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The effect of natural UV-B radiation on a perennial *Salicornia* salt-marsh in Bahía San Sebastián, Tierra del Fuego, Argentina: a 3-year field study

O.A. Bianciotto^{a,*}, L.B. Pinedo^a, N.A. San Roman^a, A.Y. Blessio^a, M.B. Collantes^b^aCentro Austral de Investigaciones Científicas (CONICET), CC. 92 (9410) Ushuaia, Tierra del Fuego, Argentina^bCEFYO (CONICET), Serrano 668, Buenos Aires, Argentina

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Abstract

The Antarctic ozone hole and a general depletion of the stratospheric ozone layer cause increased levels of ultraviolet-B solar radiation (UV-B) over Tierra del Fuego, the southernmost tip of South America. For three consecutive growing seasons (1997–2000), we studied the biological impacts (morphology, physiology, demography and phenology) of natural UV-B radiation on a perennial *Salicornia ambigua* Michx. community in San Sebastian Bay (53° S and 68° W), Tierra del Fuego, Argentina. This is the first UV-B screening experiment on a subantarctic halophytic community. The shortwave UV-B spectrum (280 to 320 nm) was excluded by covering plots with UV-B blocking film (Mylar). These plots were compared to controls covered with UV-B transparent (Aclar) plastic screens, and unshielded plots. Shoot length in *Salicornia* was not affected by UV-B. Exposure to natural UV-B reduced biomass and density (by 17% and 38%, respectively). Concentration of UV-shielding pigments and cuticle thickness were both significantly higher (25–48% and 21–40%, respectively) in plants receiving ambient UV-B. The increase in cuticle thickness persisted throughout the growing season, whereas pigment concentration was higher at the beginning of the growing season. Also, the number of dead shoots was higher in plants exposed to UV-B. At the end of the growing season (March) shoot mortality was higher in plants exposed to ambient UV-B, and post-flowering senescence was 30 days earlier. Slight changes in the relative composition of *Salicornia* to *Puccinellia* were seen. The reduction observed in *Salicornia* shoot density under ambient UV-B was cumulative over time; 23% in the first growing-season, rising to 38% by the third growing-season. A similar incremental increase in pigment absorption at 305 nm was seen; 25% in the first and 48% in the third growing season.

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1. Introduction

Studies of the biological effects of natural ultraviolet-B radiation (UV-B; 280–320 nm) have proliferated as a consequence of the springtime Antarctic ‘ozone hole’, first reported in the 1980s [1]. During the 1990s, the surface area of the ozone hole reached 24 million km² [2]. In September of 1988 NASA reported its expansion to 27.3 million km² and a minimum ozone column thickness of 90 DU (Dobson Units) over the Antarctic. During 2 weeks in 1994 and November–December 1995 ozone-depleted air from the hole, extended over South America to about 38° S [3].

Based upon TOMS (Total Ozone Mapping Spectrom-

ter) data for 1978–1999, a downward trend occurred in ozone column thickness over Ushuaia, in the spring and summer of these years [4]. This led to an increase in UV-B radiation [5], as confirmed by the significant inverse correlation between ozone column and ground level biologically effective UV doses, at this time [4,6,7].

The effects of UV-B radiation on biological systems have been evaluated by the UN Environment Programme [8]. Many studies have shown deleterious effects of natural UV-B radiation on plants via reduced photosynthesis and biomass production [9–11]. Changes in the size of leaves, plant morphology and primary productivity are common effects of UV-B radiation on plants under field conditions [12,13]. Although environmentally relevant UV-B doses in the field may have small effects on photosynthetic processes, they may alter productivity through changes in canopy structure and hence light interception (e.g. leaf expansion and plant morphology) [14]. Stem elongation in

*Corresponding author. Tel.: +54-2901-422-310; fax: +54-2901-430-644.

E-mail address: o_bianciotto@gmx.net (O.A. Bianciotto).

plants is influenced by the auxin indole acetic acid, which absorbs in the UV-B range and can be photodegraded by high UV-B radiation doses [15]. However, plants can protect their tissues from harmful radiation by accumulating UV-B protective pigments in the epidermal layer of their leaves [16] and by repairing damaged DNA [6,10]. The leaf epidermis is thought to play a major role in attenuating UV-B radiation, and in diminishing the potential of UV-B to damage the mesophyll. Flavonoids, and other phenolic compounds, located in the epidermal cells' vacuole, absorb UV-B [17]. Experiments into flavonoid formation in *Pinus sylvestris* L. seedlings have shown that diacylated flavonol glycosides are accumulated in the needles as a response to supplemental UV-B treatment [18].

The comparatively small solar angle of incidence, and the thicker ozone layer found towards the poles, provide conditions of less solar radiation than are seen at lower latitudes. When the ozone hole appeared, these conditions changed [19]. Robberecht et al. [20] compared the penetration of UV through the epidermis of Alaskan plants with low latitude plants of Hawaii and the Andean Mountains. They observed greater transmittance of the radiation through the epidermis in Arctic plants to be attributable to their lower concentration of protective pigments.

Laboratory studies are very important in defining biological responses to UV-B. However, exposure to artificial radiation sources cannot replicate changes in natural solar irradiance, and the time scale over which they occur. According to Caldwell and Flint [21], only 15% of the work carried out on plant responses to UV-B give radiation doses similar to those observed under field conditions.

