Carpinus betulus L. pollen grains in the aeroplankton of twelve selected Polish cities in 2023

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Abstract:

In Poland, Hornbeam (*Carpinus betulus L.*) is one of the trees responsible for the symptoms of spring allergy, although epidemiology of sensitivity is not well understood. The aim of this study was to analyse the *Carpinus betulus L.* pollen seasons in the following cities of Poland: Bialystok, Bydgoszcz, Cracow, Kielce, Lublin, Lodz, Olsztyn, Opole, Piotrkow Trybunalski, Szczecin, Warsaw and Zielona Gora.

Pollen grain was recorded with the standardized volumetric method with the Hirst type traps. The following parameters were included in the analysis: length of the season, peaks of pollen concentration and their date of occurrence, seasonal total values and the number of days of the pollen concentration above the threshold value that could cause symptoms.

The hornbeam pollen season in 2023 started in the third decade of March or the first decade of April and lasted until May, for 33.58 days on average. The highest total values were recorded in Szczecin, Zielona Gora, Bydgoszcz, Cracow and Kielce, while the lowest in Bialystok and Lublin. Compared to 2014, the 2023 season was generally characterized by an earlier start, higher total daily and annual pollen levels.

Key words: hornbeam, *Carpinus betulus L.*, pollen monitoring 2023, Poland

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Introduction

Systematic and botanic characteristics. *Carpinus betulus Linnaeus* was first described in 1753 by a Swedish naturalist and medical doctor Carl von Linné. Both the hornbeam and morphologically similar beech belong to the same order *Fagales*, which contains seven different families. However, due to recent molecular research hornbeam was moved from *Betulaceae* family to *Corylaceae Mirb* family, which also includes hazel and the European hop-hornbeam [1].

Flowering. *Carpinus betulus L.* is a monoecious plant. Each tree has both male and female flowers, located on separate catkins. Female flowers are pollinated by wind with the pollen from the same or other trees. Hornbeam is commonly found in Europe, Asia and North America. It can live for up to 120 years [2].

Ecological and utility values. Hornbeams are characterized by great adaptability, including regeneration and resistance to environmental stress from air pollution [3]. The species is used for air quality biomonitoring [4] and valuable for ensuring the forest biodiversity of animals [5]. Recently, it was found that hornbeam, as one of the few trees, has mechanisms enabling adaptation to dynamic changes in sunlight intensity and can tolerate shading stress even in the range of 50% to ~75% [6]. The photoprotective mechanisms discovered included rapid and significant changes in morphology and physiology. Including changes in leaf size, chlorophyll content, accumulation of relatively high concentrations of organic substances to maintain cellular osmotic balance, as well as increases in the activity of antioxidant hormones and enzymes, activation of antioxidants, etc.

Hornbeam wood is considered the hardest of the trees growing in Europe, harder than beech and oak (the ancient name of hornbeam is “iron wood”). The English name European hornbeam comes from the hardness of the wood, which was compared to a horn. Thanks to its excellent technical properties, since ancient times it has been used to build chariots, elements of traditional windmills, ships, agricultural tools, gears, sleighs, etc. Currently, its wood is used in many branches of the economy. For example, in the production of railway ties, in garden and furniture construction, in the production of parquet floors, tool handles, musical instruments, including drumsticks and other products that require very hard, durable and flexible wood [7].

Medicinal values. Hornbeam extracts have proven to be readily available sources of biomolecules with promising anticancer, immunosuppressive, anti-inflammatory, antimicrobial and antioxidant properties, considered as noteworthy potential precursors of new drugs. Comprehensive profiling of bioactive compounds in *C. betulus* enabled the characterization of nearly 200 polyphenols. Interestingly, the highest antioxidant activity was demonstrated by extracts from leaves and male flowers [4, 8–10].

