



Report No. AS 04/07

# Ozone Umbrella:

## Effects of Ground-level Ozone on (Upland) Vegetation in the UK

Defra Contract 1/3/201, CEH C02158

Interim Report May 2004



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## CEH Edinburgh

At CEH Edinburgh total and stomatal ozone deposition is being measured over upland vegetation at Auchencorth in the Scottish Borders. Instrumentation to measure fluxes in remote areas, without access to mains power, is also being developed and is currently undergoing test at the Easter Bush grassland site, near to CEH Edinburgh. Recent results from Auchencorth and the new instrumentation are described below.

### Stand-Alone Ozone Flux Monitor

The system is based around the ROFI ozone sensor, built by CEH following the method of Gusten 1992. This sensor monitors rapid changes in ozone concentration by measuring variations in light emission from a disk coated with an ozone-sensitive dye. A standard ozone analyser is also needed to monitor ambient concentrations and calibrate the response of the ROFI. Additional instruments are included to measure atmospheric turbulence, water-vapour flux, air temperature, humidity, surface wetness, radiation and soil heat flux. To allow the use of a wind turbine and solar panel all the instruments are chosen for their low power consumption and ability to run on a 12V power supply. At present a Campbell 23X is being used to log and analyse the data as a laptop or pc requires too much power. Figure 1 below, illustrates the configuration of the system.

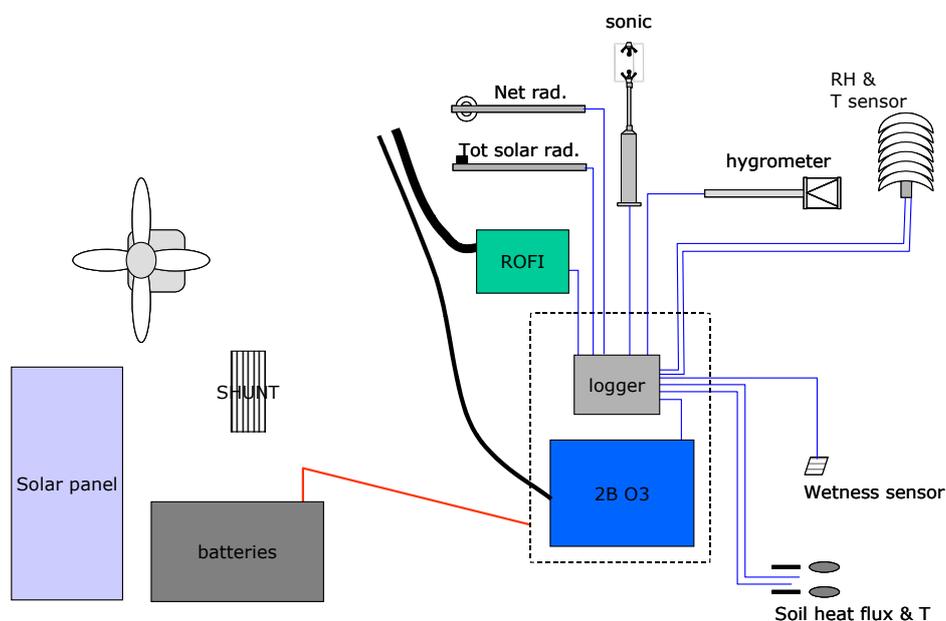


Figure 1. Outline of the stand-alone O<sub>3</sub>/H<sub>2</sub>O flux monitoring system (SAO3).

The system is currently being set-up at our Easter Bush field site, near to CEH Edinburgh, for testing.

### Results from Auchencoth Moss

The Auchencorth Moss field site just south of CEH, in the Borders region, and has been monitoring total ozone deposition for 10 years, with periods of water-vapour flux measurements which can be used to determine stomatal ozone flux. In 2002 continuous monitoring of water-vapour and carbon-dioxide fluxes was added to the site, which will allow detailed study and modelling of the partitioning between stomatal and non-stomatal ozone fluxes. The vegetation at the site is typical of rough, upland moor or bog with large amounts of *Deschampsia flexuosa*, *Eriophorum* varieties, various mosses and other wild species. Data analysis is ongoing and a preliminary results show that stomatal uptake is ~30% of total ozone deposition during the summer months.