Long-term field studies into the effects of UV-B radiation on vascular plants on the coast of the Beagle Channel (Subantarctic Region) commenced in 1997. Ambient UV-B is known to have a significant inhibitory effect on the growth of herbaceous and graminoid species of this region. In *Gunnera magellanica*, the inhibitory effect is accompanied by increased level of DNA damage in leaf tissue. The density of DNA dimers in the early spring is correlated with the dose of natural UV-B measured at ground level [6]. In Antarctica, increases of 32% in *Deschampsia antarctica* leaf length, and 55% in leaf number, in response to UV-B exclusion were reported [23]. Seedlings of *Colobantus quitensis* and *D. antarctica* exposed to natural UV-B radiation showed less biomass production, fewer leaves, and reduced leaf area compared to plants growing under attenuated UV-B radiation [24,25]. San Roman et al. [27] found significantly lower pigment concentration and increased shoot density in *Salicornia* sp. growing under attenuated UV-B radiation on a salt marsh in Tierra del Fuego.

This investigation focuses on the ubiquitous salt marsh species *Salicornia ambigua*, the dominant species in a community also containing *Puccinellia* sp. (nearly 1% coverage). This is the first long-term field experiment

involving UV-exclusion to evaluate the impact of natural UV-B radiation on the marine coastal ecosystems of Tierra del Fuego. These ecosystems are extremely important because of their high productivity, their capacity to stabilise the sediment, providing fisheries habitat and playing a major role in the filtration of coastal nutrients [28,29]. This work aims: (a) to analyse the effects of natural UV-B radiation on shoot growth and density, biomass production and the epidermal screening properties in plants of *S. ambigua* in a long-term field experiment (3 years); (b) to assess population and phenological changes at the community level and (c) to evaluate the impact of temporal environmental UV-B variability on a Tierra del Fuego salt marsh.

2. Materials and methods

2.1. Experimental site and ozone trends

The field experiment was conducted in San Sebastian Bay located in the Northeast of Tierra del Fuego, Argentina (53° 27' 52" S, 68° 4' 26" W) 230 km north from the city of Ushuaia. It is a semicircular bay 55 km long and 40 km wide. The local climate is maritime. In the coldest month (July), mean temperature is about 0 °C, while in the warmest (January), it reaches 10 °C. Annual rainfall is about 300 mm. There are frequent and strong westerly winds (ca. 200 days per year), which attain mean speeds of 60 km/h, with maximum of 150 km/h in spring and summer [30]. These winds cause high evaporation rates and increase tidal level.

Salt marsh communities in Tierra del Fuego are part of a Fuegian Steppe, and it is possible to describe three types [31,32]: saline lagoon grassland, with vegetation dominated by *Hordeum lechlerii*, *Puccinellia* spp., *S. ambigua* and *Lepidophyllum cupressiforme* scrub (Mata Verde), covering high borders of lagoons (albardones) or over the crest of cheniers. Pastures of *Puccinellia* sp. and *S. ambigua*, with salinity around 7–10 g/l, over ancient tidal plains. Carpets of *S. ambigua* with a presence of scattered graminoid *Puccinellia* sp., growing on intertidal plains nearly from the outlet of the San Martin River, with a salinity of 18–35 g/l, the site of the present study.

S. ambigua is a creeping perennial herb with much-branched jointed succulent stems, somewhat woody at the base [33]. New shoots sprout in spring (October), and in autumn (May) some lignify while others senesce. Plant height is around 7–10 cm, and lateral branches reach around 1–3 cm.

To assess the influence of ozone depletion on San Sebastian Bay, ozone data for Rio Grande (80 km south from San Sebastian Bay) were obtained from the NASA Total Ozone Mapping Spectrometer (TOMS) (<http://jwocky.gsfc.nasa.gov/TOMSmain>). The UV-B data were collected from an IL 1700 research radiometer (Internation-

tional Light, Newburyport, MD, USA) also located in Rio Grande. To calibrate the IL 1700 radiometer, measured UV-B data were compared with solar UV-B radiation estimated from the UVR channels of a spectroradiometer (GUV-511, Biospherical Instruments). This estimation of UV-B (280–320 nm) was calculated using a multiple linear regression from 305 nm and 320 nm GUV-511 data [3]:

$$\text{UV-B} = 59.5 \times E_{305} + 4.1 \times E_{320}.$$

Since we had not 24 h data, the UV-B doses were calculated by integrating the data taken from 11:30 to 15:30 h because the solar noon at Rio Grande is at 13:30 h. Peaks in UV-B doses (between 11:30 and 15:30 h) generally corresponded to days when the ozone column was the thinnest (under 300 DU) (Fig. 1a and b). The scattered UV-B peaks were the highest during late spring (December) and the lowest during early summer (January). For the same time period, similar correlations between depletion of the ozone column and increased daily UV-B doses have been reported in Ushuaia and close to Punta Arenas (Chile) by other authors [4,35,36]. It was not

possible to weigh biologically relevant UV-B doses because of the instrument used for the experiment. The IL 1700 radiometer used only records the overall UV-B dose range, but does not record UV-B doses at specific wavelengths.

2.2. Experimental set-up and treatments

A 50×30 m mapped area (topography and vegetation distribution) was enclosed to exclude the grazing cattle. A complete randomised blocked design with three replicate plots of 1.2×1.2 m was used for each treatment. To create an attenuated UV-B treatment, plots were covered by a clear polyester film (175 μm thick, optically equivalent to 'Mylar-D', Dupont); to create a near-ambient UV-B treatment plots were covered by thick 'Aclar' plastic film; and three plots were left uncovered to provide a comparison with ambient UV-B radiation under unmanipulated conditions. In the centre of each plot, small sub-plots (0.25×0.25 m) were set-up and marked for non-destructive measurements only (shoot length and density). The remaining area inside the plot was considered a buffer zone, where destructive *Salicornia* samples (for pigment extraction, cuticle thickness and biomass) could be taken.

