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Centre for  
Ecology & Hydrology

NATURAL ENVIRONMENT RESEARCH COUNCIL



# 25<sup>th</sup> Task Force Meeting & one-day ozone workshop

31 January – 2 February, 2012  
Brescia, Italy

*Università Cattolica, via Trieste 17, Brescia*

# Programme & Abstracts



## Organizers:



**Centre for  
Ecology & Hydrology**

NATURAL ENVIRONMENT RESEARCH COUNCIL

ICP Vegetation Programme Coordination Centre  
Centre for Ecology and Hydrology  
Bangor, UK.

*Harry Harmens  
Gina Mills  
Felicity Hayes*

## Local organizers



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Ecophysiology and Environmental Physics Lab -  
Math and Physics Dept., Università Cattolica del  
Sacro Cuore, Brescia, Italy

*Giacomo Gerosa  
Riccardo Marzuoli  
Angelo Finco  
Erica Cabrioli*



**PROVINCIA  
DI BRESCIA**

## With financial support of:

***Brescia District Administration –  
Department of Environmental  
Protection, Ecology and Mining, Energy***

# Programme one-day ozone workshop

*Location: SALA della GLORIA, Università Cattolica, via Trieste 17, Brescia*

**Tuesday 31<sup>st</sup> January, 2012**

**08:15**            **Registration and putting up posters**

**08:45**            Gina Mills – Introduction to the Workshop

## **Theme 1: Quantifying ozone impacts on Mediterranean Forests**

**Chair: Elena Paoletti**

**Rapporteur: Ignacio González Fernández**

*Questions to be answered:*

- *Which indicators perform best for Mediterranean Forests?*
- *How well does DO<sub>3</sub>SE perform?*
- *Can we predict flux from passive sampler measurements?*
- *Can we set new critical levels specific to Mediterranean forests yet?*
- *Is there field-based evidence of predicted impacts?*

**09:00 – 10:30: session 1** (15 min per presentation, 10 min discussion per paper)

09:00            *Mihaela Mircea et al.* – Ozone simulations over Italy with the atmospheric modelling system of the Minni project: evaluation and perspectives for vegetation.

09:25            *Angelo Finco et al.* – When stomatal flux is predictable from AOT40. Results of a 13 years stomatal flux calculation exercise at an Alpine spruce forest with the DO<sub>3</sub>SE model.

09:50            *Alessandra De Marco et al.* – FO<sub>3</sub>REST project - First application of DO<sub>3</sub>SE model on French and Italian Forests: Comparison of risk indicators for Mediterranean trees.

10:15            General discussion.

**10:30 – 11:00 Coffee/tea**

**11:00 – 12:20: session 2** (15 min per presentation, 10 min discussion per paper)

11:00            *Silvano Fares et al.* – Measured and modelled stomatal and non-stomatal ozone fluxes in a mixed Mediterranean forest.

11:25            *Rocio Alonso et al.* – Ozone critical levels for Mediterranean forests.

11:50            General discussion & answers to questions.

**12:20 – 14:00 Lunch (to be paid in cash at restaurant)**

## Theme 2: Mapping vegetation at risk from ozone at the national scale

Chair: Jürg Fuhrer

Rapporteur: Håkan Pleijel

Questions to be answered:

- *How is DO<sub>3</sub>SE being used at the national scale?*
- *Can a hybrid between the AOT40 and flux approaches be applied at national level?*
- *How well do the EMEP model predictions match those from national modelling?*
- *What do epidemiological approaches tell us?*
- *Is there field-based evidence of predicted impacts?*

**14:00 – 15:30: session 3** (15 min per presentation, 10 min discussion per paper)

- 14:00 *Per Erik Karlsson et al.* – Introduction of Environmental Objectives for ozone impacts on vegetation in Sweden based on ozone flux.
- 14:25 *Patrick Büker et al.* – Applying flux based ozone risk assessment methods in the UK.
- 14:50 *Beat Rihm et al.* – Mapping ozone fluxes for Switzerland: comparison with EMEP-maps.
- 15:15 General discussion.

**15:30 – 16:00 Coffee/tea**

**16:00 – 17:00: session 4** (15 min per presentation, 10 min discussion per paper)

- 16:00 *Sabine Braun et al.* – Flux-response relationship of *Fagus sylvatica*: what does the epidemiological data analysis tell us?
- 16:25 General discussion & answers to questions

**17:00 End**

**17:15 Welcome coffee, registration Task Force Meeting, including payment for lunches etc. and putting up posters**  
(Please note that a meal is not included and that you have to make your own arrangements for dinner if required).

# Programme 25<sup>th</sup> Task Force Meeting of the ICP Vegetation

*Location: SALA della GLORIA, Università Cattolica, via Trieste 17, Brescia*

## Tuesday 31<sup>st</sup> January, 2012

**17:15** Welcome coffee, registration Task Force Meeting, including payment for lunches etc. and putting up posters  
(Please note that a meal is not included and that you have to make your own arrangements for dinner if required).

## Wednesday 1<sup>st</sup> February, 2012

**08:15** Registration for late arrivals

**Session 1:** **08:45 – 10:30 Plenary session** Chair: **Giacomo Gerosa**

08:45 Welcome address by Prof. Francesco Lechi (Chamber of Commerce of Brescia) and Prof. Alfredo Marzocchi (chairman of the Faculty of Mathematics, Physics and Natural Sciences, Catholic University of Brescia)

09:00 *Gudrun Schütze* (vice chair of WGE) – News from the LRTAP Convention

09:15 *Harry Harmens et al.* – Overview of the achievements of the ICP Vegetation in 2011.

09:40 *Marcus Schaub et al.* – An update from the ICP Forests.

09:55 *Patrick Büker et al.* – Update on the DO<sub>3</sub>SE model and outreach activities.

10:15 *Riccardo Marzuoli et al.* – Overview on ten years of ozone research at the Catholic University of Brescia.

**10:30 – 11:00 Coffee/tea and poster viewing**

**Session 2:** **11:00 – 12:15 Plenary session** Chair: **Felicity Hayes**

11:00 *Harry Harmens et al.* – The ICP vegetation study on the effects of ozone on carbon sequestration in Europe.

11:25 *Per Erik Karlsson* – Ozone impacts on carbon sequestration in Northern and Central European Forests.

11:45 *Håkan Pleijel et al.* – Ozone risk for vegetation in the future climate of Europe based on stomatal ozone uptake calculations.

12:05 General discussion

**12:15 – 14:00 Lunch**

**Session 3: 14:00 – 15:30 Two parallel sessions: Ozone and moss survey**

**Session 3a: Ozone: Effects on food security Chair: Alessandra De Marco**

14:00 *Gina Mills* et al. – The ICP vegetation report on impacts of ozone on food security, including the first flux-based European crop loss assessment.

14:30 *Giacomo Gerosa* et al. – Does ozone negatively affect durum wheat?

14:50 *Jacques Berner* et al. – Ozone has a negative impact on the photosynthetic capability of maize and wheat.

15:10 General discussion

**Session 3b: Moss survey: temporal trends Chair: Louise Foan**

14:00 *Eiliv Steinnes* et al. – Three decades of atmospheric metal deposition in Norway as evident from analysis of moss samples.

14:20 *Marina Frontasyeva* – Moss biomonitoring in Russia: past, present and future.

14:40 *Gunilla Pihl Karlsson* et al. – Metal content in mosses continues to decline in Sweden.

15:00 Poster slides (2 min per poster).

**15:30 - 16:30 Extended coffee/tea and poster viewing (with authors at the posters)**

**Session 4: 16:30-18:00 Two parallel sessions: Ozone and moss survey**

**Session 4a: Ozone: biomonitoring with bean Chair: Rocio Alonso**

16:30 *Kent Burkey* et al. – Assessment of the snap bean ozone bioindicator system under different management regimes

16:50 *Felicity Hayes* et al. – An overview of results from the ICP Vegetation ozone biomonitoring with bean (2008 - 2011).

17:10 *Elisabetta Salvatori* et al. – A chamber fumigation study to evaluate the performance of the snap bean biomonitoring system under simulated Mediterranean climatic conditions.

17:30 *Gina Mills* – Discussion on the future of ozone biomonitoring within the ICP Vegetation.

**Session 4b: Moss survey: current status and future Chair: Harry Harmens**

16:30 Harry Harmens – Status of data submission for the 2010/11 European moss survey, outline of reporting in 2013, POPs review.

16:50 Discussion on the future of the European moss survey.

17:30 Jesus Santamaria – Preliminary results of <sup>15</sup>N analyses in mosses.

**18:00- 18:45 Musical aperitif with university choir and piano+cello suite (e = mc<sup>2</sup>)**

## Thursday 2<sup>nd</sup> February, 2011

**Session 5: 8:30 – 10:00 Two parallel sessions: Ozone and moss survey**

**Session 5a: Ozone: combined effects with other drivers on grasslands  
Chair: Eleni Goumenaki**

08:30 *Matthias Volk* et al. - Responsiveness of subalpine grassland productivity under increased N and O<sub>3</sub> deposition determined by carry-over effects and climate impacts of extreme years.

08:50 *Seraina Bassin* et al. – Effects of elevated O<sub>3</sub> and N deposition on ecosystem N pools and the fate of a <sup>15</sup>N isotope tracer.

09:10 *Hector Calvete* et al. – Response to ozone and nitrogen of Mediterranean annual pasture sown in natural soil: gas exchange, yield and biodiversity.

09:30 *Nathan Callaghan* et al. – Combined effects of ozone and drought on model mesotrophic grassland communities.

09:50 General discussion.

**Session 5b: Moss survey Chair: Zdravko Spiric**

08:30 *Louise Foan* et al. – Validation of mosses as biomonitors of POPs pollution: Spatial trends of PAH concentrations in mosses from France, Switzerland and Spain - Comparison with heavy metal concentrations, C and N content and their stable isotope signatures  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ .

08:50 *Ludwig De Temmerman* et al. – Is there any direct uptake and translocation of air borne trace elements in root and legume vegetables?

09:10 *Dinesh Saxena* – Seasonal atmospheric metal data by moss analysis of last 5 years from India.

09:30 *Izquieta* et al. – *Pleurochaete squarrosa* (Brid.) Lindb: an alternative for heavy metals and nitrogen monitoring in southern Europe.

09:50 General discussion.

**10:00 - 10:30 Coffee/tea and poster viewing**

**Session 6: 10:30-12:15 Two parallel sessions: Ozone and moss survey**

**Session 6 a: Ozone: reporting back from workshop Chair: Gina Mills**

10:30 Reporting back from the two ozone workshop sessions and discussion.

11:00 Reports from other ozone expert groups established in 2011.

11:15 Future workplan on ozone, including impacts of ozone on ecosystem services and biodiversity, followed by a general discussion.

**Session 6b: Moss survey** **Chair: Gunilla Phil Karlsson**

- 10:30 *Zdravko Spiric* et al. – Moss biomonitoring in Croatia in 2010.
- 10:50 *Trajče Stafilov* et al. – Moss biomonitoring of atmospheric pollution with heavy metals in the Republic of Macedonia.
- 11:10 *Panvera Lazo* et al. – Moss biomonitoring in Albania: present and future.
- 11:30 General discussion.

**12:15 – 14:00 Lunch**

**Session 7: 14:00 – 16:00 Plenary session**

**Chair: Harry Harmens**

- Reporting back from ozone and heavy metals sessions.
- ICP Vegetation work programme 2013 – 2015.
- Collaboration with other relevant bodies/organizations.
- Conclusions and review of the 25<sup>th</sup> Task Force Meeting.
- Next Task Force Meeting.
- Any other business.

**16:30 – 18:00 Excursion: visit to the Museal and Archaeological area of Santa Giulia (5 minutes by foot from the meeting venue, ticket €.-, to be paid at the museum)**

**19:30 Conference dinner in Paolo VI**

**Friday 3<sup>rd</sup> February, 2012**

Departure: participants are to make their own travel arrangements to the airport.



# **Ozone workshop**

**Abstracts**

**Oral**

**Presentations**

## OZONE CRITICAL LEVELS FOR MEDITERRANEAN FORESTS

Rocío Alonso\*<sup>1</sup>, Vicent Calatayud<sup>2</sup>, Ángela Ribas<sup>3</sup>, Giacomo Gerosa<sup>4</sup>, María Díaz de Quijano<sup>5</sup>, Susana Elvira<sup>1</sup>, Esperanza Calvo<sup>2</sup>, Ricardo Marzuoli<sup>4</sup>, Josep Peñuelas<sup>5</sup>, Filippo Bussotti<sup>6</sup>, Ignacio González-Fernández<sup>1</sup>

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Ozone (O<sub>3</sub>) pollution is especially relevant for the Mediterranean region where climatic conditions favor O<sub>3</sub> photochemical formation and persistence. Many studies have reported O<sub>3</sub>-induced effects on the physiology and growth of Mediterranean forest species. However, some discrepancies exist between the predicted O<sub>3</sub> effects on Southern European forests based on current O<sub>3</sub> critical levels, and the smaller effects observed in the field. Several reasons have been proposed to explain this discrepancy such as the inadequacy of the critical level or the inherent higher O<sub>3</sub> tolerance of Mediterranean vegetation. A review of O<sub>3</sub> exposure experiments performed with Mediterranean tree species has been carried out. Only those experiments estimating O<sub>3</sub>-induced effects on tree biomass or growth of evergreen tree species growing under Mediterranean climate conditions were considered. A database of 15 experiments including 7 different tree species and 4 experimental sites was used to derive exposure- and flux-based response functions. Ozone fluxes were estimated using both local and the generic stomatal conductance parameterization proposed in the CLRTAP- Mapping Manual for Mediterranean evergreen vegetation. Ozone-induced effects were better related to stomatal fluxes than to the exposure expressed as AOT40, especially in those experiments including a drought stress treatment. Mediterranean evergreen tree species seem to be more tolerant to O<sub>3</sub>, thus a higher O<sub>3</sub> critical level seems to be more relevant for O<sub>3</sub> damage risk assessment in Southern Europe. Current proposed flux-based critical level for European forests would protect Mediterranean forests but they would overestimate the effects over large areas.

## FLUX-RESPONSE RELATIONSHIP OF FAGUS SYLVATICA: WHAT DOES THE EPIDEMIOLOGICAL DATA ANALYSIS TELL US

Authors: Braun S.<sup>1)</sup>, Rihm B.<sup>2)</sup>, Schindler C.<sup>3)</sup>

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Ozone is causing growth losses in a variety of ecosystems including forests and thus affects carbon sequestration. It is, however, difficult to quantify this effect as the flux based critical level for forest trees bases on dose-response curves from experiments with saplings and young trees differ from mature trees in various aspects. Apart from expensive fumigation experiments with mature trees, epidemiological data analysis of growth data may help to fill this gap.

Epidemiological data analysis requires a model of ozone load which reflects the variations in space and time. Rihm and Braun (this volume) have established such a model for Switzerland. Annual ozone uptake was modelled for 94 permanent observation plots of beech for the years 1991-2006. In these forest plots, stem increment is measured at an interval of 4 years (60 trees per plot) whereas shoot growth data are available in annual resolution for 8 trees per plot. These data were normalized within each tree to the long term average and then subjected to a multivariate analysis including drought, climate and ozone. Using a hydrological model for each site, drought was parametrized as ratio between actual and potential evapotranspiration, as average soil water potential or as the time with soil water below a certain threshold. Drought was a confounding factor for AOT40, much less for ozone uptake.

The aim was to combine shoot and stem growth for an estimate of volume increment. The regression for stem growth revealed, however, some problems with confounding factors which could not be solved yet. The regression coefficient for shoot growth suggests a growth reduction for beech which is quite similar to the experimental dose-response curve, implying that ozone flux is predicting growth loss well.

**Acknowledgment:** This work was financed by the Federal Office for the Environment (FOEN), Air Pollution Control and NIR Division.

# FO<sub>3</sub>REST PROJECT - FIRST APPLICATION OF DO<sub>3</sub>SE MODEL ON FRENCH AND ITALIAN FORESTS: COMPARISON OF RISK INDICATORS FOR MEDITERRANEAN TREES

De Marco A.<sup>1</sup>, Sicard P.<sup>2</sup>, Paoletti E.<sup>3</sup>, Vitale M.<sup>4</sup>, Renou C.<sup>2</sup>, Taburet G.<sup>2</sup>

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In Europe, especially in the Mediterranean region, background ozone levels are gradually increasing. At present, the European standard for forest protection is the AOT40 index, based on the atmospheric ozone concentrations. Many studies have suggested that the stomatal flux-based approach is scientifically-sound and would be a useful tool for ozone risk assessment. In fact, stomatal uptake is limited by drought during the summer in Mediterranean regions where ozone concentrations are high. Ozone pollution is pronounced in regions with strong photochemical activity, such as the Mediterranean basin. O<sub>3</sub> levels regularly exceed the critical thresholds for forests in the given region.

A comparison of the maps of total stomatal ozone uptake (POD0), threshold-based phytotoxic ozone dose (POD1), and concentrations exceeding 40 ppb (AOT40) for *Pinus halepensis* and *Fagus sylvatica* was conducted in South-eastern France and North-western Italy. In order to calculate ozone fluxes, meteorological data (air temperature, relative humidity, soil water content and solar radiation), soil data and ozone concentrations for 2010 and 2011 were calculated from the coupled MM5-CHIMERE modelling system. The CHIMERE multi-scale model is primarily designed to produce daily forecasts of ozone, aerosols and other pollutants and make long-term simulations for emission control scenarios. The regional chemical and transport model CHIMERE (IPSL/LMD) is coupled with the MM5 mesoscale meteorological model (NCAR). For this work data has been provided at 1-h temporal resolution and a spatial resolution of 9×9km across a study area. The comparison of obtained maps shows the different distribution of AOT40 and flux in the selected region. FO<sub>3</sub>REST provides an opportunity to infer an evaluation of the DO<sub>3</sub>SE model parameterization through a comparison of observed and modeled total O<sub>3</sub> concentration and to extend the number of DO<sub>3</sub>SE model evaluation studies conducted under “Mediterranean style” conditions.

## **Acknowledgement**

This work has been made possible with the contribution of the LIFE financial instrument of the European Union (LIFE10 ENV/FR/208) and thanks to the good cooperation and harmony between the project partners: ACRI-ST, GIEFS, IPP-CNR, IPLA and ENEA.

## APPLYING FLUX BASED OZONE RISK ASSESSMENT METHODS IN THE UK

Emberson, L.<sup>1</sup>, Büker, P.<sup>1</sup>, Kitwiroon, N.<sup>2</sup>, Beever, S.<sup>2</sup>

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<sup>2</sup>*King's College London, Environmental Research Group, London, SE1 9NH, U.K.*

This study presents results of the application of the UNECE (2008 & 2010) Mapping Manual O<sub>3</sub> flux-based methods to assess O<sub>3</sub> damage to forests, arable crops and grasslands. These methods use O<sub>3</sub> data and associated meteorology from both the EMEP and CMAQ photooxidant models providing estimates of flux at 50x50 and 10 x10 km, respectively. The 'Atlantic Central European' parameterisations of the flux models are used where available to provide climate- specific estimates of stomatal O<sub>3</sub> flux and damage. Results are presented as PODy values, yield losses, biomass losses, and for arable crops, estimates of production and economic loss. These results are interpreted to inform the debate on national application of flux models by investigating i) the importance of climate- specific parameterisations of flux models, ii) an assessment of the consequences - across the latitudinal breadth of the UK - of moving to a thermal as compared to a fixed-time estimate of the O<sub>3</sub> accumulation period for wheat, and iii) the importance of incorporating a new soil moisture deficit model to assess the influence of drought on O<sub>3</sub> flux with consequences both for ecosystem damage and human health.

