*Folia Forestalia Polonica, Series A – Forestry, 2019, Vol. 61 (2), 112–122*

**ORIGINAL ARTICLE**

DOI: 10.2478/ffp-2019-0011

# Wood structure of Scots pine (*Pinus sylvestris* L.) growing on flotation tailings

*Marta Górska* ✉, *Edward Roszyk*

Poznań University of Life Sciences, Faculty of Wood Technology, Department of Wood Science, Wojska Polskiego 38/42, 60-627 Poznań, Poland, phone: +48 61 8487450, e-mail: marta.gorska@up.poznan.pl

## ABSTRACT

The aim of this study was to examine Scots pine (*Pinus sylvestris* L.) xylem changes caused by heavy metal pollution. Annual rings width, number of tracheids in radial rows and the length of tracheids in the wood from trees growing on post-flotation tailings have been measured. Dimensions of tracheids have been examined separately for early- and latewood of each radial increment. The most demonstrable changes are observed in the tracheids length, which appears to be shorter in the xylem from the industrial area than in the control samples. In the wood from the polluted site, the variability of length of tracheids is increased. Microscopic observations revealed numerous deformations in the xylem tissue and deviations from the standard cell arrangement. Circumferential deformations occurring in the wood structure may indicate the increasing spring frost vulnerability of these trees.

## KEY WORDS

flotation tailings, heavy metals, industrial pollution, Scots pine, tracheids

## INTRODUCTION

The environmental burden of industrial waste has been reduced since the beginning of 1990s thanks to the implementation of new technologies (e.g., pollutants-retaining filters) and increasing worldwide environmental awareness (Zwoliński and Orzeł 2000; Danek 2008; Barniak and Krąpiec 2009; Wertz 2012; Duszyński 2014). However, there are still many improvements to bring in this area. The problem of this study is the effect of industrial pollution on the wood microstructure, which has not been covered comprehensively in the published studies to date.

Relatively well known is the effect of air pollution on the physiology and growth of trees. The problem has been described as leading to the degradation of tree stands and decrease in stand volume (Niedzielska 1986; Zwoliński and Orzeł 2000) or even dying out of trees and forests (Paschalis and Staniszewski 1994; Hüttermann et al. 2004). It can result in the fluctuating asymmetry (FA) index increase, which is a stress indicator (Błocka and Staszewski 2007; Chudzińska et al. 2014). Works on structural changes in xylem tissue related to the environment pollution are fewer and mostly concern the width of annual rings, mechanical properties and density of wood (Paschalis and Staniszewski 1994; Niedzielska 1996; Kask et al. 2008; Stravinskiene et al. 2013; Duszyński 2014; Sensuła et al. 2015; Sensuła et al. 2017). The influence of pollution reflected in the annual rings has been studied using the methods of dendrochronology (Danek

Received 26 October 2018 / Accepted 13 May 2019

2008; Duszyński 2014). The least explored in this context seems to be its effects on anatomical structure of wood (Niedzielska 1986; Tulik and Kozakiewicz 2005); moreover, these studies focused mainly on the effects of air pollution, whereas the impact of soil pollution has been sparsely examined (Watmough 1999; Maćkowiak 2016; Mleczek et al. 2016; Mleczek et al. 2017).

Analysing the trace elements' impact on the wood structure, it should be considered that some heavy metals are at the same time essential for proper trees development (e.g., Cu, Fe, Mn, Zn) or can even stimulate growth of particular trees (e.g., Cd – *Betula* spp.) (Hagemeyer 2004). Character and shape of annual rings depend on many other factors that can interact with each other (Zwoliński and Orzeł 2000). On top of that, the reactions of trees may change according to their age (Hawryś 1987).

The study was performed on the wood of *Pinus sylvestris* L., since this species is of high economic significance in Poland. On the other hand, Scots pine is sensitive to the environmental pollution, so it is suitable for the observation (Hawryś 1987; Niedzielska 1996; Wertz 2012; Duszyński 2014). However, the sensitivity of different species to the heavy metals can vary within the same genus (Duszyński 2014) and even within the same species the reaction of individual population can be different (Ernst 2006).

The available data on structural changes in wood of trees growing in polluted environment are incomplete and noncoherent in some cases. The aim of this study was to supplement the present knowledge with the analysis of the industrial pollution impact on the wood structure of Scots pine tree (*Pinus sylvestris* L.), in particular, concerning the impact of the presence of heavy metals. Understanding of modification of wood macrostructure and its properties can be useful from the viewpoint of utilisation of wood obtained from trees used in the dendroremediation process (Komives and Gullner 2006). Microscopic observations of the anatomical changes within particular annual rings provide more specific analysis of the response of trees to industrial pollution.

## MATERIAL AND METHODS

Material for the study was obtained from woods growing on post-flotation tailings' reservoir in the Lower Silesia Province (Poland). The dominant tree species in this site are *Pinus sylvestris* L. and *Betula pendula* Roth. The reference material was wood from the same age trees grown in unpolluted site in a fresh pine forest in the Forest District Durowo (Wielkopolska Province). Analysis of the reference wood and trace elements' content in wood and soil have been performed earlier (Maćkowiak 2016; Mleczek et al. 2018). The contents of the majority of trace elements (i.e., As, Cd, Cr, Cu, Mn, Mo, Ni, Pb, Zn) in the soil from the vicinity of the copper smelter were significantly higher than in the soil from unpolluted site (Mleczek et al. 2018).

Three Scots pine trees of about 18 years of age from both the polluted and control sites were collected for analysis. Selected trees were of similar diameter at breast height and were collected from the same part of the reservoir. No noticeable eccentricity of the pith was observed. Two discs were cut out from each tree, one from the base end and one at breast height. The thickness of each discs (in longitudinal direction) was close to 15 mm. The age of a given tree was determined based on the number of annual rings at the base end discs. Examined samples were all sapwood as trees are felled at an age when heartwood has not yet formed or reached any significant proportion. Annual increments have been numbered from the pith to the bark. The widths of annual rings and the latewood percentage were measured along the greatest diameter of discs using a Brinell magnifying glass equipped with a scale of 0.1 mm. Samples of tree rings have been visually crossdated and the mean tree ring widths have been figured for each site. Then the percentage of latewood within annual increments was calculated from equation (1). Distinction of early- and latewood was made using magnification of 10x.

$$U_{dp}\left(\%\right) = \frac{S_{dp}}{S_{cp}} \cdot 100 \tag{1}$$

where:
$U_{dp}$ – latewood percentage [%],
$S_{dp}$ – latewood width [mm],
$S_{cp}$ – annual ring width [mm].