The microclimate under these treatments and adjacent open areas was monitored during the three growing seasons. The spectral climate under the films was measured with an IL 1700 research radiometer. The reduced UV-B treatment (Mylar) allowed ~32% transmission of the daily ambient UV-B to reach the plant canopy under the frames, while near-ambient UV-B treatment allowed 87% of the daily ambient UV-B to reach the plant canopy.

Air temperature and wind speed were recorded using an electronic thermometer (model Brand) and a digital wind meter (Kestrel, Ben Meadows, USA) every 21 days during the growing season—October to April (between 14:00 and 16:00 h)—to coincide with our measurements. Ten measurements were made outside the screens, as well as under each screen. Samples for soil humidity and soil conductivity measurements were collected from outside the plots' area. These were collected three times during the growing season (October, December and March), and at the end of the experience in all of the plots, and were analysed at Consejo Agrario Provincial Soil Laboratory, Rio Gallegos, Argentina. Outside the exclusion area the soil conductivity range was between 43 and 21 mS/cm. Soil conductivity beneath the filters did not differ from that outside the filtered areas ($P=0.135$). The general soil humidity changed between 11 and 30%. We did not find soil humidity differences between filtered plots and outside ($P=0.065$). Hence, no data are shown. Air temperature at plant canopy height (5 cm) averaged 0.2–0.4 °C higher under both filter treatments than in the open marsh. Similarly, average wind speed was 30% lower under the filters compared with open areas.

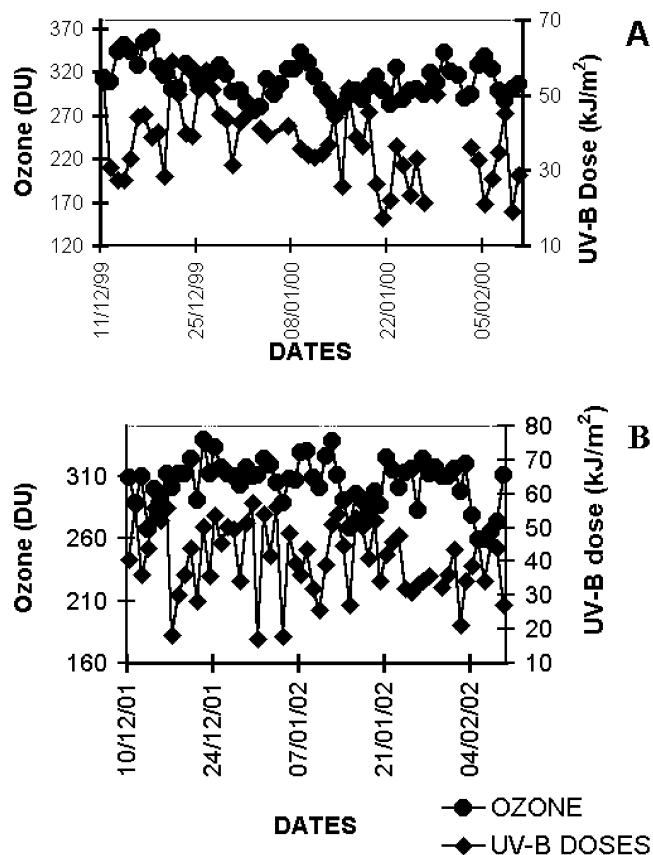


Fig. 1. Ozone column and UV-B ground level solar radiation at Rio Grande. The ozone column data are from NASA TOMS. The UV-B doses were calculated by integrating data from 11:30 to 15:30 h (solar noon at Rio Grande: 13:30 h). Missing data correspond to days not recorded by IL research radiometer. Notice when ozone column is lower than 305 DU, UV-B dose peaks occur.

2.3. Shoot length, density and phenology

Plants within each plot were cut to ~5 cm above the ground at the beginning of the experiment to avoid apical dominance. All plants emerging within the 0.25×0.25 central squares were counted to determine density of shoots every 21 days during the growing season, since flowering was discriminated between live and senescent (dead) shoots. Because of a high density of annual shoots (6400 to 19,200/m²), central sub-plots were divided into 16 equal subplots to count in the field. Concurrently, phenological observations of sprouting, flowering and senescence were made on this population. Each year, inside these permanent sub-plots, 25 randomly chosen new annually produced shoots were tagged. Length of the main shoot axis was measured at flowering with a calliper (due to the small size of the *Salicornia* sprout).

2.4. Pigment analysis and cuticle thickness

After collection, samples were preserved in a freezer at –18 °C until extraction. Tissue samples were ground with methanol:acetic acid (99:1, v/v). Extracts were centrifuged at 1000 rev./min for 5 min. Then the supernatant was decanted into a quartz cuvette for analysis. Absorbency of methanol extractable UV-B absorbing compounds was measured from 280 to 320 nm in steps of ~5 nm in a double-beam spectrophotometer (Metrolab 1700 UV, Argentina). Another one measurement was done at 665 nm for chlorophyll determination. The results were normalised by weight and solvent volume. Chlorophyll concentration did not differ between samples.

Cuticle thickness was determined every month over the three growing seasons using a light microscope (Carl Zeiss, 40 w, Germany). The shoot portion just below the mid-point was cross-sectioned and cut with a freezing microtome (Reichert 343). The cuticle was stained with Sudan IV to facilitate the identification. Six preparations of each replicate ($n=3$) were done and three readings on random cuts were taken using an ocular micrometer.

