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**The Application of the Plant Pollen for the Bioindicative
Assessment of the Environmental Pollution Level**

Wykorzystanie pyłku roślin do bioindykacyjnej oceny poziomu
zanieczyszczenia środowiska

Abstract. The experiment revealed a high positive correlation of pollen abortivity frequency of investigated species and traffic intensity. It was assumed that the increase of the concentration of the motor-cars emissions is responsible for these results. Within the experiment *Aesculus hippocastanum* demonstrated significant variability of pollen grains quality. Variance analysis showed 99.7% of this variability depend on controlled parameter and thus environmental pollution. Such statistical confirmation makes it possible to apply *Aesculus hippocastanum* pollen to environmental control.

Key words – słowa kluczowe: *Aesculus hippocastanum*, *Dactylis glomerata*, pollen – pyłek, bioindication – bioindykacja, environmental pollution – zanieczyszczenie środowiska.

INTRODUCTION

Modern analytical methods make it possible to measure the concentrations of chemical substances in the environment but do not give information about their toxic effect on the living systems. This problem can be successfully solved with bioindicative methods. Nevertheless, with bioindication we can estimate only the common effect of all factors without revealing their specific action. Often, it is enough to know the total effect of pollutants on organisms for the assessment of their further functioning.

Higher plants have already shown their ability to bioindication of the environmental chemicals toxicity (Treszou, 1988). Among the different properties of plant organisms the processes of sporogenesis and gametogenesis are very sensible to exogenous factors. Most available for bioindicative tests is the quality of the seed plants male gametophytes – pollen grains (Bessonova, 1992; Mičieta and Murín, 1997). The plants produce them in a quantity that is necessary for statistical analysis. Besides the normal pollen organisms form some quantity of sterile pollen, as a rule not more than 5%. In the cases of cytoplasmic male sterility, polyploidy, apomixis, hybridization as well as in unfavorable exogenous condition – such as extreme temperature, aridity, deficit in nutrition, heavy metal and other chemical contamination, radioactivity – it can increase significantly (Poddubnaya-Arnoldy, 1976). As it was established in another region there is a high positive correlation between the amount of passing cars and pollen sterility of some species growing near the roads (Glazunowa, 1990).

The goal of our study was to reveal the plant species with pollen whose quality depends on air and soil contamination with the motor-car emissions.

MATERIAL AND METHODS

Within the city limits a few roads with different traffic intensity were selected. We expressed the rate of environmental pollution through the average amount of cars passing by per 1 hour (Berzina, 1980). As the objects of the study *Aesculus hippocastanum* and *Dactylis glomerata* were chosen. In their embryological processes neither cytoplasmic male sterility or polyploidy, neither apomixis or hybridization were observed (Comparative embryology..., 1987).

With the matter *in vitro* viability tests of pollen grains nearly 30 closed flowers from each of 10 individual plants of *A. hippocastanum* along each experimental road were collected in paper bags. In the case with *D. glomerata* a few inflorescences from each of 10 sites along each studied road were collected for analysis. After 3–5 hours of collection spilt out and mixed pollen grains were fixed in a little quantity of ethanol: acetic acid (3 : 1) mixture. The pollen suspension was treated with 0.5% acetocarmin and observed with 400 multiple magnification of microscope. From each plant (site) about 2000 pollen grains were evaluated for size, form and color.

The observed pollen grains were polymorphous. The pollen of average sizes with well colored cytoplasm and nuclei was considered to be fertile. It was generally stated that gigantic, small, corroded and not colored pollen grains are sterile (Pausheva, 1988).

RESULTS AND DISCUSSION

The pollen grains counting and correlation analysis showed that increasing motor transport on the roads per one hour accompanies the worsening of pollen quality of both investigated species (Tab. 1, 2). Deterioration of pollen quality was observed when the damages on the macro-morphological level were absent.

A high level of correlation (0.99 with 99% confidence interval) between motor transport load and pollen condition of *A. hippocastanum* was revealed. In case of *D. glomerata* the correlation index was smaller (0.92 with 95% confidence interval).

