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Original Research

Phytoindication of the Ecogenotoxic Effects of Vehicle Emissions Using Pollen Abortion Test with Native Flora

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Abstract

This study aimed to evaluate the rate of ecogenotoxicity caused by traffic emissions at sites in the vicinity of roads and at sites near planned highway construction using a pollen grain abortion assay with higher wild plant species. The relationship between pollen grain abortivity and distance from a road also was assessed. The highest values were found at a distance of 30 m, where the prevailing wind direction was parallel to the road. Wind blowing away from the road shifted this boundary to a distance of 350 m. The results showed the highest genotoxicity at the "Ring road" site, where the frequency of abortive pollen grains was 4.05 times higher than at the control site, and at the "Dubná Skala" site, with induction factor 3.48. Based on our results we can conclude that *Chelidonium majus*, *Cichorium intybus*, and *Melilotus albus* are suitable species for the detection of genotoxicity in the environment.

Keywords: pollen abortivity, bioindication, genotoxicity, vehicle emissions, native flora

Introduction

Many experimental studies have shown that all environmental components (air, water, and soil) are contaminated by pollutants from different sources in industrial and urban areas [1-7]. Traffic is one of the main sources of air pollutants that significantly decrease environmental quality. Damage to the genetic material of plant, animal, and human cells results from huge amounts of contaminants (CO, NO_x, SO_2, VOC-volatile organic compounds and heavy metals) with toxic, carcinogenic, and mutagenic effects. Negative toxic effects on pollen grains also were observed for platinum group elements (Pd, Pt, Rh), which are used in catalytic converters and emitted to the environment, mainly such as nanoparticles [8].

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In 2008 Slovak road traffic produced 44,000 tons of NO_x (47% of total), 62,000 tons of CO (26%), 4,700 tons of particulate matter (12%), and 200 tons of SO_2 [9]. Ambient air in the vicinity of urban areas with industry and heavy traffic contains human carcinogens such as benzene, 1,3-butadiene, and polycyclic aromatic hydrocarbons, which are among the most carcinogenic substances contained in gas emissions [5]. Positive results of genotoxic effects of heavy metals (Pb, Cr, Ni, Cd, Hg) were obtained in many studies with plant bioassays [10, 11]. Negative influences of heavy metals were confirmed by Calzoni et al. [6].

Environmental degradation due to anthropogenic impacts has led to a search for suitable bioindication methods and bioindicators. Phytoindication methods allow us to evaluate environmental quality changes according to plant responses. Plants are suitable bioindicators of the genotoxicity of a polluted environment due to their high sensitivity and ability to accumulate harmful substances. Specific plant responses can be observed even at very low concentrations.

Plants have a natural integrating function in water, soil and air. Their primary position in the food chain is very important, as is the fact that a huge amount of genotoxic substances have the same effect on plants, animals, and humans [12]. Plant chromosomes are highly sensitive to chemical mutagens, giving rise to chromosomal aberrations. The organization of the plant chromosome is similar to that of humans [13, 14]. However, there are several differences in anatomy and physiology between plant cells and those of animals including humans, but DNA structure, function, and the process of protein synthesis are similar. One of the main advantages of plant test systems is that they show a wide range of genetic, chromosomal, morphological, and physiological changes by implication of mutagen treatment [8, 15].

One of the reliable plant bioassays for *in situ* biomonitoring of air pollution is pollen abortion assay with wild plants collected directly from a contaminated environment, enabling us to assess the actual situation without regulated conditions [16]. The basic criteria for selection of wild plant bioindicators were established in the study of Murín [17]. The list of suitable species of Slovak native flora has been gradually extended by Mičieta and Murín [1]. Possible plant adaptations to certain toxins are the main disadvantage of the mentioned method. Some physiological characteristics (unknown length of exposure to the substance, environmental conditions affecting pollen development, short vitality and production only in a fixed period) are disadvantageous to its application as an indicator in *in situ* conditions [18]. The high sensitivity of pollen during development causes responses to extreme natural conditions such as heat and dryness [17]. On the other hand, the pollen can accumulate harmful substances and provide information about their negative influences. Even small changes in the pollen genome can cause plant damage [19] and, consequently, fertility reduction. In addition to genotoxic effects of traffic emissions [16, 20], the test mentioned above also has been used to evaluate genotoxic hazard in areas with increased radioactivity [21], for bioindication of air pollution in the vicinity of industrial areas and landfills [4, 5], and for the genotoxicity detection of an environment contaminated by heavy metals [22].

