodiversity: Investigating Planting Inventory and Allergenicity in Urban Green Spaces in Vilnius, Lithuania

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Abstract

Background: Vascular plant rich urban spaces provide important ecosystem services such as local climate regulation, while species richness of plants is valued highly by local residents and pollinators alike. However, green spaces are also known to deliver a few disservices that may influence human health like increasing potentials for allergy problems caused by the spread of pollen from the urban nature. It is important to recognize both the benefits and detriments from these urban green spaces so as to minimize their potential negative impacts to human well-being.

The purpose of this study is a comparison of two urban green spaces in Vilnius, Lithuania in terms of the planting material inventory, while evaluating their plant species composition and allergenicity.

Methods: For the purpose of the study, two urban green spaces in Vilnius, Lithuania were selected: a sculpture garden at MO museum (area of 600 sq.m, planted in 2018), and plantings around Business Garden Vilnius (area of 5000 sq.m, planted in 2020). Both green spaces were assessed using plant material inventory surveys based on indicators derived from the literature. Both plantings were designed in a contemporary manner, with much attention given to the notion of 'right plants for a right space', making use of pollinator- and biodiversity-friendly native plants, including trees, shrubs and herbaceous perennials.

Results: In the course of this study, plant material inventories of both plantings were compared. The MO sculpture garden, while comprising 600 sq.m, had 10 trees, 2 of which are highly allergic – both *Betula utilis* ‚Doorenbos', while the rest 8 – *Magnolia stellata* and *Cornus kousa* – are classified as mildly or moderately allergenic. The herbaceous layer of the plantings, comprising 5000 pcs. herbaceous perennials, includes 1700 pcs. of ornamental grasses and sedges, none of which are deemed to be higly allergenic (*Sporobolus*, *Hakonechloa*, *Molinia*, *Carex* sp.).

The plant inventory of Business Garden Vilnius of 5000 sq.m lists 150 trees, belonging to 9 genera. Out of this, 2 anemophilic genera – *Betula utilis* ‚Doorenbos' and *Picea omorika* - are considered to be highly or moderately allergenic, respectively. The said plantings also include 850 shrubs (*Hydrangea*, *Cornus* sp.), which are not known to be of severe allergenic threat; and 39 000 pcs. herbaceous perennials, out of which number 19 000 pcs. belong to grasses and sedges. However, none of grasses or sedges used in the plantings are known to be highly allergenic.

Conclusion: Findings of this study show urban green spaces having a relevant place in the urban ecosystems. It would be neither possible, nor desirable to aim for totally allergen free urban spaces: even taxa with high allergenicity can offer positive contribution towards supporting biodiversity, or revealing a particular resistance to heat or drought. However, it is of utmost importance that allergenic plants should not be planted in great clusters or numbers. Rather, plants for green spaces should be chosen from different species and genera, thus reducing pollen concentrations, keeping high biodiversity and minimizing the risk of plant diseases.

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Unravelling the Aspergillus and Penicillium Diversity in the Air of the National Library of Greece

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Abstract

Libraries safeguard the written heritage of humanity. The preservation of books and archive material is related to the presence of airborne fungi that are potent biodeteriogens for the cultural artifacts. The National Library of Greece (NLG) was investigated to characterize the fungal aerosol spectrum with a focus on the genera *Aspergillus* and *Penicillium* that are of special interest for a library environment. They include numerous species with worldwide distribution, can grow on a wide range of substrates and may pose a threat to human health.

A portable air sampler for agar plates (Burkard Manufacturing Co Ltd) was used to sample fungal propagules during the years 2019 to 2020. Samplings were performed fortnightly and petri plates with Malt Agar (MA 2%) were exposed for three minutes each and incubated at 25° C for 2–4 weeks. The monitoring included reading rooms, book storage rooms, vaults for the rare and valuable artifacts, manuscript repositories, newspaper archives, restoration laboratories, offices etc., as well as the outdoor air for comparison. Morphologically different *Aspergillus* and *Penicillium* colonies on the sample plates were isolated in pure culture and identified to species level with a multifaceted approach that involved phenotypic analysis and molecular characterization of three genetic markers.