## CEH Bangor

In 2003, thirty three species were exposed to ozone as a screening study to investigate whether predicted increases in ambient ozone pollution may affect upland vegetation. Plants were mostly collected from Snowdonia or grown from seed of known sources. An episodic ozone regime was applied for ten weeks. Plants were assessed weekly for ozone injury and senescence and were harvested at the end of the exposure. Visible injury due to ozone was observed on seven of the species tested. Eleven of the species tested showed either increased or premature senescence in response to ozone. Some species showed a significant reduction in biomass of ozone-treated compared to control plants. As some of the species were sensitive to ozone and others showed no effects, it is possible that there may be changes in species composition in upland communities due to ozone pollution. Some plants from the screening study will be grown as small artificial communities in 2004-2005. Current ozone concentrations in Snowdonia and predicted changes in ozone concentration in upland areas will be used to devise an appropriate ozone regime and the effects of predicted ozone changes on these small communities will be investigated.

## Introduction

So far, only a small proportion of the semi-natural vegetation species have been tested for sensitivity to ambient ozone pollution. Of the species tested, many show effects of ozone including ozone-specific visible injury and alterations in biomass. There is also some evidence that natural selection has occurred for some species to favour those with increased ozone resistance. The nature of ambient ozone pollution is thought to be changing, with an increase in the background level of ozone. This is thought to be particularly significant for upland vegetation communities, as the background ozone concentration in upland rural areas is already high and near to the current threshold for ozone pollution of 40 ppb.

Currently the effects of ozone on the majority of semi-natural vegetation species of the UK uplands are not known, and therefore the consequences of predicted increases in ambient ozone pollution cannot be determined. This study aims to screen a range of species from the uplands of Snowdonia for sensitivity to ozone and to begin to characterise the nature of the responses.

## Methods

### *Plant material*

Plants were collected from Clogwyn Coch (SH 603555) and Cwm Idwal (SH 640597 – Figure 1) in May and June 2003 and potted into acidic compost mix (40 litres ericaceous compost mixed with 25 litres sharp sand), calcareous compost mix (3 parts John Innes compost No2 mixed with 1 part gritty sand and 1 part peat) or multi-purpose compost as appropriate (Table 1). Plants were raised in sheltered but outdoor conditions in Bangor.

In late July 12 plants per species were numbered from 1 (smallest) to 12 (largest). There were three size classes (1-4, 5-8 and 9-12), and one individual plant per size class was randomly selected for each of the four solardomes. At the end of July the following grass species were cut back to 7cm: *Holcus lanatus*, *Agrostis capillaris*, *Agrostis vineale*, *Anthoxanthum odoratum*. *Festuca ovina*, *Festuca rubra* and *Nardus stricta* were not cut back as they were less than 7cm tall. Plants were transferred to the solardomes on 31<sup>st</sup> July and exposure to ozone started on 1<sup>st</sup> August. Plants were sprayed to control pests as necessary during the exposure period.

### *Ozone exposure*

Two domes were controls, with ozone added to charcoal filtered air to give a total ozone concentration of 20 ppb. An episodic ozone regime was applied to the other two domes, with concentrations rising to 80 ppb on day 1, 100 ppb on days 2 and 3, and 80 ppb on day 4. Ozone concentrations remained at 20 ppb at all other times. The ozone exposure was for 10 weeks. Total ozone exposure for the ozone treatment was 18.3 ppm.h AOT40.

