

Solar UV-B radiation affects below-ground parameters in a fen ecosystem in Tierra del Fuego, Argentina: implications of stratospheric ozone depletion

JOHANN G. ZALLER*§, MARTYN M. CALDWELL*, STEPHAN D. FLINT*, ANA L. SCOPEL‡, OSVALDO E. SALO‡ and CARLOS L. BALLARÉ‡

*The Ecology Center, Utah State University, 5205 Old Main Hill, Logan, Utah 84322–5205, USA., ‡ IFEVA, Consejo Nacional de Investigaciones Científicas y Técnicas, and Universidad de Buenos Aires, Avenida. San Martín 4453, 1417 Buenos Aires, Argentina

Abstract

Stratospheric ozone depletion caused by the release of chlorofluorocarbons is most pronounced at high latitudes, especially in the Southern Hemisphere (including the so-called 'ozone hole'). The consequent increase in solar ultraviolet-B radiation (UV-B, 280–315 nm) reaching the earth's surface may cause a variety of alterations in terrestrial ecosystems. Most effects might be expected to occur above-ground since sunlight does not penetrate effectively below-ground. Here, we demonstrate that solar UV-B radiation in a fen in Tierra del Fuego (Argentina), where the ozone hole passes overhead several times during the Austral spring, is causing large changes of below-ground processes of this ecosystem. During the third and fourth year of a manipulative field experiment, we investigated root systems in these plots and found that when the ambient solar UV-B radiation was substantially reduced, there was a 30% increase in summer root length production and as much as a threefold decrease in already low symbiotic mycorrhizal colonization frequency of the roots compared with plots receiving near-ambient solar UV-B. There was also an apparent shift toward older age classes of roots under reduced solar UV-B. Such large changes in root system behaviour may have decided effects on competition and other ecological interactions in this ecosystem.

Key words: mycorrhizal symbiosis, root growth, Southern Hemisphere, ultraviolet-B radiation

Received 6 November 2001; revised version received 26 February 2002 and accepted 12 March 2002

Introduction

The Antarctic ozone hole and the associated general erosion of the ozone layer at high latitudes in the Southern Hemisphere is a dramatic alteration of the atmosphere (Shindell *et al.*, 1998) and has produced measurable increases in ground-level solar UV-B even at southern mid-latitudes (McKenzie *et al.*, 1999). While the ecological consequences are often poorly understood, UV-B-induced changes in vegetation may mediate other ecosystem changes (e.g., Caldwell *et al.*, 1999; Ballaré *et al.*, 2001). A meta-analysis of c. 100 field studies using elevated UV-B from lamp systems indicated that a consistent effect of supplemental UV-B radiation is an increase

of UV-B-absorbing compounds (phenolics and related compounds) in plant tissues but reductions in above-ground biomass production are also not uncommon (Searles *et al.*, 2001a). From individual studies, other reported changes with implications for ecosystem function include altered litter decomposition rates (e.g., Gehrke *et al.*, 1995; Cybulski *et al.*, 2000; Newsham, 2001), foliage herbivory (e.g., Rousseaux *et al.*, 1998; Rousseaux *et al.*, 2001; Zaller *et al.*, 2002) and plant–microbe interactions (e.g., Paul *et al.*, 1997; Searles *et al.*, 1999; Newsham, 2001; Searles *et al.*, 2001b). Since solar radiation does not penetrate appreciably into soils (Bliss & Smith, 1985), little attention has been paid to changes in below-ground processes (but see Klironomos & Allen, 1995; van de Staaij *et al.*, 2001). Yet, a significant proportion of the primary production is allocated to the root systems in most terrestrial ecosystems and below-ground interactions play an influential role in nutrient and water

Correspondence: Johann G Zaller, Institute of Organic Agriculture, University of Bonn, Katzenburgweg 3, D-53115 Bonn, Germany. fax + 49 (228) 73 56 17, e-mail: jzaller@uni-bonn.de

fluxes and carbon sequestration in ecosystems (Fitter *et al.*, 1985; Jackson *et al.*, 1996). Roots are a major source of energy for soil heterotrophic communities and ultimately fuel the soil ecosystem (e.g., Coleman, 1985).

Here we present results from a well replicated field experiment conducted in a sedge fen ecosystem in Tierra del Fuego (Argentina) where we selectively manipulated ambient solar UV-B radiation over a four-year period. Apart from the Antarctic continent, this region experiences more ozone reduction than any other terrestrial ecosystem. In addition to the passage of the Antarctic vortex containing the 'ozone hole' directly over the area (Diaz *et al.*, 2001), there is a general erosion of the ozone layer that persists into the late spring and summer, long after the Antarctic vortex has dissipated (Rousseaux *et al.*, 2001). The central aim of our study was to determine the effect of UV-B radiation on the growth, depth distribution, apparent age and the mycorrhizal colonization of roots in this fen ecosystem.

