

Anatomy of Leaves of *Betula pendula* (Roth.) Affected by Air Emissions in Industrial Area of Kemerovo City

^{1,2}*Olga Alexandrovna Neverova,*

²*Olga Michajlovna Legoshchina and* ³*Anatolij Alexandrovish Bykov*

¹Kemerovo Institute of Food Science and Technology, Kemerovo, Russia

²Institute of Human Ecology SB RAS, Kemerovo, Russia

³The Kemerovo branch of the Institute of computational technologies SB RAS,
Kemerovo, Russia

Abstract: Modeling of air pollution of the industrial zone of Kemerovo city using a special module of the ERA software complex. The complex index of air pollution by priority pollutants (nitrogen oxides, sulfur dioxide, carbon monoxide and polycyclic aromatic hydrocarbons, including benzopyrene and suspended solids) was calculated using the modeling data. It is determined that the zone of maximum pollution from emissions sources reaches 4 km in the prevailing wind direction. Anatomic and morphological characteristics of common birch *Betula pendula* (Roth.) leaves affected by emissions of the industrial zone of Kemerovo city were investigated. The maximum changes in these characteristics were identified at a distance of 1 km from the industrial zone. The most clear negative changes appeared as the reduction in the thickness of the cuticle and width of the lower epidermis cells. A statistically significant increase in the thickness of the palisade and spongy mesophyll, thickness of a lamina and thickness of the upper epidermis tissue were determined as the adaptive changes. There are significant correlations of the complex index of air pollution with anatomic-morphological characteristics of birch lamina: thickness of lamina, thickness of the lower epidermis, thickness of palisade and spongy mesophyll. The study results revealed the significant effect of emissions of industrial zones on the anatomical and morphological transformations of the lamina of *Betula pendula* (Roth.).

Key words: Industrial zone • A complex index of air pollution • Anatomical characteristics of leaves
• Adaptive changes

INTRODUCTION

The industrial zone of Kemerovo city including Kemerovo Hydro power plant, Khimprom Ltd. and Koks Ltd., is the general source of air pollution in the city. This zone is located on the border of Central and Zavodskoy districts of the city close to the residential zones. Nitrogen oxides, sulfur dioxide, carbon monoxide and polycyclic aromatic hydrocarbons, including benzopyrene and suspended substances are general emissions of these industries.

It is known that the wood plants characterize by high sensitivity to anthropogenic load, therefore, can

serve as the appropriate environmental indicators of the conditions of the urban territories [1]. It is noted in the literature sources that chronic effect of air toxicants causes the serious changes in the anatomical structure of plant leaves and needles [2, 3].

Therefore, the aim of our study was to examine the anatomic and morphological characteristics of leaves of *Betula pendula* (Roth.) and their relationship with the total air pollution in Kemerovo city affected by industrial emissions.

To fulfill the objectives, the conditions of pollution were modeled on the test sites and anatomic-morphological signs of leaves of *Betula pendula* (Roth.)

were studied. This work was supported by the integration project no. 84 of Siberian Branch, Russian Academy of Sciences.

MATERIALS AND METHODS

Six test (monitoring) sites (MS) located along the flame trail of distribution of emissions from industrial zone were selected for monitoring (MON) (according to southwestern wind direction) (Fig.1).

Studies were conducted during summer of 2009–2011. To estimate the average parameters over a long period of air pollution in Kemerovo city, a special module of the ERA software complex developed at the Voeikov Main Geophysical Observatory (St. Petersburg, Russia) was used to verify the conformity of the short- and long-term models [4-5]. ERA complex can be applied for standard calculations of the air pollution (www.logos-plus.ru) and allows the use of standard databases accumulated in the formats of the ERA software complex, easy and developed interface and all text and graphics capabilities for representation of the results (including creation of the digital and raster maps). For linear and site sources, there is a procedure of integrating with step depending on remote point distance from the source and a step is limited by 3% computational error.

