

POLLEN MICROSCOPIC IDENTIFICATION OF ALLERGENIC SPECIES IN ORADEA AREA

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Abstract

Palynology is the science that studies palynomorphs, which includes the spores of the inferior plants, and of the pteridophytes, as well as the pollen of the gymnosperms and of the angiosperms. The pollen released by anemophilous plants irritates the mucosa of the sensitive individuals, initiating the occurrence of allergic diseases.

Palynology, by providing with data about the bloom period of the species that produce allergenic pollen, about the possible structural and morphological changes that occur every year, hence offers the possibility to take precaution measures in order to avoid the occurrence of allergies. A structural and morphological analysis of the pollen grains of seven allergenic species from Oradea area was carried out in this paper. Some of the species presented minor deviations, morphological and structural changes, reduction of the exine reticulation, changes in the number of spores, substance sedimentation, that appear as unspecific excrescences visible with an optical microscope. These small changes, as well as other exogenous factors, are able to explain the yearly increase of the allergenic potential of the plants subjected to analysis.

Key words: pollen, allergens, pollinosis

INTRODUCTION

Palynology is the science that studies palynomorphs, which includes the spores of the inferior plants, and of the pteridophytes, as well as the pollen of the gymnosperms and of the angiosperms (Dinescu C. et al, 2005).

Anemophilous plants represent the major source of pollen, small sized pollen grains are carried away by the wind at great distances from the source.

Many of the anemophilous species are allergenic, the pollen released by them irritates the mucosa of the sensitive individuals, initiating the occurrence of allergic diseases (Szabo I, Pallag A., 2007).

Pollen allergy, pollinosis, presents a seasonal character in Romania, and it manifests as allergic rhinitis, allergic conjunctivitis, rhinoconjunctivitis or allergic asthma (Tarnavschi I. T. et al 1987).

Pollinosis, with a continuous increasing incidence in the present, are easier to prevent than to cure.

Implementing prophylactic measures, controlled decontamination of the areas with allergenic species, new sensitizations can be avoided, allergic manifestations are reduced at already sensitized individuals, also the cost of the antiallergic treatment are reduced (Tylianakis I.M. et al 2007).

Interpretation of pollen counts and their relationship to allergy symptoms is complex. The symptoms experienced by allergy sufferers vary from patient to patient and at varying level of pollen and mold concentrations.

Weather can also influence allergy symptoms. Allergy sufferers often find relief on rainy, cloudy or windless days because pollen movement is limited during these days. Hot, dry and windy conditions result in higher pollen concentrations, and increased allergy symptoms.

Because pollens are microscopic particles they are easily spread over great distances by wind, and can result in the detection of pollen which may not be “in season” in the local area (Šimić, S. et al 2007).

The aim of the present paper is the study of some allergenic species in Oradea area.

MATERIALS AND METHOD

Structural and morphological analysis of the pollen grains

Due to the small size of the pollen grains, analysis is performed at a microscope at least 400x.

Identification of the particles is carried out using identification manuals, pollen determiners.

Taking into consideration the polymorphism of the grains within the same species, there can often occur difficulties in the identification by the standard shape recorded in the manuals, especially if it is not described in the local flora.

Also, identification can be made difficult by the presence on the slide of some other particles, different from the pollen particles, like the spores of some moulds, dust particles etc. or some artefacts resulted from the execution of the preparation (water bubbles, air bubbles or stain bubbles) (Cupsa D., Telcean I, 2005).

In order to ease the identification of different types of pollen, the elaboration of a collection of reference slides is advisable, made up of the pollens of the plants collected in the area where the reading is performed, or of collections identified skillfully (Szabo I, 2004, Szabo I, Pallag A., 2007).

We have monitored the structural and morphological characteristics of pollen grains, collected from different species of allergenic plants found in Oradea area.

- *Betula verrucosa* – weeping birch
- *Carpinus betulus* – common hornbeam;
- *Platanus occidentalis* – American sycamore;
- *Sambucus nigra* – elderberry;
- *Tilia cordata* – small-leaved lime;

The morphological and structural particularities occurring at microscopic level have been studied. The structures have also been compared to the standard shapes described in the manuals.

RESULTS AND DISCUSSIONS

Changes in the structure of the pollen, and especially in the exine, that can be observed at an optical microscope, at 400x, can be of different causes.

During the atmospheric heavy metal pollution, the metal particles from the air, attach themselves at the level of the apertures and of the pores of the exine and determine changes in the morphological structure of the pollen.