Penicillium was, after *Cladosporium*, the second most abundant inside the NLG contributing with 21 % to the total count, while *Aspergillus* was the third representing 5 % of the fungal aerosol. The mean daily concentration was 21 CFU m^{-3} for *Penicillium* and 5 CFU m^{-3} for *Aspergillus*. The indoor/outdoor (I/O) concentration ratio was 1.1 for *Penicillium* and 1.4 for *Aspergillus*. Regarding their diversity, totally 16 *Aspergillus* and 15 *Penicillium* species were recovered and characterized. The presence of common, widespread species like *Aspergillus niger* and *Penicillium chrysogenum* was documented inside the National Library of Greece, but also rare ones like *Aspergillus sigurros* and *Penicillium momoii*. These species are reported for the first time from Greece. *Aspergillus fumigatus*, a species of concern according to World Health Organization, was found in the air of NLG but its presence was very rare.

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Potential Distribution Range of *Sorghum Halepense* (L.) Pers

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Abstract

The success of invasive species has often been estimated as the actual size of the range in the introduced region. However, the current range is only a snapshot of the invasion at any given time and does not inform the potential range of the species. Based on niche-based models, we use climatic, geographic and landscape information on the current distribution of the species Sorghum halepense (L.) Pers in Spain to estimate its potential distribution, using publicly accessible databases (GBIF and WorldClim). For a more local prediction, in addition to the data obtained from the previous databases, field data have been taken on the presence and/or absence of the species at different altitudes in the province of Córdoba, to determine the forecast of the distribution of this species at the local level. This species currently has a wide range of distribution throughout the peninsula, focusing on the northeast and south. A prediction model has been made about the potential distribution in the future based on presence and absence data at the peninsular and local level. It has been observed that the potential ranges of Sorghum halepense (L.) Pers will expand into higher altitude areas, where they are currently restricted by temperature. Due to climate change, which is causing an increase in temperature in the peninsula, especially in the south, our model indicates that its distribution area, until then located in grasslands and riverbanks, will change towards higher altitude areas.

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Can Pollen Affect Precipitation?

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Abstract

Large primary bioparticles such as pollen can be abundant in the atmosphere, for example near-surface pollen concentrations above 10 000 particles per cubic meter can occur during intense pollination periods. On one hand, due to their large size (10-100 micrometers), pollens can act as giant cloud condensation nuclei and enhance the collision-coalescence process in clouds that leads to drizzle formation. On the other hand, in humid conditions pollens are known to rupture and release a large number of fine particles that can increase the cloud stability by reducing the droplet size. Additionally, both whole pollen grains and the sub-pollen particles released by pollen rupture are known to act as ice-nucleating particles (INs). Due to these complex interactions, the role of pollen in modulating the cloud cover and precipitation remains uncertain.

We used the UCLALES-SALSA large eddy simulator for simulating birch pollen effects on liquid and mixed-phase clouds. Our simulations show that the pollen concentrations observed during the most intense pollination seasons can locally enhance precipitation from both liquid and mixed phase clouds, while more commonly encountered pollen concentrations are unlikely to cause a noticeable change. The liquid precipitation enhancement depended linearly on the emitted pollen flux in both liquid and mixed phase clouds, however, the slope of this relationship was case dependent. Ice nucleation happened at relevant degree only if the process of rupturing pollens producing large number of fine ice nucleating particles was included in the simulations. Resulting precipitation was not linear to pollen emission and saturated for the highest IN concentrations. Secondary ice formation by rime splintering had only minor effect in the considered one-day timescale.

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A Model for Forecasting the Prevalence of Wheat Stem Rust Fungus

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Abstract

The complex life cycles of rust fungi (*Pucciniales*) with different temperature and humidity requirements in various phases and their reliance on airborne spores for annual recolonization of areas not climatically suitable for year-round survival make them especially sensitive to climate change. A detailed mechanistic model has been developed to simulate the spatiotemporal development of the prevalence of wheat stem rust (*Puccinia graminis* f. sp. *tritici*), that has been listed among the most devastating crop diseases. It includes parameterizations to account for spore production on infected wheat fields and escape from the crop canopy, spore mortality during long range transport due to UV radiation, the spread of the infection from the deposited viable spores taking into account the influence of local meteorological conditions on spore germination, and overwintering. The model runs with sub-daily timestep for timescales up to decades and can be used in both regional and global scale.