In this study the potential effects of environmental contamination by traffic emissions in the vicinity of populated areas and their possible impact on human health were examined. We decided to evaluate the level of ecogenotoxic deterioration by pollen abortion test. We wanted to highlight the suitability of this test for determining the rate of environmental genotoxicity near high vehicular traffic. The aim of this study also was to standardize applied fytoindicator species and to find the regions of the most intensive contamination of the environment depending on distance from the road. The distances of occurrence of contamined pollen from nearby roads were measured. The hypothesis that plant genetic material effectively indicates the low concentrations of the environmental contamination also was tested.

Material and Methods

Sampling Sites

The town of Martin is situated in the northern part of central Slovakia. It is characterized by a moderately warm climate with an average annual temperature of 7-7.5ºC and average annual precipitation of 750-860 mm and north-south prevailing wind. Frequent temperature inversions, low air circulation, and high humidity are caused by its geographic location in Turčianska Valley, surrounded by mountains. These climatic phenomena increase the concentration of air pollutants in the ground layer of the atmosphere. The territory of the town is regarded as an industrial area. Stationary sources of emissions are located near the monitored sites (calandria, ZŤS TEES Martin, ŽOS Vrútky, and Sučany industrial area). The town has 163 km of roads with high traffic density. Two main roads, 1/18 (E50) and 1/65, pass through the built-up areas of the town. Average traffic density was 15,000 vehicles per 24 hours on individual sections of the 1/18 road [23].

Research and sampling sites were divided into two clusters of locations: A and B. Four sites were chosen in the urban area of Martin (Part A – urban area, from 1 to 4 in Fig. 1). All sites were located near a road with high traffic density. Vegetation was predominantly composed of ruderal plant communities.

The additional four sites (Part B – future highway area, from 5 to 8 in Fig. 1) were located near the planned construction of Highway D1 Dubná Skala-Turany, which will probably become the main line source of exhaust and dust emissions. Locating this road in the proximity of the river Váh will surely influence the littoral vegetation with its subsequent fragmentation or partial loss of eminent habitats of national importance – Kr8, Kr9 [24]. Our results offer the initial material for further monitoring of genotoxic effects of traffic pollution after construction of the highway.

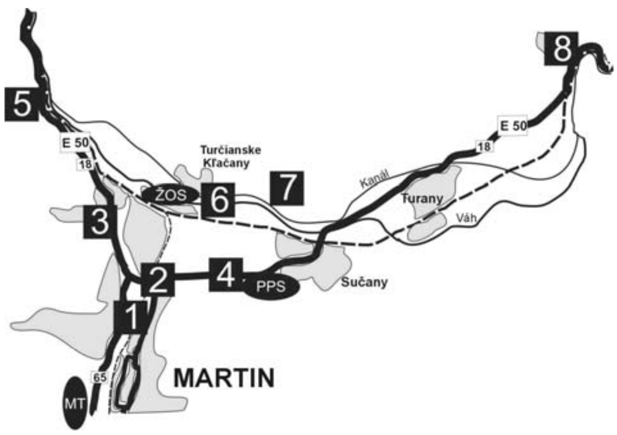

Fig. 1. Map of Martin town showing locations of sampling sites: 1 – Tulip (below as T), 2 – Košúty (K), and 3 – SNP Cemetery (C) – 17,193 vehicles per hour, 4 – Ring-road (R) – 14,859 vehicles/hour, 5 – Dubná Skala (DS) – 14,784 vehicles/hour, 6 – Váh (V), 7 – Biele Brehy (BB), 8 – Šútovo (S) – 11,618 vehicles/hour; stationary sources of emissions: calandria and machine plant, PPS – Sučany industrial area, ŽOS Vrútky – machine plant.

Table 1. Frequency of abortive pollen grains (% and SEM) in 2009 – Urban area (Part A).

