O3GRASSLAND:

a CEH-Integrating Fund Project

2001-2003

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Foreword

Project O3GRASSLAND was initiated in 2001. The project was a collaboration between three groups: *Biosystems Dynamics* and *Atmospheric Sciences*, both based at CEH-Edinburgh, and *Environmental Pollution*, based at CEH-Bangor. Most of this report is the product of the whole group, with the authorship listed on the front page of this document. However, the following chapters had different first authors:

1. Physiological experiments in solardomes: Felicity Hayes, Patrick Büker and Gina Mills
2. Flux measurements in the field: Mhairi Coyle
3. Biological measurements in the field: Peter Levy and Maureen Murray

Summary

The problem

Ozone is a major pollutant in industrialised societies. The amount of damage to vegetation by ozone has thus far been quantified mostly with simple dose-response functions, which are robust but cannot handle interactions of ozone with other environmental factors well. There is a need to assess the scope for a more ambitious approach that would permit accounting for the effect of different site conditions on ozone sensitivity. We aimed for such an approach, beginning with studying the processes through which ozone is taken up and causes damage. Four damage mechanisms can be distinguished: (1) disrupting photosynthesis, (2) increasing respiration, (3) accelerating senescence, and (4) altering root-shoot allocation. Damage varies with ozone deposition rate and how it is distributed in time, but it also depends on the plant species, its physiological state, and the environmental conditions. Because of these interactions, it is desirable to quantify damage under field conditions, particularly if the goal is to predict when real vegetation is at risk from ambient ozone. However, the four damage mechanisms can only be quantified well under controlled conditions. An instrument for scaling up leaf-scale data measured under controlled conditions to the field-scale would thus be useful for predicting ozone damage. The O3GRASSLAND project, a CEH-Integrating-Fund initiative, was aimed at developing such a tool in the form of a process-based model. The project ran for two years: May 2001 – April 2003.

The project

In O3GRASSLAND, the solardomes at CEH-Bangor were used for detailed measurements of damage caused by ozone to grass (*Lolium perenne*) and clover (*Trifolium repens*). The plants had been cloned from material taken from a grass field near CEH-Edinburgh. At this latter field site, measurements of O_3, CO_2 and H_2O fluxes between vegetation and atmosphere were made using the eddy covariance method. Vegetation growth rate was also measured at this field site.

A simulation model that accounts for the four damage mechanisms triggered by ozone was constructed and applied to the field site. Parameters of the model were taken from the solardome experiments.

Results

The solardome experiments yielded valuable information on the response of grass and clover to elevated O_3. The field flux measurements quantified intra- and inter-seasonal variation in gas fluxes between a grass sward and the atmosphere. The process-based model for effects of O_3 and other environmental conditions on grass and clover simulated the fluxes of O_3, CO_2 and H_2O observed in the field with reasonable accuracy. The effects of environmental conditions on the risk of ozone damage were assessed by application of the model to various hypothetical environmental scenarios.

The productive collaboration, in O3GRASSLAND, of experts in physiological studies under controlled conditions, field measurements of gas fluxes, and process-based modelling, confirmed the efficacy of interdisciplinary approaches to problems in environmental science.

General introduction

The ozone problem

The concentration of ozone (O_3) has increased at 1-2% per year during the last two to three decades at middle and high latitudes over Europe (Guicheret 1989), and present-day tropospheric concentrations of O_3 are considered to be damaging (Skarby et al. 1993). To estimate the damage that ozone may cause to vegetation, statistical approaches are generally adopted. Most recently, yield and biomass changes have been related to AOT40, the Accumulated Ozone exposure above a Threshold of 40 ppb O_3 (Fuhrer et al., 1997). The latter is the product of concentration and time and fits the data better than previously used parameters such as the 7 hour mean. However, AOT40 is not the absorbed physiologically active dose (Fowler et al., 1997), and thus its use still leaves much variation in dose-response data unaccounted for. For example, there is a tendency for AOT40 to overestimate the magnitude of O_3 effects because the environmental conditions leading to large O_3 concentration are also correlated with high VPD and reduced stomatal conductance and flux of ozone. Statistical approaches such as correlations of plant damage with AOT40 ignore interactions of ozone with other environmental factors, and also ignore variability in the sensitivity of the plants to ozone. The first step beyond AOT40 would be to replace "exposure" with "uptake", i.e. statistically relating damage to cumulative ozone-uptake rather than exposure. That step is already being taken for some species of crops and trees (Reich, 1987; Fowler et al., 1998, Emberson et al., 2000). Whereas this is obviously a useful next step, uptake-based methods remain statistical in nature and do not account for the physiological response of the vegetation, and how it differs between genotypes and interacting environmental conditions.

The O3GRASSLAND project

In this project, we explored the scope for new methods of quantifying and predicting ozone uptake by vegetation and the resulting damage, that have greater generality then the use of the AOT40 index and other statistical approaches. Grassland, the most common vegetation type in the UK and not yet well represented in the ozone literature, was chosen as the study vegetation type. Instead of deriving empirical damage relationships, we developed a process-based model for uptake of ozone by vegetation and the physiological response of the plants, starting from a similar type of model derived for wheat (Van Oijen and Ewert 1999). To parameterise the model, detailed physiological measurements were carried out in the solardomes of CEH-Bangor, and to assess the magnitude of the ozone deposition onto and into foliage, detailed flux measurements were carried out on a grassland site near CEH-Edinburgh. In Chapter 1, the solardome experiments will be described, Chapter 2 presents the field flux measurements, Chapter 3 describes measurements of grass growth and photosynthesis in the field, and Chapter 4 shows the integration of this information in the process-based model. The report is concluded by a general discussion of the results, and some appendices with administrative project information.

1. Physiological experiments in solardomes

Abstract

Ozone exposure experiments were conducted at CEH Bangor in 2001 and 2002 to provide information on the growth and physiological responses of the component species of grassland to ozone, for use in the mechanistic model developed by Marcel van Oijen and colleagues at CEH Edinburgh (Chapter 4).

In 2001, component species of the grassland site adjacent to CEH Edinburgh were grown from seed and screened for ozone sensitivity. A longer exposure was then carried out for *Lolium perenne, Trifolium repens* and *Cirsium arvense.* The biomass of *Trifolium repens* was significantly less in the ozone-treated domes compared to the charcoal filtered domes, but there were no significant differences in the biomass of either *Cirsium arvense* or *Lolium perenne.* In 2002, plants of perennial rye-grass (*Lolium perenne*) and white clover (*Trifolium repens*), from the grassland site adjacent to CEH Edinburgh used for flux measurements (Chapter 2), were sent to CEH Bangor several months before the start of the exposure experiment. Clonal material from these parent plants was grown into individual plants for use in solardome exposure experiments. Three large containers of each of the following combinations were placed in each of four solardomes: all clover, all perennial rye grass, or 4 clover plants on the inside, surrounded by 8 outer rye grass plants. Two solardomes were ventilated with filtered air to which 30 ppb of ozone was added (background ozone treatment), and two were ventilated with the following 7 day episodic ozone regime for 12 weeks: 7h per day at 80 ppb for day 1, at 100 ppb for days 2 and 3, and 80 ppb for day 4; 30 ppb of ozone was added at all other times. The episodic ozone treatment amounted to an AOT40 of 21.9 ppm.h over the 12 weeks.

Six weeks after the start of exposure, all of the plants in the containers were cut to 7 cm above soil level. Sixty nine percent of the clover leaves per pot showed visible injury in the high ozone treatment compared to 0.5% in the background ozone treatment. The leaves of *Lolium* showed a significant increase in senescence in the high ozone treatment compared to the background treatment, but there was not a significant ozone effect on total dry weight or leaf dry weight for this species. At the final harvest, the canopy was divided into layers: significant effects of ozone on clover occurred in the uppermost layers of the canopy, but not in the lowest layer; no significant effects on the dry weight of *Lolium* were found even though the canopy was visibly and quantifiably more senescent. Physiological measurements showed that carboxylation efficiency was reduced by ozone in both *Trifolium repens* and *Lolium perenne*, even though in the *Lolium perenne,* the biomass was not significantly reduced. These results will be used for parameterising and testing a process-based model in Chapter 4.

Introduction

Several reviews and individual papers have been published describing the sensitivity of semi-natural vegetation species to ozone (e.g. Ball et al. 1999; Bergmann et al. 1996; Bungener et al. 1999; Davison and Barnes 1998; Mortensen and Nilsen 1992; Pleijel and Danielsson 1997; Warwick and Taylor 1995). Ozone exposure-response studies have shown that biomass reductions can occur in sensitive vegetation species in ambient air conditions. Sensitive species include *Trifolium repens, Plantago lanceolata* and *Festuca rubra*. However, biomass increases are predicted for some of the semi-natural vegetation species, e.g. *Bromus erectus* and *Festuca ovina*, and other species such as *Lolium perenne* are thought to be insensitive to ozone. Studies have shown that some plants such as sheeps fescue (*Festuca ovina*) preferentially partitioned biomass to shoots rather than to roots, which may give the plants a competitive disadvantage when they are in species mixtures, even though there appears to be no significant effect (on a total weight basis) when the plants are grown singly (Cooley and Manning 1987). Biomass partitioning in clover has been shown to be affected by ozone. However, there is less information about biomass partitioning in response to ozone for *Lolium perenne* and other grasses. In white clover, damaged leaves may be replaced at the expense of the stolons (Wilbourn et al. 1995).

Reductions in yield of a *Trifolium repens* and *Lolium perenne* mixture established from seed then exposed to ozone episodes using an unenclosed field fumigation system showed significant effects on both clover yield and total yield (Wilbourn et al. 1995). The reductions in total yield were due largely to a 32.5% reduction in the yield of the clover component. However, not all grass:clover mixtures are dominated by effects on the clover component. In a study by Nussbaum *et al* (1995) the effect on total yield of a *Trifolium repens* (ladino) and *Lolium perenne* mixture grown from seed and exposed in open-top chambers was largely determined by the effect on *Lolium perenne.* This was because the yield and proportion of clover was reduced in all ozone treatments, but for *Lolium perenne* at low ozone concentrations the yield increased, whereas it was decreased at high ozone concentrations with an episodic ozone regime (an AOT40 of approximately 6000 ppb.h over 4 weeks, peak ozone concentrations of approximately 200 ppb during selected episodes).

Aims:

- To assess the component species of a grassland for sensitivity to ozone.
- To quantify impacts of O_3 on *Lolium perenne* and *Trifolium repens* using plants cloned from the CEH Edinburgh field site
- To provide parameter values and test data for modelling

Materials and Methods

Solardomes

Four solardomes (Figure 1.1) were used for exposure of plant material to ozone, whilst maintaining near-ambient meteorological conditions (except rainfall). Plants were kept well-watered using a mist irrigation system, with regular additional watering by hand during periods of warm weather. Ozone concentrations in each solardome were measured every 30 minutes using a ‘Dasibi’ ozone analyser, and temperatures were recorded every five minutes.

Figure 1.1. Solardomes at CEH Bangor.

Experiments conducted in 2001

Plant material

The plants used in 2001 were grown from seed.

Experimental design

Screening Trial

Plants were exposed three per 7.5 litre pot, 5 pots per dome, 2 domes per treatment for three weeks. The species used were *Trifolium repens, Trifolium pratense*, *Cirsium arvense, Lolium perenne, Dactylis glomerata, Rumex obtusifolius, Sonchus oleraceus* and *Sonchus apser*. All of these species were present in the Easter Bush field where ozone flux measurements were made (Chapter 2).

Main Experiment

Plants of white clover (*Trifolium repens*), creeping thistle (*Cirsium arvense*) and perennial rye-grass (*Lolium perenne*) were exposed to ozone in single species pots in the same ozone regime as the screening experiment. Plants were grown in 15 litre pots, 5 plants per pot (the same density as the screening trial), 8 pots per dome per species. *Trifolium repens* and *Lolium perenne* were exposed to the ozone regime for 10 weeks. *Cirsium arvense* was exposed for six weeks.

Ozone exposure

The ozone regime was two domes with charcoal-filtered air (CF), two domes with CF + 8h @ 80 ppb day 1, 8h @ 100 ppb day 2, 8h @ 100 ppb day 3, 8h @ 80 ppb day 4, CF at all other times, with an AOT40 of approximately 1600 ppb.h per week.

Measurements

- Final harvest – above ground biomass was determined for each species.
- Stomatal conductance measurements – using a Delta-T porometer and ambient light levels.
- Photosynthesis measurements on selected species using a PP-Systems CIRAS
- Visual estimates of senescence and ozone-specific injury

Data analysis

Statistical analysis was performed using Minitab ANOVA.

Experiments conducted in 2002

Plant material

Lolium perenne and *Trifolium repens* plants were propagated from turf samples from a field near Edinburgh (Chapter 2). Clover was propagated by taking cuttings from stolons and *Lolium* by dividing plants and planting individual tillers (with attached root).

Experimental design

Large containers were used so that plants could be exposed as part of a community rather than as individuals. The containers used were 35.5 cm x 45 cm x 25 cm deep and filled with multipurpose compost (‘Gem’ tub and planter). Holes were drilled in the bottom for drainage, and the pots were lined with perforated plastic sheeting to discourage roots from growing out through the bottom of the pots.

Each pot contained 12 plants, consisting of 4 central plants and 8 additional plants around these. Each solardome had 3 pots each of Lolium and clover monocultures, and four pots containing a mixture of Lolium and clover. In each mixture, the four central plants were clover. Plants were exposed in the solardomes for 12 weeks.

Ozone exposure

Four solardomes were used for exposure, with ozone added to charcoal filtered air to give the required ozone concentrations. Two were used as controls, with ozone added to give total ozone concentrations in each dome of 30 ppb. A computer controlled episodic ozone profile was given over the course of each week to the other two domes. The ozone exposure was programmed to reach a maximum ozone concentration of 80 ppb on days 1 and 4, and a maximum ozone concentration of 100 ppb on days two and three. Ozone concentrations increased from 30 ppb to the daily maximum over the course of 2 hours, remained at the daily maximum for 6 hours, then decreased back down to 30 ppb over the course of 2 hours. Ozone concentrations were programmed to remain at 30 ppb at all other times.

Measurements

- Intermediate harvest – all plants were cut back to a height of 7 cm after 6 weeks exposure. The plants were harvested in separate layers – material growing outside the pot perimeter, material greater than 14 cm above soil level, and plant material between 7 cm and 14 cm above soil level.

- Final harvest – above ground biomass was determined for each species, and for the component species of the mixture. The plant material was harvested in layers as above, and with the addition of a layer with plant material 0 cm to 7 cm above soil level.
- Root-shoot ratio – determined for *Lolium* only at the final harvest.
- $[N]_{leaves}$ and C:N ratio - of *Lolium* only (determined at Bangor University).
- A-C_i curves – using a PP-Systems CIRAS fitted with an automatic cuvette. Light was provided at constant 1000 PAR. Applied CO_2 was reduced to 0, then slowly increased to 2000 ppm. This gave internal CO_2 concentrations of up to approximately 1600 ppm.
- Light curves – using a PP-Systems CIRAS fitted with an automatic cuvette. CO_2 was supplied at a constant 1000 ppm. Light levels were reduced to 0, then slowly increased to a PAR of 2000.
- Stomatal conductance measurements – using a Delta-T porometer and ambient light levels.
- Chlorophyll content of clover – using an Opti-Sciences CCM-200 chlorophyll meter.
- Chlorophyll fluorescence – for *Trifolium* only using a Hansatech Chlorophyll Fluorescence meter.
- Respiration
- Visual estimates of senescence and ozone-specific injury

<u>Data analysis</u>

Statistical analysis was performed using Minitab ANOVA.

Calculation of Farquhar parameters ($V_{c,max}$, J_{max})

Peter Levy (CEH-Edinburgh) calculated these parameters from both the solardome and field gas exchange measurements (see Chapter 3).

Results and Discussion

The Screening Trial in 2001 showed no significant differences in plant biomass for any of the species tested (data not presented). Visible injury was observed on white clover (*Trifolium repens*) and red clover (*Trifolium pratense*) and very slight injury was observed on creeping thistle (*Cirsium arvense*). No ozone-specific injury was observed on ryegrass (*Lolium perenne*), cocks foot (*Dactylis glomerata*), dock (*Rumex obtusifolius*) or prickly and smooth sow thistles (*Sonchus oleraceus* and *Sonchus apser)*.

Main Experiment 2001

Ozone-specific injury, characterised as small yellow speckles on the leaf surface, was apparent on the clover plants after exposure to the ozone regime for one week and soon afterwards the plants showed moderate to severe ozone injury symptoms (Figure 1.2).

The biomass of *Trifolium repens* was significantly less in the ozone-treated domes compared to the charcoal filtered domes (Figure 1.3). There were no significant differences in the biomass of either *Cirsium arvense* or *Lolium perenne*. Physiological measurements showed that the photosynthetic rate of the ozone treated *Trifolium repens* was 40% lower than that of plants growing in filtered air after 6 weeks of exposure in the solardomes.

Figure 1.2. Ozone injury on the leaves of white clover (*Trifolium repens*).

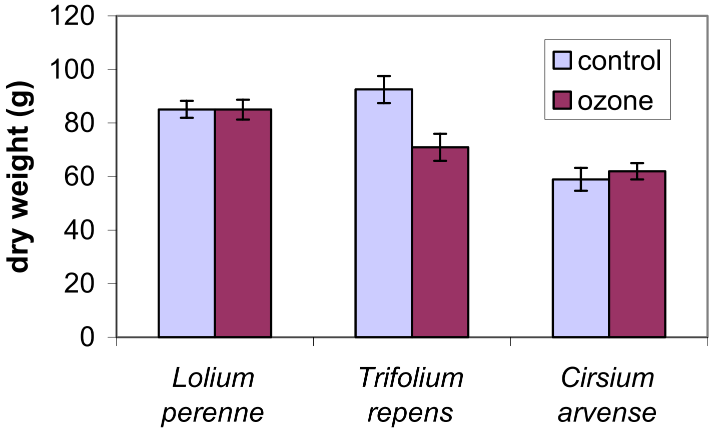


Figure 1.3. Biomass of Lolium perenne, Trifolium repens and Cirisum arvense at harvest after exposure to charcoal filtered air (control) or an episodic ozone exposure.

2002

The average AOT40 for the two domes exposed to the episodic ozone regime was 9.98 ppm.h during the first harvest interval, and 11.89 ppm.h during the second harvest interval, giving a total of 21.86 ppm.h over the 12 week exposure period (this is approximately equivalent to the exposure received in ambient air in Switzerland in a 'high' ozone year, or Italy in a 'moderate' ozone year). The difference in AOT40 between the two domes exposed to ozone was less than 2% for each harvest interval. In the two control domes, the average AOT40 was less than 0.02 ppm.h.

Biomass Measurements

Intermediate Harvest

Total biomass (above a cutting height of 7 cm) was not significantly different for either *Lolium perenne* or *Trifolium repens* when growing as monocultures (Table 1.1). *Trifolium repens* biomass was significantly reduced by ozone when grown as a mixture, whereas the *Lolium perenne* biomass was not affected.

Table 1.1 Biomass and biomass partitioning of *Lolium perenne* and *Trifolium repens* at the Intermediate Harvest.

Dry weight per pot (g)		outside pot perimeter		above 14 cm		7 to 14 cm		below 7 cm	
		control	ozone	control	ozone	control	ozone	control	ozone
Lolium perenne	mean	8.21	7.85	1.48	0.91	7.99	5.69	n/a	n/a
	significance	0.925		0.54		0.442			
Trifolium repens	mean	43.76	33.47	16.81	13.76	13.45	17.54	n/a	n/a
	significance	0.252		0.208		0.175			
Mixture *Lolium perenne*	mean	9.50	9.99	1.92	2.94	3.71	3.38	n/a	n/a
	significance	0.866		0.376		0.796			
Mixture *Trifolium repens*	mean	21.71	11.91	20.99	12.27	10.18	11.32	n/a	n/a
	Significance	0.002		0.001		0.597			

At the intermediate harvest there was a significantly reduced leaf biomass in *Trifolium repens* growing as part of a mixture. Effects on leaf biomass on *Trifolium repens* growing in monoculture were not significantly different. Differences in the biomasses of both healthy leaves and ozone injured leaves were significant in *Trifolium repens* growing as a monoculture or as part of the mixture. The biomass of injured leaves was approximately two thirds of the total leaf biomass in ozone treated plants, whereas the biomass of injured leaves was negligible in control treated plants.

Final Harvest

The total biomass of *Trifolium repens* was significantly reduced (by approximately 25%) in plants growing in domes treated with ozone (Table 1.2). *Lolium perenne* biomass was not affected by ozone treatment.

At the final harvest the total leaf biomass and the biomass of both healthy and injured leaves were significantly reduced by ozone in *Trifolium repens* growing both as a monoculture and as part of the mixture. The proportion of injured leaves was approximately 80% of the total leaf biomass in ozone treated plants. The biomass of injured leaves was negligible in control treated plants.

Lolium perenne biomass partitioning was not significantly different at either harvest. There was a significantly increased biomass of senesced leaves of ozone treated *Lolium perenne* growing as a monoculture compared to control treated plants at the intermediate harvest. Differences were not significant when *Lolium perenne* was growing as part of a mixture. At the final harvest, there was a significant increase in the biomass of the senesced leaves of this species in ozone treated plants growing as part of a mixture, but not as a monoculture.

A limited assessment of root biomass indicated that the root:shoot ratio of ozone treated *Lolium perenne* was increased by 30% (data not presented).

Table 1.2 Biomass and biomass partitioning of *Lolium perenne* and *Trifolium repens* at the Final Harvest.

Dry weight per pot (g)		outside pot perimeter		above 14 cm		7 to 14 cm		below 7 cm	
		control	ozone	control	ozone	control	ozone	control	ozone
Lolium perenne	mean	4.09	1.67	0.75	0.91	2.92	1.42	40.88	31.49
	significance	0.278		0.75		0.145		0.305	
Trifolium repens	mean	31.07	18.17	6.30	1.50	13.34	8.81	83.1	74.57
	significance	0.063		0.002		<0.001		0.426	
Mixture *Lolium perenne*	mean	2.16	2.10	0.56	0.74	1.85	2.61	*not sorted into Lolium/Trifolium*	
	significance	0.94		0.358		0.296			
Mixture *Trifolium repens*	mean	26.04	8.00	6.76	2.56	12.42	8.36		
	significance	<0.001		0.022		<0.001			

Canopy Height

There were no significant differences in the height of the canopy in ozone compared to control treated plants, apart from the *Lolium/Trifolium* mixture at the final harvest, which was significantly reduced in ozone treated plants.

Senescence of Lolium

In *Lolium perenne* plants, senescence (particularly of the ozone treated plants) started at the tip of the leaf blade and progressed back towards the main plant. The extent of the senesced portion of leaf (in mm) was significantly increased in ozone treated plants compared to control plants for both the monoculture and the mixture at both harvests (Figure 1.4). The extent of senescence was reduced in the mixture compared to the monoculture.

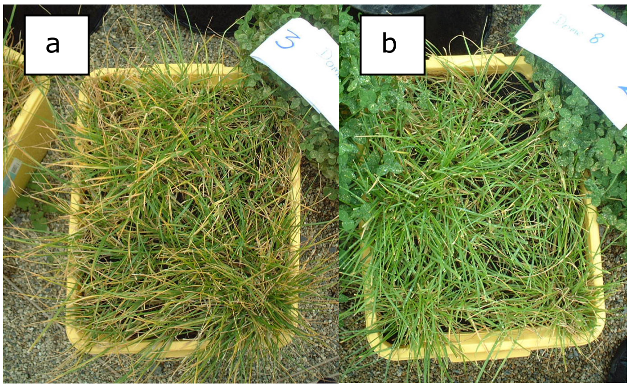


Figure 1.4. Senescence of *Lolium perenne* in ozone treatment (a) compared to control (b).