Model evaluation is challenging due to limited availability of quantitative observational data; however, the model does show some skill in reproducing the annual recolonization of United States wheat growing areas by stem rust compared to observations reported by farmers to USDA.

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WIBS-4+ Bioaerosol Sensor: Assessment of Intended-Use, and Evaluation of Alternative Aerosol Applications

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Abstract

This intensive real-time monitoring campaign was carried out over a two-month period in Saclay, a semi-urban centre, southwest of the city of Paris, France, and is an outer suburb/exurb of the city proper.

The Wideband Integrated Bioaerosol Sensor (WIBS) 4+ model was first compared to the traditional Hirst volumetric sampling method. It was evaluated for its ability to sample and detect ambient bioaerosol concentrations, namely fungal spores and pollen grains. Along with the WIBS device and the Hirst device, meteorological and pollution parameters were obtained from co-located monitoring devices at the research centre. This allowed the construction of several Multiple Linear Regression (MLR) algorithms.

For fungal spores, significant predictors of concentration were the A WIBS channel, wind from the south and south-westerly directions, and NOx emissions. For pollen grains, the WIBS 4+ additional Xenon flashlamp (distinguishing the WIBS 4 from the WIBS 4+), which allowed for the D, DE, and E WIBS channel categories, was a strong predictor of concentrations, when combined with northerly and easterly winds, and atmospheric ammonia concentrations. Additionally, the possibility of using WIBS 4+ technology to monitor aerosols that are non-biological was evaluated. Black carbon, which does fluoresce but does not need to be of biological origin, was found to strongly correlate with BC WIBS channel particles, along with various windspeed and wind direction parameters.

The work from this campaign shows the strong bioaerosol monitoring capabilities of the WIBS technology, the benefits of the additional Xenon flashlamp, and the potential alternative and novel uses for which the device can be deployed.

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Interpolation of Dataset Gaps via Use of the AeRobiology "R" package.

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Abstract

An attempt was made in Ireland at running a nationwide traditional bioaerosol monitoring campaign in the Summer of 2021. With limited financial and staffing resources, it was impossible to simultaneously run the required samplers continuously for the duration of the campaign period. Resultantly, an exparimental campaign commenced in which traditional Hirst bioaerosol monitoring devices were installed at four locations across the island of Ireland, in Dublin, Cork, Carlow, and Sligo. The Dublin location was managed by the research team, while the three remote locations were managed by volunteer participants. Due to the aforementioned constraints, only one single location (Dublin) was monitored as a continuous dataset throughout the course of the 12 week campaign, while multiple stoppages of varying lenghts were observed at the three other locations. As a result of this, several different data processing and analysis approaches were employed and compared with one another to attempt to best replace or repair the missing data.

The tool used for this campaign was the “interpollen” tool available within the “AeRobiology” R package. Methods employed and compared were as follows, the moving mean method, the lineal method, the spline regression method, the time series LOESS method, and the neighbour station (Spearman) method. The complete dataset in Dublin was used as a control, with the same transformations performed on spliced copies of the dataset, so that it matched each station. In this study, the lineal and neighbour station methods performed the worst The lineal method simply connecting the gaps with direct lines of data, so as to minimise variation. The neighbour method was unsuitable due to the large distances and considerable ecological and meteorological differences between the sites. The moving mean method, performed well, mainly due to the majority of the gaps in the dataset lasting only one week in duration, or shorter. The spline method was affected by instances in which not enough valid days of data were available immediately before or after a missing period, or if the missing data was also the start or end of the measurement campaign. The lack of historical databases for the majority of stations greatly affected the accuracy of the stations with missing data, but was able to perform strongly for Dublin, due to multiple years of collected bioaerosol values.

While the majority of the field is moving to unmanned, online monitoring devices, the issue of missing data will persist in some form, and it is vital that some appropriate statistical back-up is available, especially for the purposes of engagement and outreach through data visualisation with the general public and interested stakeholders.