The general ratio for calculation of the average value over a long period of concentration of C in the point with polar coordinates (r, φ) relatively to the source is:

$$C(r, \varphi) = \frac{p_1(\varphi)M}{r} \int_0^\infty du \int_0^\infty d\lambda p_2(u) p_3(\lambda) q(r, u, \varphi, \lambda, H_e) \quad (1)$$

where M (g/s) is average emission from the source over period of averaging. Description of definitions and formula for the integrand function q and the effective height of the source H_e are presented in [5]. The equation (1) includes three functions representing the density distribution: wind direction $p_1(\varphi)$; wind speed $p_2(u)$; dimensionless parameter λ of intensity of turbulent mixing $p_3(\lambda)$. For normative calculations, these distributions that determine the behavior of long-term air pollution in the vicinity of the source were received together with the other parameters at the Voeikov Main Geophysical Observatory. The function $p_1(\varphi)$ was obtained from the standard 8-rhumba wind rose by interpolation, which is defined in [5].



Fig. 1: Location of the test sites for monitoring on the map of the Kemerovo city.

The inventory data from the materials of the consolidated volume of maximum permissible emission (MPE) of Kemerovo city [6] of priority emissions of the enterprises of the industrial zones, nitrogen oxides, sulphur dioxide, carbon monoxide, benzopyrene and suspended solids (ash, soot, etc.) were also used in calculations.

Conditional dimensionless complex index (CI) of total average annual air pollution was calculated during modeling, $CI = C_1 / MPC_{a1} + C_2 / MPC_{a2} + \dots + C_n / MPC_{an}$, where C is the average ground concentration, MPC_{a} —average daily maximum permissible concentrations (MPC), the indices 1, 2, 3, ..., n are the pollutants mentioned above. This indicator is not hygienic standard criterion therefore only a part of the considered substances possess the unidirectional effect on a man, which is “the total anthropogenic load” created by the industry through atmospheric transport of pollution on the certain territory of the city.

Betula pendula (Roth.) served as the object of anatomical studies in average-age generative state, growing in the zone of the industrial emissions. Five trees from each test site were used in experiment. One-year shoots with leaves were collected from the middle part of the South side crown and fixed in the 60% ethyl alcohol solution. For anatomical studies, the middle part of each leaf was transected and placed in glycerin. The study of the anatomical and morphological characteristics of the leaves was performed using a microscope Aksioskop-2+ (models ZEISSN HBO103 and N XBO75 (Germany)) with

the data delivery software. The thickness of the lamina, cuticle, the upper and lower epidermis, palisade and spongy mesophyll were determined. The study results were statistically processed using Statistica ver. 6.0 software. Reliability of differences in anatomical parameters was determined by the Student *t*-test at a significance level of 0,05.

RESULTS AND DISCUSSION

The values of the complex index (CI) of air pollution on test sites are presented in Table 1. The data shows that the CI values are distributed in the decrease order of the distance from the industrial zone.

The significant difference between the values of CI was noted between 1-4 test sites, at a distance of 4 km from the pollution sources. CI values from 1 to 4 test sites are distributed in following order $17,97 > 10,43 > 8,13 > 6,87$. Significant differences in the CI values were not observed from 4 to 6 test sites (at a distance 4-6 km from the emission sources), as this indicator varies within the range from 6,87-6,21.

It was determined that the changes in anatomy and morphology of lamina of *Betula pendula* (Roth.) are nonlinear and depend on the distance from industrial zone.

More significant differences between the studied parameters were observed at a distance 2 km from emission sources at test sites 1 and 2 compared with test site 6, which is the most distanced from the industrial zone. Moreover, there are negative and adaptive changes found in *Betula pendula* (Roth.) A tendency to reduction of the thickness of the cuticle and width of the lower epidermis cells compared with test site 6 by 22 and 17 %, respectively was observed at the distance 1 km (test site 1) (Table 2).

The significant adaptive changes in anatomy and morphology of *Betula pendula* (Roth.) were identified in the adjacent zone to the industrial zone that, apparently, provides the birch with high resistance to the pollution. In particular, there was statistically significant increase in the thickness of the palisade and spongy mesophyll (by 27 and 28 %), thickness of a lamina (by 22 %) and the thickness of the upper epidermis tissue (by 19%) found in the *Betula pendula* (Roth.) on test site 1 compared with test site 6 (Table 2, Fig. 2). It is known that the leaf mesophyll is a complex structure formed by different tissues (palisade and spongy cells) and filling internal space of the leaf. The total surface of mesophyll cells determines the rate of photosynthesis [7].