Visible Injury of Trifolium repens

Visible injury symptoms caused by ozone were observed on the clover plants only one week after exposure to ozone began. At the intermediate harvest, 69% of leaves per pot showed visible injury symptoms compared to only 0.5% in the control pots. Similar proportions of injury were observed when the clover was grown in combination with *Lolium perenne.* At the final harvest the proportion of injured clover leaves per pot was slightly higher when clover was grown in the mixture compared to when grown in monoculture (74% compared to 63%).

Physiological Measurements

There was no effect of ozone on *Lolium perenne* during the first harvest interval. There was a decrease in $V_{c,max}$ for *Lolium perenne* towards the end of the experiment, with older leaves more affected than younger leaves (Table 1.3). At this stage there was also some senescence at the tip of the measured leaves in the ozone treatment, but not the control, however green leaf tissue was used to determine the A-C_i curves in all cases. As the decrease in $V_{c,max}$ only started at the end of the experiment, there had been no time for significant growth effects.

There were reductions in $V_{c,max}$ for *Trifolium repens* leaves much earlier in the course of the study and again older leaves were more affected. In this case the older leaves from the ozone treated plants had moderate injury symptoms. The chlorophyll content of ozone treated *Trifolium repens* leaves was reduced by approximately 25%.

There were effects on the light curve for *Trifolium repens*, which had a reduced maximum assimilation rate following ozone treatment, however there were no corresponding effects on *Lolium perenne*.

Conclusions

- No significant differences in *Lolium perenne* biomass or partitioning at either harvest
- Significant 44% clover biomass reduction for final harvest, therefore field-derived white clover are very sensitive to ozone
- For the *Trifolium*:*Lolium* mixture, *Lolium perenne* biomass was not significantly affected by ozone whereas clover biomass was reduced by 58%, thus altering the species balance in favour of *Lolium perenne*
- Significant increase in senescence/visible injury due to ozone in both species
- Carboxylation efficiency reduced by ozone in *Lolium perenne*, even though the biomass was not significantly reduced.
- Results were used for parameterising and testing the process-based model (Chapter 4).

Table 1.3 Calculated J_{max} and $V_{c,max}$ values for Trifolium repens and Lolium perenne using functions as in Chapter 3, and chlorophyll contents for Trifolium repens.

Species	**Harvest**	**Leaf**	**Treatment**	J_{max}	**Rd (J_{max}-meas.)**	$V_{c,max}$	**Rd ($V_{c,max}$-meas.)**	**Chlorophyll content (mg/g FW)**
TRIFOLIUM	Intermed.	1	control			48	0.1	1222
			ozone			40	-0.1	902
		2	control	139	-2.1	61	0.2	1150
			ozone	89	-4.0	50	-0.4	900
	Final	1	control			34	-0.6	
			ozone			34	-0.8	
		2	control			53	-0.9	
			ozone			36	0.2	
LOLIUM	Intermed.	1	control			87	1.0	
			ozone			104	3.9	
		2	control	178	0.0			
			ozone	135	3.5			
	Final	1	control			77	1.3	
			ozone			72	5.2	
		2	control			39	-1.9	
			ozone			25	0.8	

2. Flux measurements in the field

Abstract

Field-scale measurements of fluxes between vegetation and atmosphere of O_3, CO_2 and H_2O were made at the grassland site, near CEH-Edinburgh, during the 2001 and 2002 growing seasons. The grass was cut for silage twice during the season and so 3 periods of canopy growth were observed each year. The H_2O flux results were used to estimate bulk-canopy stomatal conductance and hence the proportion of the ozone flux that was taken up by the vegetation through its stomata or onto other plant surfaces and the soil. The results show that the percentage of the total ozone flux that is stomatal increases from ~10% to ~60% over a growth period. On average the stomatal component is only about 40% of the total flux and the flux is dominated by non-stomatal uptake.

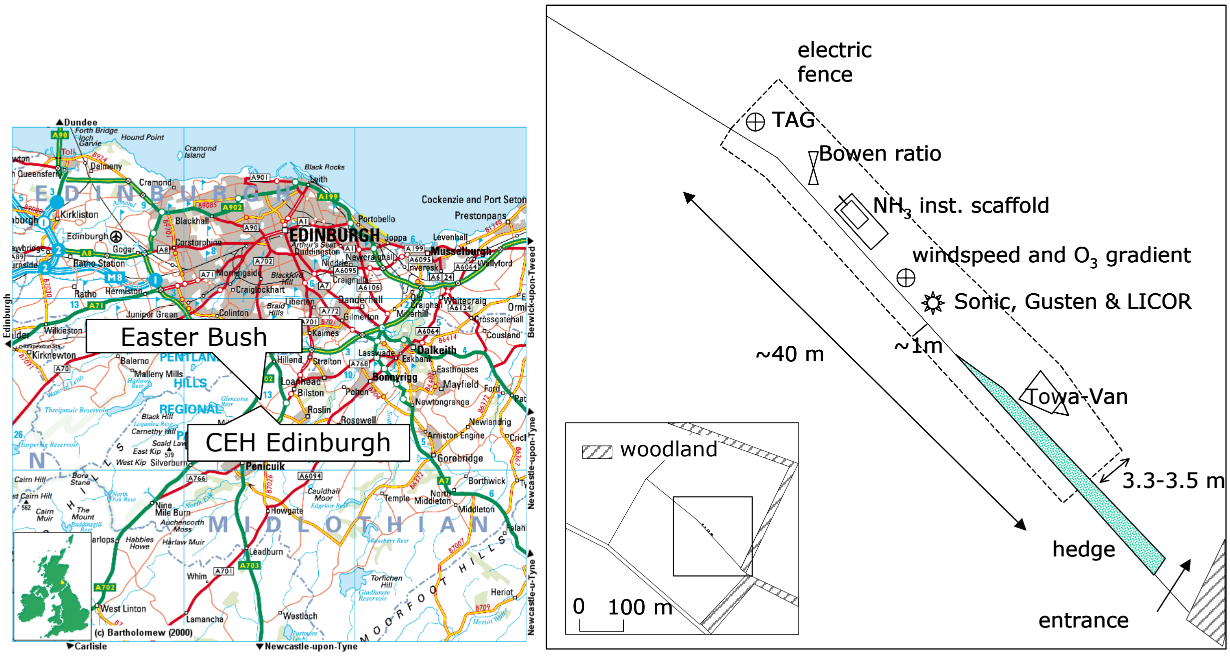
Introduction

As described earlier, ozone is a major phytotoxic pollutant, which currently reaches levels that are potentially harmful to UK vegetation (Coyle et al. 2003). The O3GRASSLAND project was designed to provide a comprehensive data set of all the parameters required to model and predict ozone damage in the field (Chapter 4). In this chapter, we report on the part of the project that involved measuring O_3, CO_2 and H_2O fluxes along with the major meteorological parameters that affect ozone fluxes and stomatal opening, such as radiation, temperature, humidity and soil moisture. The methodology used and summaries of the results obtained to date are given below.

Materials and Methods

The field site

The Easter Bush field site is located ~8 miles south of Edinburgh at the foot of the Pentland Hills, close to CEH-Edinburgh (180 m a.s.l., NT245642, 55.86 °N, 3.21 °E, Figure 2.1a). Measurements were made continuously from 26th May to 31st September 2001 and 21st March to 31st September 2002. There are two fields (total ~16 ha), separated by a wire fence and hedge with predominately *Lolium perenne* (>95%), some clover (*Trifolium pratense* and *repens*) and other wild species mixed in.



a. *b.*

Figure 2.1 a. Location of the Easter Bush Field site, b. schematic of the instrumentation layout.

They are managed for silage, normally with harvests twice during the summer. The fields are fertilized immediately after the cut grass is lifted and livestock are allowed on to graze a few weeks after the final cut. The same management regime occurred on both fields in 2001 and so measurements made over both are combined. During 2002 the management on each field varied so each field is treated separately. The time series plot in Figure 2.2 shows the variation in canopy height throughout the two measurement periods. The canopy does not grow to the same levels after each cut. This effect is often seen in grass that is harvested during the growing season, as regrowth after cutting is usually only vegetative material without flowers and seed heads (Sheaffer 2002). Particularly wet weather in 2002 significantly reduced canopy growth as the soil was waterlogged for much of the time and only the SW field was harvested a second time. Table 2.1 lists relevant management events on each field and major events, such as the installation of a new cabin in 2002.

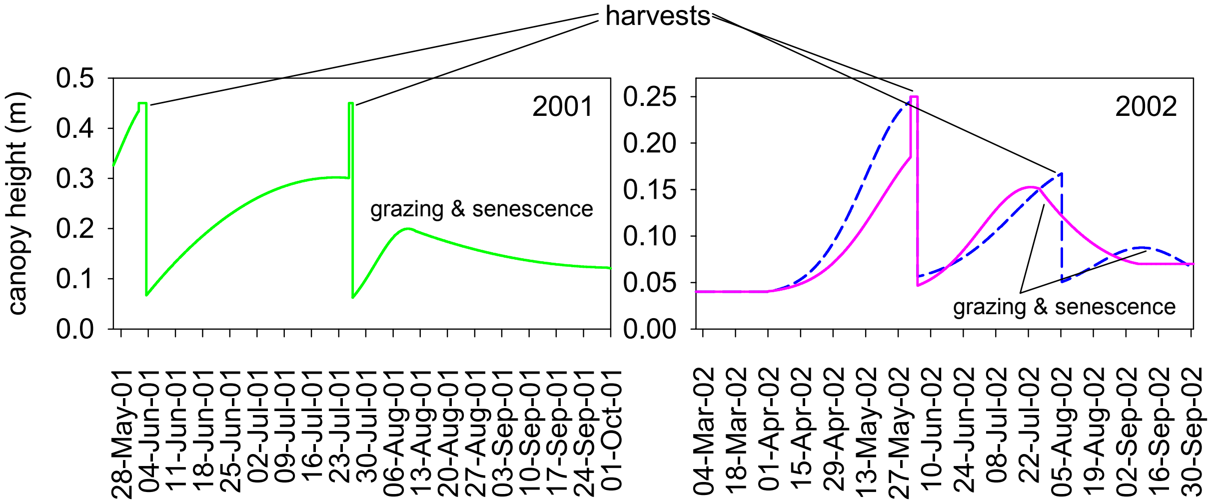


Figure 2.2 Canopy height time series (short square peaks are due to the grass being cut and left in piles before lifting). The harvests occurred on 1/6/01, 25/7/01, 1/6/02 and 5/8/02.

Table 2.1. Management events at Easter Bush during 2001 and 2002.

Date	Event	Date	Event
18/05/01	Open-path LICOR and sonic anemometer installed in Tow-A-Van	21/03/02	Gradient and GFAS reinstalled and fully functional
22/05/01	Ozone gradient mast up and operational	26/03/02	Both fields fertilised (400kg/ha of 24:5:8 +7.5S.)
01/06/01	both fields harvested for silage	17/04/02	GFAS removed
10/06/01	Both fields fertilised (300 kg/ha of 24:5:10 - N:P:K)	25/04/02	New O_3 analyser installed
20/06/01	GFAS Fast O_3 sonde operational	22/05/02	New mains power supply installed.
25/07/01	both fields harvested for silage	01/06/02	both fields harvested for silage
29/07/01	SW field fertilised (250 kg/ha of 24:5:10)	06/06/02	GFAS reinstalled
20/08/01	NE field waterlogged	08/06/02	both fields fertilised (250 kg/ha of 24:5:10 silage compound fertiliser)
		25/06/02	TDR installed and operational
		17/07/02	Everything off in the Old TAV for installation of the new cabin.
		18/07/02	New cabin in place and instrumentation back on.
		03/08/02	Floods around AMANDA
		05/08/02	SW field harvested for silage
		10/08/02	SW field fertilised (200 kg ha-1 25:5:5)
		27/09/02	SO_2 analyser installed on gradient
		22/10/02	Heavy rain, equipment flooded, all power off, TDL returned to Bush

Flux measurement methods

Trace-gas deposition or emission occur at the Earth's surface in the lowest part of the troposphere, known as the planetary boundary layer, where air flow is modified by aerodynamic friction with the surface and thermal stratification which determines the air density gradient. Within the boundary layer entities such as momentum, heat, water vapour and trace gases are transported horizontally by wind and vertically by turbulence. It is only the bottom few metres of the boundary layer, where the atmosphere is closely coupled to the surface, in which we are interested. This region, termed the surface layer, is normally defined as where fluxes vary by less than 10% with height. It is often called the constant flux layer, as fluxes within it can be described by measurements at a single height. Monin-Obukhov similarity theory is used to describe properties of the surface layer and the relevant scaling parameters with their typical orders of magnitude are defined below.

Common variables and constants:

u' = turbulent component of horizontal wind velocity, ie $u' = u - \bar{u}$ where u is the instantaneous velocity and $\bar{u}$ is the mean velocity
w' = vertical wind velocity (as above)
T = absolute temperature
θ = potential temperature
θ_v = virtual potential temperature
$k = 0.41$, von Karman's constant
$g = 9.81$ m s^{-2}, gravitational acceleration
ρ = air density

d = zero plane displacement height, where the canopy effectively becomes closed and all momentum is dissipated
λ = latent heat of vaporisation of water
χ = gas concentration
z = height above ground
h = crop height
c_p = specific heat at constant pressure for moist air
z_0 = aerodynamic roughness length, the height at which $u = 0$, typically 1 mm to 1 m.
NB over bars denote time averages

Scaling parameters:

Monin-Obukhov length,
$$L = -\frac{\overline{(u'w')}^{\frac{3}{2}}}{k(g/\bar{\theta}_v)\overline{(w'\theta'_v)}} \qquad \text{(1 m to 200 m)} \qquad (1)$$

friction velocity,
$$u_* = \overline{(u'w')}^{\frac{1}{2}} \qquad (0.05 \text{ to } 0.3 \text{ ms}^{-1}) \qquad (2)$$

potential temperature,
$$\theta_* = -\frac{\overline{(w'\theta'_v)}}{u_*} \qquad (0.1 \text{ to } 2.0\ ^{o}\text{C}) \qquad (3)$$

humidity,
$$q_* = -\frac{\overline{(w'q')}}{u_*} \qquad (0.1 \text{ to } 5\ g_{H_2O}\ kg_{air}^{-1}) \qquad (4)$$

The instantaneous eddy vertical flux of momentum, or shearing stress, τ, within the inertial sub-layer may be defined as:

$$\tau = \rho\, u'\, w' = \rho\, u_*^2 \qquad (5)$$

and by analogy the fluxes of sensible heat (H), latent heat (E) and a trace gas (F) may be written as:

$$H = \rho c_p\, w'\, \theta' \qquad (6)$$
$$E = \lambda\, w'\, q' \qquad (7)$$
$$F = w'\, \chi' \qquad (8)$$

These equations show that in a turbulent atmosphere the vertical flux of an entity is related to the vertical velocity and this is the basis of the eddy-correlation technique of measuring fluxes.

To determine the fluxes using other methods, the turbulence parameters u', w', θ', q' and χ' must also be described. To simplify the equations governing turbulent flow in the surface layer the first-order closure technique, K-theory, is used. This makes the assumption that the time averaged flux of an entity is related to its local gradient, i.e.:

$$F(z) = a_s\, K(z)\, ds/dz \qquad (9)$$

where: $F(z)$ = vertical flux density
$K(z)$ = turbulent transfer coefficient or eddy diffusivity (units, m^2s^{-1})
s = concentration of entity S
z = height above the surface
a_s = scaling factor for S

Hence, fluxes can be obtained by two methods:

1. Aerodynamic gradient, where gradients of the respective variables are measured and their diffusivities calculated (equation 9)
2. Eddy-correlation
   Equations (5) to (8) show that the flux of an entity is proportional to the product of its fluctuating component and that of vertical wind speed. Hence measurements of the turbulent fluctuations can be used to determine fluxes and this method has the advantage of being quite simple and direct. However, as the turbulent fluctuations occur very rapidly, fast response instruments are required. Sensors must be capable of responding to signals with a frequency of 0.1 Hz to 10 Hz. The use of this technique is therefore limited by the cost and availability of such fast response equipment.

These two methods were used at Easter Bush to measure the fluxes of H (sensible heat), λE (latent heat), CO_2, water vapour and O_3. However, there following conditions that must be met to allow the application of these methods:

- Winds must not be calm and the friction velocity, u_* (eqn. 2), must be greater than zero.
- There must be sufficient upwind fetch from the measurement system to allow the surface layer to develop. A ratio of 100 m of fetch to a 1 m deep surface layer is commonly used.
- The flux of the entity being measured should have stationarity ($d\chi/dt = 0$), which implies that there are no sources or sinks of the entity other than the surface and no chemical reactions with the entity in the air column.

By convention the momentum flux is positive in the downward direction and so H, E and F are negative. Further details of the methods used to calculate the fluxes from the measurements can be found in (Aubinet et al. 2000) and (Sutton 1990).

Instrumentation and data treatment

Micrometeorological measurements of atmospheric turbulence, O_3, CO_2, H_2O and NH_3 flux were made as well as rainfall, surface wetness, soil temperature, soil heat flux, total solar radiation and net radiation, during both years. Instrumentation to measure soil moisture using the TDR (time-domain reflectometry, (Topp et al. 1982)) technique was also added on the 25th of June 2002, as this is an important parameter controlling stomatal opening. All the measurements were logged on a 15-minute basis although individual instruments had different sampling frequencies. The instruments, described in Table 2.2 are situated on the boundary between the fields, as illustrated in Figure 2.1b. Table 2.2 also details the data capture obtained from each suite of instruments.

Table 2.2 Easter Bush Instrumentation and %Data Capture (filtered or filled*)	2001	2002
Eddy Correlation system		
open-path LICOR (LI-7500), CO_2 and H_2O flux	95% (72%)	100% (56%)
closed-path LICOR (LI-7000), CO_2 and H_2O flux	54%	86% (30%)
Gill Instruments, Solent Research 1012 sonic anemometer, turbulence and wind direction	96%	95%
GFAS fast-response ozone sonde OS-G-2, ozone flux	40%	70%
5-point gradient mast, ozone flux and turbulence		
5 ozone inlets (measured by high resolution UV O_3 analyser)	97% (70%)	95% (45%)
3 (5 in 2002) cup anemometers (Vector Instruments A100R)	98% (88%)	95% (73%)
3 fine-wire thermocouples (home-made)	98%	86%
Meteorology Mast (Campbell Scientific)		
2 thermocouples	96%	84%
soil temperature (Ts)	96%	88%
soil heat flux (G)	96%	88%
net radiation (Rn)	96%	88%
solar radiation (St)	100%	100%
surface wetness (Campbell Scientific 237 sensing grid)	96%	88%
rainfall tipping bucket	96% (100%)	88% (100%)
TDR Soil Moisture – 2 sets of 4 probes	-	42%

*The filters are described below. Gaps are filled using data from alternative instruments, after checking for consistency between them, for example the rainfall data is filled using data from the tipping bucket at Bush nearby.

The raw data capture for 15-minute periods from most of the instrumentation is over 80% when calibration periods and power failures are removed. However several other parameter specific and global filters have to be applied to the data before further analysis is done:

- Specific Filters
  - Wind speeds less than 0.8 $m\ s^{-1}$ measured by the cup anemometers were discarded as the anemometer could have stalled during the 15 minute period.
  - The LICOR open-path sensor cannot operate in the rain or fog as it uses an infra-red beam, the instrument indicates when the beam may be obscured.
  - The GFAS fast-response ozone sonde requires disks coated with an ozone sensitive compound to be replaced at regular intervals when the coatings response to ozone is reduced. A period of about 2 hours before and 1 hour after a new disk is inserted are therefore discarded. Data is also discarded when the output voltage from the GFAS drops below a threshold of 15 mV.
- Global Filters
  - Data from outside the good fetch sectors must be removed. Data from 200 ° to 300 ° and 30 ° to 70 ° are retained. The wind rose plot in Figure 2.3 shows the frequency of wind directions, which is mainly in the region of good fetch from the SW.
  - As stated above the Monin-Obukhov similarity theory may only be applied when the winds are not calm and the friction velocity, u_*, is not zero. Flux measurements are discarded when: $u_* < 0.08\ m\ s^{-1}$; $-5 < L < 5$.

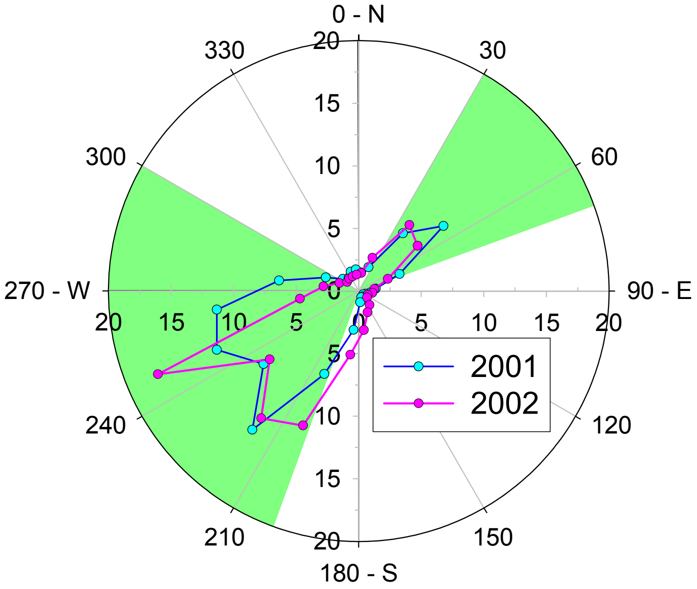


Figure 2.3 Wind rose showing the %frequency of wind directions and the good-fetch sectors (green).

The data capture achieved for 2001 is generally greater than that in 2002 (see Table 2.2) as instrument failures, restricted fetch and the weather caused some problems. For example, rainfall events were more frequent in 2002 (Figure 2.5), which led to the low data capture for the open-path LICOR7500. However, to allow the calculation of net carbon ecosystem exchange the CO_2 measurements are gapped filled using the methods outlined in (Falge et al. 2001), resulting in effectively 100% data capture. The O_3 flux time series may also be gap filled by developing a suitable model for the site, however this has not been completed as yet and so only the measured data are reported here.

Results

The Weather

The two years had quite different weather patterns, 2001 was generally warm, dry and fairly sunny whereas 2002 was slightly cooler and very wet, contributing to the poorer growth in 2002 (Figure 2.2). Figure 2.4 shows the hourly average solar radiation, ambient air temperature and soil temperature for each year. Considering the same time periods for each year (15th May to 30th Sept), although radiation levels are similar (average 514 and 512 $W\ m^{-2}$ in 2001 and 2002 respectively), temperatures were lower in 2002 (average 14 °C, maximum

26 °C in 2001; average 13 °C, max 24 °C in 2002). The most important factor causing the poor canopy growth during 2002 was the high rainfall (389 mm, 494 mm during 2001 and 2002 respectively). The field was waterlogged for much of the summer after very heavy rain early in the year, then frequent events throughout the year. In 2001 the rainfall was sufficient to ensure the canopy was well watered and the rainfall received was significantly greater then the measured transpiration, as shown in Figure 2.5.