Scientists in Japan have observed for the first time that patients who were allergic to *Abies firma* pollen (Momi fir), lived in the proximity of some roads. Although Momi fir also grows in areas that are far from heavily trafficked roads, people in those areas did not suffer from allergies (Okubo A., Levin S.A., 1989).

It was proven experimentally that vehicle exhaust gases are dispersed into the air and microscopic solid particles adhere to the surface of the pollen grains, to the ornamentations and to the pores of the exine, bringing alterations to their structure.

On the surface of the pollen grains there are pores that release proteins.

As a result of the pollution factors, the quantity and the quality of the released proteins undergo significant changes. Most of the secreted proteins are proteolytic proteins, that at modified concentrations irritate the mucosae they get in contact with, initiating allergic reactions (Gran W K, Sork V L., 2001).

This explains why many plant species that were not so allergenic or just lightly allergenic in the past, have become strongly allergenic in the last decades causing pollinosis among the sensitive patients (Tylianakis I T et al, 1987, Wodehouse R P., 1935).

Another cause experimentally proven is represented by the induction of genetic mutations by pollution agents. The pollen of the species growing in polluted environment is altered at the level of the genetic material (Gran W.K., Sork V.L., 2001).

According to scientific research, mutations occur at the level of the genes that encode the structure and the ornamentation of the exine, in such a way that these allow the viability of the pollen and the performance of the reproductive function under modified environmental conditions (Taito H et al 2006).

This example also shows that the unfavourable effects and the consequences of pollution occur in unexpected fields and affects the health of the population (Szabo I, 2004).

We have monitored the structural and morphological characteristics of the pollen grains of the species under study.

Betula verrucosa, the weeping birch, presents a specific pollen, with radial symmetry, three colpi, and superficially presents microwarts and it is presented in figure 1.

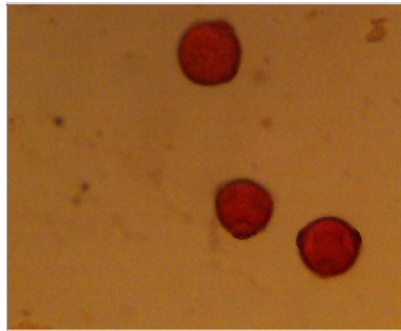


Fig 1. *Betula verrucosa* pollen (optical microscopy image, 200x)

The pollen collected corresponds to the structure typically described with no structural changes.

The pollen collected from *Carpinus betulus*, as the largest in size, can be well visualized with a 40x objective. Figures 2, 3, and 4 present the images obtained with a 10x objective and a 40x objective, respectively.

The typical pollen grains present four equatorial pores, the exine is granular, while some of the collected grains present five pores, and the exine is less granular.

Platanus occidentalis pollen grains have a radial symmetry, pollen diameter 18,6-26 μ . There are three colpi, the membrane of the aperture is granular, the surface of the exine is reticulated. The collected and analyzed grains present a typical structure, only some of the grains are slightly modified, a little flattened, and thus the three colpi are difficult to be observed. All these can be better visualized in figure 5.

The elderberry (*Sambucus nigra*) pollen grains are threecolpate, oblat-sphaeroidal shape, with perpendicular thickening on the colpi. The collected grains present numerous rugosities and atypical excrescences that can be observed in figures 6 and 7.

Tilia cordata grains are heteropolar, with ornamentations between the poles, threecolporate.

The colpi have irregular margins. Most of the collected grains correspond structurally and morphologically to the typical form, only few specimens present small structural changes, as for example the one in figure 8.

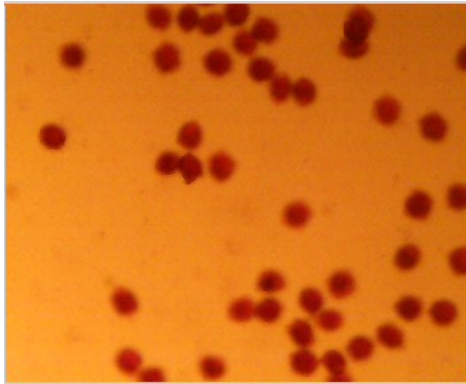


Fig 2. *Carpinus betulus* pollen (optical microscopy image, 100x)



Fig. 3. *Carpinus betulus* pollen (optical microscopy image, 400x)