**Table 1:** Origin of the species exposed in the solardomes. The compost mix used for each species is also shown.

Collection date & origin	Species	Soil type
29-05/Clogwyn Coch	<i>Arabis petraea</i> (northern rock-cress)	Calcareous
29-05/Clogwyn Coch	<i>Armeria maritima</i> (thrift)	Acidic
29-05/Clogwyn Coch	<i>Campanula rotundifolia</i> (harebell)	Acidic
29-05/Clogwyn Coch	<i>Cryptogramma crispa</i> (parsley fern)	Acidic
29-05/Clogwyn Coch	<i>Carex demissa</i> (common yellow sedge)	Acidic
29-05/Clogwyn Coch	<i>Ranunculus acris</i> (meadow buttercup)	Multi-purpose
29-05/Clogwyn Coch	<i>Rumex acetosella</i> (sheep's sorrel)	Acidic
29-05/Clogwyn Coch	<i>Senecio fluviatilis</i> (broadleaved ragwort)	Multi-purpose
12-06/Cym Idwal	<i>Cirsium palustre</i> (marsh thistle)	Acidic
12-06/Cym Idwal	<i>Galium saxatile</i> (heath bedstraw)	Acidic
12-06/Cym Idwal	<i>Oxalis acetosella</i> (wood-sorrel, sub-montane)	Acidic
12-06/Cym Idwal	<i>Oxyria digyna</i> (mountain sorrel)	Acidic
12-06/Cym Idwal	<i>Plantago lanceolata</i> (ribwort plantain)	Multi-purpose
12-06/Cym Idwal	<i>Potentilla erecta</i> (tormentil)	Acidic
12-06/Cym Idwal	<i>Saxifraga stellaris</i> (starry saxifrage)	Acidic
12-06/Cym Idwal	<i>Thymus polytrichus</i> (wild thyme)	Multi-purpose
12-06/Cym Idwal	<i>Viola lutea</i> (mountain violet)	Acidic
17-06/Cym Idwal	<i>Anthoxanthum odoratum</i> (sweet vernal grass)	Acidic
17-06/Cym Idwal	<i>Carex demissa</i> (common yellow sedge)	Acidic
17-06/Cym Idwal	<i>Carex laevigata</i>	Acidic
17-06/Cym Idwal	<i>Carex panicea</i> (carnation sedge)	Acidic
17-06/Cym Idwal	<i>Scirpus cespitosus</i> (deergrass) (= <i>Trichophorum cespitosum</i> )	Acidic
17-06/Cym Idwal	<i>Juncus effusus</i> (soft rush)	Acidic
17-06/Cym Idwal	<i>Nardus stricta</i> (mat-grass)	Acidic
17-06/Cym Idwal	<i>Eriophorum angustifolium</i> (common cotton grass)	Acidic
17-06/Cym Idwal	<i>Juncus squarrosus</i> (heath rush)	Acidic
17-06/Cym Idwal	<i>Carex echinata</i> (star sedge)	Acidic
17-06/Cym Idwal	<i>Narthecium ossifragum</i> (bog ashpodel)	Acidic
sown (Gwynedd)	<i>Agrostis capillaris</i> (common bent)	Acidic
sown (Gwynedd)	<i>Agrostis vineale</i> (brown bent)	Acidic
sown (Wiltshire)	<i>Anthoxanthum odoratum</i> (sweet vernal grass)	Acidic
sown (Oxfordshire)	<i>Festuca ovina</i> (sheep's fescue)	Acidic
sown (Somerset)	<i>Festuca rubra</i> (red/creeping fescue)	Multi-purpose
sown (England)	<i>Holcus lanatus</i> (Yorkshire fog)	Multi-purpose
Nursery stock	<i>Dryas octopetala</i> (mountain avens)	

### Assessments and Physiological Measurements

Plants were assessed weekly for ozone injury and senescence. Stomatal conductance measurements were carried out on selected species using a Delta-T porometer on days of stable weather conditions.

### Intermediate harvests

After 4 and 8 weeks of exposure to ozone, most grass species were cut back to 7cm (*Holcus lanatus*, *Agrostis capillaris*, *Agrostis vineale*, *Anthoxanthum odoratum* (wild and seed-grown forms), *Festuca ovina* and *Festuca rubra*). *Nardus stricta* was not cut back due to its low stature – material above 7cm was predominantly flowers. The clippings cut after 4 weeks of exposure were sorted into green and senesced (>25% of the leaf senesced) material. The

clippings cut after 8 weeks of exposure to ozone were not sorted as the leaves were mainly healthy or less than 25% senesced.

### Final Harvest

After 10 weeks of exposure to ozone all plants were harvested. Above ground biomass was removed and dried at 65°C for a minimum of 7 days. For some species the above ground biomass was sorted into green and senesced (>25% of the leaf senesced) leaf material and flowers (*Carex echinata*, *Carex demissa*, *Carex panicea*, *Carex laevigata*, *Nardus stricta*).