2.5. Plant cover and total biomass

At the beginning and at the end of the investigation, plant cover was evaluated inside each permanent plot. Aerial plant cover of annual shoots was assessed and compared between the first and the third year. Pictures of each subplot (0.25×0.25 m) were taken with a camera (Chinon S24) every summer. On these pictures, the *Salicornia* and *Puccinellia* cover percentage area was calculated from each sub-plot. At the end of the third growing season, circular samples were taken, with a large bore, (0.10 m deep and 0.10 m diameter) from each treatment's plot to evaluate total dry matter above and below ground. Dry biomass was determined after keeping the samples for at least 3 days at 70 °C.

2.6. Statistical analysis

The parameters were analyzed using a one-way ANOVA to test the effects of UV-B treatments. Repeated-measures ANOVA was not used to analyze main shoot length, because every year the measurements were taken on different shoots. All statistical tests used a 0.05 level of significance. Unplanned comparisons between means were done using the Scheffé method. The relative reductions in shoot density and the relative increment in absorbance at 305 nm attributable to UV-B were calculated by a regression using an adjusted linear model.

3. Results

3.1. Shoot length, density, phenology and biomass

Main shoot length measured at flowering was significantly greater in plots receiving near-ambient UV-B radiation than those receiving attenuated UV-B and the no filter control, only in the second year of treatments (Table 1).

There were consistent effects on live shoot density. In late summer of the third growing season (2000), the total number of live shoots was greater in plots receiving attenuated UV-B compared to near-ambient UV-B plots (and the no filter control) ($P<0.05$; Fig. 2). In late summer of the second and third growing seasons, UV-B attenuation also reduced the total number of dead (including senescent) shoots ($P<0.01$; Fig. 3).

The growing season for *S. ambigua* was longer under attenuated UV-B than under near-ambient (or the no filter control). Flowering was delayed by ca. 30 days and senescence by 20 days in the attenuated UV-B plots (Fig. 4). Dry mass production was 20% greater in attenuated UV-B plots than in the near-ambient UV-B plots (84% greater than the no filter controls) (Table 1).

3.2. Effects of UV-B on protective sunscreens

Concentrations of methanol extractable UV-B-absorbing compounds were consistently higher (25–48%) in plants grown under near-ambient, compared to attenuated UV-B

Table 1
Effects of UV-B radiation on height and plant dry mass of *S. ambigua*

Treatment	Main shoot length at flowering (mm)			Dry mass (g/m ²)
	2-12-97	8-12-98	6-12-99	
Ambient UV-B	18.37	10.13	9.45	741.87
Near-ambient UV-B	22.48	19.15	9.76	1142.09
Attenuated UV-B	19.98	11.89	7.22	1369.15
	$P=0.190$	$P<0.01$	$P=0.063$	$P=0.058$

Height (mm) at flowering during the 3 experimental years and plant dry mass (g/m²) measured at the end of the experiment. At December 8th 1998, the differences occurred between near-ambient and ambient UV-B, and between near-ambient and attenuated UV-B.

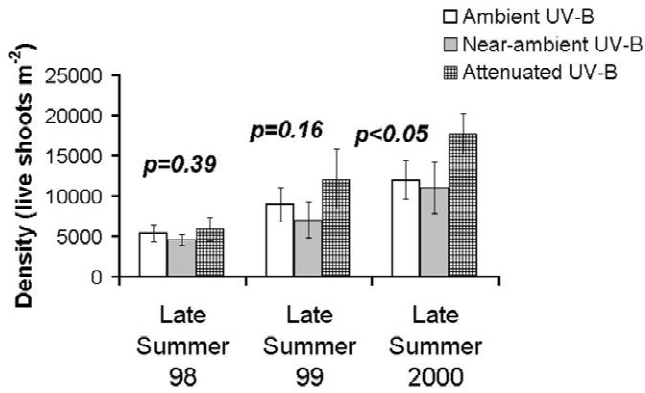


Fig. 2. Shoot density of *S. ambigua* at ambient, under near-ambient and attenuated solar UV-B radiation. Data were analysed by one-way ANOVA, results from which are shown in each figure. The differences were significant between treatments during the third period. There was a significantly greater live shoot number in plots receiving attenuated than under near-ambient UV-B (and the no filter control). $N=3$ (mean \pm 1 S.E.).

($P<0.01$, Fig. 5). Plant growth in the no-filter plots also exhibited increases in UV-B absorbing compounds in late spring and early summer (50–80% above attenuated-UV-B plants, Fig. 5).

Cuticle thickness was consistently increased under near-ambient, compared to attenuated UV-B radiation ($P<0.01$, Fig. 6). The mean (1997–2000) increase in thickness was 20% in spring, and 40% in late summer (Fig. 6).