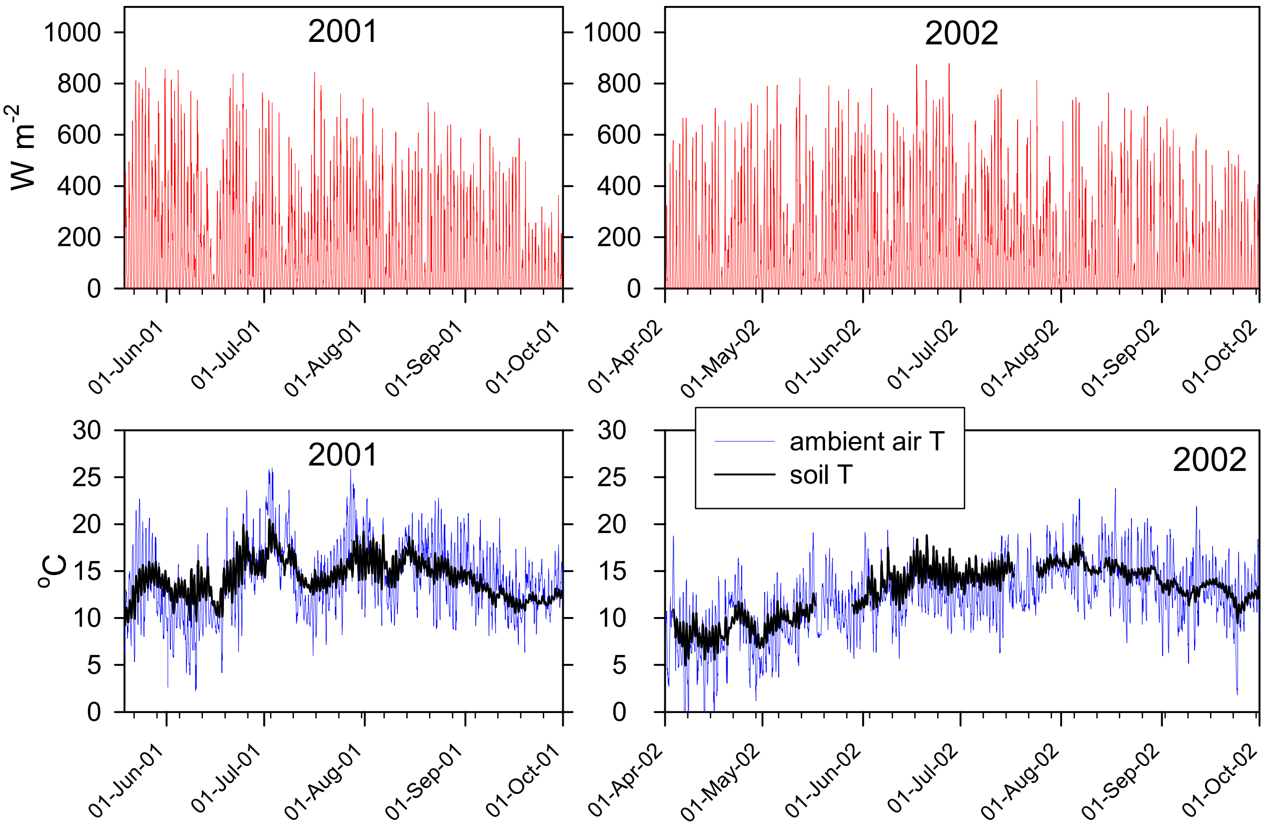


Figure 2.4 Hourly average solar radiation (top plot), ambient air temperature and soil temperature (bottom plot).

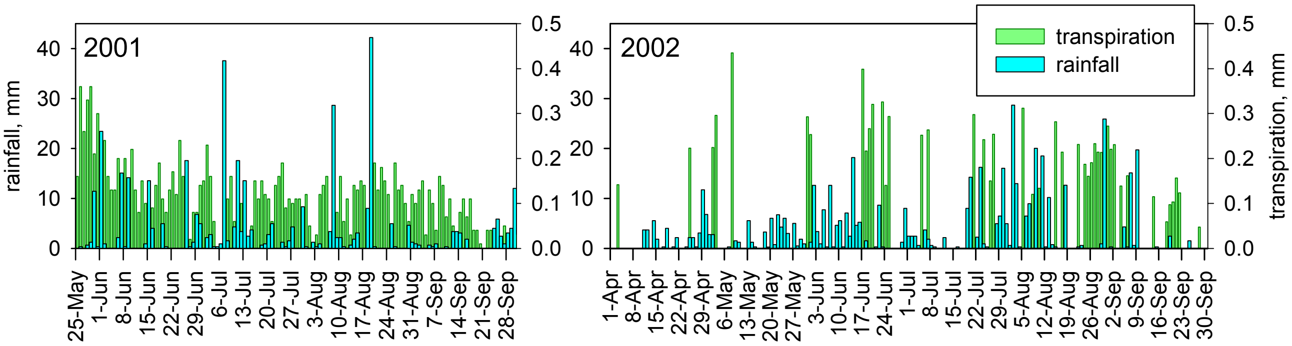


Figure 2.5 Daily total rainfall and transpiration (may include evaporation from soil) during 2001 and 2002 growing seasons.

Total Ozone Deposition

Trace deposition measurements are often reported as deposition velocity (v_d) at 1 m height, as this parameter normalises for variation in deposition with concentration (eqn. 10). For ozone this represents the ease of transfer through the atmosphere and the sink strength for the whole surface, i.e. plant stomata, external plant surfaces and soil, and the larger v_d the greater the rate of deposition. A diurnal cycle in ozone deposition velocity over vegetation is usually observed and it is generally accepted that uptake of ozone by plant stomata is the main factor controlling the cycle. At the beginning of a typical summer day the plants become active (solar radiation > 50 W m^{-2}) and start taking up ozone through their stomata. As plant activity increases during the day so does ozone uptake, then in the afternoon declines again as plants close their stomata. Thus a stomatally controlled component of ozone deposition is superimposed on the more constant surface (or non-stomatal) deposition, as illustrated in Figure 2.6.

$$v_d(1\ \mathrm{m}) = F(1\ \mathrm{m})/\chi(1\mathrm{m}) \tag{10}$$

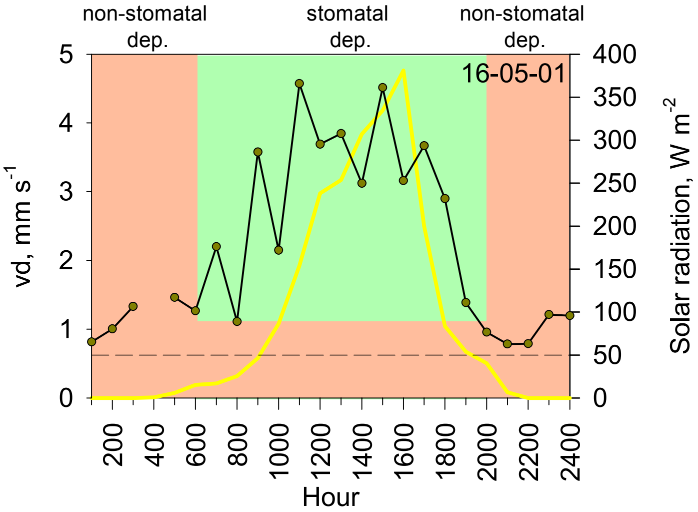


Figure 2.6 Example of a typical diurnal cycle in ozone deposition. The green area is approximately when the vegetation is active and so taking up ozone through their stomata.

In 2001 an average v_d of 3.5 mm s^{-1} was measured using the gradient system, which is consistent with other studies over grassland canopies (De Miguel and Bilbao 1999; Delany et al. 1986; Grunhage et al. 1998; Nussbaum et al. 2003; Stocker et al. 1993). The effect of canopy growth can clearly be seen in the time series of v_d with it increasing as the effective surface area and number of stomata increases. The harvests are also clearly visible as v_d drops, from ~ 3 mm s^{-1} to ~ 1.5 mm s^{-1} after the first cut and ~ 5 mm s^{-1} to ~ 3.5 mm s^{-1} after the second. This can be seen in the plot of 3 day running mean v_d shown in Figure 2.7 (bottom left plot). It was noted, however, that the deposition velocity did not always decrease fully immediately after the harvest. It is hypothesised that this is due to enhanced deposition on the newly exposed soil and grass stubble and chemical reactions with VOCs emitted from the cut grass. The second theory could be tested by measuring VOC levels before and after the harvest, which we hope to do during the summer of 2004.

The short period of eddy-correlation measurements available for 2001 show quite good agreement with the gradient data with an average v_d of 4 mm s^{-1} compared to 3.8 mm s^{-1} from the gradient. However this data needs to be fully reanalysed to incorporate changes in the flux calculation methodology and apply all the required corrections.

Analysis of the 2002 gradient system measurements are currently giving large values of ~7 mm s^{-1} on average, although the time series shows similar patterns to 2001 (Figure 2.7). The reasons for these unusually high values are being investigated at present and so no quantitative results are given for 2002. A preliminary assessment has been made of the eddy-correlation ozone measurements for 2002 and these give a more realistic result of ~1.8 mm s^{-1}. However, the time series does not show the expected variation with canopy heights and as with 2001, this data needs to be fully reanalysed before any definite conclusions can be drawn.

Stomatal vs Non-Stomatal Ozone Flux

Another useful parameter to consider when examining trace-gas fluxes is resistance. This is analogous to resistance in an electrical circuit and quantifies the ease with which a gas can pass from the atmosphere to the surface. As noted above, the total ozone deposition is a combination of deposition to plant stomata, cuticles and the soil, which can be considered as a series of resistances (Figure 2.8). Deposition velocity is simply the inverse of resistance, so $v_d = 1/R_t$, where R_t = total resistance.

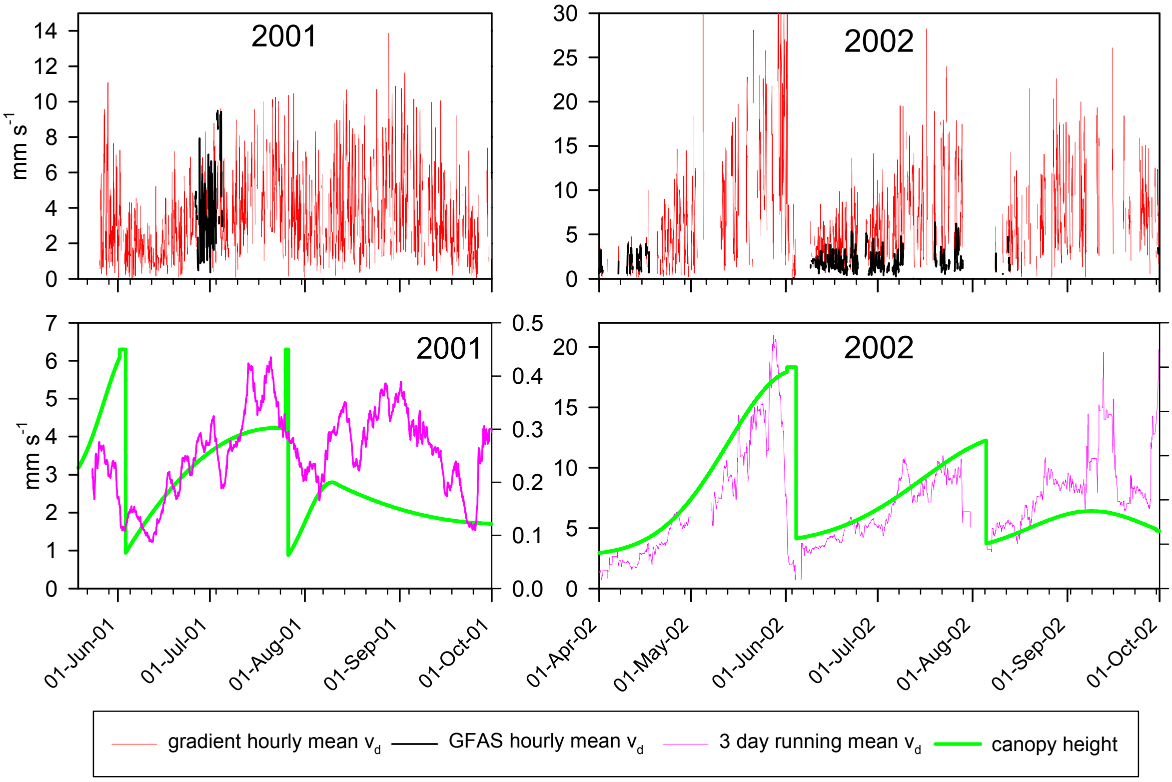


Figure 2.7 Hourly mean total ozone deposition velocities from the gradient and eddy-correlation systems (top plots). Three day running mean v_d and daily canopy height (bottom plots).

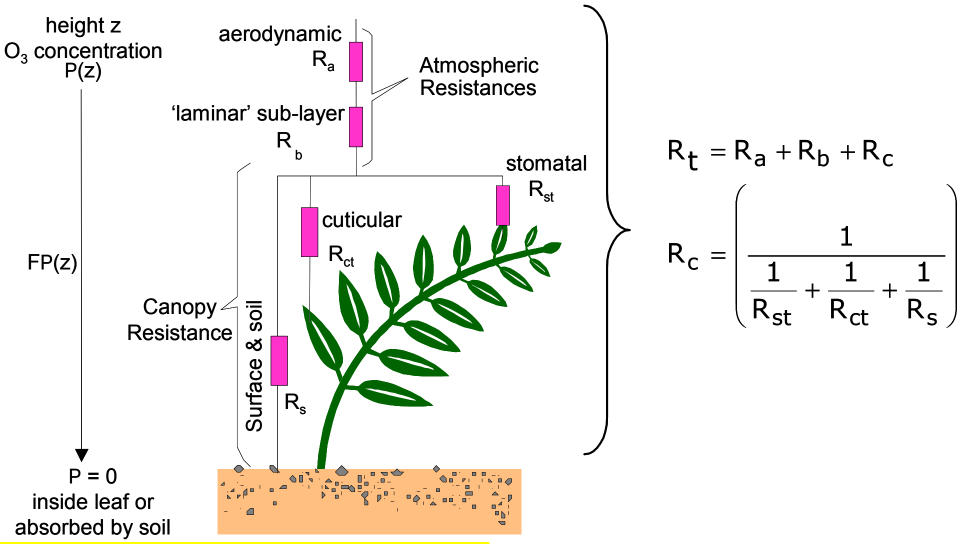


Figure 2.8 The deposition resistance analogy.

To separate the stomatal from non-stomatal sinks the water-vapour flux measurements are used. It is assumed that for each water molecule exiting a stoma, an ozone molecule enters; hence the stomatal ozone resistance (RsO_3) can be calculated using:

$$R_{sH_2O} = \frac{\rho\varepsilon}{p} \frac{e_s(T(z_{o'})) - e(z_{o'})}{E} \qquad (11)$$

where = air density (g m^{-3}), ε = ratio of the molecular weights of water vapour and air (0.622), p = atmospheric pressure (kPa), $es(T(z_o'))$ = saturation vapour pressure at the surface temperature $T(z_o')$ (kPa), z_o' = notional mean height of the canopy's surface (m), $e(z_o')$ = vapour pressure at z_o' (kPa), E = water vapour flux (g m^{-2} s^{-1})

$$RsO_3D_{O3} = Rs_{H2O}D_{H2O} \qquad (12)$$

where $\frac{D_{H_2O}}{D_{O_3}} \cong 1.5$, $D\chi$ = molecular diffusivity for χ

There are periods where the water vapour flux is from sources other than plant transpiration (evaporation from soil or wet foliage for example) and so the measurements used are restricted to periods when: the surface is dry; during daylight hours when the vegetation is active and should be transpiring; when humidity is low (< 70%). This filter retains 23% of the data in 2001 and 13% in 2002.

To examine the variation in stomatal resistance with canopy growth, daily averages during 1000-1700 hours are plotted in Figure 2.9 as this should be the period when vegetation is most active. As we would expect, Rs decreases as the canopy grows and there are more stomata, then increases after the canopy is cut or begins to senesce. During 2001 Rs increased from ~82 s m^{-1} to 290 s m^{-1} after the first cut and ~128 s m^{-1} to ~437 s m^{-1} after the second. The non-stomatal resistance (Rns) can also be estimated by taking the difference between Rc (total canopy resistance) and Rs:

$$R_{ns} = \left(\frac{1}{R_c} - \frac{1}{R_s}\right)^{-1} \tag{13}$$

As Rs can only be measured during daylight hours, the diurnal variation in the stomatal versus non-stomatal components cannot be examined without the use of a model for stomatal resistance. Such a model is being developed to allow more comprehensive analysis of the data sets, but is not complete at present. The importance of Rns can be shown, however, by plotting the daytime (1000-1700) stomatal and non-stomatal resistance as percentages of Rc, as shown in Figure 2.10. Although it is quite variable, on average stomatal deposition is ~40% of the total, decreasing from 40-50% before a harvest to ~10% just afterwards.

Measurements at another field site, Auchencorth Moss in the Scottish Borders, indicated that Rns varied with solar radiation or surface temperature (Fowler et al. 2001), decreasing as these parameters increased. It has been hypothesised that this is due to photo-dissociation of ozone mediated by the plant surface or a temperature dependant reaction with compounds on the leaf cuticle. The 2001 data from Easter Bush have been analysed in a similar manner and show a slight relationship with solar radiation (Figure 2.11). Other studies have shown that Rns may also vary with surface wetness although some found a positive variation and others negative (Fuentes 1992; Fuentes and Gillespie 1992; Pleijel et al. 1995; Zhang et al. 2002). These results indicate that the non-stomatal resistance should not be a simple parameter varying with LAI, as used in most models, but needs to incorporate some variation with meteorology as well as LAI.

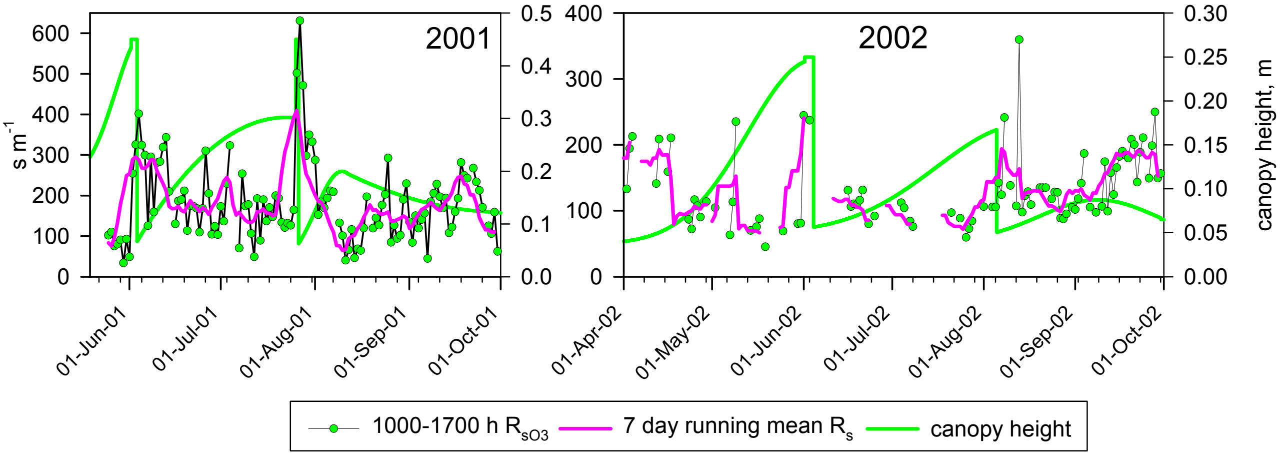


Figure 2.9 Stomatal resistance to ozone uptake, averaged over 1000-1700 h GMT and the 7 day running mean of the 1000-1700 h averages.

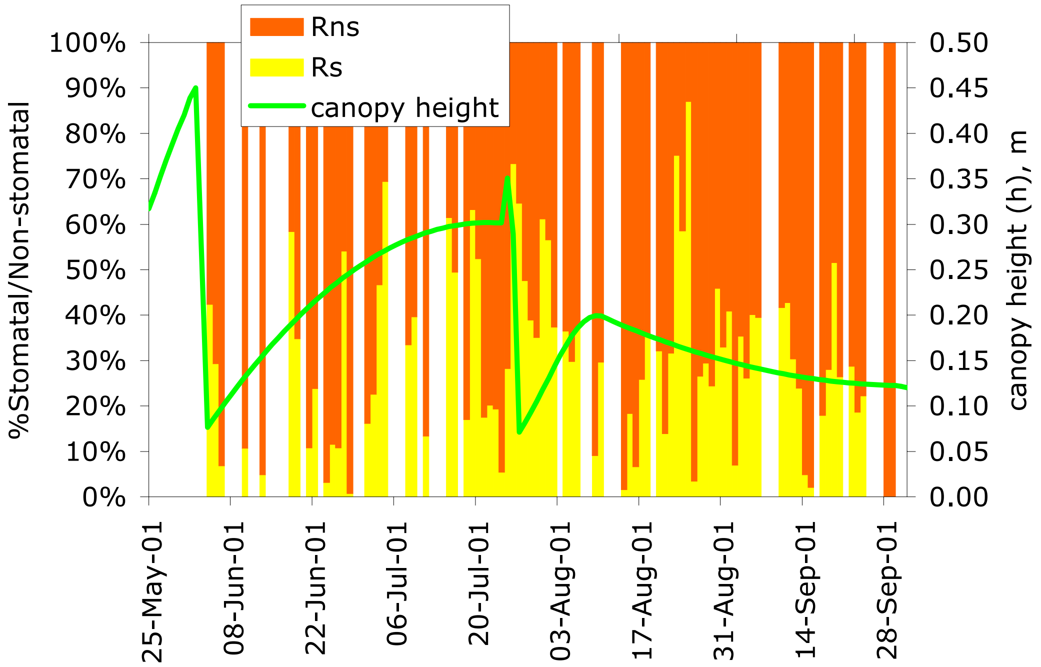


Figure 2.10 Percentage of total deposition which is stomatal or non-stomatal during 2001.

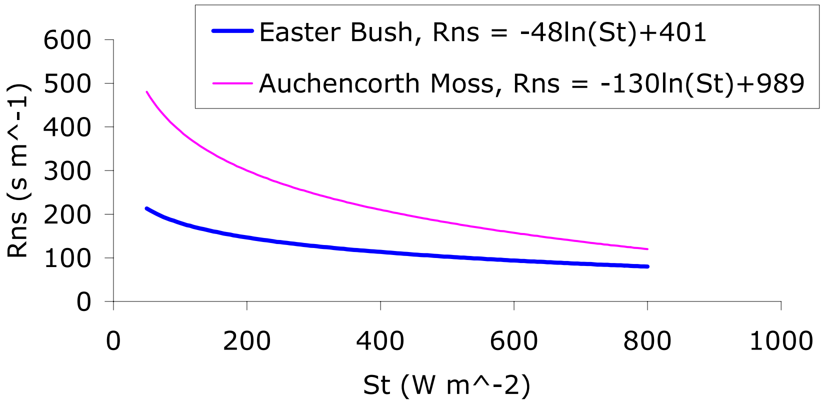


Figure 2.11 Modelled variation in non-stomatal resistance with solar radiation at Auchencorth Moss (1995) and Easter Bush (2001).

Soil Water and Ozone Flux

The TDR system for measuring soil water content (%SWC) was installed and fully operational on the 25th June 2002. It is known that soil water levels are an important factor controlling stomatal opening and so we would expect a relationship with stomatal ozone flux. Soil water pressure is more commonly used in stomatal conductance modelling (SWP, bar) and so the %SWC are converted to SWP using a relationship (14) derived from measurements of soil water release for a soil very similar to that at Easter Bush (from a nearby field).

$$\mathrm{SWP} = -b\ln\left(\frac{a}{\%\mathrm{SWC} - \%\mathrm{SWC}_0} - 1\right) + \mathrm{SWP}_0 \qquad (14)$$

where a = 47, b = 1.4, SWC_0 = 5.5, SWP_0 = 2

As noted above, the field was quite wet for much of 2002 however there where short periods during which the layer of the soil down to ~15 cm dried out, as indicated by SWP measurements over 1 bar (Figure 2.12). Plotting the block medians of SWP against Rs (25th June-02 to 30th Sep-02) shows a tendency for Rs to increase at high SWP. This relationship is more pronounced at 7.5-15 cm, where the majority of the plants' roots are, as Rs rapidly increases above 1 bar SWP. The lowest layer (30 cm) never dried out completely with a maximum SWP of only 0.3 bar, therefore the data is not plotted on Figure 2.12. However Rs did tend to decrease as SWP increased, ever over this small range of SWP.

