

Air Pollution by Pollen Grains of Anemophilous Species: Influence of Chemical and Meteorological Parameters

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Abstract One of the most important particles of biological origin present in the air is pollen grains of plants. Having basic biological function in the process of pollination, pollen grains of some plant species can cause allergic reactions among 20–30 % of the human population and thus affect their health and overall quality of life. Bearing in mind the potential influence air pollutants and meteorological parameters may have on release of pollen and granules of allergen from pollen, concentrations of air pollutants and 26 different anemophilous aeropollen types as well as meteorological parameters were established in a 5-year period (2009–2013) in Subotica, Northern Serbia. Spearman's rank correlation was made for statistical analysis of relationships between concentration of some air pollutants (sulphur dioxide, nitrogen dioxide, soot, particulate matter $(PM)_{10}$ and $PM_{2.5}$), meteorological factors (temperature of air, humidity, wind speed, cloud index) and airborne pollen. In most of the examined years, significant positive correlations were determined between temperature and total pollen concentration, while significant

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negative correlations were established between humidity as well as cloud index and total pollen concentration, clearly proving the influence these meteorological parameters have on pollination of all examined species.

Keywords Pollen \cdot Air pollutants \cdot Meteorological parameters. Spearman correlation

1 Introduction

The largest quantity of pollen grains suspended in the air are derived from anemophilous plant species pollinated by the wind. These plants produce pollen grains in a large number in order to increase their contact with the female flowers. When the pollen is released into the air, it is carried by wind from few hours to few days. The size of pollen ranges from 5 to 200 μm for the largest grains.

Interactions between pollutants of anthropogenic origin as well as meteorological conditions and biological material suspended in the air have not been fully investigated yet. Dependence of air pollution and meteorological parameters on the one side and suspended pollen in the air on the other side provide the basis for assumption of atmospheric pollen as a good bioindicator of pollution as well as physical and chemical quality of air (Cerceau-Larrival et al. [1996\)](#page-11-0). Because of specific ornamentation of pollen exine and its lipophilicity, pollen is a very good accumulator of all types of pollutants: gaseous and suspended particulate matter as well as different organic and inorganic compounds (Garrec [2006\)](#page-11-0). Accumulation itself depends on physical and chemical processes that occur on the surface of the pollen grain. The results of studies conducted in Spain (Brito et al. [2007](#page-11-0)) showed that people allergic to pollen, living in heavily contaminated regions, had more pronounced symptoms than those living in rural areas, and also led to a conclusion that other factors, including air pollutants adsorbed on the pollen particles, may increased sensitivity to aeroallergenic pollen.

About 99 % of atmospheric sulphur dioxide comes from anthropogenic sources. The main sources of sulphur dioxide in the air are traffic, industrial activities that use fossil fuels containing sulphur and domestic heating based on burn coal. The retention time of sulphur dioxide, a precursor of chemical smog, in the atmosphere is between 4 and 10 days. Nitrogen dioxide is a highly reactive gas which is formed during combustion processes at high temperatures and comes principally from motor vehicle exhaust and stationary sources such as electric utilities and industrial boilers. This gas can react in the air to form corrosive nitric acid as well as toxic organic nitrates. It also contributes to the formation of photochemical smog.

Sousa et al. ([2012](#page-11-0)) have been investigated the effects of sulphur dioxide and nitrogen dioxide on the protein content and germination rate of Acer negundo pollen. The results showed that protein content was lower in sulphur dioxide-exposed pollen samples and slightly higher in nitrogen dioxide-exposed pollen samples compared to the control sample. Their results also indicate that concentrations of sulphur dioxide and nitrogen dioxide in urban areas may cause an increase of pollen production (although this increase is not necessarily accompanied with increase of protein content, as stated before) as well as changes in the chemical composition of the compounds in pollen grains.

Nowadays, the particulate matter is among the most dangerous air contaminants—not only from the standpoint of public health. Its particle size and chemical composition are the most important parameters in the interaction of aerosols with different environmental components. Distribution of particle size depends on both the microclimate of the area and the season. Relatively recent study demonstrated that pollen from polluted regions is covered with pollutant particles, and that fact might alter their allergen content (Adhikari et al. [2006](#page-11-0)). Knox et al. ([1997](#page-11-0)) showed that grass allergen molecules tend to bind to micrometre-sized aggregates formed by soot particles, so that particulate matter can act as a carrier of allergens and could bind with airborne pollen (Risse et al. [2000](#page-11-0)). It is estimated that 70 % of the particle matter from the air can be settled by gravity, precipitation or incorporation into drops as condensing centres. One portion of atmospheric particles have hygroscopic surface and, in contact with water vapour, can dissolve if particle matter is soluble. Pollen showed enhanced mass growth at all relative humidity values larger than 0 % (Pope [2010](#page-11-0)). At 75 % of relative humidity, an average value of the mass increase due to water uptake was 16 %. Based on the results of concentration of phospholipids, Womiloju et al. ([2003](#page-11-0)) reported that the cell materials of pollen contribute 4–11 % of the total particulate matter $(PM)_{2.5}$ mass.