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The Top Pollen Taxa From an Allergological and Aerobiological Point of View Using the Example of the Pollen Spectrum of the City of Berlin, Germany

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Abstract

Introduction:
According to the results of the population-based German Health Interview and Examination Survey for Adults (DEGS1) (2008–2011), participants were most frequently sensitised to the following eight pollen: 1st *Poaceae* (specifically *Phleum*), 2nd *Betula*, 3rd *Poaceae* (specifically *Secale*), 4th *Alnus*, 5th *Corylus*, 6th *Fraxinus*, 7th *Artemisia,* and 8th *Ambrosia*. How common are these allergologically important pollen taxa (PT) in the outside air, using the pollen spectrum of the city of Berlin, Germany, as an example?

Methods:
The results of the DEGS1 are population-based, therefore they are assumed here to be valid for Berlin too (although there may exist some local variations) and are presented here as the top 8 PT from an allergological perspective (Top-8-ALL). To gain insight into the pollen spectrum composition of the Berlin air, pollen data from the pollen monitoring station of the German Pollen Information Service Foundation in Berlin were used. The air was monitored using a volumetric spore trap (Hirst type) at roof level in the city centre, with subsequent light microscopy pollen analysis of the daily samples following VDI-4252-Blatt-4:2019-03 (Germany). For the 9-year period (2014–2022), the mean annual pollen integrals (APIn) in pollen/m^3 of air were calculated for the PT of the Berlin pollen spectrum.

Results:
The top 8 PT of the Berlin airborne pollen spectrum (Top-8-AERO) were as follows: 1st *Betula* (with an average of 15,739 pollen per year [P/a]), 2nd *Pinus* (14,615 P/a), 3rd *Urticaceae* (9,636 P/a), 4th *Alnus* (8,310P/a), 5th *Quercus* (5,883 P/a), 6th *Poaceae* excluding *Cerealia* (3,426 P/a), 7th *Taxus* (2,948 P/a) and 8th *Platanus* (1,884 P/a). 3 PT out of the Top-8-ALL were represented in the Top-8-AERO. The allergologically important birch pollen (2nd in the Top-8-ALL) was also very frequent in the air (1st in the Top-8-AERO), as was *Alnus* (4th in the Top-8-ALL and 4th in the Top-8-AERO), whereas the allergologically most important *Poaceae* pollen (1st in the Top-8-ALL) was only 6th in the Top-8-AERO. The remaining PT from the Top-8-ALL were distributed in the Berlin pollen spectrum as such: *Fraxinus* in the 11th place with 777 P/a, *Corylus* 14th with 635 P/a, *Artemisia* 16th with 478 P/a, *Secale* 29th with 113 P/a, and *Ambrosia* 32nd with 95 P/a.

Conclusion:
The example of the Berlin pollen spectrum shows that only some of the PT of the Top-8-ALL

were found in the Top-8-AERO and others of the PT of the Top-8-ALL (for example *Ambrosia*) were rather rare in the Berlin air, whereas allergologically less significant PT (*Urticaceae*) or allergologically insignificant/significance unknown PT (*Pinus*) were among the most frequent PT in the air. There is no clear correlation between the frequency of sensitisation and the occurrence of the triggering PT in the air. As a take-home message, we recommend using the terms "frequent/common pollen/allergen" and "rare pollen/allergen" in clear context with the respective subject area – from an allergological point of view or an aerobiological point of view.

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Environmental Allergens Screening in Indoor Environment, in Students Houses, Using Molecular Methods

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Abstract

Indoor environments are characterized by several sources of pollution, and the health problems associated with bad indoor air quality have acquired high importance in recent years. The biological particles present in the indoor environment can be of various types including infectious (viruses, bacteria, and fungal spores) and allergenic particles generated by the house dust mites, fungal spores, and pollen. In "healthy" homes and buildings, most of the fungi come from external sources and the total number of fungi is usually lower than in the outdoor environment. The pollen detected in the indoor environment is usually dependent on the concentration of pollen in the outdoor air, but nowadays its contribution to allergic disease is undetermined. General identification techniques of pollen and fungal spores are based on optical microscopic observation; however, it is time-consuming, and it needs an experienced taxonomist. Molecular biology techniques are faster and more used for indoor allergen identification and can be useful in outbreak prevention.