Figure 1: Cwm Idwal, One of the sites for collection of plant material

## Results

Ozone specific injury was observed on *Potentilla erecta* (Figure 2a), *Carex echinata* (Figure 2b), *Dryas octopetala* (Figure 2c), *Oxalis acetosella*, *Nardus stricta* (Figure 2d), *Eriophorum angustifolium* (one leaf each of four plants), *Carex panicea* (although the same symptoms were later observed on the control plants) and *Carex demissa* (one leaf of one plant). There was possibly slight injury on *Scirpus cespitosus*, but the potential injury symptoms were very transient and senescence had occurred before the injury symptoms could be confirmed. On *Viola lutea* symptoms appeared to be ozone injury initially, but preceded senescence. The same symptoms appeared although to a lesser extent and later in the exposure period, on the control plants.

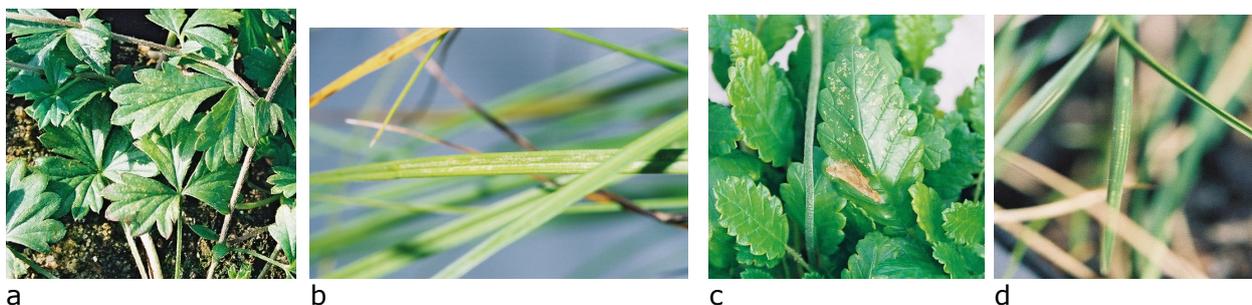
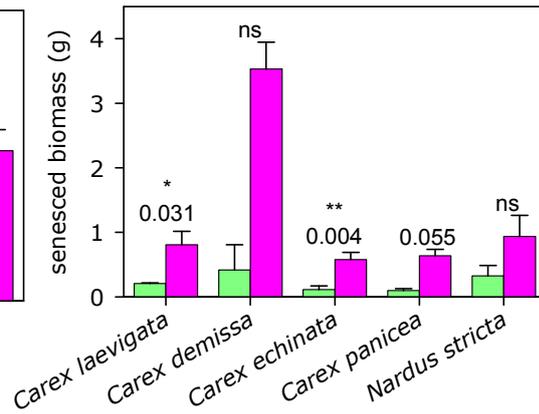
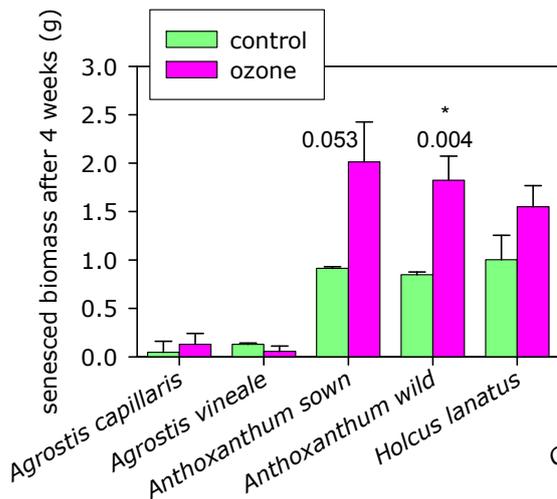


Figure 2: Ozone-specific injury on: a. *Potentilla erecta*; b. *Carex echinata*; c. *Dryas octopetala*; d. *Nardus stricta*

Weekly assessments showed increased / premature senescence on several other species, including *Scirpus cespitosus*, *Viola lutea*, *Cirsium palustre*, *Carex laevigata*, *Carex panicea*, *Carex echinata*, *Carex demissa*, *Holcus lanatus*, *Agrostis capillaris*, *Agrostis vineale* and *Anthoxanthum odoratum*.

Grasses cut after exposure to ozone or control conditions for four weeks were sorted into healthy and senesced leaves. The biomass of the senesced leaves is shown in Figure 3 and was significantly increased by ozone in *Anthoxanthum odoratum* (wild origin) and greatly but not significantly increased for *Anthoxanthum odoratum* (sown). There were no senesced leaves for either *Festuca rubra* or *Festuca ovina* at this harvest. Very little plant material of the seven species for which intermediate harvests were performed had senesced prior to subsequent harvests.