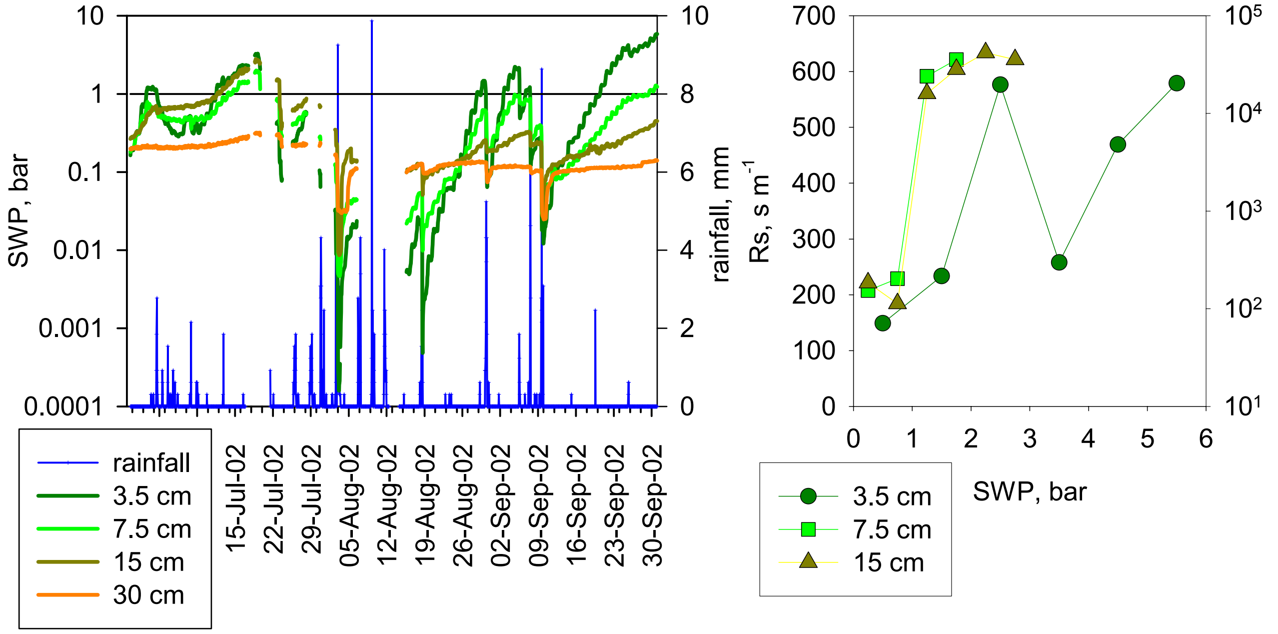


Figure 2.12 Time series of SWP and rainfall measured in the SW field at Easter Bush (left plot) and median stomatal resistance (Rs) versus SWP of the top 3 measurement points (right plot).

Conclusion

Although the data analysis is still ongoing, some interesting results have been found already, such as the delayed decrease in deposition velocity after the grass is harvested and the variation of non-stomatal resistance with solar radiation. In general the ozone fluxes vary as we would expect, increasing as the canopy grows and decreasing after the harvest or with senescence. The whole data set has and will provide the resources needed to assess and develop models of ozone deposition and stomatal uptake (see Chapter 4).

3. Biological measurements in the field

Abstract

Measurements of plant growth, nitrogen content, and leaf gas exchange were made at the Easter Bush grassland site during the 2001 and 2002 growing seasons. The growth rate of above-ground dry mass accumulation was around 4 g m^{-2} d^{-1}, reaching ~250 g m^{-2} before cuts in 2002. LAI increased up to a maximum of around 6 in 2002. Very high values of biomass and LAI were measured in 2001 (up to 900 g m^{-2} and 16, respectively), which we suspect to be erroneous. The trend in SLA was not very clear, but increased over the season, from ~100 to 400 cm^2 g^{-1}. Nitrogen content of leaves as a fraction of dry mass declined as biomass increased, though there was a slight increase in absolute terms. The C:N ratio of dry mass increased as biomass increased. The responses of photosynthesis (A) to photosynthetic photon flux density (Q) and internal CO_2 concentration (C_i) were derived using a Licor 6200 gas exchange system. Photosynthesis became light-saturated at Q >1000 μmol m^{-2} s^{-1}, reaching a value of A of ~15 μmol m^{-2} s^{-1}. Photosynthesis became CO_2-saturated at C_i >600 μmol mol^{-1}, reaching a value of A of 15-30 μmol m^{-2} s^{-1}. Fitting the Farquhar (1980) model of photosynthesis to these responses gave mean values for the $V_{c,max}$ and J_{max} parameters of 40 and 106 μmol m^{-2} s^{-1}, respectively, normalised to a temperature of 25 °C.

Introduction

To complement the micrometeorological flux measurements (Chapter 2) and the integrative modelling work (Chapter 4), a range of direct biological measurements were made at the Easter Bush field site with the project. These measurements were made for three main purposes:

- to quantify biological parameters required by the model
- to provide validation data for comparison with model output
- to quantify biological properties for use in interpreting the micrometeorological data.

Here, we describe measurements of plant growth, nitrogen content, and leaf gas exchange made at the Easter Bush grassland site during the 2001 and 2002 growing seasons.

Materials and Methods

Site characteristics

Details of site and grassland are given in Chapter 2 "Flux measurements in the field".

Growth

Harvest measurements were made every few weeks between April and August in 2001 and 2002. On each occasion, a randomly orientated transect was chosen, and ten evenly-spaced sample points were located, spanning the length of the field. A 28 x 37 cm quadrat (1036 cm^2) was placed at each sample point, and all above-ground plant material was removed with scissors. Fresh weights were determined immediately. Samples were then oven dried to constant weight (24 hours @ 70 °C).

At the same time as the quadrat harvests, a smaller amount of material was collected at each sample point for the determination of specific leaf area (SLA). The fresh weight of these SLA samples was measured immediately, and surface area was measured using a leaf area meter (either a LI-3000, Li-Cor, Lincoln, NB, USA or Delta-T Devices, Cambridge, UK). Samples were then dried and dry weights measured as above. For each sampling date, leaf area index (LAI) was calculated as the product of dry biomass (g m^{-2}) and SLA (m^2 g^{-1}). The bulk of plant material sampled in the quadrats was *Lolium perenne*, with the remaining fraction made up of other grasses (mainly *Holcus lanatus*), and other herbs (*Ranunculus* and *Rumex* species), though this fraction was negligible quantitatively.

Nitrogen and carbon contents

(determined at CEH-Merlewood by Valerie Kennedy)

Throughout the 2001 and 2002 growing season, nitrogen (N) and non-structural carbon (C) were measured in the leaves of *Lolium perenne*. A sub-sample taken from each quadrat on every harvest occasion throughout both years was oven dried at 70 ºC. Samples were then ground using a mill (Wiley – DCFH48, Glen Creston, Sranmore, UK) to less than 0.8 mm in preparation for chemical analysis. An aliquot of the ground material was re-dried in an air-circulated oven at 105 ºC for 3 h, and 350 mg of the oven-dry sample was digested by a modified Kjeldhal procedure in the presence of H_2O_2, with Li_2SO_4 to increase boiling point and Se as catalyst (Parkinson and Allen, 1975). Concentrations of N were then measured by continuous flow colorimetry (Skalar Analytical) via indolephenol blue. Samples for total C analysis were further ground to a fine particle size using a ball mill then measured using elemental analysis (Carlo Erba Strumentazione, Mod 1106, Fison Instruments, Sussex, UK).

Gas exchange

On a number of days in summer 2001 and 2002, leaf photosynthesis (A) and stomatal conductance were measured using a closed gas exchange system (LI-6200, Li-Cor, Lincoln, NB, USA). The measurements were only carried out on upper leaves in the sward, as the lower leaves could not be reached because of the size of the measurement chamber. Measurements were of two types: 1. those made rapidly after enclosing the leaf with the chamber, so as to be representative of the ambient functioning of the leaf; or 2. those made over a number of minutes, during which chamber conditions were altered, so as to derive the response of photosynthesis to changes in internal CO_2 concentration (C_i) or photosynthetic photon flux density (Q). Each individual measurement was made over 15 seconds, achieving a CO_2 differential of around 5 ppm with 4 cm^2 of leaf area enclosed. A consecutive pair of measurements was made each time to check for errors.

To achieve a range of CO_2 concentrations, the CO_2 concentration was reduced either by photosynthesis alone or by diverting air through soda lime. The CO_2 concentration was elevated above ambient by exhaling near the system intake and opening the inlet valve briefly. When conducting A-C_i responses, CO_2 was first lowered progressively below ambient to near zero, then raised to around 1600 ppm and lowered progressively back to ambient. This allowed the measurements to be made as fast as possible while maintaining stomatal opening. A range of Q values was achieved by altering the angle of the chamber relative to the sun. To achieve very low levels, cloth were used to shade the chamber. In the measurements where environmental variables were manipulated over a period of minutes, stomatal conductance was likely to still be adjusting to the changed conditions. These responses are therefore only applicable to photosynthesis, which has a much faster response time (<1 s).

Data from the A-C_i and A-Q curves were used to fit the parameters of the Farquhar (1980) model of photosynthesis. A-Q curves were used to fit the maximum rate of electron transport (J_{max}) and dark respiration rate (R_d) parameters, normalised to a reference temperature of 25 °C. A-C_i curves were used to fit the maximum rate of carboxylation ($V_{c,max}$) parameter, also normalised to a reference temperature of 25 °C. When fitting $V_{c,max}$, R_d was allowed to vary, but the R_d values were not used, as they are unlikely to be representative of the respiration rate in the dark under ambient CO_2. The model fitting procedure was performed using the Genstat statistical package. The programs are listed in the Appendices to this chapter.

Results and Discussion

Growth

Figure 3.1 shows the trend in biomass, SLA and LAI over the two seasons. Very high values of biomass and LAI were measured in 2001 (up to 900 g m^{-2} and 16, respectively), which we suspect to be erroneous. The values of SLA measured in 2001 were not unusual, and so we suspect the problem to be with the biomass measurements, rather than in the conversion of this to leaf area. However, an examination of the raw data did not yield any obvious source of error.

In 2002, the growth rate of above-ground dry mass accumulation was around 4 g m^{-2} d^{-1}, and was slightly higher before the first cut than after. The maximum value of biomass was also found just before the first cut (286 g m^{-2}), although a similar value (253 g m^{-2}) was reached before the end of the experiment. A similar pattern was seen in the 2001 data, though the absolute values are suspect.

The trend in the SLA data was not very clear, but increased markedly after the first cut in both years, from ~100 to 300 cm^2 g^{-1}. This causes a different pattern in LAI to that seen in biomass, with higher LAI values after the first cut than before it (almost 7 cf. 3.5 in 2002).

Nitrogen and carbon contents

Figure 3.2a shows a clear decline in the nitrogen content of biomass over the growth periods between cuts in all years. This is reflected in the trend in the C:N ratio of dry mass, which increased correspondingly (Fig. 3.2b). Both these trends were caused by an increase in carbon over time, rather than a loss of nitrogen. Indeed, there was a slight increase in total nitrogen on a ground area basis (Fig 3.2c).

Gas exchange

Figure 3.3 shows *A*-*Q* response curves from four leaves of *Lolium perenne* on 21 May 2002. Photosynthesis became light-saturated at *Q* ~1000 μmol m^{-2} s^{-1}, reaching a value of *A* of around 15 μmol m^{-2} s^{-1}. The dark respiration rate, inferred from the *y*-intercept, was approximately 2 μmol m^{-2} s^{-1}. Photosynthesis became CO_2-saturated at C_i >600 μmol mol^{-1}, in three of the four *A*-C_i responses shown in Figure 3.4. The asymptotic value of *A* reached varied from 15-30 μmol m^{-2} s^{-1}, and depended on the amount of ambient Q, as this could not be controlled. Where possible, response curves were carried out in full sunlight. The *A*-*Q* and *A*-C_i response curves were used to fit the parameters of the Farquhar (1980) model of photosynthesis (Table 3.1). Mean values for the $V_{c,max}$ and J_{max} parameters were 40 and 106 μmol m^{-2} s^{-1}, respectively (Fig. 3.5).

Table 3.1. Values of the parameters of the Farquhar (1980) photosynthesis model fitted to data from (a) *A*-*Q* and (b) *A*-C_i response curves for *Lolium perenne* at the Easter Bush field site. Parameters fitted were the maximum rate of electron transport (J_{max}), the dark respiration rate (R_d), and the maximum rate of carboxylation ($V_{c,max}$), all normalised to a reference temperature of 25 °C.

(a)

Leaf no.	Date	Leaf no.	J_{max}	s.e.	R_d	s.e.	*n*	Species
yymmdd##	dd/mm/yy	nn	umol m^{-2} s^{-1}		umol m^{-2} s^{-1}			
2052107	21/05/2002	7	99	17.9	5.74	2.26	10	*Holcus lanatus*
1060800	08/06/2001	pooled	73.3	33.3	-1.1	7.88	24	*Lolium perenne*
1062100	21/06/2001	pooled	115.9	0	0	0	42	*Lolium perenne*
2052101	21/05/2002	1	103	10.8	3.66	1.06	14	*Lolium perenne*
2052102	21/05/2002	2	94.8	6.88	4.589	0.883	18	*Lolium perenne*
2052103	21/05/2002	3	81.24	8.53	4.8	1.15	14	*Lolium perenne*
2052104	21/05/2002	4	90.31	4.19	2.114	0.379	6	*Lolium perenne*
2052105	21/05/2002	5	134.48	8.67	5.17	1.18	18	*Lolium perenne*
2052106	21/05/2002	6	113.7	10	4.31	1.22	10	*Lolium perenne*
2052108	21/05/2002	8	119.29	9.11	5.17	1.29	14	*Lolium perenne*
2052109	21/05/2002	9	119.14	9.88	5.95	1.38	14	*Lolium perenne*
1062200	22/06/2001	pooled	134.7	70.4	0.4	10.7	10	*Rumex* sp
		Mean	**106.57**		**4.19**			

(b)

Leaf no.	Date	Leaf no.	$V_{c,max}$	s.e.	*n*	Species
yymmdd##	dd/mm/yy	nn	umol m^{-2} s^{-1}			
2051607	16/05/2002	7	21.83	4.7	20	*Holcus lanatus*
1062208	22/06/2001	8	50.2	1.67	34	*Lolium perenne*
1062209	22/06/2001	9	42.75	1.92	18	*Lolium perenne*
1062210	22/06/2001	10	63.87	3.39	34	*Lolium perenne*
2051601	16/05/2002	1	39.52	3.42	24	*Lolium perenne*
2051602	16/05/2002	2	28.31	3.18	22	*Lolium perenne*
2051603	16/05/2002	3	49.16	2.16	18	*Lolium perenne*
2051604	16/05/2002	4	33.17	3.27	16	*Lolium perenne*
2051605	16/05/2002	5	36.4	3.87	20	*Lolium perenne*
2051606	16/05/2002	6	28.61	2.47	22	*Lolium perenne*
2051608	16/05/2002	8	39.27	1.6	22	*Lolium perenne*
2052109	21/05/2002	9	66.43	8.73	20	*Lolium perenne*
1062206	22/06/2001	6	23.96	2.95	10	*Rumex* sp
	Mean		**40.27**			

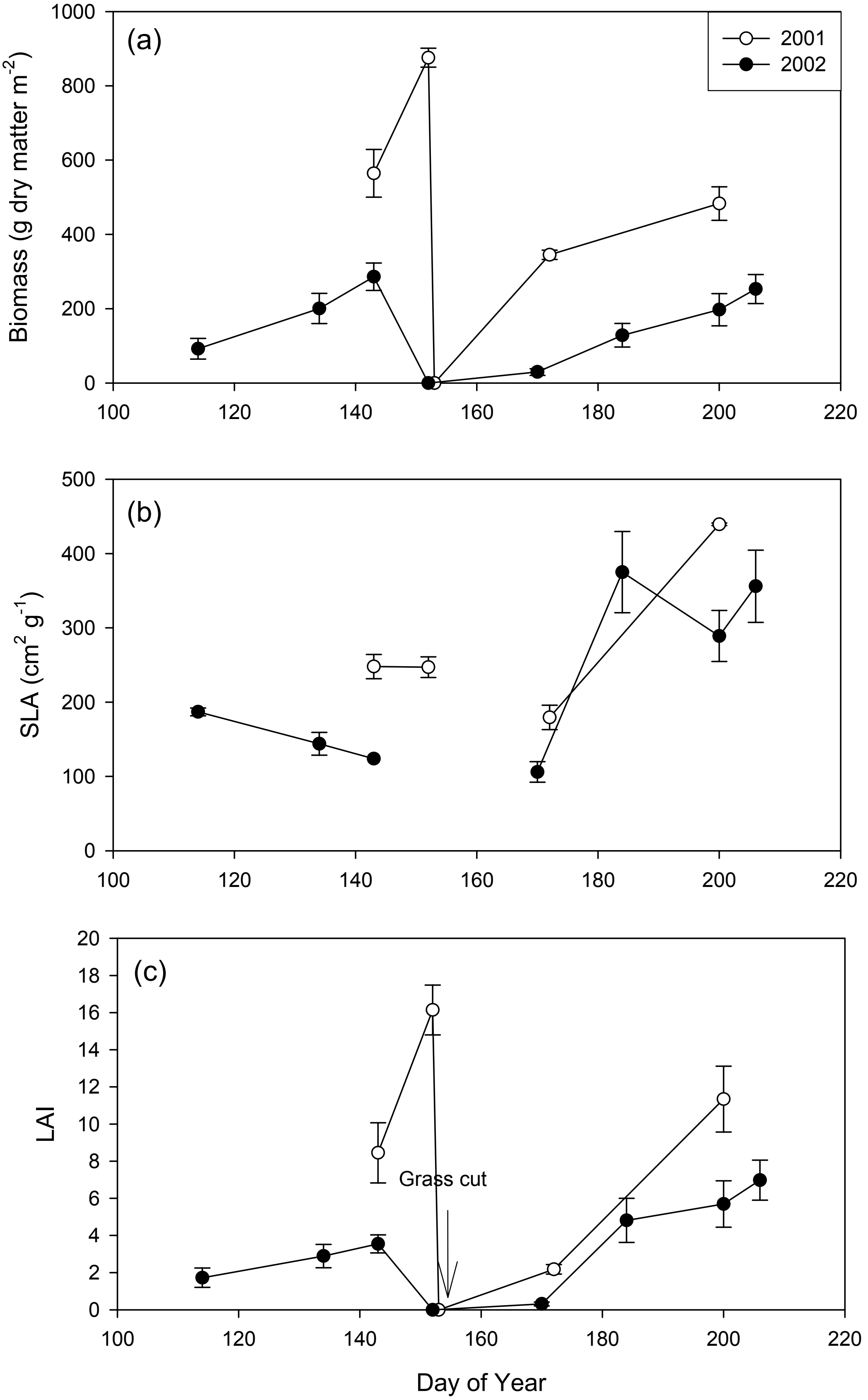


Figure 3.1. Changes in (a) biomass, (b) specific leaf area (c) and leaf area index at Easter Bush over the 2001 and 2002 growing seasons. Error bars show 95 % confidence intervals calculated from the variance in the ten sample quadrats.

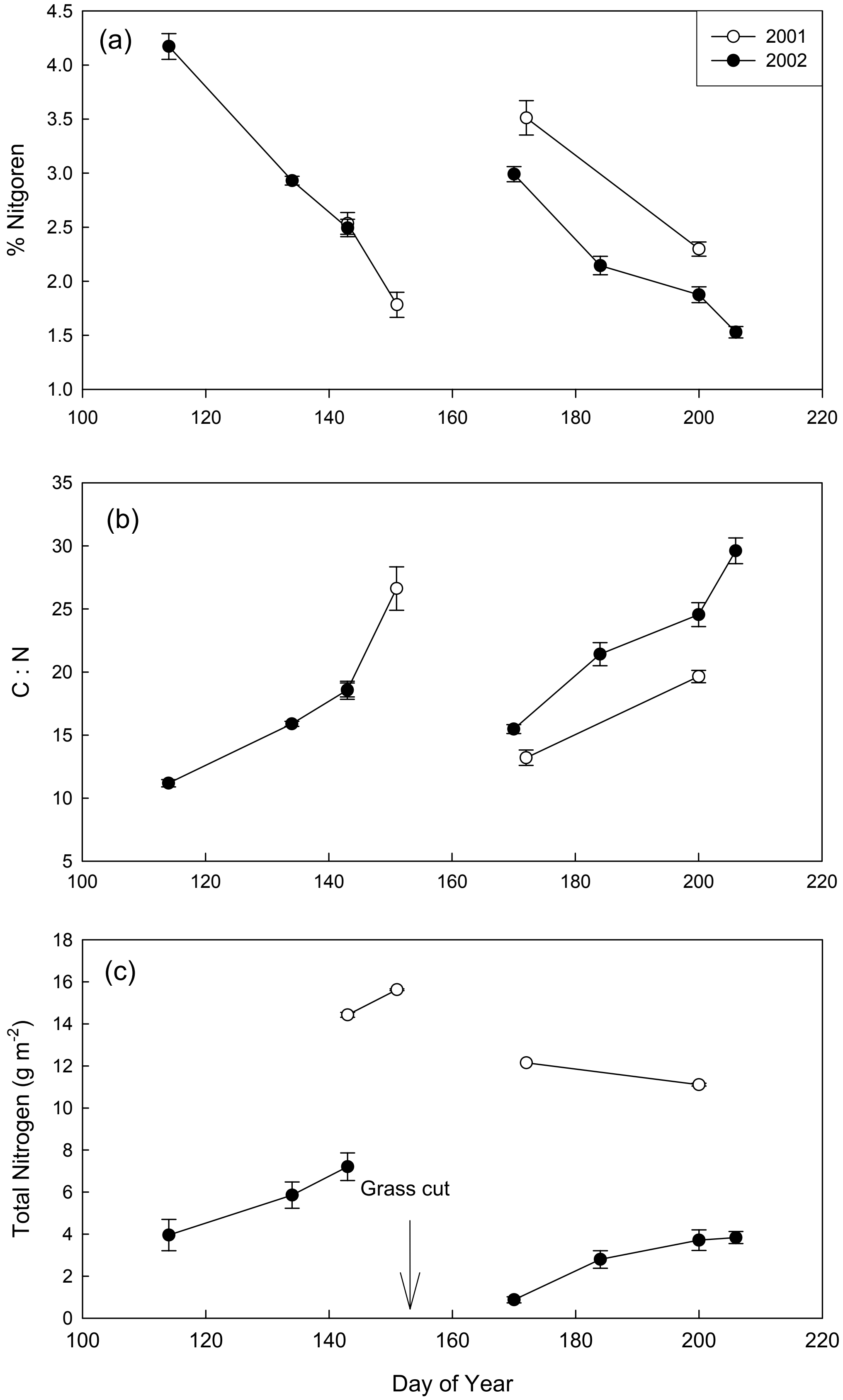


Figure 3.2. Changes in (a) nitrogen content as a percentage of dry biomass, (b) carbon:nitrogen ratio of dry biomass (c) and total nitrogen on a ground area basis at Easter Bush over the 2001 and 2002 growing seasons. Error bars show 95 % confidence intervals calculated from the variance in the ten samples.