Known hornbeam allergens and their clinical significance. Currently, official databases describe three panallergens of *C. betulus* [11–13]:

***Car b* 1** – it’s the main allergen of hornbeam pollen. It includes a group of 16 isoforms. *Car b* 1 are thermolabile glycoproteins with a molecular weight of approximately 17 kDa, associated with the pathogenesis of PR-10, as the expression of genes for their synthesis increases in response to stress. As a member of the *Bet v* 1 family, they show high homology of the spatial structure and amino acid sequence with *Bet v* 1, as well as with allergens of other trees of the *Fagales* order, even in the range of 73–88% (for the hornbeam isoallergen *Car b* 1.0109, the agreement was 86%) [13, 14]. *Car b* 1, like other *Bet v* 1-like proteins, has a characteristic hydrophobic molecular pocket which allows for connection with various ligands (and pollutants) up to 1400 kD in size, which can have a significant impact on immunogenicity and allergenicity. This is probably related to the different potential to induce IgE antibodies, regardless of the molecular similarity of hornbeam and birch allergens, which has been demonstrated in birch-free regions of Europe, e.g. Spain [15].

***Car b* 2** (profilin) – a thermolabile structural protein that binds actin in plant cells – important for cytoskeleton formation, mobility and signaling [13, 16].

***Car b* 4** (polcalcin) – a structural protein that binds calcium, occurring only in mature pollen grains. The presence of IgE antibodies against polcalcins can be read as an indicator of a long-lasting allergy, as well as its more severe course.

Methods

The study analyzed average daily concentrations of *Carpinus betulus L.* pollen recorded in 2023 from twelve Polish cities: Bialystok, Bydgoszcz, Cracow, Kielce, Lublin, Lodz, Olsztyn, Opole, Piotrkow Trybunalski, Szczecin, Warsaw and Zielona Gora. Pollen concentrations were examined continuously 24 h a day, using Hirst-type volumetric devices (Lanzoni or Burkard), on a 7-day cycle, in accordance with the requirements specified by the European Aerobiological Society (EAS) [17]. The concentration of hornbeam pollen grains was expressed in 1 m^3 of air.

The start and the end dates of the pollen season, as well as its length, were determined using the 98% method, assuming that its beginning and end, respectively, are determined by the days on which 1% and 99% of the registered annual pollen grains are found. The study presented the characteristics of hornbeam pollen seasons in partner cities, including the calculation of: annual pollen sums, the number of pollen days, their ranges and duration, maximum pollen concentrations (the day of the year on which the highest daily pollen concentration was recorded) and their dates. The annual total pollen value (APIn) was defined as the annual sum of daily values (measured in pollen grains/m^3).

The aim of the study was to compare the characteristics of the hornbeam pollen season in selected Polish cities in 2023.

Results

In the aeroplankton of twelve Polish cities, hornbeam pollen (counted using the 98% method) lasted for 25 to 46 days, on average for 33.58 days. It was observed since the third decade of March and the first decade of April (March 24th–April 17th, 2023) and lasted until May (May 2nd–May 11th, 2023). The earliest it was recorded on March 24th in Zielona Gora and Opole, and its detection ended on May 11th in Bialystok and Bydgoszcz. Meanwhile, botanical sources say that the hornbeam blooms only from mid-April to early May [18].

Pollen grain concentrations reached low and medium values. High concentrations were recorded only on single days in Szczecin, Opole, Bydgoszcz, Cracow and Kielce.

The highest risk of pollen allergy caused by the presence of airborne hornbeam pollen was recorded in the west and south of Poland. The highest total values (SPI index) occurred in Szczecin (488/m^3) and Zielona Gora (395/m^3), Bydgoszcz (359/m^3), as well as in Cracow (456/m^3) and Kielce (419/m^3). In the same cities, concentrations ≥ 16 grains/m^3 persisted for 9 days. In cities in eastern Poland, pollen sums reached low values (fig. 1, 2; tab. 1). The highest daily maximum values were obtained in Szczecin (84/m^3) and Cracow (71/m^3). Moderate values of annual sums were obtained in Opole (267/m^3), Warsaw (222/m^3), Lodz (209/m^3) and Piotrkow Trybunalski (201/m^3). Surprisingly low total concentrations of hornbeam pollen were found in eastern Polish cities: Bialystok (74/m^3) and Lublin (76/m^3).