The study from Szczecin (Poland) showed that there were significant correlations between the various meteorological factors (air temperature, humidity, wind speed, cloud index), air pollution parameters $(PM₁₀$, sulphur dioxide concentrations) and grass pollen (Puc [2011](#page-11-0)). Meteorological conditions are one of the main factors modifying pollen season, but they are not the only ones (Piotrowska and Kubik-Komar [2012](#page-11-0)). Air temperature was the most important meteorological parameter that correlated positively to daily pollen concentration increase. An increase in relative humidity and precipitation was usually related to a decrease in airborne pollen content in research that was conducted from 2008 until 2010 with a smaller number of pollen species than in our investigation (Aboulaich et al. [2013\)](#page-11-0). From statistical analysis of the correlations between meteorological factors and the start date of the birch pollen season in Lublin (Poland) in the period 2001– 2010, it can be concluded that low temperatures in February promote the occurrence of high pollen concentrations (Piotrowska and Kubik-Komar [2012](#page-11-0)). However, it was also found that the earlier start of the birch pollen season in many cities across Europe was associated with increased temperatures in the months between January and March (Spieksma et al. [2003\)](#page-11-0).

The aim of this study was to present and compare the pollen concentrations of 26 different anemophilous aeropollen species with concentrations of air pollutants and meteorological parameters in the atmosphere of Subotica (Serbia) during the 5-year period, in order to establish possible long-term influence that selected air pollutants and meteorological parameters may have on concentrations of pollen in the air. To the best of the authors' knowledge, this is the first study combining an influence to a large number of different pollen species during relatively long time frame (5 years).

2 Experimental

The research was conducted in Subotica, a city located in the Pannonian Basin, in Northern Serbia. With a population of almost 120,000 inhabitants, Subotica could be considered as a semi-rural environment, since most of the inhabitants live in family houses surrounded by plenty of green areas and agriculture fields. Air samples collected between 2009 and 2013 were analysed in Public Health Institute in Subotica in accordance with ISO standard methods [\(ISO 6767:1990;](#page-11-0) [ISO](#page-11-0) [9835:1993;](#page-11-0) [ISO 6768:1998](#page-11-0); [EN 12341:1998](#page-11-0); [EN](#page-11-0) [14907:2005](#page-11-0)). The measured parameters of air pollution were sulphur dioxide, nitrogen dioxide, soot, PM_{10} , PM_{2.5} and total pollen. Blanks as well as duplicates were also analysed throughout the years.

Sampling of gaseous pollutants were performed by appliances for air sampling (Proekos AT-801-2X and Proekos AT-801-2BP), absorbing contaminants from a known volume of air in a suitable absorption solution. Soot samples were obtained by filtering a known volume of air through the appropriate filter paper. Reflectometric measurement of soot index was done on a reflectometer (Proekos RM-02). Samples of suspended particles were taken using an ambient air sampler (Sven Leckel LSV 3), and samples of pollen were taken by 7-day pollen trap (Burkhard).

The sampling and measurement site was located in the city centre (46° 10′ 47″ N, 19° 66′ 80″ E). Regular daily measurements were performed every day (sulphur dioxide, nitrogen dioxide and soot), from February to November for qualitative and quantitative analyse of pollen and periodically for PM_{10} and $PM_{2.5}$ for the period of 5 years (2009–2013). The meteorological data (temperature of air, humidity, wind speed, cloud index) were provided by a weather station located in Palić, near Subotica.

Total pollens were studied by counting of 26 different anemophilous pollen types (Table [1](#page-3-0)): Acer, Alnus, Ambrosia, Artemisia, Betula, Cannabaceae, Carpinus, Corylus, Chenopodiaceae/Amaranthaceae, Cyperaceae, Fagus, Fraxinus, Juglans, Moraceae, Pinaceae, Plantago, Platanus, Poaceae, Populus, Quercus, Rumex, Salix, Taxus, Tilia, Ulmaceae and Urticaceae. The beginning of pollen season with trees flowering in early spring depends mainly on the nonlinear equilibrium between cold winter and warmer spring temperatures needed for breaking the winter dormancy (Emberlin [2003\)](#page-11-0).