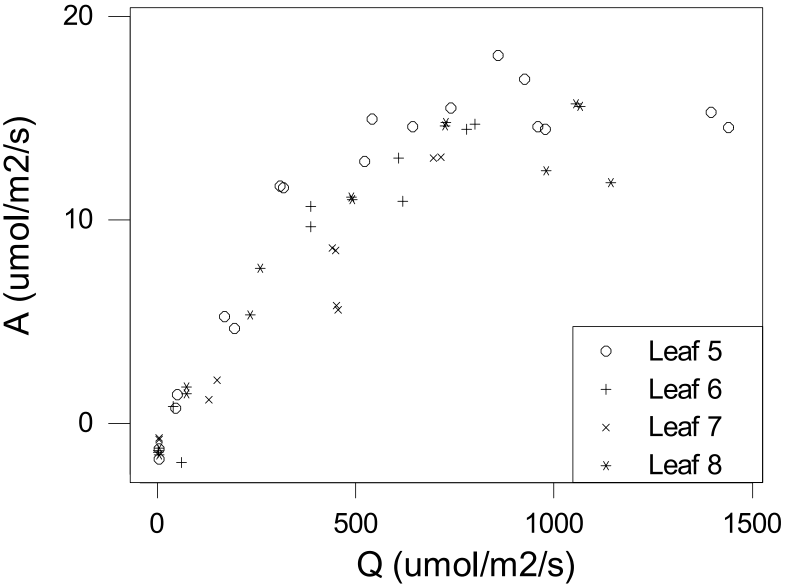


Figure 3.3. Response of photosynthetic rate, *A*, to photosynthetic photon flux density, *Q*. Response curves are shown for four leaves of *Lolium perenne* at Easter Bush on 21 May 2002 using the Licor 6200.

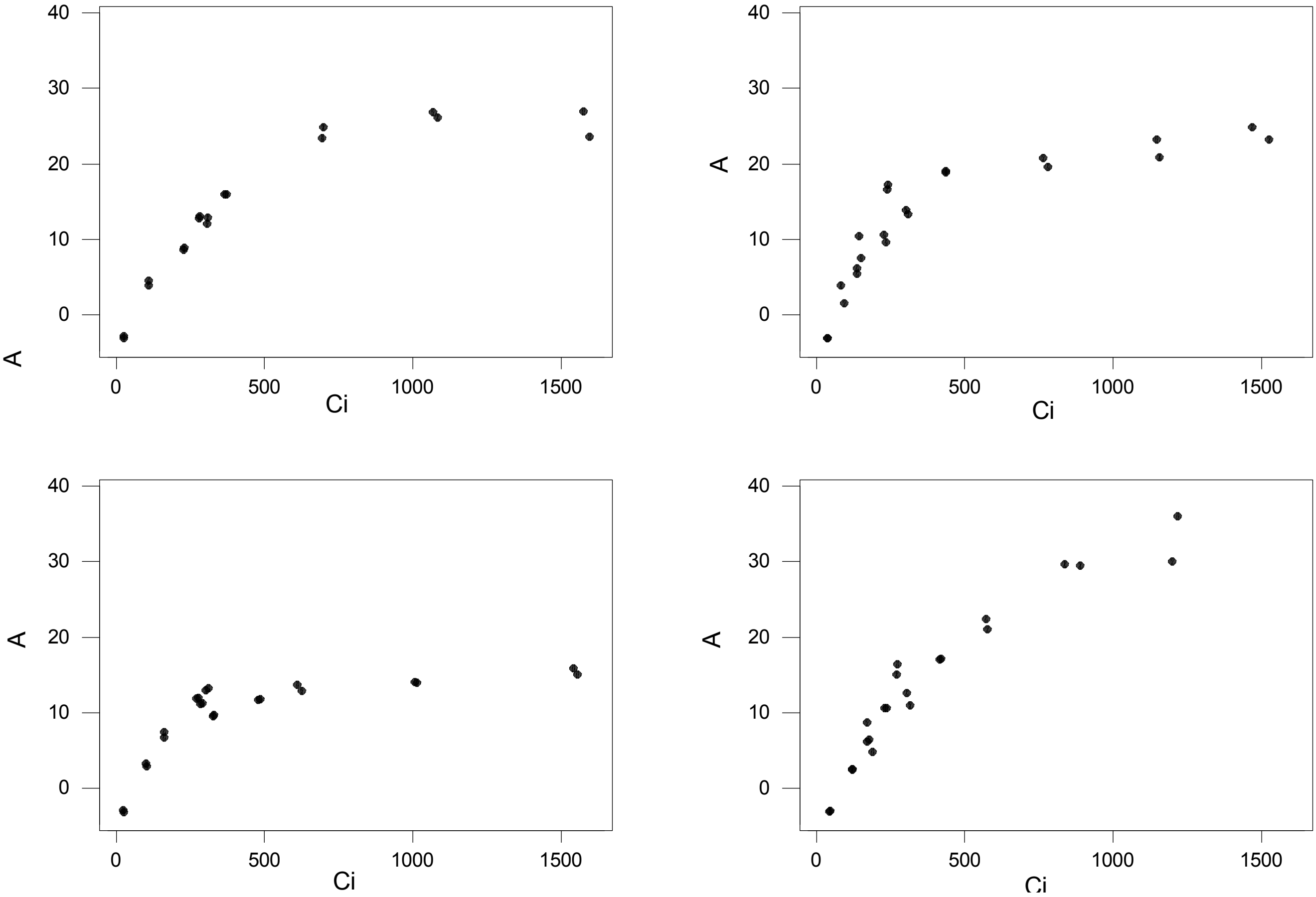


Figure 3.4. Response of photosynthetic rate, *A*, to internal CO_2 concentration, C_i. Response curves are shown for four leaves of *Lolium perenne* at Easter Bush on 16 May 2002 using the Licor 6200.

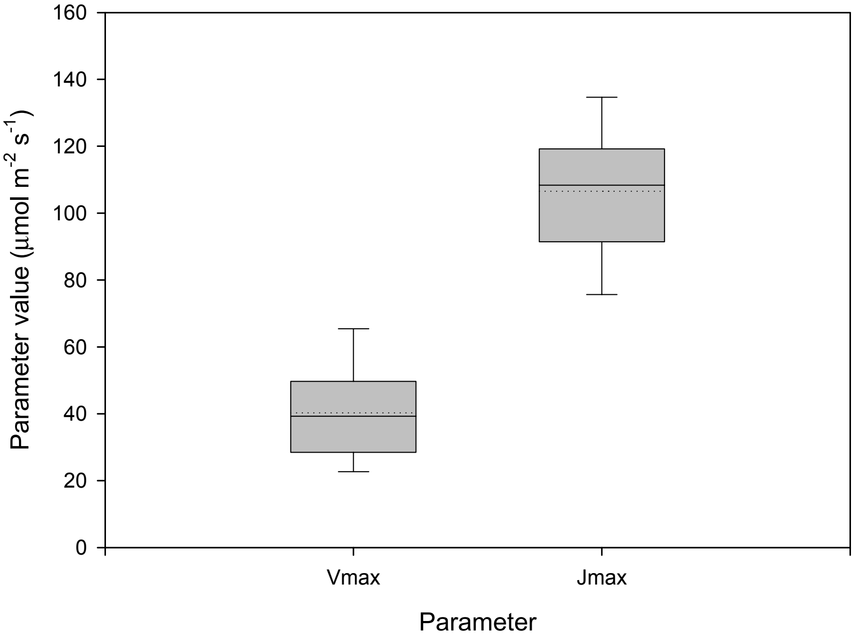


Figure 3.5. Boxplot showing the mean (solid line), median (dotted line), inter-quartile (shaded box) and range (whiskers) for the $V_{c,max}$ and J_{max} parameters of the Farquhar (1980) model fitted to gas exchange measurements on *Lolium perenne* at the Easter Bush field site.

Conclusions

- Above-ground dry biomass reached a maximum of around 250 g m^{-2} before cuts in 2002.
- The rate of accumulation of above-ground dry biomass was around 4 g m^{-2} d^{-1}, and was slightly higher before the first cut.
- LAI increased up to a maximum of around 6 in 2002.
- Very high values of biomass and LAI were measured in 2001, which we suspect to be erroneous. SLA increased towards the end of the growing season, from ~100 to 400 cm^2 g^{-1}.
- Nitrogen content of leaves as a fraction of dry mass declined as biomass increased, though there was a slight increase in absolute terms.
- Leaf photosynthesis became light-saturated at Q >1000 μmol m^{-2} s^{-1}, reaching a value of A of ~15 μmol m^{-2} s^{-1}.
- Photosynthesis became CO_2-saturated at C_i >600 μmol mol^{-1}, reaching a value of A of 15-30 μmol m^{-2} s^{-1}.
- Fitting the Farquhar (1980) model of photosynthesis to these responses gave mean values for the $V_{c,max}$ and J_{max} parameters of 40 and 106 μmol m^{-2} s^{-1}, respectively, normalised to a temperature of 25 °C.

Appendices to Chapter 3

1. Program for J_{max} from A-Q curves

```
" Genstat program for fitting the parameters of the Farquhar model of photosynthesis to gas exchange A-
Ci response curves from the Licor 6200
Peter Levy, Centre for Ecology and Hydrology
Bush Estate, Penicuik, Midlothian, EH26 0QB, UK
Tel: 0131 445 8556 (direct), Fax: 0131 445 3943
E-mail: plevy@ceh.ac.uk "
read Q, Tair,   Tleaf,  Ca,    CO2_range,   Flow_rate,    RH_range,     RH,    Ps_initial,     gs,
        Ci,     dummy1,        dc_dt,  dummy2,      dummy3,       eair,  date,   month, Leaf_area,
        Aobs

" Sample data – 2 records "
1806    25.29   27.12   333.3  3.783   105.8   0.1848 41.23   13.66   0.2322 221.6   0       -0.24860
        0       13.28   21     5               2.933333333            13.97045455
2058    25.5    27.76   328.6  3.756   105.4   0.2465 40.88   13.54   0.229  216.1   0       -0.24780
        0       13.34   21     5               2.933333333            13.84772727

" parameters "
scalar EaJ ; value = 37000
scalar EaKc ; value = 59356
scalar EaKo ; value = 35948
scalar EaV ; value = 58520
scalar EaRd ; value = 66400
scalar gammaTref ; value = 36.9
scalar gi ; value = 10
scalar H ; value = 220000
scalar KcTref ; value = 460
scalar KoTref ; value = 330000
scalar pO2 ; value = 210000
scalar R_ ; value = 8.314
scalar S ; value = 710
scalar Theta ; value = 0.7
scalar Tref ; value = 298.15
scalar VmaxTref ; value = 60.0

" Farquhar equations "
EXPRESSION FarquharEq[1]; !e( gsc = gs*1000/1.606 )
EXPRESSION FarquharEq[2]; !e( Rd = RdTref*EXP((Tleaf-(Tref-
273.15))*EaRd/(R_*298.15*(Tleaf+273.15))) )
EXPRESSION FarquharEq[3]; !e( KcT = KcTref*EXP((Tleaf+273.15-
Tref)*EaKc/(R_*Tref*(Tleaf+273.15))) )
EXPRESSION FarquharEq[4]; !e( KoT = KoTref*EXP((Tleaf+273.15-
Tref)*EaKo/(R_*Tref*(Tleaf+273.15))) )
EXPRESSION FarquharEq[5]; !e( JmaxT = JmaxTref*(EXP(((Tleaf+273.15)/Tref-
1)*EaJ/(R_*(Tleaf+273.15)))*(1+EXP((Tref*S-H)/(R_*Tref)))/(1+EXP((S*(Tleaf+273.15)-
H)/(R_*(Tleaf+273.15))))) )
EXPRESSION FarquharEq[6]; !e( J = (Q/2*0.85**2+JmaxT-SQRT((Q/2*0.85**2+JmaxT)**2-
4*Theta*Q/2*0.85**2*JmaxT))/(2*Theta) )
EXPRESSION FarquharEq[7]; !e( VmaxT = VmaxTref*EXP((Tleaf+273.15-
Tref)*EaV/(R_*Tref*(Tleaf+273.15))) )
EXPRESSION FarquharEq[8]; !e( gamma = gammaTref+1.88*(Tleaf-(Tref-273.15))+0.036*(Tleaf-(Tref-
273.15))**2 )
EXPRESSION FarquharEq[9]; !e( a_quad = -gi-gsc )
EXPRESSION FarquharEq[10]; !e( b_quad_J = gi*gsc*(Ca+2*gamma)+(gi+gsc)*(J/4-Rd) )
EXPRESSION FarquharEq[11]; !e( c_quad_J = gi*gsc*(Rd*(Ca+2*gamma)-J/4*(Ca-gamma)) )
EXPRESSION FarquharEq[12]; !e( c_quad_V = gi*gsc*(Rd*(KcT*(1+pO2/KoT)+Ca)-VmaxT*(Ca-
gamma)) )
EXPRESSION FarquharEq[13]; !e( b_quad_V = gi*gsc*(KcT*(1+pO2/KoT)+Ca)+(gi+gsc)*(VmaxT-Rd) )
EXPRESSION FarquharEq[14]; !e( Av = (-b_quad_V+sqrt(b_quad_V**2-
4*a_quad*c_quad_V))/(2*a_quad) )
```

```
EXPRESSION FarquharEq[15]; !e( Aj = (-b_quad_J+sqrt(b_quad_J**2-
4*a_quad*c_quad_J))/(2*a_quad) )
EXPRESSION FarquharEq[16]; !e( Amodel = Aj )
EXPRESSION FarquharEq[17]; !e( Cimodel = Ca-Amodel/gsc )

model Aobs; fittedvalues=Amodel
scalar VmaxTref, RdTref
rcycle JmaxTref, RdTref; initial=100.0, -5.0
" Replace the above line with following to impose parameter constraints: rcycle JmaxTref, RdTref;
initial=60.0, -5.0; lower= 10.0, -10.0; upper= 200.0, 0.0 "
fitnonlinear [print=m,s,e,f,c,monitoring; calculation=FarquharEq[]]

‶ Plot graph of fitted and observed A vs Q”
XAXIS [RESET=yes] WINDOW=1; LPOSITION=outside; LDIRECTION=parallel; MPOSITION=outside;\
 ARROWHEAD=omit; ACTION=display
YAXIS [RESET=yes] WINDOW=1; LPOSITION=outside; LDIRECTION=perpendicular;
MPOSITION=outside;\
 ARROWHEAD=omit; ACTION=display
PEN [RESET=yes] 1...2; METHOD=line; JOIN=ascending; SYMBOL=1; LINESTYLE=1
DGRAPH Y=Aobs,Amodel; X=2(Q)
PEN [RESET=yes] 1...2
```