Materials and methods

The study site is located in a *Carex* fen near Ushuaia, Tierra del Fuego, Argentina (54°47'S, 68°16'W; elevation about 200 m a.s.l.) where the climate is sub-Antarctic with a mean annual air temperature of 5.5°C and an annual precipitation of 500 mm (FAO, 1985). The fen occurs in peat soil (pH = 6.0, N = 1.7 mg/g, C/N ratio = 19.7) with a water table about 5–10 cm below the soil surface. Experimental plots (1.4 × 1.5 m) are interspersed among scattered saplings of Southern Beech trees (*Nothofagus antarctica*) and are dominated by the sedges *Carex curta* and *C. decidua* (plant nomenclature follows Moore, 1983).

The experiment was first established in October, 1996, with two types of plastic filters stretched taut horizontally over 18 plots to selectively manipulate the solar UV-B transmission. All filters were uniformly perforated with louvers to allow precipitation as well as a fraction of the unfiltered solar radiation to penetrate to the plots below (Searles *et al.*, 1999). Nine plots were under clear 38 µm thick Aclar (type 22 A, Honeywell, formerly Allied Signal, Pottsville, Pennsylvania, USA) to provide a near-ambient UV-B treatment. This material transmits 90% of the solar UV and visible wavelengths (Robberecht & Caldwell, 1986; Searles *et al.*, 1999). The other nine plots were under 100 µm thick extra-clear polyester (optical equivalent of DuPont Mylar, Wilmington, Delaware, USA). This provided a 'reduced-UV-B' treatment, with 10–20% transmittance at most UV-B wavelengths, a rapid rise in transmittance at 320 nm with high transmittance in the UV-A and 90% transmittance at the visible wavelengths (similar visible transmittance to the Aclar, Searles *et al.*, unpublished). These treatments were maintained during the growing seasons of four consecutive

years following snowmelt in early October until plant senescence began in mid-March. These plots received the same treatments each year of the study and these studies are continuing. Occasionally filters would be destroyed by wind, but these were always promptly replaced.

In the third and fourth year of treatment, we used root ingrowth cores (Böhm, 1979) constructed out of plastic mesh to measure root production (1 mm plastic mesh, 10 cm long, 5.5 cm diameter). These ingrowth cores were filled with local root-free peat soil. We inserted four randomly distributed ingrowth cores in each plot in early October, removed two in late December (i.e., spring sampling period) and replaced those with new root-free ingrowth cores. In March of the following year we removed all four cores and replaced all of them with a new set of ingrowth cores for the following season (i.e., summer sample period and entire year period, respectively). After removal of the cores, cores were cut in half to differentiate between root activity at depths of 0–5 cm and 5–10 cm. All apparently living roots of the cores were washed free of soil by using a uniform amount of water over the same time span for each core. Roots were preserved in 70% ethanol until further analysis. Root length was measured with a root length scanner (Commonwealth Aircraft Corporation Ltd., Melbourne, Australia). After the root length measurements, vesicular-arbuscular mycorrhizal colonization was determined by acid fuchsin staining (Kormanik & McGraw, 1982). We determined root length colonization (e.g., length where vesicles and/or arbuscules were present) and its relative intensity (three-level scale from scattered to solidly colonized) under a dissecting microscope using a mm-grid underneath the Petri dishes. To estimate the relative apparent age of the roots we took digital photographs of the total root samples per ingrowth core and analysed the images for luminosity and colour quality (pixels per brightness level ranging from 1–255 for each colour) with Adobe Photoshop software (vers. 4, histogram function, Adobe Inc., USA). Roots generally turn darker with age and as phenolics accumulate (e.g., Eissenstat *et al.*, 2000). Root-growth comparisons between UV-B treatments were based on length and dry mass (dried at 70°C for two days) of fine roots that accumulated in a given period in the ingrowth cores. To assess below-ground biomass, soil cores in the plots were taken to a depth of 10 cm and the roots were subsequently washed, dried and weighed. Aboveground plant biomass was assessed by clipping all shoot material in March at 3 cm above soil surface, with subsequent drying and weighing. All statistical analyses were conducted using a nested ANOVA model (cores nested within plot, soil depth nested within core) with the two UV-B treatments as independent variables.