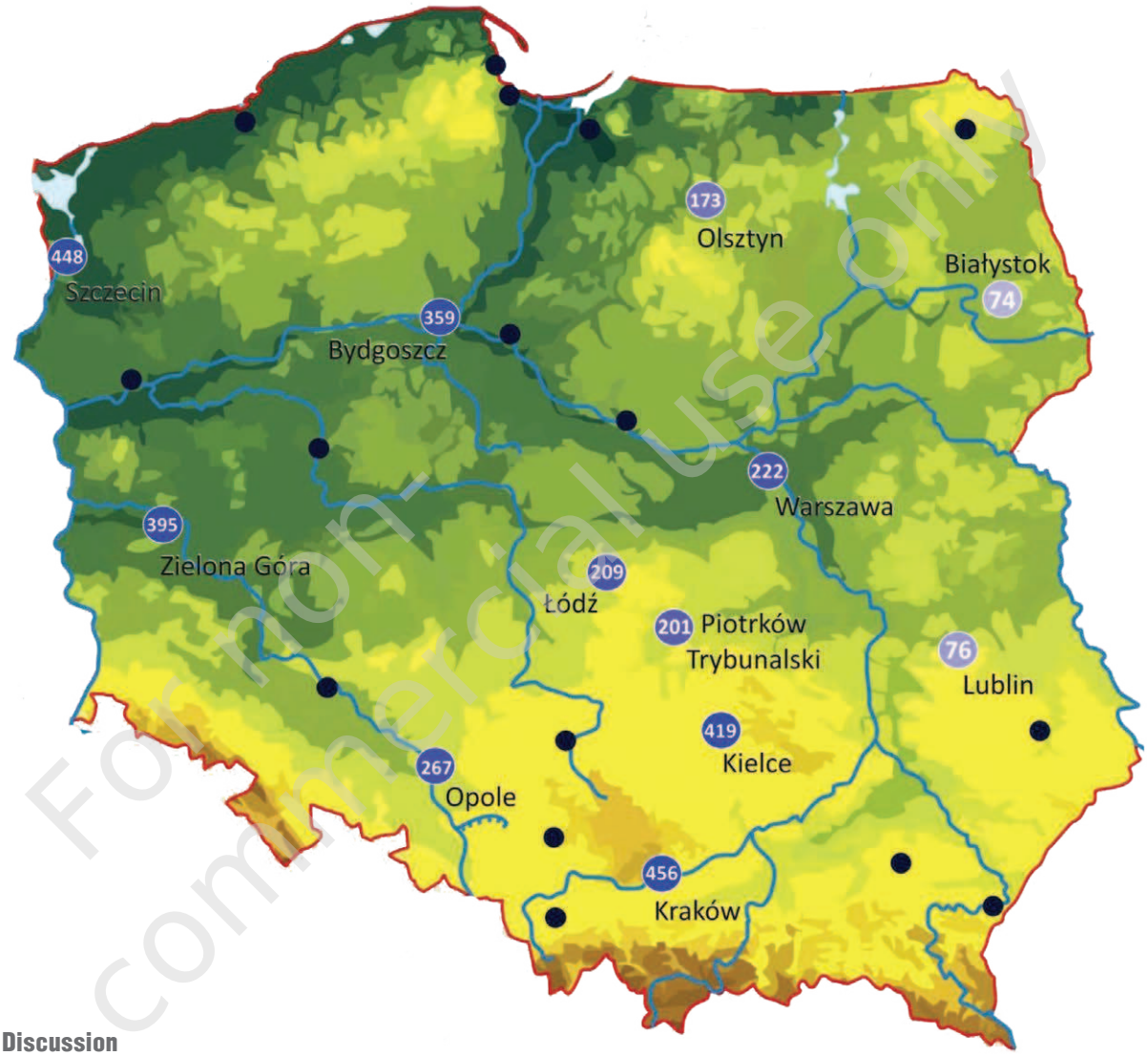
The maximum daily concentrations of hornbeam pollen were the lowest in Bialystok: 13 grains in 1 m^3 and the highest in Szczecin: 84/m^3; the average number of days with the number of grains ≥ 16 m^3 was 5.42.