# MEASURED AND MODELLED STOMATAL AND NON-STOMATAL OZONE FLUXES IN A MIXED MEDITERRANEAN FOREST

S. Fares<sup>1</sup>, G. Matteucci<sup>2</sup>, G. Scarascia Mugnozza<sup>1</sup>, A. Morani<sup>2</sup>, C. Calfapietra<sup>2</sup>, F. Manes<sup>3</sup>, E. Salvatori<sup>3</sup>, L. Fusaro<sup>3</sup>, E. Paoletti<sup>4</sup>, F. Loreto<sup>4</sup>.

<sup>1</sup>CRA (Agricultural Research Council), Research Center for the Soil-Plant System, Rome, Italy; <sup>2</sup>CNR (National Research Council), Institute of Agro-environmental and Forest Biology, Rome, Italy; <sup>3</sup> Department of Plant Biology, University of Rome "La Sapienza", Rome, Italy; <sup>4</sup>CNR, Istituto per la Protezione delle Piante, Sesto Fiorentino, Florence, Italy.

In September 2011, an experimental site has been equipped in a mixed Mediterranean forest located inside the Presidential Estate of Castelporziano, Rome, central Italy, composed by *Quercus ilex*, *Quercus suber*, *Pinus pinea*, *Laurus nobilis*, *Arbutus unedo*. The main goal was to initiate a long-term investigation of fluxes of ozone, water and CO<sub>2</sub> with the Eddy Covariance technique in order to quantify ozone deposition to the forest canopy and characterize its dependence on stomatal and non-stomatal drivers. Continuous measurements from September to December highlighted the forest canopy as a relevant ozone sink, with total ozone fluxes up to 10 nmol m<sup>-2</sup> s<sup>-1</sup> during the central hours of the day (Figure 1 a). Stomatal conductance was calculated by inversion of the Monteith equation through available water fluxes and meteorological parameters. Results show that stomata are a minor sink for ozone, contributing up to 15 % to the total ozone flux. Low levels of stomatal conductance can be explained by drought stress in the warm days of September and October, while the cold days affected the phenology of trees thus limiting the stomatal conductance. The occurrence of high levels of non-stomatal ozone fluxes suggests an important role of non-stomatal sinks (wet and dry deposition to soil and plant surfaces, reaction in the gas phase between ozone and volatile organic compounds) in the total ozone removal, and the need for a long-term measurement to understand how seasonal dynamics can affect plant ecophysiology and ozone fluxes. A second goal of the study was to parameterize the DO<sub>3</sub>SE model (Deposition of Ozone for Stomatal Exchange, Emberson et al. Env. Pol. 2000), used for ozone-risk assessment and test the model performance against the measured values with Eddy Covariance. For this purpose, for each species series of ecophysiological parameters were measured at leaf level in the field using portable infrared analyzers, and used to model stomatal conductance as a function of vapor pressure deficit, temperature, and light. Comparisons of measured and modeled stomatal conductances (Figure 1 b) showed that modeled values were underestimated by about 10 %. Current processing activities are aimed at testing alternative methods to model stomatal conductance (e.g. Ball-Berry approach) and at quantifying non-stomatal ozone sinks.

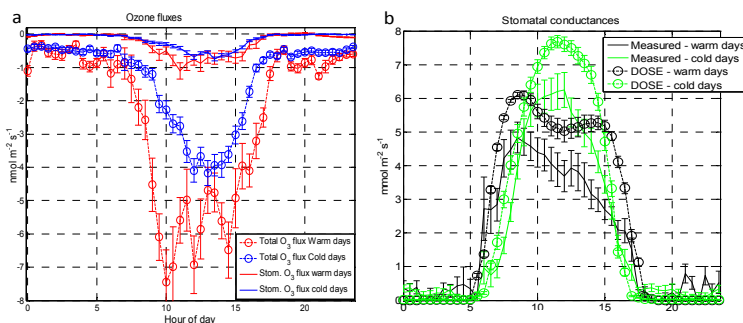


Figure 1: Ozone fluxes and stomatal conductances during warm and dry days (DOY 249-280) and during cold and wet days (DOY 281-349).

**Acknowledgments:** We are grateful to the Scientific Committee of the Presidential Estate of Castelporziano and to its staff, in particular to Ing. Alejandro Tinelli, for the scientific and financial support. Thanks also to the EU project Marie Curie-IAPP "Innovative Application of PTR-TOF mass spectrometry in plant biology (PTR-TOF)", the EU EXPEER project (GA no. 262060), and the UE LIFE project (ENV/FR/000208) for financial help. We finally want to acknowledge Roberto and Valerio Moretti and Filippo Ilardi for helping with the maintenance of the experimental site.



WHEN STOMATAL FLUX IS PREDICTABLE FROM AOT40.  
RESULTS OF A 13 YEARS STOMATAL FLUX CALCULATION EXERCISE  
AT AN ALPINE SPRUCE FOREST WITH THE DO<sub>3</sub>SE MODEL

Gerosa G.A.<sup>1</sup>, Finco A.<sup>2</sup>, Marzuoli, R.<sup>1,2</sup>, Ferretti M.<sup>3</sup>, Gottardini E.<sup>4</sup>

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The evaluation of the ozone risk for mountain forests in remote areas often starts with the measurements of in situ ozone concentrations by means of passive samplers.

Thirteen years (1996-2009) of summer ozone measurements taken with passive samplers in an Alpine spruce forest at Passo Lavazé (Italy, 1750 m a.s.l.) have been used for a stomatal ozone dose estimation exercise by means of a DO<sub>3</sub>SE type model.

The hourly ozone concentrations required by the model have been derived by disaggregation of the weekly concentration means assuming an average daily ozone concentration course typical of the mountain areas located at the same relative elevation of the testing site (Loibl et al., 1994; Gerosa et al. 2007).

Meteorological data have been taken from an in situ monitoring station running from 2002. For the years preceding 2002 a mean meteorological season has been estimated by averaging hour by hour all the meteo data from 2002 and 2009.

Model simulations have been made for a 28 m tall forest canopy with a constant LAI of 2.94. A maximum stomatal conductance value of 125 mmol m<sup>-2</sup> s<sup>-1</sup> has been employed as indicated by (UN/ECE 2009). Soil moisture has been estimated by a daily water balance taking into account the soil texture and a rooting depth set to 1 m.

The results show a high exposure of forest to ozone (AOT40 of the daylight hours between a minimum of 30,000 ppb•h and a maximum of 87,000 ppb•h) and an important stomatal flux (AFst0) between 36.3 and 49.4 mmol m<sup>-2</sup>. Even taking into account the detoxifying capacity of the plants, simulated by the introduction of the 1.6 nmol m<sup>-2</sup> s<sup>-1</sup> instantaneous flux threshold (AFst1.6), the phytotoxycal ozone dose resulted between a minimum of 22.1 and a maximum of 35.7 mmol m<sup>-2</sup>, well above the critical level of 8 mmol m<sup>-2</sup> proposed by the UN/ECE for Norway spruce.

The ozone exposure and dose are well linearly correlated ( $R^2 = 0.85$ ), revealing the almost negligible influence of water availability on the stomatal conductance for this site. Conversely, stomatal conductance, and the absorbed ozone dose, seem to be more strongly limited by the air temperature and the VPD values.

#### References

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- Loibl, W., Winiwarter, W., Kopsca, A., Zueger, J., Baumann, R., 1994. Estimating the spatial distribution of ozone concentrations in complex terrain. *Atmospheric Envir.* 28, 2557-2566
- UN/ECE 2009. ICP Modeling and Mapping. Manual on methodologies and criteria for modeling and mapping critical load and levels and air pollution effects, risks and trends.

## INTRODUCTION OF ENVIRONMENTAL OBJECTIVES FOR OZONE IMPACTS ON VEGETATION IN SWEDEN BASED ON OZONE FLUX

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16 environmental quality objectives are used in Sweden in order to ensure environmentally sustainable development. One of the objectives, “Clean Air”, is defined as that “The air must be clean enough not to represent a risk to human health or to animals, plants or cultural assets”. This objective presently includes a target value for ozone impacts on vegetation, defined as that daylight AOT40 April-Sept should not exceed 5 ppm h.

The target value should as far as possible reflect important, unacceptable ozone impacts on vegetation in Sweden. It should also be able to reflect changes in the impacts as a result of future climate changes. Finally, it should be constructed so that it is possible to be applied by local authorities.

In this study we analyze the possibilities to introduce a target value for ozone impacts on vegetation in Sweden based on, or related to, ozone flux. We consider the following concepts: AOT40, POD<sub>y</sub> and finally a partly new concept, “the flux modified AOT40”.



## OZONE SIMULATIONS OVER ITALY WITH ATMOSPHERIC MODELLING SYSTEM OF THE MINNI PROJECT: EVALUATION AND PERSPECTIVES FOR VEGETATION

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The capacity of MINNI atmospheric modelling system to simulate ozone concentrations over Italy, according to the EU criteria, has been analysed. The MINNI modelling system (Zanini et al., 2005) is the Italian Integrated Assessment Modelling System for supporting the International Negotiation Process on Air Pollution and assessing Air Quality Policies at national/local level sponsored by the Italian Ministry of the Environment. The MINNI system is composed by an Atmospheric Modelling System (AMS) and the Greenhouse Gas and Air Pollution Interactions and Synergies Model over Italy (GAINS-Italy) (D'Elia et al., 2009). The main components of AMS are the meteorological model (RAMS) (Cotton et al., 2003) for simulating the meteorological conditions, the Emission Manager and the air quality model (FARM) for simulating the atmospheric chemistry (Silibello et al., 2008). Yearly simulations were carried out using national emission inventories for Italy and EMEP emission inventories for the other countries included in the computational domain.

A comparison between model simulation and monitoring data has been carried out. At most of the background stations (rural, urban, suburban) considered in the evaluation, the comparison of simulated and measured ozone concentrations shows a good agreement. According to AOT40 outcomes, surface ozone concentrations over Italy exceed often the thresholds established in EU legislation to protect vegetation. In order to improve the assessment of ozone damages on vegetation at national scale, a specific modelling tool, integrated in the MINNI system, based on the ozone stomatal fluxes, instead of exposure, and tailored on selected species is under development.

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MAPPING OZONE FLUXES FOR SWITZERLAND: COMPARISON WITH EMEP-MAPS  
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The aim of this study was to produce maps of ozone flux that can be used for epidemiological analysis of tree growth data, for comparing with EMEP-maps and other purposes. In a first step, the DO<sub>3</sub>SE model was run at 23 rural monitoring stations (Figure 1, triangles) for the period 1991–2006 to calculate ozone flux doses POD1. The required hourly input-data (ozone and climatic parameters) were measured on-site; pressure was adjusted to altitude.

In a second step, a map of the POD1 average 1991–2006 was produced for beech (Figure 1). Given the relatively small number of stations, geo-statistical approaches like kriging are not adequate. Therefore, we tried to find predictor maps correlated with the spatial distribution of POD1. The following maps were available and examined in multiple regression analysis: AOT40 (ppm·h, mean 1998-2002), ozone concentration, NO<sub>2</sub> concentration, global irradiance, relative humidity (% , mean April-September, 1990-2006), altitude, relative altitude (km above lowest point in 5 km radius), precipitation, temperature and wind speed. The optimal equation found was:

$$\text{POD1} = -78.0 - 6.35 \cdot \text{rel\_altitude} + 0.0469 \cdot \text{AOT40} + 1.14 \cdot \text{rel\_humidity}, R^2 = 0.42, n = 23.$$

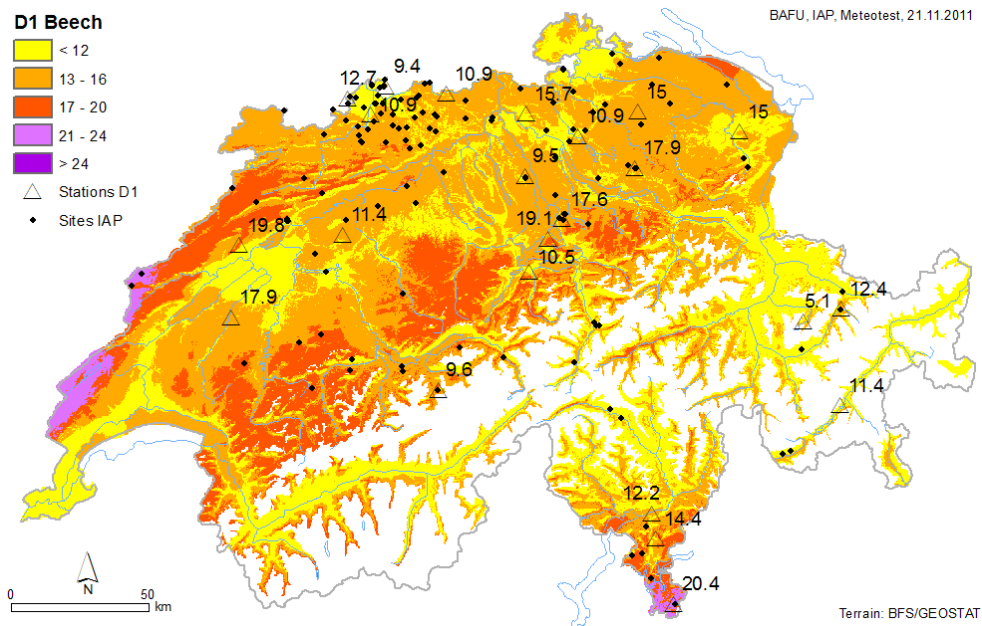


Figure 1: Map of POD1 for beech [ $\text{mmol m}^{-2} \text{yr}^{-1}$ ], average 1991–2006, cantonal and national monitoring stations ( $\Delta$ ) as well as tree observation sites of the IAP ( $\bullet$ ).

In a third step, yearly POD1 maps were produced by spatially interpolating the residuals of the monitoring stations (POD1 of the specific year divided by the average POD1) using an inverse-distance weighting method. In a similar way further yearly maps were produced: POD1 for spruce, AOT40 and flux variants with IAP-specific parameterization.

In a first comparison with EMEP-data (Dec. 19 2011), POD1 and AOT40 from EMEP seemed to be too high for Switzerland.

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# **Task Force meeting**

**Abstracts**

**Oral**

**Presentations**

## EFFECTS OF ELEVATED O<sub>3</sub> AND N DEPOSITION ON ECOSYSTEM N POOLS AND THE FATE OF A <sup>15</sup>N ISOTOPE TRACER

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To investigate the effects of elevated ozone (O<sub>3</sub>) and nitrogen (N) deposition on a subalpine pasture, 180 turf monoliths were exposed at Alp Flix, Switzerland (2000 m a.s.l.) in a free air fumigation system to a combination of three O<sub>3</sub> levels and five N loads. With the aim to study the pollutant impact on ecosystem N pools, a <sup>15</sup>N stable isotope tracer experiment was carried out in the seventh year in a subsample of 36 monoliths exposed to the most extreme treatments and their combinations: control, + 50 kg N ha<sup>-1</sup> yr<sup>-1</sup> (N50), 1.6 x ambient O<sub>3</sub> (O<sub>3</sub>++). After snowmelt in May, 0.04 g m<sup>-2</sup> <sup>15</sup>N stable isotope tracer in the form of double-labelled NH<sub>4</sub>NO<sub>3</sub> was added to the monoliths. On days 3, 62, and 112 after application, two soil cores were taken (6 cm diameter, 10 cm depth) per monolith. N pools and <sup>15</sup>N recovery (% of added tracer) were calculated for green biomass, harvested green biomass, necromass, roots, microbial biomass, soil extractable N, and immobilised soil N. Furthermore, leachate was collected in 2-4 week intervals during the growing season from a subsample of the monoliths and analysed for N content and tracer recovery.

Elevated N deposition strongly increased all plant N pools (+1.8 g m<sup>-2</sup> N in green biomass (+49%), +1.9 g m<sup>-2</sup> in necromass (+54%), +6 g m<sup>-2</sup> in root biomass (+36%)). Of the green biomass, the amount of N exported by harvest was almost doubled, namely 1.36 g N m<sup>-2</sup> more in N50 compared to the control. The other ecosystem N pools and summarised leachate N were not affected by N addition. When all N pools are summed up, the treatment induced difference accounted for approximately 9.6 g N m<sup>-2</sup>, which is substantially less than the sum of 30 g N m<sup>-2</sup> that was added to the ecosystem in the N50 treatment in the preceding six growing seasons. Tracer recovery was increased by N addition only in harvested green biomass and temporarily in soil extractable N, while the recovery of <sup>15</sup>N immobilised in soil was reduced. Overall, these results show that almost all of the additional N is accumulated in living and dead phytomass, but even less N is immobilised in soil compared to the control. By the time there is no explanation for the large gap between sum of added N and total ecosystem N pool.

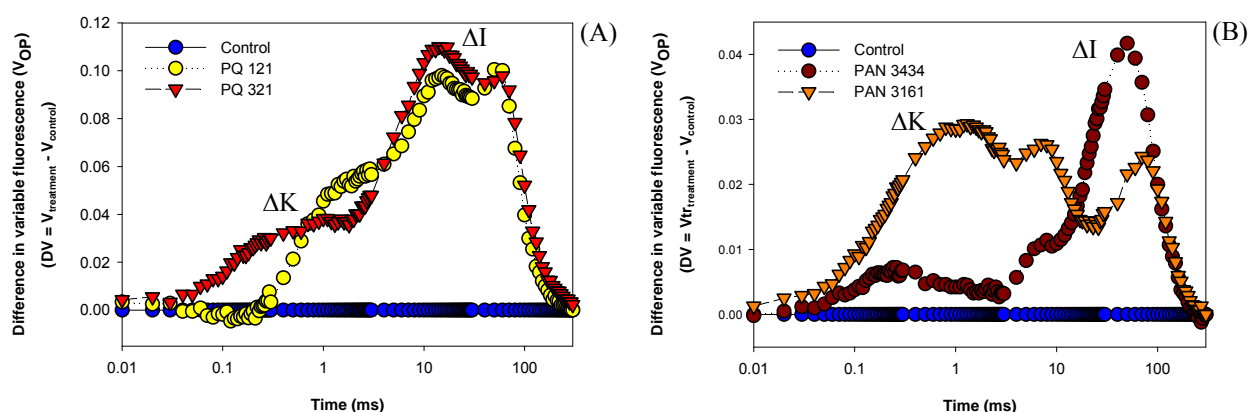
Elevated O<sub>3</sub> concentrations significantly reduced the necromass N pool by 13%, while the microbial N pool was increased by 10%. All other compartments remained unaffected. Correspondingly, tracer recovery was reduced by O<sub>3</sub> by 25% in necromass and it was increased by 25% in microbial biomass. Presumably, necromass decay rates were increased through altered litter quality, which in turn stimulated soil microbial biomass.