The section of about 15 mm in width were cut out from the discs along the longest diameter and then split into fragments of 3–6 annual rings. In order to soften the wood tissue, the samples were immersed in hot water for a few hours. After that process, cross-sectional microslides of about 20 μm in thickness were prepared. The number of tracheids in each of the annual rings was counted separately for early- and latewood using a computer image analyser (Motic Image Plus 2.0 ML). From the selected annual rings, the tangential section was also cut to determine the arrangement of tracheids in the xylem tissue.

For measuring of the length of tracheids, early- and latewood from particular rings were separated and then macerated for about 20 hours in a mixture of hydrogen peroxide and glacial acetic acid at 60°C. Subsequently, the macerated samples were neutralized with distilled water and the specimens were used to prepare microslides. The lengths were measured for 30 undamaged tracheids from both early- and latewood using a computer assisted image analyser with an accuracy of 1 μm (Motic Image Plus 2.0 ML). For statistical analysis in STATISTICA 13.3 (StatSoft, USA), Student's t-test was applied.

## RESULTS

Average annual rings from the pith to the bark for both investigated and control area have been plotted in Figure 1. It seems that at breast height in the trees from the polluted site, growth dynamics was greater than in the reference trees in the first years of growth (Fig. 1A). In subsequent years, the annual rings in trees from the industrial area are only slightly wider than those in the control samples. However, for both breast and base height, there was no statistically significant difference between tree rings from polluted and unpolluted sites.

For trees from the polluted site, the graphs of the earlywood tracheids number seem to be similar to the graph of the width of the annual rings, both at breast height and at base end (Fig. 2). The correlation coefficient is however only slightly higher for earlywood tracheids (0.62 for breast height and 0.59 for base end) than for latewood tracheids (0.51 and 0.22, respectively).