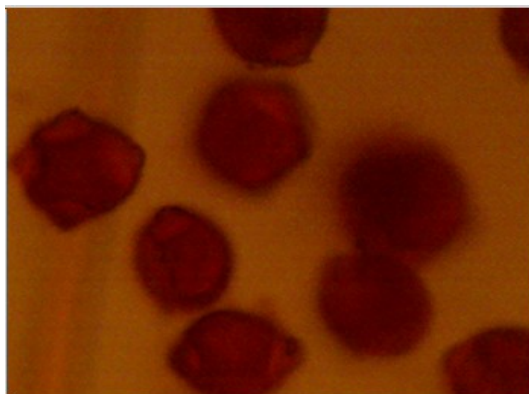


Fig 4. *Carpinus betulus* pollen (optical microscopy image, 400x)

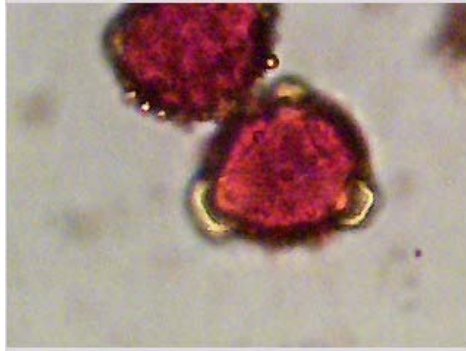


Fig 5. *Platanus occidentalis* pollen (optical microscopy image, 800x)

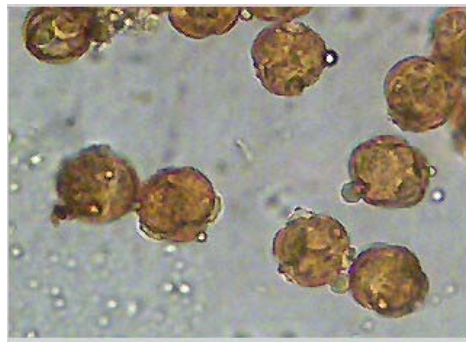


Fig 6. *Sambucus nigra* pollen (optical microscopy image, 200x)

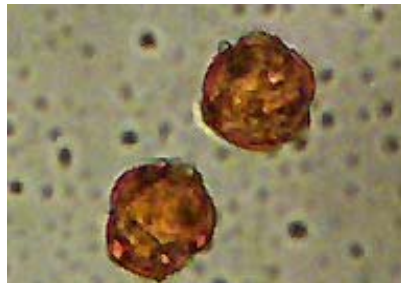


Fig 7. *Sambucus nigra* pollen (optical microscopy image, 800x)

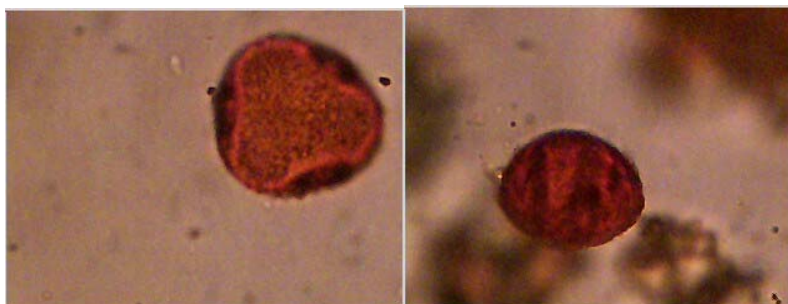


Fig 8. *Tilia cordata* pollen (optical microscopy images, 800x)

CONCLUSIONS

The number of allergenic species in the local spontaneous flora is large and it includes a series of trees from parks, gardens and vegetation curtains, as well as herbs (Dinescu C et al, 2005)

Over three million individuals suffer from allergic diseases in Romania. The most frequent are allergic rhinitis, conjunctivitis, asthma and allergic sinusitis that can occur at all age categories. The prevalence of allergy to pollen is high, and its incidence is increasing in comparison to past decades (Cupsa D., Telcean I, 2005).

Some of the species present minor deviations, morphological and structural changes, reduction of the exine reticulation, changes in the number of pores or colpi, substance sedimentation, that appear as unspecific excrescences visible with an optical microscope. These small changes, as well as other exogenous factors, are able to explain the yearly increase of the allergenic potential of the plants subjected to analysis.

According to scientific research documentation, these structural changes of the outer layer of the pollen lead to alteration of the quantity and quality of the secreted proteins. And as most of these proteins are proteolytic enzymes which at modified concentrations irritate the mucosae they come in contact with, it explains the initiation of the allergic reactions.

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