

# **Aerobiology: current stage and future perspectives**

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## **Book of Abstracts**



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1

## Pollen concentration of invasive tree of heaven (*Ailanthus altissima*) on the Southern Great Plain region, in Hungary between 2019 and 2020

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Nowadays, there is an increasing emphasis on the problem of invasive species. In areas where the tree of heaven (*Ailanthus altissima*) appears and multiplies, the original vegetation degrades and transforms. The tree of heaven is of great importance in urban environments, where it causes building damage, static problems, and endangers utilities. In addition, it is worth mentioning that the pollen of *A. altissima* is allergenic, although less important than ragweed pollen.

Pollen concentration of tree of heaven was measured in three counties of the Southern Great Plain region (Bács-Kiskun county, Csongrád-Csanád county, Békés county) with the 7-day Hirst-type (Burkard) pollen trap.

The highest annual total pollen count was detected in 2019 in Bács-Kiskun county (66 pieces) and Csongrád-Csanád county (36 pieces), while in Békés county (16 pieces) in 2020. In Békés county, a trap error was detected when measuring the pollen count of *Ailanthus altissima* in 2019, therefore the results cannot be used.

Our work draws attention to the differences in the distribution of the tree of heaven in the Southern Great Plain, based on which it can be seen that there can be more than twice the differences between the cities in terms of the total annual pollen count.

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## Real-Time monitoring of pollen and fungal spores in Dublin, Ireland

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Bioaerosols are omnipresent within all ecosystems globally and have been shown to have serious impacts on human, animal, and plant health, with additional climatic affects noted. Thus, an understanding of the seasonal variation of bioaerosols such as pollen and fungal spores and the associated species with these categories is of great interest to the public, policy makers and academics alike. Hence, the use of online spectroscopic techniques have begun to be utilized in several locations around the world. As to allow for the determination of bioaerosols in near real-time.

In this study real-time fluorescence spectroscopy devices, the WIBS NEO (Wideband Integrated Bioaerosol Sensor) and IBAC-2 along with a traditional bioaerosol sampler (Hirst volumetric trap) were all run in tandem. This was undertaken at the TU Dublin Kevin Street sampling site (53°33'67" N, 06°26'79" W) at a height of 20 m) for a 6 week period in the summer of 2019 (07/08 to 16/09).

A Spearman correlation test was then used to calculate the degree and the significance between selected variables and fungal spores/pollen concentrations and WIBS/IBAC-2 sampled particles. The statistical analysis was performed using the packages `nortest` and `corrplot` in the software R (R Core Team, 2017)

The results obtained, and subsequent comparisons between the three devices and monitoring methods, show the ability of the WIBS NEO device is being used as a real-time bioaerosol sensor and its capabilities in discriminating between different pollen and fungal spores and non-fluorescent ambient particles. Good agreement for Total pollen and *Alternaria* spores ( $R^2=0.8$ ), Basidiospores ( $R^2=0.66$ ) and Ascospores ( $R^2=0.93$ ) were seen for comparisons between the Hirst and WIBS. However, the sensitivity of the WIBS-NEO also led to anthropogenic particles interfering with the determination and differentiation of PBAP, even at increased fluorescent thresholds.

The IBAC-2 did not exhibit a good correlation with ambient fungal spore concentrations, likely as a result of interferences by anthropogenic particles and rain droplets. High amounts of rainfall were shown to impact the sampling specificity of the IBAC-2, resulting in the recording of large particle counts that did not coincide with the other instruments.

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## Alternaria and Cladosporium modelling in Ireland

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Fungal spores are considered some of the most important aeroallergens, triggering allergic symptoms and further exacerbating existing respiratory conditions. Predicting periods when ambient concentrations of allergenic fungal spores will be high, represents an important resource for allergy sufferers and those suffering from respiratory conditions such as asthma. Aerobiological surveys have reported *Alternaria* and *Cladosporium* are two of the most prevalent airborne fungal spores and are important aeroallergens. The main goal of this study was to analyse the airborne concentrations of *Cladosporium* and *Alternaria* at a number of sites in Ireland and construct a model to predict the concentrations.