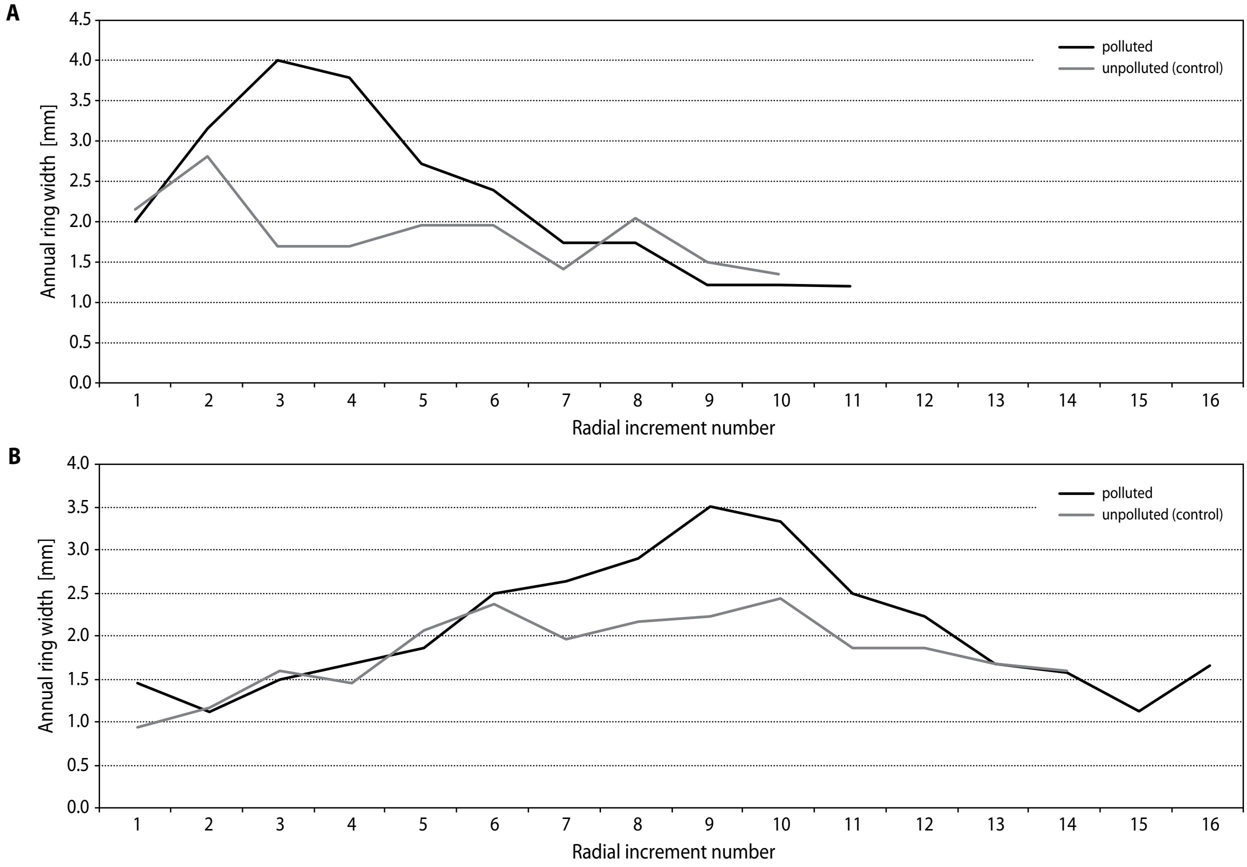

**Figure 1.** Average annual rings' width along the rays (from pith to bark) at breast height (A) and at base end (B)

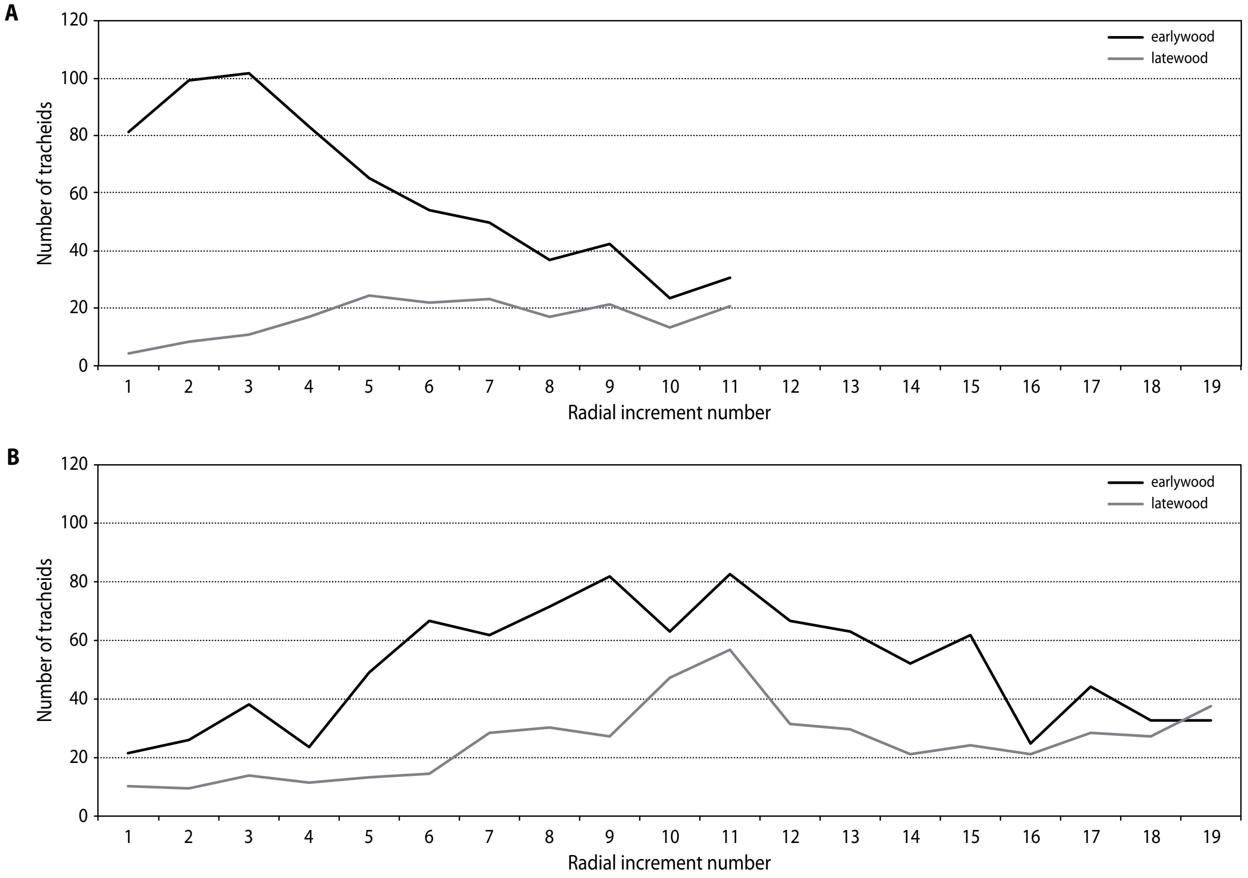

**Figure 2.** Average number of tracheids in the wood from polluted site at breast height (A) and at base end (B)

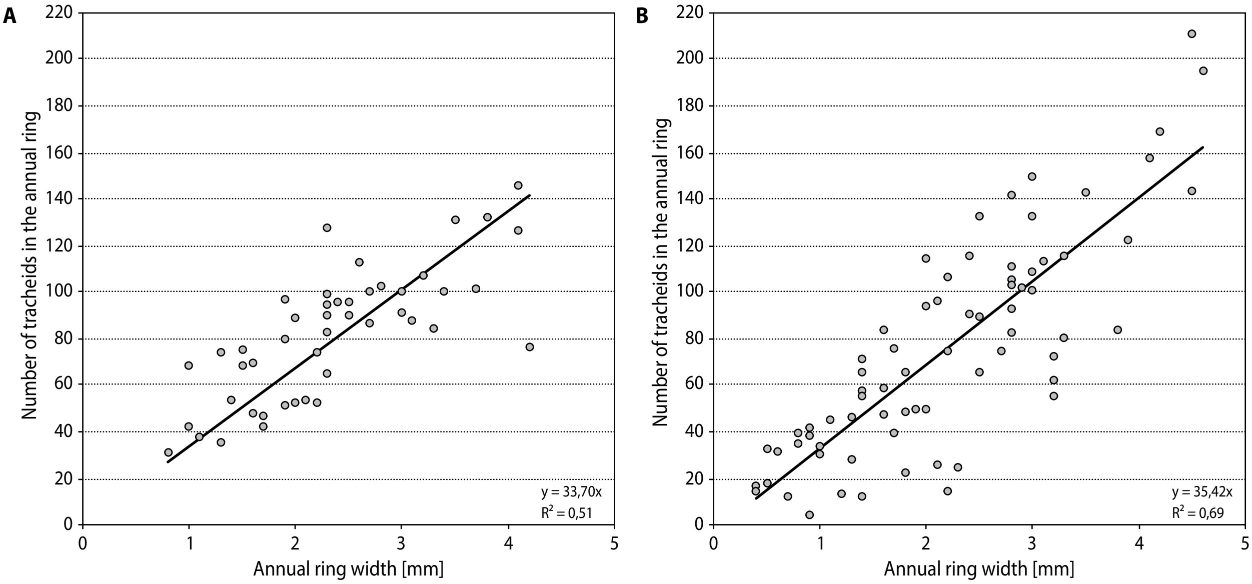

**Figure 3.** Relation between the number of tracheids in radial rows and the annual rings' width for wood from industrial area at breast height (A) and at base end (B)

The relations between the annual rings' width and the number of tracheids in radial rows for wood from the industrial area are shown in Figure 3. These relationships can be well described by the linear regression with the coefficient of determination above 0.5.

The latewood percentage within the annual increments moderately increases with cambial age in the wood from trees growing on flotation tailings. At breast height (Fig. 4A), the comparison is difficult because of unusually high latewood percentage appeared in a few annual rings in the reference wood. Statistical analysis however showed no significant differences between the control samples and the wood from the polluted site.