Results and discussion

Root length production in the top 5 cm of the soil was 30% lower over the summer months under near-ambient UV-B radiation than under reduced UV-B (Figs. 1a,b). This was consistent for the two years when the root systems were investigated. Root length production in the soil layer between 5 and 10 cm was apparently unaffected by the UV-B radiation manipulations (Figs. 1a,b). Specific root length, an indirect measure of root thickness, remained unaffected by the UV-B treatments in both depths and years, however, was significantly greater in the second year (across depths and UV-B treatments 87.04 ± 20.51 vs. 228.32 ± 37.93 m/g for 1998/1999 vs. 1999/2000, respectively; $P < 0.05$). Additionally, we observed that roots that had been grown under near-ambient UV-B were consistently more colonized with

symbiotic mycorrhizal fungi than roots under reduced UV-B radiation (Figs 1c,d). Intensity of mycorrhizal colonization remained unaffected by UV-B radiation (data not shown). However, the mycorrhizal colonization rates we measured were very low and may be due to the very moist conditions of this ecosystem and the dominance of sedges which have generally been considered nonmycorrhizal (but see Miller *et al.*, 1999). Thus, changes in mycorrhizal colonization due to manipulation of solar UV-B are unlikely to have substantial impact on plant performance in this system, however, should be tested in root systems with greater general colonization frequencies. It also appeared that the UV-B treatments affected longevity and/or chemical composition of roots in the 0–5 cm layer since the roots were significantly lighter in colour at near-ambient UV-B (Fig. 1e). Despite this considerable reduction in root length production in the upper soil layer for these two years under near-ambient solar UV-B, this was not yet reflected in total root biomass at the end of the growing season in these two years (e.g., for 1999/2000, 0–5 cm soil layer: 107.4 ± 12.2 vs. 111.8 ± 14.6 g roots/m² for reduced vs. near-ambient UV-B, respectively) or above-ground biomass production (e.g., for 1999/2000, 137.6 ± 10.5 vs. 161.4 ± 24.0 g/m² for reduced vs. near-ambient UV-B, respectively).

It was surprising to see such considerable changes in the root systems of this vegetation due to solar UV-B manipulations. There are a few studies that have reported UV-B-induced changes in root : shoot ratios (Caldwell, 1997) or a tendency for reduced mycorrhizal colonization of isolated plants (Klironomos & Allen, 1995; van de Staaij *et al.*, 2001), however, we are unaware of any reports of changes in root systems due to UV-B treatments in field studies of intact ecosystems. At this time, we lack a clear understanding of the processes causing the observed UV-B-induced changes in the root system. Above-ground, UV-B has been shown to influence growth allocation. For example, typically, stems might be shorter and leaves smaller, but more stems and leaves are commonly produced under elevated UV-B radiation (e.g., Barnes *et al.*, 1990). Thus, it might not be surprising that changes in UV-B radiation might be reflected in altered root growth. Also, since UV-B has been shown to rather consistently increase phenolic concentrations in leaf tissues (Searles *et al.*, 2001a) changes in phenolic allocation from roots to leaves might be playing a role in root development, mycorrhizal colonization and their apparent ageing (colour changes). Given the decisive role that root systems play in competitive balance of species, fluxes of water and mineral nutrients and sequestration of carbon in ecosystems (Fitter *et al.*, 1985; Jackson *et al.*, 1996), the implications of our findings can be appreciated. Solar UV-B changes due to stratospheric ozone depletion are expected to be greater in Tierra del Fuego

