ICP VEGETATION

27th Task Force Meeting and ozone workshop

28 – 30 January, 2014
Paris, France

Programme

&

Abstracts

Working Group on Effects of the Convention on Long-range Transboundary Air Pollution

Organizers:

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

Harry Harmens
Gina Mills
Felicity Hayes

Local organizers:

Jean-François Castell (AgroParisTech)
Olivier Bethenod (INRA)
Anne Repellin (UPEC)
Sébastien Leblond (MNHN)
Laurence Galsomiès (ADEME)
Sophie Formisano (INRA)

Host: ***French Environment and Energy Management Agency (ADEME)***

With financial support from*:*

The Ile-de-France Region, ADEME, City of Paris, UPEC, AgroParisTech and INRA

Programme one-day ozone workshop *(Location:* AgroParisTech)

Tuesday 28th January, 2014

08:15 **Registration**

08:45 *Harry Harmens* – Introduction to the workshop

Theme 1: Quantifying ozone impacts on trees

Chair: Harry Harmens **Rapporteur: Felicity Hayes**

Questions to be discussed:

- *Can we add new flux parameterisations for additional tree species to the Modelling and Mapping Manual?*
- *Can we set critical levels for new tree species?*
- *Is amendment of current critical levels required?*
- *Is there new field-based evidence of predicted impacts?*

09:00 – 10:30: session 1 (10 min per presentation, 5 min discussion per paper)

09:00 *Alexandra De Marco* et al. – Ozone stomatal flux-based critical levels translated into real-world forest impacts.

09:15 *Silvano Fares* et al. – Ball-Berry vs Jarvis approaches to model stomatal ozone deposition in Mediterranean forests: validation using the test sites of Castelporziano and San Rossore forests.

09:30 *Beat Rihm* et al. – Mapping of ozone flux in Switzerland with high spatial resolution for epidemiological analysis.

09:45 *Patrick Büker* et al. – The effect of ozone on the C sequestration potentials of European forests.

10:00 General discussion.

10:30 – 11:00 Coffee/tea

11:00 – 12:30: session 2 (10 min per presentation, 5 min discussion per paper)

11:00 *Per Erik Karlsson* et al. – Further developments of the concept of the Simplified Phytotoxic Ozone Dose, S-POD, to assess the risk for ozone impacts on trees on the regional scale.

11:15 *Sabine Braun* – Validation of stomatal flux in *Quercus* species.

11:30 *Patrick Büker* et al. – DO_3SE parameterisation for new forest tree species and revision of flux-response relationships for eight European trees species (*note: 15 min presentation*).

11:45 *Harry Harmens* – Structured discussion on questions, conclusions & recommendations on trees.

12:30 – 14:00 Lunch

Theme 2: Quantifying ozone impacts on crops and (semi-)natural vegetation

Chair: Håkan Pleijel **Rapporteur: Patrick Büker**

Questions to be answered:

- *Do we have new flux parameterisations for additional crops and (semi-) natural vegetation species?*
- *Can we set critical levels for new crop species?*
- *Can we set critical levels for generic crops species for IAM?*
- *Can we set new critical levels for grassland species?*

14:00 – 15:30: session 3 (10 min per presentation, 5 min discussion per paper)

14:00 *Olivier Bethenod* et al. – Ozone impact on maize.

14:15 *Patrick Stella* et al. – Two-year total, stomatal, cuticular, and soil ozone budgets of an agricultural field with winter wheat and maize crops near Paris, France.

14:30 *Giacomo Gerosa* et al. – Yield response of some Italian and Spanish cultivars of durum wheat to elevated ozone: a varietal screening.

14:45 *Felicity Hayes* et al. – Dose-response relationships for grassland species.

15:00 General discussion.

15:30 – 16:00 Coffee/tea

16:00 – 17:30: session 4

16:00 *Gina Mills, Håkan Pleijel* et al. – Application of a generic crop flux-effect relationship in integrated assessment modelling in Europe.

16:20 *Patrick Büker* et al. – Overview of the DO_3SE parameterisation for new crop species.

16:35 *Håkan Pleijel* – Structured discussion on questions, conclusions & recommendations on crop and grassland species; amendments to chapter 3 of the Modelling and Mapping Manual.

17:30 End

18:30-20:30 Welcome buffet/reception and registration Task Force Meeting (*Location:* AgroParisTech)

Programme 27th Task Force Meeting of the ICP Vegetation

Tuesday 28th January, 2014

18:30-20:30: Welcome buffet/reception and registration Task Force Meeting (*Location*: AgroParisTech)

Wednesday 29th January, 2014

***Location*: the Auditorium de la Grande Galerie de l'Evolution du Museum National d'Histoire Naturelle**

09:00 Late registration

Session 1: 09:15 – 10:30 (Plenary session at the Auditorium de la **Grande Galerie de l'Evolution** du Museum National d'Histoire Naturelle**)**

Session 1: 9:15 – 10:30 Plenary session **Chair: Jean-François Castell**

09:15 Welcome addresses:
- Laurence Galsomies (ADEME)
- Jean-Philippe Siblet (Natural History Museum)
- Speaker to be confirmed (Regional representative)

09:35 *Krzysztof Olendrzynski* (Secretariat LRTAP Convention) – Update on the activities under the LRTAP Convention.

09:50 *Harry Harmens* et al. – Overview of the achievements of the ICP Vegetation in 2013 and future workplan (2014-2016).

10:15 *Jean-Paul Hettelingh, Anne Christine Le Gall* et al. – Overview of the activities of ICP Modelling and Mapping and plans for the future.

10:30 – 11:00 Coffee/tea

Session 2: 11:00 – 12:30 Plenary session **Chair: Jürgen Bender**

11:00 *Jean-Francois Castell* et al. – Validity of usual dose-response functions in the case of wheat exposed to free-air fumigation in the Paris region.

11:15 *Thomas Verbeke* et al. – Incorporating a more mechanistic ozone impact parameterisation in the ORCHIDEE global vegetation model.

11:30 *Beat Achermann* et al. – Effects of nitrogen deposition on species richness and diversity in Switzerland.

11:45 *Marina Frontasyeva* – Transfer of the coordination of the European moss survey to the Russian Federation: plans for the future.

12:00 *Katrina Sharps* – New ICP Vegetation smart-phone App for recording incidences of ozone injury on vegetation.

12:15 General discussion.

12:30 – 14:00 Lunch and putting up posters - Caves Esclangon - University of Paris VI

Session 3: **14:00 – 15:30 Two parallel sessions: Ozone** in the Auditorium de la Grande Galerie de l'Evolution, **Moss survey** in University of Paris VI.

Session 3a: **Ozone** **Chair: Alessandra De Marco**

14:00 *Håkan Pleijel* et al. – Latitude dependence and temporal trend of the yearly peak in surface ozone in middle and northern Europe.

14:20 *Matthias Volk, Seraina Bassin* et al. – Seven years of N- and O_3 pollution × climate interaction in subalpine grassland: ecosystem carbon budget, nitrogen pools and O_3-flux modelling (POD).

14:40 *Dan Hewitt* et al. – Is ozone reducing the N fixation capacity of current managed pasture?

15:00 Demonstration and discussion on plans for use of ICP Vegetation smart-phone App for recording ozone damage to vegetation.

Session 3b: **Moss survey** **Chair: Gunilla Pihl-Karlsson**

14:00 *Harry Harmens, Lotti Thöni* et al. – Relationship between site-specific nitrogen concentrations in mosses and bulk atmospheric nitrogen deposition.

14:20 *Winfried Schröder* et al. – Correlations between nitrogen concentrations in atmospheric deposition and mosses mapped for natural landscapes in Europe.

14:40 *Michaela Kluge* et al. – Differences in concentration of nitrogen in mosses due to canopy drip effects - case study Germany.

15:00 *Mitja Skudnik* et al. – Indicative content of N, S and $\delta^{15}N$ in the moss *Hypnum cupressiforme* in the southern part of Central Europe.

15:20 General discussion.

15:30 - 16:00 **Refreshments**

Session 4: **16:00-17:15 Two parallel sessions Ozone and Moss survey**

Session 4a: **Ozone** **Chair: Silvano Fares**

16:00 *Gina Mills* et al. – Overview of progress for the EU FP7 ECLAIRE project.

16:20 *Felicity Hayes* et al. – Impacts of ozone and nitrogen on silver birch.

16:40 *Giacomo Geroso* et al. – Stomatal uptake and non-stomatal ozone removal by a mixed oak-hornbeam mature forest in the Po Valley: Results of the ECLAIRE long-term campaign.

17:00 General discussion.

Session 4b: **Moss survey** **Chair: Harry Harmens**

16:00 *Marina Frontasyeva* – Moss biomonitoring of trace elements and radionuclides in rural and urban areas experiencing environmental stress.

16:30 *Harry Harmens and Marina Frontasyeva* – Preparations for the European moss survey in 2015: monitoring manual, coordination, participation, stimulating participation more EECCA countries and Asia.

18:00- 19:30 **Poster session and buffet – Caves Esclangon – University of Paris VI**
(remember to take posters down at the end!!).

Thursday 30th January, 2014

Session 5: **9:00 – 10:30 Two parallel sessions: Ozone** in the Auditorium de la Grande Galerie de l'Evolution, **Moss survey** in University of Paris VI.

Session 5a: **Ozone** **Chair: Kent Burkey**

09:00 *Muhammad Adrees* et al. – Ambient air ozone impact on growth, yield and nutritional quality of wheat, maize and mung bean at two sites of Faisalabad-Pakistan.

09:15 *Marien Havé* et al. – Vacuolar endoproteolytic activities and protein carbonylation levels in leaf as a model for depicting ozone tolerance and sensitivity in winter wheat (*Triticum aestivum* L.).

09:30 *Samia Madkour* et al. – The role of jasmonate and salicylic acid in conferring tolerance against ozone injury in tomato plants.

09:45 *Elisa Pellegrini* et al. – Circadian profiles of photosynthetic parameters and primary metabolites in grapevine varieties exposed to ozone.

10:00 *Yuri Maia* et al. – Involvement of plastidial terminal oxidase (PTOX) and mitochondrial proteins (AOX and pUCP) in the response of Vigna unguiculata (cowpea) to combined drought stress and ozone exposure.

10:15 General discussion

Session 5b: **Moss survey:** **Chair: Marina Frontasyeva**

09:00 *Ilia Ilyin* et al. – Evaluation of heavy metal and POP pollution levels in the EMEP region using atmospheric deposition modeling and measurements in mosses.

09:20 *Martijn Schaap* et al. – Atmospheric deposition of heavy metals to terrestrial ecosystems in Germany.

09:40 *Pranvera Lazo* et al. – The survey of atmospheric deposition of Al, Cr, Fe, Ni, V and Zn in Albania by using moss biomonitoring and ICP-AES.

10:00 General discussion, recommendations and decisions.

10:30 – 11:00 Refreshments

Session 6: **11:00-12:30 Two parallel sessions: Ozone** in the Auditorium de la Grande Galerie de l'Evolution, **Moss survey** in University of Paris VI.

Session 6 a: **Ozone** **Chair: Håkan Pleijel**

11:00 *Lorenzo Cotrozzi* et al. – Response of *Quercus cerris* to combined ozone and drought stress.

11:20 General discussion, recommendations and decisions (including from the ozone workshop).

Session 6b: **Moss survey** **Chair: Sébastien Leblond**

11:00 *Zdravko Spiric* et al. – Croatia participated in the ICP Vegetation HM survey since 2005.

11:20 *Flora Qarri* et al. – Assessment of trace elements pollution around Vlora area, Albania.

11:40 *Sergio Esposito* et al. – A possible role of Heat Shock Proteins as sensor of heavy metal pollution in Bryophyta.

12:00 *Anatoliy Dunaev* et al. – Biomonitoring of trace elements in Ivanovo region (Central Russia).

12: 20 General discussion.

12:30 – 14:00 Lunch - Restaurant of the Great Mosque of Paris

Session 7: **14:00 – 16:00 Plenary session** **Chair: Harry Harmens**

- Reporting decisions and recommendations.
- ICP Vegetation work programme 2014 – 2016.
- Collaboration with other relevant bodies/organizations.
- *Patrick Büker* et al. – Outreach activities: Raising awareness of air pollution impacts on vegetation in South Asia.
- Conclusions and review of the 27th Task Force Meeting.
- Next Task Force Meeting.
- Any other business.

Excursion: **16:00 – 18:00 Grande Galerie de l'Evolution or Paleontology Gallery or Museum Greenhouses (choice of one of the three)**

20:00 **Conference dinner: dinner-cruise on the Seine River**

List of posters Task Force meeting

Pollutant	Author(s)	Title
Ozone	Agathokleous Evgenios, Saitanis Costas, Papatheohari Yolanda	Evaluation of Pinolene as protectant against ozone phytotoxicity
	Bermejo-Bermejo, Calatayud, V., Cerveró, J., Pastor, I., Calvo, E., García, H, Elvira, S., Alonso, R.	Ozone effects on yield and gas excahnge of spinach and chard
	Kent O. Burkey	Temperature effects on yield of R123 and S156 snap beans - establishing criteria for effective use as an ozone bio-indicator system
	Campanella A., Lorenzini G., Nali C.	Ozone: an elicitor of secondary compounds in *Hypericum perforatum*
	Fusaro L., Fares S., Gerosa G., Marzuoli R., Salvatori E., Finco A., Quarato D., Monga R., Manes F.	Photosynthetic performance of *Quercus ilex* L. under long-term ozone exposure probed by carboxylation efficiency, maximum apparent quantum yeld and modulated 820 nm reflection
	Gerosa Giacomo, Marzuoli R., Monga R., Finco A.	Effects of ozone and nitrogen deposition in young trees of hornbeam and oak: results from the ECLAIRE experiments in Italy
	Maamar B., Maatoug M., Dellal A, Ait Hammou M.	Study of a tropospheric ozone effect on two cultivars of tomato: Lycopersicon lycopersicum colgar (L.) Karsten ex Farw. and Solanum lycopersicum rechaiga II
	Spiric, Zdravko et al.	Effects of ozone on bean (*Phaseolus vulgaris*) in Croatia in 2013
	Sanz, J., Calvete-Sogo, H., Gonzalez-Fernandez, I., García H., Alonso, R., Bermejo-Bermejo, V.	Concentration-based critical levels for quality and yield of annual pasture
	Schröder, M. ; Grünhage, L.	Impact of climate change on stomatal ozone uptake
Heavy metals	L. Barandovski, M.V. Frontasyeva, T. Stafilov, R. Sajn, K. Baceva, A.Yu. Dmitriev	Air pollution study in Macedonia by using moss biomonitoring technique, NAA, ICP-AES, and AAS
	Lirim Bekteshi, Flora Qarri, Pranvera Lazo, Trajce Stafilov	Atmospheric deposition of heavy metal in eastern Albania by moss biomonitorig technique
	Borowiak K., Zbierska J., Barałkiewicz D., Hanć A., Kayzer D., Budka A.	Relations between accumulation of trace elements, photosynthetic acitivity, water status and growth parameters of Italian ryegrass exposed in ambient air conditions
	Bouchenafa Nadia,Maazouz Sihem, Dellal Abdelkader,	Mapping of air pollution roadside origin by some trace metals (Cd and Cu) using the transplantation of a bio accumulator lichen *Xanthoria parietina* in the city of Tiaret (Algeria)
	S.V.Gorelova, M.V. Frontasyeva, A.V.Gorbunov, S.M. Lyapunov, O.I Okina	Bioindication and monitoring of atmospheric deposition using trees and shrubs
	Koroleva Yuliya, Vakhranyova Olga, Styogantsev Vasiliy, Melnikova Irina	Accumulation of heavy metals in the Moss Pleurozium Schreberi in the West of Russia (Kaliningrad Region)
	Maňkovská, B.; Oszlányi, J.; Izakovičová, Z., Frontasyeava,M.V.	Critical evaluation of ecosystem pollution
	Albert Maxhuni, Pranvera Lazo	Atmospheric deposition of heavy metal deposition in Kosovo by using moss biomonitoring and AAS
	Jarmo Poikolainen	Changes in the heavy metal contents of dried moss samples during long-term dry storage
	I. Popescu, M.V. Frontasyeva et al.	Atmospheric deposition of major and trace elements in Romania studied by NAA and AAS: moss survey 2010/2011
	Saitanis Costas, Lappa Katerina., Ntatsi Georgia, Barouchas Pantelis, Agathokleous Evgenios	Biomonitoring of heavy metals in playgrounds in Athens, Greece
	Saxena, Dinesh (to be confirmed)	Analysis of moss Thuidium cymbifolium Doz. et Molk to retrieve past atmospheric elemental profile of Garhwal state of in India
	Urbaniak J., Budka A., Kayzer D., Borowiak K., Zbierska J., Barałkiewicz D., Hanć A.	Biomonitoring of air pollution by trace elements using italian ryegrass (*Lolium multiflorum* l. 'Lema')
Nitrogen	Boltersdorf SH, Pesch R, Schröder W, Werner W	Physiological responses: do lichens, mosses and tree bark react with different carbon and nitrogen isotope patterns along a nitrogen deposition gradient?
	I. Fumagalli and Gruening C.	One year results of soil NO and NO_2 fluxes measurements in subalpine forest
	Zdravko Špirić, Trajče Stafilov, Ivana Vučković, Marin Glad, Vladimir Kušan	Nitrogen in mosses in Croatia in 2006/2010
	Werner W. & Schlimpen K.	Some consideratons about ^{15}N-patterns in moss tissue around a poultry farm
PM	Laffray, X., Domanski, M., Castell, J.F.	Foliar surface cleaning techniques for analysis of particulate chemical composition in air pollutants monitoring

Abstracts

Presentations

Ozone workshop

OZONE IMPACT ON MAIZE

[a]Bethenod O., [c]Leitao L., [b]Castell J.F., [a]Massad R., [a]Tuzet A., [d]Le Thiec D., [c]Repellin A., [c]Bagard M.

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Ozone (O_3) is the air pollutant that currently represents one of the greatest environmental concerns in the troposphere, with particles and nitrogen oxides. Ground levels of O_3 have increased continuously over the three decades before 2000, at a rate of 0.5-2% per year[1]. Although a stagnation or slight decrease over Europe and North America was observed since 2000, O_3 concentration is still increasing in Asia and others world parts[2]. Based on dose-effect curves, Mills *et al*. (2007)[3] identified three crops groups according to ozone sensibility: ozone sensitive crops (such as wheat), moderately sensitive crops (such as maize) and ozone resistant crops (such as barley)[4].

For a given AOT40, the decrease in leaf photosynthesis (established using the A/Ci curves) is similar in maize and wheat whereas the decrease in grain dry matter is about doubled in wheat compared to maize. To understand this discrepancy, we investigated changes in maize (*Zea mays* L.) Leaf Area Index (LAI), leaf gas exchanges and harvest index, due to ozone using a new linear field fumigation device.

When fumigation occurred before flowering, leaf area decreased up to 15%[4]. If the LAI remained over 4, the intercepted radiation by the maize plants was maximal. Therefore, in this case, ozone impact on maize intercepted radiation was limited. When fumigation occcured after flowering, the decrease in leaf photosynthesis in the upper leaves of the plants negatively impacted their dry matter production. In response to an AOT40 of 22 mmol $mol^{-1}h$, leaf photosynthesis decreased by 30% whereas the grain dry matter was reduced by 5% only, as previously observed by Mills *et al.* (2007)[3]. In addition, the shoot dry matter production was reduced by more than 11% and the harvest indexes were 51.6 % in control plants and 59 % in fumigated plants. Moreover, ozone-induced moderation of stomatal conductance resulted in a limitation of gase fluxes (CO_2,O_3) entering in the leaves. This moderation of stomatal conductance was similar to that observed for leaf photosynthesis. Since the stomatal conductance of maize leaves was approximately half that of wheat leaves, the phytotoxic ozone dose (POD) [5] in maize was half that of wheat POD, for a given AOT40.

In conclusion, maize grain production was less impacted by ozone than wheat one due in part to lower maize gs and a grain sink priority.

References

[1] Vingarzan, 2004, Atmospheric Environment, 38, 3431-442.
[2] Oltman *et al*., 2013, Atmospheric Environment, 67, 331-351.
[3] Mills *et al*. 2007, Atmospheric Environment, 41, 2630-2643.
[4]Leitao *et al*., 2007, Plant Biol. (Stuttg). 9, 478–88.
[5]Grunhage *et al*., 2012, Environmental Pollution, 165, 147-157.

VALIDATION OF STOMATAL FLUX IN *QUERCUS* SP.

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Oak species (mainly *Quercus petraea* and *Q. robur*) are important in Switzerland, and it is expected that their significance in forestry will increase in future due to climate change. Ozone effects on *Quercus* are therefore an important issue, and their quantification needs estimates of ozone uptake. The recommendations for flux calculations of oaks in the Mapping Manual (UNECE, 2010) are based for several important parameters on the generic deciduous tree parameterisation. The aim of the present investigation was (1) to validate the proposed parameterisation using sap flow and (2) to test if *Q. petraea* and *Q. robur* have to be treated separately. Sap flow has been used successfully to validate stomatal uptake of ozone in *Fagus sylvatica* (Braun et al., 2010).