The studied parameters on the test sites 3 and 5 changed without clear tendency and their values were insignificant compared with test site 6.

Table 1: The values of the complex index of air pollution in the tested sites

| Number of test site | Type of test site | Complex index |
|---------------------|---|---------------|
| 1 | park at the entrance of the hydroelectric power station | 17,968 |
| 2 | Gorkiy park (near sports complex) | 10,429 |
| 3 | territory of Yunatov station (Shakhterov street, 10) | 8,132 |
| 4 | territory of Zhuravlik sanatorium (Tereshkovoi street, 7) | 6,871 |
| 5 | Shakhterov park (between Shakhterov prospekt and Institutsкая street) | 6,281 |
| 6 | the yard of rural hospital (Avrory street, 12) | 6,208 |

Table 2: The anatomical characteristics of the structure of the birch leaf lamina affected by industrial pollution of the Kemerovo city.

| Characteristics | Number monitoring site | | | | | |
|--|------------------------|-----------------|-----------------|------------------|-----------------|-----------------|
| | 1 | 2 | 3 | 4 | 5 | 6 |
| Thickness of lamina, μm | 176.1 \pm 8.0* | 140.4 \pm 5.6 | 137.9 \pm 6.5 | 165.3 \pm 3.4* | 148.1 \pm 1.3 | 144.2 \pm 4.4 |
| Height of the upper epidermis cells, μm | 23.5 \pm 1.0* | 21.6 \pm 0.5 | 19.6 \pm 0.7 | 29.4 \pm 1.4* | 19.4 \pm 0.9 | 19.7 \pm 0.7 |
| Width of the upper epidermis cells, μm | 22.1 \pm 1.0 | 19.9 \pm 0.9 | 22.4 \pm 0.7* | 22.9 \pm 0.8* | 22.8 \pm 0.9* | 20.1 \pm 0.7 |
| Height of lower epidermis cells, μm | 14.2 \pm 0.5 | 11.2 \pm 0.6* | 10.6 \pm 0.4* | 12.9 \pm 0.6 | 12.4 \pm 0.5 | 12.8 \pm 0.5 |
| Width of lower epidermis cells, μm | 17.7 \pm 0.9* | 20.1 \pm 0.9 | 21.8 \pm 1.1 | 20.0 \pm 0.9 | 19.8 \pm 0.8 | 21.3 \pm 0.8 |
| Thickness of palisade mesophyll, μm | 72.2 \pm 2.8* | 49.7 \pm 2.3 | 48.3 \pm 2.1* | 58.2 \pm 1.1 | 58.5 \pm 1.9 | 56.7 \pm 1.6 |
| Thickness of spongy mesophyll, μm | 74.6 \pm 3.1* | 62.7 \pm 3.1 | 60.0 \pm 2.2 | 66.6 \pm 2.1* | 64.4 \pm 1.4 | 58.2 \pm 1.7 |
| Width of cuticle, μm | 4.9 \pm 0.12 | 4.1 \pm 0.16 | 4.7 \pm 0.22 | 6.2 \pm 0.32 | 5.4 \pm 0.17 | 6.3 \pm 0.28 |

Note: * - significant differences from test site 6

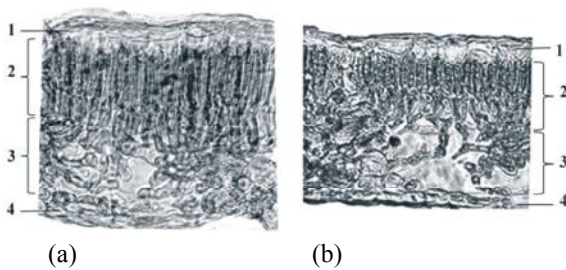


Fig. 2: Transversal section of a leaf of silver birch growing affected by industrial emissions. A, - test site 1 (park at the entrance of the hydroelectric power station), B, - test site 6 (the yard of rural hospital). 1, - upper epidermis, 2, - palisade mesophyll, 3, - spongy mesophyll, 4, - lower epidermis.