This work aimed to detect the presence of fungi and pollen in indoor environments in dwellings in Évora, south of Portugal, by molecular methods.

Electrostatic dust fall collectors (EDC), developed in the scope of the COST ADOPT action CA-18226, were used for indoor sample collection and placed near the entrance door of the house,

living room or bedroom, during the 3rd week of November 2022. The bioaerosols counting was executed using standard Hirst-type traps and identified by optical microscopy, according to the standard methodology (REA.com). Eighteen volunteers participated in this study by completing a questionnaire and placing the EDC in a specific location in the house. For Real-Time PCR fungal detection, the Internal Transcribed Spacer (ITS) region was considered, and for airborne pollen detection, the trnL gene was selected.

The results of the bioaerosol counts showed that fungi represent the largest outdoor contribution. PCR results showed an amplification curve and a respective melting peak in all the samples for the ITS gene, and 8 out of 18 were positive for the trnL gene in samples placed next to windows. Electrophoresis results showed 2 bands, demonstrating the presence of, at least, 2 different fungal species. The results demonstrated that the presence of fungi regardless of the sampled location was detected. For pollen species, only 1 band was observed.

The total DNA concentration extracted from the EDCs was around 4 ng/µL. Nevertheless, with this low amount, it was still possible to detect several bioaerosols in the indoor environment that are more difficult to identify by the standard methods.

The results have shown that it is possible to identify different biological particles, from different species, in the indoor environment, contributing to generating alert systems for the populations and, thus being useful in outbreak prevention.

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Screening for Environmental Allergens in Indoor Environment, in Different Spaces of a Public Institution, Using Molecular Methods

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Abstract

The health problems associated with indoor air quality have acquired high importance in recent years. Biological particles in the indoor environment include viruses, bacteria, fungal spores, and house dust mites (HDM). These may cause a set of different respiratory dysfunctions including infection and allergic and/or other inflammatory manifestations. There is no established methodology to identify all bioaerosols and the existing ones are time-consuming and need experienced taxonomists. This work aimed to detect the presence of HDM, bacteria, and fungal genes in indoor environments, using molecular methods.

Electrostatic dust-fall collectors (EDC) were used for indoor sample collection and placed in 15 selected rooms at the University of Évora, regularly frequented by students and staff, on the 23rd-30th of November 2023, following a protocol developed in the COST Action ADOPT CA-18226. The bioaerosols counting was carried out using standard Hirst-type traps and identified by optical microscopy, according to the standard methodology (REA.com). For Real-Time PCR, two species of HDM were considered, *Dermatophagoides pteronyssinus* (DP) and *Dermatophagoides farinae* (DF); for fungal and bacterial detection the ITS1-ITS2 and the v3-v4 regions, respectively, were selected.

The results of the bioaerosol counts showed that fungi represent the largest outdoor contribution. The PCR and electrophoresis results showed that DP was detected in 5 samples (33%) (1 canteen, 1 classroom, 1 laboratory and 2 outdoor samples). DF was detected in 13% (1 canteen and 1 classroom). The 16S rRNA gene was detected in 20% of the samples (1 classroom, 1 laboratory and 1 outdoor sample). Sixty per cent of the samples tested positive for the fungal genes, namely 2 libraries, 2 classrooms, 1 canteen, 1 laboratory, 1 toilet, 1 animal reception room and 1 outdoor sample.

The total DNA concentration extracted from the EDCs was around 4 ng/µL. Nevertheless, with this low amount, it was still possible to detect several bioaerosols in the indoor environment that are more difficult to identify by the standard methods.

In conclusion, molecular methods are potent tools to identify different components of the bioaerosol using affordable collection devices, in the indoor environment. This approach establishes the basis for the development of monitoring strategies, thus useful in disease prevention.