4. Discussion

The occurrence of similar spring and summer ozone depletion over Rio Grande (Fig. 1) to that seen over the rest of southern South America, was confirmed using TOMS data. Some days of reduced ozone column thick-

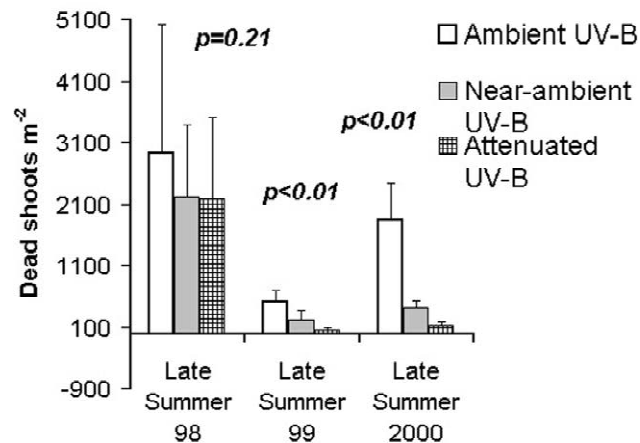


Fig. 3. Dead shoot density of *S. ambigua* with no filter, under near-ambient and attenuated solar UV-B radiation. Data were analysed by one-way ANOVA, results from which are shown in each figure. We found significant differences between attenuated and near-ambient/ambient treatments. $N=3$ (mean \pm 1 S.E.).

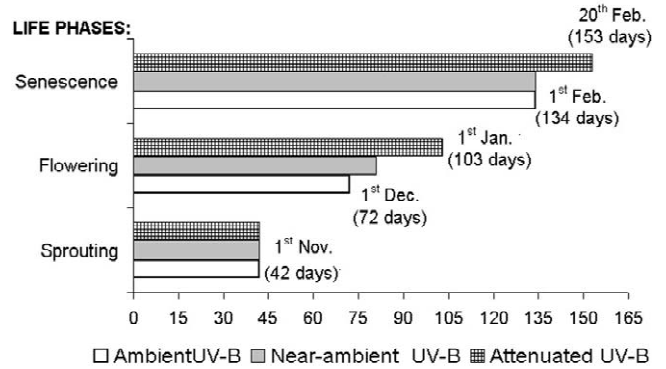


Fig. 4. Phenological changes in *S. ambigua* with no filter, under near-ambient and attenuated UV-B solar radiation (emergence; day 1=21st September, beginning sprouting). Flowering was delayed by ca. 30 days and senescence by 20 days in the attenuated UV-B plots. Final senescence happened between 15 April and 15 May in relation to daily average temperatures below 0 °C and snowing. Days from emergence are averaged from 3 experimental years.

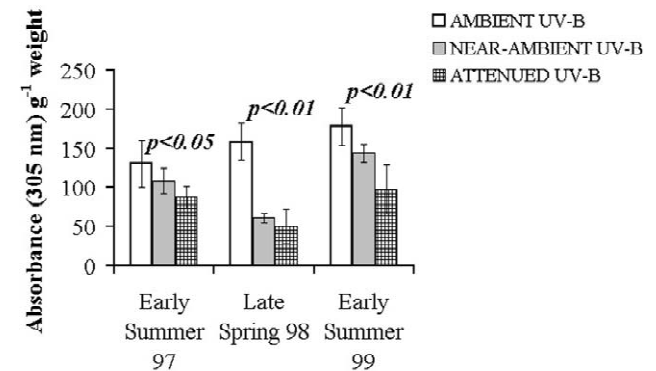


Fig. 5. Effects of UV-B treatment on absorbing compounds extracted from *S. ambigua* during early summer and late spring over the 3 years. The absorbance values at 305 nm were normalised for 1 ml of methanol and by the sample weight. The results from one-way ANOVA show consistent differences between treatments (mean \pm 1 S.E.) ($n=3$).

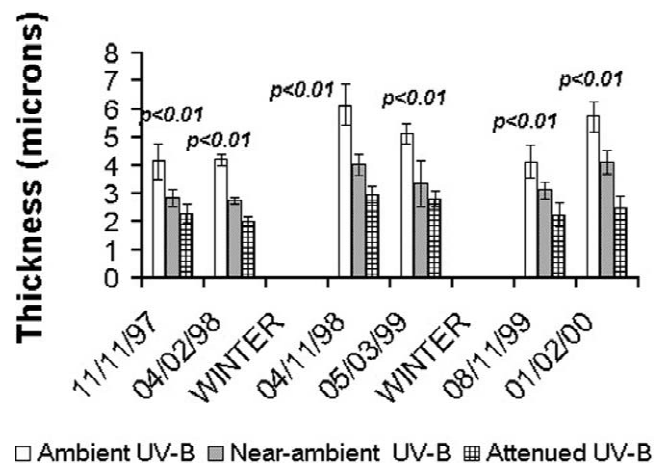


Fig. 6. Effects of solar UV-B on cuticle thickness of *S. ambigua* measured over 3 growing seasons. Data analysed by one-way ANOVA show significant differences during spring and summer with thicker leaves under near-ambient UV-B radiation (mean \pm 1 S.E.) ($n=3$).

ness (<305 DU) were observed during every month of the growing season. Ozone depletion caused a clear increase in ground level UV-B radiation in Rio Grande, which occurred in spite of the frequently heavy cloud cover (Fig. 1).

Organisms may be adapted to the levels of UV-B radiation where they exist. Prior to ozone depletion, levels of UV-B radiation in Tierra del Fuego were low, but the large relative increase now seen may be most relevant [4], given that the magnitude of UV-B fluctuation has been shown to affect the degree of damage caused to the plant. Plants subjected to very low levels of UV-B radiation and subsequently exposed to high doses display UV-B sensitivity [37,38]. Reduction of plant growth rates, shoot-leaf density, plant biomass, and an increase in UV-B-absorbing pigments and cuticle thickness have been observed in response to augmentation of UV-B radiation.