There are many papers in international literature on the effect of environmental factors on the anatomical and morphological characteristics of leaves of woody plants. The literature analysis shows a great number of contradictory data on the anatomical and morphological transformations of the plants affected by technogenic pollution of the environment that, apparently, is determined by the species features of the trees and the degree of anthropogenic impact. In particular, Bruno Francisco Sant'Anna-Santos *et al.* [8] has investigated the action of acid rain on the leaf structure of *Genipa americana* L. and determined the necrotic spots on the leaf lamina, the cuticular ruptures in the necrosis zone, the destruction of the cells of the epidermis and palisade mesophyll and hypertrophy of the spongy mesophyll. Liezel, M.M. *et al.* [9] have found the changes in the length of the leaf of *Tithonia diversifolia* (Hemsl), length of internodes, thickness and density of the epidermis affected by transport emissions in Baguio city. Kurteva M. and Stambolieva K. [10] have noted the reduction of the linear growth of annual shoots and leaves of *Acer pseudoplatanus* L., *Acer platanoides* L. and *Betula pendula* (Roth.) affected by industrial pollution in urban zones of Sofia city (Bulgaria). Ksanen *et al.* [11] has revealed the negative impacts of ozone and CO₂ on anatomical parameters of the leaves of *Betula pendula* (Roth.) Higher concentrations of CO₂ and ozone caused a reduction in the thickness of leaves in particular; ozone raised a reduction of the thickness of palisade mesophyll. Riikonen *et al.* [12] has found that the influence of ozone and CO₂ on the birch reduces the size and density of the cells of the epidermis.

However, there are numerous studies, which describe the adaptive changes of morphological, anatomical and physical-chemical characteristics of the plants affected by unfavorable environmental factors, allowing them to survive unfavorable conditions [13-19].

In our study, the results of the correlation analysis revealed a significant relationship between anatomical indicators of the birch leaves with CI of the air pollution as well as with thickness of leaf lamina ($r = 0,37, p < 0,05, n = 82$), the thickness of the lower epidermis ($r = 0,22, p < 0,05, n = 106$) and thickness of palisade and spongy mesophyll ($r = 0,43, p < 0,05, n = 100$ and $r = 0,39, p < 0,05, n = 100$). This evidences the significant influence of industrial emissions on anatomical and morphological transformations of the leaf lamina of *Betula pendula* (Roth.).

CONCLUSIONS

Modeling of air pollution in industrial zone of Kemerovo city has revealed the zone of the maximal pollution at the distance of 4 km from pollution sources in wind direction. The changes of anatomic and morphological characteristics of the leaf lamina of *Betula pendula* (Roth.) were determined in conditions of industrial emissions of Kemerovo. The maximum changes in anatomy and morphology of the birch leaves were identified at a distance of 1-2 km from the industrial zone. Among negative changes, we have determined the reduction of thickness of the cuticle and width of the cells of the lower epidermis. The maximum adaptive changes of the birch were revealed at the distance of 1 km from industrial zone, which included a statistically significant increase in the thickness of the palisade and spongy mesophyll, thickness of a leaf lamina and thickness of the upper epidermis that, apparently, ensures the high adaptation of the birch to pollutants.

The reliable correlation of the complex index of air pollution with the thickness of the leaf plate, thickness of the bottom of the epidermis, thick palisade and spongy mesophyll determined in our study, confirms the significant impact of the emissions of the industrial zone on the anatomical and morphological transformations of the leaf lamina of *Betula pendula* (Roth.).

REFERENCES

1. Nikolaevskiy, V.S., 2002. Assessment of the Environmental Pollution and the Condition of Land Ecosystems by Phytoidication Methods. Pushkino: VNIILM, pp: 220.