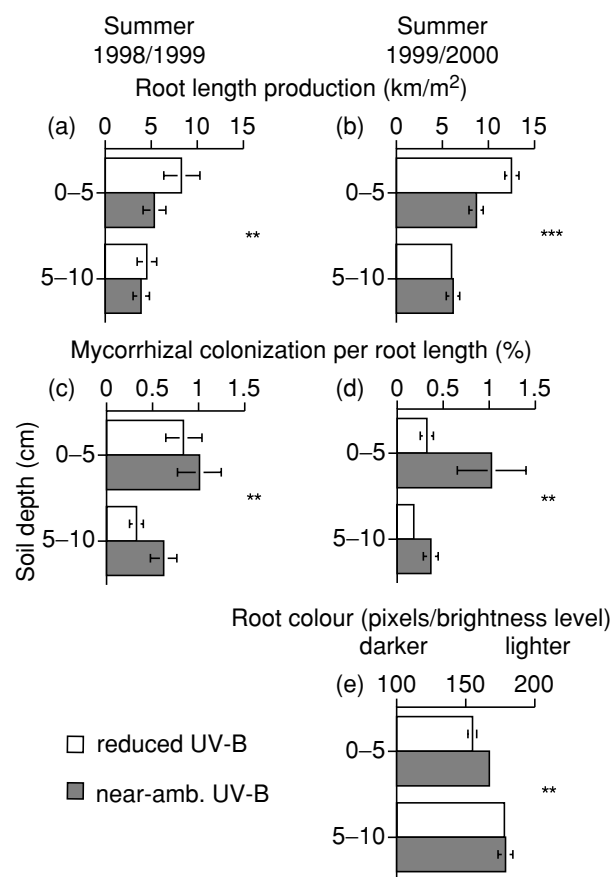


Fig. 1 Root length production (a, b), mycorrhizal colonization (c, d) and root colour (e) in experimental plots of a *Carex fen* ecosystem in Tierra del Fuego (Argentina) maintained at either reduced or near-ambient UV-B radiation. Bars represent means \pm SE ($n = 9$). Significance of results derived from nested ANOVAs among the two soil layers: *** $P < .01$, ** $P < 0.05$. No interactive effects between UV-B and year or soil depth occurred.

than in most locations outside of Antarctica, however, these sizeable changes in the root systems provide impetus for more general attention to the effects of solar UV-B changes on the below-ground component of ecosystems.

Acknowledgements

We are grateful to Oscar Bianciotto who allowed us to conduct our experiments on his property, Eduardo Olivero and his team at the Centro Austral de Investigaciones Científicas (CADIC), Ushuaia, Argentina, and especially Susana Díaz for providing logistical support. Cecilia Rousseaux and Peter Searles maintained the experiment during the first years and provided critical review of an earlier version of the manuscript. Nancy Lozano, Nicolas Garibaldi, Mayme Seng-Ercanbrak, Usha Menon-Spaulding, H.S. Satyanarayana, and Amy Kaplan-Stevenson provided technical assistance. Thanks also to Bradley Kropp, Dane Hansen, Eugene Schupp and Dani Or for their help and use of laboratory facilities and Susan Durham for statistical advice. This research was supported by the NSF/DOE/NASA/USDA/EPA/NOAA Interagency Program on Terrestrial Ecology and Global Change (TECO) by the United States National Science Foundation (IBN-9524144 and IBN-9814357).