Shoot elongation of *S. ambigua* was not affected by natural solar UV-B (Table 1). In another 3-year exclusion experiment (using filters) along the latitudinal gradient between Bahía San Sebastian, Tierra del Fuego (Argentina, 52° S, 68° W), outlet of Río Chubut (Argentina, 43° S, 64° W), Lagoa dos Patos (Brazil, 32° S, 52° W) and Chesapeake Bay (USA, 38° N, 75° W), Koch and Col [39] found that exposure to solar UV-B did not affect growth parameters (such as leaf/shoots elongation), in salt marsh vegetation (*Spartina*, *Juncus*, and *Salicornia*). The high flavonoid concentration of *Juncus* and *Spartina* leaves may confer protection on these plants. In *S. ambigua*, the concentration of UV-B absorbing pigments is even higher, and cuticle thicker, than in the other two species, thus explaining the lack of response of shoot elongation to solar UV-B. In contrast, Day et al. [40] found that Antarctic solar UV-B radiation led to slower leaf elongation rates and shorter fully expanded leaves of *D. antarctica* and *C. quitensis*, in a long-term study.

Near-ambient solar UV-B reduced the density of *S. ambigua* live shoots (Fig. 2) only after the third growing season of our study. New shoots arise from buds formed during the previous growing season, so differences were not anticipated until after the first year of treatments. Several other plant species in Tierra del Fuego and Antarctica have also shown no difference in vegetative growth after one growing season, followed by subtle responses during the second year of UV-B treatments [25–27,34,50].

In salt marshes at medium and low latitudes, *Juncus roemerianus*–*Spartina alterniflora* in USA, and *J. effusus* in Brazil did not display any change in shoot/leaf density as the result of UV-B exclusion. *Spartina longispica* in Puerto Madryn increased tillering above ambient condition [39], which we saw in *S. ambigua*.

Ruhland and Day [25] observed a reduced growth under near-ambient solar UV-B radiation in two higher plant species of the Antarctic Peninsula; *D. antarctica* and *C. quitensis*. They reported 7–16% fewer leaves and 65–82%

fewer branches, respectively, in seedlings grown under ambient and near-ambient UV-B condition than attenuated UV-B. Xiong and Day [23] also found a reduction of 11–22% in total biomass, and 24–31% of total leaf area.

We can contrast the substrate required by *Salicornia*, against the requirements of *Colobantus* and *Deschampsia*: *Salicornia* requires high salinity and pH>7, whereas *Colobantus* and *Deschampsia* need high acidity (pH<5) arising from a high concentration of aluminium or hydrogen ions in the soil [41,42]. The three genera give similar responses to near-ambient solar UV-B radiation: i.e. thicker leaf-cuticle and increased pigment concentration, reduction in the number of shoots, lower biomass production, fewer leaves and reduced total leaf area, slower leaf elongation rates and shorter fully expanded leaves. We suspect that these similar responses of plants from these three genera could be a consequence of their adaptation to two environmental extremes, although further studies are needed to confirm these preliminary conclusions.

The timing of life phases in plant communities is a combination of response to environmental factors and the genetic constitution of plants. The timing of events such as flowering or senescence of foliage can be altered by UV-B radiation [11,43], and in *S. ambigua* we observed shorter flowering and early senescence under near-ambient solar UV-B (Fig. 4).

UV-B absorbing compounds in the leaf cuticle and epidermis reduce penetration of UV-B radiation into the leaf. Increased UV-B can alter the composition of plant secondary compounds; for example, increasing flavonoids and related phenolic compounds [44,45]. Changes in epidermal UV-B screening compounds, leaf and cuticle thickness potentially provide protection to plants against UV-B damage, and reduce the penetration to underlying tissues [46]. *Salicornia* showed an increase in UV-B absorbing pigments under near-ambient UV-B radiation (Fig. 5). There were consistently higher concentrations of methanol-extractable UV-absorbing compounds in plants under near-ambient UV-B conditions. In some conifers, young leaves attenuate less UV-B than more mature leaves [47]. If also true for *S. ambigua*, this could explain why younger shoots produced in November and February showed the largest treatment effects (Fig. 5, other months not shown). At mid and high latitudes (Puerto Madryn and Tierra del Fuego) UV-absorbing pigment concentrations were highest in *S. ambigua* growing under solar UV-B levels at the beginning of the growing season. This tendency may be related to changing solar UV-B doses during spring and summer.

There appeared to be a cumulative response, through successive years, in some responses to UV-B: these were the number of live shoots and absorbance at 305 nm (as indicative of the concentration of UV-B absorbing compounds). To assess whether UV-B effects on this variable showed an increment with the year, we examined temporal trends in the relative reduction in these parameters that

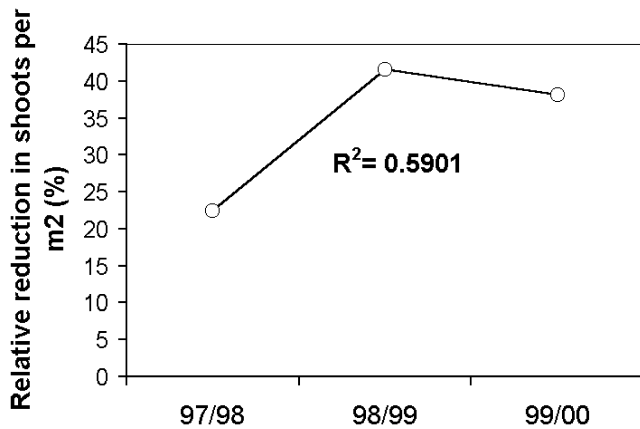


Fig. 7. Relationship between relative reduction in density (shoots per m^2) of *S. ambigua* attributable to UV-B exposure and growing season. Relative reductions were calculated as the change under near-ambient UV-B expressed as a percentage of the value.