# OZONE HAS A NEGATIVE IMPACT ON THE PHOTOSYNTHETIC CAPABILITY OF MAIZE AND WHEAT

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The effect O<sub>3</sub> and on wheat and maize cultivars were investigated by making use of an open-top chamber (OTC) facility. A wick irrigation system was employed to control the water regime of the potted plants located inside the OTCs. Both the wheat and maize plants were fumigated for 8 hours a day, 7 days a week with 80 ppb O<sub>3</sub>. Two wheat cultivars PAN 3434 and PAN 3161 and two maize cultivars PQ 121 and PQ 321 were used. The effect O<sub>3</sub> on photosynthesis was measured by means of chlorophyll *a* fluorescence. Analysis of the O-J-I-P transients revealed  $\Delta K$ , and  $\Delta I$  peaks (Fig. 1) appearing within both the wheat and maize cultivars. A  $\Delta K$  peak is a consequence of an increase in fluorescence resulting from the uncoupling of the oxygen evolving complex of PSII. The formation of a  $\Delta I$  peak indicates the inhibition of electron flow downstream of PSI. Further analysis of the chlorophyll fluorescence transients, showed that PSII function was mainly inhibited with respect to the multiple turnover phase of primary photochemistry, i.e. electron transport beyond the quinone electron acceptor QA. Ozone exposure resulted in disengagement of the oxygen-evolving complex of photosystem II of both wheat and maize. A significant ( $P < 0.05$ ) decrease in the performance index ( $PI_{ABS, TOT}$ ) of both the wheat and the maize cultivars were observed indicating that the vitality of the plants are reduced with O<sub>3</sub> fumigation. Comparing the two wheat cultivars to each other, it was evident that there were differences in the photochemistry in response to O<sub>3</sub>. The manner, in which the photochemistry was affected by O<sub>3</sub>, seems to be dependent of the genetic background of the cultivar. The differences in the photochemistry of the maize cultivars were less significant.



**Figure 1** Difference (DV) in the relative variable chlorophyll *a* fluorescence transients, normalized between 0.05 and 300 ms [ $F_0$  and  $F_P$  ( $V = (F - F_0) / (F_P - F_0)$ ),  $\Delta V_{OP} = V_{OP, treatment} - V_{OP, control}$ ] for (A) two maize cultivars and (B) two wheat cultivars after 3 weeks' fumigation with O<sub>3</sub>.

## ASSESSMENT OF THE SNAP BEAN OZONE BIOINDICATOR SYSTEM UNDER DIFFERENT MANAGEMENT REGIMES

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**Introduction:** Ozone-sensitive (S156) and tolerant (R123) snap bean genotypes described previously (Burkey, Miller, and Fiscus 2005 J Environ. Qual. 34: 1081-1086) are being evaluated under different management scenarios as a potential ozone bio-indicator system. Results from pot studies and field trials are reported here. **Methods:** In Raleigh, S156 and R123 were grown in pots according to the experimental protocol recommended by the ICP-vegetation Program with an irrigated in-ground study established in an adjacent field. In Urbana, a field study was conducted under ambient air and elevated ozone treatments in the absence of irrigation at the SoyFACE site (<http://soyface.illinois.edu>). **Results and Conclusions:** Under similar AOT40 exposures in Raleigh, ozone suppression of pod yield in S156 relative to R123 was not found in the ICP pot study (Table 1), but was observed in the field (Table 2). This may be attributed to large numbers of undeveloped R123 pods (Table 1) at 79 days after planting (DAP) so that R123 yields were underestimated. This may not have been a factor in the field study harvested two weeks later at 94 DAP. Local conditions may affect timing of S156 and R123 pod development, requiring a growth period greater than 12 weeks to fully realize ozone effects on yield. Under non-irrigated conditions, the S156/R123 system detected yield effects at an AOT40 of 16.2 ppm h relative to 5.3 ppm h (Table 2).

Table 1. S156 and R123 snap bean yield at Raleigh, NC in 2011 with plants grown in pots. Planting was on June 8 with final harvest on August 28 at 79 days after planting (DAP). The seasonal AOT40 was 10.1 ppm h.

Parameter (per plant basis)	S156	R123	S156/R123
Number of pods > 4 cm with seeds	61 ± 6	49 ± 4	1.24
Dry weight (g) of pods > 4 cm with seeds	34 ± 4	28 ± 3	1.21
Number of pods > 4 cm with no seeds	5 ± 1	46 ± 10	0.11
Dry weight (g) of pods > 4 cm with no seeds	0.3 ± 0.1	3.2 ± 1	0.08

Table 2. S156 and R123 snap bean pod yield in field grown plants at Raleigh, NC (2011) and the SoyFACE site at Urbana, IL (2006) using different management regimes.

Site	Management Regime	Harvest DAP	Seasonal AOT40 (ppm h)	S156 (g DW plant-1)	R123 (g DW plant-1)	S156/R123
Raleigh	ambient air irrigation	94	11.2	7.0 ± 1.0	17.2 ± 2.1	0.47 ± 0.06
Urbana	ambient air no irrigation	96	5.3	5.2 ± 0.8	6.9 ± 1.1	0.78 ± 0.05
Urbana	1.4x ambient ozone no irrigation	96	16.2	1.9 ± 0.3	5.8 ± 0.9	0.35 ± 0.05

# COMBINED EFFECTS OF OZONE AND DROUGHT ON MODEL MESOTROPHIC GRASSLAND COMMUNITIES

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Possible future global climate change scenarios include the co-occurrence of increased background tropospheric ozone and more frequent droughts during the plant growing season. This study exposed mixed plant community mesocosms containing the dominant functional groups from a mesotrophic grassland (grass / legume / forb), to a range of atmospheric ozone concentrations (0 - 90 ppb) and 2 watering regimes (south east England 30 year average vs. year 2003 drought) for a 16 week period during 2009 and for an 18 week period in 2010. The stomatal behaviour of the constituent grassland species was measured and used to model the phytotoxic ozone dose ( $POD_1$ ). Commutative seasonal ozone dose was related to green, senescent and total biomass at species, functional group and whole canopy level for the well watered and droughted mesocosms. Despite there being few interactions between drought and ozone stress in most species, co-occurring stresses generally resulted in an additive reduction in, total plant biomass and often increases in senescent and dead biomass. As well as a decrease in productivity at the canopy level, ozone and drought stress are also drivers of species change at the community level; changes in plant community composition were driven by competition between species of differing stress sensitivities and the associated changes in canopy level ozone uptake. Drought and ozone stress drove the mixed grass-legume-forb community to one dominated by grasses, a process amplified when these stresses occurred together. There was also some benefit to understory plants through a reduction in competition for light associated with reduced biomass in the upper canopy. This study shows that when investigating the impacts of future climate scenario on ecosystems, it is important not only to consider the stresses but also the effect that competition between species and resultant changes in canopy-atmosphere interaction can have on both productivity and biodiversity.

## RESPONSE TO OZONE AND NITROGEN OF MEDITERRANEAN ANNUAL PASTURE SOWN IN NATURAL SOIL: GAS EXCHANGE, YIELD AND BIODIVERSITY

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Pastures are among the most important ecosystems in Europe considering the high biodiversity of its plant communities and the extension covered in the European territory. In the last decade, a big effort has been done to characterize the response of these communities to increasing tropospheric ozone (O<sub>3</sub>) levels and nitrogen (N) deposition, two of the main drivers of global change. Under the framework of the LRTAP Convention, critical levels and loads of O<sub>3</sub> and N deposition have been defined for different plant communities. However more research is still needed to define these values for Mediterranean annual pastures.

Aiming to study in depth the response of the annual Mediterranean pastures to atmospheric pollution considering the whole atmosphere-plant-soil system, a new experimental study was performed in an Open-Top Chamber (OTC) experimental facility located in the Experimental Farm “La Higuera/CSIC”, Toledo (central Spain). Six species representative of annual pastures in the Central Iberian Peninsula (*Trifolium striatum*, *Trifolium cherleri*, *Ornithopus compressus*, *Cynosurus echinatus*, *Briza maxima* and *Silene gallica*) were sowed in the field (1000 seeds m<sup>-2</sup>). Four applications of NO<sub>3</sub>NH<sub>4</sub> were performed every 15 days aiming to reach N integrated doses of “background”, +20 or +40 Kg N ha<sup>-1</sup>. Plants were exposed for 30 days to four ozone treatments: filtered air (FA), non-filtered air (NFA) reproducing ambient levels, NFA supplemented with 40 ppb O<sub>3</sub> (NFA+) and NFA supplemented with 60 ppb O<sub>3</sub> (NFA++). Three replicated OTCs were used for each O<sub>3</sub> and N treatment. Meteorological parameters, O<sub>3</sub>, NO<sub>x</sub> and SO<sub>2</sub> concentrations were continuously monitored during the growing cycle. Three harvests were performed from plant emergence to senescence. Different parameters were measured related to plant growth and physiology (total yield, biomass production per species, gas exchange at the leaf and canopy level, plant nutrition, VOCs emissions) and related to soil processes (emission of CO<sub>2</sub>, CH<sub>4</sub>, N<sub>2</sub>O and NO<sub>2</sub>, mineral N, water-filled pore space). First results corresponding with total yield of the pasture, gas exchange at canopy level and biodiversity are presented.

Ozone significantly reduced the total green biomass of the pasture. Biomass decreased by 14% and 25% in NFA+ and NFA++ treatments respectively compared with FA. The total senescent biomass was also affected by the pollutant, causing an increase close to 40% in NFA+ and NFA++. The studied species presented different O<sub>3</sub> sensitivity patterns *T. striatum* being the most sensitive, affected even by ambient ozone levels (NFA). The differences in O<sub>3</sub> sensitivity produced a shift in the species abundance of the pasture, since the resistant *O. compressus* took advantage over the sensitive clover.

Ozone also altered soil processes, increasing the emissions of N<sub>2</sub>O. Therefore O<sub>3</sub> in the Mediterranean areas must be considered as a stress factor for annual pastures that can potentially have detrimental effects on the yield, structure and composition of these ecosystems.



## IS THERE ANY DIRECT UPTAKE AND TRANSLOCATION OF AIRBORNE TRACE ELEMENTS IN ROOT AND LEGUME VEGETABLES

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Atmospheric deposition has an impact on the concentration of trace elements in vegetables. Leafy vegetables accumulate most efficiently deposited trace elements on their well exposed leaf blades but what about root vegetables and legume vegetables? Exposure of vegetables using a biomonitoring technique proved to be a good tool to study the impact of atmospheric deposition (De Temmerman & Hoenig, 2004).

The vegetables studied were carrots (*Daucus carota*), celeriac (*Apium graveolens* var. *rapaceum*) as root crops and bean (*Phaseolus vulgaris*) as a legume crop. The test plants were grown in a reference area with low atmospheric deposition and than exposed in a polluted area around a lead smelter for 42 days (green beans), for 70 days (white beans), for 56 days (carrots), or for 112 days (celeriace). In most cases the containers were placed near kitchen gardens where vegetables were cultivated. At each experimental plot, the containers were exposed in triplicate allowing statistical treatment of the results. After exposure, the leaves of celeriac and the storage organs of carrot and celeriac were cleaned, washed thoroughly and peeled. The peels and the inner part (pulp) were analyzed separately. The beans were harvested as green beans, but some plants were left at the experimental plots until complete ripening in order to harvest the white beans. Also the unwashed leaves of carrot and bean were analyzed for comparison reasons. At each site, bulk deposition was measured in order to be able to link the results obtained for the edible parts to atmospheric deposition.

The leaves appear to be excellent accumulators of As, Cd and Pb but also the storage organs of celeriac and carrot accumulate to some extent airborne As, Cd and Pb. In the unexposed inner part of both root crops, a significant accumulation of As and Cd was observed as well, but not for Pb. As analysis of trace elements in pore water of the rooting substrate did not show any significant difference in trace element contents after long term exposure at the polluted sites in comparison with the reference site. As such direct uptake by the aboveground plant parts and translocation within the plant is most likely as far as Cd and As is taken into consideration. If there is any translocation of Pb in the plant, it will be extremely low.

In spite of the development of green beans, below an umbrella of leaves, there is a significant accumulation of Pb and As but not Cd. Is there also any significant effect on the white beans after ripening, as the seeds are not exposed at all?

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VALIDATION OF MOSSES AS BIOMONITORS OF POPS POLLUTION: SPATIAL TRENDS OF PAH CONCENTRATIONS IN MOSSES FROM FRANCE, SWITZERLAND AND SPAIN – COMPARISON WITH HEAVY METAL CONCENTRATIONS, C & N CONTENT AND THEIR STABLE ISOTOPE SIGNATURES  $\delta^{13}\text{C}$  &  $\delta^{15}\text{N}$

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Regulation of persistent organic pollutant (POP) emissions and reliable monitoring of POP concentrations in ambient air is of paramount importance because of their slow rates of degradation, toxicity and potential for long-range transport and bioaccumulation in living organisms (Aarhus protocol, 1998). Polycyclic aromatic hydrocarbons (PAHs) in particular are ubiquitous and appear to be carcinogenic, mutagenic and immunotoxic. Their concentrations in mosses have shown similar spatial patterns and temporal trends to ambient air concentrations (Holoubek et al., 2007).

This study was carried out to investigate whether mosses can be used as biomonitors of atmospheric deposition of PAHs at a European scale. Mosses *Hypnum cupressiforme* Hedw. were collected during October 2010 in 3 regions of Europe: Île-de-France (France), Navarra (Spain) and the Swiss Plateau (Switzerland). PAH concentrations were determined by High-Performance Liquid Chromatography (HPLC) associated with fluorimetric detection.

Stable isotopes at natural abundance levels in mosses also provide a powerful approach for understanding environmental interactions. Isotopic composition of elements, such as carbon and nitrogen, changes in predictable ways during their course through the biosphere, which makes them ideal tracers of the pathways and origins of these elements (Liu et al., 2008). C and N content were determined by an elemental analyzer and stable isotopic ratios  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  with an isotope ratio mass spectrometer operating in continuous flow mode.

Spatial trends of PAH concentrations, C & N contents and  $\delta^{13}\text{C}$  &  $\delta^{15}\text{N}$  isotopic ratios were plotted and treated statistically with heavy metal concentrations obtained during the 2010 ICP-Vegetation survey, as well as emission values (EPER), soil occupation (Corine Land cover), geomorphological and meteorological data.

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## MOSS BIOMONITORING IN RUSSIA: PAST, PRESENT AND FUTURE

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The use of mosses as biomonitors of atmospheric deposition of heavy metals and radionuclides in Russia started more than 30 years ago in connection with the development and problems of the nuclear and military-industrial complexes in Siberia and the Urals. In the 1990s, within the framework of UNECE ICP Vegetation programme, systematic studies using moss were carried on in north-western Russia (Karelia, Kola Peninsula, Kaliningrad, Pskov and Leningrad regions), and the results were presented in the European Atlas *Atmospheric Heavy Metal Deposition in Europe – Estimations Based on Moss Analysis*. In 1998–2002, JINR participated in the IAEA-coordinated research project “Biomonitoring of air pollution in the Chelyabinsk region (South Ural Mountains, Russia) through trace elements” in one of the most contaminated areas of the world experiencing strong ecological stress from heavy metals and radionuclides.

JINR took part in the 2000/2001 European moss survey reporting data on some areas of Central Russia (Tula, Yaroslavl and Tver regions) and in the 2005/2006 moss survey in the north-east of the Moscow region and the Republic of Udmurtia.

The active moss biomonitoring (moss bags technique) was used to study air pollution in street canyons of the intensely growing megapolice of Moscow.

A combination of instrumental ENAA at the IBR-2 reactor at JINR, Dubna, and AAS at counterpart laboratories provides data on concentrations of about 40 chemical elements (Al, As, Au, Ba, Br, Ca, Cd, Ce, Cl, Co, Cr, Cs, Cu, Dy, Eu, Fe, Hf, Hg, I, In, La, Lu, Mg, Mn, Na, Nd, Ni, Pb, Rb, Sb, S, Sc, Se, Sm, Ta, Tb, Ti, Th, V, W, Yb, Zn), which substantially exceeds the requested by the European Atlas number of elements (given in bold). Distribution of the determined elements over the sampled areas is illustrated by the contour maps produced by the Russian software package GIS-INTEGRO with raster and vector graphics.

The 2010/2011 moss survey extended study areas in Russia as PhD students, teachers and pupils of secondary schools in the Smolensk, Ivanovo, Kostroma, and Yekaterinburg regions, Stavropol area, and some districts of the Moscow and Leningrad regions were involved in terrestrial moss sampling.

The moss technique is supposed to be used for assessing sequences of the Fukushima disaster in the Far East of Russia (mapping of radionuclide distribution around the city of Vladivostok). It is also planned to use moss as natural planchette for tracing deposition of cosmic dust in peat bog cores in Western Siberia and some mountainous areas of Russia.

## DOES OZONE NEGATIVELY AFFECT DURUM WHEAT?

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Agricultural production is supposed to be negatively affected by ozone, particularly in the Mediterranean countries where the photochemical formation of this pollutant is high. However, the sensitivity of crops to the consequent oxidative stress largely depends on species and variety.

In this work the ozone sensitivity of two cultivars of durum wheat (Neodur and Virgilio) was tested in an Open-Top Chambers experiment at Curno (Po Valley, Italy). A factorial design was used with two levels of ozone concentrations: charcoal filtered air (ambient air -50%) and ozone enriched air (+35%). A total number of 8 OTCs was used in order to have 4 replicates of the two ozone treatments. Seven pots of each cultivar were placed within each OTC and three durum wheat plants were grown in each pot, keeping the irrigation at field capacity. The experiment was carried on between February and the end of June in 2011.

Onset and diffusion of leaf visible symptoms were measured with periodical visual assessments; flag-leaves samples were collected for microscopic and histochemical analysis. Stomatal conductance to water was measured on flag-leaves at morning, midday and afternoon, in six different dates between 1<sup>st</sup> May and 15<sup>th</sup> June. Main agronomical yield parameters were evaluated after the harvesting (28th June).

At the end of the growing season the ozone enriched plants experienced an AOT40 ozone exposure of 23'000 ppb·h vs an AOT40 of 700 ppb·h of the control plants.

Although *cv* Virgilio shown considerable leaf injuries in the ozone enriched treatment, no significant effects on yield parameters were detected. The same result was obtained for *cv* Neodur which developed visible symptoms with a lower intensity and diffusion than Virgilio.

Ozone reduced the number of empty ears (*n.s.*) and increased the dry weight of the stems (*n.s.*), while negligible differences on total grain weight in both cultivars were found.

Plant's *gs* response to ozone was contrasting in the two *cultivars*, being decreased in ozone treated Neodur and increased in the *cv* Virgilio with the same treatment.

The opposite stomatal behavior could explain the different susceptibility of the two cultivars to ozone in relation to leaf injuries onset.

Nevertheless the onset of leaf visible injuries and their diffusion could not be taken as a reliable predictor of yield losses at all. On the contrary, being the *cv* Virgilio more productive than the *cv* Neodur (+12% in the non-treated plants) the farmers should prefer Virgilio regardless of the foliar symptoms development.