2. Zotikova, A.P., O.G. Bender, R.O. Sobchak and T.P. Astafurova, 2007. Comparative Assessment of Structural-Functional Organization of Leaf Apparatus of Coniferous on the Territory of Gorno-Altaysk City. *Vestnik TGU*, 299(1): 197-200.
3. Kurovskaya, L.V., 2002. Morphological and functional peculiarities of coniferous in the urban environment, Cand. Sci. (Biol.) Dissertation, Tomsk, pp: 22.
4. A Method of Calculation of the Concentration of Harmful Substances of Industrial Emissions in Atmosphere, 1987. Leningrad: Gidrometeoizdat, pp: 92.
5. A Method of Calculation of the Average Concentrations of Harmful Substances in Atmosphere over a long period (Supplement to OND-86), 2005. St. Petersburg: Voeikov Main Geophysical Observatory, pp: 15.
6. Azhiganich, T.E., T.G. Alekseichenko, A.A. Bykov *et al.*, 2003. Summary results of calculations of environmental pollution of Kemerovo city for standardization of emissions and diagnostics. In Proceedings of V city scientific-practical conference "Urban ecology: Problems and solutions". Kemerovo, pp: 41-45.
7. Ivanova, L.O. and V.I. P'yankov, 2002. The Influence of Environmental Factors on Structural Parameters of the Leaf Mesophyll. *Botanicheskiy Zhurnal*, 12(87): 17-28.
8. Sant'Anna-Santos, B.F., L.C. da Silva, A.A. Azevedo and R. Aguiar, 2006. Effects of Simulated Acid Rain on Leaf Anatomy and Micromorphology of *Genipa americana* L. (Rubiaceae). *Braz. Arch. Biol. Technol.*, 49(2): 313-321.
9. Liezel, M.M., G.M. Deemson and P.L. Fideliz, 2013. Morpho-Anatomical Characterization of *Tithonia diversifolia* (Hemsl.) Gray Growing on Sites Exposed to Vehicular Emissions. *International Journal of Plant, Animal and Environmental Sciences*, 3(3): 168-172.
10. Kurteva, M. and K. Stambolieva, 2007. *Acer pseudoplatanus* L., *Acer platanoides* L. and *Betula pendula* (Roth.) as bioindicators of urban pollution in Sofia. *Silva Balcanica*, 8(1): 32-46.
11. Ksanen, E.O., J.R. Nenwz, S. Kaakinenz, T.H. Enw and E. Vapaavuoriz, 2005. Structural Characteristics and Chemical Composition of Birch (*Betula pendula*) Leaves are Modified by Increasing CO₂ and Ozone. *Global Change Biology*, 11: 732-748. doi: 10.1111/j.1365-2486.2005.00938.x
12. Riikonen, J., K.E. Percy, M. Kivima, M.E. Kubiske, N.D. Nelson, E. Vapaavuori and D.F. Karnosky, 2010. Leaf Size and Surface Characteristics of *Betula papyrifera* Exposed to Elevated CO₂ and O₃. *Environmental Pollution*, 158: 1029-1035.
13. Wyszowski, M. and J. Wyszowska, 2003. Effect of Soil Contamination by Copper on the Content of Macro Elements in Spring Barley. *Polish Journal Natural Sciences*, 14: 309-320.
14. Kovacic, S. and T. Nikolic, 2005. Relations between *Betula pendula* (Roth.) (Betulaceae) Leaf Morphology and Environmental Factors in Five Regions of Croatia. *Acta Biologica Cracoviensia*, 47: 7-13.
15. Gomes, M.P., T.C.L. Marques, L.S.M., Nogueira, E.M. Castro and A.M. Soares, 2011. Ecophysiological and anatomical changes due to uptake and accumulation of heavy metal in *Brachiaria decumbens*. *Scientia Agricola*, 68: 566-573.
16. Ristic, Z. and M.A. Jenks, 2002. Leaf Cuticle and Water Loss in Maize Lines Differing in Dehydration Avoidance. *Journal of Plant Physiology*, 159: 645-651.
17. Kapitonova, O.A, 2002. Specific Anatomical Features of Vegetative Organs in Some Macrophyte Species under Conditions of Industrial Pollution. *Russian Journal of Ecology*, 33(1): 59-61.
18. Uribe-Salas, D., C. Saenz-Romero, A. Gonzalez-Rodriguez, O. Tellez-Valdez and K. Oyama, 2008. Foliar Morphological Variation in the White Oak *Quercus rugosa* Nee (Fagaceae) along a Latitudinal Gradient in Mexico: Potential Implications for Management and Conservation. *Forest Ecology and Management*, 256(12): 2121-2126.
19. Snejana, B.D., 2005. Leaf Blade Structure and the Tolerance of *Acer negundo* L. (Box elder) to the Polluted Environment. *Dendrobiology*, 53: 11-16.