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Growth and development of the leaf blades of *Acer tataricum* in industrially contaminated environment

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Abstract: During one vegetative season, the growth and the development of the leaf blades of Tatarian maple (*Acer tataricum* L.) from heavily polluted area has been studied. The region under investigation was contaminated mainly with SO₂, N_xO_x, Pb, Zn, and Cu etc. The aim of the study was to compare the growth and the development of the leaf blades of *Acer tataricum* L. (Tatarian maple) from polluted field with those from non-polluted. Base on this to assess its tolerance to polluted conditions of the atmosphere, as well as to look for adaptive responses. The conducted study registered shorter time of the vegetative growth of the leaf blades in the commencement stages of the vegetative development. The leaves from polluted site had emergence approximately two weeks earlier compared with these from the control area. Faster linear growth of the leaf surfaces in the commencement stages of the development had been noted for the trees from polluted field. The spongy mesophyll had been reduced, as well as the common thickness of the leaf blades of the tree plants from the contaminated region. The observed changes are regarded as adaptation of the plant to the polluted environment, i.e. as tolerance.

Key words: *Acer tataricum* L., Tatarian maple, air pollution, and sulphur dioxide, leaf blade structure

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Introduction

Gaseous pollutants include sulfur dioxide, nitrogen oxides, organic compounds, and trace metals. After entering the atmosphere, they may be oxidized (sulfur dioxide going to sulfate, the nitrogen oxides going to nitrate), associate with aerosols, or be degraded. Plants have a very large surface area and their leaves function as an efficient pollutant-trapping device. Vegetation naturally cleanses the atmosphere by absorbing gases and some particulate matter through leaves. Therefore, their assimilative organs are directly affected from the industrially polluted air.

Polluted air is a stress factor that contributes to the decline of trees. Whether or not a pollutant causes

acute or chronic damage depends upon tree species. Individual trees within a species often display varying degrees of sensitivity to the same pollutant. Sensitivity to damage by simulated acid rain has been reported to vary with species (Evans & Curry 1979; Haines et al. 1980; Evans, Gmur & Mancini 1982), growth conditions (Evans 1982) and with age of leaves (Evans & Lewin 1981). In general, developing young leaves are more affected by acid rain than full-developed mature leaves (Evans et al. 1978; Evans & Curry 1979; Paparozzi & Tukey 1983; Adams et al. 1984; Evans 1984; Crang & McQuattie 1986; Rinallo et al. 1986).

Effects of acid rain on plant growth are often species-specific. Some plants show enhanced growth

(Wood & Bormann 1977; Evans & Lewin 1981; Lee et al. 1981; Raynal et al. 1982; Troiano & Jacobson 1982), some no effect (Lee et al. 1981) and other reduce growth (Lee et al. 1981; Evans et al. 1981; Raynal et al. 1982; Johnson et al. 1982).

The aim of the investigation was to trail and compare the dynamics of the growth and the development of the leaf blades and their structures from the trees of Tatarian maple (*Acer tataricum* L.) growing in polluted and non-polluted environment, as well as to look for adaptive responses. Base on this to evaluate the sensitivity of the *Acer tataricum* L. (Tatarian maple) to the industrially contaminated air. There are disagreements in the opinion of the tolerance of *Acer tataricum* L. to industrial contamination. More than 50% of the authors assess *Acer tataricum* L. as high resistant species (Mamaev 1969; Ilkun 1971; Kachalov 1970; Antipov 1974; 1975). Nevertheless, there are some reports that refereed Tatarian maple (*Acer tataricum* L.) as less resistant or sensitive one (Krasinskii 1950; Tolonnikova 1970). These dissensions are due to the lack of the uniform method or criteria for assessment as well as from the different conditions of the environment in that the tolerance of plants finds its extrapolation.

Materials and methods

Characteristics of the regions

The region of metallurgical plant Kremikovtzi (42°47'N; 23°30'E) is heavily polluted with SO₂, N_xO_x, Pb, As, Zn, Cu etc. The major industrial processes generated particles (dust). The main air pollutant is the sulfur dioxide. During the investigation period, the concentration of sulfur dioxide in the observed area was 0.5 mg/m³.

The National Park Vitosha (42°30'N; 23°15'E) was chosen as a control, both fields have tempered climate.