Detailed results regarding the characteristics of seasons from individual analyzed cities are presented in table 1. Additionally, figure 1 shows the graphical distribution of annual hornbeam pollen sums in the cities included in the analysis against the background of the map of Poland, and figure 2 contains a summary of annual pollen sums, enabling comparison of concentration gradations. The course of pollen in neighboring cities is shown on figures 3–6.

Table 1. *Characteristics of Carpinus betulus L. pollen season in 2023.*

Site	Pollen season (98% method)	Number days of pollen season (98% method)	Maximum value	Date of maximum value	Daily average for days with *Carpinus b.L.* pollen	Sum of daily concentrations (SPI)	Days number with the grains level ≥ 16/m^3
Bialystok	2023-04-17–2023-05-11	25	13	2023-04-21	2.96	74	0
Bydgoszcz	2023-04-05–2023-05-11	31	69	2023-04-22	11.58	359	6
Cracow	2023-03-31–2023-05-03	34	71	2023-04-06	13.41	456	9
Kielce	2023-03-25–2023-05-05	42	67	2023-04-14	9.98	419	9
Lodz	2023-03-25–2023-05-02	39	32	2023-04-20	5.36	209	6
Lublin	2023-04-11–2023-05-10	30	20	2023-04-21	2.53	76	1
Olsztyn	2023-04-12–2023-05-07	26	40	2023-04-22	6.65	173	4
Opole	2023-03-24–2023-05-08	46	54	2023-04-21	5.80	267	3
Piotrkow Trybunalski	2023-04-12–2023-05-02	21	38	2023-04-14	9.57	201	4
Szczecin	2023-03-28–2023-05-07	41	84	2023-04-21	10.93	448	9
Warsaw	2023-04-07–2023-05-03	27	37	2023-04-20	8.22	222	5
Zielona Gora	2023-03-24–2023-05-03	41	48	2023-04-17	9.63	395	9
Average value		33.59	47.75		8.05	261.33	5.42

Figure 1. *Distribution of the sums of annual concentrations (SPI) of hornbeam pollen in 2023 season in twelve Polish cities.*