2. Program for J_{max} from A-C$_i$ curves

```
" Genstat program for fitting the parameters of the
  Farquhar model of photosynthesis to gas exchange A-Ci response curves from the Licor 6200
Peter Levy, Centre for Ecology and Hydrology
Bush Estate, Penicuik, Midlothian, EH26 0QB, UK
Tel: 0131 445 8556 (direct), Fax: 0131 445 3943
E-mail: plevy@ceh.ac.uk "
read Q, Tair,   Tleaf,  Ca,     CO2_range,      Flow_rate,      RH_range,       RH,     Ps_initial,  gs,
        Ci,     dummy1,         dc_dt,  dummy2,         dummy3,         eair,   date,   month, Leaf_area,
Aobs

‶ Sample data – 2 records ”
1161    20.43   21.2    289.1   5.254   113.7   0.3973 61.55    55.75   4.6     235.4   0       -0.33150
        0       14.78   16      5               3.483333333             16.00478469
1116    20.6    21.34   282.6   5.106   113.4   0.2838 61.14    54      4.507   230     0       -0.32160
        0       14.83   16      5               3.483333333             15.50239234
" parameters "
scalar EaJ  ; value = 37000
scalar EaKc ; value = 59356
scalar EaKo ; value = 35948
scalar EaV  ; value = 58520
scalar EaRd  ; value = 66400
scalar gammaTref ; value = 36.9
scalar gi ; value = 10
scalar H ; value = 220000
scalar KcTref ; value = 460
scalar KoTref ; value = 330000
scalar pO2 ; value = 210000
scalar R_ ; value = 8.314
scalar S ; value = 710
scalar Theta ; value = 0.7
scalar Tref ; value = 298.15
scalar JmaxTref ; value = 100.0

" Farquhar equations "
EXPRESSION FarquharEq[1]; !e( gsc = gs*1000/1.606 )
EXPRESSION FarquharEq[2]; !e( Rd = RdTref*EXP((Tleaf-(Tref-
273.15))*EaRd/(R_*298.15*(Tleaf+273.15))) )
EXPRESSION FarquharEq[3]; !e( KcT = KcTref*EXP((Tleaf+273.15-
Tref)*EaKc/(R_*Tref*(Tleaf+273.15))) )
```

```
EXPRESSION FarquharEq[4]; !e( KoT = KoTref*EXP((Tleaf+273.15-
Tref)*EaKo/(R_*Tref*(Tleaf+273.15))) )
EXPRESSION FarquharEq[5]; !e( JmaxT = JmaxTref*(EXP(((Tleaf+273.15)/Tref-
1)*EaJ/(R_*(Tleaf+273.15)))*(1+EXP((Tref*S-H)/(R_*Tref)))/(1+EXP((S*(Tleaf+273.15)-
H)/(R_*(Tleaf+273.15))))) )
EXPRESSION FarquharEq[6]; !e( J = (Q/2*0.85**2+JmaxT-SQRT((Q/2*0.85**2+JmaxT)**2-
4*Theta*Q/2*0.85**2*JmaxT))/(2*Theta) )
EXPRESSION FarquharEq[7]; !e( VmaxT = VmaxTref*EXP((Tleaf+273.15-
Tref)*EaV/(R_*Tref*(Tleaf+273.15))) )
EXPRESSION FarquharEq[8]; !e( gamma = gammaTref+1.88*(Tleaf-(Tref-273.15))+0.036*(Tleaf-(Tref-
273.15))**2 )
EXPRESSION FarquharEq[9]; !e( a_quad = -gi-gsc )
EXPRESSION FarquharEq[10]; !e( b_quad_J = gi*gsc*(Ca+2*gamma)+(gi+gsc)*(J/4-Rd) )
EXPRESSION FarquharEq[11]; !e( c_quad_J = gi*gsc*(Rd*(Ca+2*gamma)-J/4*(Ca-gamma)) )
EXPRESSION FarquharEq[12]; !e( c_quad_V = gi*gsc*(Rd*(KcT*(1+pO2/KoT)+Ca)-VmaxT*(Ca-
gamma)) )
EXPRESSION FarquharEq[13]; !e( b_quad_V = gi*gsc*(KcT*(1+pO2/KoT)+Ca)+(gi+gsc)*(VmaxT-Rd) )
EXPRESSION FarquharEq[14]; !e( Av = (-b_quad_V+sqrt(b_quad_V**2-
4*a_quad*c_quad_V))/(2*a_quad) )
EXPRESSION FarquharEq[15]; !e( Aj = (-b_quad_J+sqrt(b_quad_J**2-
4*a_quad*c_quad_J))/(2*a_quad) )
EXPRESSION FarquharEq[16]; !e( Amodel = Av )
EXPRESSION FarquharEq[17]; !e( Cimodel = Ca-Amodel/gsc )

model Aobs; fittedvalues=Amodel
scalar VmaxTref, RdTref
rcycle VmaxTref, RdTref; initial=60.0, -5.0
" Replace the above line with following to impose parameter constraints: rcycle JmaxTref, RdTref;
initial=60.0, -5.0; lower= 10.0, -10.0; upper= 200.0, 0.0 "
fitnonlinear [print=m,s,e,f,c,monitoring; calculation=FarquharEq[]]

“ Plot graph of fitted and observed A vs Ci”
XAXIS [RESET=yes] WINDOW=1; LPOSITION=outside; LDIRECTION=parallel; MPOSITION=outside;\
 ARROWHEAD=omit; ACTION=display
YAXIS [RESET=yes] WINDOW=1; LPOSITION=outside; LDIRECTION=perpendicular;
MPOSITION=outside;\
 ARROWHEAD=omit; ACTION=display
PEN [RESET=yes] 1...2; METHOD=line; JOIN=ascending; SYMBOL=1; LINESTYLE=1
DGRAPH Y=Aobs,Amodel; X=2(Cimodel)
PEN [RESET=yes] 1...2
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4. Integrative modelling

Abstract

Ozone damages plants in various ways. We can distinguish four damage mechanisms: (1) disrupting photosynthesis, (2) increasing respiration, (3) accelerating senescence, and (4) altering root-shoot allocation. Damage depends on the ozone deposition rate and how it is distributed in time, but it also depends on the plant species, its physiological state, and the environmental conditions. Because of these interactions, it is desirable to quantify damage under field conditions, particularly if the goal is to predict when real vegetation is at risk from ambient ozone. However, the four damage mechanisms can only be quantified well under controlled conditions. An instrument for scaling up leaf-scale data measured under controlled conditions to the field-scale would thus be useful for predicting ozone damage.

In this project (O3GRASSLAND), the solardomes at CEH-Bangor were used for detailed measurements of damage caused by ozone to grass (*Lolium perenne*) and clover (*Trifolium repens*) (Chapter 1). The plants had been cloned from material taken from a grassland field near CEH-Edinburgh. At this latter field site, measurements of fluxes between vegetation and atmosphere of O_3, CO_2 and H_2O were made by the eddy covariance and gradient methods (Chapter 2). Vegetation growth rate was also measured at this field site (Chapter 3).

In this chapter, we describe how a simulation model that accounts for the four damage mechanisms triggered by ozone was constructed and applied to the field site. Parameters of the model were taken from the solardome experiments. We report the accuracy with which the model was able to simulate the fluxes of O_3, CO_2 and H_2O in the field during two consecutive years. An analysis is presented of the conditions identified by the model as conducive to high ozone risk.

Introduction

The concentration of ozone (O_3) has increased at 1-2% per year during the last two to three decades at middle and high latitudes over Europe (Guicheret 1989). Present-day tropospheric concentrations of O_3 are considered to be damaging (Skarby et al. 1993), and are expected to continue increasing.

The ozone damage mechanisms are not yet completely clarified. Knowledge of the damage mechanisms is needed to identify climatic conditions and plant characteristics that enhance tolerance to O_3. Mechanistic simulation of vegetation growth may be a tool to investigate these issues. However, no model exists that incorporates the mechanisms by which O_3 affects growth and yield, nor the mechanisms by which the plants adapt.

Monteith (1977) showed an often close to linear correlation between cumulative light interception and yield of crops. This induced a trend towards building simple and robust growth models, based on the light interception and light-use efficiency (LUE) approach. Modelling papers have appeared in which the relationship between LUE and physiological characteristics, such as foliar nitrogen content, is explained (Charles-Edwards 1982; Dewar 1996). However, no clear concepts have been given for the effects of atmospheric gases, such as O_3, on LUE. For these gases detailed short-term physiological models at the cellular and leaf level do exist, from which one could derive LUE after upscaling to the integration level of the vegetation (e.g., for O_3: (Schut 1985)). Also available are very simple regression models linking yields to atmospheric gas concentrations.

In this paper, we aim to present a model in which LUE and grassland growth are linked in a mechanistic manner to ambient concentrations of O_3. We feel that there is a need for such instruments of analysis, to allow results from different fumigation experiments to be compared in a more quantitative manner than thus far, and to act as a precursor for operational prediction of ozone damage. The grassland growth model is parameterised using the data presented in the three preceding chapters, and additional data from the literature on the effects of O_3 on plants. The sensitivity of the model to conditions differing in light intensity, temperature and atmospheric CO_2 concentration will be tested.

Model structure

The O3GRASSLAND-model was based on two predecessors: LINTULCC, a model for the effects of climate, CO_2 and O_3 on wheat (Rodriguez et al. 1999; Van Oijen and Ewert 1999), and LINGRA-Timothy, a model for the effects of climate, CO_2 and drought on timothy grassland (Hoglind et al. 2001). In O3GRASSLAND, these two parent models were simplified, merged and a routine for competition between two species for light and water was added to allow simulation of grass-clover interaction. The resulting model is deterministic, operates at a daily time step, and has 36 parameters for characteristics of the plants, and 5 for the soil.

Growth

Growth rate is calculated by multiplying the amount of light intercepted by the canopy by the light-use efficiency of the sward (Monteith 1977):

$$\mathrm{GR} = \mathrm{LUE}\,\mathrm{I}_0 (1 - \mathrm{e}^{-k\ \mathrm{LAI}}) \qquad (1)$$

where GR = growth rate (g biomass m^{-2} d^{-1}); LUE = light-use efficiency (g biomass MJ^{-1}); k = light extinction coefficient (-); LAI = leaf area index (m^2 m^{-2}); I_0 = incoming photosynthetically active radiation (MJ PAR m^{-2} d^{-1}).

Biomass allocation and leaf area dynamics are modelled as in LINGRA-Timothy (Hoglind et al. 2001). Allocation patterns, i.e. the partitioning of GR over organs, depend on source-sink relations within the sward. The light-use efficiency depends on several factors, including light intensity, leaf Rubisco and chlorophyll content, atmospheric CO_2 concentration, and the physiological effects of O_3. We will first present a general equation for the LUE, and then show how O_3 can modify it.

Light-use efficiency

Light-use efficiency (LUE) is defined as the ratio of biomass growth and light interception:

$$\mathrm{LUE} = \frac{\mathrm{P_c}\,\gamma}{(1 - \mathrm{e}^{-k\ \mathrm{LAI}})\,\mathrm{I}_0} \qquad (2)$$

where P_c = daily integral of gross canopy photosynthesis (g CO_2 m^{-2} d^{-1}); γ = efficiency with which gross assimilated CO_2 is converted to biomass (g biomass g^{-1} CO_2). Equation (2) is difficult to solve because it explicitly contains a term for daily photosynthesis. However, Rodriguez et al (1999), building on the work of Charles-Edwards (1982), presented a simplified equation:

$$\mathrm{LUE} = \frac{\gamma\,\alpha}{1 + \dfrac{\alpha\,k\,\mathrm{I}_0}{\mathrm{P_{max}}\,\delta}} \qquad (3)$$

where α = leaf quantum yield (g CO_2 MJ^{-1}); P_{max} = photosynthetic rate of upper leaves at light saturation (g CO_2 m^{-2} d^{-1}); δ = photoperiod duration (d d^{-1}). Three assumptions underly Eq. (3). First, light and P_{max} follow the same negative exponential curve from the top of the canopy to the bottom, whereas α is assumed to be independent of leaf position (Beyschlag et al. 1990; Grindlay 1997). Second, the leaf photosynthetic light-response curve is a rectangular hyperbola. Third, the diurnal course of radiation is sinusoidal.

The parameters of the photosynthetic light-response curve (P_{max} and α) in Eq. (3) are derived from the biochemistry of leaf photosynthesis of C_3-plants (Eq. 40 in (Farquhar et al. 1980); Eqs 16.32 and 16.62 in (Farquhar and Von Caemmerer 1982)), and depend on leaf Rubisco and leaf chlorophyll content, respectively. The Farquhar-equations operate on the internal CO_2

concentration in the leaves (C_i), and stomatal regulation is assumed to maintain C_i at 0.7 times the ambient concentration (Wong et al. 1979).

Influx of ozone

Ozone uptake by the canopy occurs mainly through the stomata, during the photoperiod (Chapter 2) (Allen 1990; Polle 1996). Canopy stomatal conductance for CO_2 influx can be calculated as gross canopy CO_2 uptake (= GR/γ) divided by the concentration difference between ambient and internal CO_2 concentration. Because ozone follows the same path as CO_2, and because ozone and CO_2 molecules are of similar size with similar molecular diffusivities, canopy conductance for O_3 is approximately the same as that for CO_2:

$$g_s = \frac{CGR}{\gamma (C_a - C_i)} \qquad (4)$$

where g_s = canopy stomatal conductance for CO_2 and O_3 (g gas m^{-2} ground d^{-1} / (ml gas m^{-3})). Note that Eq. (4) implies that ozone has no direct effect on stomatal conductance and only affects conductance indirectly by decreasing the photosynthetic capacity of the mesophyll (Balaguer et al. 1995; Farage et al. 1991; Lehnherr et al. 1988).

Ozone damage relations

In the literature, one can find four apparent ways in which ozone damages plants:

1. Decreased rate of photosynthesis
2. Increased rate of respiration
3. Accelerated leaf senescence
4. Decreased allocation to roots

Instead of incorporating these four apparent damage mechanisms simply as modifiers of LUE, leaf longevity and allocation, a more mechanistic representation of the ozone damage relations was chosen. We consider the increased respiration to be the result of energy-requiring detoxification of O_3 and repair of damage, the changes in photosynthesis and senescence to be caused by leaf protein breakdown caused by ozone, and the allocation changes to be the result of the increased energy-demand in the shoot (Fig. 4.1).

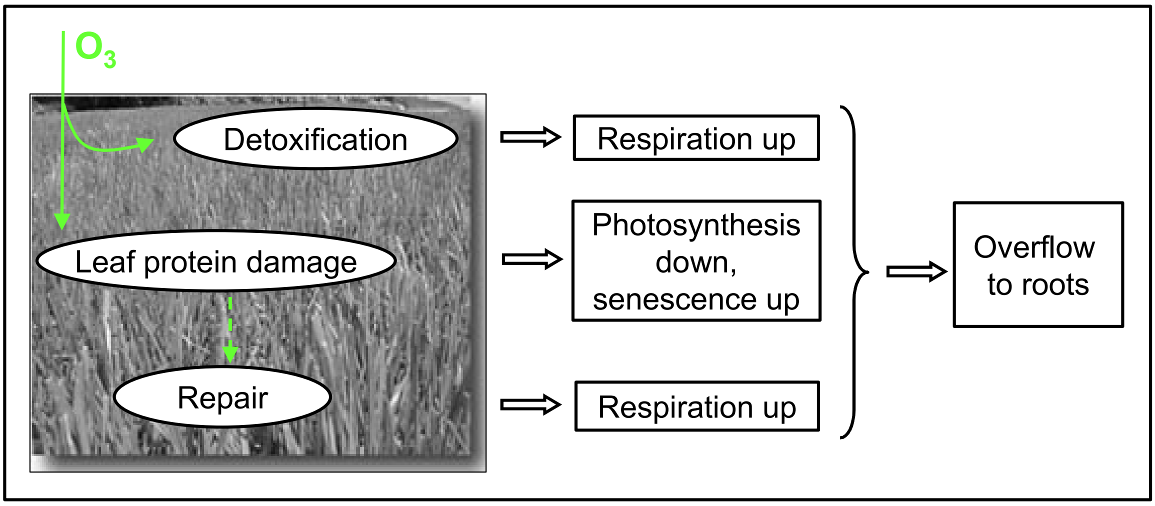


Figure 4.1. Ozone damage relations.

In the following, we will give a more detailed description of the modelling of the ozone damage relations. We begin with ozone uptake. The internal ozone concentration in leaves is nearly zero (Laisk et al. 1989), so the rate of ozone uptake is proportional to the ambient ozone concentration, but only part of the influx is effectively damaging:

$$F_{O3} = g_s \, O_{3,a} \, (1-f_{detox}) \qquad (5)$$

where F_{O3} = effectivse O_3 uptake by the canopy, after accounting for that part of the flux that is detoxified by inert plant material as cell walls and by biochemical detoxification mechanisms (g O_3 m^{-2} ground d^{-1}); $O_{3,a}$ = the photoperiod average of ambient ozone concentration (ml O_3 m^{-3}); f_{detox} = the detoxified fraction of the ozone flux entering the stomata (-).

Detoxification in plants exposed to ozone proceeds mainly by the reaction of antioxidants, such as ascorbic acid and various polyamines, with radicals formed due to ozone. The rate of formation or regeneration of antioxidants implies metabolic costs for the plants, which we capture in the model parameter c_{detox} (g CH_2O g^{-1} detoxified O_3).

Ozone damages leaf tissue in various ways, but most effects are secondary to an initial decrease in Rubisco concentration (Farage et al. 1991; Lehnherr et al. 1988; Pell et al. 1994); the mechanisms of light capture and electron transport are not affected (Nie et al. 1993). In the model, we quantify the damage to Rubisco by the ozone molecules that escaped detoxification by means of the O_3-damage coefficient φ (g Rubisco denatured g^{-1} O_3).

Plants can repair some of the damage. Schut (1985) developed a model for short-term effects of ozone, which included repair processes. He made use of the observation by Sutton and Ting (1977) that repair of foliar tissue from ozone injury required light, and showed that the rate of repair was proportional to the photosynthetic rate. However, in long-term ozone damage experiments, repair decreases with leaf age (Nie et al. 1993). We therefore modified Schut's approach by assuming that the repair rate is directly proportional to the amount of photosynthates normally allocated to the leaves, rather than to overall assimilation rate. The proportionality constant as well as the cost coefficient for repair are the two final parameters needed in our model: f_{repair} = fraction of assimilates, allocated to leaves, that is used in repair (g g^{-1}); c_{repair} = the repair cost coefficient (g CH_2O g^{-1} Rubisco repaired).

The metabolic costs of detoxification and repair reduce the amount of assimilates available for growth, and thus decrease biomass formation efficiency (γ) (Amthor 1988; Pell et al. 1994).

Soil water balance

A soil water balance was incorporated in the model. If the model is run with one species only (e.g. grass without clover), one soil layer is simulated, which increases in depth with root depth growth. Water is added to this layer by rainfall, and is lost by plant transpiration, soil evaporation and drainage. In the two-species case, i.e. grass-clover competition, two soil layers are simulated, an upper one with roots of both species, and a lower one with roots of the deepest rooting species only.

Modelling competition

The model simulates competition between clover and grass for light and water. The competition for light depends on the positioning of the foliage in space (height and angle), so height becomes an additional variable that the model must calculate. Height is calculated from aboveground biomass by means of an allometric equation: height increases with the square root of biomass.

Model inputs

Besides parameters that quantify plant and soil characteristics, the model requires as input daily weather data (temperature, light intensity, rain, humidity, wind speed) and daily data for atmospheric composition (O_3, CO_2).

Model parameterisation

Measurements of leaf N-content from the solardomes (Chapter 1) and field (Chapter 3) were used to estimate leaf Rubisco contents. Values for the parameters of the Farquhar-equations underlying the calculation of sward LUE were based on the measurements of $V_{c,max}$ (μmol CO_2 m^{-2} leaf s^{-1}) and J_{max} (mol m^{-2} s^{-1}) in the solardomes. Leaf chlorophyll contents were estimated from the Bangor J_{max} data, assuming that a J_{max} value of 100 mol m^{-2} s^{-1} corresponds to a chlorophyll content of 300 μmol chlorophyll m^{-2} leaf.

Model implementation

The parent models LINTULCC and LINGRA, on which the O3GRASSLAND model was based, had been programmed in the code-based simulation software FST. In O3GRASSLAND, a more modern and flexible simulation environment was chosen, i.e. the graphical modelling tool Simulink, which is an add-on to the numerical analysis software MATLAB.

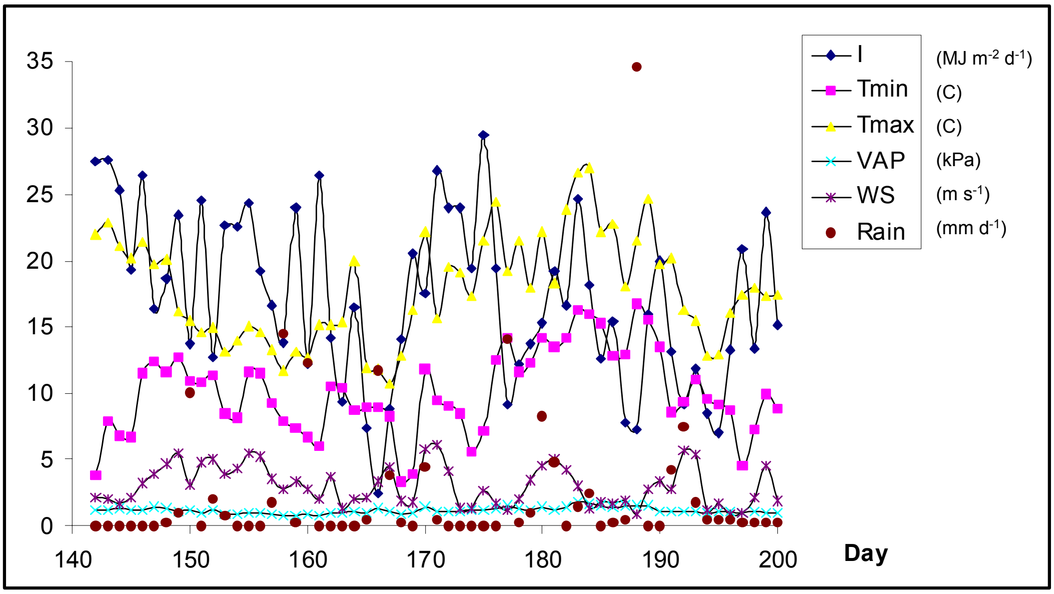
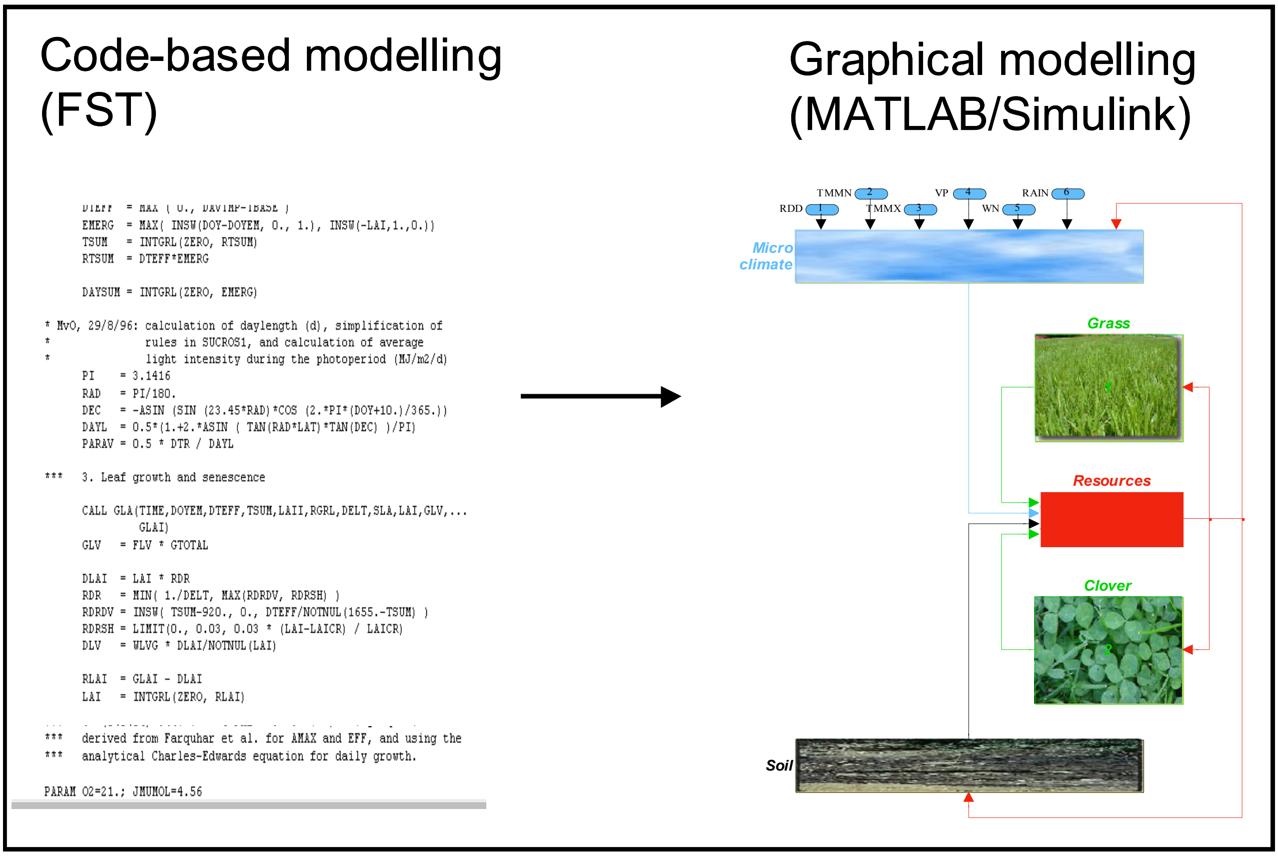


Figure 4.2. Weather input data, Easter Bush 2001. The six daily weather input variables required by the model are: light intensity (I), minimum and maximum temperature (Tmin, Tmax), vapour pressure (VAP), wind speed (WS) and rain.



```
      DTEFF  = MAX ( 0., DAVTMP-TBASE )
      EMERG  = MAX( INSW(DOY-DOYEM, 0., 1.), INSW(-LAI,1.,0.))
      TSUM   = INTGRL(ZERO, RTSUM)
      RTSUM  = DTEFF*EMERG

      DAYSUM = INTGRL(ZERO, EMERG)

* MvO, 29/8/96: calculation of daylength (d), simplification of
*               rules in SUCROS1, and calculation of average
*               light intensity during the photoperiod (MJ/m2/d)
      PI    = 3.1416
      RAD   = PI/180.
      DEC   = -ASIN (SIN (23.45*RAD)*COS (2.*PI*(DOY+10.)/365.))
      DAYL  = 0.5*(1.+2.*ASIN ( TAN(RAD*LAT)*TAN(DEC) )/PI)
      PARAV = 0.5 * DTR / DAYL

***   3. Leaf growth and senescence

      CALL GLA(TIME,DOYEM,DTEFF,TSUM,LAII,RGRL,DELT,SLA,LAI,GLV,...
               GLAI)
      GLV   = FLV * GTOTAL

      DLAI  = LAI * RDR
      RDR   = MIN( 1./DELT, MAX(RDRDV, RDRSH) )
      RDRDV = INSW( TSUM-920., 0., DTEFF/NOTNUL(1655.-TSUM) )
      RDRSH = LIMIT(0., 0.03, 0.03 * (LAI-LAICR) / LAICR)
      DLV   = WLVG * DLAI/NOTNUL(LAI)

      RLAI  = GLAI - DLAI
      LAI   = INTGRL(ZERO, RLAI)

***   derived from Farquhar et al. for AMAX and EFF, and using the
***   analytical Charles-Edwards equation for daily growth.