appeared attributable to UV-B radiation effects (the reduction under near ambient UV-B treatment expressed as a percentage of data under attenuated UV-B). The results showed that the relative reduction in number of live shoots (green shoots) tended to increase from 22% (1998) to 41% (1999) with a light decline in the third growing season (38%). We found a tendency to a positive correlation between the relative reduction and year (linear model adjusted $r^2=0.59$; Fig. 7). There was no clear cumulative

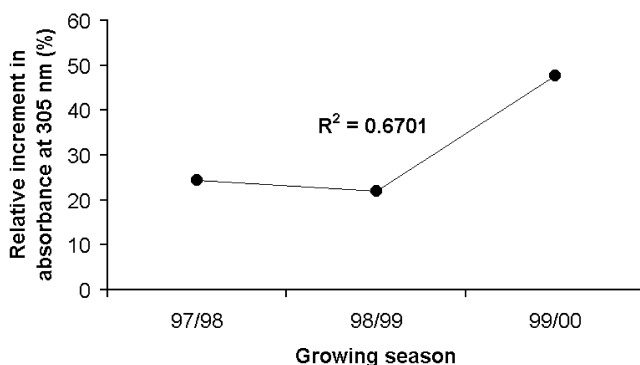


Fig. 8. Relationship between relative increments in absorbance at 305 nm of pigment compounds extracted from *S. ambigua* attributable to UV-B exposure and growing season.

Table 2
Effects of UV-B radiation on percentage of ground area covered

Treatment	Ambient UV-B			Near-ambient UV-B			Attenuated UV-B		
	1998	2000	$\Delta\%$	1998	2000	$\Delta\%$	1998	2000	$\Delta\%$
Uncovered soil	53.32	51.84	-1.48	42.85	43.60	0.76	16.80	14.20	-2.60
<i>S. ambigua</i>	46.01	46.65	0.64	54.97	50.04	-4.93	82.40	85.00	2.60
<i>Puccinellia</i> sp.	0.50	1.50	1.00	2.34	6.69	4.35	0.90	0.90	0.00

Percentage of ground area covered in mid summer by *S. ambigua* and *Puccinellia* sp. and changes in cover between the first and the last growing season of the experiment. Cover was calculated on small subplots (0.25×0.25 m) (details in Section 2). UV-B exclusion increased *Salicornia* cover with no changes in species composition.

UV-B effect on absorbance, though overall it increased from 25% (1997) to 48% (2000). Again, a tendency of positive correlation between these two parameters with a linear model adjusted $r^2=0.671$ (Fig. 8). For perennial plants, it has been proposed that the effects of UV-B on growth might accumulate over the years [40,48,49].

Differences in the response to solar UV-B among plant species could be the result of the adaptation of inherent mechanisms to tolerate only a certain maximum threshold of UV-B exposure. Plants respond to UV-B radiation by synthesising pigments that absorb in these wavelengths, or by morphological and physiological changes. Salt marsh species living under high environmental stress appear to adopt this dose-dependent response to UV-B radiation [39]. *S. ambigua* responded to UV-B levels independently of other environmental stresses; perhaps this is a stress-tolerant plant that did not yet reach the threshold of its stress response. Current levels of solar UV-B radiation have measurable inhibitory effects on a perennial *Salicornia* salt marsh. *S. ambigua* showed changes in some growth parameters (density of live and dead shoots, and biomass production), also increased concentration of epidermal UV-B screening compounds and cuticle thickness as mechanisms to reduce the penetration of UV-B into the mesophyll. A slight decrease in cover (shoot density) of *S. ambigua* (and corresponding increase in *Puccinellia*) under near-ambient solar UV-B (Table 2) indicates that dominance of *S. ambigua* in this community may decrease if current trends for increased solar UV-B continue. In conclusion, *Salicornia*, along with *Deschampsia* and *Colobanthus*, appears to be a good indicator species, to assess the impact of ozone depletion on Austral ecosystems.