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Chemical Elements and Free Radicals in Pollen: a Case Study

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Abstract

A new era in aerobiology research has begun with the transition from manual to real-time automatic bioaerosol monitoring in Europe. This shift relies on advanced technologies such as laser systems and image recognition, enabling the detection of particles based on fluorescence, particle shape, signal strength, and other characteristics, which means that pollen grain characteristics directly impact the result of recognition. The high-quality real-time pollen and spore recognition remains a challenge. In parallel with the automatization of bioaerosol monitoring, laboratory experiments on pollen grains are performed in Europe. Different microscopic studies on pollen are important for several reasons, both for (potential) improvement of automatic bioaerosol monitoring and for advancing human health studies. Including: (1) investigating if pollutants (debris) accumulate on airborne pollen, affecting recognition properties during automatic monitoring; (2) assessing the differences between clean and contaminated pollen; (3) determining if urban and rural plants produce pollen with differing chemical compositions; (4) supporting fundamental research in aerobiology; (5) exploring new applied directions in pollen research. This study aims to compare the chemical characteristics of pollen from various taxa and environments and to evaluate the methods used for such analyses.

Several case studies on the chemical composition of pollen were conducted in Latvia. Samples of *Betula*, *Pinus*, and *Salix* pollen from different environments were analyzed using a scanning electron microscope (SEM) at Glasgow University to identify visual changes of polluted pollen and chemical elements present. Additionally, the analysis of free radicals in *Ulmus, Populus, Salix, Corylus, Alnus*, and *Betula* pollen was carried out. Sample preparation was performed in the Laboratory of Atmospheric Processes and Aerobiology at the University of Latvia, while electron paramagnetic resonance (EPR) spectroscopy was performed at the Latvian Institute of Organic Synthesis.

SEM analysis of clean pollen samples showed the presence of chemical elements such as oxygen (O), carbon (C), phosphorus (P), potassium (K), and calcium (Ca) on the surfaces of hazel, alder, willow, pine, and birch pollen. Hazel pollen showed the presence of tin (Sn). Polluted willow and birch pollen surfaces contained silicon (Si), sulfur (S), and copper (Cu). EPR spectra identified Fe^{+3} and Mn^{+2} signals alongside organic free radicals, with typical g-values around 2.006 ± 0.002. The concentration of organic radicals varied significantly among pollen samples from different tree species, with the highest concentrations in *Salix* and *Ulmus* pollen and the lowest in *Populus* pollen. *Corylus*, *Alnus*, and *Betula* pollen exhibited similar concentrations of organic radicals.

The study shows the potential for using chemical and free radical analyses to better understand the pollen composition, the impacts of environmental pollution on pollen and its implications for human health.

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Personalized Approach In Pollen Allergy Forecasting

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Abstract

A personalized approach is becoming more common in numerous healthcare directions, including preventive medicine. The noticeable diversity in the intensity of individual reactions under the same environmental conditions forces scientists to develop personalized symptom forecasts, for example, in the direction of pollen allergy. The well-known importance of having the preventive medical treatment two weeks before the start of pollen season, allows the introduction of drug therapy and prevents severe allergic reactions in allergy sufferers. Together with that, in everyday life, people quite often tend to forget to follow the timing of pollen forecasts and are forced to deal with sudden and unexpected health reactions.

The personalized pollen allergy symptom forecasting system PASYFO was developed as the supportive tool to manage these issues. The expansion of PASYFO to the European level has been performed in the Horizon project, "AI-augmented ecosystem for Earth Observation data accessibility with Extended Reality User Interfaces for Service and data exploitation" EO4EU, funded by the European Commission.

The system is available as a mobile application as well as a web page, https://pasyfo.eu. The mobile application includes sections of:

- personalized symptom forecasts - developed based on statistical and persistence models;
- general allergy risk for the selected area - calculated considering all the symptoms and air quality data from the particular area;
- plant guidebook - providing end-users with relevant information on allergenic plants.
- animated pollen forecasting maps;
- access to the personalized symptom entries;
- technical section- for language, country, area and allergen selection.

The website, the second essential component, which serves as an international platform for sharing applied pollen research and allergy-related information. This collaborative approach enriches our understanding of allergies and enhances the quality of information available to end-users.

The development and expansion of PASYFO offer a promising solution to the challenges faced by allergy sufferers, delivering timely and personalized information that can substantially reduce the impact of pollen allergies. PASYFO empowers individuals to manage their health proactively and facilitates collaborative research across Europe.