Compared to the bread wheat, the durum wheat resulted an ozone tolerant species. The blanket application of the critical levels based on dose-response relationships for bread wheat, hence, could be very penalizing for those Mediterranean countries, such as Italy, where 2/3 of the arable land used for wheat are assigned to the cultivation of durum wheat.

## THE ICP VEGETATION STUDY ON THE EFFECTS OF OZONE ON CARBON SEQUESTRATION IN EUROPE

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The aims of this study were:

- To review current knowledge on the impacts of ozone on C sequestration and ozone absorption by vegetation and implications for climate change;
- To estimate the impacts of ozone on C storage in forests and grasslands in Europe using flux-based methods and an offline global land surface model.

We reviewed current knowledge on:

- Vegetation as a sink for ozone: non-stomatal and stomatal deposition;
- The role of biogenic volatile organic compounds (BVOCs) in the deposition, detoxification and impacts of ozone;
- Physiological processes via which ozone might ultimately reduces C sequestration;
- C sequestration potential of croplands, grasslands and forests;
- Variation in sensitivity of species to ozone: implications for C sequestration;
- Ozone effects in a changing climate (elevated CO<sub>2</sub>, reduced N deposition, enhanced drought frequency) and the implications for C sequestration;
- Implications for climate change, the global water cycle and human health.

In addition, two modelling case studies were conducted to provide either a ‘bottom up’ or ‘top down’ approach to estimate ozone-induced loss of C from living biomass of forests and grassland vegetation under both present (year 2000) and future (year 2040) climate conditions. In the ‘bottom up’ approach the DO<sub>3</sub>SE model (<http://www.sei-international.org/do3se>) was applied and in the ‘top down’ approach the JULES (<http://www.jchmr.org/jules/>) land surface model was applied. Preliminary results of these case studies will be discussed.

Furthermore, Karlsson et al. (see abstract in this issue) reported on ozone impacts on C sequestration in the living biomass of forests for some northern and central European countries, i.e. Sweden, Finland, Norway, Denmark, Estonia, Latvia, Lithuania, Poland, Czech Republic and Germany.

### **Acknowledgement**

We thank the UK Department for Environment, Food and Rural Affairs (Defra) for funding the ICP Vegetation Programme Coordination Centre. Further financial support was provided by the UNECE and the UK Natural Environment Research Council (NERC).

## OVERVIEW OF THE ACHIEVEMENTS OF THE ICP VEGETATION IN 2011

Harmens, H.<sup>1</sup>, Mills, G.<sup>1</sup>, Hayes, F.<sup>1</sup>, Norris D.A.<sup>1</sup>,  
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The ICP Vegetation is an international programme that reports on the effects of air pollutants on natural vegetation and crops [1]. It reports to the Working Group on Effects (WGE) of the UNECE Convention on Long-range Transboundary Air Pollution (LRTAP). In particular, the ICP Vegetation focuses on the following air pollution problems: quantifying the risks to vegetation posed by ozone pollution and the atmospheric deposition of heavy metals, nitrogen and persistent organic pollutants (POPs) to vegetation. In addition, the ICP Vegetation studies the impacts of pollutant mixtures (e.g. ozone and nitrogen), consequences for ecosystem services (including biodiversity and carbon sequestration) and interactions between air pollutants and climate change. When possible, economic evaluations of the impacts of ozone are included.

At the 25<sup>th</sup> Task Force Meeting we will report on the achievements of the ICP Vegetation in 2011 [1] and progress made with items to be reported to the LRTAP Convention in 2012 [2], including:

- Supporting evidence for ozone impacts on vegetation, including the 2011 biomonitoring exercise;
- Ozone impacts on food security;
- Impacts of ozone on carbon sequestration;
- Impacts of black carbon on vegetation;
- Progress with European heavy metals and nitrogen in mosses survey 2010/11;
- Mosses as biomonitors of POPs;
- Relationship between (i) heavy metal and (ii) nitrogen concentrations in mosses and their impacts on ecosystems.

In addition, we will report on the contribution of ICP Vegetation to the common work plan items of the WGE for 2012 [2].

Apart from looking back to our achievements in 2011, throughout the Task Force Meeting we will be discussing our future plans, in particular the medium-term work plan of the ICP Vegetation (2012 – 2015).

### **Acknowledgement**

We thank the UK Department for Environment, Food and Rural Affairs (Defra) for funding the ICP Vegetation Programme Coordination Centre. Further financial support was provided by the UNECE and the UK Natural Environment Research Council (NERC).

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- [2] Draft 2012-2013 Workplan for the implementation of the Convention on Long-range Transboundary Air Pollution <http://www.unece.org/fileadmin/DAM/env/documents/2011/eb/eb/ECE.EB.AIR.GE.1.2011.10.e.pdf>

## AN OVERVIEW OF RESULTS FROM THE ICP VEGETATION OZONE BIOMONITORING WITH BEAN (2008 – 2011)

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Participants of the ICP Vegetation have used ozone sensitive (S156) and ozone-resistant (R123) genotypes of *Phaseolus vulgaris* (Bush bean, French Dwarf bean) to monitor ozone impacts in ambient air for the last four growing seasons. The genotypes were selected at the USDA-ARS Plant Science Unit field site near Raleigh, North Carolina, USA and kindly provided to the ICP Vegetation by Kent Burkey and colleagues. Plants were grown individually in 12-15 litre pots and exposed to ambient air for approximately 2-3 months (depending on the development of plants at individual sites). Some participants also exposed plants to ozone treatments using their own ozone exposure facilities such as open-top chambers and solardomes. Participants followed a common Protocol (modified slightly between years) and assessed plants for the occurrence of visible injury (all years) and yield (2009 onwards only). Many participants measured stomatal conductance of their plants in ambient air conditions at several stages of the growing season, to allow development of a stomatal flux model for bean based on over 3000 measurements.

Using hourly climate and ozone data, stomatal fluxes were calculated for the majority of experimental sites (hourly meteorological data was not available for all sites). Calculated fluxes ( $POD_0$ ) for the ambient air experiments ranged from 10.1  $mmol\ m^{-2}$  (Italy-Rome, 2008) to 18.9  $mmol\ m^{-2}$  (Austria-Seibersdorf, 2010), whereas 12h mean ozone ranged from 24 ppb (UK-Ascot, 2010) to 53 ppb (Spain-Villar, 2009). Unexpectedly high S/R ratios for pod weight and pod number (of pods > 4 cm) were found at many ambient air sites. These high ratios were not found in chamber experiments. Although there was generally a good fit of pod weight ratio to 12h mean ozone using the chamber experiments, and broad agreement with the previous USA studies, there was a poor fit using the ambient air experiments data. When using the whole dataset, there was a poor fit of pod weight ratio to ozone flux. Results will be presented using subsets of the data.

### **Acknowledgement**

We thank the UK Department for Environment, Food and Rural Affairs (Defra) for funding the ICP Vegetation Programme Coordination Centre. Further financial support was provided by the UNECE and the UK Natural Environment Research Council (NERC).

*PLEUROCHAETE SQUARROSA* (Brid.) Lindb.: AN ALTERNATIVE FOR HEAVY METALS AND NITROGEN MONITORING IN SOUTHERN EUROPE?

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Biomonitoring of element atmospheric deposition using terrestrial mosses is a well-established technique. Species as *Hylocomium splendens*, *Pleurozium schreberi*, *Scleropodium purum* and *Hypnum cupressiforme* have been widely used in the European surveys of heavy metal and nitrogen accumulation in mosses for more than 20 years. It has been normally assumed these species could cover the whole of the European territory. Nevertheless, different species distribution is found in southern and northern/central Europe. Thus, in the Mediterranean region, the aforementioned mosses are scarce and difficult to find whereas other species like *Pleurochaete squarrosa*, become frequent and abundant. The aim of this research was to compare the capability of two mosses species (*Hypnum cupressiforme* Hedw. and *Pleurochaete squarrosa* (Brid.) Lindb) to monitor the heavy metal and nitrogen atmospheric deposition and determine the suitability of *Pleurochaete squarrosa* to be used in biomonitoring surveys. To that end, twenty samples of both *Hypnum cupressiforme* Hedw. and *Pleurochaete squarrosa* (Brid.) Lindb. were collected in a Mediterranean region.

The mean elemental concentrations were of comparable magnitude in both species, although *Pleurochaete squarrosa* showed higher concentrations than *Hypnum cupressiforme* for all the analyzed elements except for Cd and Hg. The highest differences between both species were found for lithogenic elements (Al, Ti, V, Mn and Fe), probably due to the different places where the mosses grew. Thus, whereas the presence of *Hypnum cupressiforme* was reduced to shaded places under trees and bushes, *Pleurochaete squarrosa* grew in open areas receiving large quantities of re-suspended soil material. Both mosses showed similar spatial distributions for non-lithogenic heavy metals (As, Cd, Cu, Hg, Ni, Pb) and N in the studied area which suggested a similar capacity to monitor the atmospheric deposition. Lastly, Cd, Cu, Hg, Pb and Zn showed high enrichment factors in both species regarding the soil contents. This fact agreed with the results obtained in the PCA where those elements along with nitrogen were differentiated from the rest of elements showing an anthropogenic origin.



# OZONE IMPACTS ON CARBON SEQUESTRATION IN NORTHERN AND CENTRAL EUROPEAN FORESTS

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The ozone impacts on forest carbon sequestration were assessed for some northern and central European countries, i.e. Sweden, Finland, Norway, Denmark, Estonia, Latvia, Lithuania, Poland, Czech Republic and Germany. Since the most important increase in forest carbon stocks today is to the living biomass, the analysis focused on ozone impacts on the living biomass carbon stock changes. The analysis was based on UN-ECE statistics regarding forested areas, forest growth and harvest rates for the 2000-2005 period. This was combined with ozone exposure – response relationships for different forest types and age-classes in combination with nation-wide values for the AOT40 calculated from April – September.

The following conclusions were made:

1. The by far most important countries for carbon sequestration in the living biomass carbon stocks are Sweden, Finland, Poland and Germany.
2. The estimated annual increase in the living biomass carbon stocks under current ozone levels for the ten countries was 171 M t CO<sub>2</sub>e yr<sup>-1</sup>, while it was estimated to have been 190 M t CO<sub>2</sub>e yr<sup>s</sup> under pre-industrial ozone levels.
3. The difference caused by current ozone exposure on the annual living biomass carbon stock change was 19 M t CO<sub>2</sub>e yr<sup>-1</sup>, which was a reduction of 10%.
4. The magnitude of the predicted ozone effect for the different countries strongly depended on the gap between forest growth and harvest rates.
5. The knowledge regarding ozone impacts on mature trees under stand condition is to a large extent incomplete and further research is strongly needed.

## MOSS BIOMONITORING IN ALBANIA: PRESENT AND FUTURE

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The use of mosses as biomonitors of atmospheric deposition of heavy metals and radionuclides in Albania is used for the first time the moss from the year 2010, when our country for the first time joined the framework of UNECE ICP Vegetation programme, systematic studies. 32 moss samples were collected during the period September – October 2010 on the south part of Albania and in Tirana city, capital city of Albania. The University of Tirana, faculty of Natural Sciences, Dept. of Chemistry, Anal. Chem. Section was involved in sampling campaigns and the analysis of Hg via CVAAS method.

The ICP/AES analysis of 19 elements (**Al, B, Ba, Ca, Cd, Cr, Cu, Fe, Hg, Mg, Mn, Na, Ni, P, Pb-US, Sr, V and Zn**) was performed by the Institute of Chemistry, Faculty of Science, Sts. Cyril and Methodius University, Skopje, Republic of Macedonia. **The rest of elements (21 elements may fill the table of 40 elements**, like: **As, Au, Br, Ce, Cl, Co, Cs, Dy, Eu, Hf, I, In, La, Lu, Nd, Rb, Sb, S, Sc, Se, Sm, Ta, Tb, Ti, Th, W and Yb**), are in the process of analysis by Frank Laboratory of Neutron Physics, Joint Institute for Nuclear Research Dubna, Russian Federation.

The results are generally expressed as pollution gradients. The target elements in this study are heavy metals, as most toxic elements. For better interpretation of data, the elements Al, Ca, Fe, K, Na and P, as typically Earth crust elements, were also included.

The intensity of metal content based on the mean values in moss samples follows the trend Cd<Li<Pb<V<Cr<Cu<Zn<Ni<B<Sr<Mn<Ba<Na <P< Fe<Mg<Al<K<Ca. The range of variation of Cd, Li, V, Zn, B, Mn and Na in moss samples is larger than of other minor elements, while Al, Fe and Ca appear large range of variation, too.

After the normalization using Mg as natural component, and because it appear the smallest range of variation among Al, Fe, K and P, the intensity of metal content based on the mean values in moss samples follows the trend Cd<Li<Pb<V<Cr<Cu< **Ni < Zn** <B<Sr< **Ba < Na < Mn** <P< Fe<Al<K<Ca. The position of Ni, Zn, Ba, Na and Mn was changed, mean Zn and Mn may related to lithogenic and anthropogenic source. After normalization of the elements, the ranges of variation of Cd and Mn in moss samples remain to be the highest, compare to other elements.

The main sources of cadmium and manganese include the emission from the engine of old cars, the use of leaded petrol, waste incineration and industry. Intensive traffic in Tirana could be the main factor of high Pb content in about 47% of the analyzed moss samples. Proximity to the road and wear and tear of the automobile parts could be also the reason (Pearson et al. 2000) and the main reason of cadmium contamination.

To distinguish different geochemical mobility of elements and geochemically bounded elements, correlation analysis was carried out. Two groups of elements were found: **the first** group of elements with high correlation coefficients, belongs to lithogenic elements, such as Al, Ba, Ca, Cr, Fe, Li, Mn and Ni, as typically elements of Earth crust and of ultrabasic origin. The **second group** of elements, with high correlation coefficient between them like: Cd, Cu, Li, Mn and Cu, Ni, Zn, are mostly related to anthropogenic source.

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On the occasion of the tenth years of activity of the Catholic University of Brescia research group on ecophysiology and environmental physics, an overview of the main research themes developed will be given.

The actual research on ozone is mostly focused on 3 topics:

- 1) Characterization of gas exchange dynamics (O<sub>3</sub>, CO<sub>2</sub>, H<sub>2</sub>O) between vegetation (plant and ecosystems) and atmosphere.
- 2) Evaluation of the effects of ozone on vegetation and definition of flux-effect relationship.
- 3) Risk mapping at regional level.

Regarding gas exchanges dynamics, measurements of ozone, energy and water fluxes were performed with the eddy-covariance technique during several field campaigns. These measurements were conducted in different locations across Italy (most of them in the Po Plain) and over different vegetation ecosystems, including crops (barley, soybean, wheat, alfalfa) and forests (poplar, larch, holm oak). In this context, one of the main efforts of our group has been the partition of ozone fluxes between stomatal and non-stomatal components. As regards the second field of research, an experimental facility of Open-top chambers was built and managed with support and funding of Lombardy Region government, Regional Agency for Agriculture and Forests (ERSAF), and Lombardy Foundation for the Environment (FLA). This facility allowed us to conduct ozone experiments with controlled conditions (filtration or active fumigation). Several forest species (beech, oak, ash and poplar) and agricultural species (wheat, bean, tomato and lettuce) were investigated, while the latest experiments have been performed on Mediterranean macchia species (holm oak and strawberry tree).

The objectives these experiments were initially aimed at the characterization of the ozone visible leaf symptoms and at the evaluation of negative effects on plants' photosynthetic performance in relation to ozone dose. Later, the research was focused on the effects on productivity (yield and biomass losses) for the possible definition of dose-response relationship based on ozone fluxes.

In this context it should be noted the contribution of our group to the new proposal of ozone critical level for tomato.

In more recent years, the experimental work was devoted to the study of interaction effects between ozone and other abiotic stresses, such as drought and saline stress, in maquis ecosystems species and in durum wheat.

Concerning the risk mapping works, our research group was involved in an intense activity of ozone monitoring across the regional territory, with a mobile laboratory and with passive samplers, in order to provide and update geostatistical maps of the ozone exposure risk for ecosystems in different orographic contexts, including alpine valleys and natural reserves.

These skills have recently found a new development in the FP7 ECLAIRE project, in which the Catholic University of Brescia was asked to organize a monitoring campaign of gas exchanges over a lowlands forest in a natural reserve of the Po Plain (Bosco Fontana), and new OTC experiments on the evaluation of ozone effects on broadleaves species.

# THE ICP VEGETATION REPORT ON IMPACTS OF OZONE ON FOOD SECURITY, INCLUDING THE FIRST FLUX-BASED EUROPEAN CROP LOSS ASSESSMENT

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For the first time, ozone impacts on crop yield in Europe were quantified for wheat and tomato using the flux-based methodology. Applying the national emissions projections scenario for 2000, ozone pollution in EU27 (+ Norway and Switzerland) was predicted to be causing an average of 13.7% yield loss for wheat (an ozone sensitive crop), with an economic loss of €3.2 billion predicted if soil moisture is not limiting (Table 1). Economic losses per grid square in 2000 were greatest for wheat in the highest producing areas in France, Germany, Belgium, Denmark and the UK, indicating that ozone flux was high enough in these central and northern areas to have an impact on wheat production. For tomato (a moderately ozone sensitive crop) economic losses of €1.02 billion representing 9.4% of production value were estimated for 2000, with the highest total losses predicted for Italy, Spain, Greece and the Netherlands. Predicted effects for 2020 were generally lower than those in 2000 (Table 1). For both wheat and tomato, economic impacts were predicted to decrease by 38% to €1.96 billion and €0.63 billion respectively by 2020. However, for wheat, critical level exceedance remained high at 82.2% of EMEP grids for the wheat growing areas.

**Table 1.** Predicted impacts of ozone pollution on wheat and tomato yield and economic value, together with critical level exceedance in EU27+Switzerland+Norway in 2000 and 2020 under the current legislation scenario (NAT scenario). Analysis was conducted on a 50 x 50 km EMEP grid square using crop values in 2000 and an ozone stomatal flux-based risk assessment.

	Wheat		Tomato	
	2000	2020	2000	2020
Total production, million t	133.53		17.68	
Total economic value in 2000, billion Euro	15.87		6.85	
Mean % yield loss per grid square	13.7 <sup>1</sup>	9.07 <sup>1</sup>	9.4 <sup>2</sup>	5.7 <sup>2</sup>
Total production loss, million t	26.89	16.45	2.64	1.62
Total economic value loss, billion Euro	3.20	1.96	1.02	0.63
EMEP grids exceeding critical level (%)	84.8 <sup>1</sup>	82.2 <sup>1</sup>	77.8 <sup>2</sup>	51.3 <sup>2</sup>

<sup>1</sup> based on all grid squares with wheat production, <sup>2</sup> based on grid squares with > 1 tonne of production

Also included in the full report [1] are reviews of: i) ozone sensitivity in crop species using a concentration-based approach, ii) impacts on food and feed quality, iii) effects in a changing climate, iv) concerns for northern and southern Europe, v) case studies of national- and local-scale risk assessments and vi) impacts on food crops in South Asia.

## Acknowledgement

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## METAL CONTENT IN MOSSES CONTINUES TO DECLINE IN SWEDEN

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For the majority of the heavy metals included in the study 2010, concentrations in mosses have decreased since 2005.