The tracheids in the wood coming from the industrial area were shorter than those in the control samples (Fig. 5). The variability of results described by the coef-

**Table 1.** Coefficient of variation of tracheid length (blank cells – no measurement was carried out)

| Coefficient of variation CV [%] | | | | |
|---|---|---|---|---|
| radial increment number | earlywood tracheids length | | latewood tracheids length | |
| | unpolluted | polluted | unpolluted | polluted |
| 3 | 10.9 | 18.9 | 12.8 | 15.7 |
| 5 | | 21.9 | | 21.4 |
| 6 | 16,9 | | 8.0 | |
| 7 | | 28.9 | | 23.1 |
| 8 | 13.6 | | 15.5 | |
| 9 | | 25.3 | | 24.1 |
| 11 | | 22.2 | | 19.6 |
| 12 | 17.8 | | 12.8 | |
| Mean | 14.8 | 23.5 | 12.3 | 20.8 |

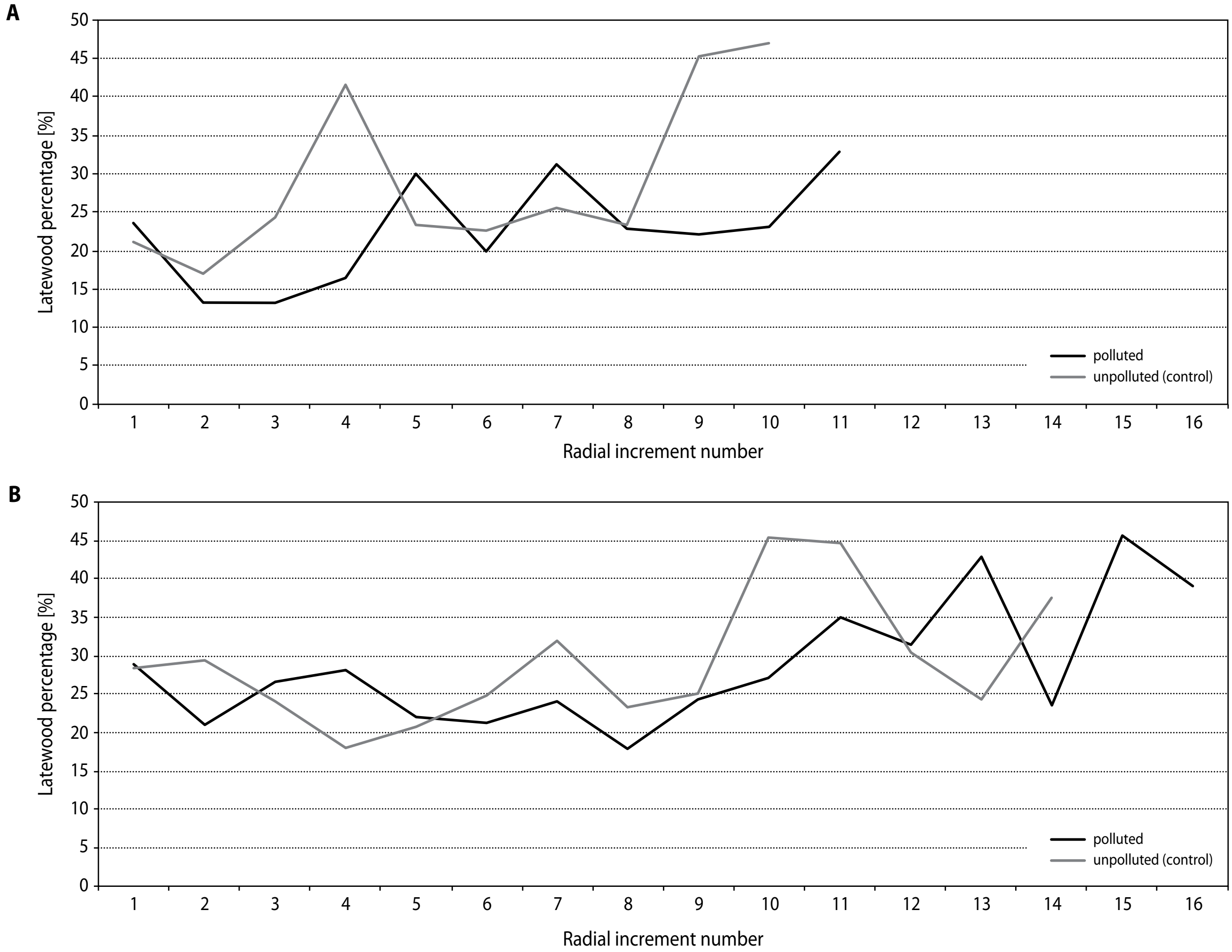

**Figure 4.** Mean latewood percentage within annual increments at breast height (A) and at base end (B)

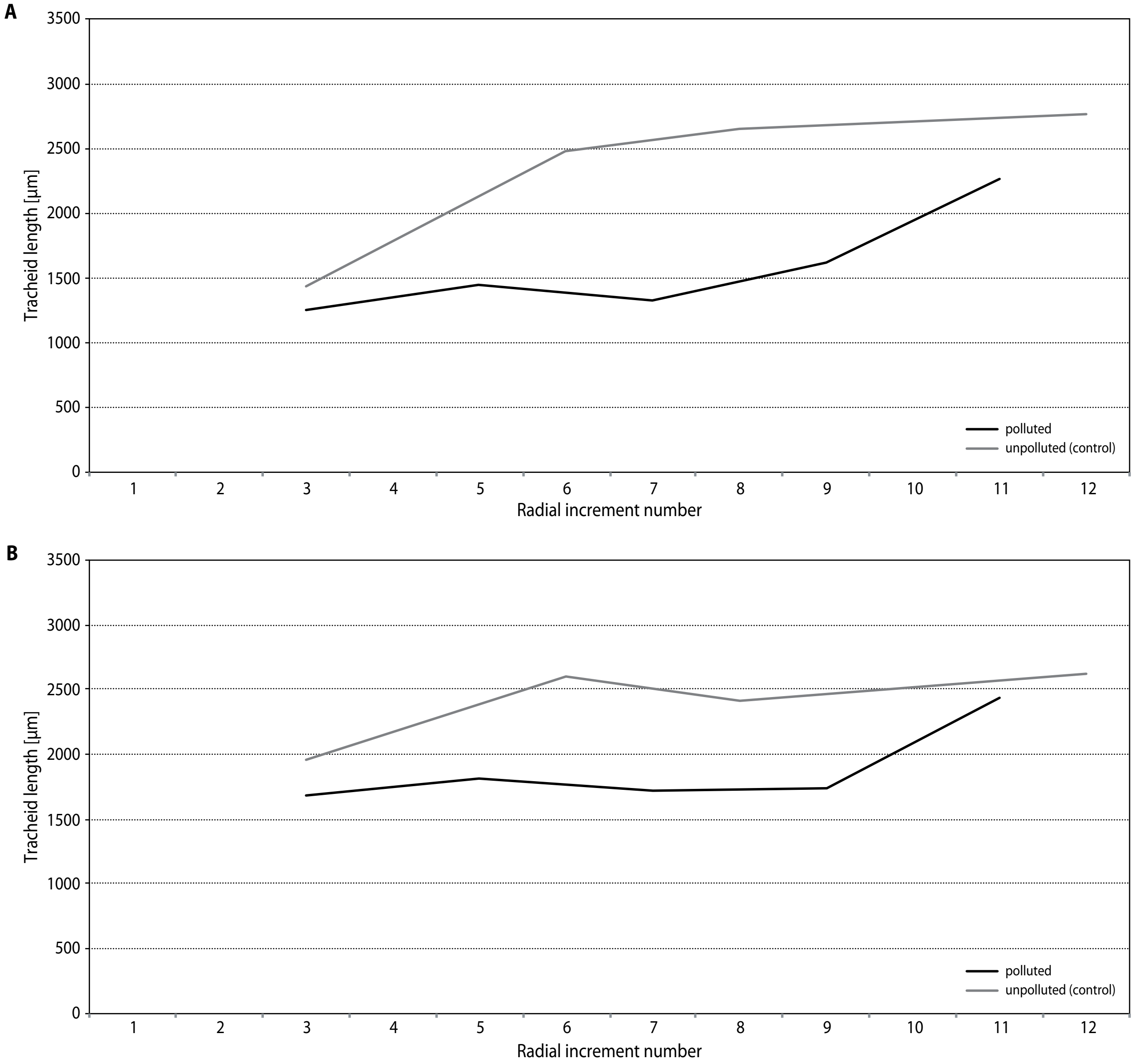

**Figure 5.** Mean, minimum and maximum tracheid length in earlywood (A) and latewood (B) at breast height

ficient of variation (CV) was by 8.5 percentage points higher for tracheids from the trees growing near the copper smelter in both early- and latewood (Tab. 1).