The ambient monitoring was carried out between the years 2017 to 2021 in Dublin and from March to August 2021 in Sligo, Carlow and Cork. Fungal spore monitoring was conducted via Hist-type spore traps in each of the locations following the recommendations of the European Aerobiological Society.

Two different types of models were constructed, classification models and regression models. Two methods of classification models were developed, Random Forest (RF) and support vector machine (SVM) models. A 3 level concentration classification was used for *Alternaria* (<10, 10-99 and >100 fungal spores/m<sup>3</sup>) while a two-level concentration break up to differentiate between high and low was used for *Cladosporium* (<500 and >500 fungal spores/m<sup>3</sup>). With regard to the regression type models used, Least Trimmed Squares Robust Regression (Robust Model), Generalized Linear Ensemble (Ensemble Model) and Generalized Additive Models (GAM Model) were developed. Dublin monitoring data was used for model training while validation of the models was done with data obtained from Sligo, Cork and Carlow.

Overall, the SVM models showed a greater accuracy when applying the developed *Alternaria* models to the data collected from the Cork and Sligo sampling sites, with model accuracy exceeding 60%

and 75% accuracy, respectively. The Random Forest models, on the other hand, performed best for the Carlow sampling site with model accuracy exceeding 60%.

Moving to the modelling of *Cladosporium* at the sites, the developed SVM model performed best for both Carlow and Sligo, yielding model accuracies of greater than 75% and 40%. Whereas the random forest model performed best for Cork with model accuracy results exceeding 60%.

The robust model performed best for *Alternaria* prediction in Carlow, whereas the ensemble model achieved the best results for *Alternaria* prediction in Cork and Sligo. Overall, models were less accurate in predicting *Cladosporium* concentrations, especially for Carlow. On the other hand, the robust model and GAM model performed best for *Cladosporium* prediction in Cork and Sligo, respectively.

Any notable deviations observed between predicted and expected values can be explained due to deviations introduced by applying the models to different sampling locations. As these sites likely experience different changes in environmental factors. In addition, at this early stage in model development, it is expected that periods of extreme deviations in spore concentration will not be accurately predicted by the developed models using the available training data alone. The use of classification models for current *Alternaria* and *Cladosporium* forecasting is advised rather than the use of regression models.

This is due to the improved accuracy experienced when broader concentration levels are predicted over specific numerical values. As monitoring efforts continue to grow and expand, the incorporation of additional data will greatly improve model performance and accuracy in the future. Increased monitoring will also provide an opportunity to develop location-specific models for the other sampling locations and spore types.

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## The sources of long-term trends of airborne birch and grass pollen levels in Belgium

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Airborne pollen may have a substantial contribution to respiratory allergies. In Belgium, ~10% is sensitive to birch pollen and ~15% to grass pollen. Since climate change and land-use change tend to enlarge the amount of allergenic airborne pollen and prolong the pollen seasons, even more people might be affected in the future.

Here we apply the pollen transport model SILAM (System for Integrated modeLLing of Atmospheric coMposition) for attributing the long-term changes in the releases of pollen by birches and grasses to meteorology and vegetation dynamics in Belgium. The pollen transport model is driven by ECMWF ERA5 meteorological data in a bottom-up emission approach for the period 1982-2019. The pollen emission maps make out the dynamic vegetation component and are based on merging multi-decadal datasets of spaceborne NDVI with forest inventory data and grass distribution maps for 1982-2019.

Temporal trends are computed using Theil Sen slopes and the Area Under the Curve (AUC) of the modelled seasonal birch and grass pollen cycles based on daily pollen levels, and of 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 coefficient.