Three forest plots in Switzerland including *Fagus sylvatica*, *Quercus petraea* and/or *Q. robur* were equipped with sap flow sensors in the years 2012 and 2013. Analysis of the oak sap flow data was performed in a similar way as described earlier for beech. The multivariate nonlinear regression approach was applied successfully but some questions remained open. Some of the *Q. petraea* showed a strange sapflow pattern in the outer sapwood, and it seems that the optimum temperature may vary from year to year depending on the climatic conditions. The first results suggest that *Q. petraea* and *Q. robur* need different parameterisations for calculating ozone uptake.

References

Braun, S., Schindler, C., Leuzinger, S., 2010. Use of sap flow measurements to validate stomatal functions for mature beech (*Fagus sylvatica*) in view of ozone uptake calculations. Environmental Pollution 158, 2954-2963.

UNECE, 2010. Mapping Critical Levels for Vegetation. Revised Chapter 3 of the UNECE Manual on methodologies and criteria for Modelling and Mapping Critical Loads & Levels and Air Pollution Effects, Risks and Trends. Prepared under the Convention on Long-range Transboundary Air Pollution (UNECE) by the International Cooperative Programme on Effects of Air Pollution on Natural Vegetation and Crops (ICP Vegetation, Centre for Ecology & Hydrology (CEH), Bangor, UK.

URRENT DAY TROPOSPHERIC OZONE CONCENTRATIONS SUBSTANTIALLY REDUCE THE CARBON SEQUESTRATION POTENTIAL OF LIVING TREE BIOMASS ACROSS EUROPE

Patrick Büker[1], Harry Harmens[2], Gina Mills[2], Lisa Emberson[1]

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Tropospheric ('ground level') ozone (O_3) is not only the third most important greenhouse gas, it is also an important air pollutant that reduces plant growth and crop yield. This damaging impact of O_3 on vegetation reduces the amount of carbon (C) sequestered in living biomass and therefore may enhance climate change by increasing atmospheric CO_2 concentrations. Trees have the greatest C sink capacity of any vegetation and represent the largest stock of stored biomass C.

In this study we estimate the impacts of O_3 on sequestered C - expressed as net annual increment (NAI) - of European forest trees using re-analysed UNECE LRTAP (Convention of Long-range Transboundary Air Pollution) flux-based dose-response relationships. The methodology is applied for the year 2000 using EMEP (European Monitoring and Evaluation Programme) meteorology and O_3 data and the effect of soil water conditions and species' sensitivity (distinguishing only conifer and deciduous vs. specific parameterisation for six tree species) on O_3 flux and C sequestration response is investigated.

Ambient stomatal O_3 fluxes in 2000 reduced C storage in the living biomass of trees on average by approximately 19% across Europe, with the highest and lowest relative reductions in Central/Western Europe and Southern Europe, respectively. The highest absolute reductions generally occurred in forest-rich countries such as France, Germany, Poland, Finland and Sweden. The inclusion of the potential for soil water to limit O_3 uptake reduced the O_3 fluxes and hence C sequestration losses especially in the Mediterranean countries. The species-specific parameterisation led to generally slightly higher fluxes, suggesting that a parameterisation only accounting for deciduous and coniferous trees might underestimate the risk O_3 poses to C sequestration.

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

OZONE STOMATAL FLUX-BASED CRITICAL LEVELS TRANSLATED INTO REAL-WORLD FOREST IMPACTS

De Marco A. [(1)], Sicard P. [(2)], Dalstein-Richier L. [(3)], Paoletti E. [(4)]

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Ground-level ozone (O_3) is an important atmospheric pollutant, a pressing sanitary problem for human and ecosystem health, and a serious climate forcer. The European region at highest O_3 risk is the Mediterranean area. Indeed, the climate change is expected to be more pronounced in the Mediterranean Basin than in most other regions of the world. In the Western European Mediterranean area, we demonstrated that the ozone pollution is a major air quality issue in both remote and urban atmospheres.

To date, most experiments to establish biologically relevant plant responses to ozone have been performed under controlled conditions, not representative of actual field conditions, and the results may not provide realistic results for developing standards. An assessment based on phytotoxic ozone dose (PODY) and on real plant damage is more appropriate. The LIFE+ FO_3REST project allows testing the current exposure-based critical levels (AOT40) and suggesting new ozone flux-based critical levels for Mediterranean forest protection against ozone pollution.

A field survey of ozone-induced symptoms was carried out (ICP-Forests protocol) in 54 plots in South-eastern France and North-western Italy. Stomatal ozone fluxes were modelled and correlated to real-world forest impacts in terms of visible injury (stippling/mottling, crown discoloration and leaf loss) in 2012 and 2013. The indicators POD0 and POD1 were calculated thanks to the DO3SE model using the available parameterization for Mediterranean and continental deciduous broadleaf forests and conifers. Spearman test was carried out to understand the relative contribution of O_3 (concentrations or stomatal uptake) to visible injury occurrence. From the flux-effect relationships we derived and proposed species-specific flux-based critical levels CLef for forest protection. In a climate change context, with information in hand, policymakers can make decisions about new legislation for air pollution control strategies in Europe.

Acknowledgment
This work was made possible with the contribution of the LIFE financial instrument of the European Union (LIFE10 ENV/FR/208) for the FO_3REST project.

BALL-BERRY VS JARVIS APPROACHES TO MODEL STOMATAL OZONE DEPOSITION IN MEDITERRANEAN FORESTS: VALIDATION USING THE TEST SITES OF CASTELPORZIANO AND SAN ROSSORE FORESTS

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Models to predict stomatal uptake require a proper parameterization. Field sites where ozone fluxes are measured continuously with Eddy Covariance are ideal to extract fundamental information on stomatal ozone fluxes to validate models. In this study, we collected ozone flux measurements from two representative Mediterranean forest ecosystems in Italy: A Holm Oak forest (*Quercus ilex*) located inside the Presidential Estate of Castelporziano, Rome, and a stone pine (*Pinus pinea*) forest in San Rossore, Pisa. Measurements were carried out during all seasons in order to capture ecophysiological responses to seasonal changes in meteorology and stomatal fluxes were calculated using the Monteith evaporative/resistive methods.

Two modelling approaches were used: the first is the empirical Jarvis approach based on multiplicative algorithm. We used the most recent literature to parameterize reducing functions in the algorithm reported in the UNECE Mapping Manual (release 2010). A second and process-based approach involved use of Ball-Berry equation to estimate stomatal conductance as a function of carbon assimilation, testing the equation with classical parameterization retrieved from literature and with data directly measured in the field using the eddy covariance technique. Results show that the Jarvis approach better predicts absolute magnitude of stomatal ozone fluxes, while the Ball-Berry approach better reproduced the dynamics of stomatal ozone fluxes during the day but it diverged from the measured fluxes. Current research is aimed at refining parameterization of the Ball-Berry approach in order to obtain a good agreement with measurements.

YIELD RESPONSE OF SOME ITALIAN AND SPANISH CULTIVARS OF DURUM WHEAT TO ELEVATED OZONE: A VARIETAL SCREENING

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Durum wheat is the main wheat species grown in Southern Europe countries. Hence, the evaluation of the response of the main cultivars of durum wheat to tropospheric ozone is a priority for Southern Europe. The present work investigated the O_3 effects on five cultivars of durum wheat, three of them from Italy (*Colombo, Sculptur, Faraon*) and two from Spain (*Gallareta* and *Vitron*) in terms of yield and related physiological drivers.
The experiment was performed in the open-top chambers (OTC) facility of C.R.IN.E.S at Curno, Italy, from 09th March to 15th June 2013. Each *cv* was sown in 12 pots (three plants per pot) and pots were randomly assigned to 4 OTC (three pots per OTC for each *cv*), two of them supplied with charcoal filtered air (-50% of ambient air ozone = CF) and two with ozone enriched air (+50% of ambient air ozone = EN). During the experiment the ozonated plants were exposed to a daylight AOT40 value of 11.561 ppm•h, well above the UN/ECE critical level of 3 ppmh, while the control (CF) plants experienced an exposure of 0.432 ppm•h.
Periodical measurements of stomatal conductance, chlorophyll-a fluorescence and growth were performed during the growing season. Furthermore, macroscopic (chlorotic/necrotic spots) and microscopic (cell death and H_2O_2 deposits in mesophyll tissue) symptoms development were followed on the flag leaves. At the end of the season plants were harvested and the stems and the ears of each plant were separately dried and weighted. Then the grain yield was assessed per each OTC as total grain weight and hectoliter weight.
The Italian *cv Sculptur*, the most productive one without ozone, together with the *cv Colombo* resulted the most ozone sensitive *cvs*, with a grain yield decrease of 16% ($p<0.05$) and 10% (ns) respectively in the EN treatment. Nevertheless very small differences were observed in the hectoliter weight between the two ozone treatments. The remaining *cvs* did not show any negative effect of ozone but rather a slight yield stimulation (between 1 and 5%, ns). The Spanish *Gallareta* resulted the best performing *cv* when both CF and EN yield are considered.
Ozone affected the earing and the plants growth: the number of ears was quite uniformly reduced by ozone, with a remarkable low for *Sculptur*, and the losses of the aboveground biomass confirm the ozone sensitivity of *Sculpur* and *Colombo* (-20% and -25% respectively, both $p<0.01$).
The stomatal conductance was greatly decreased by ozone in all *cvs* (between 11% and 37%, $p<0.01$), but it is notable that the most sensitive *cvs* showed the highest *gs* values, even under the ozone treatment, which implies a highest ozone dose. This would confirm that the stomatal dose is an important driver of the plants' response to this pollutant.
On the contrary the overall performance of the photosynthesis (PI) - assessed by the OJIP test on the chlorophyll fluorescence measurements – was not affected by ozone. Thus, the observed reduction of productivity in the sensitive *cvs* is not to be ascribed to a damage of the photosystyems but rather to a dissipation of the available energy for the metabolic defense against the oxidative stress.
Finally, the observation of the the *Sculpure cv* behaviour suggests that the breeding strategy of selecting a greater gs to increase the yield may not be appropriate under increasing ozone conditions.

FURTHER DEVELOPMENTS OF THE CONCEPT OF THE SIMPLIFIED PHYTOTOXIC OZONE DOSE, S-POD, TO ASSESS THE RISK FOR OZONE IMPACTS ON TREES ON THE REGIONAL SCALE

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A new concept was developed, based on the "Simplified Phytotoxic Ozone Dose", S-POD. The exposure index in intended to be used to assess ozone impacts on vegetation on the regional scale in northern Europe and it should be possible to estimate the ozone exposure based on relatively few, low cost, in situ measurements.
The S-POD is estimated from measured hourly ozone concentrations according to:
$S\text{-}POD_x$; ∑daylight (($[O_3]$ * f) >x); f = max [f_{min},(f_{phen} * f_{temp} * f_{VPD} * f_{gmax}).
The functions used are as far as possible similar to what is stated in the LRTAP Mapping Manual. The f_{gmax} is introduced as a possibility to normalize between different plant species based on the combination of g_{max} and g_b. The S-POD concept was tested on experimental data from Sweden and Finland on young Norway spruce and Silver birch and compared with POD and AOT40. The strongest dose-response relationships (D-R) were still obtained with POD (with a threshold >2). However, the D-R derived from S-POD was almost as good as for POD and far better than for AOT40. It is concluded that the S-POD concept is well suited to be used e.g. as an indicator in the Swedish Environmental Quality Objectives.

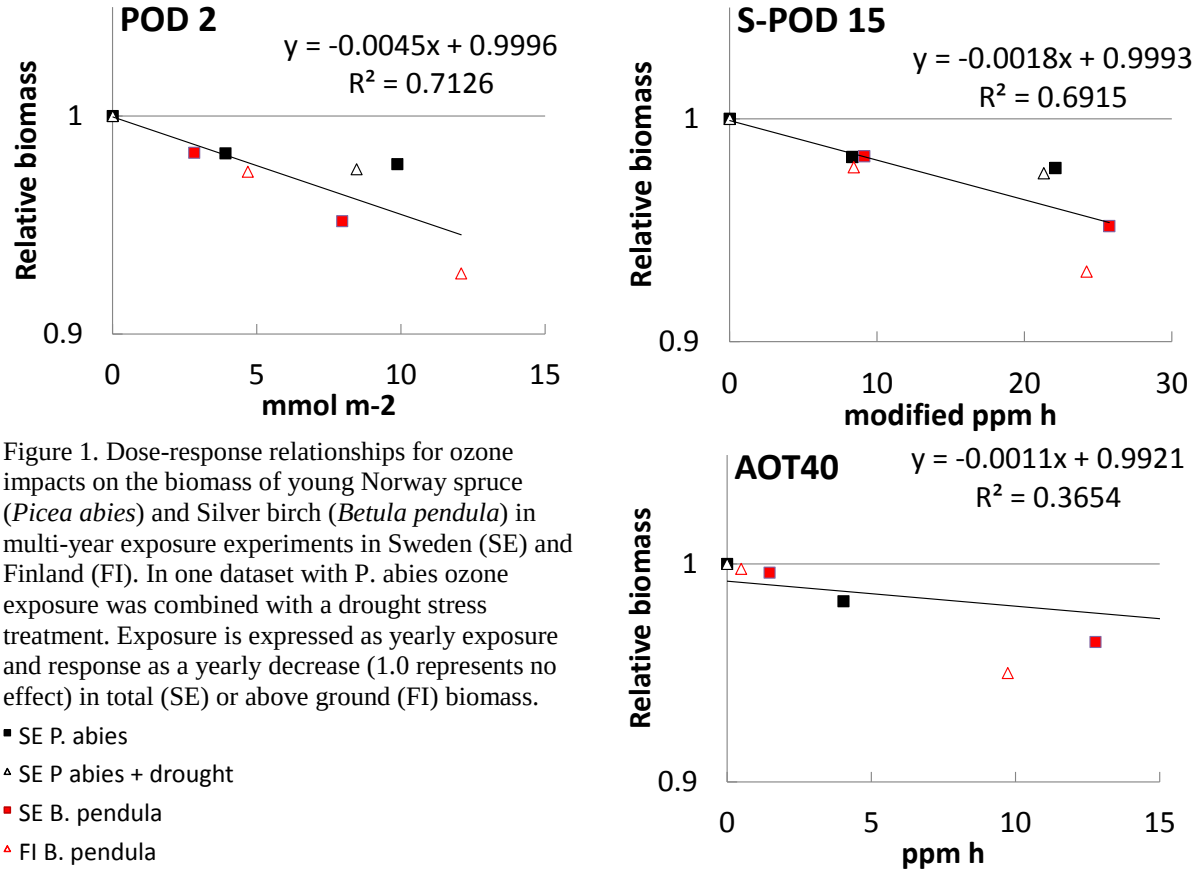

Figure 1. Dose-response relationships for ozone impacts on the biomass of young Norway spruce (*Picea abies*) and Silver birch (*Betula pendula*) in multi-year exposure experiments in Sweden (SE) and Finland (FI). In one dataset with P. abies ozone exposure was combined with a drought stress treatment. Exposure is expressed as yearly exposure and response as a yearly decrease (1.0 represents no effect) in total (SE) or above ground (FI) biomass.

- SE P. abies
- SE P abies + drought
- SE B. pendula
- FI B. pendula

APPLICATION OF A GENERIC CROP FLUX-EFFECT RELATIONSHIP IN INTEGRATED ASSESSMENT MODELLING IN EUROPE

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The full parameterisation of the DO_3SE flux model for ozone is data intensive, including a requirement for hourly resolution climate and soil moisture data on a 50 x 50 km (or smaller) grid. At the request of integrated assessment modellers within the LRTAP Convention, participants at the most recent critical levels workshop (Ispra, October, 2009) and follow-on ICP Vegetation Task Force Meeting in Tervuren (Belgium, January, 2010) discussed a simplified modelling approach for a generic crop and tree species that is suitable for application in scenario analysis within GAINS. Parameterisations were derived that excluded soil moisture, a particularly problematic variable for modelling, and were included in the LRTAP Convention's Modelling and Mapping Manual (LRTAP Convention, 2010). Guidance was provided that these models should be used to identify areas at risk of damage rather than to quantify the economic impacts of such damage.

More recently, the Task Force on Integrated Assessment Modelling have requested a mechanism for quantifying the extent of risk within GAINS using a dose-response relationship for the generic flux models. Hakan Pleijel and Helena Dannielsson re-visited the wheat database used to derive the full flux model and have calculated a yield-effect relationship for the generic crop flux model which is based on wheat. This relationship (Figure 1a) can be applied to assess risk to crops on the assumption that soil moisture is not limiting. To meet the needs of the LRTAP Convention to quantify effects and benefits for the various scenarios discussed during the recent revision of the Gothenburg Protocol, the response function was applied spatially to the wheat-growing areas of EU27+CH+NO. The larger countries with the highest potential percentage effects were Hungary, Italy and Poland, with the mean percentage yield loss for EU27+CH+NO being 12.4% for GP2005, the baseline scenario for 2005 and 10.3% for GP CLE2020, the Gothenburg Protocol current legislation scenario for 2020.

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).

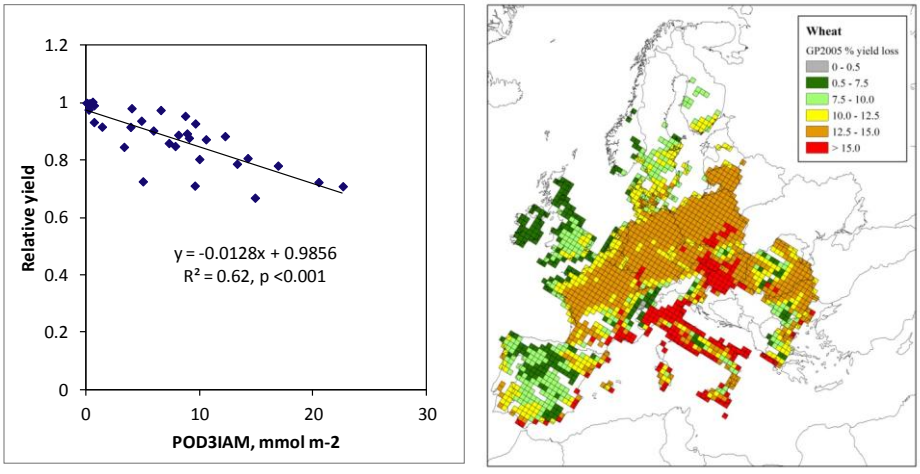

Figure 1(a) New response function for integrated assessment modelling based on wheat and accumulated over 45 days; (b) Percentage yield loss for GP2005 for wheat-growing grid squares, calculated using the equation in (a).

MAPPING OF OZONE FLUX IN SWITZERLAND WITH HIGH SPATIAL RESOLUTION FOR EPIDEMIOLOGICAL ANALYSIS

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This study aims for maps of ozone flux that (1) can be used for epidemiological analysis of ozone effects on tree growth, i.e. have a sufficient spatial resolution (250 m raster) to reflect local meteorological conditions and pollutant levels, (2) cover the whole country allowing for national assessments of ozone-effects and (3) show possibilities of simplifying the flux calculation in order to reduce data demand and to make it more manageable in the practice of air pollution control.

In a first step, the DO_3SE model was run at 24 rural monitoring stations in the period 1991–2006 and at 37 stations in the period 2007–2011 to calculate ozone flux doses POD1 for beech and spruce. The required hourly input-data (ozone and climatic parameters) were measured on-site. Medium water storage capacity was used uniformly for all calculations as flux differences due to soil water variation were small.

For producing maps two approaches are pursued. In the first approach, the fluxes calculated at the monitoring stations were spatially interpolated: The average fluxes of each period were related to maps of predicting parameters (long-term ozone concentration, global radiation and wind speed) by regression analysis. The mean stomatal flux in the resulting map for deciduous forest was 20.5, for the coniferous forest 27.8 mmol m^{-2} $year^{-1}$. The area-weighted average growth reduction for all forests was 11% (Figure 1). In a further step, provisional yearly POD1 maps were produced by spatially interpolating POD ratios (POD1 of the specific year divided by the average POD1) of the monitoring stations using an inverse-distance weighting (IDW) method. The yearly fluxes were successfully used for provisional epidemiological analysis of stem increment data.

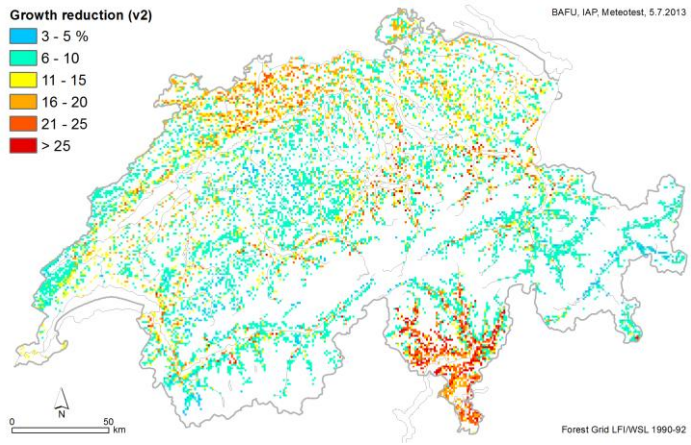

Figure 1: Growth reduction, average 1991-2011.