PARAM O2=21.; JMUMOL=4.56
```

Figure 4.3. Implementation of the O3GRASSLAND model. In contrast to its parent models, the model was not implemented in a code-based language (left panel), but in a graphical model (right panel, showing the upper level of the hierarchically structured model).

Results and Discussion

Three simulation experiments were performed:

1. Simulation of sward growth and fluxes as in the field 2001 & 2002
2. Sensitivity analysis for the effects of ambient [CO_2], temperature and light intensity on the response of the sward to ozone (using as default conditions those in the field 2001)
3. Simulation of grass-clover interaction.

Simulations and measurements for the field conditions in 2001 and 2002

In both years, the model accounted well for observed variation in fluxes of CO_2 and water between the sward and the atmosphere. O_3-fluxes were only measured accurately in 2001, and the model explained a quarter of the variation in the data. That is low, but still an improvement on models that assume constant stomatal conductance and thus calculate ozone-uptake as proportional to atmospheric concentration only (a linear regression of measured O_3-flux on atmospheric concentration has $r^2 < 0.1$ for the 2001 data). Note that we can evaluate the simulations of CO_2- and O_3-fluxes only by inspecting the correlations between observed and simulated fluxes, not by comparing the absolute values, because the observations include the contributions from both vegetation and soil, whereas the simulations only account for the vegetation. Therefore the simulated CO_2-uptake is higher than the measured fluxes (because the CO_2-loss by soil microbial respiration is not simulated) and simulated O_3-deposition is lower than the measured deposition (because non-stomatal deposition and deposition onto soil are not simulated).

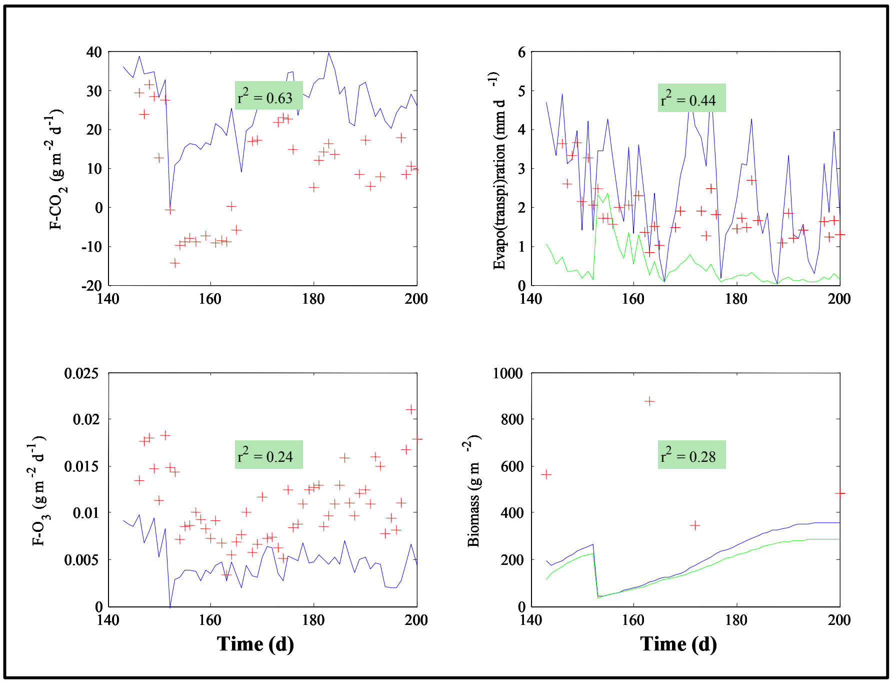


Figure 4.4. Simulations and measurements for the Easter Bush grassland, 2001. Top-left: CO_2-uptake by sward. Top right: water loss from sward (blue) and ground (green). Bottom-left: O_3-uptake by sward. Bottom-right: above-ground vegetation dry matter (including and excluding reserves). Crosses: measurements (see Chapters 2 and 3). The r^2-values indicate the fraction of the variation between data-points accounted for by the model.

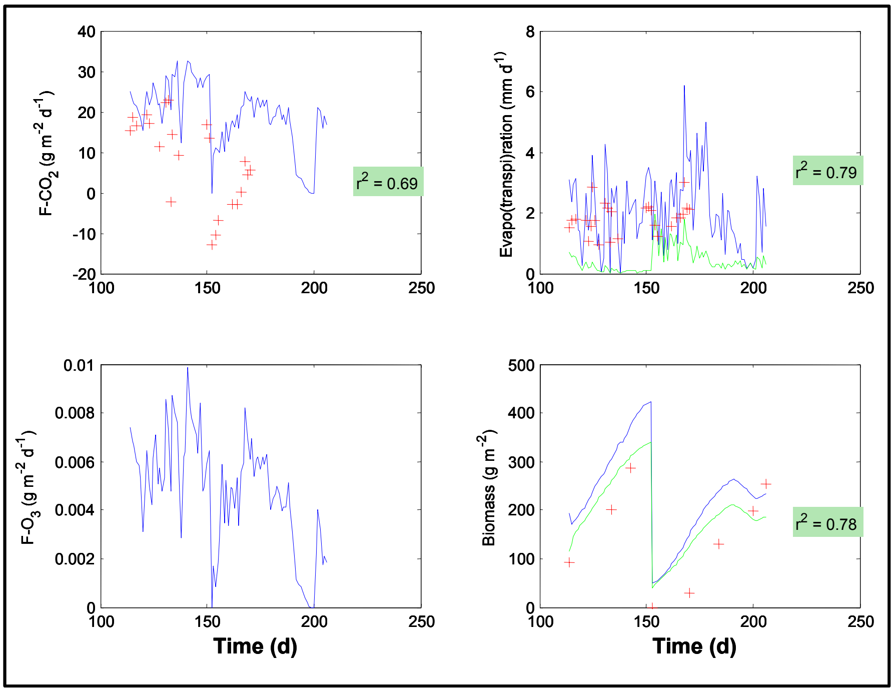


Figure 4.5. Simulations and measurements for the Easter Bush grassland, 2002. Symbols as in Fig. 4.4.

The simulation of the time course of biomass compared very well to the measurements in 2002, but were less close for 2001. The mismatch in 2001 was likely due to errors in measurement, as grass growth appeared to be similar in both years, on visual inspection, while the data suggested a threefold higher biomass density in 2001.

Simulations of O_3-CO_2-temperature-light interaction

In this experiment 12 simulation runs were carried out. The default run was the same one reported above for the field conditions in 2001. The set of 12 runs was formed by combining two levels of ozone (default and five times default), two levels of CO_2 (default and doubled) and three types of weather (default, "warm" (= default temperature + 5 ºC), "dark" (= default light intensity x 0.75)).

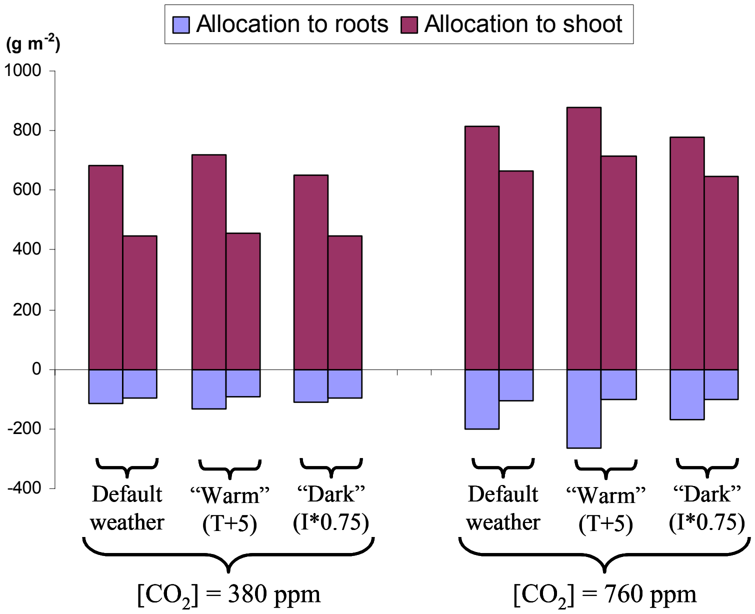


Figure 4.6. Simulated cumulative grass yields under 12 different sets of environmental conditions. Leftmost column: default conditions (Easter Bush, 2001). The right column of each pair shows the effect of a five-fold increase in atmospheric O_3 concentration. Other environmental conditions (CO_2, temperature, light intensity) are indicated under the columns.

The simulations corresponded well with observations in the literature that elevated CO_2 not only increases growth, but can also afford some protection against damage by O_3 (Rudorff et al. (1996), McKee et al. (1995), Rao et al. (1995)). Simulations showed smaller percentages yield loss due to O_3 at elevated CO_2 (Fig. 4.6). In the model, the causes for decreased O_3-damage under elevated CO_2 are (1) decreased stomatal conductance, which itself is a consequence of the mechanism that keeps the C_i/C_a ratio constant, and (2) decreased source-limitation in the vegetation.

The model suggested that root growth, like shoot growth, was always hampered by high levels of ozone. However, ozone only caused a decrease of root-shoot ratio at elevated CO_2.

In contrast to CO_2, differences in light intensity and temperature did not markedly affect the sensitivity to O_3, nor the allocation between root and shoot (Fig. 4.6).

Simulation of grass-clover competition

In the simulations of competition, differences in height are important because higher plants have better access to light. However, this aspect of the simulations was difficult to test using the Bangor data. In the Bangor data, weight increased while height decreased in *Lolium* between intermediate and final harvest. One minor cause for this is that some leaves fell over the side of the containers, but the major cause will have been that *Lolium* got more tillers during the growing period, i.e. there was an increase in biomass near the soil surface rather than in height.

Figs 4.7 and 4.8 show the results of the grass-clover simulations, with the same output-variables shown in Figs 4.4 and 4.5. The two species were initialised at the same biomass density, but the simulated grass was the stronger competitor, which is most clearly seen in the time courses of biomass. Note, however, that in these simulations nitrogen is not assumed to be limiting growth, so the outcome of the competition is dominated by the capacities of the species for CO_2-assimilation, which is highest in the grass (because of higher Rubisco and chlorophyll contents).

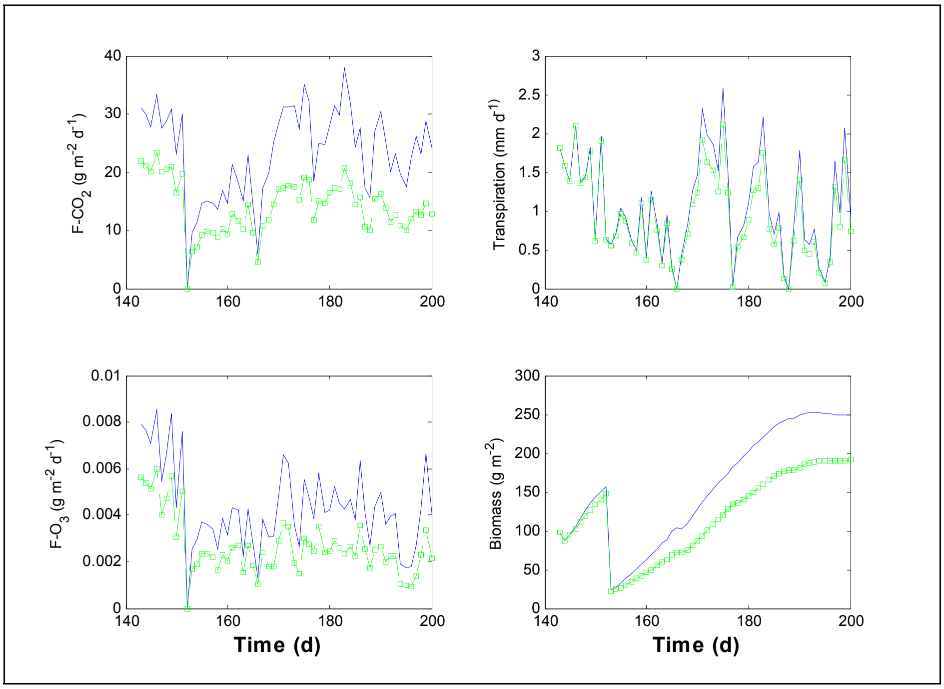


Figure 4.7. Simulation of grass-clover competition under default conditions (Easter Bush, 2001). The panels show the same variables as in Figs 4.4 and 4.5. The simulations start from equal biomass densities of grass (solid blue lines) and clover (marked green lines) at day 143.

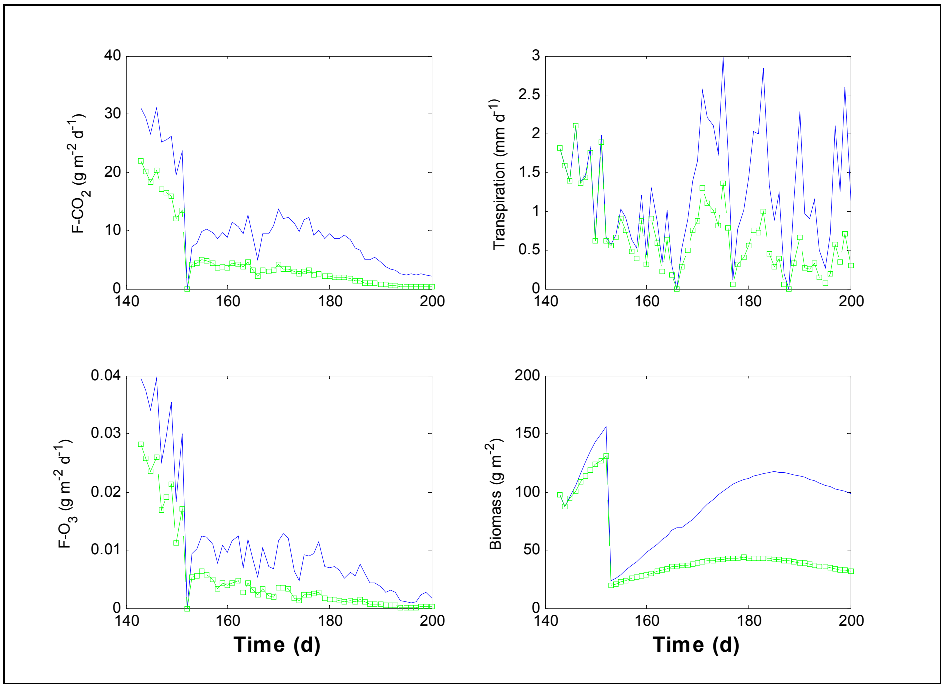


Figure 4.8. Simulation of grass-clover competition under high ozone-conditions (= 5 times the default conditions of Easter Bush, 2001). Variables and symbols as in Fig 4.7.

The comparison of Fig 4.7 (low, actual O_3-levels) and 4.8 (fivefold-O_3 levels) shows that the competitive advantage of the grass was exacerbated by high levels of ozone: relative biomass-loss was least in *Lolium*. The model results suggest that the tolerance of *Lolium* to ozone was due to its high initial contents of Rubisco and chlorophyll. Growth is determined by the source-sink balance in both plant species, and some loss of source strength (i.e. loss of Rubisco and chlorophyll) can be tolerated if source strength is high to begin with and growth therefore mostly sink-limited.

Conclusions

- A simple, dynamic, process-based model was constructed. The model accounted for much of the variation in fluxes and growth observed in the field.
- Parameterisation of the model from measurements under controlled growing conditions in solardomes appeared successful, even though the model was applied to field conditions.
- The simulation of ozone-uptake was an improvement over simple correlations with ambient O_3-concentration but still needs further improvement.
- The model predicted that biomass growth would be decreased less by O_3 at elevated CO_2, which is consistent with the literature.
- There is little information in the literature on the effect of ozone on allocation patterns in grassland species. The model suggests that the root-fraction will only decrease at elevated CO_2.
- No weather conditions conducive to ozone-damage were identified: interactions of O_3 with light and temperature were insignificant.
- In the solardomes, *Lolium* was shown to be more tolerant to ozone than clover. This was confirmed and explained by the model as the result of differences in leaf composition and source-sink relations between the two plant species.
- The simulations with high-ozone levels in the field used hypothetical pollution scenarios, as the ozone-levels on the grassland site were generally low. There is a need to test the model more extensively under actual field conditions with higher levels of ozone. To assess the capacity of the model to predict not only fluxes, but also damage, a field with more ozone-sensitive vegetation than grassland might need to be chosen.

5. General discussion

Model development

During the past four decades, many process-based models of the growth of crops, forests and grasslands have been made. Such models have only rarely been used for the analysis of the effects of air pollution on vegetation, and statistical approaches have become the standard for prediction of damage. The strength of the statistical approaches, e.g. regressions of damage on AOT40, is that they are easy to parameterise and use. However, it is difficult to extend the regression equations with interacting factors, like the presence of competing species or the effects of different weather conditions, especially when the interactions change over time. Therefore, in O3GRASSLAND, we chose process-based modelling as the tool to analyse ozone damage to grassland. To keep model structure as simple as possible, we captured all of the ozone relations in only five parameters, which define the amount of damage to leaf tissue per unit of ozone and the rates and costs of detoxification and repair. Competition between grass and clover, and how this is affected by ozone, were modelled as well. The two main lessons learned from the model development were, first, that it was relatively straightforward to build such a model of ozone relations in grassland, and second, that even this simple model could not be parameterised from literature data only.

Physiological measurements and model parameterisation

In the solardome experiments carried out at CEH-Bangor, the effects of ozone on gas exchange, growth and senescence of grass, clover and grass-clover mixtures were determined. The experiments provided insight into the differences in ozone tolerance of both species, and helped parameterise the process-based model. Ozone fumigation increased senescence in both species, affected gas exchange strongly in clover but less so in grass, and decreased biomass yield only in clover. The fact that ozone decreased carboxylation capacity of the grass but not its biomass yield was a surprising finding. Solardome data were used to parameterise, in the process-based model, the different gas exchange characteristics of the two species, and the growth data were used for model testing. The solardome experiments were labour-intensive, but provided much information on the effects of ozone on the competition between grass and clover. The experiments did not provide parameter values for the rates and costs of ozone detoxification and repair of damage. Such parameter quantification will require different experiments with more short-duration high-ozone episodes, and with more precise quantification of the changes in leaf respiration associated with the processes of detoxification and repair. In the solardome experiments, gas exchange was always measured after ozone-fumigation was switched off, but measuring the immediate effects of ozone on gas exchange would allow a better assessment of the dynamics of damage. Quantifying changes in root-shoot allocation may require experiments on soil solution where the root system can be more easily isolated, although such a set-up is probably irreconcilable with the aim to study grass-clover competition.

Field measurements and model testing

The field site where gas fluxes and grass growth were measured, was also the source of the plant material used in the solardome experiments. Measurements of leaf photosynthesis parameters showed similar results under both conditions, which supported the approach taken whereby the model was parameterised using solardome information before being tested against field data. Overall, the model simulated the field fluxes of CO_2 and H_2O and the growth of the grass well. The correlation between measured and simulated ozone flux was only a moderate improvement over a simple linear regression of flux on ambient concentration. However, it is difficult to evaluate the simulation of the ozone flux, as the simulations and the measurements quantified different processes: the model only simulated stomatal uptake by the vegetation, whereas the measurements quantified the overall flux, which included non-stomatal fluxes to vegetation and soil. There clearly remains a further need for testing the model against measurements of the precise processes that it simulates.

Model application

When the model was applied to the growing conditions in the solardomes, it helped explain the puzzling observation that carboxylation capacity had been reduced in grass but not its biomass yield, suggesting that the experiment had been in fact too short to reveal any effects on growth. The model further explained the differences between grass and clover in ozone tolerance as the result of differences in leaf composition that make clover more source-limited than grass.

A key objective of O3GRASSLAND was to identify environmental conditions conducive to ozone damage. Application of the model led to the perhaps surprising finding that temperature and light intensity, albeit affecting growth itself strongly, had only small effects on the growth response to ozone. This in fact suggests that simple linear regressions of ozone damage on ozone concentration or flux may have wider validity than expected when the project was started, although better model parameterisation and testing is of course needed before such a statement can be reliably made.

Model simulations did show that increases in atmospheric CO_2 concentration are likely to afford some protection against ozone. This interaction between the effects of the two gases had already been repeatedly demonstrated in the literature and been attributed to reduced stomatal conductance at elevated CO_2. The model confirmed the role of stomatal closure but also attributed part of the protective effect of CO_2 to its decreasing source-limitation in the vegetation.

Future activities

Project O3GRASSLAND showed a proof of principle: a process-based model of the effects of ozone on vegetation is feasible with present-day understanding of the system, if sufficient information at the plant and field level is available. However, the project duration of two years was too short to fully test the mechanisms implemented in the model. In a future project, more time should be allocated to measurements aimed at model parameterisation and testing, to the extent that a full uncertainty analysis of the model becomes possible. There is a need for further model development too. We have indicated some mismatches between what the model simulates and what can be measured. The field flux measurements included non-stomatal ozone deposition onto vegetation and soil, and for improved testability of the model these two fluxes could be included in the model. Furthermore, the measurements had high temporal resolution, in many cases 15-minute intervals, whereas the model operates on a daily time step. Developing a model version that simulates diurnal time courses of fluxes may be useful to allow using more of the information in the measured time series. However, keeping the model simple and fast is a goal in itself, so a separate daily-time step model may need to be maintained as well.

The field flux measurements initiated in O3GRASSLAND are continuing with different sources of funding, but the simultaneous physiological study and integration by means of process-based modelling have been stopped. Publications are in preparation (see Appendix 3).

Acknowledgements

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Appendix I. Project Proposal 2001

Note: this appendix shows the proposal for O3GRASSLAND as submitted by the project group on 17 January 2001. On 28 March 2001, the group received notice that the proposal was accepted, with the proviso that funding was at the lower level of £100k and the work should be carried out in two years rather than three. The consequently revised Project Initiation Form (PIF) is shown in Appendix 2.

Page 1

INTEGRATING FUND: ROUND 7

17/1/2001

PROJECT TITLE: Bridging the gap: From ozone effects on single plants to predicting ecosystem effects based on field-scale fluxes
PROPOSER AND SITE: (A) M. van Oijen (Project Leader), P. Levy & M. Murray (CEH-Edinburgh, Section Ecosystem Modelling)
COLLABORATORS AND LOCATION: (B) G. Mills, T. Ashenden & F. Hayes (CEH-Bangor, Section Air Pollution & Climate Change)
(C) D. Fowler, M. Coyle & K. Hargreaves (CEH-Edinburgh, Section Trace Gas Fluxes and Air Pollution)
Dates: **Start:** 1 / 4 / 2001
End: 31 / 3 / 2004
CEH Science Programme: Programme 7: Pollution

Cost Summary	Year 1 2001-2	Year 2 2002-3	Year 3 2003-4
Site/Division A	£ 17K	£ 17K	£ 17K
Site/Division B	£ 17K	£ 17K	£ 17K
Site/Division C	£ 25K	£ 12K	£ 12K
Total	£ 59K	£ 45K	£ 47K

Objectives of the Research:

Overall objective:
To develop a mechanistic model to predict ozone damage to grassland from plant, weather and ozone parameters

Specific objectives:
1 To construct a process-based simulation model of the physiological responses of the plants of a grassland community to ozone (M. van Oijen, P. Levy & M. Murray).
2. To parameterise and test the model by measuring field-scale fluxes of ozone, carbon dioxide uptake and water vapour above a managed grassland, quantifying interactions between the three gaseous fluxes and meteorological variables (D. Fowler, M. Coyle & K. Hargreaves).
3. To evaluate the model by measuring effects of ozone on the physiological processes of the individual species under controlled exposure conditions (G. Mills, T. Ashenden & F. Hayes).
4. To use the model, and the field measurements, to identify weather conditions under which managed grassland is most sensitive to ozone (M. van Oijen, P. Levy & M. Murray).
5. To derive recommendations for future experimental and modelling research on air pollution in CEH, based on (1) knowledge gaps identified during the construction of the model; (2) the major environmental interactions identified in both experiments and modelling (M. van Oijen, P. Levy, M. Murray, G. Mills, T. Ashenden, F. Hayes, D. Fowler, M. Coyle, K. Hargreaves).

Pages 2 – 4

Project Description (methodology and approach):

1. Introduction

Ozone can damage plants and is **the pollutant that currently represents the greatest threat to crop production in Europe**. To estimate the potential for ozone effects on vegetation, statistical approaches are generally adopted. Most recently, yield and biomass changes have been related to AOT40, the Accumulated Ozone exposure above a Threshold of 40 ppb O_3 (Fuhrer et al., 1997). The latter is the product of concentration and time and fits the data better than previously-used parameters such as the 7h mean. However, AOT40 is not the absorbed physiologically active dose (Fowler et al., 1997), and thus its use still leaves much variation in dose-response data unaccounted for. For example, there is a tendency for AOT40 to overestimate the magnitude of O_3 effects because the environmental conditions leading to large O_3 concentration are also correlated with high VPD and reduced stomatal conductance and flux of ozone. Statistical approaches such as correlations of plant damage with AOT40 ignore interactions of ozone with other environmental factors, and also ignore variability in the sensitivity of the plants to ozone. The first step beyond AOT40 would be to replace "exposure" with "uptake", i.e. statistically relating damage to cumulative ozone-uptake rather than exposure. That step is already being taken for single species such as some crops and trees (Reich, 1987; Fowler et al., 1998, Emberson et al., 2000). However, the challenge remains to determine ozone uptake for a mixed species grassland where individual species will respond differently to ozone both in terms of uptake and physiological effect. Such grassland is the dominant land-cover for the UK, and thus is very important for the consideration of national uptake/deposition budgets for ozone. In the proposed project, we intend to include a study of the physiological damage relationships for the component species of managed grassland, and show how they relate to field-scale processes and fluxes by **the development of a mechanistic model**.

We distinguish three steps in ozone stress: 1 ambient ozone concentration causes a certain deposition rate; 2 part of the deposited ozone is taken up by the vegetation through the stomata; 3 ozone uptake by the vegetation causes damage. Progress in the mechanistic modelling of these three steps has been hampered so far because of lack of empirical information. However, steps 1 (ozone deposition) and 2 (ozone uptake) have become the subject of **field-scale studies in CEH** recently (Fowler et al., 1997, 1998, 1999), and step 3 (damage relations) has become the subject of **physiological research in CEH** (Ball et al., 1998; Mills et al., 2000). **Preliminary physiological models of ozone damage** have also been made in the recent past (Van Oijen & Ewert, 1999; Ewert et al., 1999) but, as indicated above, these have been hampered in their applicability because of insufficient underlying empirical information. Better models may now be made if the two lines of investigation of field-scale flux quantification and detailed physiological studies of damage relationships for individual species are tied together. We will use mechanistic modelling to help integrate the two approaches and allow better analysis and, ultimately, prediction of ozone-damage.

The field-scale and physiological measurements will complement each other and provide data that can be used to develop and test the model. Experiments will be set up where physiological and field-scale measurements quantify the processes that the model simulates. Physiological studies as well as field-scale studies will be carried out with species from grasslands. Fields dominated by perennial ryegrass are available close to Collaborator C and grasses are well suited to growth in controlled environments. Furthermore, use may be made of ozone research that has already been done with other Gramineae such as wheat (e.g. Fangmeier et al., 1999; Mitchell et al., 1999; Ommen et al., 1999). Several of the annual and perennial broad-leaved species that are also present in the grassland have already been tested for sensitivity to ozone at CEH-Bangor (Mills, unpublished), and experimental methods have been established. The existing knowledge will help make the transition from a statistical to a more data-demanding mechanistic model feasible.