Results show that for the period 1982-2019 the increasing radiation, the decreasing precipitation and the decreasing horizontal wind speed are associated with a strong increase in birch pollen levels. The decreasing grass pollen levels in the air between 1982 and 2019 are associated with decreasing precipitation. This is mainly induced by the decreasing trend in grass pollen sources. The associations between meteorology and airborne birch pollen levels are much stronger than for grass pollen. Birch pollen production dynamics and wind speed and precipitation contribute to the higher amount of birch pollen in the air. The inter-seasonal variations in birch pollen production dampen the overall increase rate by ~7%. The grass pollen production dynamics introduced in SILAM resulted in 3.5 times less airborne grass pollen levels over the studied period.

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## ITS1, ITS2, 5'-ETS, and trnL-F barcodes comparison for metabarcoding of the Poaceae pollen

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Grass pollen is one of the major causes of allergy, affecting 10-30% of the population around the globe. About 100 species of grass could be found in the European part of Russia, but the time of flowering and the allergenicity of different grass species are not the same.

The primary source of information on grasses' pollen content in the air is classical aerobiological monitoring, but microscopic pollen analysis does not allow identifying species and even genera within Poaceae because of the stenopalynous nature of the family. A possible solution to this problem could be the metabarcoding approach employing DNA barcodes for taxonomical identification of species in complex mixes by high-throughput sequencing of the pollen DNA.

To evaluate the possibility of a metabarcoding approach to discriminate species in airborne pollen mixes, we have selected 14 grass species widespread in Central Russia. Nuclear ITS1, ITS2, 5-ETS, and plastome trnL-F DNA barcodes of the selected species were Sanger-sequenced from live field and herbarium specimens and collected from GenBank, if available, to create a local reference barcode database. Novel Poaceae-specific primers for 5-ETS have been designed and tested to obtain a 5'-ETS region no more than 600 bp long, suitable for high-throughput sequencing. Barcode sequences were compared by intra- and interspecific distances, and barcoding gaps were measured. The ability of barcodes to correctly identify pollen mixes' composition was tested on artificial pollen mixes of various complexity using a subset of 7 reference grass species (*Calamagrostis epigeios*, *Phleum pratense*, *Bromus inermis*, *Festuca pratensis*, *Elymus repens*, *Alopecurus pratensis*, and *Lolium perenne*). We have optimized the DNA extraction method for single-species pollen samples and mixes to increase yield for better amplification and sequencing of pollen DNA.

Metabarcoding analysis of artificial pollen mixes showed that nuclear DNA barcodes proved to be more efficient than plastome one in PCR amplification and resolution of grass species on genus level. Although the barcodes demonstrated correct qualitative results of the composition analysis, the quantitative results rarely matched the actual ratio of pollen in the mix. Barcodes' ability to quantitatively predict correct pollen ratio in mix goes from best to worst in this order ITS1>ITS2>ETS>trnL-F.

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# Influence of atmospheric pollutants on pollen concentrations of Chenopodia-Amaranthaceae, Fraxinus and Myrtaceae

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Air plays an important role in the health of the population (World Health Organization, 2021). Airborne bioaerosols such as pollen grains cause pulmonary and cardiovascular diseases in allergic citizens, around 30% of the world's population (Brunekreef et al., 2000). Among the most allergenic pollen grains are those of the families of Poaceae, Oleaceae, Urticaceae, Compositae, and Chenopodia-Amaranthaceae. The allergenicity of these pollen types can be increased due to atmospheric pollution (Cuinica et al., 2015).

On the other hand, atmospheric compounds such as carbon dioxide can increase pollen production (Zhang et al., 2013), and pollutants as ozone, sulfur dioxide, carbon monoxide, nitrogen oxides, and particulate matter, as well as meteorological variables show effects on pollen concentrations, highlighting the importance of environmental conditions on pollen levels (Cariñanos et al., 2021). The aim of our study is to analyze the influence of O<sub>3</sub>, NO, NO<sub>2</sub>, NO<sub>x</sub>, SO<sub>2</sub>, CO, PM<sub>1</sub>, PM<sub>2.5</sub> and PM<sub>10</sub> on the concentrations of Fraxinus, Chenopodia-Amaranthaceae and Myrtaceae pollen types in the city of Badajoz (SW, Spain).