The occurrence of most metals in moss samples follows a general gradient with the lowest levels in the mountains and inland parts of northern Sweden. The highest concentrations are generally found in the southern parts of Sweden. There are, however, for some metals local effects on the metal concentrations in mosses, e.g. in the ore fields and along the coastline in northern Sweden.

During the past 35 years, between 1975 and 2010, the heavy metal content in mosses in Sweden declined significantly with between 47% and almost 100%. The largest decrease of the metal concentrations was found for lead, followed by chromium, nickel, cadmium, vanadium, arsenic, copper and zinc. Since 1990, the corresponding statistical analysis was not as pronounced and only the concentration of lead and cadmium show a statistically significant decrease. However, for iron, copper, chromium, vanadium a statistically significant decrease in concentrations was found in some parts of Sweden between 1990 and 2010.

## OZONE RISK FOR VEGETATION IN THE FUTURE CLIMATE OF EUROPE BASED ON STOMATAL OZONE UPTAKE CALCULATIONS

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The negative impacts of surface ozone ( $O_3$ ) on vegetation are determined by external exposure, leaf gas exchange and plant antioxidant defence capacity, all dependent on climate and  $CO_2$  concentrations. In this study the influence of climate change on simulated stomatal  $O_3$  uptake of a generic crop and a generic deciduous tree at ten European sites, ranging from Northern Finland to Southern Spain, was investigated using the LRTAP Mapping Manual stomatal flux model.  $O_3$  concentrations were calculated by a chemistry transport model (MATCH) for three 30-year time-windows (1961-1990, 2021-2050, 2071-2100), with constant precursor emissions (year 2000) and meteorology from a regional climate model (RCA3). RCA model runs for the IPCC A2 and B2 scenarios were used. Comparing the AOT40 index (integrating the exceedance of the ozone concentration 40 ppb) with the POD (Phytotoxic Ozone Dose) resulted in very a different geographical distribution of estimated ozone risk for vegetation over Europe. Despite substantially increased modelled future  $O_3$  concentrations in central and southern Europe, the flux-based risk for  $O_3$  damage to vegetation was predicted to remain unchanged or decrease at most sites, mainly as a result of projected reductions in stomatal conductance under rising  $CO_2$  concentrations. The  $CO_2$  effect is uncertain, especially for trees, but will in principle promote a reduced stomatal uptake of ozone at a certain concentration of ozone. If the  $CO_2$  effect will turn out to become small, many areas in Europe will experience an increasing risk for vegetation damage. Drier conditions (higher vapour pressure deficit, lower soil moisture) in southern Europe, limiting stomatal ozone uptake, were also important, indicating that  $O_3$  risk for vegetation will not increase over the next century despite rising  $O_3$  concentrations. At northern latitudes, the current parameterisation of the stomatal conductance model suggest  $O_3$  uptake to be mainly limited by temperature. In this part of Europe an altered onset of the growing season may become very important for ozone effects on vegetation in the future.

This study demonstrates the importance of accounting for the influences by meteorological variables and rising  $CO_2$  on stomatal  $O_3$  uptake, and of developing their representation in models used for  $O_3$  risk assessment considering climate change.

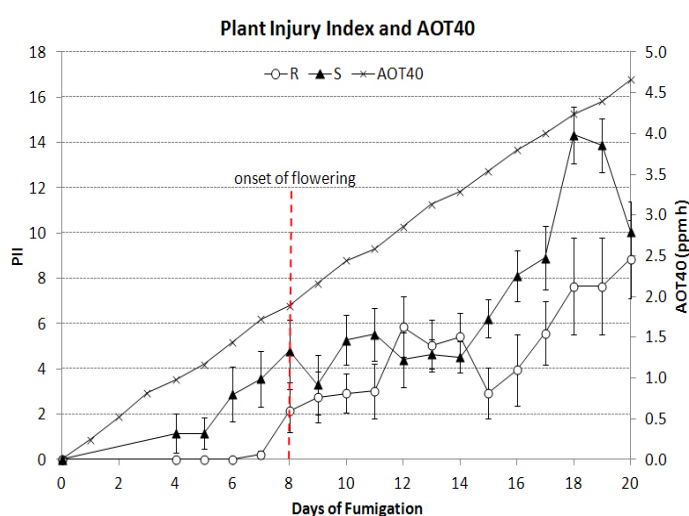
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Klingberg, J., Engardt, M., Uddling, J., Karlsson, P.E. & Pleijel, H. (2011). Ozone risk for vegetation in the future climate of Europe based on stomatal ozone uptake calculations. *Tellus* 63A, 174-187.

# A CHAMBER FUMIGATION STUDY TO EVALUATE THE PERFORMANCE OF THE SNAP BEAN BIOMONITORING SYSTEM UNDER SIMULATED MEDITERRANEAN CLIMATIC CONDITIONS.

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The O<sub>3</sub> biomonitoring system based on sensitive (S156) and resistant (R123) genotypes of snap bean (*Phaseolus vulgaris* L.) (Burkey et al. 2005), is being field-tested under the ICP Vegetation since 2008. Although OTCs studies have found clear relationships between O<sub>3</sub> exposure, physiological parameters, leaf injury, and yield of the S156/R123 system (Flowers et al., 2007), some biased results have emerged during field campaigns carried out in southern Europe. In a chamber fumigation experiment, we have tested the performance of the snap bean system under simulated Mediterranean microclimatic conditions. Sixteen seeds per genotype were grown in 12 l pots inside a walk-in chamber (mean day/night T: 28.2/24.0 °C; RH: 60%; leaf-level PAR: 350  $\mu\text{E m}^{-2}\text{s}^{-1}$ ; Photoperiod: 8:00-19:00 h; daily irrigation at field capacity); after the development of the 2<sup>nd</sup> trifoliate leaf, eight plants per genotype were transferred in a twin chamber under the same conditions, and fumigated 7 hours per day with 80 ppb of O<sub>3</sub> for 20 days, for a POD<sub>6</sub> of 4.7 mmol m<sup>-2</sup> and 3.0 mmol m<sup>-2</sup> for S and R, respectively. In the fumigated chamber, both genotypes showed visual O<sub>3</sub> symptoms (ICP Vegetation Experimental Protocol 2011). However, symptoms appeared earlier on the S plants, and were more severe at the end of the experiment than in the R plants (Fig. 1). Net photosynthesis (Pn) and stomatal conductance (gs) were significantly reduced in both cultivars (Pn: -43.4% and -54.1%; gs: -42.8% and -31.6% than the control for S and R, respectively), with no difference between S and R at the end of the fumigation. Interestingly, despite the Pn reduction, the total photosynthetic Performance Index (PI<sub>tot</sub>, Strasser et al., 2010) increased significantly in the fumigated S plants since the onset of flowering, suggesting the triggering of detoxification processes (Bussotti et al., 2011; Mereu et al., 2011). The final pod harvest showed that, although a significant O<sub>3</sub>-induced reduction of pod yield was evident in both genotypes, the S/R total pod weight ratio was lower than 1 both in the control and in the fumigated sets (0.63 and 0.57, respectively). In conclusion, our data suggests that, although prudence is needed in



**Fig. 1.** Daily evolution of Plant Injury Index (PII, Calatayud et al., 2007) and AOT40 during the experiment.

generalizing the results obtained in closed chambers, further studies are required to clarify the O<sub>3</sub> response of the S156/R123 system before its application in the field, particularly under Mediterranean climatic conditions.

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## SEASONAL ATMOSPHERIC METAL DATA BY MOSS ANALYSIS OF LAST 5 YEARS FROM INDIA

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Moss *Rhodobryum giganteum* (Schwaegr.) Par is being used to evaluate the intensity and trend of atmospheric depositions of Zn, Cu, Cd and Pb in the region of Uttarakhand hills (India) for a period of five years, 2006-2011. Before inducting moss for bio monitoring purpose, local commonly available mosses were validated for tolerance against metals by measuring their photosynthetic efficiency upon metal treatment using Photosynthetic Efficiency Analyser and tolerance species identified as *Rhodobryum giganteum* (Schwaegr.) Par, was inducted as same did not show change in efficiency (Fig 2) while same dropped down in *Pholia sp.* (Fig.1) and in other species *Grimmia sp.* The biomonitoring experiment was performed by transplanting moss bags prepared from fresh moss patches of *Rhodobryum giganteum* (Schwaegr.) Par.

The seasonal analysis (winter, summer and of rainy season) of transplanted moss *Rhodobryum giganteum* (Schwaegr.) Par. provided time integrated patterns of metal bioavailability including rural and urban metal deposition loads during five years as well as metal load in the proximity of traffic density areas. The findings also showed significant variations in metal deposition over the seasons i.e. high values of metals were recorded in moss exposed during summer (representing March to June) followed by winter (November to February) and low values in monsoon season (July to October). The metal deposition load was in the order of Zn>Pb>Cu>Cd during three subsequent years. Annual average percent increase in metals was 7%, 19%, 11% and 0.4% respectively in 2011 over 2006 in urban sites while an increase 12% for Zn, 11% for Pb, 14% for Cu, and 0.6% for Cd was observed in rural sites. The study area as a whole, shows an increasing trend each year as well in respective seasons and source could not be ruled out from increasing tourist activity as well as urbanization.

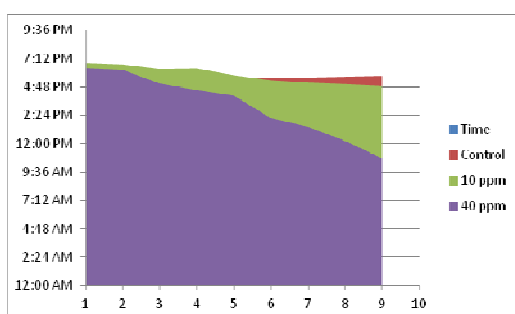


Figure:1 Moss *Pholia sp* showing decrease in photosynthetic efficiency upon metal treatment.

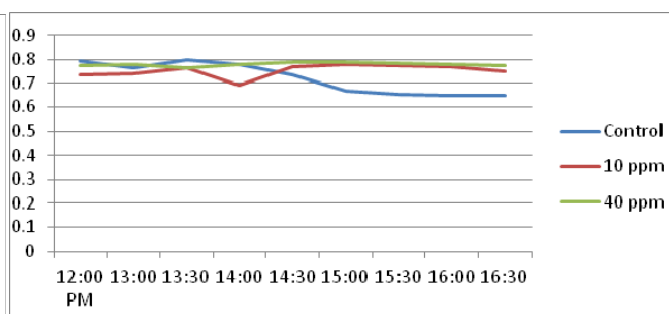


Figure:2 No change in photosynthetic efficiency upon metal treatment in *Rhodobryum giganteum* (Schwaegr.) Par



## MOSS BIOMONITORING IN CROATIA 2010

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Key words: air pollution, moss, biomonitoring, ICP Vegetation, Croatia

For the second consecutive time, Croatia participates in a moss survey in the framework of the International Cooperative Programme on Effects of Air Pollution on Natural Vegetation and Crops with heavy metals in Europe (UNECE ICP Vegetation) <http://icpvegetation.ceh.ac.uk>. Moss samples were collected during the summer/autumn of 2010 from 129 sites evenly distributed over the country.

Moss sampling was carried out according to the guidelines of the ICP Vegetation using a modified EMEP network. Areas with expected higher air pollution were covered with a denser sampling network. Samples were prepared for analytical measurements and will be analyzed using INAA, ICP-AES and AAS.

This study is undertaken in order to provide a reliable assessment of air quality throughout Croatia and to better characterize the pollution sources identified in the first moss survey in Croatia in 2006 [1]. Preliminary results on Al, Ba, Ca, Cr, Cu, Fe, K, Li, Mg, Mn, Na, Ni, Sr, Pb, Sr, V and Zn by using ICP-AES and As and Cd by ETAAS for Croatia moss 2010 survey will be presented.

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## MOSS BIOMONITORING OF ATMOSPHERIC POLLUTION WITH HEAVY METALS IN THE REPUBLIC OF MACEDONIA

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Some results from the third survey of atmospheric deposition of heavy metals and other trace elements over the territory of the Republic of Macedonia are reported. Samples of the terrestrial mosses *Homalothecium lutescens* and *Hypnum cupressiforme* were collected in August and September 2010 at 72 sites using standard procedures. Atomic emission spectrometry with inductively coupled plasma (ICP-AES) and atomic absorption spectrometry (AAS) allowed determination of the concentrations of 20 elements (Al, As, Ba, Ca, Cd, Cr, Cu, Fe, Hg, K, Li, Mg, Mn, Na, Ni, P, Pb, Sr, V and Zn). An overall comparison of data from three consecutive surveys in 2002, 2005 and 2010 is presented.

The results of factor analysis confirmed previously observed pollution sources characterized by emissions of Cd, Pb, Zn and Ni. Relatively high median value was found for Ni in 2005 and 2010, probably due to a renewed activity of a ferro-nickel smelter plant in the town of Kavadarci [1, 2]. GIS maps based on factor scores, and distribution maps for some elements will be shown.

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## THREE DECADES OF ATMOSPHERIC METAL DEPOSITION IN NORWAY AS EVIDENT FROM ANALYSIS OF MOSS SAMPLES

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Moss biomonitoring was used for the first time in 1976 (1) to study atmospheric deposition of metals in Norway, and nationwide surveys have been conducted since 1985 as a part of the national monitoring system. The main temporal and spatial trends apparent for key trace metals until 2005 are described in a recent paper (2). In the present talk these trends are briefly reviewed, and results from the 2010 moss survey are discussed in relation to the previous data.

In 2010 two additional studies employing sampling and analysis of *Hylocomium splendens* were carried out:

- A. Monitoring of metal deposition around 15 Norwegian industries, in most cases a repetition of corresponding studies in 2000 and/or 2005.
- B. Sampling of moss at 20 sites distributed over mainland Norway and analysis for POPs (PCB, HCB, HCH, DDT/DDE, PAH, brominated flame retardants, perfluorinated compounds).

Examples from these studies will also be presented and briefly discussed.

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RESPONSIVENESS OF SUBALPINE GRASSLAND PRODUCTIVITY UNDER  
INCREASED N AND O<sub>3</sub> DEPOSITION DETERMINED BY CARRY-OVER EFFECTS  
AND CLIMATE IMPACTS OF EXTREME YEARS

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Air pollution effects still threaten to alter natural and semi natural vegetation even in remote areas. The magnitude of ecosystem responses largely depends on interactions between pollutants and local environmental conditions.

In a seven year study we tested effects of increased N and O<sub>3</sub> deposition on subalpine grassland productivity and the interacting effects of climate on the response to the above pollutants. The fully factorial field experiment had five levels of N deposition (0, 5, 10, 20 und 50 kg ha<sup>-1</sup> a<sup>-1</sup>) and three levels of [O<sub>3</sub>] (ambient, amb.×1.4 and amb.×1.7) in a free air fumigation system. We hypothesized that aboveground net primary production (ANPP) is higher under N deposition and lower under O<sub>3</sub> deposition, with favorable climatic conditions increasing the magnitude of the response.

With N deposition ANPP increased up to 60% and even the lowest rate led to a significantly increased ANPP (+30%) in the seventh year. ANPP response had not yet reached a new steady state by the end of the trial. Thus, plant available N may still increase in all N treatments. But in contrast, even the highest level of O<sub>3</sub> enrichment had no effect on ANPP.

Independent of treatments, ANPP for individual years was best correlated with warmth in the beginning of the growth period (Growing Degree Days May; GDD<sub>May</sub>). The sum of precipitation May-July as a proxy for water availability can act as a co-limiting factor (2006), but may be overcompensated by favorable physical factors (2009). Surprisingly, growth response to N deposition was not correlated to GDD<sub>May</sub> or precipitation, but is positively correlated with previous year's yield. This implies, that large perennial plant parts, that serve first as a large nutrient source for early growth and later as a large assimilate sink, are prerequisite to avoid sink limitation in an N enhanced CO<sub>2</sub> assimilation system.

Thus, even N deposition rates at current critical loads, set to protect natural ecosystems, strongly increase subalpine grassland ANPP with unknown consequences for ecosystem services. Despite evidence for physiological damage to leaves, ANPP is very tolerant against high background O<sub>3</sub> concentrations. GDD<sub>May</sub> determine ANPP, but the responsiveness to N deposition depends on carry over effects of previous year's yields.

**Abstracts**

**Poster**

**Presentations**

# LARGE-SCALE ANALYSES OF GENE EXPRESSION AND METABOLITE LEVELS REVEAL THE CONTRASTING SENSITIVITY OF TWO WINTER WHEAT CULTIVARS EXPOSED TO OZONE UNDER FIELD CONDITIONS

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Two winter wheat cultivars (*Triticum aestivum* L., cv Premio and Soissons) were exposed to ozone in field conditions by means of a linear free-air ozone enrichment system at the INRA research station of Grignon (30 km west of Paris). The experimental set up was designed in such a way that plants located downwind were exposed to decreasing ozone levels as their distance from the ozone source increased. As ozone concentrations and water fluxes were monitored at the study site, the Phytotoxic Ozone Dose (POD) were calculated following the instructions given in the EMEP mapping manual. At the end of the experiment, after 2 months of ozone enrichment, flag-leaves of plants at 2m, 3.5m and 5m from the ozone source were exposed to POD<sub>6</sub> of 5.42, 4.49 and 4.03 mmol.m<sup>-2</sup>, respectively. Control plants, which were located 15m upwind from the ozone source, were subjected to a POD<sub>6</sub> of 3.5 mmol.m<sup>-2</sup>. Grain yield was negatively correlated to the POD<sub>6</sub> index for both cultivars. At 2m, grain yield was reduced by 12 % in Premio and 18 % in Soissons as compared to the control. For Premio, the linear relationship between POD<sub>6</sub> and grain yield agreed well with the impact function for wheat described in the EMEP mapping manual, but the older cultivar Soissons appeared to be more sensitive.

To unravel early responses to ozone, large-scale analyses of gene expression and metabolite levels were carried out on leaf samples harvested after 34 days of ozone fumigation, before the flowering stage, on control plants (POD<sub>6</sub> = 1.46 mmol.m<sup>-2</sup>) and on plants located at 2m (POD<sub>6</sub> = 3.26 mmol.m<sup>-2</sup>) and 3.5m (POD<sub>6</sub> = 2.23 mmol.m<sup>-2</sup>) from the ozone source. Overall, the ozone treatment hardly modified gene expression in Premio while the older cultivar Soissons showed a higher degree of deregulation that was correlated to the POD<sub>6</sub> index. Similarly, metabolite levels were modified only in response to the highest ozone exposure in Premio, whereas Soissons showed stronger responses that were related to the POD<sub>6</sub> index. The identification of differentially expressed genes in Soissons revealed that approximately 20 % of the down-regulated genes were transcription factors, notably ethylene-responsive ones, while 20 % of the up-regulated genes were linked to intracellular signal transduction and calcium homeostasis. These results emphasize the prominent role of cellular signaling processes in early responses to ozone. In addition, 6 % of the up-regulated genes were associated to plant-pathogens interactions, which underlines the similarity between early events triggered by ozone and pathogens. The identification of metabolites which abundance was affected by ozone showed features of the impact of the pollutant on plants, such as reduced levels of photosynthetic and plastid membrane lipids.

Large-scale analyses of gene expression and metabolite levels highlighted the differential responses to ozone of two wheat cultivars with contrasting sensitivity. Such analyses are helpful in dissecting molecular and cellular aspects of the impact of ozone on plants and in finding relevant indicators of early responses to ozone.