A drawback of that approach is that the yearly flux maps attained by the IDW method do not include further local information. Therefore, existing monthly meteorological maps were included in the second mapping approach: The calculated hourly stomatal functions (for temperature, VPD and light) and fluxes at the monitoring stations as well as the hourly input data were aggregated to monthly values. The relation between monthly averages of the stomatal functions and the input parameters was analyzed using nonlinear regression. The analysis showed that the fluxes can be estimated based on monthly means of radiation, temperature, dew point and ozone concentrations. The monthly ozone maps are produced by geo-statistical interpolation of monitoring data.

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

TWO-YEAR TOTAL, STOMATAL, CUTICULAR, AND SOIL OZONE BUDGETS OF AN AGRICULTURAL FIELD WITH WINTER-WHEAT AND MAIZE CROPS NEAR PARIS, FRANCE

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In this study, we evaluated ozone (O_3) deposition to an agricultural field with winter-wheat and maize crops over a period of two years. The Surfatm-O_3 model (Personne et al., 2009, Stella et al., 2011), a two-layer soil-vegetation-atmosphere-transfer model, was used to partition the O_3 flux between the soil, the cuticular and the stomatal pathways.

The comparison between measured and modeled O_3 fluxes exhibited a good agreement, independently of the canopy structure and coverage and the climatic conditions. This statement implicitly validates the O_3 flux partitioning. Indeed, sensitivity analysis using other parameterization for soil and cuticular resistances induced underestimations of the total O_3 deposition ranging between 0% to around 20%. Therefore, the Surfatm-O_3 model is used to establish the total, soil, cuticular and stomatal O_3 budgets.

Over the two-year period, total ecosystem O_3 deposition was 87.5 kg ha^{-1}. Non-stomatal deposition, i.e., soil and cuticular pathways, dominated the deposition budget. In particular, soil deposition accounted for up to 50% of the total deposition. However, phenological and physiological differences between maize and winter-wheat induced large differences in the stomatal deposition budgets of these two crops: while it represented 5.4 kg ha^{-1} (22% of the total deposition) for the maize crop, stomatal sink accounted for 10.5 kg ha^{-1} (36% of the total deposition) for the winter-wheat from sowing to harvest.

Finally, crop yield losses were estimated for winter-wheat and maize crops using the AOT40 and POD6 approaches. According to these methods, crop yield loss was 1.5-4.2% for the winter-wheat whereas maize was not affected by O_3.

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Abstracts

Oral

Presentations

Task Force meeting

EFFECTS OF NITROGEN DEPOSITION ON SPECIES RICHNESS AND DIVERSITY IN SWITZERLAND

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Relationships between atmospheric nitrogen deposition and biodiversity monitored at the habitat (plots of 10 m^2) and landscape (plots of 1 km^2) level in Switzerland were analysed on the basis of vascular plants and bryophytes data from the Swiss Biodiversity Monitoring (BDM, www.biodiversitymonitoring.ch) and nitrogen deposition data at a high spatial resolution. Higher mean nutrient values (according to Landolt et al. 2010) of recorded plant species at the habitat level are observed with increasing nitrogen deposition in alpine pastures, mountain habitats, forests, meadows and pastures. In mountain hay meadows (EUNIS code E2.3) the analysis showed a decrease of oligotrophic vascular plant and bryophyte species richness with increasing nitrogen deposition and a decrease of community uniqueness of vascular plant species (Roth et al. 2013). Increasing nitrogen deposition was also negatively related to species richness of target plant species for the agricultural sector. On the other hand an increase in species richness and a decrease in community uniqueness was detected for eutrophic bryophyte species with increasing nitrogen deposition. Overall the results indicate a trend towards floristic homogenization with increasing nitrogen deposition mainly due to the decrease and loss of oligotrophic plant species. On the basis of the results the Critical Load for Nitrogen for mountain hay meadows should not be set higher than 10-15 kg N $ha^{-1}yr^{-1}$. First results from investigations at the landscape level indicate as well a decrease of the species richness of oligotrophic plant species with increasing nitrogen deposition.

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AMBIENT AIR OZONE IMPACT ON GROWTH, YIELD AND NUTRITIONAL QUALITY OF WHEAT, MAIZE AND MUNG BEAN AT TWO SITES OF FAISALABAD-PAKISTAN

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Ground level ozone (O_3) is the most important atmospheric pollutant that likely to become a threat to global food production by damaging crops because of its high level of toxicity. Ambient O_3 concentrations are reducing the quality and total production of crops in many countries worldwide including Pakistan, East Asia, India and Europe. Rolling industrialization and economic growth over much of Asia has resulted in increased level of O_3 emission precursor pollutants and consequently elevated concentrations of O_3. The concentration of ambient O_3 is high enough in Faisalabad city to pose serious threat to the agricultural crop growth and yield.

A project was designed to study the impact of ambient air ozone on the yield and quality of wheat, maize and mung bean at two sites of Faisalabad-Pakistan. Suitable varieties were selected and sown in pots at research area of Ayub Agricultural Research Institute, Faisalabad. Half of the pots were exposed to elevated ambient ozone levels at GTS Chowk near Faisalabad Railway Station (referred to as high ozone treatment), while the other half were kept at Farm Area of Ayub Agricultural Research Institute, Faisalabad (referred as control). During the whole exposure period, ambient air O_3 was recorded. Data regarding O_3 visible injury, stomatal conductance, photosynthetic rate, and total chlorophyll contents were collected at different stages of crop growth. At the time of harvest, the data of plant yield and its attributes were recorded for further analyses.

Results revealed that exposure of plants of wheat, maize and mung bean plants at elevated pollution site caused significant decreases in stomatal conductance, photosynthetic rate, total chlorophyll content and yield of these crops. The grain protein content of these crops showed significant reduction in response to elevated O_3 exposure. These results suggest that O_3 could be a hidden threat to global food security.

Keywords: ozone pollution, food security, mung bean, wheat

VALIDITY OF USUAL DOSE-RESPONSE FUNCTIONS IN THE CASE OF WHEAT EXPOSED TO FREE-AIR FUMIGATON IN THE PARIS REGION

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Two wheat varieties (Premio and Soissons) were grown in the field in the Paris region and exposed to moderate levels of ozone through a 50m long linear free-air fumigation device. Plants were ozone-fumigated daily for 56 days (from 12 May to 6 July 2009). O_3 exposures at 3 different distances from the fumigation source: 2 m, 3.5 m and 5 m downwind of the source, were compared to control located 15m upwind of the linear source. Hourly average concentrations and cumulative exposure index (AOT40) were calculated. Stomatal conductance measurements and modelling allowed the calculation of the Phytotoxic Ozone Dose (POD06) absorbed by the upper leaves of the canopy.

The results show strong correlations between grain yield and both ozone exposure (AOT40 or POD06) indicators. However, there are differences in the dose-response relationship obtained for each variety. Moreover, these relationships are also quite different from the dose-response relationships found in the literature.

This can be partly explained by the fact that the use of free-air fumigation systems does not allow to achieve a true control treatment (plants exposed to ozone-free air). The highest production values that could be achieved in an atmosphere without ozone must therefore be extrapolated from experimental data, and this may increase the inaccuracy of calculations of relative yield losses.

RESPONSE OF *QUERCUS CERRIS* TO COMBINED OZONE AND DROUGHT STRESS

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Urban plants can help to improve air quality, and consequently human health. Due to the negative future prospects in 2050 for the urban environment caused by global climatic change [such as elevated ozone (O_3) concentration and drought]. There is a need to monitor and proactively manage urban forests, adjusting them to change and using them to help cities to adapt to change. Special attention must be paid to the species that populate environments characterized by above-optimal temperatures in summer, such as the Mediterranean ones. Under field conditions, the study of the response to environmental stresses is difficult because environmental factors can be synergistically or antagonistically modified. For these reasons, the aim of this work is the analysis of the impact of a combined stress (O_3 and drought) to simulate under controlled conditions the impact of a 2050 environmental scenario on the physiological performance of the Mediterranean *Quercus cerris*. Profiles related to leaf gas exchange, chlorophyll *a* fluorescence and leaf water potential were analyzed in 2-years old *Q. cerris* saplings exposed to O_3 (80-100 ppb, 4 h day^{-1} for 12 consecutive weeks), to drought (daily irrigated with 30% of effective evapotranspiration) and to O_3 combined with drought in order to improve the response to oxidative stress. At the end of the exposure, plants do not exhibit any foliar symptoms. Photosynthetic processes, however, were significantly changed. Since the first week of treatment, oxidative stress induced decrease in net photosynthesis (A), above all in drought-stressed plants (about 2-fold), in comparison to controls. This trend unvaried until the end of the exposure due to a reduction of the stomatal conductance (g_s). g_s was lower in drought and combined-stressed plants during the whole exposure period (until a minimum about 5-fold after 5 weeks when compared to controls). In O_3-treated plants, intermediate A [8 *vs* 3-11 (other three thesis' range) µmol CO_2 m^{-2} s^{-1}] and g_s [0.10 *vs* 0.07-0.22 (other three thesis' range) mol H_2O m^{-2} s^{-1}, after 4 weeks] values were observed in comparison to the other three thesis explained by a gas avoidance mechanism activated by a stomata regulation causing a medium decrease of the net photosynthesis. Combined stress-treated plants did not show significant changes in comparison to drought stressed individuals in terms of photosynthetic status. Although intercellular CO_2 concentration (C_i) significantly changed after every treatment. Although the variable and maximal fluorescence ratio (F_v/F_m) did not show significant change during the exposure, all the plants showed values inside the optimal range. A regulatory adjustment of photosynthetic processes was highlighted during the exposure by the higher values of no-photochemical quenching (qNP) of all plants in comparison to controls and therefore suggests a tendency to increase the efficiency of thermal energy dissipation within PSII. Measurements of predawn leaf water potential revealed a leaf drought stress (around -2.0 *vs* -0.5 MPa in controls) in plants grown under water deficit conditions especially in combined stress plants suggesting a synergistic effect on the hydric status. On this basis drought stress (single and combined) should be considered more harmful to *Q. cerris* than O_3.

Acknowledgement
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BIOMONITORING OG TRACE ELEMENTS IN IVANOVO REGION, CENTRAL RUSSIA

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The results on biomonitoring of atmospheric deposition of trace elements in Ivanovo Region, Central Russia, in moss survey 2010/2011 are reported. An attempt to assess air quality in the large cities of the Ivanovo Region and wildlife preserve "Klyazminskiy" was made. A total of 47 elements were determined in 96 moss samples, collected evenly over the study area, by two complementary analytical techniques, neutron activation analysis performed at JINR, Dubna, and atomic absorption spectrometry in ISUCT, Ivanovo. GIS distribution maps of element-pollutants and some statistical data treatment are presented.

In autumn 2013 the Ivanovo State University of Chemistry and Technology initiated the development of the environmental protection strategy for the Ivanovo Region till 2020. It includes creation of an open-access database comprising data both from the state monitoring system and scientific research based on biomonitoring. Such policy will result in reliable assessment of air quality and will allow conversion of trace element concentrations in moss to absolute deposition values for trace elements from precipitation collected near the same moss sampling sites. Thus, interpreting data for use in baseline human health *risk assessments* will be possible.

A POSSIBLE ROLE OF HEAT SHOCK PROTEINS AS SENSORS OF HEAVY METAL POLLUTION IN BRYOPHYTA

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Bio-monitoring represents a highly precise and low-cost strategy method(s) to achieve rapid and affordable results in the examining of the enrichment of pollutants in the environment; in this aim, Bryophyta show a high capability to accumulate metals because of the high surface/volume ratio and the presence of a thin cuticle. Bryophytes do not have roots, and their rhizoids do not primarily contribute to the uptake of substances; therefore, most of the elements measured in mosses are due to atmospheric depositions: thus, elements concentrations in epiphytic bryophytes reflect the amount of atmospheric depositions, and mosses are used as air pollution indicator. Therefore, different Bryophyta are currently studied in relation to uptake, and subcellular localisation, of heavy metals.
Heat Shock Proteins 70 (Hsp70) represent a group of chaperones involved in protein folding, but it has been assessed that they play a central role in counteracting the toxic effects of heavy metals on proteins and enzymes, protecting them from misfolding and proteolysis. Even if the understanding of roles and functions of Hsp70 are yet to be completely described, recent studies evidenced their role(s) in the response to abiotic stress.
The aim of this work is to indicate Hsp70 as simple and powerful indicators of heavy metal pollution in Briophyta and other aquatic plants. The subcellular localisation of pollutants can be investigated using X- ray microanalysis, associate to transmission electron microscopy (TEM). The damages caused by pollution were observed using TEM and SEM microscopy, in specimens collected in undisturbed and polluted areas, and compared to changes observed in HSP70 levels and occurrence measured using western blotting. The results were compared and related with the effects observed after *in vitro* exposition of specimens to different heavy metals (Cd, Pb, Cu, Zn).
The results will be discussed in the aim to indicate Hsp70 as a possible bioindicator able to measure the biological effects of atmospheric pollution in plants.

MOSS BIOMONITORING OF TRACE ELEMENTS AND RADIONUCLIDES IN RURAL AND URBAN AREAS EXPERIENCING ENVIRONMENTAL STRESS

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The application of mosses as biomonitors of trace elements and radionuclides in selected rural and urban areas affected by intense anthropogenic activity is reviewed. These include the western part of the Kola Peninsula (NW Russia), the most heavily industrialized area in the entire Arctic, and the South Urals (Karabash) considered amongst the most polluted areas in the world where human impact on the environment is largely irreversible. The major pollutants are heavy metals and long-lived radionuclides from full-scale activities and accidents at the radiochemical "Mayak" Production Association (PA). Moss was successfully used to study spatial deposition of ^{137}Cs and ^{210}Pb in Belarus and Slovakia 20 years after the Chernobyl accident. A combination of analytical data (NAA and AAS in our case) with principle component analysis and GIS technologies allowed pollution source characterization and apportioning in the sampled areas around (1) copper mines in Karabash (RF), Bor (Serbia), South of Poland; (2) Zn-Pb smelters in Baia Mare (Romanai), Krdjali (Bulgaria) and Veles (Macedonia); (3) Fe-V plant in Tula (RF); (4) Fe-Cr industry in Tikhvin (RF) and Mo-i-Rana (Norway); (5) oil refinery industry in Yaroslavl (RF), and (6) thermal power plant in a Moscow district. Besides passive (terrestrial) moss biomonitoring, active moss biomonitoring (moss bags technique) proved effective in biomonitoring air pollution in large cities (street canyons of Belgrade and Moscow). The results obtained at a local scale in the areas experiencing environmental stress can be used to establish emission levels of pollutants and to provide information for health-related institutions.

STOMATAL UPTAKE AND NON-STOMATAL OZONE REMOVAL BY A MIXED OAK-HORNBEAM MATURE FOREST IN THE PO VALLEY: RESULTS OF THE ECLAIRE LONG-TERM CAMPAIGN

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In the framework of the ÉCLAIRE Project of the 7th EU framework programme a joint field campaign and a following long-term one had been conducted at an Oak-Hornbeam mixed mature forest in the Po Valley (Bosco Fontana, Mantova, Italy). Inside the forest, a 42 m tall micrometeorological tower was installed to measure the mass (O_3, CO_2, H_2O) and energy exchange between the ecosystem and the atmosphere. The focus of this work will be on the ozone flux measurements which started in June 2012 and are currently running.
The ecosystem behaved as a relevant sink for tropospheric ozone, both in summer and in winter, with an average monthly ozone removal of 5.8 kg ha^{-1} and a total annual deposition of more than 70 kg ha^{-1}. The stomatal flux, estimated by deriving the bulk stomatal resistance through the inversion of the Monteith equation. The maximum stomatal fraction was observed in July 2013 (55%) and the minimum in October 2012 (13%). A strong interannual variability about the stomatal deposition between summer 2012 and 2013 was observed. The phytotoxic ozone doses taken up by the plants were respectively 13.5 mmol m^{-2} in 2012 and 29 mmol m^{-2} in 2013.
The non-stomatal deposition, obtained as a residual between the total and the stomatal ozone fluxes, was investigated too. In the joint field campaign Neimitz et al. (2013) found that half of the non-stomatal deposition, was due to NO soil emission, which were on average above 100 mg N m^{-2} s^{-1}. A minor part of the non-stomatal fluxes (6%) was attributed to isoprene reactions with ozone. A large part of the non-stomatal deposition (44%) was not explained. An investigation on this part, with particular attention on the deposition on non-transpiring surfaces, will be showed.
Additional measurements of O_3, NO and NO_2 concentrations along the tower profile, allowed to estimate the ozone storage inside the trunk space. The ozone storage can be seen as a part of the deposition flux which is temporarily stored into the trunk space. Taking it into account, the daily course of the non-stomatal deposition significantly changed, with important consequences for non-stomatal interpretation and for proper modelling of the non-stomatal processes.
An estimation of the ozone impact on the whole ecosystem was assessed by looking at the departure from the carbon dioxide balance closure. First results of CO_2 exchange seem to indicate a net emission of CO_2 from the ecosystem, as it should not be expected from a healthy mature forest ecosystem where, typically, the carbon balance is zero. Again, a general limitation of the net carbon uptake has been observed at high ozone stomatal fluxes.
Finally, the great differences observed in the two years highlight the need for long time series of flux measurements. Moreover, this kind of studies will help in raising the awareness of the policy makers on the ecosystem services offered by the vegetal ecosystems.

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OVERVIEW OF THE ACHIEVEMENTS OF THE ICP VEGETATION IN 2013 AND FUTURE WORKPLAN (2014 – 2016)

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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. The ICP Vegetation also studies the impacts of pollutant mixtures (e.g. ozone and nitrogen), consequences for ecosystem services and biodiversity, and interactions between air pollutants and climate change. When possible, economic evaluations of the impacts of ozone are included.

At the 27th Task Force Meeting we will report on the achievements of the ICP Vegetation in 2013 [1], including:

- Impacts of ozone pollution on ecosystem services and biodiversity [2];
- Outcome of the European heavy metals and nitrogen in mosses survey 2010/11 [3] and the pilot study on POPs;

In addition, we will report on the contribution of ICP Vegetation to the common work plan items of the WGE for 2013, such as:

- Report [4] and brochure [5] on benefits of air pollution control for biodiversity and ecosystem services;
- Guidance document on health and environmental improvements [6].

We will also discuss the future workplan, including progress made with items to be reported to the LRTAP Convention in 2014 [7], such as:

- Report on air pollution deposition to, and impacts on vegetation, in EECCA/SEE countries and South-East Asia;
- Update chapter 3 of the Modelling and Mapping Manual of the LRTAP Convention.

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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[7] http://www.unece.org/fileadmin/DAM/env/documents/2013/air/eb/ECE_EB.AIR_2013_6_E.pdf

RELATIONSHIP BETWEEN SITE-SPECIFIC NITROGEN CONCENTRATIONS IN MOSSES AND BULK ATMOSPHERIC NITROGEN DEPOSITION

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To assess the relationship between nitrogen concentrations in mosses and bulk nitrogen deposition or concentrations in precipitation, both moss tissue and precipitation were sampled within a distance of 1 km of each other at sites in seven countries across Europe. As previously described for modelled nitrogen deposition [1], the relationship between nitrogen concentration in mosses and measured (total) bulk deposition or concentration in precipitation across Europe is best described by an asymptotic relationship. The asymptotic relationship is much stronger for ammonia-N than for nitrate-N in bulk deposition or precipitation, with high ammonium deposition rates at German sites being most influential in providing evidence of the asymptotic behaviour. Saturation appears to occur at bulk nitrogen deposition rates of ca. 15 – 20 kg ha^{-1} y^{-1}. Up to such deposition rates, linear relationships have been observed in some countries (Finland and Switzerland) but not in others. Considerable scatter was observed in the relationship at the European level, although less than previously found with modelled deposition [1]. Nitrogen concentration in mosses can be applied to identify areas at risk of high nitrogen deposition at the European scale.

