EVALUATION OF THE OZONE INJURY TO GROUND VEGETATION WITHIN THE PLOTS OF INTENSIVE MONITORING IN THE CZECH REPUBLIC

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The impact of ozone on vegetation is assessed by mapping and evaluating the visible symptoms of injury on assimilation organs in the herb, shrub and tree layer. In the Czech Republic, this survey is done regularly, once a year, in autumn, on eight selected plots of Intensive Monitoring of the Forest Ecosystem within the ICP Forests Program.

The contribution deals with the practical experience of the assessment of ozone effects on vegetation and the description of the symptoms of ozone injury that are most frequent in the Czech Republic. It shows typical symptomatic and problem species, tries to find a way how to compare the results of ozone impact on vegetation in different locations, and evaluates the occurrence of the symptoms in connection with ozone concentrations within the monitored locations.

Key words: *ozone, symptoms of visible ozone injury, monitoring, ground vegetation*

Vliv přízemního ozonu na vegetaci se mapuje vyhledáváním a hodnocením viditelných symptomů vyvolaných účinky zvýšených koncentrací ozonu na asimilačním aparátu bylinného, keřového i stromového patra. V ČR probíhá toto hodnocení pravidelně jednou ročně v podzimním aspektu na osmi vybraných plochách zařazených do sítě ploch mezinárodního monitoringu lesních ekosystémů. Při hodnocení je postupováno podle platného sub-manuálu programu ICP Forests, část X.B.

Příspěvek se zabývá praktickými zkušenostmi s hodnocením vlivu ozonu na vegetaci, popisem symptomů, které se na plochách v ČR nejčastěji v souvislosti s účinky přízemního ozonu vyskytují, poukazuje na typické symptomatické i problematické druhy, zabývá se hledáním postupů, jak vzájemně srovnat výsledky hodnocení vlivu ozonu na vegetaci z odlišných lokalit a hodnotí výskyt symptomů v souvislosti s koncentracemi přízemního ozonu na sledovaných lokalitách.

Klíčová slova: Přízemní ozon, symptomy poškození, monitoring lesa

1. Introduction

In recent years, ozone, as important harmful atmospheric substances that can negatively affect the ecosystems, is studied intensively. Conclusions of the performed studies, however, often differ significantly. While e.g. KARLSSON *et al.* (2005) work on estimates of concrete figures expressing the decrease of the forest growth and lower economic viability of the forest production due to the negative effect of ground ozone, DITTMAR *et al.* (2004) or STRIBLEY and ASHMORE (2002) state that the negative effect of ozone is not a global problem and that it is observed only in the stands of more favourable water regime of soil.

The relation of ozone uptake with the opening of the stomata in real conditions can show an increased risk for the mountain forest ecosystems. Ozone concentrations in the mountain locations are not of such high (peak) values as in industrial regions, but their development is more stable and often increased for a long time. The trees in such locations are not stressed by insufficient water uptake and the stomata conductivity is higher when compared to lower altitudes (WIESER, HAVRANEK 1995, 1996). The relation of ozone concentration and altitude has been proved already and verified many times; that is why the impact of ozone in higher altitudes, including visible symptoms of ozone injury, is more expected. On the other hand, the natural detoxification capacity of trees growing in such conditions is higher, and the period of ozone uptake is shorter (WIESER *et al.* 2000).

The negative impact of the ground ozone on vegetation is visible in the colour changes and necroses on the assimilation apparatus. In the beginning spot chloroses can be observed, together with colour changes; later they spread to bigger areas or to whole leaves (SKELLY *et al.* 1998, UHLÍŘOVÁ, KAPITOLA 2004). Chronic ozone injury can result also in smaller leaves, needles or sprouts (KELLER 1981).