# SCREENING OF TEN BANGLADESHI WHEAT VARIETIES FOR THEIR SENSITIVITY TO OZONE

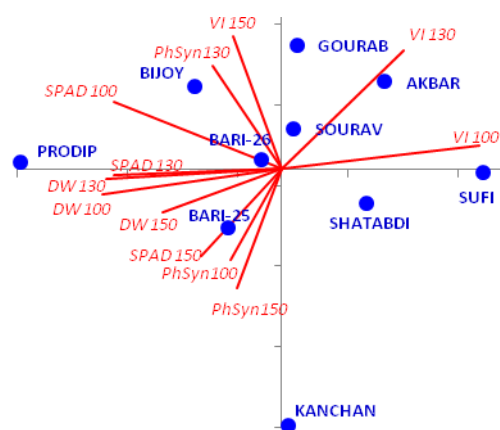
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**Introduction:** Tropospheric ozone is recognized as the most world spread phytotoxic pollutant; plenty of researches have demonstrated its negative effects on cultivated plants and natural vegetation. Although many research have been conducted to study the sensitivity to ozone of the cultivated varieties of main crops no such research has been conducted for any of the Bangladeshi main crops. Wheat constitutes a major crop and thus is of particular research interest. We study the relative sensitivity to ozone of the following ten Bangladeshi wheat varieties: Akbar, Bijoy, Gourab, Kanchan, Prodip, Shatabdi, Sourav, Sufi, Bari-25, Bari-26. **Method:** The experiments were conducted in controlled environment chambers. In three experiments, plants of the tested varieties were exposed to ozone levels of 100, 130 and 150 ppb. In each experiment, for each pot with plants exposed to ozone one pot with exactly the same number of plants was maintained in ozone-free chamber; this allowed a pair wise testing. Data of the following parameters were collected: visible leaf injury (VI), dry weight (DW), chlorophyll content evaluated by greenness (SPAD) and quantum yield of photosynthesis (PhSyn). For each parameter (except visible injury), for each variety, the ratio between O<sub>3</sub>-exposed and control plants was analyzed (the lower the ratio the higher the ozone effect). Before analysis data were tested for ANOVA assumption and, when necessary, were submitted to Box-Cox transformation.

**Results:** Both “ozone level” ( $p < 0.001$ ) and “variety” ( $p < 0.001$ ) factors affected the measured parameters; the interaction between the two factors was also statistically significant ( $p < 0.001$ ). The overall performance of the varieties for each variable in each experiment is summarily depicted in the PCA-biplot shown below (Fig. 1).

Fig. 1. PCA-Biplot, after Varimax rotation; the lines stand for the measured variables (VI, DW, SPAD, PhSyn) at each ozone level (100, 130, 150 ppb); the dots stand for the tested wheat varieties.



The first axis (X) explains 35.27 % of the observed variability while the second axis (Y) explains 25.57%. Kanchan showed the lowest visible injury at 130 and 150 ppb and the highest photosynthesis ratio at 100 and 150 ppb. Sufi showed the highest visible injury at 100 and 130 ppb. Prodip showed high ratios in SPAD at 100 and 130 ppb, and in dry weight at 100, 130 and 150 ppb, and the smallest visible injury at 100 and 130 ppb. Gourab and Akbar showed high visible injury at 100, 130 and 150 ppb.

**Conclusion:** This first pilot screening of the ten Bangladeshi wheat varieties showed that most of them are relatively sensitive or of intermediate sensitivity to ozone. Comparatively more resistant (less sensitive) seem to be Prodip and Kanchan while the relatively more sensitive seem to be Sufi, Gourab and Akbar. The screening is being continued by exposures to even lower ozone levels that hopefully discriminate the varieties further.

# TRENDS IN N QUANTITY AND $\delta^{15}\text{N}$ SIGNATURES OF DIFFERENT N DEPOSITION TYPES IN GERMANY INDICATED BY LICHENS AND BARK SAMPLES

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The assessment of air quality is an important topic. Nowadays the intensification of agriculture is responsible for a huge input of nitrogen (N) into ecosystems. Epiphytic lichens have a set of characteristics that make them well qualified for biomonitoring purposes.

In recent years, besides research of nitrogen concentrations in lichen thalli, research on isotopes in lichens increased. A lot of studies proved that it is possible to use isotope signatures for estimating different nitrogenous sources.

In the present study we tested whether epiphytic lichens and the respective bark of their carrier trees are useful indicators for reflecting nitrogen affected areas and detecting different kind of nitrogenous deposition by dint of nitrogen concentrations and  $\delta^{15}\text{N}$  signatures in lichen tissues and bark samples.

In winter 2008/09 epiphytic lichens (*Hypogymnia physodes* (L.) Nyl. and *Xanthoria parietina* (L.) Th. Fr.) were sampled from 18 sites located in Germany (permanent grid established by Federal Environment Agency). Samples were collected within a radius of 2 km around field stations for deposition measurement, from trees that met the requirements for bioindication with lichens. Besides, bark samples were collected at the same sites and furthermore characteristics of carrier trees (e.g. circumference in 1.50 m height, tree height, crown diameter, characteristics of bark and N related concentrations) and other parameters (e.g. land use, altitude and traffic influence) were registered.

As a result of being present and absent, lichens have the ability to reflect deposition circumstances at different sites. The sensitivity of epiphytic lichens is also shown in this study. Upon investigation, we also used the function of lichens as bio-accumulators. We found ranges between high N concentrations in *Xanthoria parietina* under high N deposition and lower N concentrations in *Hypogymnia physodes* in unpolluted areas. As expected, the  $\delta^{15}\text{N}$  ratios are in both lichen species more negative under high ammonia deposition, where N deposition is from agricultural origin, especially from animal husbandry. Bark samples could reflect a similar trend in highly agricultural areas.

In this study we could underline that mainly lichens can be used for monitoring atmospherical characteristics and that their ability of accumulation shows nitrogenous situation at different nitrogen affected areas. By comparing different  $\delta^{15}\text{N}$  signatures in lichens we could highlight that source attribution is possible.



# LEAF STOMATAL CONDUCTANCE AND PHOTOSYNTHESIS OF FIELD-GROWN MAIZE EXPOSED TO CHANGING OZONE CONCENTRATIONS

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The impacts of moderate ozone levels on leaf conductance and photosynthesis of maize were assessed from field fumigation experiments carried out at Grignon (France) in July-August 2011. The exposition of the plants to ozone was achieved by a linear fumigation device. Continuous measurements of ozone concentration were taken inside the vegetation, close to the ears. The mean daylight ozone levels ranged from 31 ppb (control) to 59 ppb.

Although photosynthesis was significantly affected by long-term ozone exposure, stomatal conductance appeared to be moderately sensitive to this pollutant. Furthermore, grain yields were very slightly impacted. A Jarvis-type model of stomatal conductance was used in order to establish an empirical relationship between stomatal uptake of ozone and photosynthetic capacity.

## APPLICATION OF THE LOIBL FUNCTION IN ITALY: CAN WE CALCULATE AOT40 FROM PASSIVE MONITORS?

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The AOT40 index is the actual European standard to assess when and where ozone (O<sub>3</sub>) concentrations exert a potential risk for vegetation. It can be calculated by using O<sub>3</sub> data coming from either automatic devices or passive samplers, offering thus practical advantages. The Loibl function describes the O<sub>3</sub> daily profile as a function of site elevation, relative altitude and day-time. The use of passive samplers and the Loibl function allow to calculate the AOT40 index everywhere. The question is: is the Loibl function working well in complex terrains as in most of the Italian country? Previous studies have been carried out to answer this question. In this work 89 monitoring stations (32 rural and 57 suburban) were analysed in Italy for the year 2005. AOT40 was calculated from hourly O<sub>3</sub> data measured by passive monitors, then, weekly concentrations were calculated in order to apply the Loibl function. In order to analyse the effects of other parameters that can be important for O<sub>3</sub> concentrations, distance from the closest city, distance from the sea and minimum, maximum, and mean elevation around a 5 km buffer were calculated for each station by GIS. A model describing the O<sub>3</sub> effect on vegetation was developed by using a non-linear statistical method, based on the Generalised Linear/non-Linear Model (GLZ), with the main aim of understanding which among environmental parameters mentioned above are most important for O<sub>3</sub> concentrations. GLZ model is built taking into consideration either linear linkages or non-linear ones, combining predictors and enhancing thus the predictive capacity of the model. Residual between AOT40 estimated by the Loibl function and AOT40 measured by passive devices were high for each site, and in some cases the difference reached -241%. The GLZ model highlights that elevation played a relevant role in affecting the AOT40. However, elevation does not act singly but only combined with others parameters. Elevation (and correlated parameters) is an important predictor but it has not the same “weight” for different station type and locations. As a consequence, AOT40 can be predicted in an effective way by using a non-linear approach such as the GLZ models, although this approach should be better validated by using cross-validations performed on climatically different years.

OZONE -INDUCED ACTIVE OXYGEN SPECIES IN BEAN (*Phaseolus vulgaris*)  
CULTIVARS EXHIBITING DIFFERENTIAL O<sub>3</sub> SENSITIVITY

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In recent times, the assessment of possible harmful effects of atmospheric O<sub>3</sub> on vegetation has been greatly improved by comparisons between O<sub>3</sub>-sensitive and O<sub>3</sub>-tolerant genotypes or selections of the same species. The present study was initiated as an attempt to screen commonly cultivated Egyptian bean genotypes for sensitivity to O<sub>3</sub>. Our aim was to identify the initial molecular events involved in conferring O<sub>3</sub> sensitivity and tolerance. Four Egyptian genotypes were selected for this purpose (Nebraska, Paulista, Bronco and Contender). Tests for O<sub>3</sub>-sensitivity were conducted using short-term acute exposure to O<sub>3</sub> at 200-250 ppb for 3 days (6h/d). Screening tests yielded one sensitive genotype: Nebraska, two intermediate candidates: Paulista and Bronco and one tolerant: Contender, when ranked on the basis of % foliar injury to the 1<sup>st</sup> trifoliolate leaves. Solute leakage from leaf discs of control and O<sub>3</sub>-stressed plants was used to study the extent of membrane damage sustained by plants (4 and 5 weeks old) as a result of ozone exposure (100-150ppbv and 200-250ppbv). Membrane leakiness was proportional to the level of O<sub>3</sub> used and correlated positively with the magnitude of cell damage and lesion development on the bean genotypes under study. Histochemical staining experiments were carried out immediately after O<sub>3</sub> fumigation to elucidate the accumulation sites of the major active oxygen species (AOS) mainly superoxide anions (O<sub>2</sub><sup>•-</sup>) and hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>). AOS were found to accumulate in O<sub>3</sub>-sensitive but not in O<sub>3</sub>-tolerant genotypes. In all the genotypes tested, O<sub>2</sub><sup>•-</sup> were the predominant AOS accumulated in response to O<sub>3</sub>. O<sub>2</sub><sup>•-</sup> anions reflected the shape, extent and distribution of necrotic lesions and their production intensified with the increase in ozone dose. Sites of H<sub>2</sub>O<sub>2</sub> production were significantly fewer than those of O<sub>2</sub><sup>•-</sup> and did not reflect the respective genotype's O<sub>3</sub> sensitivity. Net photosynthetic CO<sub>2</sub> assimilation (Pn) was followed throughout the experiment. A dramatic decrease in Pn was recorded on the more sensitive genotypes (Nebraska and Paulista) immediately after the first O<sub>3</sub> fumigation. The more tolerant genotypes (Bronco and Contender) showed an initial resistance to O<sub>3</sub> after the first fumigation episode, followed by a significant Pn reduction. None of the genotypes studied showed recovery of Pn in the absence of O<sub>3</sub> between fumigation sessions. This ruled out the possibility that the decrease in Pn was due to stomatal closure as a defense mechanism against O<sub>3</sub>. We concluded that the sensitivity of bean genotypes in this study resulted from the absence or ineffectiveness of an antioxidative defense mechanism, hence the burst of AOS which could be readily correlated to the relative level of genotype sensitivity.

HEAVY METAL AND NITROGEN DEPOSITION IN POLAND – MOSS SURVEY  
2010/2011

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Within the framework of the 2010 European moss survey nearly 300 sites evenly distributed across Poland were investigated in terms of trace metals and nitrogen accumulation in mosses. The sites were established at the same locations as in 1995. In 2010, average concentrations of cadmium, copper, nickel and lead in mosses were found significantly lower as compared to the values obtained in 1995 ( $p < 0.0001$ ). Median values decreased from 0.46 to 0.30 (Cd), from 7.57 to 5.93 (Cu), from 1.44 to 1.16 (Ni) and from 13.61 to 4.79 (Pb)  $\text{mg kg}^{-1}$  between the years. The trend was dependent on the region; for some provinces decreases were insignificant. Average concentrations of other metals – iron and zinc – did not alter in time ( $p > 0.05$ ). In 2010, median values of these metals were 339 and 46.7  $\text{mg kg}^{-1}$ , respectively. The spatial patterns of the accumulation of trace metals in mosses were similar in both surveys; southern parts of Poland, industrialized and densely populated, are contaminated more than the rest of the country. Nitrogen concentrations in mosses, estimated for the first time for Poland during the 2010 moss survey, ranged from 0.78 to 2.86 % (the average value was 1.56%). The western part of Poland (having more intensive agriculture and transport) seems to be influenced by N deposition to a greater extent than the eastern one. However, significant differences between provinces were not found due to high local variation.

## OZONE INDUCED CHANGES IN RUBISCO AND RUBISCO ACTIVASE OVER LETTUCE LIFE-SPAN

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The present study examined the effects of ozone on the photosynthetic capacity of two ‘Cos’ lettuce (*Lactuca sativa* L.) varieties grown widely in Greece. The role of Rubisco and Rubisco activase in mediating the O<sub>3</sub>-enhanced decline in carboxylation efficiency was explored. The transcript and protein abundances of Rubisco activase plus Rubisco large and small subunits were monitored over the leaf life-span in plants subject to ozone levels similar to those experienced regularly during the spring and summer time in Southern Europe (100 ppb ozone 8 h d<sup>-1</sup>). Ozone exposure led to a decline in net CO<sub>2</sub> fixation (i.e. a decline in the maximum light- and CO<sub>2</sub>-saturated rate of assimilation, i.e.  $A_{max}$ ), a reduction in carboxylation efficiency ( $V_{cmax}$ ) and a decline in the maximum modelled capacity for RuBP regeneration ( $J_{max}$ ).

The decline in carboxylation efficiency induced by ozone was linked to a decrease in the amount of Rubisco LSU and SSU protein, an effect resulting—at least in part—from a decrease in the transcript abundance of *rbcS1* and to a lesser extent of *rbcL*. The work outlined in this paper shows that members of the *rbcS* gene family respond differentially to ozone-induced oxidative stress. Exposure to ozone also resulted in a reduction in the amount of Rubisco activase isoforms and this was accompanied by a decline in the transcript abundance of *rca* genes.

Our findings are consistent with the view that ozone exposure triggers a general decline in the expression of photosynthetic genes. It would be interesting to ascertain whether, and at what stage in relation to leaf phenology, this situation is reversible in ozone-treated leaves.

STOMATAL CONDUCTANCE MODELING OF *ZELKOVA SERRATA*  
TO ESTIMATE STOMATAL OZONE UPTAKE IN JAPAN

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The stomatal flux-based approach for assessment of ozone damage to plants was hardly developed in East Asia. To parameterize stomatal conductance or ozone uptake modeling in the Eastern Asian species *Zelkova serrata*, measurements of stomatal conductance were carried out in several Japanese sites across the growing season. The new parameterization improved the stomatal conductance model performance relative to a previously proposed model. The results were used to compare the spatial distribution of AOT40 and *Z. serrata* stomatal ozone uptake in Japan. Although annual precipitations are usually elevated in Japan, including a soil moisture function improved the model in the short periods with low precipitation. In addition, elevated vapor pressure deficit and soil moisture deficit due to high temperature decoupled stomatal ozone uptake from high ozone levels. Therefore, the use of a flux-based approach, rather than AOT40, is recommended for forest protection in Japan.

## MOSS BAG BIOMONITORING IN ALBANIA

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The use of mosses as biomonitors of atmospheric deposition of heavy metals and radionuclides in Albania is used for the first time the moss from the year 2010, when our country for the first time joined the framework of UNECE ICP Vegetation programme, systematic studies. 32 moss samples was collected during the period September – October 2010 on the south part of Albania and in Tirana city, capital city of Albania. The University of Tirana, faculty of Natural Sciences, Dept. of Chemistry, Anal. Chem. Section was involved in sampling campaigns and the analysis of Hg via CVAAS method.

Active biomonitoring of the air quality was performed using the moss *Hypnum cupressiforme* within and around Tirana and Vlora urban areas, Albania. Moss bags were exposed with irrigation for 6 months, respectively, at 7 sites of Tirana city and 9 sites of Vlora City along the main streets.

Heavy metals, such as Cu, Pb, Zn, Mn, Fe and Cd were determined by AAS using flame/and or electro thermal system. CVAAS was used for mercury determination and AES for K and Na determination. The results are generally expressed as pollution gradients.

The target elements in this study are Cd, Hg, Cu, Pb, Zn and Mn. For better interpretation of data, the elements Fe, K and Na were also included. The cities are being moderately polluted due to the high vehicular emissions, use of adulterated fuel in vehicles. Therefore, we have been tried to categorize different places in the city on the basis of mentioned metal concentrations in the mosses and data statistical treatment. Mercury shows most sounding results (13.39 µg/g, DW in ST\_V9), near Vlora Hot Spot site, polluted by metallic mercury. The comparison with unexposed moss allowed us assessing the enrichments factors in exposed moss samples for all determined elements. To distinguish different geochemical mobility of elements and geochemically bounded elements, correlation analysis was carried out. Two groups of elements was found: the first one (Zn, Fe, Cu, K and Na) do not display high accumulation factors (mean AF<3), while Pb and Cd in Tirana sites, as well as Hg and Mn in Vlora sites, display moderate accumulation factors (mean AF >3).

## FIELD STUDY OF THE IMPACT OF OZONE ON MAIZE YIELD USING A NEW FACE SYSTEM

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Ozone (O<sub>3</sub>) is the air pollutant that currently represents one of the greatest environmental concerns, with particles and nitrogen oxides. Ground levels of O<sub>3</sub> have increased continuously over the past three decades at a rate of 0.5-2% per year (Vingarzan, 2004). O<sub>3</sub> in the troposphere is responsible for negative effects on crop production. However the responses to O<sub>3</sub> vary between plant species due to variable stomatal conductances and differences in the ability to detoxify O<sub>3</sub>-derived reactive oxygen species. Regarding their sensitivity to O<sub>3</sub>, crop species are usually divided into three groups: O<sub>3</sub> sensitive crops; moderately sensitive crops and O<sub>3</sub> resistant crops (Mills et al., 2007). Maize (*Zea mays* L.) is generally considered a moderately sensitive crop, even though information on the sensitivity of this species in the field is scarce. Here, we present the results of a field ozone fumigation experiment on maize.