Acknowledgements: This study was supported by the European Commission Horizon project "**AI-augmented ecosystem for Earth Observation data accessibility with Extended Reality User Interfaces for Service and data exploitation"** EO4EU.

AutoPollen

Pollen allergies affect between 10 to 30% of the population, and their prevalence is expected to continue rising over the coming decades. Real-time measurements and forecasts of allergenic pollen are helping to improve the diagnosis and management of these allergies and thus potentially affecting the quality of life of millions of people. Such methods can also be used to study climate change impacts, track invasive species, and better understand agricultural pathogens.

The AutoPollen programme (www.autopollen.net), established under the framework of EUMETNET (the European network of national meteorological services www.eumetnet.eu), brought together meteorological services, environmental and health institutes, and scientists to develop a European network for the real-time detection of pollen and fungal spores. AutoPollen focuses not just on the measurement techniques, but the whole information chain, all the way through to innovative products and services for the wide range of end-users of pollen and fungal spore information. During the 2016-2023 phase of the programme, the community developed methods, recommendations, and helped create a strong momentum across Europe. The main results were published in a special issue in Aerobiologia, ensuring that knowledge and expertise is available to all. AutoPollen also strongly engaged with stakeholders to better understand end-user needs and collaborated with a number of other projects such as the ADOPT COST Action and Copernicus CAMS-23 to organise training schools and further develop techniques and infrastructure across Europe. Finally, the AutoPollen community continues to campaign at the European level to promote real-time pollen and fungal spore measurements, with the long-term goal of developing a legal basis for these observations.

The 2024-2028 phase of AutoPollen is continuing to grow the automatic monitoring network across Europe. This includes creating a quality label for measurement sites, further developing algorithms to identify more pollen and fungal spore taxa, and improving data quality and comparability. Jointly with the Horizon Europe SYLVA project, AutoPollen is helping to develop infrastructure that can support the growing network, ensure better access to data, and standardise the application of methods, algorithms, and quality control. Improving access to information, including enhanced forecasts based on real-time observations, will enable medical doctors and allergy sufferers to better diagnose and manage their symptoms, potentially helping reduce related health costs. With further developments focused on fungal spores, agricultural and sylvicultural pathogens may also be better managed, particularly through more timely and precise application of fungicides. Finally, continuing work from the first phase of AutoPollen, a European standard for automatic pollen and fungal spore monitoring should be completed by the end of the current phase of the programme.

This session aims to provide information and a forum for discussion for all those interested in automatic methods for real-time pollen and fungal spore monitoring. The AutoPollen Programme will be briefly introduced, but the main focus will be on engaging with all stakeholders interested in the topic – answering questions, providing information about available infrastructure, and discussing issues related to automatic bioaerosol detection methods.

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EO4EU

UNLOCK THE POTENTIAL OF EARTH OBSERVATION DATA WITH EO4EU

The EO4EU Platform revolutionizes the accessibility and usability of Earth Observation (EO) data. Developed by a European Commission-funded consortium of 20 partners, EO4EU streamlines and enhances EO data use for various applications. Whether you are a researcher, policymaker, business leader, or citizen scientist, EO4EU offers unparalleled tools and resources to advance your projects.

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EO4EU connects users with major EO data repositories such as DestinE, GEOSS, INSPIRE, Copernicus, and Galileo. The platform's user-friendly interface and advanced search capabilities make finding and utilizing specific data effortless. Leveraging machine learning, cloud computing, and high-performance computing, EO4EU ensures quick and accurate data processing. Its extended reality interfaces allow intuitive visualization of complex data, turning raw information into actionable insights.

Diverse Use Cases Demonstrating Impact

EO4EU supports seven pilot use cases across various areas:

- Personalized Healthcare: The PASYFO system provides allergy forecasts, enhancing quality of life.
- Civil Protection: Improved EO datasets aid in managing natural and man-made risks.
- Environmental Pest Control: Monitoring and predicting locust plagues to mitigate agricultural damage.
- Soil Erosion Management: Assessing soil susceptibility to inform prevention strategies.
- Forest Ecosystems: Insights into forest services like timber production and climate regulation.
- Food Security: Addressing food security through climate indicators and risk forecasts.
- Ocean Monitoring: Optimizing shipping routes to reduce travel time and environmental impact.