The concentrations of atmospheric pollutants were obtained from the studied city air quality monitoring unit from 2010 to 2019. The concentrations of the pollen types under study were determined using a volumetric sampler with the Hirst methodology. The relationship between atmospheric pollutants and pollen concentrations was studied through Spearman correlations, testing for a significance level of 95 and 99% with the R studio software.

The obtained correlations for the different pollen types vary depending on the pollutant. Ozone showed the highest correlation, being positive for Chenopodia-Amaranthaceae (0.42) and Myrtaceae (0.37) and negative for Fraxinus (-0.36). The positive and negative values obtained are in line with the literature (Oduber et al., 2019; Rahman et al., 2019). Nitrogen oxides had statistically significant negative correlations with Chenopodia-Amaranthaceae (-0.26 NO, -0.07 NO<sub>2</sub> and -0.12 NO<sub>x</sub>) and with Myrtaceae (-0.20 NO, -0.10 NO<sub>2</sub> and -0.13 NO<sub>x</sub>) positive correlations for the pollen type Fraxinus (0.24 NO, 0.19 NO<sub>2</sub> and 0.23 NO<sub>x</sub>), the latter being similar to a previous study (Puc, 2012). Particulate matter levels had statistically significant positive correlations for Chenopodia-Amaranthaceae and negative correlations for Fraxinus as well as the values published in the previous studies (Oduber et al., 2019; Puc, 2012; Rahman et al., 2019). Carbon monoxide levels had statistically significant positive values for Chenopodia-Amaranthaceae, similar to studies cited above. The results showed different influences of pollutants on pollen grain concentrations. In general, pollutants have a similar correlation on Chenopodia-Amaranthaceae and Myrtaceae pollen types but the opposite correlation on Fraxinus pollen types.

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## Keywords

Pollen, Pollutants and Correlation

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## High temporal resolution monitoring of *Ambrosia* pollen in ambient air

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The pollen grains of invasive *Ambrosia* species contain very potent allergens and can be transported vast distances when conditions are favourable. In this study we present the first airborne *Ambrosia* pollen measurements recorded by an automatic sampler with 1-hour and sub-hourly resolution (i.e. 1-minute and 1-second data).

The data were collected by traditional Hirst-type methods and a state-of-the-art Rapid-E real-time bioaerosol detector. Airborne pollen data, for Total Pollen and *Ambrosia*, were collected during the 2019 pollen season in Novi Sad, Serbia. Pollen data with daily, hourly and sub-hourly temporal

resolution were analysed in terms of their temporal variability. The impact of turbulence kinetic energy (TKE) on pollen cloud homogeneity was investigated.

Variations in Seasonal Pollen Integrals produced by Hirst and Rapid-E show that scaling factors are required to make data comparable. Daily average and hourly measurements of atmospheric Ambrosia pollen recorded by the Rapid-E and Hirst were highly correlated and so examining Rapid-E measurements with the sub-hourly resolution is assumed meaningful from the perspective of identification accuracy. Sub-hourly data provided an insight into the heterogeneous nature of pollen in the air, with short peaks lasting max. 11 minutes, and mostly single pollen grains recorded per second. Short-term variations in 1-minute pollen concentrations could not be wholly explained by TKE.

The new generation of automatic devices has the potential to increase our understanding of the distribution of bioaerosols in the air, provide insights into biological processes such as pollen release and dispersal mechanisms, and allow us to conduct investigations into dose-response relationships and personal exposure to aeroallergens.

**Keywords:** airborne pollen, Ambrosia, high temporal resolution, 1-minute data, 1-second data, laser spectroscopy

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## Disparity between olive fruit production and pollen integrals in Malaga (southern Spain)

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Olive trees usually have an alternate bearing in their fruit production. This phenomenon consists of alternating high and low fruit productivity between consecutive years and, usually, it is caused by the high demand for nutrients that the plants require to fructify. This biannual cycle constitutes a major economic problem for olive growers and, consequently, a lot of interest is put in elaborating forecasts models of the annual olive production.