Evaluating the injury of adult trees (solitary, in urban area and/or in forest stand) is complicated, as the observer usually cannot see the upper part of the crown where the changes are most frequent. That is why the symptoms of visible injury are monitored and assessed in the ground vegetation, which in many cases reacts more sensitively than adult trees (Vollenweider, Ottinger, Günthardt-Goerg 2003, Manning, Godzik 2004). However, regional evaluation of the ozone impact is complicated due to variability of the floristic composition in different conditions, and also due to the fact that the evaluation of the symptoms in most herb species was not tested experimentally (Bussoti, Cozzi, Ferretti 2006). The quality of the assessment thus depends significantly on the experience of the observer (Bussoti *et al.* 2006).

The presented contribution deals with practical experience in the evaluation of visible symptoms of ozone injury to vegetation and the description of the symptoms of ground ozone injury that are most frequent within plots in the Czech Republic. It shows typical symptomatic and problem species, tries to find how to compare results of the evaluation of ozone effect in different locations, and evaluates the occurrence of the symptoms in connection with ground ozone concentration in the studied monitoring plots.

2. Material and Methods 2.1. Monitoring plots

Eight plots of intensive monitoring (ICP Forests/Forest Focus – level II) were selected in different regions of the Czech Republic, at altitudes ranging from 350 to 1300 m (Fig. 1, Tab. 1). ICP Forests was launched in 1985 under the Convention on Long-range Transboundary Air Pollution (CLRTAP) of the United Nations Economic Commission for Europe (UNECE), due to the growing public awareness of possible adverse effects of air pollution on forests. Intensive monitoring (level II) is the key for providing insight into causes affecting the condition of forest ecosystems and into effects of different stress factors. More is under www.icp-forests.org.

Altitude of the plots selected is shown in the Table 1. Plots Míšečky, Buchlovice and Všeteč are situated in the beech stands, the rest of the plots are in the spruce stands. Detailed information about the plots is presented under www.vulhm.cz/docs/monitoring.

2.2. Visual leaf injury assessment

The method of symptoms assessment is described in detail in the Sub-manual for the assessment of ozone injury on European forest ecosystems of the ICP Forests programme, revised in 2004 (UN/ECE, 2004). The level of injury is assessed in four steps: 0 -not injured; 1 -visible symptoms on 1 - 5% of leaves; 2 -symptoms on 6 - 50% of leaves; 3 -symptoms on more than 50\% of leaves.

The assessment was performed on the LESS plots (light exposed sampling site). The sites are exposed to full sunshine on open plot area, i.e. forest edges, clear-cuts, and meadows etc., preferably south or southwest exposition. Visible ozone injury is assessed on fully developed leaves that are exposed to full sunlight. LESS plots were established according to the manual, within the vicinity of the ozone measurement device, with the exception of the plot in Jizerské hory, where the distance of 500 m could not be complied.

The evaluation was performed in 2005–2008, in the late summer aspect, mostly in the first half of September, before the autumn yellowing.



Fig. 1. The number of symptomatic species on the plots assessed in 2005–2008 in correlation to the altitude.

Literature and photographic documentation is also used for ozone-like visible symptoms identification (MANNING *et al.* 2002, MANNING 2004, INNES *et al.* 2001).

2.3. Measuring of ground-level ozone

Ozone concentration was measured using passive diffusion tubes (Gradko International Ltd.), with a recommended exposition period of up to 4 weeks. The measurements are based on the principle of nitrite oxidation on the nitrate within the active part of the filter. The producer using ion chromatography does the analysis of the exposed filters. A one-micron porosity Teflon filter is fitted to the sampler to prevent the ingress of airborne particulate nitrate. Thanks to this construction, the resulting concentration is also only minimally affected by the wind. According to the producer, the measuring precision is \pm 6.8%, and the detection limit 1.42 ppb (approx. 3 µg.m⁻³) within a two-week exposition. The measuring range is 6.8 – 200 µg.m⁻³ and the uptake rate is 0.93 cm.h⁻¹. Other technical information is presented on the producer's website (http://www.gradko.co.uk).