It should be emphasized that the structure of wood coming from the polluted site revealed many anomalies. Besides the typical reaction wood, tracheids strongly modified in shape and size in some sections (Fig. 6). These disorders made it difficult to reckon the number of tracheids in the radial rows. Defeatured tracheids were observed in the circumferential clusters. Lesions were often observed at the beginning of the annual increments (Fig. 7). In some areas, the deformations of xylem were so expanded that it was hard to establish the boundary of a given annual ring.

Disturbances in wood from the industrial area were also noted in the radial section. Figure 8 presents the normal xylem structure and irregular arrangement of tracheids observed in the investigated wood.

In the xylem of trees from the polluted site some tracheids of unnormal shapes, nontypical endings or unusually short were observed. Illustrations of such abnormal tracheids are given in Figure 9.

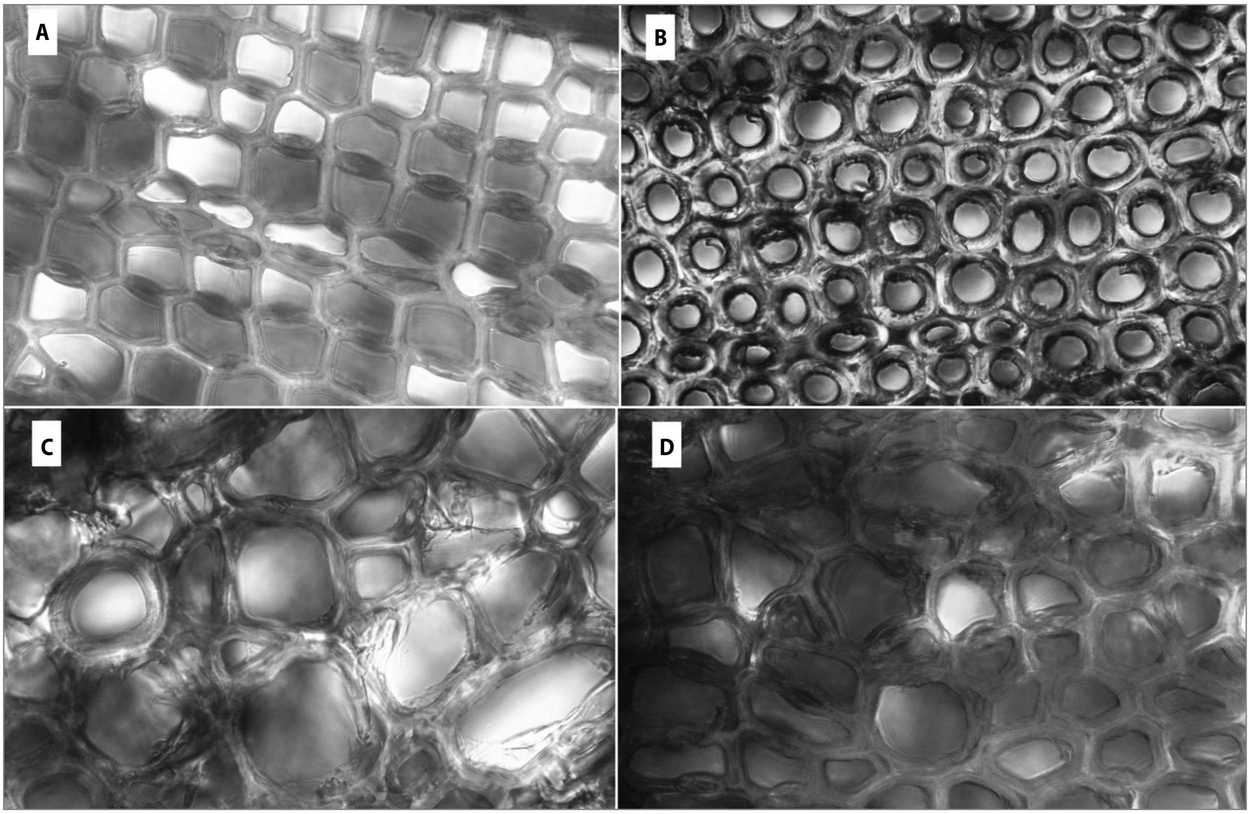

**Figure 6.** Cross-section of a healthy wood tissue (A), reaction (compression) wood (B) and distorted wood tissue (C and D)