SPSS 12.0 application was used for statistical analysis. The degrees of correlation between pollen data with meteorological parameters and air pollution parameters were described using Spearman's rank correlation coefficient. Due to the fact that data set was not normally and linearly distributed, a non-parametric method was used in order to show whether the two set of parameters were covariate or independent.

3 Results and Discussion

The results obtained for air pollution in Subotica for the examined period are presented in Table [2](#page-4-0) (sulphur dioxide, nitrogen dioxide, soot, PM_{10} and $PM_{2.5}$ with minimum, maximum, average and median values and number of days), while the processed parameters for air quality for the period 2009–2013 are shown in Fig. [1](#page-5-0) (sulphur dioxide, nitrogen dioxide, soot, total pollen).

The amount of sulphur dioxide in the air of Subotica is acceptably low since the maximal daily value was 34 μ g/m³ (24-h EU air quality standard is 125 μ g/m³). In 2009, the annual average concentration of sulphur dioxide in the air was 10 μ g/m³, while in 2013, it was under the limit of quantification (Fig. [1\)](#page-5-0). The observed decrease is mainly due to the reduction in sulphur dioxide emissions in neighbouring EU countries (Hungary, Croatia) resulting from their regulations on sulphur content in fuels.

Monthly variation patterns of nitrogen dioxide during 5 years of the study (Fig. [2\)](#page-6-0) show maximal values during late spring and summer. Daily variations are significant, with peaks occurring during traffic rush hours. With increased concentrations of nitrogen dioxide, sufferers from pollen allergy usually find their symptoms getting worse [\(www.aha.ch/swiss-allergy](http://www.aha.ch/swiss-allergy-centre/info-on-allergies/allergies-en/pollen-allergy/pollen-and-airpollutants)[centre/info-on-allergies/allergies-en/pollen-allergy/](http://www.aha.ch/swiss-allergy-centre/info-on-allergies/allergies-en/pollen-allergy/pollen-and-airpollutants) [pollen-and-airpollutants](http://www.aha.ch/swiss-allergy-centre/info-on-allergies/allergies-en/pollen-allergy/pollen-and-airpollutants)). Annual mean concentrations of nitrogen dioxide in Subotica during 2009–2013 generally do not exceed 90 μ g/m³. Increase of concentrations during the observed period can be attributed to a lack of reduction in nitrogen dioxide emissions from vehicles. From 1116 days of total count during 2009–2013, there were 191 days or 17 % when the measured concentrations of nitrogen dioxide were higher than daily allowed value of 50 μ g/m³ (Ambient Air Quality Directive [2008/](#page-11-0)50/EC).

Concentrations of soot were high throughout the observed period. Average daily values of soot, PM_{10}

Table 1 Registered types of pollen in the province of Vojvodina (Igić et al. [2012](#page-11-0))

Table 2 Processed parameters for air quality for the period 2009– 2013

		Minimum Maximum Average Median Count ^a	values	values	
$SO_2 \left(\mu g/m^3\right)$					
2009	1	26	10	9	188
2010	$\overline{0}$	34	5	4	228
2011	$\overline{0}$	23	$\overline{2}$	1	237
2012	$\mathbf{0}$	12	1	1	241
2013	θ	10	θ	$\mathbf{0}$	243
2009-2013	θ	34	3	1	
$NO2 (\mu g/m3)$					
2009	6	63	34	34	
2010	1	59	29	30	231
2011	1	63	28	27	242
2012	$\overline{0}$	88	31	30	237
2013	$\overline{0}$	90	32	31	229
2009-2013	θ	90	31	30	
Soot $(\mu g/m^3)$					
2009	0	157	39	35	
2010	θ	103	35	33	227
2011	$\mathbf{0}$	123	33	28	239
2012	θ	125	21	15	237
2013	$\overline{0}$	124	22	16	223
2009-2013	$\mathbf{0}$	157	30	26	
$PM_{10} (\mu g/m^3)$					
2012	13	65	33	29	
2013	10	81	36	33	32
2009-2013	10	81	35	31	47
$PM_{2.5} (\mu g/m^3)$					
2012	6	233	33	18	15
2013	3	210	33	17	34
2009-2013	3	233	33	17	49

a Number of days

and $PM_{2.5}$ concentrations for the 5 years were 30, 35 and 33 μ g/m³, respectively. During the period observed in this experiment, there were 7 out of 47 days in which daily PM_{10} concentration exceeded the daily standard of 50 μg/m³ (Ambient Air Quality Directive [2008](#page-11-0)/50/EC).