Discussion

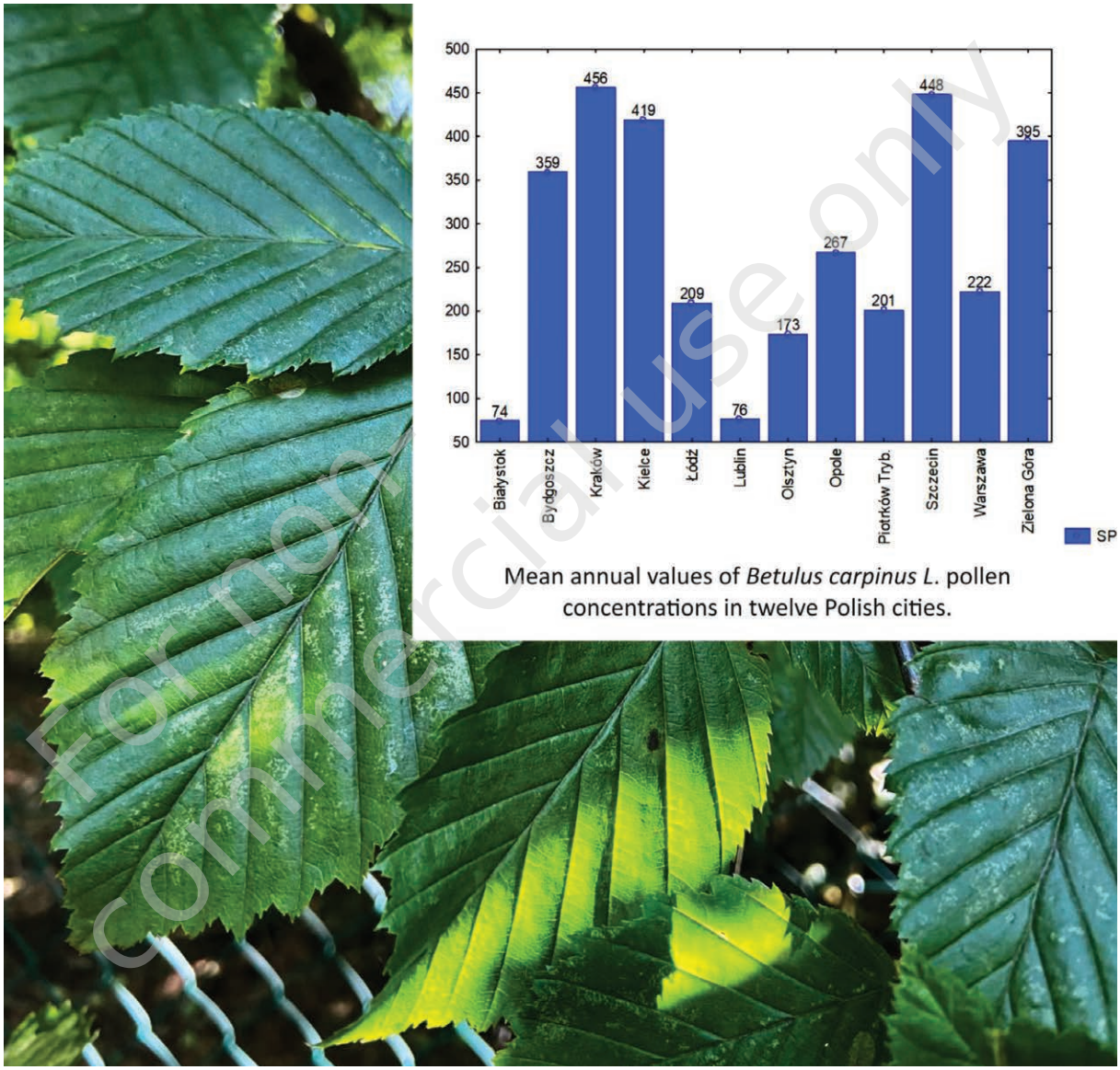
In Polish cities, analyzes of hornbeam pollen concentrations are not common. The available literature shows that the first one was published in 2007 [18] and the next one was in 2014. Comparison of hornbeam pollen fall from 2014 and 2023 [19] indicates a significant increasing trend of exposure to hornbeam pollen. But the references cannot be authoritative. Despite the overall small share of hornbeam pollen in aeroplankton, in most cities subjected to repeated analyses a two-, three- or several-fold increase in hornbeam pollen concentrations was found in 2023. It can be expected that the share of hornbeam will gradually increase in our country.

Until recently, the importance of allergies to hornbeam pollen allergens has been ignored both in scientific and clinical research, even in molecular diagnostics, the symptoms during hornbeam flowering being attributed to cross-reactions, mainly with birch pollen or food allergens [17].

Recently, scientists have questioned the previous view that the *Bet v* 1 protein is the main, proverbial "Trojan horse" causing sensitization to allergens of hornbeam pollen and related plant taxa. Molecular tests revealed only 25% match between IgE epitopes of *Betuloideae* and *Coryloideae* allergens, which are capable of cross-reacting. Laboratory cross-reactivity studies with specific T cells showed limited proliferative responses to *Car b* 1 (38%) against homologous *Coryloideae* and *Fagaceae* allergens. The rest were considered unique to a given subfamily. Therefore, studies of humoral and cellular responses to allergens indicate the possibility of allergic reactions independent of *Bet v* 1, also in people from areas dominated by birch trees [20].

Molecular studies of cross-reactive epitopes have shown that cross-reactivity of T lymphocytes (assessed by ELISA inhibition and basophil activa-

Figure 2. *The summary of the sums of annual concentrations (SPI) of hornbeam pollen in the 2023 season in twelve Polish cities.*
Photo 1. *Hornbeam leaves in private garden. Lodz, August 2023.*



tion tests) specific to pollen allergens from the *Bet v* 1 family, to which hornbeam *Car b* 1 belongs, is lower than expected based on structural similarities [16]. Therefore, the effects of exposure to hornbeam allergens require further, thorough research. Although molecular diagnostics have been introduced into clinical practice, which makes it possible to learn about the personalized sensitization profiles of allergy patients, available panels often do not allow for the diagnosis of allergy to all known hornbeam allergens.