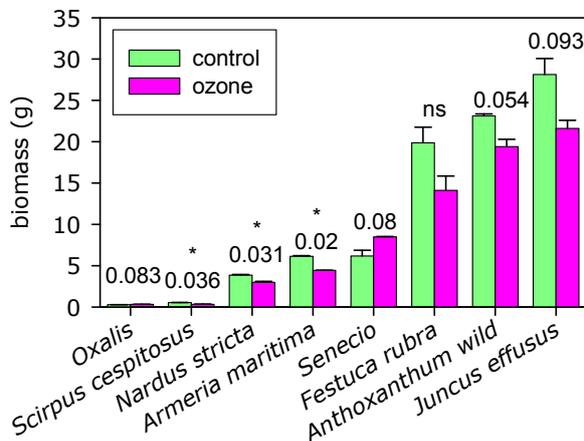


**Figure 3:** Biomass of senesced leaves of selected grasses after exposure to ozone or control conditions for 4 weeks.

**Figure 4:** Biomass of senesced leaves of selected species after exposure to ozone or control conditions for 10 weeks.

The leaves of several species were sorted into green and senesced (>25% of leaf senesced) at the final harvest. There was a significant increase in the dry weight of senesced material of plants exposed to ozone compared to control plants for *Carex echinata* and *Carex laevigata* (Figure 4). There was also a large but not significant increase in the dry weight of senesced material in *Carex panicea* and *Carex demissa*. There was an increased dry weight of flowers in *Carex echinata* for plants treated with ozone.

Significant decreases in biomass in response to ozone were observed for some species. *Scirpus cespitosus* (collected from wild, 42%); *Armeria maritima* (28%); *Nardus stricta* (22%), and a large but not significantly different decrease for *Juncus effusus* (23%) and *Anthoxanthum odoratum* (collected from wild, 16%). There was also a large but not significant increase in the biomass of *Senecio fluviatilis* exposed to ozone compared to those grown in control conditions (Figure 5).



**Figure 5:** Total above ground biomass of selected species exposed to ozone or control conditions for 10 weeks.

The grasses cut after four weeks were cut back to 7cm again after a further 4 weeks, then again after a further 2 weeks. Relative growth rates were calculated and analysis of the relative growth rate showed no significant differences in relative growth rate with ozone treatment compared to control for any of the species tested between the harvest after 4 weeks and the harvest after 8 weeks, although the relative growth rate was reduced in ozone treated *Festuca ovina* plants (data not shown).

Relative growth rate of *Holcus lanatus*, *Festuca rubra* and *Festuca ovina* was decreased in ozone treated plants compared to control treated plants. However, these differences were not statistically significant (data not shown).

After a further 2 weeks (exposure to control or ozone conditions for a total of 10 weeks), the relative growth rate of all plants had decreased compared to during the previous harvest interval.

Stomatal conductance was reduced in *Carex laevigata* plants growing in ozone treated domes compared to control domes. No differences in the stomatal conductance in response to ozone were detected in the other 11 species tested.

## Discussion

These data indicate that a wide range of species from upland vegetation communities may be sensitive to future predicted ambient ozone scenarios. Eighteen of the thirty-three species tested in this study showed one or more effects of ozone in ozone-treated compared to control conditions. Visible injury represents a biochemical response in the plant, but may not represent a loss in ecological fitness, whereas increased/premature senescence and biomass reductions due to ozone may result in reduced fitness. This may be particularly important in upland communities, where growth is often slow and vegetative persistence is particularly important. The differential extent of sensitivity in different species could potentially lead to changes in community composition and biodiversity.

The data from this screening study shows that the sedges (*Carex* spp) appeared particularly sensitive to ozone, as each of the sedge species tested showed one or more symptoms of ozone sensitivity. In addition, many of the species characteristic of upland vegetation showed some symptoms of ozone sensitivity (e.g. *Scirpus cespitosus*-Deergrass; *Potentilla erecta*-tormentil) so that potentially the nature of some of the characteristic upland communities could change.

## Conclusions

Many of the upland species screened show effects of ozone pollution. The effects of ozone pollution include visible injury, increased/premature senescence and effects on biomass. Some species are more sensitive to ozone than others, which could potentially lead to changes in competitive ability and biodiversity based on future predicted ozone scenarios for the uplands. Effects of predicted ozone scenarios on small communities will be investigated in 2004/2005.