References

- Ballaré CL, Rousseaux MC, Searles PS *et al.* (2001) Impacts of solar ultraviolet-B radiation on terrestrial ecosystems of Tierra del Fuego (southern Argentina). An overview of recent progress. *Journal of Photochemistry and Photobiology B-Biology*, **62**, 67–77.
- Barnes PW, Flint SD, Caldwell MM (1990) Morphological responses of crop and weed species of different growth forms to ultraviolet-B radiation. *American Journal of Botany*, **77**, 1354–1360.
- Bliss D, Smith H (1985) Penetration of light into soil and its role in the control of seed germination. *Plant, Cell and Environment*, **8**, 475–483.
- Böhm W (1979) *Methods of studying root Systems*. Springer-Verlag, Heidelberg.
- Caldwell MM (1997) Alterations in competitive balance. In: *Plants and UV-B: Responses to Environmental Change* (ed. P Lumsden), pp. 305–315. Cambridge University Press, Cambridge.
- Caldwell MM, Searles PS, Flint SD, Barnes PW (1999) Terrestrial ecosystem responses to solar UV-B radiation mediated by vegetation, microbes and abiotic photochemistry. In: *Physiological Plant Ecology* (eds. Press MC, Schole JD, Barker MG), pp. 241–262. Blackwell, London.
- Coleman DC (1985) Through a ped darkly: an ecological assessment of root-soil microbial-faunal interactions. In: *Ecological Interactions in Soil* (eds Fitter AH, Atkinson D, Read DJ, Usher MB), **Vol. 4**, pp. 1–21. Blackwell, Oxford.
- Cybulski WJ, Peterjohn WT, Sullivan JH (2000) The influence of elevated ultraviolet-B radiation (UV-B) on tissue quality and decomposition of loblolly pine (*Pinus taeda* L.) needles. *Environmental and Experimental Botany*, **44**, 231–241.
- Diaz S, Deferrari G, Booth C *et al.* (2001) Solar irradiances over Ushuaia (54.49 degrees S, 68. 19 degrees W) and San Diego (32.45 degrees N, 117.11 degrees W) geographical and seasonal variation. *Journal of Atmospheric and Solar-Terrestrial Physics*, **63**, 309–320.
- Eissenstat DM, Wells CE, Yanai RD *et al.* (2000) Building roots in a changing environment: implications for root longevity. *New Phytologist*, **147**, 33–42.
- FAO (1985) Agroclimatic data for Latin America and the Caribbean, Rome.
- Fitter AH, Atkinson D, Read DJ *et al.* eds. (1985) *Ecological Interactions in Soil*. Blackwell, Oxford.
- Gehrke C, Johanson U, Callaghan TV *et al.* (1995) The impact of enhanced ultraviolet-B radiation on litter quality and decomposition processes in *Vaccinium* leaves from the Subarctic. *Oikos*, **72**, 213–222.
- Jackson RB, Canadell J, Ehleringer JR *et al.* (1996) A global analysis of root distributions for terrestrial biomes. *Oecologia*, **108**, 389–411.
- Klironomos JN, Allen MF (1995) UV-B-mediated changes on below-ground communities associated with the roots of *Acer saccharum*. *Functional Ecology*, **9**, 923–930.
- Kormanik PP, McGraw A-C (1982) Quantification of vesicular-arbuscular mycorrhizae in plant roots. In: *Methods and Principles of Mycorrhizal Research* (ed. Schenck NC), American Phytopathological Society, St. Paul, Minnesota, USA.
- McKenzie R, Connor B, Bodeker G (1999) Increased summertime UV radiation in New Zealand in response to ozone loss. *Science*, **285**, 1709–1711.
- Miller RM, Smith CI, Jastrow JD *et al.* (1999) Mycorrhizal status of the genus *Carex* (Cyperaceae). *American Journal of Botany*, **86**, 547–553.
- Moore DM (1983) *Flora of Tierra del Fuego*. Anthony Nelson, Shropshire, England.
- Newsham KK (2001) UV-B effect on *Quercus robur* leaf litter decomposition persists over four years. *Global Change Biology*, **7**, 479–483.
- Paul ND, Rasanayagam S, Moody SA *et al.* (1997) The role of interactions between trophic levels in determining the effects of UV-B on terrestrial ecosystems. *Plant Ecology*, **128**, 296–308.
- Robberecht R, Caldwell MM (1986) Leaf UV optical properties of *Rumex patientia* L. & *Rumex obtusifolius* L. in regard to protective mechanism against solar UV-B radiation injury. In: *Stratospheric Ozone Reduction, Solar Ultraviolet Radiation and Plant Life* (eds Worrest RC, Caldwell MM), Vol. G8, pp. 251–259. Springer-Verlag, Berlin.
- Rousseaux MC, Ballaré CL, Scopel AL *et al.* (1998) Solar ultraviolet-B radiation affects plant-insect interactions in a natural ecosystem of Tierra del Fuego (southern Argentina). *Oecologia*, **116**, 528–535.
- Rousseaux MC, Scopel AL, Searles PS *et al.* (2001) Responses to solar ultraviolet-B radiation in a shrub-dominated natural ecosystem of Tierra del Fuego (Southern Argentina). *Global Change Biology*, **7**, 467–478.
- Searles PS, Flint SD, Caldwell MM (2001a) A meta-analysis of plant field studies simulating stratospheric ozone depletion. *Oecologia*, **127**, 1–10.
- Searles PS, Flint SD, Diaz S *et al.* (1999) Solar ultraviolet-B radiation influence on *Sphagnum* bog and *Carex* fen ecosystems:

- first field season findings in Tierra del Fuego Argentina. *Global Change Biology*, **5**, 225–234.
- Searles PS, Kropp BR, Flint SD *et al.* (2001b) Influence of solar UV-B radiation on peatland microbial communities of southern Argentina. *New Phytologist*, **152**, 213–221.
- Shindell DT, Rind D, Lonergan P (1998) Increased polar stratospheric ozone losses and delayed eventual recovery owing to increasing greenhouse-gas concentrations. *Nature*, **392**, 589–592.
- van de Staaij J, Rozema J, van Beem A *et al.* (2001) Increased solar UV-B radiation may reduce infection by arbuscular mycorrhizal fungi (AMF) in dune grassland plants: evidence from five years of field exposure. *Plant Ecology*, **154**, 171–177.
- Zaller JG, Searles PS, Rousseaux MC *et al.* (2002) Solar ultraviolet-B radiation can affect slug feeding preference for some plant species native to a fen ecosystem in Tierra del Fuego, Argentina. *Plant Ecology*, in press.