Species/location	T	K	C	R	Control	B
Artemisia vulgaris	12.4±0.8 **	7.8±0.9 **	8.1±0.6 **	9.4±0.8 **	2.4±0.7	VII-VIII
IF	5.2	3.3	3.4	3.9		
Barbarea vulgaris	2.5±0.4	3.2±0.4 **	4.3±0.6 **	8.2±1.1 **	1.4±0.4	V-VIII
IF	1.8	2.3	3.1	5.8		
Chelidonium majus	4.2±0.5 **	3.2±0.3 **	4.3±0.6 **	6.8±0.7 **	2.0±0.3	V-VIII
IF	2.1	1.6	2.15	3.4		
Cichorium intybus	9.9±0.8 **	5.3±0.5 **	4.0±0.5 **	4.6±0.4 **	1.0±0.2	VII-X
IF	9.9	5.3	4	4.6		
Daucus carota	5.6±0.5	17.1±2.2 **	2.2±0.3	5.6±0.9	4.4±1.2	VI-XI
IF	1.3	3.9	0.5	1.3		
Lamium maculatum	1.4±0.2 **	1.6±0.2 **	2.1±0.3	7.0±0.6 **	2.8±0.2	IV-IX
IF	0.5	0.6	0.75	2.5		
Lactuca serriola	1.8±0.3 *	2.3±0.3 **	3.0±0.4 **	5.7±0.9 **	0.9±0.2	VII-IX
IF	2	2.6	3.3	6.3		
Melilotus albus	5.1±0.8 **	10.4±1.1**	7.2±1.3 **	7.7±1.5 **	1.0±0.3	VI-IX
IF	5.1	10.4	7.2	7.7		
Pastinaca sativa	9.7±1.1 **	5.5±0.5 **	4.3±0.4	3.5±0.5	3.4±0.4	VII-VIII
IF	2.85	1.6	1.3	1		

T – Tulip, K – Košúty, C – SNP Cementery, R – Ring road, B – blooming period (months).
*Statistical significance (P≤0.05)
**Statistical significance (P≤0.01)
IF – induction factor

Záhorska lowland (between the villages of Moravský Ján and Závod) was used as a control site where abortion of pollen grains over the last 70 years has not exceeded 5% [1].

Sampling and Evaluation of Pollen Abortion

A pollen grain abortion assay was used to detect the rate of phytotoxicity and mutagenicity of a polluted environment. Twelve species of native flora satisfying all criteria for selection [17] were chosen from the list of suitable species. Collection of samples (90) was done during the 2009 growing period from early April to September. At each site (1-8), 10-20 young and closed flowers and flower buds from at least 10 individuals from each species were collected. The samples were fixed in a mixture of ethanol (96%) and acetic acid (3:1). After 24 hours the fixing solution was replaced with 70% ethanol. Pollen grains were printed from anthers and stained with 0.05% aniline blue in lactophenol. The abortion pollen grains were determined on the basis of form (altered form, unformed, undeveloped), size, and staining deficiency [1, 17]. The evaluated set of 3,000 pollen grains per sample (per species) was evaluated under a light microscope Nikon YS2 Alphaphot (400-times magnification). The induction factor (IF) was used as an indicator of increased genotoxic load. The ratio of abortivity found at the polluted/control site provided information for how many times the abortivity at the polluted site was higher than in the control site [5]. Statistical significance of differences between frequency of the abortive pollen grain at the control and polluted sites were evaluated by Student's T-test at 0.05 and 0.01 significance levels.

Results

Urban Area (Part A)

The frequencies of abortive pollen grains were significantly increased at all sites with high genotoxic effect of traffic emissions in the vicinity of road section (Table 1). The species *Artemisia vulgaris*, *Chelidonium majus*, *Cichorium intybus*, and *Melilotus albus* showed statistically highly significant increase (P≤0.01) of pollen abortivity at all monitored sites. The most significant increase of the frequency of abortive pollen grains was found in *Melilotus albus*. Its pollen abortion exceeded the limit of 5% at all polluted sites ("T" 5.1±0.8, "K" 10.4±1.1, "C" 7.2±1.3, and "R" 7.7±1.5). The genotoxic effect of air pollution was 10.4 times higher at "K" site than at the control site (1.0±0.3).

Table 2. Frequency of abortive pollen grains (% and SEM) in 2009 – Future highway area (Part B).