Acknowledgement
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VACUOLAR ENDOPROTEOLYTIC ACTIVITIES AND PROTEIN CARBONYLATION LEVELS IN LEAF AS A MODEL FOR DEPICTING OZONE TOLERANCE AND SENSITIVITY IN WINTER WHEAT (*TRITICUM AESTIVUM L.*)

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Current global background concentrations in tropospheric ozone (O_3) have been shown to have significant adverse effects on crop yield, including wheat. As selection of O_3 resistant cultivars (cvs) could help reduce O_3-induced agricultural losses (Avnery et al., 2013), we investigated whether protein oxidation (carbonylation) and vacuolar endoproteolytic activities measured in the flag leaf could differentiate between ozone sensitive and tolerant wheat cvs. Two winter wheat (*Triticum aestivum* L.) cvs released in 1986 (Soissons) and 2006 (Premio) were grown in the field and exposed to ambient and semi-controlled chronic O_3 concentrations, from pre-anthesis to harvest, using a new linear O_3 fumigation device that generates gradients of the pollutant. Determination of resistance to O_3 from grain yield and quality showed that Soissons was the most sensitive cv. For both cvs, an important decline in total chlorophyll contents was observed in response to O_3 suggesting it induced a premature development of leaf senescence, causing the shortening of the assimilation and grain-filling periods. O_3 induced an increase in protein carbonyl groups (PCO) for both cvs. However, Soissons was characterized by a higher intrinsic level of protein carbonyls. Increased PCO levels and losses in total chlorophyll contents were concurrent, suggesting a link between the extent of oxidative stress and senescence development. Consistent with a premature induction of leaf senescence, O_3 induced a stimulation of endoproteolytic activities that resulted mostly from increases in cysteine protease (CP) activities, for both cvs. However, CP stimulation was more important in the resistant cv. Increased endoproteolytic activities were associated with increased PCO and with a decline in total soluble protein contents. In conclusion, intrinsic PCO levels and induction of CP in response to O_3 could help distinguish between resistant and sensitive wheat cvs to O_3.

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IMPACTS OF OZONE AND NITROGEN ON SILVER BIRCH

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Birch trees (*Betula pendula*) were exposed to factorial combinations of seven ozone and four nitrogen regimes for five months in solardomes at CEH Bangor in 2012 and 2013. The ozone regime was designed to investigate the benefits of changes in air quality policy that are anticipated to reduce both background and peak ozone concentrations, but with a larger reduction for the peaks. The range of ozone exposure seasonal means was 35 ppb to 70 ppb (24h mean) and the nitrogen treatments were applied weekly as ammonium nitrate to give treatments equating to 10, 30, 50 and 70 kg ha^{-1} yr^{-1}. Measurements were made at both the leaf level and at whole-tree level to investigate whether:

- nitrogen modifies the response to ozone;
- nitrogen and/or ozone treatment alter the DO_3SE parameterisations for birch;
- fluxes of ozone and carbon become uncoupled;
- whole-tree alterations in fluxes are a consequence of individual leaf physiological responses or via alterations in tree biomass.

The main findings in the second season of ozone exposure where:

- Reduced tree size and leaf number;
- Premature leaf loss
- Tendency to reduced leaf photosynthetic capacity ($V_{c,max}$, J_{max}) later in the season, linked to reduction chlorophyll index due to early leaf die-back.

In the first year of exposure we had shown that whole tree carbon fluxes were reduced with increasing ozone treatment.

Effects of nitrogen included:

- Increased tree size, leaf number and bud length;
- Increased stomatal conductance;
- Increased chlorophyll content and photosynthetic capacity, particularly early in the season.

Potential interactions between ozone and nitrogen were also observed, including:

- Impacts on tree size and woody biomass;
- High nitrogen seemed to delay early leaf die-back at high ozone exposure later in the season, delaying the reduction in photosynthetic capacity due to ozone.

Conclusions

- Ozone pollution decreases growth and therefore carbon sequestration of birch trees;
- Nitrogen treatment affects stomatal fluxes of birch and therefore nitrogen deposition should be accounted for when calculating ozone fluxes to inform assessments of vegetation at risk of ozone pollution;
- The cumulative effects of ozone and nitrogen pollution on trees require further study over several years as leaf-level measurements indicate that effects and interactions may occur over longer timescales.

Acknowledgement

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OVERVIEW OF THE ACTIVITIES OF ICP MODELLING AND MAPPING AND PLANS FOR THE FUTURE

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The Coordination Centre for Effects (CCE) is the programme centre of the ICP Modelling and Mapping (ICP-M&M; www.icpmapping.org) and its network of National Focal Centres developing methods and databases on critical thresholds. These thresholds are used in the **G**reenhouse Gas and **A**ir Pollution **In**teractions and **S**ynergies (**GAINS**)-model in support of abatement policies that aim to protect European ecosystems against adverse effects of atmospheric deposition. In recent protocol negotiations, the focus of effects-based knowledge by the ICP-M&M and ICP-V as used in GAINS, was on geo-chemical modelling of exceedances of critical loads for eutrophication and acidification (Figure 1) and ozone critical levels respectively.

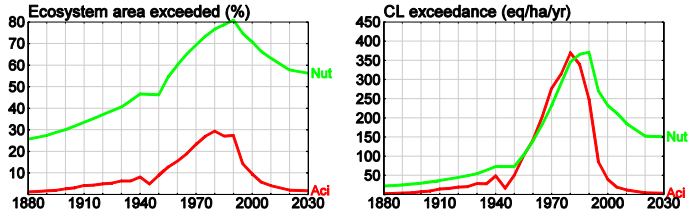

Figure 1: *The temporal development since 1880 of the area at risk (in %, left) and magnitude (in eq ha^{-1}yr^{-1}, right) of exceedance (AAE) of acidification (red) and eutrophication (green), using Gothenburg Protocol – Current Legislation (GP-CLE) scenario depositions from 2010 onwards* (Source: Hettelingh *et al.* EEA technical report XX/2013 *in press*)

Critical thresholds are embedded in the "GAINS-*model"* to interactively optimize emission reduction patterns to regionally diminish adverse ecosystem effects. Alternatively, critical thresholds can be used outside of the model, in a "GAINS-*system"* mode to post-analyse adverse ecosystem effects of air pollution for selected emission scenario-outputs of the GAINS-model. This approach resulted in collaboration between most ICPs to produce values for a range of indicators in two collaborative reports entitled "Guidance document on health and environmental improvements"[1] and "Benefits of Air Pollution control for benefits and ecosystem services"[2]. This collaboration between ICPs in general and with ICP-V in particular is further strengthened under the LRTAP Convention workplan for 2014-2015. Near future work of the ICP-M&M and ICP-V focusses on the identification and application of novel indicators and thresholds within the EU Framework VII project *Effects of Climate Change on Air Pollution and Response Strategies for European Ecosystems* (ECLAIRE: http://www.eclaire-fp7.eu). Vegetation change dynamics are coupled to soil chemistry to model changes of plant species diversity using selected indicators that capture adverse effects of air pollution under climate change. The challenge for the ICP-M&M and ICP-V is to address the trade-offs between (a) the nitrogen induced increase, and ozone induced decrease, of plant productivity in the short term and (b) plant productivity in the short term and the N induced impoverishment of soil chemistry in the long term.

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IS OZONE REDUCING THE N FIXATION CAPACITY OF CURRENT MANAGED PASTURE?

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Clovers (*Trifolium* spp.) are an important component of temperate pasture, providing a source of fixed nitrogen (N) to soil and improving forage quality and productivity. Although known to be highly sensitive to ozone pollution, surprisingly little is known about the effects of ozone on the modern cultivars of clover sewn by farmers for use in intensively grazed pasture. In 2012, we exposed monocultures of white clover (*T. repens* cv. Crusader) and red clover (*T. pratense* cv. Merviot) to a range of ozone profiles for 12 weeks. We found reductions in root biomass and root nodule number in response to ozone in both cultivars, with Crusader also displaying reductions in the size and mass of nodules and N-fixation rates. Applying the AOT40-based response function spatially to the UK, we found that there was potential for a median 9% reduction in clover root nodule mass (mean for all 10 x 10km grid squares containing pasture) in a high ozone spring such as the year 2006.

In 2013, we exposed Crusader, in mixture with a current cultivar of high-sugar ryegrass (*L. perenne* cv. AberMagic), to current and near-future ozone scenarios for a period of 16 weeks. We investigated effects on N-fixation, and associated physiology and growth throughout the growing season. The results are discussed in relation to potential mechanisms and impacts on agricultural sustainability.

DIFFERENCES IN CONCENTRATION OF NITROGEN IN MOSSES DUE TO CANOPY DRIP EFFECTS - CASE STUDY GERMANY

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Background. Atmospheric nitrogen (N) deposition into terrestrial ecosystems is frequently considered as a threat to phyto-diversity. In Germany, N concentrations in mosses were recorded in 2004 as part of a regional investigation at 54 locations in the North-West (Schröder et al. 2007) and in 2005 at 726 locations across the whole territory. Following the methodology presented by Schröder et al. (2013), differences of correlations between N concentrations in atmospheric deposition and in mosses between landscapes across Europe could be corroborated (Schröder et al. 2014). These investigations (Schröder et al. 2007, 2013, 2014) were accomplished by studies focusing on spatial differences of N concentrations in mosses at the site level. ***Methods***. To this end, N concentrations were determined in mosses sampled at sites with and without canopy drip effects in several European states (see *Discussion*), amongst them Germany. This article deals with that study conducted in 2012 comparing N concentrations in mosses sampled within 30 forest stands and in 26 adjacent open fields in Northwestern Germany (Kluge et al. 2013). The N concentrations were converted to N atmospheric deposition values by use of a regression model derived from Schröder et al. (2014). These deposition estimations enabled to calculate N critical loads exceedances. ***Results.*** Compared to the average N concentration in mosses sampled in open fields 2012 (7.4 kg / ha*a), the average N concentrations in mosses within adjacent forests were almost four times higher (26.6 kg / ha*a), and the maximum within the stands accounted for ~ 56 kg / ha*a. Compared to 2005, there was a slight decline of the average N deposition by 2.4 kg / ha*a in open fields. However, the average N concentrations in mosses within forests stands in 2012 remained nearly the same since 2004 (29 kg / ha*a). The atmospheric N deposition as estimated from the N concentration in mosses ranged between the minimum and maximum N critical load at 71 % of the 56 sites investigated. At 14 % of the sites the N deposition was close to the maximum N critical load value which was exceeded in 11%. ***Discussion.*** The results presented could be compared to concentrations of N in mosses sampled at both, canopy drip influenced sites and open areas, in several European countries (Austria, Estonia, France, Finland, Germany, Switzerland, The Netherlands, and Slovenia). In addition, results of a re-sampling in 2013 in Northwestern Germany and of mosses collected at two German ICP Integrated Monitoring and EMEP deposition monitoring sites (Neuglobsow, Bayerischer Wald) could be presented and discussed. ***Conclusions.*** The study at hand revealed statistically significant differences between N concentrations measured in mosses sampled within forests and in open fields (Kluge et al. 2013). The presented findings should be accounted for both modelling and mapping atmospheric N deposition into terrestrial ecosystems on the one hand and related estimations of N critical load exceedances on the other hand.

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THE SURVEY OF ATMOSPHERIC DEPOSITION OF AL, CR, FE, NI, V AND ZN IN ALBANIA BY USING MOSS BIOMONITORING AND ICP-AES

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The atmospheric deposition of Al, Cr, Fe, Ni, V and Zn in Albania was investigated by using carpet-forming-moss species (*Hypnum cupressiforme*) as bioindicators. This research is a part of the international programme (ICP Vegetation Programme, UNECE) carried out in most of European countries since 1987 investigating the impacts of air pollutants on crops and natural vegetation. Sampling was done in dry seasons of autumn 2010 and summer 2011 at 62 sites distributed over Albania. Unwashed, dried samples were totally digested by using microwave digestion and the concentrations of metal elements were determined by ICP-AES. The results reflect local emission points. The elements with high values of concentration compare with other European countries, such as Al, Cr, Fe, Ni, V and Zn, were separately treated in this paper. The median values of chromium (4.75 mg/kg, DW), iron (1618 mg/kg, DW), nickel (5.85 mg/kg, DW), vanadium (3.51 mg/kg, DW), zinc (13.77 mg/kg, DW) and aluminum (6974 mg/kg, DW) are similar to those of neighboring countries, but higher to those of European countries. EWMA and univariate control chart was used to investigate the moving range of successive observations and to estimate the variability of the data.

Certain local emitters were identified like, iron-chromium metallurgy and cement industry, oil refinery, mining industry, and transport. In addition, the natural sources, from the accumulation of these metals in mosses caused by metal-enriched soils, associated with wind blowing soils particularly in south-east direction of the country, was pointed as another possibility of local emitting factors.

Keywords: air pollution, metals; moss biomonitoring; ICP-AES, EWMA, multivariate analysis, Albania.

THE ROLE OF JASMONATE AND SALICYLIC ACID IN CONFERRING TOLERANCE AGAINST OZONE INJURY IN TOMATO PLANTS

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Enhancing stress tolerance in plants has major implications in agriculture. However, a simple method for inducing tolerance in plants without undesirable side effects has not been available till now. Both Salicylic acid (SA) and Jasmonic acid (JA) signaling pathways were found to be activated in response to O_3 exposure. The endogenous accumulation of SA and JA during O_3 – fumigation was reported repeatedly in the literature. In the present study exogenous applications of SA and JA prior to O_3 exposure were used to elucidate their effect in altering the response of tomato plants to O_3-induced oxidative stress, to investigate whether plants exhibiting different levels of O_3–tolerance exhibited different responses to signal transduction; and to test the hypothesis that treatment with SA and JA is able to alter or reduce O_3 injury symptoms. Eight different tomato cultivars were tested for O_3 – sensitivity. Tests were undertaken with different O_3 concentrations (100-150ppb and 150-200ppb) at different stages of vegetative growth (4-5 leaves and 7-8 leaves). Data collected from those screening tests yielded two sensitive cultivars, Strain B and Peto86. There were no significantly tolerant species in the group, rather different degrees of intermediate responses. Chico (intermediate /sensitive), Ace 55 and Castle rock (intermediate/tolerant) were selected on the basis of their consistent responses in the repeated experiments. Electrolyte leakage which is an early event leading to cell death and lesion formation was investigated. Dramatic increases in electrolyte leakage were recorded on both the O_3 – sensitive cultivars (Strain B and Peto86),while significant but less striking enhancement of e-leakage and cell death were the results of O_3 exposure of the intermediate cultivars (Chico, Ace 55 and Castle Rock). Ethylene (ET) emission has been shown to correlate to O_3 sensitivity in several plant species. Induction of ET synthesis in O_3 – sensitive tomato plants occurred within 0.5-2hr after the onset of ozone exposure and reached its peak value at 6h from the start of O_3 fumigation. Data collected 18hr afterwards (24hr sampling) were dramatically lower but remained stable in the subsequent sampling time (48h). The amount of ET produced generally correlated well with the degree of visible lesion formation that developed at least 18-24hr from the end of O_3 exposure. Exogenous SA induced a clear and significant increase in O_3 – sensitivity in all the tomato cultivars studied. Visible symptoms were more severe and spread over larger leaf areas. The increase in foliar symptoms was supported by the enhancement of ET emission from SA – treated plants, especially in O_3 – sensitive cultivars. A general increase in loss of cell integrity and cell death was also observed. On the other hand, pre-treatment with exogenous JA dramatically reduced the magnitude of visible foliar injury caused by O_3 and foliar lesions were significantly attenuated on all the cultivars investigated. Electrolyte leakage decreased significantly pointing to a lower rate of cell death. Cell death was almost completely abolished in all the cultivars except for Strain B, the most sensitive cultivar. ET levels in JA – treated plants declined sharply, but the general ET emission trend was maintained. We concluded that SA is most probably involved in lesion propagation and would thus promote cell death, while JA influences lesion attenuation and containment, therefore, would play a role in enhancing stress tolerance.

INVOLVEMENT OF PLASTIDIAL TERMINAL OXIDASE (PTOX) AND MITOCHONDRIAL PROTEINS (AOX AND PUCP) IN THE RESPONSE OF *VIGNA UNGUICULATA* TO COMBINED DROUGHT STRESS AND OZONE EXPOSURE

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Mitochondria and chloroplasts possess energy-wasteful pathways mediated by proteins (AOX, pUCP) and PTOX, respectively. Alternative electron pathways linked to these enzymes could prevent reactive oxygen species (ROS) formation during stress and alleviate oxidative damage to cell organelles, membranes and proteins. In this paper we assess the expression of these proteins under drought, ozone and their combination (D+O). *VuPTOX*, *VuUCP1b*, *VuAOX1* and, to a lesser extent, *VuAOX2b* showed the strongest responses to the treatments. Two expression patterns were identified: on the short term (3 days), these genes were generally up regulated regardless of the constraint, while on the longer term (14 days), the responses differed according to the treatment. Under ozone exposure, the stimulation of mitochondrial protein expression was maintained, whereas *VuPTOX* expression decreased. Under drought stress, only the plastidial protein (*VuPTOX*) remained up regulated. Our result led us to suggest that under D, to cope with the ROS formation into the chloroplasts, the up-regulation *VuPTOX* and secondarily pUCP, as well as higher antioxidant content was triggered. This early response to cope with drought would protect the plants from the ROS formation by the O_3 fumigation in the D+O plants. Ozone applied alone, generates ROS that would trigger a mitochondrial response leading to an up-regulation of AOX and pUCP genes. But this response, concomitant with low antioxidant content seems to be no efficient to avoid the accumulation of H_2O_2 and the subsequent necrosis formation.

NEW ICP VEGETATION SMART-PHONE APP FOR RECORDING INCIDENCES OF OZONE INJURY ON VEGETATION

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In 2007, the ICP Vegetation published a synthesis report documenting over 500 incidences of ozone injury on crops, grassland species and shrubs growing in the field under ambient air conditions in 17 countries of Europe (Hayes et al., 2007). We plan to revisit this study by compiling new spatial data on current incidences of ozone injury. Using smart-phone technology for i-phones and android phones, and web-based recording methodology we are developing a new way of recording incidences of ozone injury in the field. A new App is nearing completion that will allow participants to upload photographs of ozone injury direct from the field together with the coordinates for the location where the injury was detected (derived from a zoom-able Google map). Participants will be taken through a series of questions designed to assist with quality assurance, including being asked if they have previous experience of identifying ozone damage or plant diseases and recent weather conditions. The App could be used anywhere in the world and thus is suitable for use in the outreach activities as well as the European activities of the ICP Vegetation. The aim is to finalise the App before the forthcoming growing season allowing it to be rolled out to participants in March/April. We would like to encourage as many people as possible to participate in using the App to record ozone injury in 2014 allowing us to fully test the App by developing a large database for analysis.

The web-based recording facility will be available at our webpage: http://icpvegetation.ceh.ac.uk. News of the completion and launch of the web-based and smart-phone App will be provided by email and on the web.

Acknowledgement
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OVERVIEW OF PROGRESS FOR THE EU FP7 ECLAIRE* PROJECT

Mills, G.[1], Sutton, M.[2], and many colleagues from the ECLAIRE project

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Running for just over two years, the EU FP7 project ECLAIRE brings together expertise from many specialists to develop new ways of modelling the risks posed to European ecosystems from the combined effects of air pollution and climate change. This unique combination of scientists covering effects at the leaf scale through to modelling impacts on European ecosystem services and climate, and including experts on ozone, nitrogen, aerosols and climate change met recently in Zagreb, Croatia to discuss progress at the half-way stage of the project. This presentation will provide a brief overview of progress with the project, with particular emphasis on aspects of relevance to ICP Vegetation and policy makers at the LRTAP Convention.

The project is split into five main scientific components: (1) Emissions and Exchange processes; (2) Emissions and exchanges at local to EU and global scales; (3) Ecological response processes and thresholds; (4) Ecological responses at regional and European scales; and (5) Integrated risk assessment and policy tools. Many representatives of bodies of the LRTAP Convention are participating in this project, e.g. ICP Vegetation, CCE (ICP Modelling and Mapping), ICP Forests, Task Force on & Centre for Integrated Assessment Modelling, NEBEI, EMEP/MSC-West, Task Force on Reactive Nitrogen.

ICP Vegetation participants have greatest involvement in component 3, including analysis of data mined from the published literature to derive dose-response relationships for physiological and growth responses, experimental exposure of crops, grassland and tree species to combinations of ozone and nitrogen pollutants, investigations of leaf-scale processes and model development including photosynthesis-based ozone uptake and growth models, MADOC for species changes and JULES (a land-surface climate model). As well as providing an overview, this paper will also include results from the data mining activities showing how cross-species response functions are being derived from a data extraction exercise involving over 300 papers. The following two papers will describe results from tree O_3 and N exposure experiments at Curno, Italy (Gerosa *et al.*) and Bangor, UK (Hayes *et al.*).

* Effects of climate change on air pollution impacts and response strategies for European ecosystems.