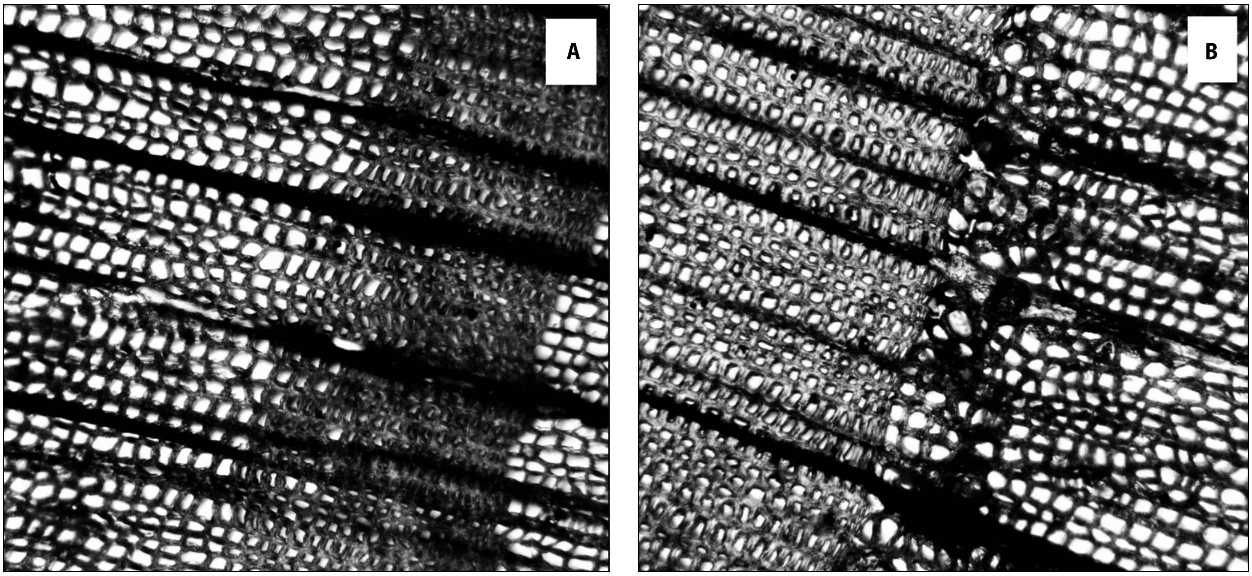

**Figure 7.** Correct (A) and disturbed (B) arrangement of tracheids at the beginning of the annual ring (cross-section)

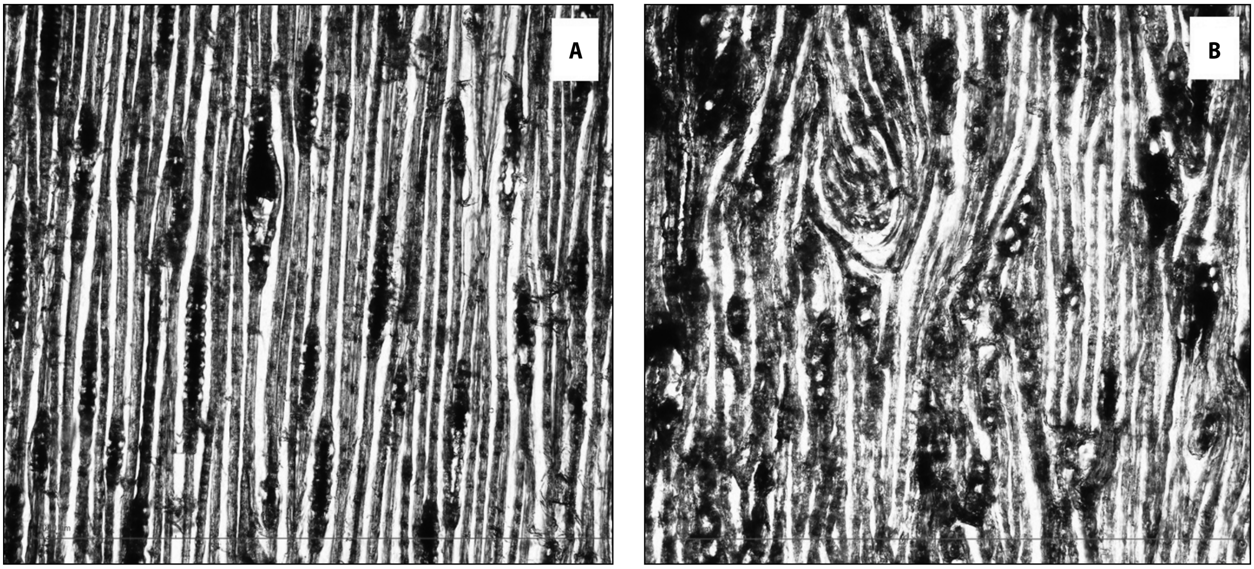

**Figure 8.** Tangential section through the regular (A) and disturbed (B) xylem

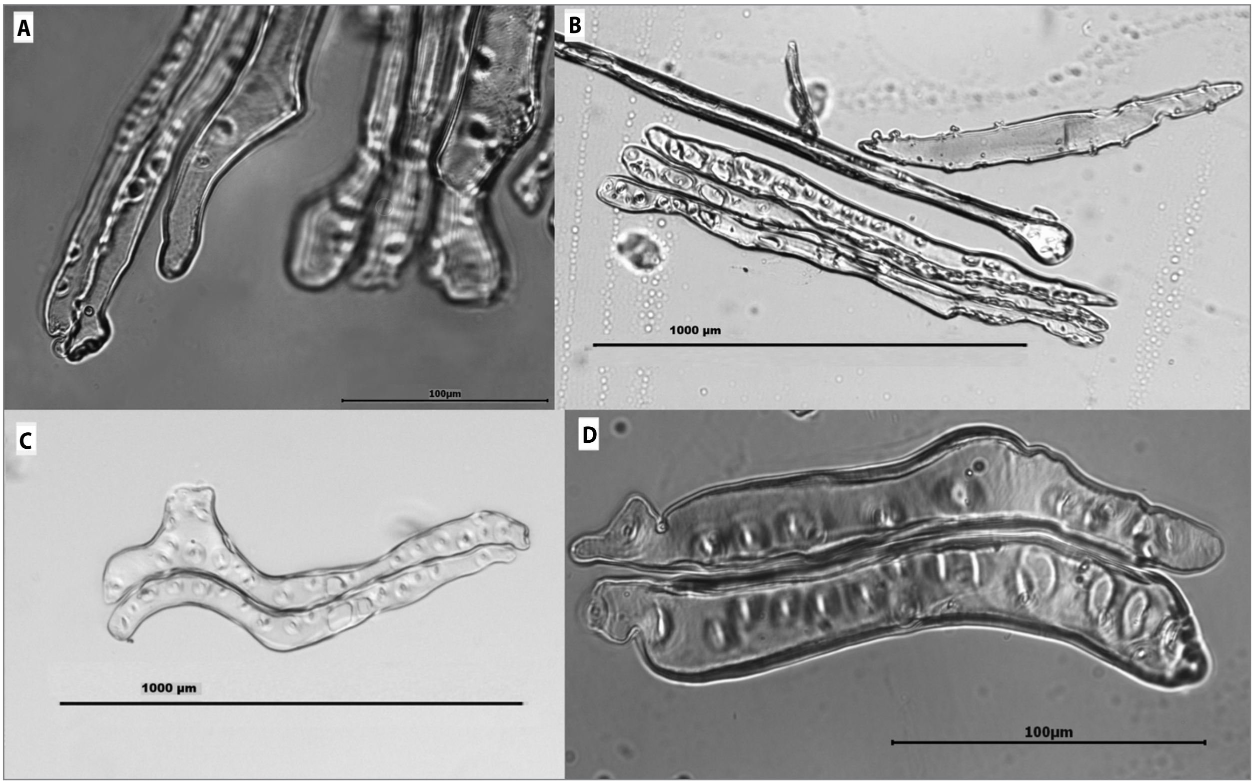

**Figure 9.** Examples of unnatural shapes (B, C, D) and abnormal endings (A) of cells and unusually short tracheids (C, D)

## Discussion

Trying to explain the results of this study at first, the response of Scots pine to the pollutants manifested in annual rings width should be considered since it is one of the most common wood constitution indicators (Kask et al. 2008; Duszyński 2014). According to the published data, all types of industrial pollution lead to the decreasing of annual ring widths (Białobok et al. 1993; Niedzielska 1996; Danek 2008) and increasing latewood percentage as a consequence. Increasing of annual ring width in the first years of growth observed in the trees from the flotation tailings can be explained by the fact that to some extent, some elements present in the soil can have stimulating effect on the growth of certain plants (Hagemeyer 2004). The structure of xylem depends on a number of factors and interactions between them. The complexity of wood forming process makes the interpretation of response of trees to the environmental pollution not clear. It is hard to ascertain if the changes are caused by the presence of heavy metals. The reduction of the further annual rings' width can be a result of increasing density of the soil (substrate) with time and water availability limitation. Decreasing growth can also be a result of the lack of mycorrhiza in the rhizosphere (Białobok et al. 1993).

Tracheids' dimensions are species specific, that is, determined mainly by genetic constitution, but they can be altered by external factors such as water availability, insolation and soil condition or environment pollution (Kozłowski and Pallardy 1997). The last reason could occur in case of this study, since the site on which the examined tree was growing was extremely polluted by heavy metals. Enlarged variation in tracheid length data set from trees growing in industrially polluted area was found. In one of the studies, Niedzielska (1996) have noted similar tendency – tracheid lengths from trees growing farther from the smelter were more uniform than those growing close to the emitter. This may indicate individual response of trees to the pollution. The same