Species/location	DS	V	BB	S	Control	B
Artemisia vulgaris	5.7±0.6 **	9.6±1.1 **	6.9±0.7 **	9.2±1.0 **	2.4±0.7	VII-VIII
IF	2.4	4	2.9	3.8		
Calystegia sepium	10.5±3.4 **	6.2±1.2 **	7.8±0.5 **	0.9±0.2 *	1.6±0.2	VI-IX
IF	6.6	3.9	4.9	0.6		
Cichorium intybus	4.3±0.4 **	2.3±0.37 **	3.2±0.6 **	4.8±0.4 **	1.0±0.2	VII-X
IF	4.3	2.3	3.2	4.8		
Daucus carota	7.3±0.7 *	9.9±0.9**	6.5±0.4	7.6±0.5 *	4.4±1.2	VI-XI
IF	1.7	2.3	1.5	1.7		
Lactuca serriola	2.1±0.4 **	2.4±0.4 **	2.6±0.4 **	1.7±0.3 *	0.9±0.2	VII-IX
IF	2.3	2.7	2.9	1.9		
Melilotus albus	4.1±0.8 **	3.8±0.4 **	4.0±0.5 **	11.2±2.5 **	1.0±0.3	VI-IX
IF	4.1	3.8	4	11.2		
Melilotus officinalis	2.4±0.3	5.9±0.9 **	8.2±0.7 **	2.1±0.4	1.4±0.4	VI-IX
IF	1.7	4.2	5.9	1.5		
Pastinaca sativa	8.9±0.9 **	3.5±0.4	3.4±0.4	4.5±0.5	3.4±0.4	VII-VIII
IF	2.6	1.02	1	1.3		
Trifolium pratense	21.2±2.7 **	8.6±0.7 **	15.0±1.7 **	12.7±1.3 **	3.8±0.6	VI-XI
IF	5.6	2.3	3.9	3.3		

DS – Dubná Skala, V – Váh, BB – Biele Brehy, S – Šútovo, B – blooming period (months).
*Statistical significance (P≤0.05).
**Statistical significance P≤0.01).
PPIF – induction factor

In contrast, the lowest frequency of abortive pollen grains ("T" 1.4±0.2, "K" 1.6±0.2, and "C" 2.1±0.3) was observed in samples of *Lamium maculatum*. Its value at the control site was 2.8±0.2. Even high statistically significant decrease of abortive pollen grains was seen at two sites ("T" and "K") with induction factors 0.5 and 0.6. From the evaluated sites, the strongest increase was observed at "R" site, where the environmental genotoxic load was 4.05 times higher in comparison to the control site, and the lowest genotoxic effect (average IF 2.85) of air pollution was seen at the "C" site. "K" became the second most genotoxic burdened site where the average induction factor suggested increasing frequency of abortive pollen grains by 3.51 times. A similar result was found at the "T" site (average IF 3.42) (Table 1, Fig. 2).

Future Highway Area (Part B)

From the results summarized in Table 2 it is apparent that environmental genotoxic load was elevated at all monitoring sites. A highly statistically significant increase (P≤0.01) of frequency of abortive pollen grains was observed at all sites for *Artemisia vulgaris*, *Cichorium intybus*, *Melilotus albus*, and *Trifolium pretense*. The strongest

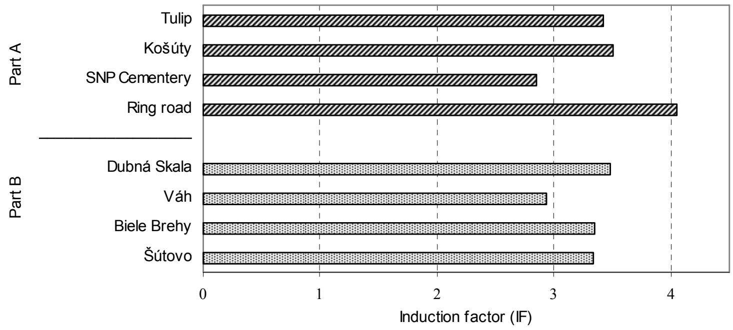

Fig. 2. Average induction factors of the evaluated sites.

increase was found at site “S” in *Melilotus albus* (IF 11.2). No statistically significant differences between percentages of abortive pollen grains at polluted versus control sites were found in *Pastinaca sativa* (“V” 3.5±0.4; “BB” 3.4±0.4; “S” 4.5±0.5), *Melilotus officinalis* (“DS” 2.4±0.3; “S” 2.1±0.4), and *Daucus carota* (“BB” 6.5±0.4) (Table 2). The same value as at the control site was found at “BB” sites in *Pastinaca sativa* (3.4±0.4). The highest environmental genotoxicity was registered at the “DS” site (average IF 3.48) and the lowest at the “V” site (average IF 2.94) (Fig. 2).