In Malaga (southern Spain), from 1992 to 2010 there was a strong correlation between the annual pollen integral and the annual olive fruit production but, since 2011, this correlation was no longer detected. Most forecast models elaborated for predicting olive production are based on the pollen integrals detected during the spring prior to harvesting, but it is expected that these models diminish their accuracy rates due to these changes.

In this work, we studied the trends in the alternate bearing followed by both the olive airborne pollen and the fruit production, as well as the main environmental and human factors that could be conditioning them.

In fact, we observed that the implementation of different agricultural techniques together with climate change could be the reasons behind the interruption in the pollen alternate bearing. These variables should be integrated into the prediction models of olive production in order to maintain their accuracy rates.

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## Achievements and perspectives of aerobiology in the Spanish Aerobiology Network (REA)

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The Spanish Aerobiology Network (REA) was funded in 1992 (<http://www.uco.es/rea/>). This is an academic-research network, comprising teaching and/or research staff specialising in different scientific areas, i.e., Botany, Mycology, Palynology and Atmospheric Dynamics. This is a Technical Network within the Spanish Aerobiology Association (AEA) since the Association was formed in 1995 (<http://www.aea.aerobiologia.org/>). The REA is involved in the European Aeroallergen Network (EAN) <https://ean.polleninfo.eu/Ean/>, at the HNO-Klinik, University of Vienna, Austria. On the other hand, REA is based on a federated network where different regional networks are working closely with the Health or Environment Councils [https://www.uco.es/investiga/grupos/rea/?page\\_id=22](https://www.uco.es/investiga/grupos/rea/?page_id=22): Andalusia Aerobiology Network (RAA), Aragon Aerobiology Network, Asturias Aerobiology Network, Balearic Islands Aerobiology Network, Cantabria Aerobiology Network, Castilla la Mancha Aerobiology Network (AEROCAM), Castilla y Leon Aerobiology Network (RACYL), Cataluña Aerobiology Network (PIA), Extremadura Aerobiology Network (AeroUEX), Galicia Aerobiology Network (RIAG), Community of Madrid (PALINOCAM) Aerobiology Network, Region of Murcia Aerobiology Network, Navarra Aerobiology Network, Aragon Aerobiology Network and País Vasco Aerobiology Network.

From the beginning, REA has been involved in the development of a standardised methodology, following the EAN Minimum Requirement in the methodology for Routinely Performed Monitoring of Airborne Pollen Recommendations (Jäger, 1995). This methodology has been validated and published in different papers that have also supported the publication of the REA Management and Quality Manual (Galán et al., 2007). REA is also involved in different quality control inter-laboratory surveys for proficiency testing, trying to determine the performance of technician staff for reading slides (Oteros et al. 2013), and has been involved in different inter-laboratory surveys and exercises, in the frame of the Working Group of Quality Control from the European Aerobiology Society (EAS) <http://www.eas-aerobiology.eu/>. Results from these external exercises have made possible the publication of different papers for improving Quality Control, i.e., the minimum requirements and reproducibility of pollen (Galán et al 2014) and fungal spores (Galán et al 2021) monitoring in Europe; inter-laboratory ring tests for counting pollen with similar morphology, as an education process for aerobiologists (Sikoparija et al 2017); and, recently, the use of virtual slides to facilitate it (Smith & Sikoparija, 2020). Members of REA are also members of the Air Quality working group of the Spanish Standardization Association (UNE), being involved in the French Standardization Association (AFNOR) for working in the Working Group on Legislation. Today we count with the European standard norm on Ambient Air- sampling and analysis of airborne pollen grains and fungal spores for networks related to allergy- Volumetric Hirst method (EN 16868) and starting to work on a Technical Specification (TS) to define the standard, requirements, and procedures for pollen and fungal spores automatic monitoring.