Common characteristic of *Acer tataricum* L. – Tatarian maple

Tatarian maple is a small deciduous tree or large shrub with a slow growth rate (Dirr 1983; Little 1980). It is a small rounded to broad-rounded tree similar to but slightly larger than Amur maple (*Acer ginnala*), and having leaves that are either unlobed or barely lobed.

Tatarian maple (*Acer tataricum* L.) is found in deciduous oak scrub, rocky slopes and river valleys at altitudes between 500–1700 m (Davis 1965). It prefers a good moist well-drained soil on the acid side and sunny positions (Bean 1981; Thomas 1992). Grows well in heavy clay soils (Bean 1981). Bad companion plant, inhibiting the growth of nearby plants (Philbrick & Gregg 1979; Riotte 1978). Tatarian ma-

ple (*Acer tataricum* L.) has been reported as high resistant (Lipa 1952; Mamaev 1969; Voloshin 1968; Kachalov 1970; Ilkun 1971; Chuvaev 1972; Erohina 1973; Antipov 1974; 1975), resistant or less resistant and even sensitive species to the air contamination (Krasinskii 1950; Tolonnikova 1970).

Plant material and methods

The study examined the leaf blades of *Acer tataricum* L. The plant material was collected monthly from April to October. Samples were taken randomly (30 leaf blades from each tree) from the south side of the crown at 160–200 cm of the trees (10–15 trees) from both fields. The trees were of a similar age, sun exposure and had uniform height and growth form. The middle parts of the leaf blades were cut and fixed in 90% ethanol – 90 cm³, ice acetic acid – 5 cm³ and formalin – 5 cm³. Standard histological techniques and light microscopy were used to examine the anatomical characteristics of the leaf blades. The cross-sections of the leaf blades were prepared and observed under light microscopy, measured, drawn and photographs were taken. The measurements were repeated 30 times per one parameter. Cell size and thickness of the layers were assessed statistically with t-test. The influence of the pollution on the linear growth of the leaf blades, length and width, was evaluated with ANOVA (two way).

Results

The field's observations revealed that the leaf blades of the trees of *Acer tataricum* L. (Tatarian maple) from the polluted region have faster growth in the commencement stages of the vegetative development compared with the growth of the leaf blades from the control trees (Table 1).

In polluted field, the leaf blades were appeared earlier approximately two weeks before these from the control field. The expansion of the leaves from polluted region was accelerated significantly compared with those that were registered for the control. During April the control, leaves had 3,3 times shorter length than polluted leaves (Table 1). The anatomical measurements of the plant material collected during April discovered that the leaf thickness value from the polluted site was significantly lower than that from the non-polluted. The difference of the leaf thickness was mainly due to the smaller thickness of the spongy mesophyll in the leaf blades of the tree plants growing in the polluted region than that measured for the control (Table 1). The registered coefficient of palisadness was 45% for polluted plants and 32% for non-polluted ones.

In May, the surfaces of the leaves of the control plants were significantly bigger in length and width than these registered for the tree plants from the pol-

Table 1. Results of the linear growth of the leaf blades – ANOVA (two way)