The most significant increase in the frequency of abortive pollen grains was found in *Melilotus albus* for both clusters of localities: A and B. In contrast, the lowest values were observed in samples of *Lamium maculatum* for cluster A and *Calystegia sepium* for cluster B.

Pollen grains of *Salix caprea*, collected at the sites with growing distances from the road, showed significant increasing of pollen damage up to the threshold limit with the highest genotoxic burden of the environment (Fig. 3). This value was 30 m at the urban area (Part A), where the prevailing wind direction was parallel to the road. Frequency of abortive pollen grains there was (28.4±3.19). While the highest value (7.7±1.0) was recorded at a distance of 1,000 m at the site contaminated to a great extend by pollution from a nearby machine plant (ŽOS Vrútky), the threshold value was considered at a distance of 350 m (6.9±0.6) from the road for the future highway area with prevailing wind direction away from the road (Part B). A negative correlation was observed for distances above this value.

Discussion

Ambient air in urban and industrial areas with busy roads contains substances that have negative effects on all living organisms, including humans. Examination of the rate of environmental genotoxicity to which these organisms are exposed is key in terms of the possible occurrence of carcinogenic effects or DNA damage.

Our results confirm that pollen abortion assay allow us to effectively assess the extent of the genotoxic hazard of carcinogenic and mutagenic traffic emissions to the environment, significantly affecting human health. In most studies on the genotoxicity of a large spectrum of mutagens, including traffic emissions, this assay is used simultaneously with a Tradescantia micronucleus assay (Trad MCN test) [4, 5, 16]. Despite the many advantages of using pollen grains as an evaluating material for monitoring environmental contamination, the micronucleus test is more often used for the evaluation of genotoxicity of traffic emissions. A significant decrease of the environmental genotoxicity during the bus strike showed that disruption of one source of pollutants has the ability to change the mutagenic potential of these substances [3]. The genotoxic effect of atmospheric pollutants in urban areas without the impact of industry but with high traffic density also was documented in the study of Meireles et al. [25]. The sensitivity of *Tradescantia* to air pollution [26, 27] and to traffic emissions [28] showed the genotoxic potential of these substances. These studies confirm that a high degree of vehicular traffic is one of the most important stressors affecting the toxicity of the environment.

The results of the present study show the strong genotoxic effects of air pollutants, especially traffic emissions, on native plant species. As expected, a statistically significant increase in the frequency of abortive pollen grains was observed at all sites in an urban area (Part A). Even higher values of average induction factors in the range between 2.85 and 4.05 were observed there in comparison with a site near a highway in Bratislava, where the average value caused by vehicle emissions was 1.7 [16]. The highest genotoxicity of the environment from all evaluated sites was found at site “R” with an average induction factor of 4.05. Traffic in this part of the 1/18 road is significantly held up, which gives rise to increased amounts of exhaust and abrasive emissions. A similar case was spotted at site “T” due to the construction of shopping centers in the immediate vicinity of the busy 1/65 road. In addition to traffic emissions, a north-south wind direction carries emissions to