Acknowledgement
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CIRCADIAN PROFILES OF PHOTOSYNTHETIC PARAMETERS AND PRIMARY METABOLITES IN GRAPEVINE VARIETIES EXPOSED TO OZONE

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A comparative study on the diurnal courses of photosynthetic gas exchange parameters, chlorophyll *a* fluorescence characteristics and photoprotection mechanisms has been conducted on two *Vitis vinifera* varieties [black-berried cv. Aleatico (ALE) and white-berried cv. Trebbiano giallo (TRE)] exposed to chronic ozone (O_3) fumigation simulating the typical circadian profile of this pollutant in Central Italy. Experiments started in early spring before budbreak. Two-year-old plants grown in five liter pots were acclimated inside a greenhouse ventilated with charcoal filtered air and treated for 28 days with a pulse of O_3 concentration between 8:00 a.m. and 1:00 p.m., with a target concentration of 80 ppb and between 1:00 p.m. and 6:00 p.m., at 40 ppb. At the constitutive level, TRE showed both higher average value of whole leaf thickness (157 ±3.5 TRE, *vs* 139 ±2.3 µm ALE) and higher stomatal conductance to water vapour (G_w) levels (+29% in term to daily values) in comparison to ALE. On the other hand, stomatal density, violaxanthin content and abscisic acid concentration were lower in TRE compared to ALE (143 ±30.8 stomata mm^{-2} TRE, *vs* 249 ±31.8 ALE, -75 and -37%, respectively), suggesting a different stomatal behaviour of the varieties. At the end of the treatment, both varieties showed visible spots (Ø 1-2 mm) of yellowing tissue located in the interveinal area of fully expanded leaves. SEM observations of the abaxial surface of treated samples showed an irregular presence of deformed swelling areas between veins. These modifications were more frequent and developed in ALE. Oxidative stress modified the diurnal pattern of leaf CO_2 assimilation rate (A): in both varieties, A values were significantly reduced during all day and a single-peak was obtained at 4:00 p.m. In ALE, this reduction was mainly due to a reduced mesophyllic functioning, as evidenced by the marked increase in intercellular CO_2 concentration (+33% in terms of daily values). On the contrary, in TRE, insufficient stomatal conductance was found to be the main reason for the alteration of photosynthesis, as confirmed by the decrease of G_w levels (-29% in terms of daily values). In both varieties, the alteration of photosynthetic performance observed at the end of the exposure was accompanied by PSII damage. The decrease of maximum PSII photochemical efficiency (F_v/F_m ratio) recorded in the early afternoon (at 4:00 p.m. and between 2:00 and 4:00 p.m., respectively in ALE and TRE) is indicative of photoinhibition. The reversible change of this parameter found during the day (in both varieties, F_v/F_m levels recovered at the constitutive values in the noon) suggests that photoprotection rather than photodamage occurred. At the end of the exposure, both varieties displayed mechanisms that play a role in dissipating the excess energy, as evidenced by: (*i*) increased content of total carotenoids (+64 and +30% in term to daily values, respectively in ALE and TRE); (*ii*) enhanced efficiency of thermal energy dissipation within PSII (+32 and +20%) that closely correlated with the increased de-epoxidation index (+26 and +22%), and (*iii*) partitioning of carbon to total carbohydrates (+14 and +9%) in comparison to controls. These results indicate a different efficiency of the thermal dissipation mechanisms and photorespiratory pathway among varieties and a major need for carotenoid-mediated photoprotection in ALE.

LATITUDE DEPENDENCE AND TEMPORAL TREND OF THE YEARLY PEAK IN SURFACE OZONE IN MIDDLE AND NORTHERN EUROPE

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The yearly dynamics of the surface ozone concentration [O_3] was studied for 25 monitoring stations ranging from Austria and Switzerland in the south, Ireland in the west to the Arctic Circle in Sweden and Finland. The aim was to investigate the level and timing of the annual peak of [O_3] in relation to latitude and growing season. Further, the change in the timing of the annual O_3 peak was studied, since this may affect risk assessment for O_3. The [O_3] yearly maximum and maximum 8h-mean significantly declined with increasing latitude as did the number of days with 8h-mean [O_3]>120 µg m^{-3}. However, the yearly minimum in [O_3] (lowest running two-week average) correlated positively with latitude, thus being higher in the north. The day number of maximum [O_3] declined strongly and significantly with increasing latitude. This means that that the annual peak in [O_3] is shifted from the summer in the southern part of the region to spring in the north (Figure 1). This is of particular importance in the north, since the yearly O_3 peak today occurs partly before the onset of the growing season. Climate change is likely to lead to an earlier onset of the growing season, for which there is already evidence in the last few decades e.g. in northern Sweden. Thus, the growing season may increasingly overlap with the O_3 peak in the boreal/subarctic region in the future. It has, however, been suggested, that the pronounced spring peak in [O_3] in the north is largely a result of low deposition to snow, ice and vegetation which is not physiologically active and the spring peak would be reduced or occur earlier if the growing season starts earlier. Our study shows that there is currently a time trend for an earlier yearly maximum in [O_3] in the whole region covered by the study with no latitude dependence. Preliminary results indicate that the rate of change in the timing of the yearly [O_3] peak is smaller than that in onset of the growing season. This suggests a development towards increasing risk for O_3 damage to vegetation in northern Scandinavia with respect to phenology and timing of the yearly [O_3] peak, the reasons for which will be further discussed in the presentation.

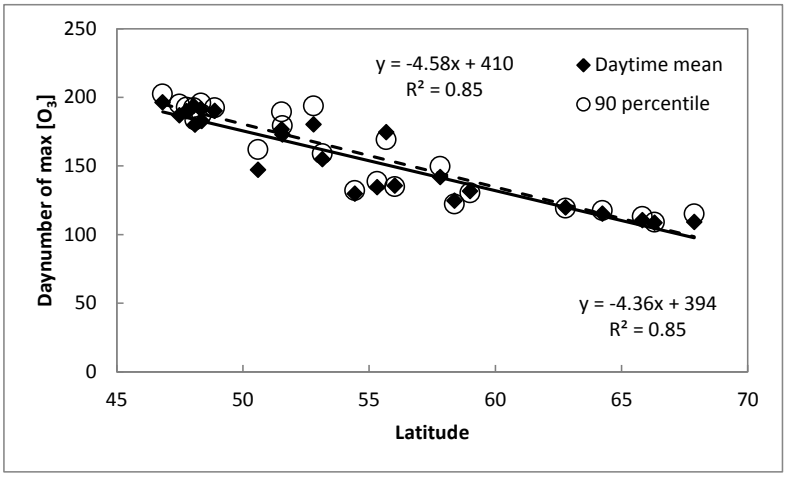

Figure 1. Day number of maximum daytime mean and 90-percentile of the ozone concentration in relation to latitude. For both ozone variables running two-week means were used. Both relationships are strongly significant ($p<0.001$).

ASSESSMENT OF TRACE ELEMENT POLLUTION AROUND VLORA AREA, ALBANIA

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The environmental assessment of air and seawater pollution was carried out on coastals part of Vlora Bay, Albania. The air quality survey was carried out by using the moss Biomonitoring technique, where the moss Hypnum cupressiforme was employed as a long-term biomonitoring system. The urban area of Vlora city was investigated by using the active biomonitoring technique. The moss bags were exposed parallel with ditches for six months at nine sites in Vlora, along the main streets. Moss Biomonitoring at coastal and inland sites was used for investigation of the distribution of heavy metals in the air. The air deposition of mobilized traces metals was investigated by using ICP/AES analysis of moss samples. The seawater quality survey was also carried out from April–May 2011 and in February 2012 and heavy metals content in water samples from 15 sites in Narta and Orikumi lagoons and Vlora Bay were determined.

Heavy metals (Cu, Pb, Zn, Mn, Fe and Cd) in seawater and moss bag samples were determined via Atomic Absorption Spectrometry (flame, electrothermal systems). The area studied is moderately polluted as a result of high vehicular emissions and use of adulterated fuel in vehicles. Locations in the city were categorized on the basis of metal concentrations in the mosses and data statistical treatment. Comparison of exposed and unexposed moss helped assess the factors that adulterated the exposed moss samples. Correlation analysis helped determine the geochemically mobile elements and geochemically bound elements.
The heavy metal concentrations in seawater samples (Cd, Cr, Cu, Fe, Hg, Mn, Ni, Pb and Zn) were quite similar with the values reported for other regions in the European coastal waters.
This study proved that the multivariate analysis is a powerful tool in environmental study. It was found that the Bay receives particulate matter, nutrient and dissolved element inputs from Vjosa and Shushica Rivers, as well from industrial, urban and agricultural activities along this river and the estuary around the Vlora Bay, that caused an increase of the concentration of Fe, Mn and Zn in the vicinity of the delta of the Vjosa River.
Harbors activities, such as dredging and cargo handling, the dumping of ship waste and other coastal activities are reflected on the concentrations of Fe, Cd, Cr, Cu and other metals in seawater of Vlora Bay, that cause anthropogenic pollution of the bay.
Some other anthropogenic factors such as atmospheric sources of dust generated by the erosion of land surfaces, wind transported dusts and by direct emission from anthropogenic activities, such as industry, agriculture and transportation, were found as less important factors for dissolved metals content in seawater of the Vlora Bay.
The dissolved form of the metals in the upper layer of seawater depended on weather conditions and red/ox process in seawater that may cause the variations in the concentration of dissolved form of heavy metals in seawater.

Keywords: active biomonitoring, urban area, seawater, heavy metals, accumulation factor, multivariate analysis, moderately polluted.

ATMOSPHERIC DEPOSITION OF HEAVY METALS TO TERRESTRIAL ECOSYSTEMS IN GERMANY

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Quantifying the atmospheric deposition of heavy metals to ecosystems remains a challenge as experimental data for many metals are few and, if present, emission estimates are highly uncertain. University of Vechta, TNO, and ÖKO-DATA started a project to establish the atmospheric deposition of heavy metals to terrestrial ecosystems in Germany and to identify ecosystems at risk. In total, 17 metals are in focus. As the information availability on the different metals differs substantially, the metals have been divided in three groups, indicating the effort put into their assessment from high to low.

Group A	*Group B*	*Group C*
Cd, Pb, Hg	As, Cr, Cu, Ni, Zn	V, Mn, Sb, Ti, Th, Co, Mo, Pt

The first step in the project is to quantify the atmospheric input to ecosystems. For this purpose two methodologies will be used. For all metals a first order assessment will be made based on experimental data. Available air concentration and wet deposition data will be classified as function of station type. Through combination of air concentrations with effective dry deposition velocities typical ranges for dry deposition fluxes can be estimated for each land use class for rural, urban and industrial areas. Combination with wet deposition data will provide first order estimates for the total deposition.
In the second methodology the chemical transport model LOTOS-EUROS will be used to calculate land use dependent dry deposition distributions across Germany for the metals that allow explicit modeling. Calculations will be performed using dedicated existing emission databases or through the modeling of PM in combination with metal contents. An example for the latter is the estimation of Vanadium levels based on modeled primary PM from heavy oil combustion. The observations from methodology 1 will be used for validation. As systematic biases are anticipated between model and observations the modeled fields may be scaled up or down towards the measured concentrations provided that the spatial gradients are resolved well. Wet deposition fluxes will be determined through *Residual Kriging* of observed fluxes.
The second step in the project will entail a comparison to other studies, e.g. EMEP, and an exploration to the use of moss monitoring data to improve the deposition estimates. At the meeting we will present the set-up of the study and illustrate the first results for deposition of Cadmium, Lead and Vanadium to German ecosystems.

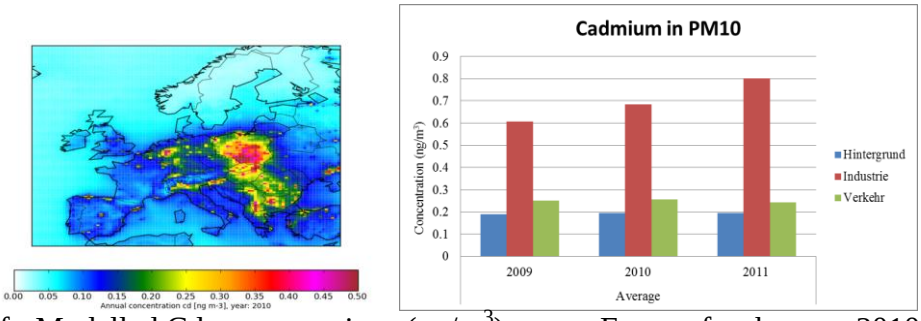

Figure 1. Left: Modelled Cd concentrations (ng/m^3) across Europe for the year 2010, Right: Observed Cd concentrations across Germany as function of station type for 3 years.

CORRELATIONS BETWEEN NITROGEN CONCENTRATIONS IN ATMOSPHERIC DEPOSITION AND MOSSES MAPPED FOR NATURAL LANDSCAPES IN EUROPE

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Nitrogen (N) concentrations in mosses are primarily determined by atmospheric deposition. The correlations are country- and N compound-specific and agree well with spatial patterns and temporal trends across Europe as a whole and in single European countries. Following the methodology presented by Schröder et al. (2013) this study investigates whether correlations between the concentration of N in atmospheric deposition and mosses within the units of an ecological land classification of Europe can be established (Schröder et al. 2014). To this end, N measurements from the 2005 European moss survey and modelled N atmospheric deposition in 2005 were intersected with a map of European landscapes. Then, considering minimum numbers of sampling sites required across Europe, in single European countries and within the landscapes of Europe and accounting for spatial auto-correlation, the correlations between the N concentration in mosses and corresponding deposition were calculated and mapped for each of those landscape units containing moss sampling sites. Using an example of one landscape with positive correlation and one landscape with no correlation between N concentrations in deposition and in mosses, influencing factors were ranked based on investigating the multivariate interactions between moss concentrations and, amongst others, atmospheric deposition, land use, elevation or moss species by classification and regression trees. From this study it could be concluded that the numbers of sampling sites within Europe and most participating countries as well as within most of the landscapes covering Europe are sufficient for reliable statistics. Spatial patterns of correlations between the atmospheric N deposition and N concentration in mosses proven to vary across the landscapes of Europe. Where clear positive correlations between N concentrations in deposition and mosses exist in landscapes, multivariate ranking identifies the deposition as main influencing factor. In cases with no correlation between deposition and N concentrations in mosses, other factors such as e.g. moss species collected may be of importance. Therefore, mosses were proven to serve as biological indicators for atmospheric depositions and ecologically defined land classes could be identified as complex indicators which allow relating exposure monitoring with effects assessment. This investigation, enabling to quantitatively describe differences of correlations between N concentrations in atmospheric deposition and in mosses between landscapes, was accomplished by studies concentrating on spatial differences of N concentration in mosses at the site level. To this end the N concentration in mosses sampled at sites with and without canopy drip effects in several European states, amongst them Germany (Kluge et al. 2013). The respective results are presented in the talk given by Michaela Kluge.

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INDICATIVE CONTENT OF N, S AND δ^{15}N IN THE MOSS *HYPNUM CUPRESSIFORME* IN THE SOUTHERN PART OF CENTRAL EUROPE

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In years 2005/6 almost 3000 moss samples were collected in 16 European countries and used for N analysis (Harmens et al., 2008). With the goal of testing if mosses could be appropriately used to assess atmospheric N deposition, Harmens et al. (2011) compared the N content in mosses and modelled atmospheric N deposition (EMEP model). For countries in the south-eastern part of Europe, they found a weak relationship between EMEP modelled N deposition and N in mosses.
With the aim of evaluating the monitoring programme in the southern part of central Europe in 2010, we collected samples of *Hypnum cupressiforme* Hedw. at 18 sites near locations where the quality of deposition (bulk and throughfall) are also assessed. From 18 sites, 4 were near locations of Slovenian national quality of deposition sites and 14 near locations of UN-ECE ICP-Forest Level II plots in Slovenia and neighbouring countries (Austria, Italy and Croatia). To test the influence of site characteristics, especially of the canopy on element content in moss samples, mosses were additionally collected at 91 sites at intersections of the regular 8x16 km grid throughout the country. At each sampling site two types of samples were taken, namely below the canopy and in the open and analysed for N and S content and δ^{15}N signature. Additionally, in the field the distances between moss collecting locations and the nearest tree crown were assessed.

Exploring the influence of atmospheric element deposition measured in bulk deposition on the element content in mosses showed that mosses collected in the open reflect atmospheric bulk N deposition, but only for cumulative one year deposition and not for the three year average. No influence of N in throughfall deposition on N in moss sampled under the canopy, nor influence of S deposition on S in moss (open and under canopy) was found. All sites showed no dependence between the δ^{15}N values of the mosses and the NH_4^+/NO_3^- ratio in the deposition. However, if 4 of the 18 sites with a cumulative yearly precipitation of less than 1000 mm were excluded, the dependence was significant for the δ^{15}N values in mosses collected in the open as well as under the canopy.
Evaluation of data for systematic sampling of mosses below the canopy and as far away as possible from the nearest tree from all 91 sites showed that canopy drip and litterfall bias the relationship for N and S, but not for δ^{15}N. Significantly higher values of N and S were found in mosses under the canopy in comparison to those at least 1 m away from the nearest tree. Namely, the moss samples collected at a least 3 m distance from the nearest tree crown have on average 41% and 34% lower values of N and S, respectively, in comparison to those sampled below the canopy.

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CROATIA PARTICIPATED IN THE ICP VEGETATION SURVEY SINCE 2005

Špirić Z. [1], Stafilov T. [2], Vučković I. [2], Kušan V. [1],
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Key words: biomonitoring, moss, air, Croatia

Croatia participated 2006 and 2010 in moss survey in the framework of the International Cooperative Programme on Effects of Air Pollution on Natural Vegetation and Crops heavy metals in Europe. http://icpvegetation.ceh.ac.uk

Moss samples were collected during the summer/autumn of 2006 and 2010, from locations evenly distributed over the country. The most dominant moss species in this study area were *Hypnum cupressiforme, Pleurozium schreberi, Brachythecium rutabulum and Homalothecium sericeum.*

In addition to the comprehensive qualitative and quantitative chemical analysis of all samples collected (*Špirić et al., 2012 and Špirić et al., 2013*), laboratory research by using the Kjeldahl analytical method was conducted in order to determine nitrogen concentration in all collected moss samples. In addition to that, 22 out of 161 moss samples from 2010 were subjected to gamma-spectrometric analyses for assessing activity of the naturally occurring radionuclides. Also, some selected samples were analysed for PAHs, PCBs, PCDDs and PCDFs.

According to this research and in comparison of the results in 2006 with 2010, it is obvious that the state of anthropogenic pollution in the last five years in Croatia has not changed significantly, although it is obvious that the anthropogenic influence in decreasing.

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IMPACT OF O_3 ON TERRESTRIAL ECOSYSTEMS: A MORE MECHANISTIC PARAMETRIZATION IN THE GLOBAL VEGETATION MODEL ORCHIDEE

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Tropospheric ozone is a major phytotoxic agent which can severely damage vegetation functioning. We propose a semi-mechanistic parameterization to integrate the impact of ozone on ecosystems in the dynamic global vegetation model ORCHIDEE. This study promotes an approach of ozone-induced effect on photosynthesis using flux-response relationships, in replacement to the former empirical function based on the external concentration AOT40 (Accumulated exposure Over a Threshold of 40 ppb). The new concept links a direct response of the maximum rate of photosynthesis V_{cmax} to the phytotoxic ozone dose (POD), also influenced by the accelerated senescence and countered by detoxification processes. Calibration of the parameters for C3 crops, grassland and deciduous forest was performed with sites measurements provided by field fumigation experiments. Furthermore, the correlation between observed and modelled relative reduction of V_{cmax} is higher basing the damage function on POD than on the ozone exposure indicator AOT40. As our photosynthesis scheme is coupled to a stomatal conductance module, we also compared the indirect effect on conductance with the impact simulated by the UNECE multiplicative conductance model, updated by the LRTAP 2010 convention. We found similar level of performance supporting the integration of the process-based flux-response module of impact in ORCHIDEE.

SEVEN YEARS OF N- AND O_3 POLLUTION × CLIMATE INTERACTION IN SUBALPINE GRASSLAND: ECOSYSTEM CARBON BUDGET, NITROGEN POOLS AND O_3-FLUX MODELLING (POD)

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We tested the effects of increased N and O_3 deposition on subalpine grassland 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^{-1} a^{-1}) and three levels of [O_3] (ambient, amb.×1.4 and amb.×1.7) in a free air fumigation system.

Carbon budget: Climatic conditions and management change have led to increased productivity and increased soil C content in the Alp Flix grassland. The N-treatment increased productivity, while O_3-treatment did not affect plant growth.
Net ecosystem exchange measurements, covering two complete years of contrasting climate, detected extreme short-term effects on the CO_2 C-balance, with antagonistic climate effects on ecosystem respiration and gross primary productivity.
Carbon isotope analysis revealed that O_3- and N-treatment made soil air, root material and bulk soil $\delta^{13}C$ less negative, but independent of the pollutant deposition treatment bulk soil became more negative over the years. While the bulk soil overall C content increased over time, soil density fractionation revealed a decreased C content in the oldest soil fraction.
In summary, single parameters of climate and atmospheric pollution are not sufficient to predict aboveground yield responses, and yield responses are not sufficient proxies for the ecosystem C-budget. Instead, the interaction of multiple parameters needs to be evaluated.