## University of Bradford

This part of the programme is focussed on assessment of impacts of ozone on different plant communities within the Yorkshire Dales National Park (YDNP). One year's work has now been completed. Progress in meeting the key objectives for this first phase of the project can be summarised as follows.

### (a) Identify field sites representing different communities of high conservation significance within YDNP

Six field sites of high conservation significance have been identified, including woodlands, limestone pavements and traditionally managed hay meadows (Table 1). These sites are all within 30km of each other, providing the basis for the planned evaluation of the variation small-scale impacts of ozone in upland areas. They also provide the basis for comparison of variation in sensitivity of different populations to ozone.

Table 1: Summary of field sites in YDNP (<http://www.atmosci.ceh.ac.uk/projects/dales.htm>)

Site	Communities
Yockenthwaite Meadows	Traditionally managed hay meadows
Conistone Old Pastures	Limestone pavement Meadow
Bastow Wood	Open pasture woodland Limestone pavement
Grass Wood	Diverse woodland
Ribblehead	Diverse limestone grassland
Scar Close	Limestone pavement

In addition, a field station for measurement of ozone concentrations and stomatal conductance has been established at Ribblehead, in collaboration with English Nature.

Each site has been surveyed and species lists prepared. Seed collections were made for 60 species during the summer of 2003, with 36 species being collected from more than one site, thus allowing the responses of different populations to be compared.

### (b) Assess the sensitivity to ozone of species characteristic of different habitats within YDNP

Plants grown from the field seed collections have been exposed to ozone in a closed chamber system that allows the maintenance of controlled temperature and humidity. A standard fumigation regime (80ppb; 8 hours day<sup>-1</sup>; 15 days) has been used; this has been selected as characteristic of UK ozone episodes. Difficulties with germination and growth from field-collected seed have been experienced in the case of some species; this was anticipated, but it is essential in this project to work with field populations rather than commercial seed. Within the first year, experiments have been completed for 13 species, and we expect that this will increase to 25-30 in the next six months.

For this reason, the main findings to date must be treated as preliminary. The most consistent finding across species is that the strongest effects of ozone are on root biomass and root:shoot ratio; relatively few populations have shown significant effects on growth above ground. The physiological and ecological significance of effects on root:shoot partitioning in the different habitats is a potential factor for further investigation; such effects might, for example, be more significant in limestone pavements than in hay meadows.

There is some evidence of significant differences in response to ozone between populations of the same species. For example, the woodland population of *Lotus corniculatus* is more sensitive to ozone than the limestone pavement population, and there are differences between populations of *Plantago lanceolata* in the extent to which ozone causes reductions in leaf width.

Work over the next year will aim to:- investigate the sensitivity of a wider range of species; compare ozone exposures and stomatal behaviour at contrasting field sites; compare populations for more species; and investigate the influence of ozone exposure on herbivory.

# University of Lancaster

## Experimental set-up

8 growth cabinets (75 x 75 x 75 cm each) in total: 4 control, 4 "2050" O<sub>3</sub> scenario. These are in a greenhouse with artificial light; all cabinets are air conditioned (20-25°C) with approximately one air change per minute.

- control = unfiltered air (will include current daytime pollution episodes but not the high background expected in uplands in the future)
- "2050" = background of up to 50 ppb (i.e. even at night) with daytime peaks (up to 80 ppb)
- 2 levels of water supply (well-watered and moderate drought)

## Choice of species for preliminary work:

(numbers in brackets are the Relative Sensitivity Index to ozone from the review by Fuhrer *et al.* – see <http://www.ozoneworkshop.ivl.se/> , workshop report, pp. 183-198)

### Grasses

Phleum pratense (0.64)

Dactylis glomerata (0.99)

Lolium perenne (1.04)

Bromus erectus (1.11)

### Legumes

Trifolium repens (0.87)

Lotus corniculatus (0.96)

Onobrychis sativa (1.02)

These were chosen on the basis of contrasting RSIs within species, which could be considered to be included as "upland" vegetation (with the exception of Onobrychis). However, in general, the variability in RSI was not very great.

At a later stage, we will narrow this down to maybe one or two species (sensitive and insensitive) to concentrate on investigating potential mechanisms for ozone damage/tolerance and the interaction with drought.