Figure [2](#page-6-0) shows two maximums of total pollen concentration during the 5 years: on spring (March–May) and late summer (first decade of September) due to flowering of the largest number of trees species during spring and *Ambrosia*, which produces an enormous number of pollen grains, during summer. In Table [3,](#page-6-0) basic aerobiological parameters during 2009–2013 are shown, while annual changes of measured parameters are presented in Fig. [3.](#page-7-0) The meteorological parameters are variable, and their values depend on a large number of the processes in the atmosphere that thus influence their mean and seasonal values as well. Average annual meteorological parameters during 2009–2013 are shown in Table [4](#page-7-0).

To examine the interaction between total pollen concentrations and different parameters examined in this research, we used Spearman's rank correlation for statistical analysis of these relationships. Spearman's correlation parameters between concentrations of total pollen and meteorological parameters are shown in Table [5,](#page-8-0) while Spearman's correlation parameters between concentrations of total pollen and concentrations of air pollutants are shown in Table [6](#page-8-0).

As it can be seen in Table [5](#page-8-0), there are strong significant correlations between most of examined physicochemical and meteorological parameters and total concentrations of investigated pollen species in the air in the investigated years. In all examined years (2009–2013), there were positive correlations between the values of air temperature and total pollen concentrations. In years 2010, 2011 (Fig. [4a](#page-9-0)), 2012 and 2013, these correlations are significant $(p<0.01)$, while in year 2009, the correlation is still positive and of high significance but lower than the threshold value. Unlike temperature, the values of humidity and cloud index are negatively correlated with the values of total pollen concentration. The values of significance of correlations are high ($p \le 0.01$) in cases of correlations of humidity and total pollen concentrations in all examined years (correlation graph for year 2013 is shown in Fig. [4b](#page-9-0)). In the case of cloud index, these correlations are significant in all but the year 2012 (correlation graph for year 2013 is shown in Fig. [4c\)](#page-9-0), when, although the correlation is still negative, it is not as highly significant as in the other examined years. When it comes to correlations between the values of wind speed and total pollen concentrations, they are ambiguous, since the negative (and significant) correlations were established in 2010, 2011 and 2013, while positive correlations were established in 2009 and 2012. The information obtained from statistical analysis clearly proves that an increase of temperature increases the quantity of total pollen in the air and that this connection is relatively strong in the case of all, or almost all, species (and thus in their sum) in which pollen was examined. Totally opposite can be stated for

Fig. 1 Values of sulphur dioxide, nitrogen dioxide, soot and total pollen during 5 years of the study

the influence of humidity and cloud index, an increase of which decreases as a rule the concentration of the pollen in the air and it does so in cases of all, or almost all, investigated species.

The linear regression analysis was used for determination of trends in average over time and for description of direction of the trend. It is useful because it gives the proportion of the variance of one variable that is

Fig. 2 Monthly variation patterns of sulphur dioxide, nitrogen dioxide, soot, PM_{10} , $PM_{2.5}$ and total pollen during 5 years of the study

predictable from the others. The obtained results for air pollutants (Table [7\)](#page-9-0) show constant decreasing trends for three measured parameters (sulphur dioxide, nitrogen dioxide and soot).

When correlation coefficients (r) and significances of correlations (p) between physico-chemical parameters (concentrations of sulphur dioxide, nitrogen dioxide, soot, PM_{10} and $PM_{2.5}$) and pollen concentrations were

Table 3 Pr biological p 2009–2013 2009–2013

Fig. 3 Annual variation pattern of sulphur dioxide, nitrogen dioxide, soot, PM₁₀, PM_{2.5} and total pollen during 5 years of the study

Table 5 Spearman's correlation coefficients between the concentrations of total airborne pollen and meteorological parameters during 2009–2013

 $*_{p<0.03}$, significant coefficients; **p<0.01, significant coefficients

Table 6 Spearman's correlation coefficients between the concentrations of total airborne pollen and measured parameters of air pollution during 2009–2013