REA members use the Hirst type volumetric spore trap (Hirst 1952) for pollen and fungal spore monitoring, following the minimum requirements proposed by the EAS and EAN. Members collect the samplers on Monday and send the data on Wednesday to the coordinating center, located in the University of Cordoba, Spain. Data are automatically entered into the National Database, using a file created for each sampling site. Data is disseminated on the web <https://www.uco.es/rea/>, and in the App "Polen REA" (in Spanish) [https://www.uco.es/investiga/grupos/rea/?page\\_id=310](https://www.uco.es/investiga/grupos/rea/?page_id=310), which can be downloaded by Google Play and by Apple Store. Authorized staff at the REA Coordinating Centre send the data to the European Aeroallergen Network; this information is weekly updated at [www.polleninfo.org](http://www.polleninfo.org). Members from the Coordinating Centre have also participated in the Spanish translation for the Pollen-App <https://pollendiary.com/Phd/>, where allergy patients can record their daily allergy symptoms and these data can be compared with the actual pollen concentrations of the main allergenic plants.

REA has developed a model for pollen forecasting. This is a statistical model using real pollen data assimilation for continuous execution with daily surface concentrations. It uses time series analysis in combination with the influence of different meteorological parameters, such as precipitation, to predict atmospheric pollen in the short term. Also uses different geostatistical techniques

and climatic influence to calculate the concentration of pollen in the air at any point in our geography. REA also participates in both, training and validation, through the European Aeroallergen Network (EAN) for other European models, e.g., SILAM, COPERNICUS models... Today, REA belongs to the AutoPollen Program, in agreement as a third party, and the coordinator is chair of the WG on Quality Control. Nowadays some REA members are working on automatic pollen and spore monitoring using instruments based on different technologies, i.e., image recognition or fluorescence. It is a new step in REA to participate in the future European automatic pollen monitoring network using high temporal-resolution real-time measurements (AutoPollen: <https://www.eumetnet.eu/activities/miscellaneous/current-activities-mi/autopollen/>) Today the REA has the status of a National Pollen Allergy Prevention Service. However, even when the main research application is focused on atopy or asthma, some research projects are related to agriculture, forestry, green urban spaces, and modelling.

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## Patterns of sensitization to birch pollen of children in Ukraine

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Pollen of allergenic species is an important issue in the healthcare system in Ukraine. It is especially important in cities and suburbs as potential sources of allergenic pollen. What is more, allergenic tree species were planted in the urbanized territories before and their planting continues, despite the potential ability of their pollen to cause allergies. Thus, at the moment, large territories are occupied by birches, which are known as an important source of allergenic pollen. What is more, birches are grown at kindergartens and schools and on their territories too. This, in turn, provokes sensitization to birch pollen allergens from early childhood.

However, it is well-known that people who are sensitive to birch pollen may develop allergies to the pollen of other tree species of the Betulaceae family and to some foods (so-called “pollen-fruit syndrome”) over time. Consequently, the symptoms of pollinosis may begin before the flowering of the birch starts and continue after it completes.

“Pollen-fruit” syndrome caused by the Bet v 1-related allergens the most likely associated with the consumption of Rose family fruits: apples, peaches, plums – and vegetables of the carrot family. This condition is manifested by the syndrome of oral allergy.

To determine the level of sensitization of the child population to birch pollen allergens, we analysed the results of molecular diagnostics obtained using the Alex test in 2018-2020. The analysis included data from 8016 patients, including 3549 children under the age of 10.

Results: Analysis of the results showed that 27.07% of these children had sensitization to birch pollen with IgE antibody concentrations of 0.30 kU/l and above.

By age, this sensitivity was distributed as follows. Among children sensitized to birch pollen, children under 1 year of age constituted 0.42 %, children aged 1 year – 1.14 %, 2 years – 4.58 %, 3 years – 7.28 %, 4 years – 10.09 %, 5 years – 11.86 %, 6 years – 14.88 %, 7 years – 13.01 %, 8 years – 12.59 %, 9 years – 12.38 % and 10 years – 11.76 %.

Based on these data, we see a trend towards an increase in the sensitivity of children to birch pollen. From birth to 6 years of age, the number of sensitive individuals constantly increases until this indicator reaches a plateau (7-10 years).