researcher (Niedzielska 1986, 1996) released reduced tracheid lengths in wood coming from industrial area, which tallies with our observations. The unusual endings and shapes of tracheids are similar to the anomalies reported by Tulik and Kozakiewicz (2005) in Scots pine from the vicinity of the Chernobyl Exclusion Zone. All these reports have revised the earlier opinions that the environment pollution has no significant effect on the wood anatomic structure (Białobok et al. 1993).

Irregular shapes of cells and their arrangement deformation, which were reported earlier for Scots pine wood from the area close to the mining and metallurgical plant in Małopolska Province (Niedzielska 1986) appear to be similar to these observed in wood growing on flotation tailings. The deformations of the xylem structure occurring at the beginning of the annual increments are comparable to the frost rings described by Schweingruber (2007). Also, other authors have indicated that trees' sensitivity to frost and freezing increase in the polluted environment (Keller 1981; Danek 2008). According to Sheppard (1991), higher concentration of certain toxic ions (e.g., sulphur ones) reduces the saplings resistance to freezing. Young trees are more vulnerable than the older ones of thicker bark.

There is no doubt that the most reliable are comparisons between samples from the same age trees, especially when we look at juvenile wood, which differs from that produced in later growth periods. It would be desirable to perform the analogous measurements in the control material from trees of similar age and growing in the same calendar years.

## Conclusions

Tracheids in both early- and latewood from trees growing on flotation tailings are shorter than in the control samples. Parameters of wood from trees growing in the industrial area were characterized by heightened dispersion. In the xylem, many abnormal tracheids' shapes and structure deviations have been observed. Some circumferential disturbances can indicate the increasing spring frost vulnerability of these trees.

## References

Barniak, J., Krąpiec, M. 2009. The influence of industry on Scots pine stands in the Tarnobrzeg area on the basis of dendrochronological analysis. *Sylwan*, 153 (12), 825–835.

Białobok, S., Boratyński, A., Bugała, W. 1993. The biology of Scots pine. Sorus, Poznań – Kórnik.

Błocka, A., Staszewski, T. 2007. Fluctuating asymmetry of needles as a non-specific stress indicator of Scots pine (*Pinus sylvestris* L.). *Leśne Prace Badawcze*, 4, 125–131.