Nitrogen pools: N contents were calculated for green phytomass, necromass, roots, microbial biomass, soluble soil N, extractable soil N, and immobilised soil N. In addition, soil microbial communities were analysed by parallel 454 pyrosequencing of bacterial and fungal ribosomal sequences tags.
N deposition increased all plant N pools: +38% N in green phytomass, +73% in necromass and +33% in root biomass (growing season average). Thus, added N was primarily stored in phytomass. In contrast, most of the N pools were unaffected by O_3 exposure; only soil microbial biomass N was slightly increased by 0.85 g m^{-2} (+8%). However, a highly significant O_3 x N interaction was found for root N pool, revealing a negative effect of O_3 in unfertilized plots and a positive effect of O_3 in fertilized plots. A similar response pattern was observed in microbial biomass and soluble soil N, potentially related to changes in nutrient allocation and/or altered root and litter decomposition. However, the first reason can be excluded since the belowground responses were not accompanied by reciprocal changes in aboveground N pools. Rather, decomposition processes were affected, as indicated by the strong separation of fungal and bacterial communities among the four treatment combinations. However, a straightforward explanation for the interactive O_3 x N effect is missing yet.

Ozone flux modelling: On average, POD0 and POD1 were increased by a factor of 1.33 and 1.37 (O_3+) and 1.83 and 1.91 (O_3++), respectively, compared to the O_3 control treatment. Both maximum (2006) and minimum (2009) POD values occurred in the two warmest and driest years, respectively. In high O_3, POD reached values found to be detrimental to plant growth elsewhere. Thus, based on our data, the high O_3 tolerance of the investigated vegetation cannot be explained by limited stomatal O_3 uptake.

Abstracts

Posters

EVALUATION OF PINOLENE AS PROTECTANT AGAINST OZONE PHYTOTOXICITY

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Introduction: Tropospheric ozone (O_3), a widespread air pollutant, constitutes a real menace to plants, threatening food security. A major - still unsolved - issue is the lack of some appropriate (in terms of effectiveness and cost) protectant of plants against ozone that would be widely used in the agricultural practice. EDU is an effective protectant but is not commercially available, and it is quite expensive to be produced for laboratory use. Pinolene (Di-1-*p*-menthene) has been suggested as a potential protectant.

Purpose: In the framework of this research we evaluated the efficacy of EDU and Vapor Gard (containing 96% pinolene) to protect plants of the sensitive to O_3 Bel-W3 tobacco (*Nicotiana tabacum L.*) plants against ozone.

Materials and methods: Two months old plants were exposed either to ozone ([O3+]: 90 ppb x 12 days x 8 hours/day) or to ozone free air ([O3-]) and treated either by Water (control group) or by EDU (at 10, 100 and 500 mg/lit) or by Vapor (at 1, 5, 10 and 50 ml/lit). The measured parameters were: leaf area (LA), foliar visible injury (VI), plant height (Height) & dry weight (DW), total chlorophylls (ChlT) & total carotenoids (Car), ChlT/Car, Chlα/Chlβ ratio, quantum yield of photosynthesis (QY), leaf greenness (SPAD) and stomatal resistance (SR).

Results and discussion: EDU, when applied at low concentration (10 mg/lit) did not protect the plants against O_3. However, the plants treated by the higher EDU concentrations (100 and 500 mg/lit) exhibited much fewer or not at all symptoms: plants were protected in all the measured parameters. In contrast, pinolene only at 1ml/lit prevented ozone-induced reduction of the parameters LA, DW, ChlT and Car. The ChlT did not differ between [O3+] and [O3-] groups. Pinolene, at 50 ml/lit, caused reduction of height and ChlT in both [O3+] and [O3-] groups. In [O3+] group, all of the subgroups of plants treated by substances (except those treated by EDU at 500mg/lit) the Chl/Car ratio was reduced when compared pairwise with that of the corresponding subgroups in the [O3-], suggesting a higher reduction of carotenoids in comparison with chlorophylls; however, this difference was statistically significant only in plants treated by pinolene at 10 ml/lit. Chlorophylls α/β ratio did not differ neither between [O3+] and [O3-] groups nor among the substances subgroups. As to the QY, Car and SR, pinolene did not exhibit any effect. Concerning SPAD within the [O3+] group, only those plants treated by pinolene 1 ml/lit had statistically significantly higher SPAD value in comparison with those sprayed only with water. However, pinolene, when applied at 10 and 50 ml/lit caused phytotoxic symptoms - necrotic spots very similar to those caused by the O_3; these phytotoxic symptoms were developed only a few hours after the pinolene application. At the end of the experiment (after 12 days) the plants injured by the highest concentration of pinolene were shorter and stunted with distorted leaves.

Conclusions: EDU remains the best available protectant against ozone; pinolene may has some protective role on Bel-W3 plants at low concentrations (1 ml/lit) that should be further investigated for several plant species and ozone levels. At higher concentrations (10 and 50 ml/lit) pinolene is phytotoxic, at least to Bel-W3 tobacco plants.

AIR POLLUTION STUDY IN MACEDONIA BY USING MOSS BIOMONITORING TECHNIQUE, NAA, ICP-AES, AND AAS

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The third moss survey in the Republic of Macedonia took place in August and September 2010 when 72 samples of the terrestrial mosses *Homalothecium lutescens* and *Hypnum cupressiforme* were collected over the whole territory of the country using the same sampling network grid as for the previous surveys in 2002 and 2005. Using neutron activation analysis (NAA), inductively coupled plasma-atomic emission spectrometry (ICP-AES), and atomic absorption spectrometry (AAS), a total of 47 elements (Al, As, Au, Ba, Br, Ca, Cd, Ce, Cl, Co, Cr, Cs, Cu, Dy, Eu, Fe, Hf, Hg, I, In, K, La, Li, Mg, Mn, Mo, Na, Nd, Ni, P, Pb, Rb, Sb, Sc, Se, Sm, Sr, Ta, Tb, Th, Ti, U, V, W, Yb, Zn, and Zr) were determined. Distribution maps were prepared to point out the regions most affected by pollution and related to known sources of contamination. To reveal hidden multivariate data structures and to identify and to characterize different pollution sources Principal Component Analysis (PCA) was used. A comparison of the results obtained in 2010 and 2005 [1, 2] surveys was carried out to examine temporal trends in the elemental depositions.

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ATMOSPHERIC DEPOSITION OF HEAVY METAL IN EASTERN ALBANIA BY MOSS BIOMONITORIG TECHNIQUE

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The moss biomonitoring technique was applied for assessing environmental situation of air pollution in eastern Albania. The carpet-forming-mosses have some particular properties that make them suitable for monitoring air pollutants. Sampling was performed in accordance with the LRTAP convention-ICP protocol and sampling strategy of the European Program on Biomonitoring of HM Atmospheric Deposition. The *Hypnum cupressiforme* moss species were used as bioindicator for air pollution, that were collected during June-July, 2011 at 19 sites distributed over the eastern part of Albania. A total of 19 elements (Al, As, Cd, Cr, Cu, Fe, Ni, Pb, V and Zn) were determined by ICP-AES technique. The results reflect local emission point.

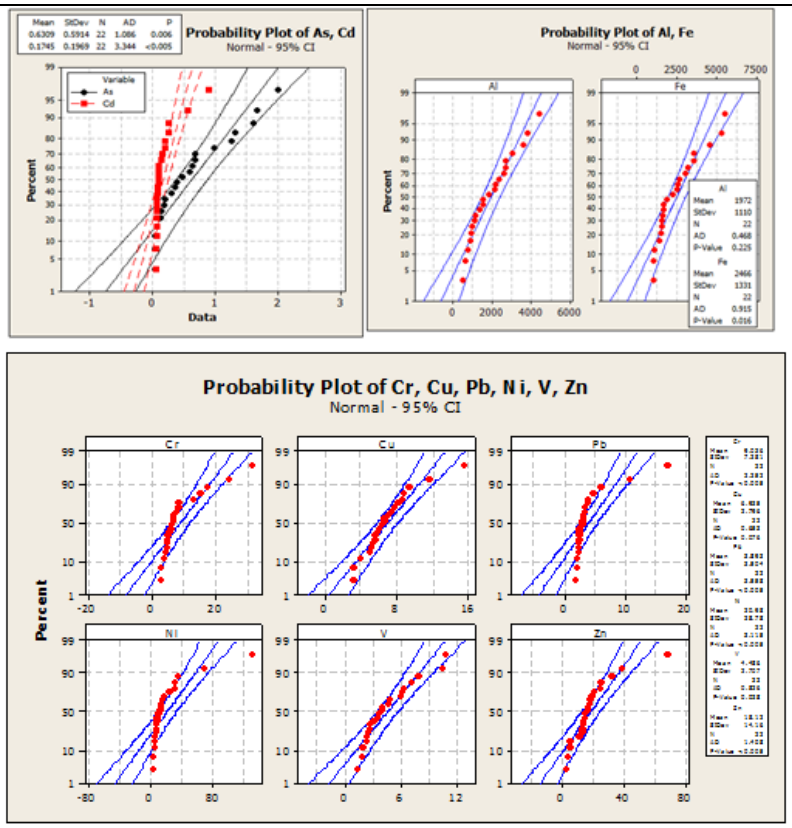

Figure 1 Normal distribution of Al, As, Cd, Cr, Cu, Fe, Ni, Pb, V and Zn

These elements show high variability compare to their mean values. High values of skeweness and curtosis are also evident that mean the data are strongly skewed, are not normal distributed, except Al (Fig. 1, $P<0.05$). The data matrix was treated statistically by using Minitab 15 software. Factor analysis with VARIMX rotation was used to identify and characterize different pollution sources and to identify the areas receiving the highest metal pollution load. The data of the factor analysis were applied to distinguish elements mainly of anthropogenic origin from those predominantly originating from natural sources.
Geogenic factor, wind soil dust and weather conditions were identified as main factors of contamination of this area.

Key word: *mosses, biomonitoring, heavy metal, ICP/AES, factor analysis, Albania.*

PHYSIOLOGICAL RESPONSES: DO LICHENS, MOSSES AND TREE BARK REACT WITH DIFFERENT CARBON AND NITROGEN ISOTOPE PATTERNS ALONG A NITROGEN DEPOSITION GRADIENT?

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Mankind has dramatically influenced the N fluxes between soil, vegetation, water and atmosphere – the global N cycle. Many problems are caused by the human-accelerated environmental change. Long-term ecological monitoring programs are fundamental to evidence-based environmental decision-making. Biomonitoring is essential, particularly where less deposition measurement sites exist. In Germany still key societal threats of excess reactive nitrogen (N_r) are on hand.
An overview is given of the physiological response of different bioindicators towards different anthropogenic N impacts in the environment. Abundance of ^{13}C and ^{15}N contents and total C and total N of lichens, mosses and tree bark, were reported.

Lichen, moss and bark samples were collected at 16 deposition measurement sites of the Air Monitoring Network of the Federal Environment Agency of Germany (UBA) in September and October 2008 (lichens and tree bark samples) and from September to October 2006 (moss samples). The investigation has tested 442 lichen samples, 203 tree bark samples and 16 moss samples and considered the epiphytic lichen species *Hypogymnia physodes*, *Parmelia sulcata*, *Xanthoria parietina*, *Physcia tenella* and *Physcia adscendens* (pooled *Physcia* sp.), the moss species *Hypnum cupressiforme*, *Pleurozium schreberi* and *Pseudoscleropodium purum*, and the tree species *Acer platanoides*, *Acer pseudoplatanus*, *Alnus glutinosa*, *Betula pendula*, *Carpinus betulus*, *Corylus avellana*, *Fraxinus excelsior*, *Malus domestica*, *Pinus sylvestris*, *Populus* × *canadenisis* agg., *Prunus avium*, *Prunus domestica*, *Quercus petraea*, *Quercus robur*, *Salix* sp., *Sorbus torminalis*, *Tilia cordata* and *Tilia platyphyllos*. The various tree species were not taken into account separately.

Isotope specificity of species and of taxanomic groups relating to $\delta^{15}N$ and $\delta^{13}C$ values were indicated, probably due to differences in plant physiology and biochemistry.
All bioindicators have shown an increase of the N content and simultaneously a decrease of the C/N ratio while enhanced agriculture-related N impact exist. C pool turnover rates greatly increased in response to N impact which point to N deposition as a stress factor.
Furthermore, lichens and bark samples were characterized by an increase of less depleted $\delta^{15}N$ values in highly agricultural areas which proves the existence of particulate ammonium, originating from agricultural immissions.

RELATIONS BETWEEN ACCUMULATION OF TRACE ELEMENTS, PHOTOSYNTHETIC ACTIVITY, WATER STATUS AND GROWTH PARAMETERS OF ITALIAN RYEGRASS EXPOSED IN AMBIENT AIR CONDITIONS

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The aim of the study was to evaluate an effect of heavy metals to Italian ryegrass (*Lolium multiflorum* L.) exposed in ambient air conditions. Plants were cultivated for six weeks in the greenhouse conditions and afterwards were transported to five exposure sites located in Wielkopolska region. Two sites were located in the Poznan city, one in suburban area, one in Landscape Park and the last one in the rural area. Plants were exposed for four weeks and four exposure series were performed in 2012 growing season, beginning in the middle of May. Selected trace elements (Ni, Cr, As, Cd, Pb) were analysed before and after exposure, as well as relative water content (RWC) and specific leaf area (SLA). While photosynthesis activity parameters were measured before, in the middle and at the end of every exposure series.

The experiment revealed an increase of leaf thickness due to increased heavy metal concentrations in ambient air, while higher trace element concentrations caused a decrease of relative water content of exposed plants. Hence, based on our results, we can conclude that trace element concentrations in ambient air were high enough to affect *Lolium multiflorum* L. water relations, but too low to influence leaf blade thickness.
Net photosynthesis rate and stomatal conductance were mostly negatively related with all analysed trace elements, while intercellular CO_2 concentrations revealed opposite tendency. However, there were some exceptions, such as for arsenic in two exposure series, when lack or low relations with P_N and g_s were observed. The lowest levels of P_N and gs were mostly noted at two sites located in the city, where high concentrations of Cd and Pb was recorded in *L. multiflorum* leaves. Moreover, the lowest levels of P_N were noted at all sites during third exposure series, when the highest Cr and Ni levels were recorded.

Acknowledgement
Presented investigations were supported by project grant No.: N 0540/B/P01/2011/40 from the National Science Centre.

MAPPING OF AIR POLLUTION ROADSIDE ORIGIN BY SOME TRACE METALS (CD AND CU) USING THE TRANSPLANTATION OF A BIO ACCUMULATOR LICHEN *XANTHORIA PARIETINA* IN THE CITY OF TIARET (ALGERIA*)*

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This study seeks to quantify air pollution by two trace metals Cd and Cu picked by lichen Xanthoria parietina bio accumulator in the city of Tiaret (Algeria). This measure of pollution is by using the method of atomic absorption spectrophotometry provides results highlighting low levels of air pollution by the two elements mentioned above. This does not mean that air pollution is absent but that these two trace elements are spread by wind over the rugged terrain of the region.

Keywords : pollution, trafic, Cu, Cd, lichen, Xanthoria parietina, bio accumulation, Tiaret, Algérie

TEMPERATURE EFFECTS ON YIELD OF R123 AND S156 SNAP BEANS – ESTABLISHING CRITERIA FOR EFFECTIVE USE AS AN OZONE BIO-INDICATOR SYSTEM

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Snap bean recombinant inbred lines derived from a cross between ozone (O_3)-sensitive and tolerant parents (Reinert and Eason, 2000) are being tested as an O_3 bio-indicator system. The S156 (O_3-sensitive) and R123 (O_3-tolerant) snap bean genotypes were originally identified based on similar yield under clean air conditions and differential yield responses reflected in declining S156/R123 yield ratios when grown in elevated O_3 environments. The S156/R123 pair has been tested previously in open-top chambers (Burkey et al., 2000) and is currently being utilized in the ICP-Vegetation snap bean project.

While S156 and R123 consistently exhibit differential foliar injury in response to O_3 exposure, differential yield responses to assess O_3 impacts appear to be more complex. At the 2013 Task Force meeting, we reported observations that suggested high temperature may limit mature pod formation in a manner that could complicate interpretation of O_3 impacts in terms of S156/R123 yields ratios. To directly test this hypothesis, mature pod yield in the absence of ozone was measured over a range of growth temperatures. Mature pod yield declined significantly at a growth temperature of 32 °C and was practically zero at a growth temperature of 36 °C (Table 1). The decline in mature pod yield at increasing growth temperature was associated with large increases in small undeveloped pods (Table 1), supporting the hypothesis that high temperatures can inhibit pollination and limit pod yield potential. Given that ambient O_3 is highest in the summer months, the use of the S156/R123 pod yield ratio as an O_3 bio-indicator may not be appropriate in regions with average temperatures high enough to impact mature pod development. Foliar injury ratings may still be useful in these situations.

Table 1. Snap bean genotypes S156 and R123 were grown from planting through harvest in charcoal-filtered air (≤ 2 ppb O_3) under 4 temperature regimes during the fall of 2013. Plants were grown in 15-liter pots of Sunshine MVP mix with Osmocote Plus slow-release fertilizer using Outdoor Plant Environment Chambers (OPECs) to control day (D)-night (N) temperature (T). The experimental design (4 T x 2 blocks) considered each of the eight OPECs as an experimental plot with 4 plants per genotype randomly assigned within each plot. Pod yield (mature pods with seeds) and counts of undeveloped pods were measured at 82 days after planting. Values are chamber means ± standard error (n = 2).

Target T D/N (°C)	Actual T D/N (°C)	S156 pod yield (g plant^{-1})	R123 pod yield (g plant^{-1})	S156/R123 yield ratio	S156 undeveloped pods plant^{-1}	R123 undeveloped pods plant^{-1}
24/20	24/21	68 ±10	79 ± 6	0.87 ± .20	13 ± 1	9 ± 2
28/24	27/25	48 ± 8	64 ± 13	0.80 ± .29	20 ± 4	32 ± 4
32/28	32/28	23 ± 6	18 ± 1	1.30 ± .25	28 ± 4	115 ± 63
36/32	34/31	3 ± 3	1 ± 1	---	73 ± 7	71 ± 12

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OZONE: AN ELICITOR OF SECONDARY COMPOUNDS IN *HYPERICUM PERFORATUM*?

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Ozone (O_3) is considered the most ubiquitous and phytotoxic air pollutant in industrialized and in developing countries. Many secondary metabolites are active compounds of medicinal plants and show a wide range of biological activities. They play important role in the adaptation of the plant to the environment, contributing to its resistance to biotic and abiotic stresses. It is interesting to study the interaction of O_3 with wild medicinal plants, with special regard to the effect on the secondary metabolism. In this work, attention has been given to a species of great scientific and economic interest, *Hypericum perforatum* L.. Commonly called St. John's wort, it is an herbaceous perennial plant, long known for its putative medicinal properties. Its macerate in oil has been traditionally used as a vulnerary to treat minor burns, wounds, skin inflammation and nerve pain, but its alcoholic extracts are gaining increasing popularity as an alternative treatment for mild to moderate depression. Considering O_3 as elicitor of secondary metabolites, the study evaluated the change of bioactive compounds (flavonoids, the naphthodianthrone hypericin and the acylphloroglucinol hyperforin) of plants obtained from local germplasm in vegetative stage. Six-month old plants were exposed under controlled environmental conditions to O_3 (100 ppb, 5 h); control plants were maintained in charcoal filtered air. Samples were taken immediately at the end of treatment and after 24 h. In all cases, there were no visible symptoms. The search of the metabolites was performed by HPLC on methanolic extracts obtained from apical shoots. Twenty-four h after the end of the exposure, it was observed a significant increase of hyperforin and chlorogenic acid (+21.6% and 49.7% compared to controls, respectively). Moreover the total levels of phenols increased significantly (+44.5%). They were correlated to an increase in the activity of L-phenylalanine ammonia-lyase, that rises 1.5-fold 24 h after the fumigation and 2-fold already at the end of the fumigation. Also total content of flavonoids and anthocyanins increases (+60.6% and +1.7%, respectively). On the contrary, isoquercitrin concentration decreases (-41.4%). No significant change has been found for hypericin, quercetin, hyperoside and amentoflavone levels. This study shows that O_3 can distinctly affect the secondary metabolism of this species and therefore its medicinal value, changing the ratio amongst the active compounds. Biotechnological applications of O_3 in the field of medicinal plants deserve attention.

ONE YEAR RESULTS OF SOIL NO & NO2 FLUXES MEASUREMENTS IN SUBALPINE FOREST

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In the framework of the FP7 project ECLAIRE (Effects of Climate Change on Air Pollution and Response Strategies for European Ecosystems) an automated dynamic chamber system for measure NO & NO_2 fluxes from the soil has been installed during 2012 at the JRC.
In particular 5 replicate chambers measuring soil fluxes and one chamber that was closed to the ground and thus serves as a measurement blank have been located in a little forest, while the analysers, pumps, control and data acquisition systems are installed nearby in a small trailer. Air temperature and humidity probes were also placed in each chamber.