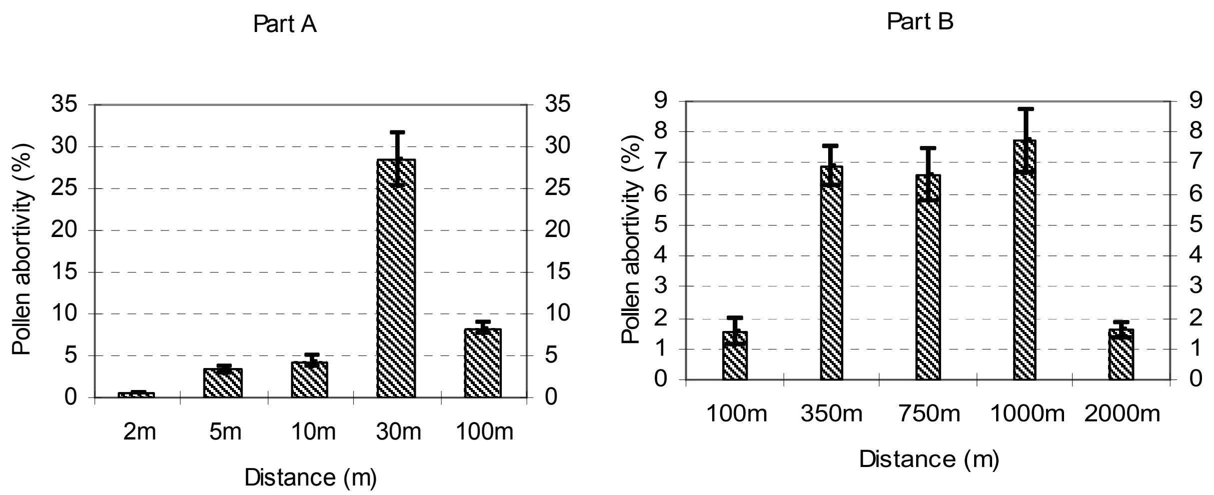

Fig. 3. The results of percentage of pollen abortivity with ±SEM in *Salix caprea* in relation to distance from the road in different prevailing wind directions (blowing parallel to the road in Part A and away from the road in Part B). At the control site a value of 3.9±1.1 was found.

the monitored site from stationary sources: Calandria Martin produce huge amounts of harmful substances such as SO_2 (817 t/yr 2008) and NO_x (317 t/yr 2008) [23], and the ZŤS TEES Martin machine plant.

Traffic volume, vehicle speed, and landscape (urban/rural) greatly affect traffic dust emissions [29, 30, 35]. Emission factors sharply decrease with increasing speed until a minimum is reached (CO – 40 $km \cdot h^{-1}$, HC – 55 $km \cdot h^{-1}$, NO – 50 $km \cdot h^{-1}$) [31].

The lowest plant responses to genotoxic agents were observed at site "C," with an average induction factor of 2.85. This result was not expected in regard to heavy traffic on the 1/18 road, where there were often traffic jams and traffic delays in the proximity of the sampling site. The highest traffic density (17,193 vehicles per 24 hours) was measured in this road section in comparison with other sections of this road [23]. The lowest environmental genotoxicity could be caused by adaptability of some species to chronic exposure [1, 25]. Results from the "K" site confirm that not only air pollution cause enhanced environmental genotoxicity [5] but partially also other sources such as contamination of water and soil by pesticides or communal waste (e.g. a settlement and cultivated area were located in the vicinity of the collection site).

The elevated environmental genotoxicity at all locations from future highway area was caused by traffic emissions from the roads passing near them. The highest rate of mutagenity (average IF 3.48) was found at the "DS" site as a result of exposure to high concentrations of traffic emissions. This pollution is maintained at ground level for a long time due to the valley end, frequent inversions, and light winds. They affect living organisms for a longer period, which causes increased frequency of abortion [4]. There is a higher traffic density than on the road sections passing near the other monitored sites. Frequent traffic accidents can cause contamination of water, soil, and air by mutagenic and carcinogenic substances [5]. The lowest rate of environmental genotoxicity at site "V" (average IF 2.94) was in a manner caused by traffic emissions from a less busy road and from the ŽOS Vrútky machine plant, which produced large quantities of SO_2 (73.7 t/yr 2008), nitrogen oxides (17.7 t/yr 2008), and CO (57 t/yr 2008). The results should be considered initial material for subsequent comparative studies during construction and after opening of the highway. We assume that the toxicity of these sites will be increased after construction of the highway, whereas the traffic will be diverted there to a great extent from the populated urban areas.

The results show a various sensitivity of individual species to the same environmental load caused by traffic emissions. As confirmed by this study, *Melilotus albus*, *Cichorium intybus,* and *Chelidonium majus* are considered suitable plant species for biomonitoring of a polluted environment. The most significant differences were found in *Melilotus albus* (IF ranged from 3.8 to 11.2) and *Cichorium intybus* (2.3-9.9), which showed higher sensitivity to air contaminants in comparison to *Chelidonium majus* (1.6-3.4). The same results were described by Mišík et al. [5]. A highly statistically significant increase ($P \leq 0.01$) in frequency of abortive pollen grains was observed at all monitoring sites in both clusters (A and B) in *Artemisia vulgaris*, *Cichorium intybus*, and *Melilotus albus*. The results described by Solenská et al. [4] confirmed this. The high sensitivity of *Cichorium intybus* also was documented by Mišík et al. [16]. Despite these results, we considered *Artemisia vulgaris* to be a less suitable species for detection of environmental quality by pollen abortion tests as in the above species, because the collection of usable material (undamaged, undried) for evaluation was quite difficult. The low response of *Lamium maculatum* (IF ranged from 0.5 to 2.5) to high concentrations of mutagenic substances indicated high plant resistance caused by its adaptation and tolerance to the pollutants concerned. *Pastinaca sativa* also has such mechanisms as a result of which the same rate of genotoxic load as at the control site (3.4±0.4) was observed at several contaminated sites. No significant differences in this species between samples from polluted versus control sites were documented in the study of Mičieta and Murín [1].