Chudzińska, E., Pawlaczyk, E. M., Celiński, K., Diatta, J. 2014. Response of Scots pine (*Pinus sylvestris* L.) to stress induced by different types of pollutants – testing the fluctuating asymmetry. *Water and Environment Journal*, 28 (4), 533–539. doi:10.1111/wej.12068

Danek, M. 2008. The influence of industry on the treering width of pines (*Pinus sylvestris* L.) living in the Olkusz region. *Sylwan*, 152 (11), 56–62.

Duszyński, F. 2014. The record of air pollution in tree rings. *Przegląd Geograficzny*, 86 (3), 317–338. doi:10.7163/przg.2014.3.2

Ernst, W.H. 2006. Evolution of metal tolerance in higher plants. *Forest Snow and Landscape Research*, 80 (3), 251–274.

Hagemeyer, J. 2004. Ecophysiology of plant growth under heavy metal stress. In: Heavy metal stress in plants (eds.: M.N.V. Prasad, J. Hagemeyer). Springer, Berlin, 201–222. doi:10.1007/978-3-662-07743-6_8

Hawryś, Z. 1987. Survival rate and growth of trees and shrubs under conditions of heavy air pollution with sulfur and heavy metal compounds. In: Reakcje biologiczne drzew na zanieczyszczenia przemysłowe. Materiały II Krajowego Sympozjum (ed.: R. Siwecki). Adam Mickiewicz University Press, Poznań, 247–255.

Hüttermann, A., Arduini, I., Godbold, D.L. 2004. Metal pollution and forest decline. In: Heavy metal stress in plants (M.N.V. Prasad, J. Hagemeyer). Springer, Berlin, 295–312. doi:10.1007/978-3-662-07743-6_12

Kask, R., Ots, K., Mandre, M., Pikk, J. 2008. Scots pine (*Pinus sylvestris* L.) wood properties in an alkaline air pollution environment. *Trees*, 22 (6), 815. doi:10.1007/s00468-008-0242-7

Keller, T. 1981. Folgen einer winterlichen $SO_2$-belastung für die Fichte. *Gartenbauwissenschaft*, 46, 170–178.

Komives, T., Gullner, G. 2006. Dendroremediation: the use of trees in cleaning up polluted soils. In: Phytoremediation Rhizoremediation (eds.: M. Mackova, D. Dowling, T. Macek). Springer, Dordrecht, 23–31. doi:10.1007/978-1-4020-4999-4_3

Kozłowski, T.T., Pallardy, S.G. 1997. Growth control in woody plants. Elsevier, Amsterdam.

Maćkowiak, M. 2016. The influence of contaminated soil on the increment dynamics and wood properties of Scots pine (*Pinus sylvestris* L.). Master's thesis, Poznań.

Mleczek, M., et al. 2016. The role of selected tree species in industrial sewage sludge/flotation tailing management. *International Journal of Phytoremediation*, 18 (11), 1086–1095. doi:10.1080/15226514.2016.1183579

Mleczek, M., et al. 2017. Phytoextraction of potentially toxic elements by six tree species growing on hazardous mining sludge. *Environmental Science and Pollution Research*, 24 (28), 22183–22195. doi:10.1007/s11356-017-9842-3

Mleczek, M., et al. 2018. The importance of substrate compaction and chemical composition in the phytoextraction of elements by *Pinus sylvestris* L. *Journal of Environmental Science and Health*, *Part A*, 53 (11), 1029–1038. doi:10.1080/10934529.2018.1471116

Moliński, W. 2010. Variability of the microfibril angle in the tangential walls of the cells and the tensile strength in the direction of the grain within the individual annual rings of *Pinus sylvestris* L. wood. The final report of the research project Nr N N309 1693 33. Poznań.

Niedzielska, B. 1986. The impact of air pollution on anatomical structure of wood of Scots pine (*Pinus silvestris* L.) growing within imissions mills "Bolesław" near Olkusz. *Acta Agraria et Silvestria, Series Silvestris*, 25, 131–141.

Niedzielska, B. 1996. Comparative research on the impact of industrial pollution based on the properties of Scots pine (*Pinus sylvestris* L.) wood. *Acta Agraria et Silvestria*, *Series Silvestris*, 34, 105–120.

Paschalis, P., Staniszewski, P. 1994. Changes in some indicators of properties of pine wood originated from industrially polluted regions. *Sylwan*, 138 (8), 35–41.

Schweingruber, F. 2007. Wood structure and environment. Springer, Berlin, 87–92. doi:10.1007/978-3-540-48548-3

Sensuła, B., Opała, M., Wilczyński, S., Pawełczyk, S. 2015. Long- and short-term incremental response of *Pinus sylvestris* L. from industrial area nearby steelworks in Silesian Upland, Poland. *Dendrochronologia*, 36, 1–12.

Sensuła, B., et al. 2017. Variations of tree ring width and chemical composition of wood of pine growing in the area nearby chemical factories. *Geochronometria*, 44 (1), 226–239.

Sheppard, L.J. 1991. Causal mechanisms by which sulphate, nitrate and acidity influence forest hardiness in red spruce: review and hypothesis, *New Phytologist*, 127 (1), 69–82. doi:10.1111/j.1469-8137.1994.tb04260.x

Stravinskiene, V., Bartkevicius, E., Plausinyte, E. 2013. Dendrochronological research of Scots pine (*Pinus sylvestris* L.) radial growth in vicinity of industrial pollution. *Dendrochronologia*, 31 (3), 179–186. doi:10.1016/j.dendro.2013.04.001

Tulik, M., Kozakiewicz, P. 2005. Some physical and mechanical properties of pine wood (*Pinus sylvestris* L.) from excluded zones around the Chernobyl power station. *Folia Forestalia Polonica, Series B – Wood Science*, 36, 3–14.

Watmough, S.A. 1999. Monitoring historical changes in soil and atmospheric trace metal levels by dendrochemical analysis. *Environmental Pollution*, 106 (3), 391–403. doi:10.1016/s0269-7491(99)00102-5

Wertz, B. 2012. Dendrochronological evaluation of the impact of industrial imissions on main coniferous species in the Kielce Upland. *Sylwan*, 156 (5), 379–390.

Zwoliński, J., Orzeł, S. 2000. Productivity of Scots pine stands (*Pinus sylvestris* L.) along an industrial pollution gradient. *Prace IBL, Ser. A*, 1 (892/894), 75–98.