Plant pollen concentrations have a direct impact on health and the quality of life of people suffering from allergies. Therefore, it is important to analyze current hypotheses and carefully determine the risk of developing IgE allergy symptoms related to hornbeam pollen, especially in the regions of western and southern Poland, where pollen concentrations were the highest in 2023. Moreover, hornbeam is generally considered to be a low-allergenic taxon, which may be the reason for the growing interest and investment in planting hornbeams, especially in urbanized urban areas.

It is even considered a marvel of Polish nature. In 1995, the hornbeam was announced as "Tree of the Year" for its phenotypic plasticity, resistance to drought, shade, high/low temperature (down to -30°C), outstanding resistance to pruning, dense growth allowing for privacy, and its important role in shaping the landscape and forest communities [21, 22]. Therefore, further aerobiological, phenological and allergological research is necessary, which could enable comparison

Figure 3. *Carpinus betulus L. pollen concentrations in Lodz, Warsaw, Piotrkow Trybunalski in 2023. Pollen grains/m³. Maximum values are marked.*

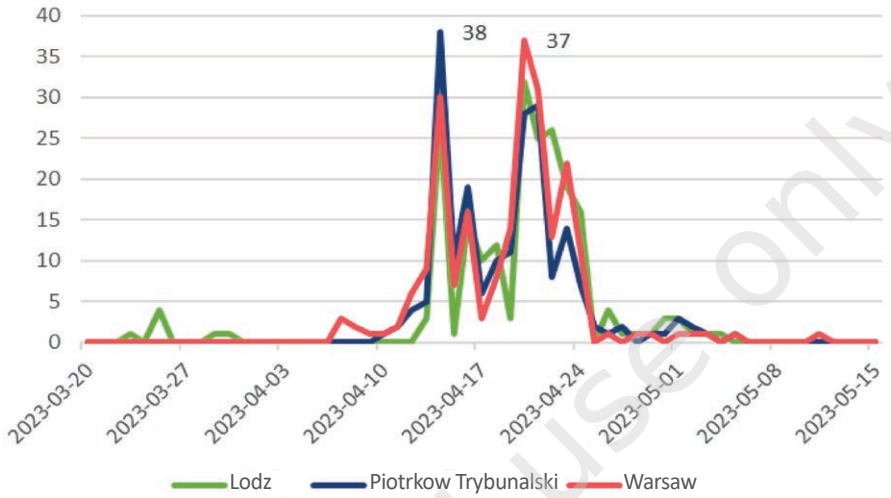


Figure 4. *Carpinus betulus L. pollen concentrations in Cracow and Kielce. Pollen grains/m³. Maximum values are marked.*

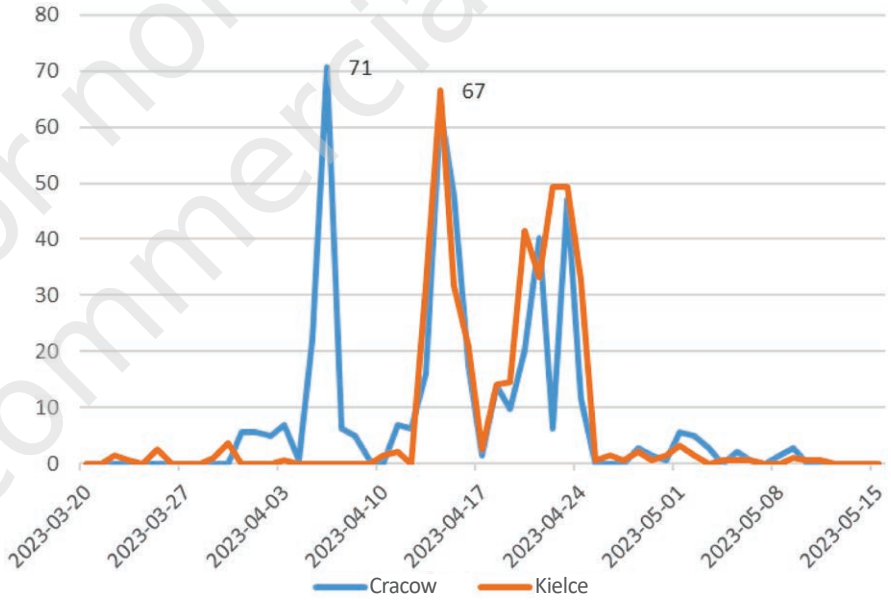


Figure 5. *Carpinus betulus L. pollen concentrations in Szczecin, Bydgoszcz, Zielona Gora in 2023. Pollen grains/m³. Maximum values are marked.*