Factors such as prevailing direction and speed of air circulation are important for evaluating the correlation between increasing distance from the road and frequency of abortive pollen grains. The generally known inversely proportional relationship between wind speed and concentration of air pollution was confirmed in the work of Hitchins et al. [32]. Traffic emissions drift directly onto sampling sites at greater distances when the wind blows away from the road. The highest genotoxic load at a distance of 30 m from the road in Part A (Fig. 3) was caused by wind blowing parallel to the 1/18 road in the vicinity of the evaluated sites. The percentage of abortive pollen grains and distance from the road were positively correlated after the threshold limit with highest genotoxic effect (28%). Beyond this, a gradual decrease in these values was observed. Decrease in concentration of traffic emissions with increasing distance from the road began beyond this limit with the highest effect, which was at a smaller distance when the wind blew parallel to the road. Carneiro et al. [20] also observed a decrease in the abortion pollen grains as the distance increased from traffic, but they found the highest genotoxic load in 0m from the road. For comparison, the highest genotoxic effect (6.9%) was observed at a distance of 350 m in Part B. Dislocation of this effect to high distances from the source of emissions was caused by wind blowing perpendicular to the road. It was obvious that a distance of 1,000 m was too far and high a value of abortive pollen grains (7.7%) caused by another source of pollution (ŽOS Vrútky). As this study was focused on evaluation of traffic emissions, the value measured here was therefore not found to be useful for this assessment. For further results it would be necessary to evaluate in more periodic intervals.

Evaluating the influence of factors that can significantly manipulate the rate of the genotoxic effect is necessary for further assessment. Climatic conditions (temperature and relative humidity) affect spontaneous and pollution-induced mutations [33]. A high increase of mutation rates (spontaneous as well as pollution-induced) can be due to temperature shock during the regeneration phase. High temperature and low relative humidity cause the stomata to open, which results in a higher transport of pollutants to the target cells and subsequent increase of pollution-induced mutations [34]. A sudden and abrupt warming in the spring could also partly contribute to the increased pollution-induced mutations observed in this study. The average monthly temperature in the monitored town increased from 2.3ºC in March to 11.3ºC in April. In comparison with laboratory experiments, *in situ* monitoring provides results in a manner influenced by external factors such as fluctuating meteorological parameters or variations in airflow [28]. Not only air pollution, but also contamination of the ground, water, or soil can partly evoke increased genotoxicity at a monitored site [5]. Turčianska Valley is characterized by often calm temperature inversions and low average wind speeds that keep emissions in the ground layers of the atmosphere. The high values obtained in this study in both parts (A and B) were also affected by the above-mentioned factors. The environmental genotoxicity was only evaluated for one season (2009), but in order to record dynamic changes it is necessary to monitor in regular time periods over several years. Many studies have aimed to assess contamination of environmental components by chemical analyses or technological systems that determine the exact concentrations of pollutants [6, 29-31, 35]. However, these analyses do not allow us to detect the mixture of toxicity that plant bioassays do [4], which represent an additional method of providing basic information about the biological effects of pollutants [18].

Conclusions

According to results of the present study, the use of a pollen abortion assay with native flora in *in situ* conditions provides valuable information about the rate of contamination of the monitored environment. *Melilotus albus*, *Cichorium intybus*, and *Chelidonium majus* may be considered the most suitable of the species applied for detection of air quality in an urban area highly contaminated by traffic and industrial emissions. Markedly higher values of pollen abortivity were observed at sites with a significant impact of traffic emissions in comparison with sites away from their effects. Thus it can be concluded that vehicle traffic is one of the major sources of air pollution.

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