Trait	Control area							Polluted area							ANOVA (P>F)		
	April	May	June	July	October	April	May	June	July	October	Pollution	Time	Interaction (P×T)				
Length of leave (cm)	1.9 (0.3)	8 (0.4)	7.5 (0.8)		6.4 (0.8)	7.9 (0.4)	8.2 (0.9)				< 0.001	< 0.001	< 0.001				
Width of leave (cm)	0.9 (0.2)	5.5 (0.3)	5 (0.8)		3.6 (0.9)	4.6 (0.4)	5 (0.6)				< 0.001	< 0.001	< 0.001				
Leaf thickness (µm)	147 (2)	127.7 (2.4)	191.5 (2)	163 (2)	136.91 (2.9)	109 (2)	160.6 (2.4)	199.6 (5.28)	197.16 (2.8)		< 0.001	NS	< 0.001				
Upper cuticle thickness (µm)	3.25 (0.0)	3.125 (0.0)	3.5 (0.7)	3.6 (0.7)	3.5 (0.7)	4.65 (1.02)	3.7 (0.8)	5.3 (0.5)	5.58 (1.1)		< 0.001	< 0.001	< 0.001				
Upper epidermis thickness (µm)	15.03 (1)	15.8 (0.96)	16.8 (1.5)	15.3 (1.3)	17.4 (3.1)	14.2 (1.5)	15.5 (2.5)	16 (3.5)	14.5 (1.05)		< 0.001	< 0.001	< 0.001				
Palisade mesophyll thickness (µm)	37.63 (2.9)	41.38 (6.01)	77.8 (3.41)	71.9 (5.2)	43.09 (4.9)	37.8 (6.5)	80.3 (9.7)	110.4 (19.5)	107.26 (8.6)		< 0.001	< 0.001	< 0.001				
Spongy mesophyll thickness (µm)	78.12 (10.5)	55.18 (6.47)	78.4 (10.4)	59.52 (6.32)	59.2 (6.9)	45.57 (3.9)	46.81 (4.9)	54.87 (6.85)	57.66 (4.8)		< 0.001	< 0.001	< 0.001				
Lower epidermis thickness (µm)	9.76 (1.9)	9.14 (1.33)	11.8 (1.05)	9.3 (1.3)	10.5 (1.7)	9.76 (1.3)	11.2 (1.5)	9.9 (1.3)	8.99 (1.6)		< 0.001	< 0.001	< 0.001				
Lower cuticle thickness (µm)	2.79 (0.3)	3.03 (0.185)	3.05 (0.18)	3.1 (0)	3.1 (0)	3.12 (0.00)	3.1 (0.0)	3.1 (0)	3.1 (0)		< 0.001	< 0.001	< 0.001				
K – Coefficient of palisadness (%)	32%	43 %	49%	54%	42%	45%	60 %	63%	66%	64%							

luted field (Table 1). The rate of the linear growth of the leaves from polluted site was slow down. There were no significant differences in the structure of the leaf blades, with exception of the thicknesses of the spongy mesophyll (Table 1). The coefficient of palisadness grew with 15% for polluted trees and with 11% for the control. During June, there were no significant differences of the measured parameters of the leaf surfaces (Table 1). The only significant difference registered for the structure of the leaves between the polluted and the control samples was the width of the spongy parenchyma. The thickness of the leaf blades from polluted site was smaller than those measured for the control. The tree plants developed less width of spongy tissue under contaminated conditions than those from the unpolluted area had. The difference of the coefficient of palisadness between polluted and control was 14 %. During the all observed period the coefficients of palisadness, stayed greater in the tree plants from the industrially contaminated field in comparison to these measured for the plants from the unpolluted region (Table 1).

In July the main differences that had been registered between polluted and control plants were the increasing of the upper cuticle layer and the width of palisade mesophyll (Table 1). In October the significance of these differences had been kept and in addition for the plants from polluted field were noted reducing of the size of epidermal cells for both upper and lower epidermis (Table 1).

The difference of the coefficients of palisadness grew up to 22% at the end of the vegetative period of the development of the leaf blades (Table 1).

On Figure 1 are given the dynamics of the growth of the different parameters of the structure of the leaf blades of Tatarian maple (*Acer tataricum* L.) in one vegetative period.

Discussion

The conducted investigation noted that in the polluted field, the leaf blades were appeared earlier approximately two weeks before these from the control field. The acceleration of vegetative development was manifested as earlier appearing of the leaves and faster growth, expanding and forming of the leaf surfaces in the woody trees from the polluted field.

The predominant emphasis on harmful effects of environmental stresses on growth of woody plants has obscured some very beneficial effects of such stresses. Slowly increasing stresses may induce physiological adjustment that protects plants from the growth inhibition and/or injury that follow when environmental stresses are abruptly imposed. In addition, short-term exposures of woody plants to extreme environmental conditions at critical times in their development often improve growth (Kozłowski &

Pallardy 2002). The acceleration of the commencement stages of the development and the registered faster linear growth of the leaves of *Acer tataricum* L. (Tatarian maple) from polluted trees, in April, might be accepted as adaptation of the tree plants to the industrially contaminated environment, i.e. as tolerance.