3. Results

The results of the evaluation of visible symptoms caused by the negative effect of ground-level ozone in 2005–2008 are presented in Table 1 as well as the respective altitude and average ozone concentration in vegetation period (April – September) calculated on the basis of measurements made by Gradko passive samplers.

| - | | | | | | | | | |
|--|----------------------------|---------|---------------|---------|------------------|----------------|------|--------|---------|
| | Term of asses- sment | Mísečky | Švýcár- na | Želivka | Luisino údolí | Medlo- vice | Lazy | Všeteč | Jizerka |
| Altitude | | 940 | 1300 | 440 | 930 | 350 | 875 | 600 | 950 |
| Average ozone concentration IV. – IX. | IX. 2005 | 83 | 115 | 66 | 94 | 81 | 103 | 73 | 89 |
| | IX. 2006 | 111 | 137 | 86 | 129 | 98 | 121 | 92 | 112 |
| | IX. 2007 | 93 | 151 | 75 | 125 | 93 | 116 | 93 | 109 |
| | IX. 2008 | 109 | | 74 | 136 | 99 | 119 | 96 | 110 |
| Number of assessed species | IX. 2005 | 24 | 19 | 3 | 15 | 8 | 9 | 0 | 14 |
| | IX. 2006 | 47 | 51 | 16 | 24 | 10 | 25 | 3 | 26 |
| | IX. 2007 | 50 | 50 | 20 | 28 | 14 | 26 | 9 | 27 |
| | IX. 2008 | 50 | 51 | 16 | 33 | 15 | 28 | 11 | 28 |
| Number of symptomatic species | IX. 2005 | 24 | 19 | 3 | 14 | 8 | 7 | 0 | 14 |
| | IX. 2006 | 23 | 20 | 15 | 19 | 4 | 11 | 2 | 20 |
| | IX. 2007 | 31 | 22 | 11 | 14 | 8 | 1 | 0 | 6 |
| | IX. 2008 | 25 | 24 | 2 | 16 | 0 | 12 | 0 | 5 |
| Percent of symptomatic species | IX. 2005 | 100 | 100 | 100 | 93 | 100 | 78 | 0 | 100 |
| | IX. 2006 | 49 | 39 | 94 | 79 | 40 | 44 | 67 | 77 |
| | IX. 2007 | 62 | 44 | 55 | 50 | 57 | 4 | 0 | 22 |
| | IX. 2008 | 50 | 47 | 13 | 48 | 0 | 43 | 0 | 18 |

Table 1. The results of ground vegetation assessment on the plots studied in 2005–2008, using the LESS/mini LESS method given in the ICP Forest manual (UN-ECE, 2004) and ground level ozone concentrations measured by passive samplers Gradko

The recorded visible symptoms usually correspond to the experience of the international cross-calibrating courses. On the tree leaves (*Acer, Fraxinus, Carpinus, Alnus, Populus, Tilia* and others) and shrub leaves (*Salix, Sambucus, Syphoricarpos* and others) dark dots (points, stippling) are observed, which in higher injury connect into bigger spots or areas. On European beech (*Fagus sylvatica*), the ozone injury is observed in the form of brownish, bronze colour of the leaves. For beech this is a typical symptom.

On most of the herb layer species, the symptoms were visible in the form of red, red-brown, brown to black dots or necroses.

Typical for the visible ozone injury is, that symptoms occurred on the sun-exposed, middle-aged and older, fully developed leaves more than on the young leaves. Shaded parts of the leaves (i.e. overlapped leaves) usually do not show any injury. Visible injury normally does not go throughout the leaf-tissue, visible symptoms being usually confined to the upper leaf surface, typically expressed as tiny purple-red, yellow or black spots or sometimes as a discolouration, reddening or bronzing. Both stippling and discolouration occur only between the veins (UN-ECE, 2004).

On coniferous trees, the symptoms were observed in the form of light but not sharply marked dots or blurred spots on the needles. Usually on sun exposed part of the branches. The increased occurrence of such symptoms on the older needle year classes is typical, because of accumulation of negative effects.

The results of the evaluation (Tab. 1) show the difference in the number of the species recorded and evaluated on individual plots. According to the site conditions, the number ranges from few individuals to several dozen. In general it can be stated



Fig. 2. The number of symptomatic species on the plots assessed in 2005–2008 in correlation to the altitude.