The division of tasks will be such that Bangor-Pollution carries out the detailed physiological studies, Edinburgh-Pollution does the field-scale flux studies, and the integrative modelling is done by Edinburgh-Modelling.

2. Research questions and project aims

The two general questions of this project are: (1) Which component species have the greatest fluxes of ozone, and for these, which vegetation processes are affected by ozone fluxes of different magnitude? (2) Are there interactions between weather and growth stage, and if so, which weather and crop conditions lead to the greatest sensitivity to ozone?
From these two general questions the following specific working questions were derived:

1. To what extent is the temporal variability of field-scale ozone fluxes correlated with specific meteorological variables?
2. What fraction of field-scale ozone fluxes is taken up by the individual species, how is the deposition flux partitioned between stomata and non-stomatal sinks and to what extent do these two sinks interact?
3. To what extent does ozone decrease the rate of photosynthesis, increase the rate of respiration, accelerate leaf senescence and decrease allocation to the root system in the component species?
4. Can we develop a physiological model that helps answer questions 2 and 3 above by: (a) simulating the net fluxes of O_3 and CO_2 and their interactions; (b) quantifying the relations between O_3 stomatal flux and meteorological variables; (c) identifying the weather and growth stages which lead to greatest sensitivity to ozone?

The aims of the project are to provide both the required data and the model to answer the listed questions.

3. Methodology and approach

The project involves modelling, solardome experiments and field measurements. The measurement activity will be integrated with the mechanistic approach used in the modelling, so the generated data must be usable for model parameterisation and testing. The following approach is planned for the modelling and the different experiments.

A. Physiological modelling (Edinburgh-Modelling)

We will develop a dynamic model that simulates grassland growth and response to its environment in a mechanistic and deterministic manner. The model will be based on a model developed earlier for spring wheat (Van Oijen & Ewert, 1999). This model simulates the processes of light absorption, biomass formation, thermal-time dependent biomass allocation to roots, leaves, stems and ears, leaf area expansion, and leaf senescence. The model has a one-day time step, and calculates daily growth rate by multiplying absorbed photosynthetically active radiation, on a ground-area basis, with a canopy light-use efficiency, which varies with the environmental conditions. This is a relatively simple approach, because it ignores within-day changes in process efficiencies and also simplifies the treatment of physiological differences between leaf layers. The present project may show that this approach is an oversimplification if we identify diurnal processes with important consequences for the response to ozone at the field-scale. The earlier model will be expanded to simulate in greater detail the physiological processes that are measured in the controlled exposure experiments, and will integrate those processes to allow prediction of the field-scale fluxes that are measured in the field study.

The four most common responses of plants to ozone are: (1) decreased rate of photosynthesis; (2) increased rate of respiration; (3) acceleration of senescence; (4) decreased allocation to the root system. In the model, these four processes will be mechanistically linked to that part of the ozone taken up by the plants that is not detoxified. Ozone-induced increased rate of respiration is of special importance as it may signify changes in plant maintenance rate due to increased need for repair (Amthor, 1988). Therefore, the model will have to explicitly simulate rates of ozone detoxification and repair. This is a strongly underdeveloped area of plant physiological modelling, and close cooperation with the experimentators in this project is essential.

The model predicts damage to the vegetation as a result of ozone-induced changes in the underlying physiological processes. The model thus needs data with which to parameterise the processes as well as data on vegetation growth to test the model predictions. These requirements will be met by the partners in the project. Bangor (Collaborator B) will provide the physiological process parameters for the model (see next section), and the Air Pollution group in Edinburgh (Collaborator C) will provide the field-level growth and flux data to test the model.

Both the experimental work and the modelling explore relatively unknown territory and the model thus will be extensively evaluated by parameter sensitivity analysis and model structure uncertainty analysis. After this evaluation, the model will finally be used to identify the major environmental conditions and plant characteristics which affect grassland response to ozone episodes of different intensity and duration.

B Physiological studies in solardomes (Bangor-Pollution)

These studies will be carried out in the solardome facility at CEH-Bangor (a set of hemi-spherical glasshouses with near-natural light conditions and well-controlled air flow cooled to ambient air), using the species that comprise the grassland used in the ozone flux measurements (see C). Preliminary exposure experiments have indicated that six of the nine dominant species are sensitive to ozone, with the most sensitive being *Trifolium repens*.

The experiments aim to determine which physiological processes are affected by ozone and to what extent in each of the species.

<u>Ozone-exposure.</u> In years one and two of the project, the plants will be exposed throughout their growth cycle to an episodic ozone regime. Each species will be grown individually in pots of size 10 x 10 x 30 cm (depth) containing agricultural loam soil, watered to field capacity, and randomly arranged in groups of 9 within insulated boxes with 8 boxes of pots per solardome. Each solardome will be supplied with two air changes per minute of air cooled to ambient temperature. Two domes will receive a <u>low</u> ozone regime of 30 ppb added to charcoal-filtered air whilst the other two will receive a <u>high</u> episodic ozone regime of 7h per day at 80, 100, 100, and 80 ppb added to charcoal-filtered air for days 1 - 4 respectively, followed by 3 days at 30 ppb for a 10 week period. These exposure regimes will provide 10 week AOT40s of 0 and 14,000 ppb.h for the two treatments respectively, and will be repeated twice per season allowing the ozone response to be monitored in four sets of climatic conditions. The design of experiments for year 3 will depend on the results of previous experiments and the requirements of the modelling process; it is likely to include responses of selected species to different ozone doses.

<u>Resource allocation and growth analysis.</u> Destructive harvests will be carried out for growth analysis on the whole plant scale: partitioning between leaves, roots and reproductive structures, and leaf area dynamics will be quantified.

<u>Stomatal conductance, photosynthesis and respiration.</u> Time-courses of diurnal changes in conductance, photosynthesis and respiration of the youngest fully expanded leaf will be made at intervals throughout each experiment.

<u>Ozone flux/deposition</u>. The partitioning of the applied ozone will be quantified between the three sinks in the solardomes: stomatal uptake by the plants, deposition on outer plant surfaces, and deposition on non-vegetation surfaces in the domes. This assessment of the relative strengths of the three ozone sinks in the chambers is needed to verify consistency with the partitioning of fluxes that occurs in the field, as measured at site C (see next section). Representatives from Collaborator C will travel to Bangor to assist in this activity.

<u>Ozone detoxification.</u> In year 3, the activity of apoplastic ascorbate, the main detoxicant of ozone in the laeves, will be determined for *Lolium perenne* plus one other species deemed to be important from the studies conducted in years 1 and 2, using methods developed by Lyons et al. (1998).

C Quantification of fluxes in the field (Edinburgh-Pollution)

Measurements of O_3-, CO_2- and H_2O-fluxes will be carried out in a field near Edinburgh (close to the location of Site C) during three growing seasons. The field is strongly dominated (>90%) by perennial ryegrass (*Lolium perenne*), with minor presence of *Urtica dioica*, *Trifolium repens*, *Ranunculus repens*, *Anthriscus caucalis*, *Dactylis glomerata*, *Cirsium pratense*, *Cirsium arvense* and *Rumex obtusifolius*. There will be no ozone applied to the field: the study focuses on natural variability of weather and ozone, under representative conditions. The fluxes will be monitored continuously during the period of most active grassland growth from April to September.

The measurement of O_3-, CO_2-, and H_2O-fluxes by micrometeorological methods will partly rely on existing equipment available at CEH-Edinburgh. Field measurements of this nature are in progress in a wheat field at Sutton Bonnington as a part of the DETR Ozone umbrella. The methods used there rely on the aerodynamic, flux-gradient approach with vertical gradients in ozone, wind velocity and air temperature forming the primary data to calculate the vertical flux density within the constant flux layer which develops above extensive, uniform vegetation. This proposal would extend the measurements to include CO_2- and H_2O-fluxes, measured using a LICOR fast response IRGA and application of eddy co-variance methods to measure the vertical flux. A LICOR CO_2 analyser must be bought for these measurements. Another requirement is the use of a sonic anemometer, which will greatly increase the quality of the underlying micrometeorological measurements and will allow the eddy diffusivity and sensible heat flux to be measured independently at the site by two methods.

The measurements of large-scale gas fluxes will be complemented by a small number of physiological gas exchange measurements, to ensure consistency with the more extensive programme of physiological measurements carried out by Collaborator B (see above).

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Programme and/or Plan of Research

		A: CEH-Edinburgh (Modelling)	***B: CEH-Bangor (Pollution)***	***C: CEH-Edinburgh (Pollution)***
2001	Apr	**KICK-OFF MEETING, Edinburgh** Aim: to finalise protocols for experimental studies. Specific goal: to refine treatments and measurement plan for both field-scale flux measurements and the solardome experiments to guarantee that the measurements are consistent and generate data that are required for model development.		
	Apr-Aug	Model development, with 1-2 VISITS TO BANGOR AND FIELD SITE to observe the experiments and discuss the physiological content of the model	Year 1 physiological experiments in the solardomes	Year 1 flux measurements in the field
	Aug-Nov	Model development	Data analysis	Data analysis
	Nov	Collection of data from the two experimental partners	Transfer of data to Edinburgh-Modelling	Transfer of data to Edinburgh-Modelling
	Dec	**SECOND MEETING, Bangor** Aims: (1) Preliminary evaluation of year 1 physiological experiments and field-scale flux measurements; (2) Verifying consistency of experimental and modelling work		
2002	Jan-Mar	Parameterisation with data from Bangor, testing of simulations on data from Edinburgh-Pollution	Preparation for year 2 solardome experiments	Preparation for year 2 field-scale flux determination
	Mar	**THIRD MEETING, Edinburgh** Aims: (1) Discussion of model simulation results and identification of areas of improvement of the model; (2) Identification of any required changes to the set-up of the measurement programmes.		
	Apr-Aug	Model development, VISITS TO BANGOR & FIELD SITE	Year 2 physiological experiments	Year 2 flux measurements
	Aug-Nov	Model development	Data analysis	Data analysis
	Nov	Collection of data from the two experimental partners	Transfer of data to Edinburgh-Modelling	Transfer of data to Edinburgh-Modelling
	Dec	**FOURTH MEETING, Bangor** Aims: (1) Discussion of year 2 results; (2) Planning for physiological experimentation and modelling in year 3.		
2003	Jan-Aug	Analyses of parameter sensitivity and structural uncertainty	Preparation and execution of year 3 experiments	Preparation and execution of year 3 flux measurements

	Aug-Nov	Identification of major ozone-effect determinants	Data analysis	Data analysis
	Nov	Collection of data from the two experimental partners. Last simulations.	Transfer of data to Edinburgh-Modelling	Transfer of data to Edinburgh-Modelling
	Dec	**FIFTH MEETING, Edinburgh** Aim: Discussion of results from year 3 modelling and experiments		
2004	Jan-Mar	Writing report and publications	Writing report and publications	Writing report and publications
	Mar	**SIXTH MEETING, Bangor** Aims: (1) Evaluation of project; (2) Discussion of future cooperation; (3) Discussion of research recommendations to CEH		

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Justification (relevance and benefit to CEH)

Relevance of the project

1. Successful completion of the work will advance understanding of air pollution relationships at both the plant and field scale.
2. The increased understanding is planned to be implemented in a mechanistic model. This will provide a valuable step towards developing an ozone damage prediction instrument that is better equipped than the statistical AOT40 approach to handle environmental interactions.
3. The project focuses on O_3, but its novel approach in combining field and solardome experiments with modelling may set a useful example for other vegetation-air pollution studies.

Benefit to CEH

1. The 'Programme Vision' of CEH Programme 7 (Pollution) identifies integrated modelling of pollutant processes as a priority.
2. Theme 7.3 of the Pollution Programme includes 'the investigation of exposure-response relationships for vegetation and materials' with the objective of formulating appropriate critical levels for exposure.
3. The work will provide scientific underpinning in an area of pollution research which is of growing importance, as background ozone levels increase. It will therefore strengthen CEH's position with DETR, UNECE, the EU and other customers concerned with ozone.
4. The project will strengthen links between the air pollution teams in CEH-Edinburgh and CEH-Bangor, and stimulate collaboration between experimentalist groups working at different integration levels (field, plant) and between experimentalists and modellers.
5. The project will identify important environmental and plant factors that modify the damaging effect of ozone. We also expect to encounter knowledge gaps, especially during the construction of the mechanistic model. Important damage-modifying factors, but also the knowledge gaps, are natural targets for future experimental and modelling research on air pollution. The project thus may help formulate research recommendations for the Air Pollution Programme of CEH.

Outputs & Deliverables

1. Data.
2. Model.
3. List of recommendations for future CEH-Research on air pollution.
4. Publications.

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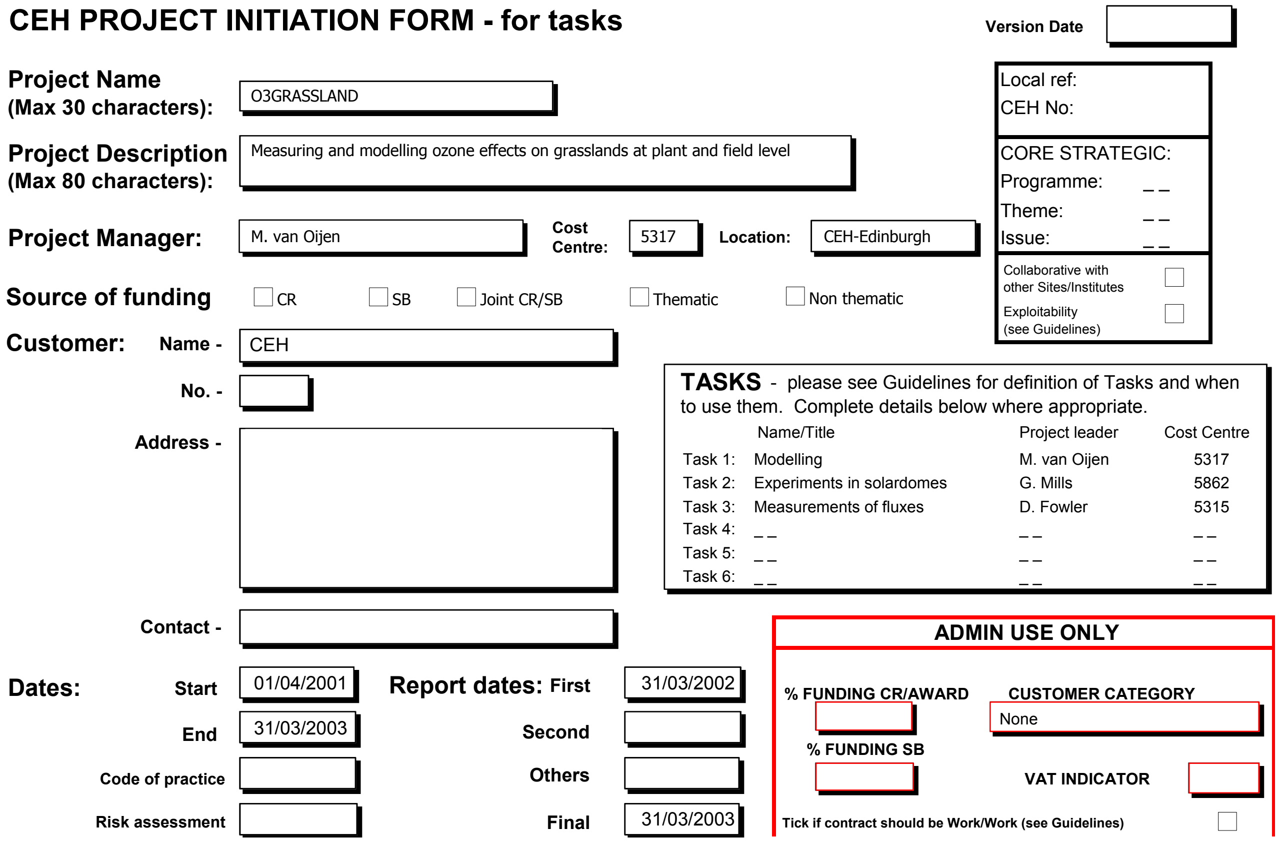
COST PROFILE

Staff Breakdown **[Salary + NI]**			**Year 1**	**Year 2**	**Year 3**
Site A (Edinburgh-Modelling, Project leader)					
Band 6	x	32%	£ 8.9 K	£ 9.3 K	£ 9.7 K
Site B (Bangor-Pollution)					
Band 4	x	2%	£ 0.8 K	£ 0.9 K	£ 0.9 K
Band 5	x	2%	£ 0.7 K	£ 0.8 K	£ 0.8 K
Band 7	x	28%	£ 6.3 K	£ 6.6 K	£ 6.9 K
Site C (Edinburgh-Pollution)					
Band 2	x	1%	£ 0.6 K	£ 0.6 K	£ 0.6 K
Band 5	x	2%	£ 0.8 K	£ 0.8 K	£ 0.9 K
Band 7	x	19%	£ 4.2 K	£ 4.4 K	£ 4.6 K
Total Salary			£ 22.4	£ 23.3 K	£ 24.3 K
Overheads at 70%					
Site A			**£ 6.3 K**	**£ 6.5 K**	**£ 6.8 K**
Site B			£ 5.5 K	£ 5.7 K	£ 6.0 K
Site C			£ 3.9 K	£ 4.1 K	£ 4.3 K
Total overheads			£ 15.7 K	£ 16.3 K	£ 17.0 K
Recurrent					
Site A (T&S and software)			£ 1.9 K	£ 0.9 K	£ 1.0 K
Site B (T&S and equipment)			£ 3.2 K	£ 3.0 K	£ 3.0 K
Site C (T&S and equipment)			£ 15.7 K	£ 2.0 K	£ 2.0 K
Total recurrent			£ 20.8 K	£ 5.8 K	£ 5.9 K
Capital					
Site A					
Site B					
Site C					
Total capital					
TOTAL COSTS			**£ 58.9 K**	**£ 45.5 K**	**£ 47.2 K**

Appendix II. Project Initiation Form (PIF)



CEH PROJECT INITIATION FORM - for tasks

Version Date

Local ref:
CEH No:

CORE STRATEGIC:
Programme: _ _
Theme: _ _
Issue: _ _

Collaborative with other Sites/Institutes ☐
Exploitability (see Guidelines) ☐

Project Name (Max 30 characters): O3GRASSLAND

Project Description (Max 80 characters): Measuring and modelling ozone effects on grasslands at plant and field level

Project Manager: M. van Oijen **Cost Centre:** 5317 **Location:** CEH-Edinburgh

Source of funding ☐ CR ☐ SB ☐ Joint CR/SB ☐ Thematic ☐ Non thematic

Customer: **Name -** CEH

No. -

Address -

Contact -

TASKS - please see Guidelines for definition of Tasks and when to use them. Complete details below where appropriate.

	Name/Title	Project leader	Cost Centre
Task 1:	Modelling	M. van Oijen	5317
Task 2:	Experiments in solardomes	G. Mills	5862
Task 3:	Measurements of fluxes	D. Fowler	5315
Task 4:	_ _	_ _	_ _
Task 5:	_ _	_ _	_ _
Task 6:	_ _	_ _	_ _

Dates:

Start	01/04/2001	**Report dates: First**	31/03/2002
End	31/03/2003	**Second**	
Code of practice		**Others**	
Risk assessment		**Final**	31/03/2003

ADMIN USE ONLY

% FUNDING CR/AWARD

CUSTOMER CATEGORY None

% FUNDING SB

VAT INDICATOR

Tick if contract should be Work/Work (see Guidelines) ☐

Financial Year 1: 2001/02

0	Name	Cost Centre No.	Grade	Days	Hours	Staff Cost Rate	Total SCR	Charge Rate for Billing	Total CRB
Task 1	M. van Oijen	5317	BAND 6	21	155.4	16.57	2575	28.17	4377
	P. Levy	5317	BAND 6	21	155.4	16.57	2575	28.17	4377
	M. Murray	5317	BAND 6	21	155.4	16.57	2575	28.17	4377
			Task 1 - Total	63	466.2		7724		13131
Task 2	TW ASHENDEN	5862	BAND 4	4	29.6	28.18	834	47.90	1418
	G MILLS	5862	BAND 5	10	74.0	21.63	1601	36.77	2721
	F HAYES	5862	BAND 7	75	555.0	13.22	7336	22.47	12471
			Task 2 - Total	89	658.6		9771		16610
Task 3	D. Fowler	5315	BAND 2	1	7.4	39.80	295	67.66	501
	M. Coyle	5315	BAND 7	27	199.8	13.22	2641	22.47	4490
	K. Hargreaves	5315	BAND 5	4	29.6	21.63	640	36.77	1088
			Task 3 - Total	32	236.8		3576		6079

Financial Year 2: 2002/03

0	Name	Cost Centre No.	Grade	Days	Hours	Staff Cost Rate	Total SCR	Charge Rate for Billing	Total CRB
Task 1	M. van Oijen	5317	BAND 6	21	155.4	17.23	2678	29.29	4552
	P. Levy	5317	BAND 6	21	155.4	17.23	2678	29.29	4552
	M. Murray	5317	BAND 6	21	155.4	17.23	2678	29.29	4552
			Task 1 - Total	63	466.2		8033		13656
Task 2	TW ASHENDEN	5862	BAND 4	4	29.6	29.31	867	49.82	1475
	G MILLS	5862	BAND 5	10	74.0	22.50	1665	38.24	2830
	F HAYES	5862	BAND 7	75	555.0	13.75	7630	23.37	12970
			Task 2 - Total	89	658.6		10162		17275
Task 3	D. Fowler	5315	BAND 2	1	7.4	41.39	306	70.36	521
	M. Coyle	5315	BAND 7	27	199.8	13.75	2747	23.37	4669
	K. Hargreaves	5315	BAND 5	4	29.6	22.50	666	38.24	1132
			Task 3 - Total	32	236.8		3719		6322

Pricing + Expected Income + Project Approval + Notes

B/f Totals	Year 1	Year 2	TOTAL
STAFF RAW COSTS	21071	21913	**42984**
NON-LABOUR EXPENDITURE	18962	5009	**23972**
SERVICES AND FACILITIES	1500	1500	**3000**
STAFF OVERHEADS (135%)	28445	29583	**58028**
FULL ECONOMIC COST (FEC)	69978	58005	**127984**

PRICING

				Year 1	Year 2	TOTAL
1	**CUSTOMER/AWARD OVERHEADS**	**70**	**%**	**14749**	**15339**	**30089**
	COST TO CUSTOMER/AWARD			**56282**	**43762**	**100044**
2	**CUSTOMER CONTRIBUTION ***	**100**	**%**	**56282**	**43762**	**100044**
	SB CONTRIBUTION TO PROJECT	**22**	**%**	**13696**	**14244**	**27940**

Summary Charge Costs	Task 1	Task 2	Task 3	TOTAL
Year 1: 2001/02	15027	19793	21462	**56282**
Year 2: 2002/03	14952	20231	8578	**43762**
TOTALS by TASK	**29980**	**40025**	**30040**	**100044**

Appendix III. Output from the project

Journal articles

Van Oijen, M., Dreccer, M.F., Firsching, K.-H. & Schnieders, B.J. (submitted). Simple equations for dynamic models of the effects of CO_2 and O_3 on light-use efficiency and growth of crops. Submitted to Ecological Modelling.

Büker/Coyle/Hayes/Mills et al (in preparation). Deposition of ozone onto grass plants in controlled conditions and in the field. [*Primarily based on chapters 3 and 4 in this report*]

Van Oijen, M., Büker, P., Coyle, M., Fowler, D., Hargreaves, K., Hayes, F., Levy, P.E., Mills, G., & Murray, M.. (in preparation). Modelling ozone fluxes and damage in grassland: from controlled environment experiments to the field scale. [*Primarily based on chapters 3 and 4 in this report*]

M. Coyle, L.D Emberson, P. Levy, M. Murray, E Nemitz, M.R Ashmore and D Fowler. Measurements of conductance, at leaf and field scales, and results from a regional dry deposition model. Submitted to Atmospheric Environment

Conference proceedings and abstracts

Coyle, M., Fowler, D., Hargreaves, K., Hayes, F., Levy, P.E., Mills, G., Murray, M. & van Oijen, M. (2003). The CEH O3GRASSLAND Project: Flux measurements at the field scale. CAPER - Natural Environment Research Council Committee on Air Pollution Effects Research. Manchester 14-16 April 2003.

Hayes, F., Büker, P. & Mills, G. (2003). The CEH O3GRASSLAND Project: Impacts of ozone on grass:clover mixtures. CAPER - Natural Environment Research Council Committee on Air Pollution Effects Research. Manchester 14-16 April 2003.

Van Oijen, M., Coyle, M., Fowler, D., Hargreaves, K., Hayes, F., Levy, P.E., Mills, G., & Murray, M.. (2003). The CEH O3GRASSLAND Project: Modelling ozone fluxes & damage. CAPER - Natural Environment Research Council Committee on Air Pollution Effects Research. Manchester 14-16 April 2003.

Mhairi Coyle, Ron Smith and David FowlerOzone Deposition and Stomatal Uptake Over Grassland in Central Scotland; ICP-Vegetation Workshop Establishing Critical Levels II, Gothenburg, Sweden. Nov. 2002.