In general, developing young leaves are more affected by acid rain than full-developed mature leaves (Evans et al. 1978; Evans & Curry 1979; Cragg & McQuattie 1986; Rinallo et al. 1986). As emphasized by Keever and Jacobson (1983), rapidly expanding leaves may have low rates of wax production and thereby develop discontinuities in the coverage of waxes over the leaf surface (Baker & Hunt 1981), i.e. they might be more susceptible to injury by acid rain. Rainfall acidity is often higher in spring and summer (Hornbeck et al. 1977), a period coinciding with rapid leaf expansion, and the potential for foliar damage would therefore be greatest at this time of the year. That's way the faster development of the leaves in April that slowed down in May in trees from polluted region might be accepted as adaptive response to the polluted conditions, because in this way the woody plant escape the peak period of acidic rainfalls. The plant met the highest peak of acidic rainfalls with fully developed mature leaves, which many authors decided that are less sensitive to adverse environmental conditions (Evans et al. 1978; Cragg & McQuattie 1986; Rinallo et al. 1986). In the end of April, the leaves of *Acer tataricum* L. (Tatarian maple) from polluted field were approximately fully expanded and in May the rate of growth slowed down.

The degree of damage, which trees and herbs suffer when exposed to acid rain, varies widely among species (Wood & Bormann 1974, 1975; Evans & Curry 1979; Lee & Weber 1979; Haines et al. 1980; Neufeld et al. 1985), but the underlying mechanisms are poorly understood at present. Plant adaptation to changing environmental factors involves both short-term physiological responses and long-term physiological, structural, and morphological modifications (Gravano et al. 2003; Novak et al. 2003; Vander et al. 2001). These changes help plants minimize stress and maximize use of internal and external resources. The structure of the leaf blades is important place in determines the response of the tree to the air contamination. Gaseous pollutants, such as ozone and sulfur dioxide, enter plants through natural openings, usually stomates. After passing through the stomates of the lower epidermis, the toxic gas meets the spongy mesophyll (Nikolaevski 1963; Ilkun 1971, 1978). Once inside the leaf, gases diffuse into intercellular spaces and may be absorbed by water films to form acids or react with inner-leaf surfaces (Smith 1990). Therefore, many authors consider that one of the main criteria of resistance to air pollution is the higher coefficient of palisadness

(Nikolaevski 1963; Ilkun 1971, 1978; Bennett et al. 1992; Ferdinand 2000). The observed anatomical structures of the leaf blades of *Acer tataricum* L.

(Tatarian maple) during the entire vegetative period were characterized with high coefficients of palisadness for the woody trees under contaminated en-