Fig. 3. The percentage of symptomatic species within the plots assessed in 2005–2008 in relation to the altitude.

that the number of species is higher with higher altitude. On the same gradient, also the number of species showing injury symptoms that could have been caused by ground ozone is increasing (Fig. 2). The relative number of symptomatic species does not confirm any trend of relation with the altitude (Fig. 3).

Among the main problem species in the survey are the representatives of *Vaccinium*, *Rumex*, *Cornus*, *Frangula*, due to natural reddening of the leaves; difficult to assess are also *Trifolium*, *Taraxacum* or *Hieracium*.

4. Discussion

On the plots situated at higher altitudes (Švýcárna, Mísečky, Jizerka, Luisino údolí, Lazy) during the evaluation period of 2005–2008, the symptoms of ground ozone injury were found on more species of the shrub and herb layer as compared to the plots under 800 (600) meters above sea level (Fig. 2). The number of monitored species growing on the plots and in their surroundings differs significantly, however, and the conclusions based only on the various numbers of assessed and symptomatic species can hardly be fully reliable. A certain guideline can be provided by the percentage of species showing visible ozone injury. The relation between the number of symptomatic species is used for comparison (Fig. 3). Way of evaluation or recording respective, within the localities assessed, can be a problem in this case. Mainly in 2005, most probably only the symptomatic species were recorded, other species in the localities assessed, showing no symptoms of ozone injury, were not recorded.

The percentage of symptomatic species and the percentage of the sub-plots with the occurrence of symptomatic species are among the obligatory parameters according to the ICP Forests (UN-ECE, 2004) method. So the assessment results show that the intensity of visible ozone injury is not connected only with the altitude, as even on the Buchlovice (350 m a.s.l.) and Želivka (440 m a.s.l.) plots, injury of 2nd class was recorded in 2005 and 2006. Only on the Všeteč (600 m a.s.l.) plot no 2nd class of injury was recorded on any of the species growing on the stand edge.

This relation was graphically expressed in the simplified version as an average value of the injury level on the plot in each assessment period. These averages are shown in graph Figure 4. Plots at higher altitudes are in light colours, while plots at lower altitudes are marked with dark symbols.

The relation of the level of injury to vegetation and the altitude, expressed in this way, shows the highest index in 2005, and the highest average value is within the two plots of the lowest altitude. The Všeteč (600 m above sea level) plot was not assessed in 2005; in 2006 it was roughly in the middle among the plots assessed, and in 2007, together with the plot Lazy (875 m a.s.l.) it was among the localities with the lowest injury level. At the same year, the ground-level ozone concentrations measured every year, e.g. on the Želivka plot, are the lowest or among the lowest within the plots studied.



Fig. 4. The average level of injury on individual plots (altitude) in individual years.



Fig. 5. The number of species in individual locations according to the level of injury in the 2005–2008 assessment periods.

This can be explained by the above-mentioned fact that in 2005 only the species showing visible symptoms of ozone injury were recorded – with a few exceptions, class 0 is missing in the results table. This means that species not showing visible injury were most probably not recorded. This practice was developed in subsequent years and applied mainly in 2007 and 2008.

When a higher level of injury is recorded in a lower number of individual species that were assessed, a higher average value of the injury level can be calculated. This can distort the results and the mutual comparison of the plots does not precisely reflect reality.

Histograms are another way of comparing individual locations, where individual classes in their frequency correspond to different levels of injury (0 - 1 - 2). We attempt to show this relation in Figure 5. Yellow and orange colours correspond to the plots less than 800 m above sea level, and grey and black colours represent the mountain locations. The figure shows that a higher intensity of injury was found mainly on the mountain plots and only exceptionally on lower altitude plots. This figure better reflects reality, i.e., the fact that on the plots situated at higher and mountain altitudes, the symptoms of vegetation injury caused by the negative effect of ground-level ozone were more frequent and also of higher intensity than at lower altitudes.