Pic. 1and 2: Example of measuring chamber system (open and closed) used to measure NO & NO_2 fluxes from soil during the experiment at the JRC Ispra forest site.

Sequentially each chamber was closed and flushed with air for approx. 55 l/min for 6 minutes. At the same time, the concentrations of NO, NO_2 are measured at the outflowing air of chamber. To account for concentration changes due to the reaction $NO + O_3 \rightarrow NO_2 + O_2$ in the chamber and sampling lines, the concentration of O_3 is measured as well. Combining the measured concentrations, air flow and chamber size, raw soil fluxes of NO and NO_2 are calculated.
The NO and NO_2 soil flux data from the first year of measurements are showed and discussed. First results indicate some differences in the fluxes for every chamber probably due to slightly different soil conditions in terms of micro-organisms or illumination conditions. During the different seasons, fluxes of NO are clearly higher in summer compared to winter. This indicates that main drivers for NO emissions are soil temperature and soil water content. Regarding NO2, the situation regarding drivers is less defined.

PHOTOSYNTHETIC PERFORMANCE OF *QUERCUS ILEX* L. UNDER LONG-TERM OZONE EXPOSURE PROBED BY CARBOXYLATION EFFICIENCY, MAXIMUM APPARENT QUANTUM YIELD AND MODULATED 820 NM REFLECTION.

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Assessing ozone (O_3) effects on photosynthetic machinery is a key issue to understand how the oxidative stress influences vegetation functionality and the ecosystem services provided (Manes et al., 2012). Moreover, data about the RuBisco and photosystem functionality measured during long term experiments could be important to improve the accuracy of ozone flux models to be use for risks assessment (Fares et al., 2012). Trends towards tolerance to O_3 have been studied for Mediterranean tree species and contrasting results were obtained over a large number of experiments, probably due to different levels of O_3 exposure and growth conditions. Moreover, only few experiment assessed the O_3 effects on evergreen species in terms of dynamic response of CO_2 assimilation to varying conditions of temperature and photosynthetic photon flux density (Mereu et al., 2011) and, to the best of our knowledge, no data is available about fluorescence of photosystems I (PSI), that plays a key role in response to oxidative stress. In this experiment, we have assessed long-term effects of O_3 exposure on *Quercus ilex*, an evergreen species with great ecological relevance in Mediterranean Basin for its wide natural distribution, as well as widespread use in urban context. Two-years-old seedlings were continuously exposed for 5 months in open-top chambers facility at the Centre for Research on Effects of Pollutants on Ecosystems, in Curno, to three O_3 treatments: charcoal filtered air (F, - 41% of ambient air O_3), and non-filtered air supplemented with + 35% (NF^+) and +71% ppb O_3 (NF^{++}) of ambient air O_3. Interestingly, it was found that after 40 days of starting experiment, an early response to O_3 leads to an increase in leaf dark respiration and photorespiration. Fluorescence parameters were also affected, and in particular the PSI functionality was involved in the first phase of oxidative stress response.

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EFFECTS OF OZONE AND NITROGEN DEPOSITION IN YOUNG TREES OF HORNBEAM AND OAK. RESULTS FROM THE ECLAIRE EXPERIMENTS IN ITALY

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An Open-Top Chambers (OTC) experiment with ozone enrichment and increased nitrogen deposition has been performed during two consecutive years (2012 and 2013) in Northern Italy on young trees of *Quercus robur* and *Carpinus betulus*. Two hundreds and sixteen saplings of each species were potted and placed in 12 Open-Top Chambers following a split-plot design with 3 randomized blocks and two factors: ozone concentration, the main factor, at 4 different levels (CF-45%, NF, NF+35%, NF+70%), and nitrogen irrigation (NDep), the nested factor, at 2 different levels (tap water for control, tap water +NDep of 70Kg of $N*ha*y^{-1}$). These treatments were applied for two consecutive growing seasons. In both years stomatal conductance (g_s) measurements and CO_2 assimilation response curves have been made throughout the season to assess the impacts on physiological and photosynthetical parameters. Half of the saplings was harvested at the end of 2012 season, while the remaining half was harvested at the end of 2013, in order to estimate the effects of both stresses, alone and in combination, on the total biomass production and on the root/shoot partition of the plants.

After two years of treatments, a general positive effect of nitrogen deposition on biomass production was found in both species, as it could be easily expected. This biomass increase was particularly intense in *C. betulus* (+76% of the total biomass, + 65% of roots biomass).

Q. robur showed a greater response to ozone than *C. betulus* in the control Ndep conditions. Oak plants showed a 10% and 18% of reduction of the total and root biomass in NF+70% treatment. This response was also found in the plants subjected increased to nitrogen deposition (-15% for total biomass, -16% for roots biomass). Hornbeam seemed stimulated by O3 when no nitrogen was added (+5% of total biomass). However, NDep treatment made hornbeam more susceptible to ozone, which caused a 30% biomass decrease in both shoots and roots.Looking at g_s as a possible driver of the plants' response to ozone, we found that ozone lead to a 21% decrease of g_s in oak plants when the had a total biomass reduction of 10%. This response is confirmed in NDep treatment (-18% in g_s, -15% in total biomass). Thus, oak seemed to be more vulnerable to g_s limitation because of the consequent reduction of CO_2 assimilation.In hornbeam, without nitrogen addition, ozone caused a slight reduction on g_s leading to a decrease of stomatal dose and a small increase in total biomass (+5%).

On the contrary, nitrogen deposition caused a significant increase of g_s (+23%) that lead to an increased ozone uptake, thus suggesting an overwhelming of the detoxifying defence (-30% in total biomass).Stomatal conductance, thus, reveals to be a key driver of the plant's response to ozone, but the final manifestation of the effect seems to be modulated by the detoxifying capacity of the plants.

The general decrease of g_s in both species caused by ozone (also found in 2012 measurements), suggests the need to include an $f(O_3)$ modifying function in the stomatal conductance models which will be defined for these two species. Some of biomass responses are partially in disagreement with the results of the first year, likely for the presence of a carry over effect. This fact highlights the importance of performing long-term experiments (more than 1 year) for the investigation on ozone and nitrogen effects on biomass.

BIOINDICATION AND MONITORING OF ATMOSPHERIC DEPOSITION USING TREES AND SHRUBS

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Vitality and morphological changes of woody plant leaves collected in the area affected by emissions from the metallurgical enterprises was studied. It was shown that *Tilia cordata, Aesculus hippocastanum, Crataegus, Symphoricarpos albus* (see Figure) can be used for bioindication of the atmospheric deposition of trace elements by morphological parameters (necrosis, leaf morphology change).

Crataegus monogyna *Aesculus hippocastanum* *Tilia cordata*

Figure. Necrotic and chloritic changes of shrubs and tree leaves caused by emissions from the metallurgical enterprises

Scanning electron microscopy allowed establishing the ratio of conglomerate components adsorbed on the leaves surface from aerosol emissions. Proportion of the organic components in the adsorbed particles was maximal (up to 87−95%); percentage of soil particles was 4−5%, and the amount of heavy metals was of the order of 2−4%. The dominant elements in the adsorbed particles of the technogenic origin were Fe and Mg (1.5−4.5%). Such elements as Mn, Cu, Pb, Cd, V, Mo, and Zn, which are the main components (with Fe) of industrial emissions of metallurgical enterprises in the city of Tula, were present in the range of 0.1−0.6 %. Their concentration exceeds the MPL in atmospheric air studied by means of neutron activation analysis and atomic absorption analysis of air aspirator air filters. Thus, the analysis of dust deposited on the woody plant leaves surface is an express method to assess the presence and ratio of the components of atmospheric deposition that can be used to judge on air quality exhaled by human respiratory tract.

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ACCUMULATION OF HEAVY METALS IN THE MOSS *PLEUROZIUM SCHREBERI* IN THE WEST OF RUSSIA (KALININGRAD REGION)

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The moss biomonitoring technique was applied to air pollution studies in the West region of Russia (Kaliningrad region). Samples of the terrestrial moss *Pleurozium schreberi* have been collected every 5 years, from 2000 in accordance with the sampling strategy of the European moss survey program.
Moss samples were collected on regular network of 10x10 km during the August – September in 2000, 2005, 2010. Metals such as Pb, Cd, Cr, Ni, Cu, Fe, Mn were determined by AAS technique (by flame – Mn, Fe; ETA – others).
In general, the concentration of Cd, Pb, Ni and Cu in mosses decreased between 2000 and 2010; the concentration of Cr increased in the same period.

Table 1 - Means of mediana in moss *Pleurozium shreberi* in Kaliningrad region in 2000 – 2010 year (mg/kg DW)

year	Cu	Ni	Pb	Cr	Cd	Mn	Fe
2000	5,3	4,11	13,2	0,14	0,14	256	192
2005	9,34	3,8	4,63	0,88	0,18	132	222
2010	2,84	1,21	1,91	1,41	0,05	245	168

The values of metals vary widely, but low values prevail. Highest concentrations were determined in the west part, lowest ones in the center of region.

Table 2 - Concentration of Heavy metals in moss *Pleurozium shreberi* in Kaliningrad region in 2010 year (mg/kg DW)

	Cu	Ni	Pb	Cr	Cd	Mn	Fe
mean	4,46	1,20	2,51	2,16	0,11	346	217
min	1,68	0,36	0,39	0,69	0,006	73	113
max	19,7	2,54	9,33	12,1	1,12	960	700

Principal component factor analysis was used to identify and characterize different pollution sources and to point out the most polluted area. There were identified two factors of metals emission.
The maps of metals atmospheric deposition were created. Cartography analysis revealed spatial patterns of metals distribution. Probably highest levels of metals in moss in the west part of region (Sambia peninsula) and elevated levels in the North, North-East and South-West parts are indicating trans-boundary pollution.

FOLIAR SURFACE CLEANING TECHNIQUES FOR ANALYSIS OF PARTICULATE CHEMICAL COMPOSITION IN AIR POLLUTANTS MONITORING

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Major contributor to the adverse health effects of air pollution on the respiratory system, airborne particles are the main drivers of the increased short and long-term frequency of respiratory ailments for humans, and especially for children, with allergies, acute asthma and other respiratory diseases inducing exacerbated responses of the immune system. Moreover, accumulation of organic and inorganic pollutants in above-ground plant parts (i.e. leaves, fruits) via wet or dry deposition can menace human health after ingestion (intake through the food chain).

Acting as a protective barrier, the hydrophobic or only moderately hydrophilic epicuticular waxes of leaf surfaces of most plants can also efficiently trap airborne particles; particles remaining adsorbed or incorporated into the hydrophobic wax layer of the foliage. Over the last three decades, chemical foliar analysis was strongly privileged as a diagnostic tool of tree nutrition over chemical soil analysis, with an increasing interest as environmental indicator of air pollution. The amount of deposited particles can be modified by rainfall, which washed off the trapped particles and plant exudates, leading to an increase of element concentrations in througfall. Therefore, the wax trapping ability of plant leaves needs attention in soil/plant transfer rate analysis for air quality monitoring studies. Thus, washing sampling techniques are necessary to distinguish between absorbed and accumultated particles.

Several techniques were used to clean both dry or moistened plant tissues with various washing chemicals (i.e. distilled or tap water, chloroform, toluene, tetrahydrofuran, hydrochloric acid, EDTA) and with or without mechanical shaking. Mechanical cleaning techniques were, to a lesser extent, also used by stripping a resin film deposited on leaf surfaces.

The foliar surface cleaning methods depend on the physical and chemical properties of the studied elements trapped on the leaf surface (surface contamination risks). The protocole should be adapted to 1) plant species and 2) surface properties and age of the leaf, since they are determinant factors (washing chemical product choice, and extraction time).

STUDY OF A TROPOSPHERIC OZONE EFFECT ON TWO CULTIVARS OF TOMATO: *LYCOPERSICON LYCOPERSICUM COLGAR (L.) KARSTEN EX FARW.* AND *SOLANUM LYCOPERSICUM* RECHAIGA II

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Two cultivars of tomato: tomato Colgar and Rechaiga II were exposed to different concentrations of O3 (50, 80 and 100 ppb) in a fumigation chamber to evaluate their sensitivities and check some physiological parameters. The cultivar Rechaiga II (Solanum lycopersicum) appeared particularly sensitive to O3 at an early stage of growth. Typical responses chlorotic in 24 hours after exposure to a single pulse of 50 ppb for 4 h, for a period of 7 days were recorded spots. Respondents physiological parameters (stomata conductance, the membrane integrity, chlorophyll a, b, total, carotenoids and soluble sugars), have suffered alterations following exposure. The cultivar Rechaiga II was sensitive to ozone while cultivar Colgar (Lycopersicon lycopersicum (L.) Karsten ex Farw) has remained asymptomatic (resistant) throughout the entire experiment.

Keywords
Tropospheric ozone, fumigation chamber, Solanum lycopersicum , Lycopersicon lycopersicum (L.) Karsten ex Farw, necrosis , physiological parameters.

CRITICAL EVALUATION OF ECOSYSTEM POLLUTION

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Pollution problems in the forest ecosystems resulting from 100 year of operation of three smelter complexes in the Central Spiš are reviewed. Original data are presented with respect to temporal and spatial trends of nitrogen, sulphur, and heavy metal pollution. INAA at the IBR-2 reactor has made it possible to determine the content of 40 elements in mosses, foliage of forest tree species, lime and spruce wood, soil and wildlife in marginal Slovak hot spot - Central Spiš. Instrumental neutron activation analysis (INAA) at the IBR-2 reactor has made it possible to determine the content of 40 elements in mosses, foliage of forest tree species, lime and spruce wood, soil and wildlife in the marginal Slovak hot spot – the Central Spiš. In addition to NAA, flame atomic absorption spectrometry (AAS) was applied to determine the contents of S, Cd, Cu, Hg and Pb.

The polluted air impact on forest ecosystems is a major question at present and in the future. 1.7-fold exceeding of critical values and high concentrations of As, Fe, Hg, and N in spruce needles were found in the Central Spiš. The Low Tatras National Park served as a pristine (baseline) region. The concentration of Hg and As in humus and horizont A_o from Central Spiš is increased markedly. Other determined elements (Cd, Co, Cu, Ni, Pb, and Zn) do not exceed the limits. In the area of Central Spiš we have found in comparison with the Norwegian limit values (the Central Norway- as relatively pristine region) exceeded levels for Al, As, Ca, Cd, Cl, Co, Fe, K, Mn, Sb, Sm, Sr, W, and Zn. In comparison with Magnitogorsk in the Ural Mountains (the most polluted area in Europe) we found in Spiš higher values for Ca, Cl, Co, K and Mn. We found the highest concentrations of Au, Br, Co, and Sb in the oldest 70–80 years old spruce wood, and the highest concentrations of Al, As, Au, Br, Cl, K, Na, Rb, Sb and Sm in the oldest 80–90 years old lime wood. We found the highest concentration of Al, As, Ba, Br, Ce, Co, Cr, Cs, Fe, Hf, Hg, K, La, Mg, Mo, Na, Ni, Pb, Rb, Sb, Sc, Sm, Ta, Tb, Th, U, V and W in soil samples; the highest concentration of Ca, Cd, Cl and S in spruce roots; Mn and S in spruce needles; Sr in mosses and Zn in lichens. In comparison with limit values the concentration of Ba, Co, Hg, Mg, Mn, and Sr in the teeth of roe deers is increased markedly. Other determined elements (Al, As, Ca, Cd, Cu, Fe, Na, Pb and Rb) do not exceed the limits.

Key words: air pollution, biomonitoring, metals, foliage, mosses, soil, wood

ATMOSPHERIC DEPOSITION OF HEAVY METAL DEPOSITION IN KOSOVO BY USING MOSS BIOMONITORING AND AAS

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Bryophytes act as precise and sensitive bioindicators as well as bioaccumulators of metal deposition in the environment. The atmospheric deposition of Cd, Cr, Cu, Fe, Hg, Ni, Mn, Pb and Zn in Kosovo was investigated by using moss species (*Hypnum cupressiforme)* as bioindicators. This research is a part of the international programme (ICP Vegetation Programme, UNECE) investigating the impacts of air pollutants on crops and natural vegetation. Moss samples were collected from 25 sampling sites evenly distributed over the territory of Kosovo during the dry period of summer 2011. The most dominant moss species in this study area were *Hypnum cupressiforme*. The map of sampling points is shown in Fig. 1.

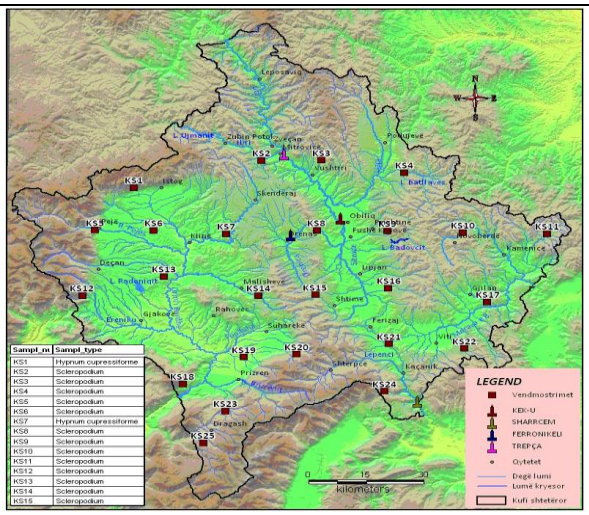

Figure 1 The map of sampling points and coordinates

Unwashed, dried samples were totally digested by using microwave digestion system and the concentrations of metal elements were determined by AAS equipped with flame and/or furnace systems. The results reflect local emission points. Descriptive statistics were made to valuate contamination level. The data obtained in this study were compared with those obtained in the similar studies in neighboring countries and Europe in 2010. The estimated level being highest for Fe, Mn and Zn followed by Pb, Cu, Cr, Ni, Cd and Hg. Overall, Fe, Zn and Pb are responsible for causing major pollution in the studied area. Relatively high lead concentrations were also found in Bulgaria Kosovo (Harmens et al. 2013).

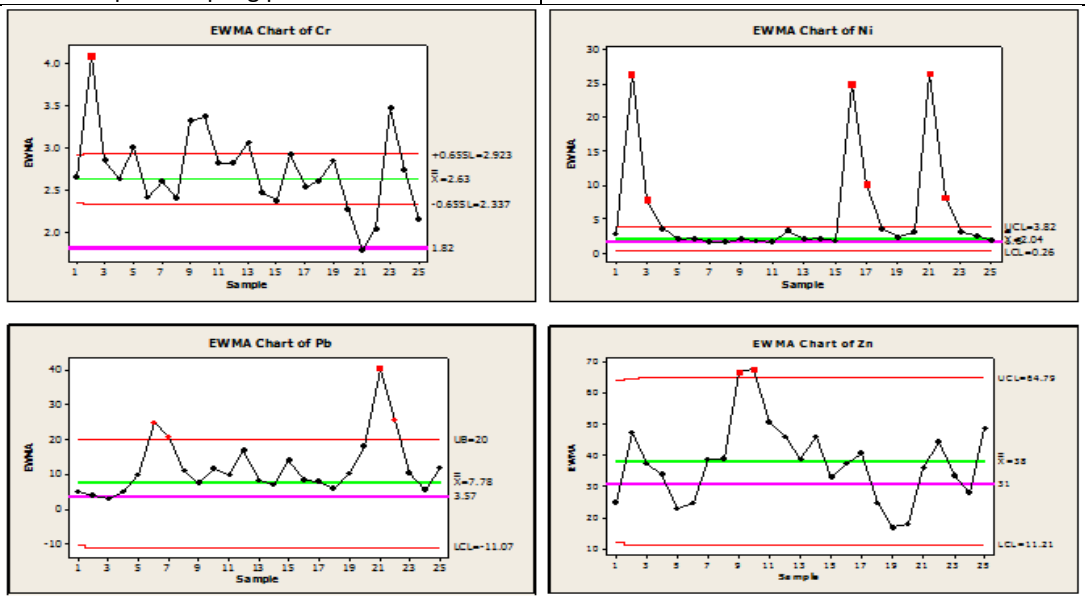

Figure 2 Cr, Ni, Pb and Zn distribution

Statistical analyses also revealed that correlation exist between the metal content in mosses and degree of pollution in studied sites. High values of positive skewness and kurtosis were found for the elements (except Cu and Zn) by determining that the data of these elements are skewed right. EWMA chart reflected the spatial distribution of the elements, and multivariate analysis applied to the data matrix of the concentrations of the elements identified the major factors that affect on the elements distribution

Key words: air pollution, Heavy metals; moss biomonitoring; AAS analysis, Kosovo area.