It is also necessary to take into account the physical and geographical zoning of Ukraine. The lowest number of children sensitive to birch pollen was found in the south of Ukraine in the Steppe zone (Odesa – 11.11 %, Kherson – 14.58 %) and in the forest zone in the Ukrainian Carpathians (Ivano-Frankivsk – 12.12 %, Lviv – 18.63%). This can be explained by the fact that there is no large number of natural arboreal associations in the Steppe. Concerning the Carpathians, other representatives of the flora predominate there. They are oak, beech, hornbeam, fir, spruce, and pine. The exception to this tendency is the city of Dnipro, located in the Steppe zone, where 28.59 % of children were sensitive to birch pollen. We believe that the reason for this is the artificial plantation of birch for city greenery.

The highest sensitivity was found in cities located in the zone of Mixed forests (Zhytomyr – 42.11%, Kyiv – 32.31%) and in the Forest-steppe zone (Kharkiv – 27.69%, Ternopil – 26.32%). Both territories are the natural area of birch.

Conclusions: Our results suggest that in cities, especially those located in the birch natural area, it is necessary to control the artificial planting of birch. It is especially important for the places of children activity. Such places, in particular, include the territories of kindergartens, schools, playgrounds, and areas around them.

It is necessary to look for the safe alternative of allergenic plants for city greenery and to inform the public about the high probability of developing pollen allergy and the “pollen-fruit” syndrome following primary sensitization to birch pollen.

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## Fungal spores as autumn allergens in Ukraine

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Intermittent allergic rhinitis has very obvious seasonality, which is caused by the periodical nature of the vegetative season of plants and fungi. It is also known that despite a drastic decrease in the pollen concentrations in autumn, symptoms of allergy and asthma persist during the autumn period too. It can be explained by the relatively high concentrations of fungal spores, which are observed in autumn. To prove this hypothesis and to evaluate the possible impact of climate change on the levels of airborne fungal spores in autumn we analyzed the seasonal dynamics of sporulation of various fungi and investigated the diversity and concentration levels of airborne fungal spores in order to improve the accuracy of allergy forecasts.

The study was performed at the National Pirogov Memorial Medical University, Vinnytsya, Ukraine (VNMU), from 2011 to 2021. Fungal spores were collected using a Burkard impact pollen trap of a Hirst type located at the roof of the VNMU at the 25 m altitude. Slides were read by 3 horizontal

transects in the years 2009-2011 and by 12 vertical transects in the years 2012-2020 under magnification of 400X.

Sporulation of fungi of Oomycota (Peronospora), Ascomycota (Alternaria, Cladosporium, Epicoccum, Leptosphaeria, Stemphylium, Pithomyces, Pleospora, Periconia, Helminthosporium) and Basidiomycota Divisions, namely Agrocybe, Coprinus, Fusarium, Ganoderma, Puccinia, and other Uredo-, Ustilago- and other unclassified Basidiospores was taken into account.

The study revealed that aggregated concentrations of these spores were the highest in the middle and end of summer. But in autumn, when there is not much pollen in the air, reproduction of most fungi continued with relatively high concentrations.

This pertains to most Basidiospores, which are known by the formation of their sporulating fruiting bodies in autumn. Namely, stable high concentrations in autumn are observed for Agrocybe, Coprinus, Uredinales, Ustilaginales, and unclassified Basidiospores. Their concentrations fluctuate between a couple of dozens to 400 hundred spores / m<sup>3</sup>.

Among Ascospores the most numerous was Cladosporium with concentrations exceeding 3000 spores/m<sup>3</sup>. Concentrations of around 100 spores / m<sup>3</sup> were common in autumn for Alternaria, Epicoccum, Periconia, Stemphylium were also continuously present but the concentration of it was low – around 10 spores / m<sup>3</sup>.

Continuous sporulation of spores lasts until the end of the sampling period, which stops at the end of the first 10-day period of November.