Evaluation of the ozone damage raises many questions – this is the result of the experience, every year widely discussed within the international calibration courses, focused on the problem of ozone injury on vegetation. Besides the above mentioned (rich species composition in different countries and forest vegetation zones, experience of the observers, combination of the symptoms caused by different agents etc.) it is

also capability to determine precisely individual species evaluated – mainly in the herb layer. Specialized botanist had recorded more species than other field experts, which is also reflected by the percentage of the proportion of symptomatic species recorded.

In 2009, all these practical experience has resulted in the corrected method of assessment. Obligatory assessment of all the wood species is the main change, herb layer assessment is voluntary. The new scale (0 - no symptoms, 1 - ozone symptoms only, 2 - symptoms due to other causes, no ozone, 3 - 1 and 2, 4 - unknown, not validated) is the second important change. Also the way of validation will be recorded, where, besides consulting of the validation centre, also atlas and other photo-guides, including the internet database, can be used (www.ozoneeffects.org). This should enable better comparability of the results among individual countries.

5. Conclusions

In the field investigation connected with the assessment of visible symptoms of injury caused by ground ozone, it is easier, as usual, to find the symptoms of higher intensity injury in the mountain areas. In the Czech Republic, the zone of more frequent damage starts from about 800 m a.s.l. At higher altitudes, the symptoms can be observed on a wider spectrum of species of the herb, shrub and tree layer. This is connected with the fact that at these sites also higher concentrations of ground ozone are recorded during the vegetation season and the water stress resulting in stomata closure is less frequent than in lowlands. Vegetation at higher altitudes is also exposed to other types of oxidation stress, which can have similar injury symptoms.

The symptoms recorded correspond to the lower load of the ecosystem by ozone when compared to some regions in Western and Southern Europe (Southern Switzerland, Northern Italy and Spain). The symptoms are often hard to be identified, as they are of lower intensity and often in combination with other biotic and abiotic factors. That is why most of the symptoms have to be validated with the help of scientific literature, photo documentation available on the Internet or at the validation centre.

It is hard to find a simple and unique method to compare different locations. It is necessary to create lists of symptomatic species, and to use various possibilities of "indexing" for their comparison. And last but not least, it is necessary to comply the valid method at all of the locations and to make a record if it is changed. The method of the assessment and recording of symptomatic and non-symptomatic species can affect and limit the possibility of the mutual comparison of the locations assessed.

Practical experience of the field assessment results in the correction of the methods used to evaluate the symptoms in the field conditions. Recent changes were done in 2009, in connection with the solution of the problem within the FutMon project.

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Resumé

Na plochách ve vyšších nadmořských výškách byly v hodnoceném období (2005 – 2008) nalezeny příznaky poškození přízemním ozonem na větším počtu druhů bylinného i keřového patra než na plochách pod 800 (600) m n. m. Výsledky hodnocení přitom naznačují, že intenzita viditelného poškození zřejmě není pouze funkcí nadmořské výšky, protože i na plochách v nadmořské výšce 350, resp. 440 m n. m. byly v roce 2005 a 2006 zaznamenány druhy se stupněm poškození 2 (na stupnici 0–3).

Obecně přesto platí, že při terénních šetřeních spojených s hodnocením viditelných symptomů poškození vyvolaných přízemním ozonem jsme se s různými projevy poškození a s vyšší intenzitou symptomů setkávali na horských lokalitách. V ČR je za hranici pro vyšší (horské) polohy považována nadmořská výška cca 800 m. V polohách nad touto hranicí jsou symptomy pozorovatelné na širším spektru druhů bylinného, keřového i stromového patra. To souvisí s faktem, že ve vyšších nadmořských výškách jsou během vegetační sezóny zaznamenávány vyšší koncentrace přízemního ozonu. Vegetace ve vyšších polohách je také více vystavena i dalším typům oxidativního stresu, který se může projevit podobnými symptomy poškození.

Při hodnocení symptomů v terénu je nutné dbát na dodržování stanovené metodiky hodnocení na všech lokalitách a při její změně o tom mít záznam. Způsob hodnocení a zaznamenávání symptomatických a nesymptomatických druhů může zásadně ovlivňovat a omezovat možnost vzájemného srovnání hodnocených lokalit.