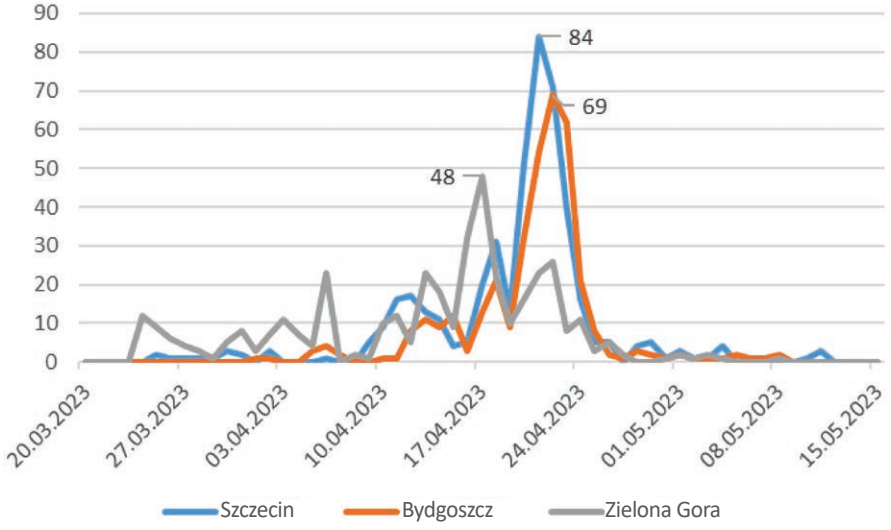
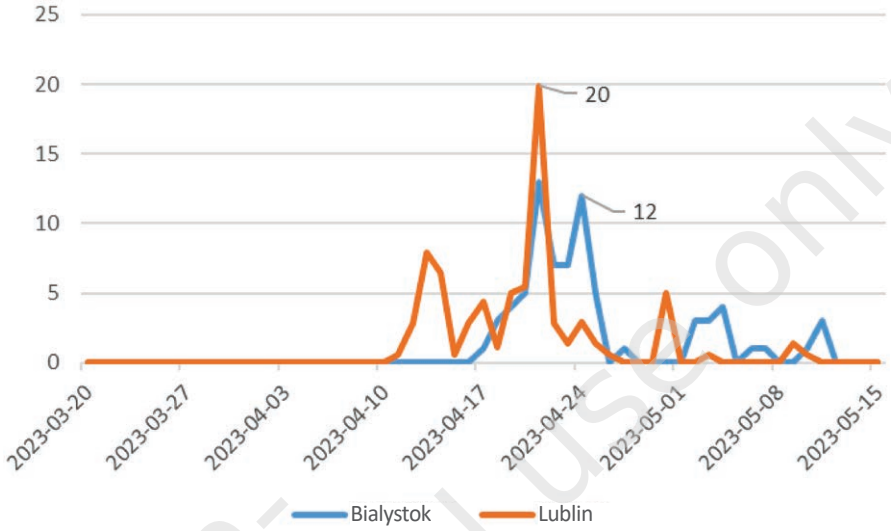


Figure 6. *Carpinus betulus L. pollen concentrations in Bialystok and Lublin in 2023. Pollen grains/m³. Maximum values are marked.*



of long-term trends in this area, as well as the allergenic potential of hornbeam on the Polish population.

Conclusions

- Exposure to hornbeam pollen in Poland in the 2023 season was relatively low (in regard to the concentrations of other tree taxa), but it started earlier, lasted from the end of March to the beginning of May and increased in number compared to the last previously analyzed 2014 season.
- The highest concentration values were recorded in the cities of western and southern Poland, in Szczecin, Zielona Gora, Bydgoszcz, Cracow and Kielce.
- The lowest risk of allergy related to sensitization to hornbeam pollen was found in the eastern cities of Poland: Lublin and Bialystok.

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