Top Features

- Easy-to-Use Interface. Designed for both technical and non-technical users, the platform offers personalized dashboards, a smart search engine, real-time and historical data visualization, analysis, interpretation, and interactive mapping. Users can also write and execute code in Python, Node.js, PHP, and C# with ease.

- AI and ML Resources. The platform provides AI and machine learning tools for various tasks such as crop recognition, locust identification, and fire detection. These resources support the development of task-specific models.
- Data Fusion. Interpolate and extrapolate datasets from different spatiotemporal snapshots using the Fusion Engine, allowing for comprehensive data analysis.
- Access to Raw EO Data. Use natural language queries to inspect and retrieve raw EO datasets, and forward them to an OpenEO-based API for further processing.
- Ease of Repetition. Select and execute ML algorithms, Fusion processes, or FaaS functions from the Marketplace with clear descriptions and user guides, ensuring seamless repetition of tasks.
- Immersive VR Experience. Experience EO data in a new dimension with a virtual reality interface powered by augmented and extended reality technologies.

Join the EO4EU community at www.eo4eu.eu and shape a sustainable future through data-driven innovation.

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SYLVA

More than 80 million Europeans are allergic to s pollen and pollen-related health impacts cost society over €50 billion per year. Meanwhile, the technology used to monitor pollen release dates back to 1950. SYLVA is a Finnish-coordinated European Union (EU) Horizon Europe (https://sylva.bioaerosol.eu/) project aiming to support the development of bioaerosol monitoring technologies and infrastructure across Europe, considering environment- and climate-related bioaerosol changes. SYLVA aims to radically improve the temporal resolution, timeliness, coverage, and availability of information about aeroallergens and other bioaerosols by pairing technological innovations with new infrastructure, distribution, and exploitation pathways. This information is key for agriculture, forestry, and human and plant health.

To go beyond the state-of-art, SYLVA combines several cutting-edge technologies, big data and machine learning techniques supported by innovative Information and Communication Technology (ICT) infrastructure, data collection, and Demonstration Pilot campaigns to construct and evaluate a versatile prototype of a Europe-wide bioaerosol monitoring system that provides game-changing information to end-users (Figure 1).

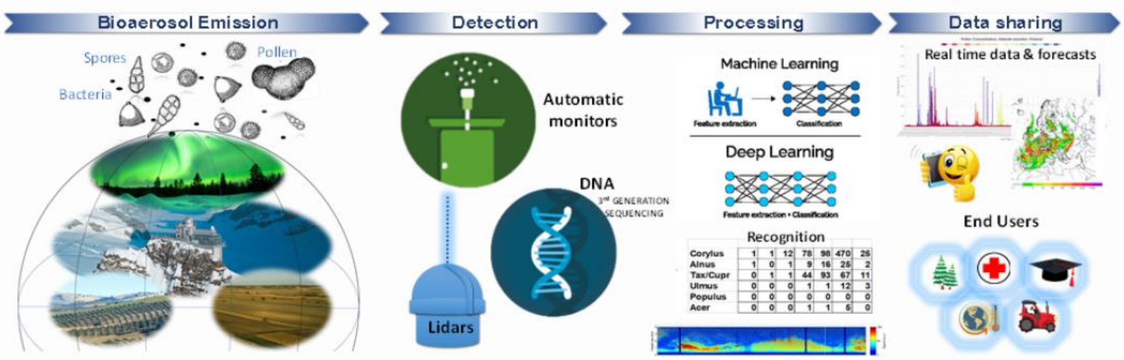

Figure 1 SYLVA concept and scope

The main goal of the Demonstration Pilot campaigns is to ensure the seamless application of automatic bioaerosol monitoring technology across Europe, including in key biogeographic and bioclimatic areas with extreme conditions, going beyond the conditions where these technologies have been tested to-date. The Pilots include (i) cold and humid Northern Europe, (ii) hot and dusty Southern Europe, and (iii) high-altitude Alpine areas, and 5 different types of automatic bioaerosol monitors. The field trials will demonstrate the added-value of SYLVA innovations for public health, agriculture, forestry, and monitoring climate change impacts.

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