Fumigation systems, such as open top chambers (OTC) or field fumigation rings (FACE), were developed to obtain experimental data on crop yield losses. In the present experiment, we used a linear fumigation device derived from FACE systems. It consists in two linear stainless fumigation cylinders oriented North-South. Under daylight conditions, when dominant winds blew (westward), the system continuously generated O<sub>3</sub> and the prevailing winds acted as natural dispersion vectors, without disturbing the other environmental and agricultural conditions. The generator was automatically turned off during the night and when the wind direction was different from that prevailing wind. The O<sub>3</sub> concentrations were slightly higher than natural levels near the fumigation source and decreased regularly with the distance to the source. Plants were exposed to O<sub>3</sub> enriched air daily for 65 days (30/06 to 3/09 2011). O<sub>3</sub> exposure at different distances from the fumigation source (2 m, 3.5 m and 5 m downwind of the source compared to control plants located 15 m upwind of the source) was calculated (i) as an hourly average concentrations and (ii) as the cumulative concentration over a threshold of 40ppb (AOT40). The yield was obtained through destructive measurements of biomass.

Table: O<sub>3</sub> concentrations and AOT40 values measured at different distances from the fumigation line (measurements were taken inside the canopy)

Distance from the line	2m	3.5m	5m	Control
mean [O <sub>3</sub> ] ppb	53	42	39	31
AOT40 (ppm h)	19.6	10.8	8.5	3.1

The results showed that the device was capable of providing realistic ozone levels. The relative yield loss (y) induced by this long-term exposition to ozone (x = AOT40) was very low:  $y = -0.0017 (\pm 0.0045)x + 1$ ,  $R^2=0.7788$ . This relationship was not significantly different from previous data (Mills *et al.*, 2007). These results confirmed that maize is less sensitive than wheat to ozone (Mills *et al.*, 2011). The use of a “level II” exposure index (POD) appears to be a good tool to analyse stomatal and non-stomatal sensitivity of these two species.

*Mills et al., 2007. Atmospheric Environment 41, 2630– 2643*

*Mills et al., 2011. Global Change Biology (2011) 17, 592–613*



## CONCENTRATION OF ELEMENTS IN TEETH OF ROE DEER (*Capreolus capreolus* L.) AND MOSSES IN SLOVAK INDUSTRIAL POLLUTED SITES

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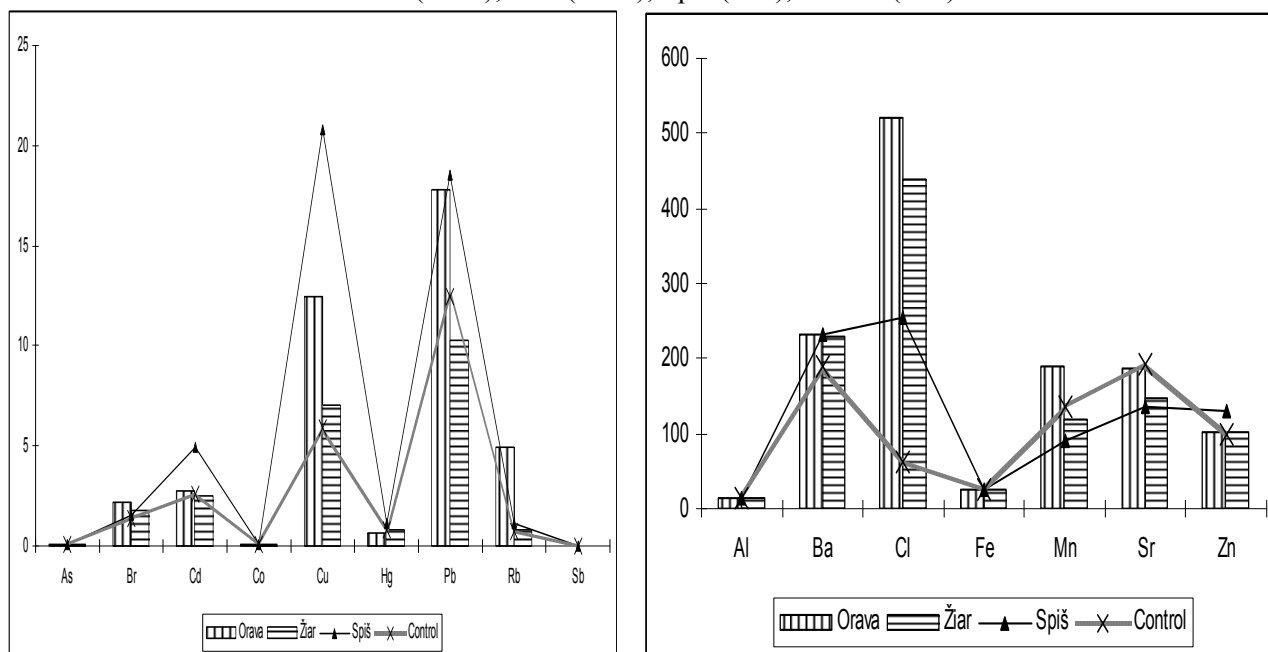
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Element analysis in roe deer teeth from the 3 polluted sites and control locality NAPANT are discussed in a context of their values. We found the highest concentration in the roe deer teeth: As, Ba, Br, Cl and Na in Žiar nad Hronom (aluminium plant); Cd, Cu, Hg and Zn in area Spiš (iron ore and mercury plant); Al, Ca, Co, Fe, Mg, Mn, Pb, Rb, Sb and Sr in Orava (smelter complex from Czech republic). There is statistically significant difference between the concentration of of As, Cd, Co, Cu, Hg, Na, Pb, Rb, Sr and Zn in roe deer teeth from the sites Orava, Žiar, Spiš and control locality NAPANT. Results are compared with European bryophyte analyses. The obtained data can be useful as a reference level for comparison with the future measurements of air pollution in the examined area, whenever a hazard due to accumulation of heavy metals along the food chain is assessed.

**Keywords:** Roe deer teeth; Heavy metals; Bioindication

Concentration of elements in teeth of roe deer in mg.kg<sup>-1</sup>  
Orava (n=27); Žiar (n=16); Spiš (n=5); control (n=9)



DEPENDENCE OF EXTENT OF OZONE INJURIES ON *TRIFOLIUM REPENS*  
FROM CLIMATIC FACTORS IN LATVIA

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The increase of ground level ozone concentration in the atmosphere is one of the most important environmental problems nowadays. Within *ICP- Vegetation* framework, investigations of interaction effects of ground level ozone and meteorological factors on white clover *Trifolium repens* L cv Regal were performed at five meteorological stations (Rūjiena, Zosēni, Dobele, Mērsrags and Rucava) located in rural areas of different climatic regions of Latvia.

Two batches of cuttings of ozone-sensitive (NC-S biotype) and ozone-resistant (NC-R biotype) white clover (*Trifolium repens* cv Regal) clones were received from the Coordination Centre of the ICP Vegetation at the Centre for Ecology and Hydrology, Bangor (UK), at an interval of 14 days between the batches. Plants were grown according to the ICP-Vegetation protocol. Two clones of white clover - an ozone sensitive (S) and ozone resistant (R) were grown in specially prepared pots in the territories of meteorological stations from 27 June to 3 October 2006. The leaves of S clones were inspected for ozone damage once a week.

28 days after transplantation the plants were cut back to 7cm above the soil level. The ratio of dry biomass S/R was calculated by cutting the clover each month. Visible injury occurred on the sensitive clone at each of the experimental sites and was present on up to 10% of the leaves in pot. Results of our investigations demonstrated that leaf injuries were strongly dependent from weather conditions. Multiple regression analysis of field experimental data showed that pure ozone effects explained only 13-16% of the data variation, while the relative air humidity explained 23-25% and sums of positive temperatures – 3-6% of the data variation. We also were able to get some preliminary insight to which extent regional climatic peculiarities may affect the plant response to the ground ozone because earlier studies have pointed on the possibility of such effects on European scale. Clover leaf injuries increased from NE to SW and positively correlated ( $R= 0.954$ ,  $P<0.05$ ) with mean seasonal relative air humidity calculated for different meteorological stations.

## TROPOSPHERIC OZONE BIOMONITORING WITH SNAP BEAN PLANTS IN RALEIGH, NORTH CAROLINA, USA

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**Introduction:** Ozone-sensitive (S156) and ozone-tolerant (R123) genotypes of snap bean (*Phaseolus vulgaris* L.) are being developed as a biomonitoring system for the evaluation of ambient ozone phytotoxicity potential. Foliar injury and yield ratios are considered good indices of ambient ozone effects on plants. This system has been adapted by many research teams involved in the ICP-vegetation Programme. We utilized this system in order to: a) evaluate the ambient ozone phytotoxicity potential in Raleigh, North Carolina; and, b) validate its creditability. **Method:** Eight plants per genotype were used according to the experimental procedure suggested by ICP-vegetation Programme. The experiment began (seeds sowing) on June 8, 2011 and lasted (up to harvest) about 11 weeks. Data of the following parameters were collected: visible leaf injury, yield, stomatal conductance and hourly mixing ratios (in ppb) of ambient ozone. Midday stomatal conductance (adaxial + abaxial) was measured using a steady-state porometer (Li-Cor Model 1600). **Results:** The AOT40 value over the 9 week growing period was 8 ppm h. S156 plants exhibited gradually increasing visible leaf injury. The percentages of leaves which exhibited injury between a) 5-25% and b) above 25%, at the end of the first week was 14% and 5% respectively while in the week before the harvest they were 7% and 59% respectively. Concerning R123, only a few symptoms were recorded on July 7 (5% and 7% respectively). These symptoms were attributed to the highest ozone levels monitored during the previous days (the highest daily AOT40 (415 ppb\*h) was monitored on July 1<sup>st</sup>). Otherwise, foliar injury in R123 was less than 5%. Stomatal conductance during vegetative growth and anthesis in S156 and R123 averaged 940 and 1013 mmol H<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup>, respectively. During pod-fill, conductance decreased to 340 mmol H<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup> in both genotypes, but diurnal measurements indicated that conductance was higher in S156 than in R123 by late July when S156 was severely injured. The differences in yield parameters between the two genotypes are shown in Table 1.

*Table 1. The comparison of the S156 and R123 snap bean genotype yield parameters. Asterisks indicate statistically significant differences between the two genotypes, at a level of significance  $\alpha=0.05$ , after t-test.*

Parameters	S156	R123	S156/R123 p (value)	
Number of pods > 4cm with seeds	61 ± 6	49 ± 4	1.24	0.06*
Dry wt (g) of pods > 4cm with seeds	34 ± 4	28 ± 3	1.21	0.12
Number of pods > 4cm with no seeds	5 ± 1	46 ± 10	0.11	0.003*
Dry wt (g) of pods > 4cm with no seeds	0.3 ± 0.1	3.2 ± 1	0.08	0.002*

**Conclusion:** Results showed that ambient ozone levels in Raleigh, North Carolina caused extensive visible leaf injury in S156 but not in R123. Differences in sensitivity did not appear related to ozone uptake as midday stomatal conductances were similar between genotypes, except during late pod-fill in injured S156 leaves. Loss of stomatal control likely occurred due to ozone injury in these plants. Pod dry weights were not significantly different between genotypes at time of harvest. The number of unfilled pods in R123, however, was much greater than in S156. Seed-fill was slower in R123 than in S156, possibly due to genotype differences in development or sensitivity to environmental factors such as temperature or water stress. Differential responses of these snap bean lines to ozone appears related to genetic differences in ozone sensitivity as evidenced by foliar injury. Differences in yield between genotypes may require longer experimental duration to become apparent.

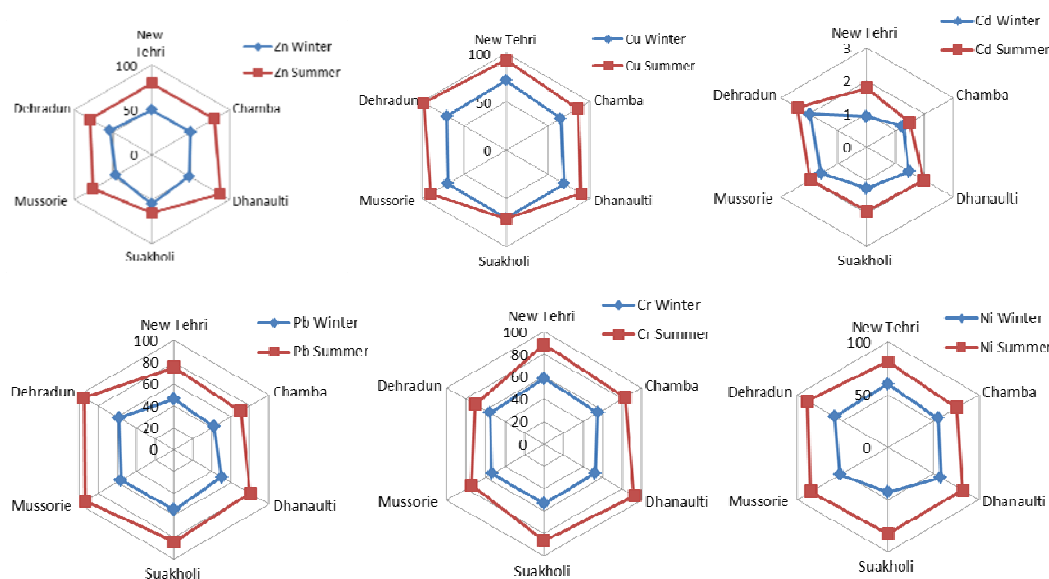
SEASONAL MONITORING OF ATMOSPHERIC METALS BY MOSS *HYPNUM CUPRESSIFORMAE* FROM SURROUNDING OF MUSSORIE TOURIST PLACE GARHWAL OF UTTARAKHAND, INDIA

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There is lack of sufficiently sensitive and inexpensive techniques with protocol that permit the simultaneous measurements of metals from atmosphere. Biomonitoring is a good option and amongst biological organism and moss are the best inexpensive choices to quantify the atmospheric metal load. Contrary to vascular plants mosses have no cuticle and epidermal layer differentiation and also have no organs for the uptake of mineral from the soil as a result contact between the moss and its substrate is mostly poorly developed. Mosses absorb minerals and moisture by their whole surface therefore are considered an ideal and are recommended for metal monitoring. Another advantage is their wide distribution and no care is required after they are exposed for an active monitoring.

Bioaccumulation ability for metals was evaluated seasonally, exhibiting maximum in summer followed by winter. Analysed moss exhibited high values for Zn ( $55.26 \mu\text{g gm}^{-1}$ ) from Suakholi during winters and same was measured highest in summer ( $88.47 \mu\text{g gm}^{-1}$ ) from Dhanaulti. These were minimum ( $46.25$  and  $65.36 \mu\text{g gm}^{-1}$ ) from Mussorie and Suakholi. On the contrary, Pb and Cd values were  $95.23$  and  $2.41 \mu\text{g gm}^{-1}$  in moss harvested from Dehradun during summers and their winters values were measured as  $1.98$  and  $58.26 \mu\text{g gm}^{-1}$ . Maximum values for Cu and Ni during summer was from Dehradun ( $98.25$  and  $88.25 \mu\text{g gm}^{-1}$ ) and for winter same was  $72.25$  and  $60.23 \mu\text{g gm}^{-1}$  from New Tehri. Minimum values were measured from Chamba ( $65.26 \mu\text{g gm}^{-1}$ ) and Suakholi ( $43.02 \mu\text{g gm}^{-1}$ ) during winters while same during summer was  $75.25 \mu\text{g gm}^{-1}$  again from Chamba. The present study allows us to determine the extent of the area affected by metal precipitation load in order of  $\text{Cu} > \text{Ni} > \text{Pb} > \text{Cr} > \text{Zn} > \text{Cd}$ .



Seasonal concentration of metals in various sites of Garhwal region of Uttarakhand, INDIA during 2009-2010.

## RESPONSE OF *BRACHYPODIUM DISTACHYON* (L.) BEAUV TO OZONE EXPOSURE IN CONTROLLED ENVIRONMENT; A PILOT EXPERIMENT

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Gramineous plants are widespread in many and different environments and are often the cause of health problems and disorders in humans. In particular the pollen produced by these species can cause strong allergic reactions in persons with high sensitivity to pollen constituents. Despite all projects and activities in progress at the moment on this issue (i.e. UNECE Task Force on Health), literature is poor in studies concerning plant species as *Brachypodium distachyon*, especially about its ecophysiological and functional characterization.

Aim of these study was to evaluate the physiological response of *Brachypodium distachyon* to acute ozone dose and to investigate the effects of ozone on pollen production and vitality; this second part of the research is already running and results will not be discussed in the present paper. In this experiment selected seeds of *Brachypodium distachyon* (L.) Beauv were placed in special microenvironments in order to keep high humidity levels and to facilitate their germination. After first root was produced (about 10 days), puppies were placed into small



pots (5-6 plants per pot) within a hydroponic system, equipped with a micro-aerator and fertilized with NPK solution. Plants were placed in a semi-controlled environment in order to limit temperature and humidity changes, equipped with a MH lamp as light source, at 15 – 25°C, 50-60% of relative humidity and 12h photoperiod. This first part of the research was carried out at the Laboratory of Aerobiology of Environmental Sciences Department of University of L’Aquila. At mature stage of growth (after about 4 weeks), plants were transplanted, with their mini-pots, into biggest pots with a mixture of ground and placed in growth walking chambers at the Environmental Biology Department of the

University of Rome “La Sapienza”, for an acclimatization period of about one week (Fig.1). After that, ecophysiological analysis (gas exchange, chlorophyll fluorescence and chlorophyll content) were carried out on plants in order to set a checkpoint and to split plants into two groups, “control” and “treated” for ozone exposure. Conductance data were processed in aim to set ozone dose for the treatment, then treated set was fumigated in growing chamber at 160 ppb x 7h x 4 days. After fumigations, a new round of physiological measurement was carried out together with biometric assessment (height of plants and leaf visible injury).

Preliminary results showed significant differences in gas exchange and relative chlorophyll content between exposed and control plants. Whereas, poor relationships and differences were found in basic chlorophyll fluorescence parameters (as F0, Fv/Fm, ABS/Rc) between the two sets. Finally, after the fumigations, ozone-like injury was shown on some leaves of treated plants, especially in older leaves (Fig.2b). Further studies will be necessary to improve the knowledge of this interesting species especially about ozone physiological response also in relation to human allergic diseases.



# ATMOSPHERIC DEPOSITION OF AIRBORNE RADIONUCLIDES IN CROATIA STUDIED BY THE MOSS BIOMONITORING TECHNIQUE

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Within the UNECE ICP Vegetation survey in 2010 the 15 moss samples out of 160 ones collected over the entire Croatia (Figure) were subjected to radiometry for assessing activity of the naturally occurring radionuclides <sup>40</sup>K, <sup>232</sup>Th, <sup>226</sup>Ra, and <sup>238</sup>U, as well as the technogenic <sup>137</sup>Cs.