 $*_{p<0.03}$, significant coefficients; **p<0.01, significant coefficients Fig. 4 Correlation graphs showing the correlation between concentration of total pollen and air temperature in 2011 (a), concentration of total pollen and humidity in 2013 (b) and concentration of total pollen and cloud index in 2013 (c)

examined (Table [6\)](#page-8-0), it was shown that there were no significant correlations in most of the examined years, or that even, in some cases when significant correlation existed, they were of different nature in examined years. For example, in the case of a correlation between concentration of sulphur dioxide and concentration of total pollen, out of five examined years, positive correlations were found in 2009, 2010 and 2012 (though only one with $p \le 0.01$ was in 2012), while negative correlations were established in 2011 and 2013 (in 2013 with $p \leq$ 0.01). Similar but not the same trend was found in the correlation between concentrations of nitrogen dioxide and total pollen: three of them were positive (in 2009 and with $p \le 0.01$ in 2010 and 2013), and two were negative (in 2012 and with $p \le 0.05$ in 2011). There was only one significant correlation between concentration of soot and total pollen: negative one in 2013, while in other years, they were positive (2010 and 2011) or negative (2009 and 2012) but not significant (with $p \leq$ 0.05). Finally, both PM_{10} and $PM_{2.5}$ values had such concentrations that, when correlations with concentrations of pollen were performed, they gave significant

Table 8 Values of meteorological parameters significantly correlated with the concentration of total pollen, obtained from correlation equations, at concentration values of pollen equal to zero

Year parameter	2009	2010	2011	2012	2013
Temperature $(^{\circ}C)$	19.7	14.7	14.2	16.6	12.1
Cloud index	3.2	4.5	3.6	3.1	4.3
Humidity $(\%)$	65	77	73	66	73

negative results in 2013 but positive and with significance $p \le 0.05$ in 2012. For all of the measured parameters (wind speed and all measured air pollutants: sulphur dioxide, nitrogen dioxide, soot, PM_{10} and $PM_{2.5}$) that gave ambiguous statistical results (in some years, positive. in the other, negative) when correlated with concentrations of pollen in examined years, the conclusion can be drawn that, even if there was a dependence between the examined parameters and the concentration of pollen, these dependences were of different nature between measured parameters and specific pollen species. As a result, correlations between total pollen concentrations and mentioned parameters were either not significant or are positive in one and negative in other year, depending on the contribution of specific species (that can be larger in one or almost non-existing in other year) out of 26 examined.

Finally, it should be noted that when coefficient values from correlation equations describing correlations between three parameters that are undoubtedly significantly correlated with concentrations of total pollen (air temperature, humidity, cloud index) are extracted and compared (Table 8), another conclusion can be drawn. Values of air temperature that are related to total pollen concentration equal to zero in 2009 and 2012 are higher, while the similar values of cloud index and humidity are lower than the same values in the other examined years. Since 2009 and 2012 were the years with much higher precipitation than the other three examined years, this clearly proves than the level of precipitation influences the dependence between total pollen on the one hand and temperature, humidity and cloud index on the other hand.

4 Conclusion

In order to establish the potential influence of some air pollutants (sulphur dioxide, nitrogen dioxide, soot, PM_{10} and $PM_{2.5}$) and meteorological factors (temperature of air, humidity, wind speed, cloud index) at concentrations of total pollen of different anemophilous aeropollen types (Acer, Alnus, Ambrosia, Artemisia, Betula, Cannabaceae, Carpinus, Corylus, Chenopodiaceae/Amaranthaceae, Cyperaceae, Fagus, Fraxinus, Juglans, Moraceae, Pinaceae, Plantago, Platanus, Poaceae, Populus, Quercus, Rumex, Salix, Taxus, Tilia, Ulmaceae and Urticaceae), concentrations of examined air pollutants, total pollen and meteorological parameters were established for 5 years (2009–2013) in Subotica, Northern Serbia (46° 10′ 47″ N, 19° 66′ 80″ E). Correlation analysis proved strong dependence of total pollen concentration on air temperature (the pollen concentration increases with the increase of air temperature), humidity and cloud index (the total pollen concentration decreases with the increase of humidity and cloud index). Since the total measured pollen concentration consists of pollen of 26 different species, it may be assumed that air temperature, humidity and cloud index are influencing all of them (or almost all of them) in the same manner. The amount of rain in the year does not influence these dependences and significances of correlation but does so with the values of coefficients in correlation equations. When it comes to examined air pollutants (sulphur dioxide, nitrogen dioxide, soot, PM_{10} and $PM_{2.5}$) and a wind speed influence on total pollen concentrations, there were no significant correlations in most of the examined years, or in some cases, the correlation coefficients were of different nature, proving that, even if the influence of some of these parameters on single pollen species existed, they did not influence the concentration of all of the pollen species in the same way, thus disabling existence of significant correlations.

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Conflict of Interest The authors herewith confirm that they do not have and did not have any conflict of interest during the research described in this paper.

Compliance with Ethical Standards None of the research described in this paper was done with the involvement of human participants and/or animals.

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