CHANGES IN THE HEAVY METAL CONTENTS OF MOSSES DURING LONG-TERM DRY STORAGE

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A heavy metal survey by means of mosses has been carried out in Finland every five years since 1985. The moss samples (*Hylocomium splendens*, *Pleurozium schreberi*) are collected from the plots of the 8th National Forest Inventory. Small amount of each moss sample is analysed soon after collection and the remainder has been stored at the Paljakka ESB. The 108 samples from same 27 plots stored in moss surveys 1985-86, 1990, 1995 and 2000 were reanalysed in 2008. Storage effects were investigated for the heavy metals Cd, Cr, Cu, Fe, Ni, Pb and Zn.

Overall, mean Fe, Pb and Cr concentrations decreased significantly. The drop was greatest on plots with initially high concentrations. The decrease of Cd, Ni and Zn contents was less pronounced. The loss of heavy metals is likely due to drying when cell membranes rupture and surface material is lost. Mean Cu contents increased during storage. Cu is the important nutrient, and probably it is moved in early stage of storage from the old shoots to new ones. Results emphasize the importance of establishing the intended use of stored moss prior to sampling, in order to select and optimize an appropriate storage technique.

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ATMOSPHERIC DEPOSITION OF MAJOR AND TRACE ELEMENTS IN ROMANIA STUDIED BY NAA AND AAS: MOSS SURVEY 2010/2011

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Biomonitoring of air pollutants using mosses has been employed in Romania for the last 20 years by applying nuclear as well as non-nuclear analytical techniques. Prior to 2010/2011 moss survey, surveys in 1995 and 2000 as well as some individual projects ("NATO Science for Peace", bilateral Romania-JINR projects) covered different areas on a local scale with the network varying from 35 to 70 sampling sites. A nationwide deposition moss survey in 2010/2011 undertaken by four Romanian Universities in Targoviste, Galati, Iasi, and Baia Mare, comprised 330 sampling sites evenly distributed over 75% of the Romanian territory. A total of 34 elements (Na, Mg, Al, Cl, K, Ca, Sc, Ti, V, Cr, Mn, Fe, Co, Ni, Cu*, Zn, As, Br, Rb, Sr, Cd*, Sb, Ba, Cs, La, Ce, Sm, Tb, Hf, Ta, W, Pb*, Th, and U) were determined in a wide concentration range from 10000 mg/kg for Al and K down to 0.001 mg/kg for some rare earths by two complementary methods: instrumental epithermal neutron activation analysis (INAA) at the reactor IBR-2 in Dubna, RF, and graphite furnace/flame atomic absorption spectrometry (GFAAS /FAAS) (marked with *) in the Multidisciplinary Research Institute for Science and Technologies from Valahia University of Targoviste, Romania. Principal component (factor) analysis was used to identify the most polluted areas and to characterize different pollution sources. Four factors were revealed. Factor 1 is a mixture of light and heavy crustal components. Factor 2 is of anthropogenic origin with Zn (0.82), As (0.73), and Sb (0,92). Relevant scores point out that the main contributors to this factor are sites from Maramures and Ruschita areas. Factor 3 is a mixture of "marine elements" with Cl (0.79) and Br (0.62) while iodine is not reported as it was not determined in all samples. Ca (0.78), K (0.57), and Sr (0.45) may originate from fertilizer components. Factor 4 comprises of Pb (0.63), Cd (0.66), and Cu (0.64) which may arise from gasoline usage to the copper mining industries. From the present results it can be concluded that atmospheric deposition of trace metals is a considerable problem in the northern and western parts of Romania. This study contributes to the national monitoring system of Romania for long range transported elements of air pollutants, and along with epidemiological data it may serve for baseline human health risk assessments.

BIOMONITORING OF HEAVY METALS IN PLAYGROUNDS IN ATHENS, GREECE

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The purpose of the study was to explore the spatial pattern of heavy metals' load in playgrounds located in the greater region of Athens. To this purpose, a biomonitoring exploration of Fe, Mn, Zn, Mg and Ca was conducted, using leaves of *Platanus orientalis* L. and *Cercis siliquastrum* L. trees, grown in playgrounds.

Methods used: **i)** Sampling and samples' treatment: Leaves of the plants were collected from 16 places scattered throughout Attica, namely: Agia praskevi, Argyroupolis, Aspropyrgos, Votanikos, Dafne, Ekali, Alimos, Glyfada, Heraklion, Keratsini, Patisia, Perama, Pireas, Ilion (Tritsis' park) Chaidari, Zographou. Samples from Ekali served as control (low pollution) region. Three to five shrubs per plant species per region were sampled. The leaves of each sample were further divided to two parts; the leaves of the one part were washed with bidistilled water while the leaves of the other part were not washed. **ii)** Analytical procedure: For the determination of the metals a Perkin- Elmer (Model 1100B) Atomic Absorption Unit was used. **iii)** Statistical analysis: Pearson correlation, ANOVA and multivariate methods were used for the analysis of the data.

Results and discussion: Significant correlations were found between Mg_Mn (r=0.2353, p=0.000), Mg_Ca (r=0.2224, p=0.000), Fe_Zn (r=0.5053, p=0.000), Fe_Mg (r=0.1557, p=0.005), Fe_Ca (r=0.2146, p=0.000), Zn_Mg (r=0.1592, p=0.004), Zn_Ca (r=0.2037, p=0.000) and Mg_Ca (r=0.6727, p=0.000). Multivariate analysis revealed highly structured data. Statistically significance difference was observed between washed and unwashed leaves, between tree species and between regions for all the elements. The highest concentrations of Fe were observed at Aspropyrgos regions, obviously due to the metallurgical industry established there. The highest concentrations for Mn and Zn were observed at Votanikos and Keratsini respectively while for Mg and Ca at Agia Paraskevi. For all the elements the lowest concentrations were observed in the low polluted Ekali region.

Conclusion: Leaves of *Platanus orientalis* L. and *Cercis siliquastrum* L. can serve as useful materials for biomonitoring for heavy metals. A high spatial variability of the levels of the measured elements was observed across the playgrounds in the greater region of Athens. The study is still in progress; the samples are further analyzed for the determinations of the other elements.

Keywords: Heavy metals; Biomonitoring; Playgrounds; *Platanus orientalis; Cercis siliquastrum*

ANALYSIS OF MOSS *THUIDIUM CYMBIFOLIUM* DOZ. ET MOLK TO RETRIEVE PAST ATMOSPHERIC ELEMENTAL PROFILE OF GARHWAL STATE OF IN INDIA

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This study is the continuation of previous study on retrospective monitoring and systematic gathering of information about the atmospheric elemental profile of the environment (Pb, Fe, Zn, Cd, Fe and Ni) over the past 100 years, i.e. 1895–1999 from Garhwal region in India by analyzing moss herbarium vouchers specimens as a tool loaned from different intentional herbaria, as well as analysis of fresh moss samples of *Thuidium cymbifolium*, (Doz. et Molk.) Doz. et Molk, harvested during 1999-2013,

A significant increase in atmospheric elemental profile was observed during analysis of moss species of *Thuidium cymbifolium* (Doz. et Molk.) Doz. et Molk. collected from Garhwal states when compared with moss voucher specimens of the same species barrowed from different herbaria on loan of this region. Analyzed elemental profile seems to be a reflection of atmospheric metal load. The lowest metal concentrations were present in analyzed mosses of the voucher specimens belongs to the period, i.e. 1895 and the level increased gradually over a period of time. However, difference in increase in trend was also observed from 0.01 to 5.9 times for different metals over 1895. The maximum significant increase observed for metals Pb was 5.9 times with respect to first metal data of the year 1895 in *Thuidium cymbifolium* (Doz. et Molk.) Doz. et Molk. Major source for the same can not be ruled out from automobiles followed by generators.

For rest of the metals same could be due to overall an increase in metal load resulting from progressive commercialization and unwisely urbanization. The ratios between the content of the various metals in bryophyte tissue increased was not identical for all metals examined. An increase in metal Ni and Cd was quite low till 1999 (1.09 and 0.03 times) while same increased to 1.7 and 0.3 times in 2013. Its source can not be ruled out from Ni Cd batteries are being used in rechargeable cells. While increased values of Cu and Zn (3.1 & 3.9 times) is due to its use in agricultural production in the country.

Present study is an effort to bridge the existing gap in terms of information and analysis of the elemental data of current and past that it provides information on retrospective atmospheric elemental data and it's an increasing trend over the past 100 year from Garhwal region.

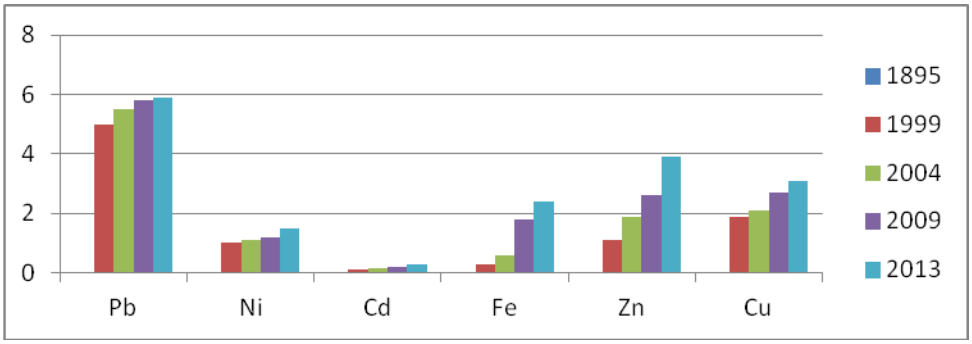

Figure: Percent incease in atmospheric elemental concentrations in moss *Thuidium cymbifolium* (Doz. et Molk.) Doz. et Molk from 1895.

IMPACT OF CLIMATE CHANGE ON STOMATAL OZONE UPTAKE

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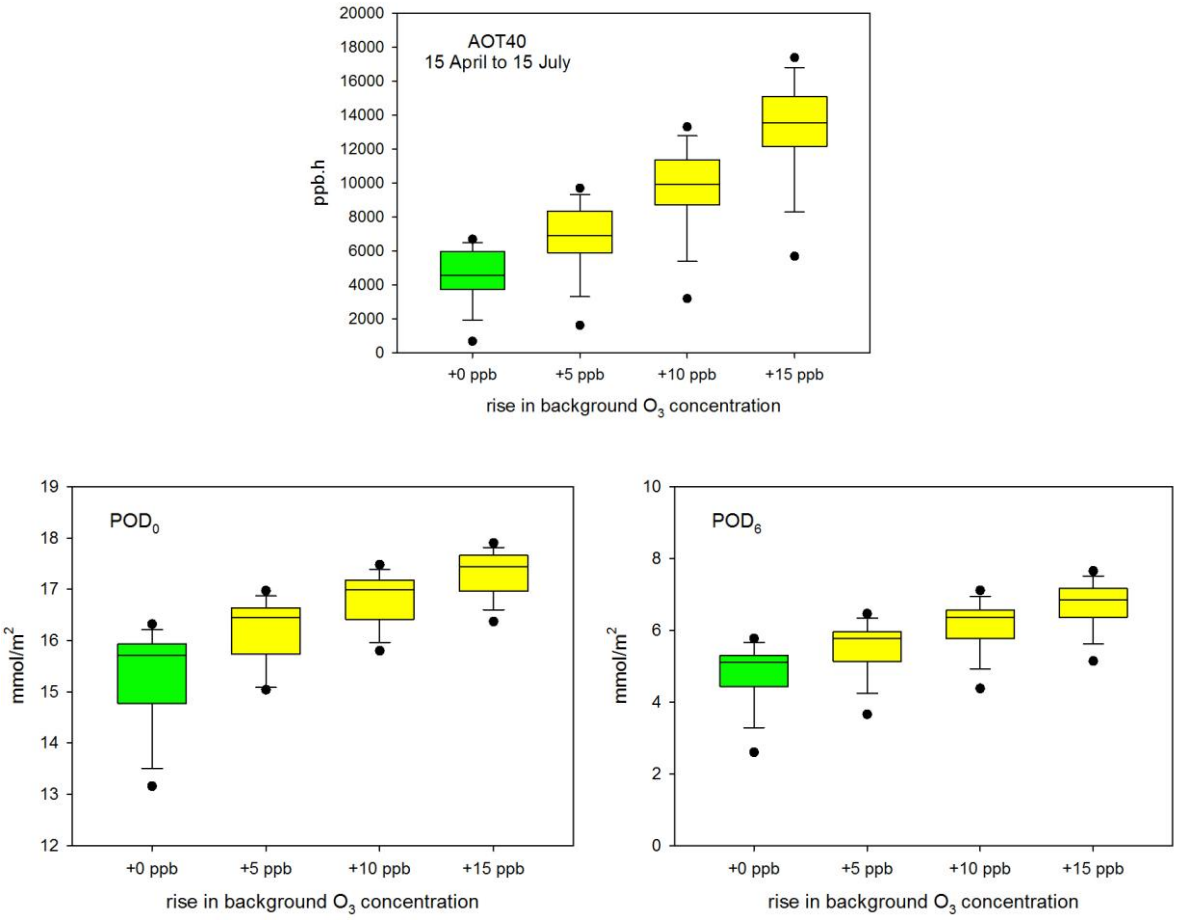
The impact of increasing background O_3 concentrations as well as the impact of increasing air temperatures and changes in precipitation patterns on wheat stomatal O_3 uptake and AOT40 are estimated exemplarily for the Hessian air quality monitoring station Linden, Germany.

O_3 concentration, air temperature and precipitation anomalies are added to the measured data of the years 1997 – 2010. Background O_3 concentrations are increased linearly by 5, 10 and 15 ppb. Mean monthly anomalies of air temperature and precipitation for the time periods 2031-2060 and 2071-2100 are derived from 13 climate change projections for scenario A1B and 3 projections for scenario E1, which stabilises global air temperature change at about 2 °C.

The box-and-whisker plots below illustrate the effect of increasing background O_3 concentration on AOT40 and POD_6 at current climate conditions.



The impact of future climate conditions will be presented and discussed.

EFFECTS OF OZONE ON BEAN *(Phaseolus vulgaris)* IN CROATIA IN 2013

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This paper presents results of the study of impact of ozone on growth and yield on bean (Phaseolus vulgaris), which was implemented in Croatia during 2013 under the International Cooperative Programme on studying the effects of air pollution on natural vegetation and crops (the UNECE ICP Vegetation. http://icpvegetation.ceh.ac.uk/research/Ozone.html)

The research results achieved in field trials in Zagreb, Križevci, M. Lošinj and Jastrebarsko will help to provide a better understanding and resolve numerous and complex problems that result from the harmful effects of ozone on vegetation at local, regional and global scale.

Key words: ICP Vegetation, Croatia, biomonitoring, ozone, bean

NITROGEN IN MOSSES IN CROATIA IN 2006/2010

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During 2006 moss samples were collected from 98 sampling sites evenly distributed all over the territory of Croatia. Moss sampling was repeated in 2010 when 23 additional sampling sites were added and analysed.
Kjeldahl method was used to determine the nitrogen content in all samples.
Descriptive statistics and distribution maps were prepared. Data obtained from these two surveys were compared. The median value of N content in the samples collected in 2006 is 1.60 % and varies from 0.79 % to 3.16 % while the content of N in samples collected in 2010 ranges between 0.71 % and 2.93 % with the median value of 1.49 %. High contents of N (2.32 % - 3.17 %) were found in the regions of Slavonia, Podravina, Posavina and cities Zagreb and Sisak as a result of agricultural and industry activities and traffic.

Key words: Croatia, nitrogen, Kjeldahl method, moss biomonitoring,

BIOMONITORING OF AIR POLLUTION BY TRACE ELEMENTS USING ITALIAN RYEGRASS (*LOLIUM MULTIFLORUM* L. 'LEMA')

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Results of investigations and assessment of air pollution by cadmium, lead and arsenic using Italian ryegrass are presented in this paper. The experiment was carried out in the 2011 growing season in Poznan city and surroundings areas. *Lolium multiflorum* L. 'Lema' exhibits several properties useful for active biomonitoring of air pollution. Plants were exposed at sites varying in environmental characteristics. High cadmium and lead concentrations in leaves were noted in plants exposed within the city area. Canonical variate analysis illustrated variability in concentrations of elements in certain exposure series. The highest arsenic concentrations were observed in the first exposure series, while the highest lead concentrations were observed during the second series. Comparison of heavy metal concentrations at exposure sites to the control site revealed that comparable levels occurred in the city sites and the Agro-ecological Landscape Park. This was an effect of high cadmium and lead levels at city sites, and arsenic at the rural site. The lowest level of measured heavy metals was observed at an exposure site located 15 km from Poznan city in a rural area.

Acknowledgement
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SOME CONSIDERATONS ABOUT ^{15}N-PATTERNS IN MOSS TISSUE AROUND A POULTRY FARM

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A case study in moss monitoring close to a poultry farm in the Eifel mountains shows high accumulation of nitrogen in active exposed moss tissue of *Hypnum cupressiforme* and a parallel increase of δ^{15}N signature. Both accumulation of nitrogen and the δ^{15}N ratio decreased by increasing distance to the immission source (poultry farm).
Most authors who investigate δ^{15}N Patterns in Moos tissue exposed to rural (agricultural) immissions report ^{15}N enrichment in Moss tissue.
Could this be right?
Theoretically, rural N-immission should be primarily deposited as gaseous NH_3. Therefore it would be fractionated and depleted to lighter $^{14}NH_3$ molecules that are energetically favoured to be volatilised into the atmosphere than heavier $^{15}NH_3$ molecules.
If only gaseous ammonia would be immitted, the δ^{15}N signature in moss tissue would decrease and not increase.
But we and most other authors find an enrichment of ^{15}N. The consequence is, that nitrogen would be deposited in different fraction than the gaseous. This would be if the gaseous ammonia would react with particles and during this gas to particle reaction (ammonia with sulphate or chloride in aerosol droplets to $(NH_4)_2SO_4$ or NH_4Cl) the heavier ^{15}N isotope will be favoured (see Fig. 1).
So we will demonstrate that in exposed plant tissue, by increasing δ^{15}N ratio, the particulate deposition predominates the gaseous deposition. Atmospheric reaction processes are important for isotope fractionation.

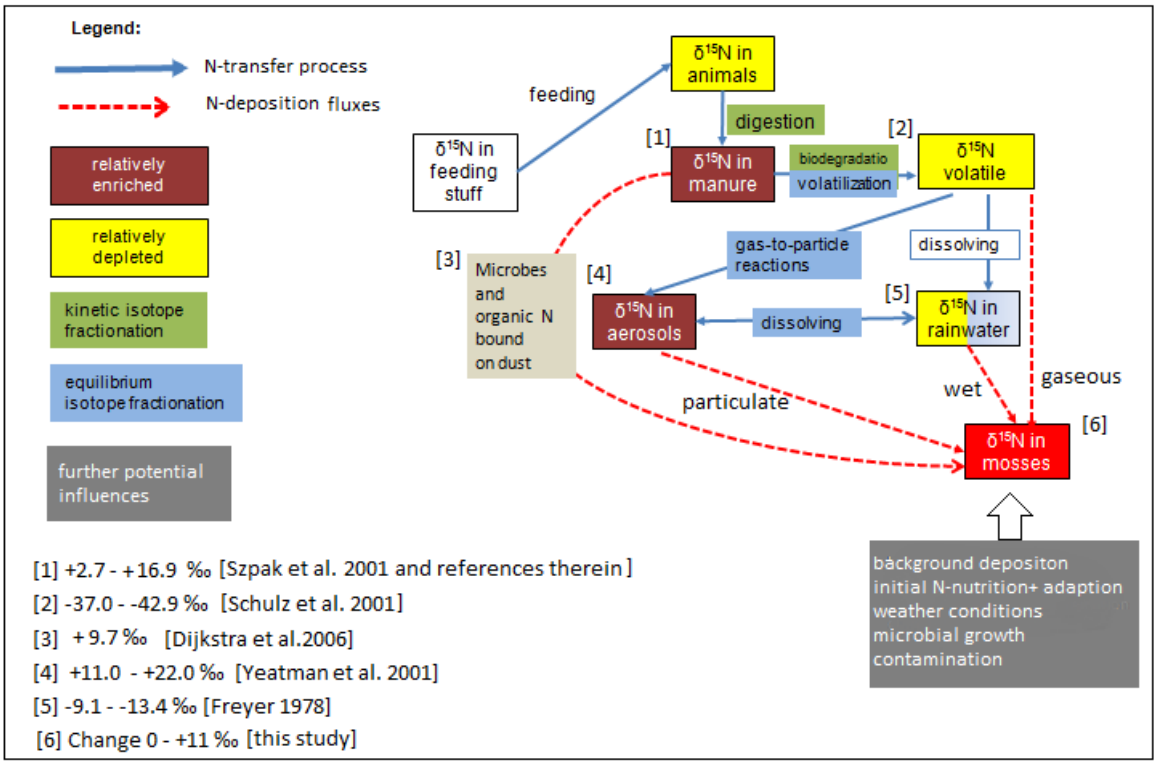

Figure 1: Pathways of N in a livestock farm from feeding to hypothetical deposition fluxes influencing isotopic N signature of moss transplants. Numbers in brackets refer to δ^{15}N values from literature.