Tab. 1. Quantity of aborted pollen grains of *Aesculus hippocastanum* planted near the roads with different traffic intensity

Traffic intensity (average amount of cars passing per 1 hour)	Average amount of sterile pollen grains (%)	Standard error	Coefficient of variation (%)
50	7.3	0.9	11.8
150	11.1	0.5	4.9
400	16.8	0.7	3.9
700	20.8	0.8	3.9
1500	37.1	1.9	5.2

Tab. 2. Quantity of aborted pollen grains of *Dactylis glomerata* growing near the roads with different traffic intensity

Traffic intensity (average amount of cars passing per 1 hour)	Average amount of sterile pollen grains (%)	Standard error	Coefficient of variation (%)
170	8.6	0.4	10.5
180	14.6	0.3	5.5
400	14.7	0.2	4.1
1500	17.6	0.3	4.5
2000	18.7	1.3	4.3
2500	22.5	0.5	5.8

Although differences between individual trees were observed, the coefficient of variation inside each variant was low. Its maximum did not exceed 11.8%. Variability range among the different gradation of the studied factor was significantly larger: 62% in the case of *A. hippocastanum* and 29% in *D. glomerata*. Therefore, in the condition of different motor-car emission load the pollen quality of *A. hippocastanum* was found very changeable, which is an obligatory requirement using it as an indicator of environmental conditions. Through the middle variability level in the condition of higher range of investigated factor gradation (from 170 to 2500 passing cars per 1 hour) *D. glomerata* pollen was assumed as more tolerant to this kind of contamination.

Since it is impossible to standardize the experimental conditions strictly, some part of the pollen sterility changes between the variants could be caused by other factors. We conducted a one-factor variance analysis to find this. It was shown that 99.7% of all changes of *A. hippocastanum* pollen sterility and 97% of *D. glomerata* depend on motor transport load of the roads with 99% confidence interval.

Hence, we can add a new taxon of *D. glomerata* to the list of herbaceous plant species of Central Europe (Glazunowa, 1990; Mičieta and Murín, 1997) that reacted to air contaminating emissions with the rise of pollen anomaly frequency. As addition to previous research (Bessonova, 1992; Kalinovich, 2001), the newly cultivated tree species of *A. hippocastanum* was tested with a positive result for its ability as bioindicator for environmental control. In our study, the dependence of pollen quality on different rates of air pollution was revealed with strictly statistical confirmation. These results make it possible to test not only the presence of pollutants in the environment but their comparative level.

CONCLUSIONS

1. In our experiments the increase of motor transport load on the roads is accompanied by the rise of pollen sterility of *A. hippocastanum* and *D. glomerata* growing near the roads. Hence, the increase of pollen abortivity frequency is a response of plants to air contamination with motor-car emissions.

2. Worsening of both the studied species' pollen quality was observed when injuries on the macro-morphological level were absent. Therefore, pollen can serve as a criterion of early plant damage diagnostics.

3. Pollen of *A. hippocastanum* is more sensitive than *D. glomerata* to motor-car emission.

4. Strictly statistical confirmation of the obtained results makes it possible to apply *A. hippocastanum* pollen in environmental control especially in the analysis and evaluation of air quality, in the detection of the presence of pollutants when the visible injuries of plants are absent, in identification of their zone of impact, in mapping the air pollution and having an idea of its concentration in the atmosphere.

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STRESZCZENIE

Doświadczenie wykazało wysoką dodatnią korelację między poziomem sterylności pyłku badanych gatunków roślin a stopniem obciążenia dróg ruchem transportu samochodowego. Okazało się, że wzrost sterylności pyłku spowodowany został przez zwiększenie zanieczyszczenia środowiska wydzielinami silników samochodowych. W doświadczeniu stwierdzono, że pyłek *Aesculus hippocastanum* odznaczał się zmienną jakością. Ponadto jednoczynnikowa analiza dyspersyjna dowiodła, że 99,7% wszystkich zmian zależy właśnie od stopnia zanieczyszczenia środowiska, dlatego też poziom sterylności pyłku *A. hippocastanum* może być wykorzystany do oceny stanu środowiska.