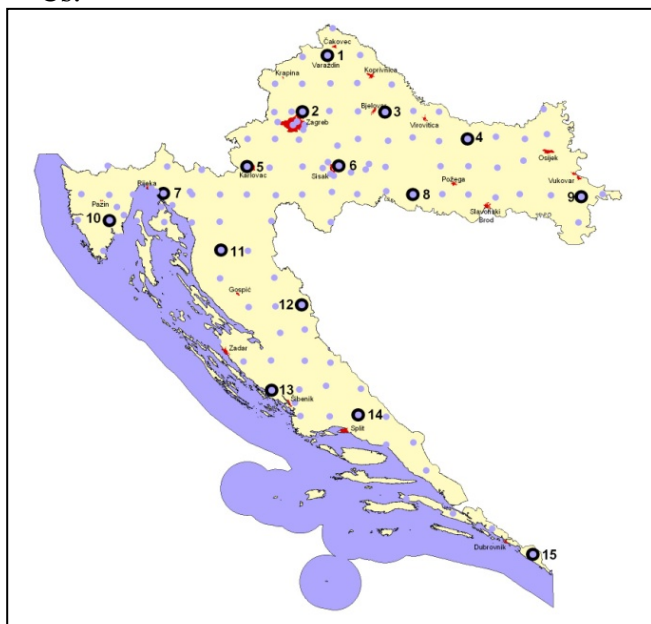


Figure: Moss sampling sites in Croatia

The activities of <sup>40</sup>K, <sup>232</sup>Th, <sup>137</sup>Cs, <sup>226</sup>Ra, and <sup>238</sup>U were determined using a low background HP Ge spectrometer (CANBERRA). For calibration gamma mixed standards supplied by Eckert & Ziegler (Analytcs USA) were used.

Efficiency of the system was regularly checked during intercomparison runs. Precision and accuracy of the system were checked additionally by simultaneous measurement of IAEA Reference Material.

Preliminary results on the selected samples from CRO 2010 moss survey (Table) may, besides the standard monitoring of radioactivity, serve as a baseline – "background pollution levels" – related to the possible effects from the Fukushima (Japan) nuclear disaster in 2011.

Table: Radionuclide activity (Bq/kg) in mosses (dry weight)

No.	Location	<sup>40</sup> K	<sup>232</sup> Th	<sup>137</sup> Cs	<sup>226</sup> Ra	<sup>238</sup> U
1	Varaždin	218 ± 36	9±4	5.7±1.6	<4.5	<18
2	Zagreb	152 ± 35	<4	5.6±1.6	<4	<18
3	Bjelovar	236±40	<4	74±8.5	<5.4	<22
4	Slatina	127±28	4±3	19±2.6	2.7±2.5	<14
5	Karlovac	222±32	11±4	50±6	15±3	<13
6	Sisak	216±39	<4	20.5±3.2	<5.4	<22
7	Rijeka	156±32	<4	27±3.8	<5	30±16
8	Okučani	<b>385±68</b>	15±7	38±5	12±5	<26
9	Vinkovci	304±43	<b>19±6</b>	5±1.6	9±3	23±13
10	Labin	162±33	6±3	<b>4.3±1.4</b>	11±3	<15
11	Otočac	108±23	<3	<b>632±63</b>	<4	<b>63±20</b>
12	Donji lapac	127±24	8±4	6±1.4	<4	40±14
13	Pirovac	90±25	4±3	306±31	6±2.4	<13
14	Sinj	<b>66±25</b>	5±3	57±7	<4	<15
15	Konavli	139±26	8±4	117±12	<b>15±3</b>	<12

- uncertainty expressed with 2k (approx 95% reliability of the results)
- results with highest value - **bold** and lowest – **green**

# COMPARISON OF DIFFERENT SAMPLING MEDIA (MOSS, LICHEN, ATTIC DUST) FOR DETERMINATION OF AIR POLLUTION WITH HEAVY METALS IN THE VICINITY OF COPPER MINE

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Moss and lichen species and attic dust samples were used for monitoring distribution of heavy metals and other trace elements in the vicinity of intensively exploited copper mine. *Hypnum cupressiforme*, *Camptothecium lutescens* and *Homolothecium sericium* were collected as characteristic moss species and *Hypogymnia physodes*, *Pseudevernia furfuracea* and *Evernia prunastri* were collected as characteristic lichens species for comparative analysis [1, 2]. Attic dust samples were collected from the same region in order of historical record for metals distribution [3]. The analysis of 17 elements (Al, Ba, Ca, Co, Cr, Cu, Li, Fe, K, Mg, Mn, Na, Ni, Pb, Sr, V and Zn) was performed by atomic emission spectrometry with inductively coupled plasma (ICP-AES). As and Cd were analyzed by electrothermal atomic absorption spectrometry (ETAAS).

The comparative analysis between elements content from mosses and lichens showed: (a) collected species were very tolerant to high content of Cu, Pb, Fe (>0.01%, >0.01% and 1% respectively); (b) moss species accumulate higher amounts of lithophile elements (Al, Ba, Cr, Fe, Mn, Na, Ni, Sr) and lichen more easily accumulates atmophile elements (Pb, V, Zn); (c) mosses and lichens cannot be used interchangeably as biomonitors of metals in areas with Cu mineral deposits and ore processing. Attic dust samples enabled recording the historical emission of heavy metals in air with emphasis of Cu, where was found that there is a deposition of large amount of copper. The highest values for copper were obtained from lichen, moss and attic dust (134, 198 and 415 mg kg<sup>-1</sup> respectively) samples collected very close to the mine.

The ultimate effect is that spatial distribution for element deposition (with emphasis on heavy metals) is not disturbed with the significant differences in sampling media matrix. Using of different types of environmental samples unable covering deficiencies one with another in order of monitoring long term and long distance metal distribution and its deposition in the environment.

1. Balabanova, B., Stafilov, T., Bačeva, K., Šajin, R. (2010). Biomonitoring of atmospheric pollution with heavy metals in the copper mine vicinity located near Radoviš, Republic of Macedonia, *Journal of Environmental Science and Health, Part A, Toxic/Hazardous Substance & Environmental Engineering*, 45, 1504–1518.
2. Balabanova, B., Stafilov, T., Šajin, R., Bačeva, K. (2011). Distribution of chemical elements in attic dust as reflection of lithology and anthropogenic influence in the vicinity of copper mine and flotation, *Archives of Environmental Contamination and Toxicology*, 61, 173–184.
3. Balabanova, B., Stafilov, T., Šajin, R., Bačeva, K. (2012). Characterisation of heavy metals in lichen species *Hypogymnia Physodes* and *Evernia Prunastri* due to biomonitoring of air pollution in the vicinity of copper mine, *International Journal of Environmental Research*, in press.

# SULPHUR AND NITROGEN MEASUREMENT IN MOSSES IN SWITZERLAND 2010, FIRST RESULTS

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At the TFM-meeting in Belgium 2010 we suggested, in addition to nitrogen, to compare also sulphur concentration in moss and sulphur deposition through precipitation.

In the year 2010, either *Hypnum cupressiforme* or *Pleurozium schreberi* was collected from 14 sites which were less than 100 m away from LWF (Thimonier et al. 2005) and FUB plots where deposition is being measured. The 3-year old part of the moss was analysed and the N- and S-concentrations were then plotted against the N- and S-deposition (average 2008-2010).

**Results and Discussion:** The relationship of nitrogen in moss and precipitation is significant, similar to the 2005 survey, although the  $R^2$  is lower (0.52; 2005 = 0.90) (fig. 1). Sulphur however, shows no dependency (with a slight trend to a negative correlation) (fig. 2). As S is a macronutrient the background level is high; the lowest measured value in moss was about  $600 \mu\text{g g}^{-1}$ . Even in very remote regions such as Spitsbergen (Grodzinska & Godzik 1991) and Antarctica (Ganeva & Yurukova 2004) S-concentrations between 700 and  $1'800 \mu\text{g g}^{-1}$  were found.

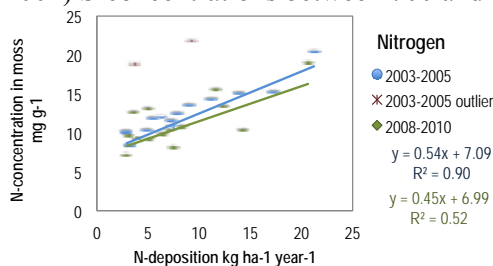


Fig. 1: N: comparison moss conc. to deposition

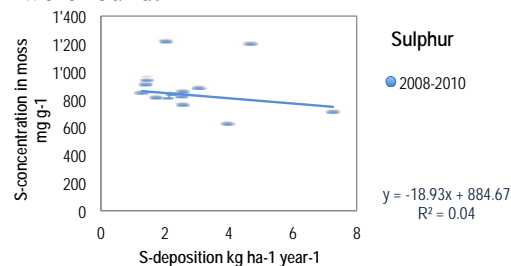


Fig. 2: S: comparison moss conc. to deposition

No change of N-concentration in moss or only a small reduction tendency was found between 1995 and 2010 (fig. 3). The same was found when analysing deposition (BAFU 2011). Sulphur as  $\text{SO}_2$  in the air and  $\text{SO}_4^{+}$  in PM10 and precipitation, however, was distinctly higher in the late eighties and nineties, but no significant change was found in S concentration in moss (fig. 4).

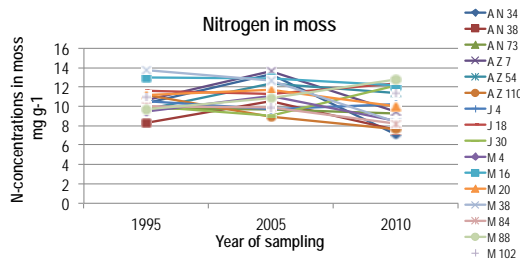


Fig. 3: Nitrogen: Course over the years (1995, 2005 analysed 2006)

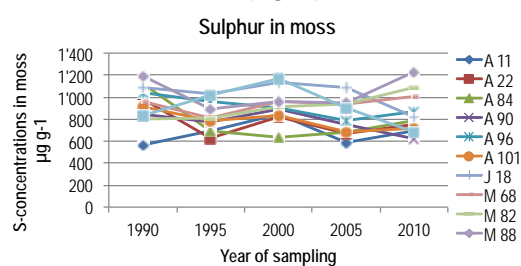


Fig. 4: Sulphur: Course over the years (all analysed 2011)

**Conclusion:** In contrary to N, we found no relationship between S-concentration in moss and S-deposition, in the order of magnitude we measured in Switzerland. Could a dependency be found when the S-deposition is much higher?

Lit: Thimonier A. et al., 2005 Environmental Monitoring and Assessment 104, 81-118  
Grodzinska, K., Godzik, B., 1991. Polar Research 9, 133-140  
Ganeva, A., Yurukova, L., 2004. Herzogia 17, 199-206  
BAFU 2011: NABEL – Luftbelastung 2010. Umwelt-Zustand Nr. 1118, 126 S.



# ANTHROPOGENIC SILVER IN SWITZERLAND: IS THE NANOINDUSTRY A POTENTIAL NEW EMISSION SOURCE?

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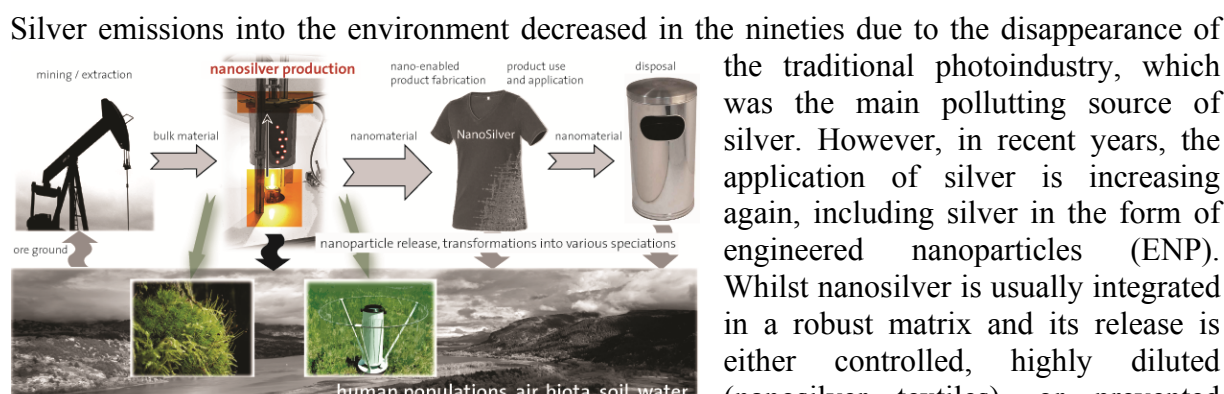


Figure 1. Life cycle of engineered nanomaterials. Our study focused on the production phase and assessed nanoparticle emissions to the environment by means of biomonitoring (mosses) and precipitation samplers.

the traditional photoindustry, which was the main polluting source of silver. However, in recent years, the application of silver is increasing again, including silver in the form of engineered nanoparticles (ENP). Whilst nanosilver is usually integrated in a robust matrix and its release is either controlled, highly diluted (nanosilver textiles), or prevented (computer modules), high point source silver emissions may occur at the production sites or during disposal of nanosilver-containing products.

Atmospheric emissions are of particular concern due to the deeply inhalable fraction with corresponding potential long-term adverse effects to humans.

The aim of this study was to quantify silver deposition in the immediate surroundings of a nanosilver production company, (i) in terms of concentration in mosses (*Brachythecium rutabulum* and *Hypnum cupressiforme*) over three years, (ii) to measure wet and dry silver deposition on a monthly basis with the Bergerhoff method, assuming that eventually all silver particles in the air are deposited (dry deposition) or washed out (wet deposition), (iii) to analyze spatially explicit deposition patterns with increasing distance from the potential emission source, (iv) to compare morphological changes of the nanosilver from production to deposition, and (iv) to compare the detected silver concentrations with known background concentrations in other Swiss regions and with historical data (142 sites). This step provides valuable information on potential anthropogenic silver sources.

Silver concentrations in the mosses were between 0.002 and 0.033  $\mu\text{g g}^{-1}$ . The silver concentration at the sampling point >8km away from the production facility was below the quantification limit, indicating that the silver concentrations decrease with increasing distance from the production facility. We found lower silver concentrations in the opposing average wind direction than in the prevalent direction. The absolute deposition rate of airborne silver was between 0.05  $\mu\text{g d}^{-1} \text{m}^{-2}$  and 0.44  $\mu\text{g d}^{-1} \text{m}^{-2}$ . In comparison to other regions in Switzerland, the deposition rates were slightly higher, but in the same order of magnitude. The same holds for the silver concentrations in the mosses. Concerning the morphology, electron microscopy showed silver of 80 nm diameter in the form of silver chloride.

# ESTIMATING STOMATAL OZONE UPTAKE OF ALPINE GRASSLAND – A COMBINED FIELD AND CLIMATE CHAMBER STUDY

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Air pollution by ozone ( $O_3$ ) occurs over wide areas and represents a major threat to vegetation across Europe and elsewhere. Adverse effects of elevated  $O_3$  on agricultural crops and forest trees have been documented [e.g. 1], while much less is known about effects on semi-natural vegetation, including grasslands [2].

A long-term field experiment was carried out at 2000 m a.s.l. in the central Swiss Alps (Alp Flix) with the aim to investigate effects of elevated  $O_3$  and N deposition, and of their combination, on subalpine grassland. While clear effects of elevated N deposition on growth and community composition were found, elevated  $O_3$  did not show market effects [3]. We

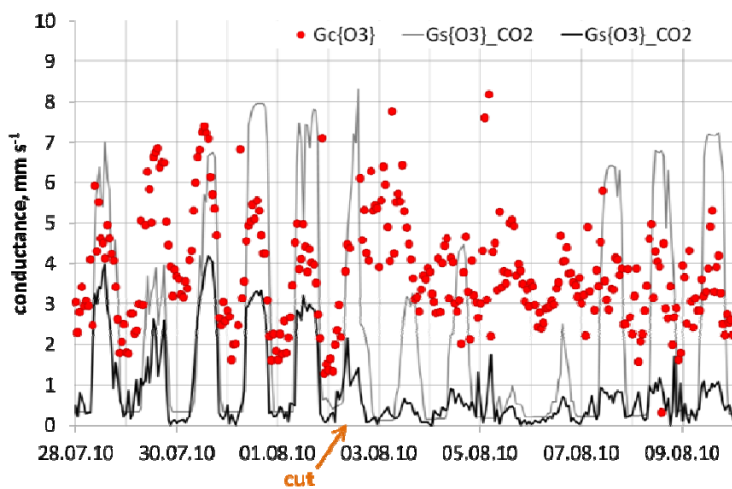


Figure 1: time series of canopy conductance of  $O_3$  ( $G_c\{O_3\}$ ), and estimated stomatal conductances ( $G_s\{O_3\}_{CO_2}$ ,  $G_s\{O_3\}_{H_2O}$ ).

hypothesized that low stomatal uptake and/or high detoxification capacity was responsible for the lack of an  $O_3$  response. To determine stomatal  $O_3$  uptake, an intensive observation study was conducted in 2010. We performed dynamic chamber exchange measurements [4] of  $O_3$ , water vapour ( $H_2O$ ) and carbon dioxide ( $CO_2$ ) (i) at the field site and (ii) in a climate-controlled chamber using monoliths from the site. At the field site, prior to the cut canopy conductance for  $O_3$  ( $G_c\{O_3\}$ ) showed a pronounced diurnal cycle and ranged from  $1.5 \text{ mm s}^{-1}$  during night up to  $7 \text{ mm s}^{-1}$  during daytime (see Figure 1). After the cut, the diurnal cycle lost in shape and amplitude, and ranged from  $2.0 \text{ mm s}^{-1}$  at night to  $5.0 \text{ mm s}^{-1}$  during the day. Stomatal conductance for  $O_3$  ( $G_s\{O_3\}$ ) was derived from  $G_s\{H_2O\}$  as estimated using two different approaches: (a)  $G_s\{H_2O\}_{H_2O}$ : a Jarvis-type parameterisation [5] on temperature, radiation, and soil water content; (b)  $G_s\{H_2O\}_{CO_2}$ : from  $CO_2$  exchange measurements [6]. While both approaches followed well the overall measured  $G_c\{H_2O\}$  (under dry conditions, when soil and other surface evaporation was negligible),  $G_s\{H_2O\}_{CO_2}$  seemed to represent stomatal activity better than  $G_s\{H_2O\}_{H_2O}$ , as clearly visible following the cut. Comparing  $G_s\{O_3\}_{CO_2}$  and  $G_c\{O_3\}$ , stomatal  $O_3$  uptake accounted for 30 to 60% of total  $O_3$  deposition. The results indicate that stomatal  $O_3$  uptake was comparable to other grasslands at lower altitude.

Further analysis will include the results of the climate chamber measurements, comparison of the results with deposition schemes used in  $O_3$  deposition models (e.g.  $DO_3SE$ ), and a tentative upscaling of our results to estimate stomatal  $O_3$  uptake for the whole vegetation period.

[1] Fuhrer, J. et al., 1997. *Environmental Pollution* 97, 91-106. [2] Bassin, S. et al., 2007a. *Environmental Pollution* 146: 678-691. [3] Bassin, S. et al., 2007b. *New Phytologist* 175: 523-534. [4] Pape, L. et al., 2009. *Biogeosciences* 6, 405-429. [5] Jarvis, P.G., 1976. *Phil Trans Roy Soc. London B273*: 593-610. [6] Lamaud, E. et al., 2009. *Agricultural and Forest Meteorology* 149, 1385-1396.

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