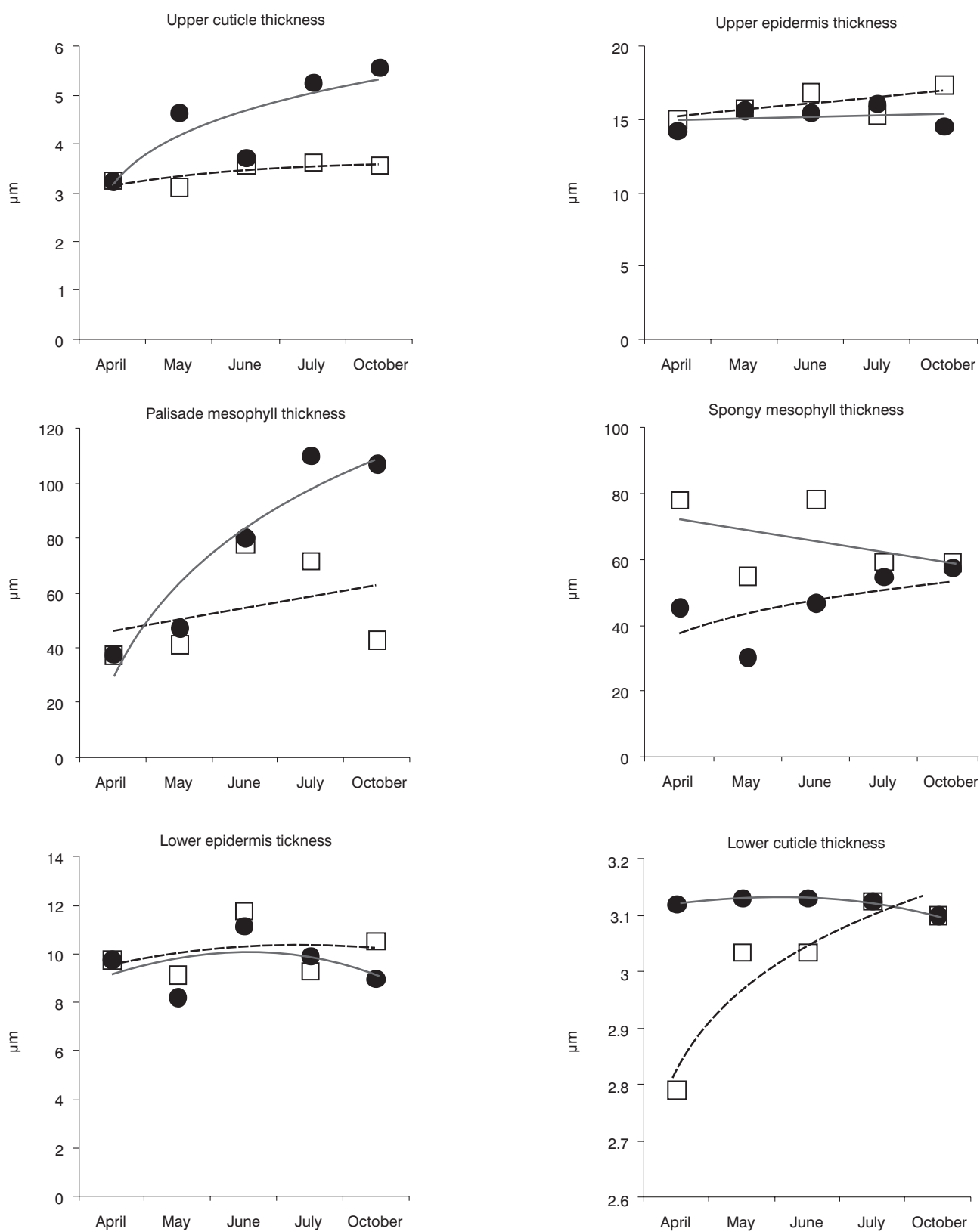


Fig. 1. Dynamics of the anatomical characteristic of the leaf blades of *Acer tataricum* L. during on vegetative season from polluted – solid line (●) and unpolluted – dashed line (□) environment

vironmental conditions. In April, May and June the high value of the coefficient of palisadness was due to the low rate of the thickness of spongy parenchyma and the thickness of the leaf blades were smaller in the tree plants from the polluted region. In the rest of the vegetative period (from July up to October), the high level of the coefficient of palisadness was caused by the expansion of the thickness of the palisade mesophyll.

Maturation of the leaves cause changes in wax and cuticle development (Juniper 1959; Martin & Juniper 1970; Baker & Hunt 1981). Young leaves of one isoline of *Glycine max* had correlate with greater damage by acid rain than older leaves (Keever & Jacobson 1983) and the reason might be was as a result of these changes of the leaf-covered tissues. The conducted investigation revealed that in July the thickness of the upper cuticle layer grew significantly in polluted plants of Tatarian maple (*Acer tataricum* L.). In October had been noted that the tree plants from polluted area not only kept those difference of the mean of the upper cuticle layer, but they were reduced the size of the epidermis cells in both – adaxial and abaxial side of the leaves. Ferrenbaugh (1976) reported that cell sizes were smaller and intercellular leaf spaces reduced in leaves of *Phaseolus vulgaris* exposed to acid rain. Many authors were reported similar results of reducing the sizes of epidermis cells of the leaf blades in plants exposed to different levels of air pollutions (Ferrenbaugh 1976; Pal et al. 2000; Aggarwal 2000).

Conclusion

The conducted study registered acceleration of the vegetative growth of the leaf blades of *Acer tataricum* L. in the commencement stages of the vegetative development. The leaves from polluted site had emergence approximately two weeks earlier compared with these from the control area. Faster linear growth of the leaf surfaces in the commencement stages of the development had been noted for the tree plants from polluted field. The leaves from polluted site were fully expanded in the end of April. The spongy mesophyll had been reduced, as well as the common thickness of the leaf blades of the tree plants from the contaminated region. The coefficient of palisadness was with higher value for the trees from the contaminated region, than those measured for the control. The palisade tissue grew during summer as well as the upper cuticle layer. The observed changes are regarded as adaptation of the plant to the polluted environment, i.e. as tolerance.

The obtained results showed that the tree plants of Tatarian maple (*Acer tataricum* L.) were well adapted to the current environmental conditions and the plants could be accepted as tolerant. Under contaminated conditions of the environment *Acer tataricum* L.

obtained physiological and anatomical changes in the growth and the development of the leaf blades.

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