1. Climate change leads to changes in the length of the fungal season of vegetation.
2. Spore load is observed in the autumn period, which makes fungal spores a specific agent of the airborne respiratory diseases at that time, especially at the background of low pollen concentrations.
3. This information should be considered while forecasting seasonal allergy symptoms in autumn.

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## Utility of pollen information searchers in internet in the context of prevention of seasonal allergy

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One of the most common allergens in Ukraine is ragweed pollen. It intervened here from the American continent a hundred years ago. The most ragweed-contaminated regions are Kherson, Odessa, Mykolaiv, Zaporizhia, Donetsk, Dnipropetrovsk, and Kirovohrad regions. According to our preliminary data, approximately 40% of people with hay fever are susceptible to ragweed. And it is believed that this percentage will increase. This is associated with the deterioration of ecological and climatic conditions, which, in turn, impair the immunity of the population. Also, climate changes and air pollution increase the allergenicity of pollen and promote the expansion of ragweed over the Ukrainian territory.

To determine, the population of which regions are most likely to suffer from allergy symptoms during the ragweed pollen season and how these symptoms may coincide with ragweed pollination timing, we analyzed queries by words “ragweed” on Internet resources, including Google Trends of Google Analytics, as well as on the Allergy.org.ua website – according to the word “allergy forecast”. The information was collected for the period 2017-2021 years.

To determine the seasonality and peak periods of Ambrosia pollination, data on the airborne ragweed pollen concentrations in Ukraine, obtained by the Laboratory for the Allergenic Environmental Factors Investigation of the National Pirogov Memorial Medical University, Vinnytsya, were used.

According to aerobiological data, there are two peaks of activity for ragweed in Ukraine: mid-late August and early-mid-September. These peaks coincided with the increase in the number of requests for ragweed information on Internet resources, including Google Trends and the website Allergy.org.ua.

According to the analysis of search engine queries, the following data were obtained. The peak of the requests for information about ragweed and allergic reactions to it occurred in 2017 on August 23-29 and September 3-9; in 2018 – on August 26 – September 1; in 2019 – on August 4-10 and on August 18-24; in 2020 – on August 23-29 and on August 30-September 5; in 2021 – on August 15-21 and on August 29-September 4. This coincides with the peak periods of ragweed pollination.

Also, the peak of inquiries for pollen forecast traditionally was the highest during the first week of September, which reflects the importance of ragweed allergy in Ukraine.

The highest number of inquiries regarding the word “ragweed” in Ukrainian (more than 50 % of all) was observed in Odesa (it took 100 %), Kyiv, Mykolaiv, Lugansk, Kherson, Cherkasy, Sumy, and Kharkiv regions. Requests in Russian reflected the same picture. It coincides with the known excessive spread of Ambrosia in these regions, except Cherkasy and Kyiv. The rise in the number of requests on Ambrosia in these 2 regions may reflect both active invasion of ragweed to the Center and, consequently, North of Ukraine and the suffering of people from seasonal allergy, which is not necessarily caused by ragweed in the summer-autumn period.

Notably, interest in Ambrosia in Zakarpattya, the most Western region of Ukraine, was higher than in other Western regions. It can be explained by the ragweed infestation of Zakarpattya from Hungary.

The lowest number of requests were seen in Western regions, which are not infested by ragweed, and in Donetsk regions, which are heavily occupied by Russia. This is despite the fact of known areas of excessive ragweed infestation in this region.

Based on the data obtained, it can be concluded that the population of the South and East of the country is more interested in ragweed topic than other citizens. This may coincide with the known areas of ragweed in Ukraine.

And the timing of requests coincides with the peak periods of ragweed pollination too.

The increase of requests for ragweed in Cherkasy and Kyiv may reflect the migration of Ambrosia from the South through Cherkasy to the capital of the country by water route as Cherkasy connects Kyiv with the Black Sea shore by the river Dnipro.

To prevent allergic reactions to ragweed pollen, quarantine measures should be taken to reduce the spread of these plants; improve public and health information.

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