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References

- [1] J.C. Farman, B.G. Gardiner, J.D. Shanklin, Large losses of total ozone in Antarctica reveal seasonal ClO_x/NO_x interaction, *Nature* 315 (1985) 207–210.
- [2] WMO scientific assessment of ozone depletion, in: WMO Ozone Report 37, WMO, Geneva, 1994.
- [3] V.L. Orce, E.W. Helbling, Latitudinal UVR-PAR measurements in Argentina: extent of the 'ozone hole', *Global Planetary Change* 15 (1997) 113–121.
- [4] M.C. Rousseaux, A. L. Scopel, P. S. Searles, M.M. Caldwell, O.E. Sala, C.L. Ballaré, Responses to solar ultraviolet-B radiation in a shrub-dominated natural ecosystem of Tierra del Fuego (southern Argentina), *Global Change Biol.* 7 (2001) 467–478.
- [5] K. Stamness, J. Slusser, M. Bowen, Biologically effective ultraviolet radiation, total ozone abundance, and cloud optical depth at McMurdo Station, Antarctica, September 15, 1988, through April 15, 1989, *Geophys. Res. Lett.* 17 (1990) 2181–2184.
- [6] M.C. Rousseaux, C.L. Ballaré, C.V. Giordano, A.L. Scopel, A.M. Zima, M. Szwarcberg-Bracchitta, P.S. Searles, M.M. Caldwell, S.B. Diaz, Ozone depletion and UVB radiation: impact on plant DNA damage in southern South America, *Plant Biol.* 96 (26) (1999) 15310–15315.
- [7] C.L. Ballaré, M.C. Rousseaux, P.S. Searles, J.G. Zaller, C.V. Giordano, T.M. Robson, M.M. Caldwell, O.E. Sala, A.L. Scopel, Impacts of solar ultraviolet-B radiation on terrestrial ecosystems of Tierra del Fuego (southern Argentina), an overview of recent progress, *J. Photochem. Photobiol. B: Biol.* 62 (2001) 67–77.
- [8] United Nations Environment Programme, Scientific assessment of ozone depletion: 1991, in: *Global Ozone Research and Monitoring Project Report Number 25*, UN, Geneva, 1991.
- [9] M. Tevini, Effects of UV-B radiation on terrestrial plants, in: M. Tevini (Ed.), *UV-B Radiation and Ozone Depletion: Effects on Humans, Animals, Plants, Microorganisms and Materials*, Lewis Publishers, London, 1993, pp. 125–153.
- [10] M.M. Caldwell, A.H. Teramura, M. Tevini, J.F. Bornman, L.O. Bjorn, G. Kulandaivelu, Effects of increased solar ultraviolet radiation on terrestrial plants, *Ambio* 24 (3) (1995) 166–173.
- [11] M.M. Caldwell, S.D. Flint, P.S. Searles, Spectral balance and UV-B sensitivity of soybean: a field experiment, *Plant Cell Environ.* 17 (1994) 267–276.
- [12] E.L. Fiscus, F.L. Booker, Is increased UV-B a threat to crop photosynthesis and productivity?, *Photosynth. Res.* 43 (1995) 81–92.
- [13] U. Johansson, C. Gehrke, L.O. Björn, T.V. Callaghan, The effects of enhanced UV-B radiation on the growth of dwarf shrubs in a subantarctic heathland, *Funct. Ecol.* 9 (1995) 713–719.
- [14] C.J. Beggs, U. Schneider-Ziebert, E. Wellmann, UV-B radiation and adaptive mechanisms in plants, in: R.C. Worcester, M.M. Caldwell (Eds.), *Stratospheric Ozone Reduction, Solar UV Radiation and Plant Life*, Springer Verlag, Berlin, 1986, pp. 313–325.
- [15] M. Tevini, J. Braun, G. Fieser, The protective function of epidermal layer of rye seedlings against ultraviolet-B radiation, *Photochem. Photobiol.* 53 (1991) 329–333.
- [16] E.H. DeLucia, T.A. Day, T.C. Vogelmann, Ultraviolet-B and visible light penetration into needles of two species of subalpine conifers during foliar development, *Plant Cell Environ.* 15 (1992) 921–929.
- [17] C.J. Beggs, E. Wellmann, Photocontrol of flavonoid biosynthesis, in: R.E. Kendrick, G.H.M. Kronenberg (Eds.), 2nd Edition, *Photomorphogenesis in Plants*, Kluwer Academic, Dordrecht, 1994, pp. 733–751.
- [18] J.P. Schnitzler, T.P. Jungblut, W. Heller, C. Langebartels, M. Kofferlein, H. Sandermann, Effects of ozone and UV-B radiation on Scots pine, *Acta Horticult.* 381 (1994) 463–469.
- [19] J.E. Frederick, S.B. Diaz, I. Smolskaia, W. Esposito, T. Lucas, C.R. Booth, Ultraviolet solar radiation in the high latitudes of South America, *Photochem. Photobiol.* 60 (1994) 356–362.
- [20] R. Robberecht, M.M. Caldwell, W.D. Billings, Leaf ultraviolet optical properties along a latitudinal gradient in the arctic alpine live zone, *Ecology* 61 (1980) 612–619.
- [21] M.M. Caldwell, S. Flint, Stratospheric ozone reduction, solar UV-B radiation and terrestrial ecosystem, *Climatic Change* 28 (1994) 375–394.
- [22] F.S. Xiong, T.A. Day, Effect of solar ultraviolet-B radiation during springtime ozone depletion on photosynthesis and biomass production of Antarctic vascular plants, *Plant Physiol.* 125 (2) (2001) 738–751.
- [23] T.A. Day, C.T. Ruhland, C.W. Grobe, F. Xiong, Growth and reproduction of Antarctic vascular plants in response to warming and UV radiation reductions in the field, *Oecologia* 119 (1999) 24–35.
- [24] C.T. Ruhland, T.A. Day, Size and longevity of seed banks in Antarctica and the influence of ultraviolet-B radiation on survivorship, growth and pigment concentrations of *Colobanthus quitensis* seedlings, *Environ. Exp. Bot.* 45 (2) (2001) 143–154.
- [25] E.M. Koch, The effect of UV-B radiation on salt-marsh vegetation along a latitudinal gradient, in: *Progress Report Inter-American Institute for Global Change Research Meeting*, Quebec, Canada, 1998.
- [26] N. San Román, O. Bianciotto, L. Pinedo, Effects of UV-B solar radiation on salt-marsh vegetation in San Sebastian Bay, Tierra del Fuego, Argentina, in: *Proceedings of the Sixth International Symposium of Cold Regions Development*, Hobart, Tasmania, Australia, 2000, pp. 5–8.
- [27] C.J. Dawes, Salt marsh communities, in: C.J. Dawes (Ed.), *Marine Botany*, John Wiley, New York, 1981, pp. 494–515.
- [28] A.J. Davy, G.F. Bishop, C.B. Costa, in: *Biological Flora of the British Isles, Salicornia L.*, 2000, p. 37.
- [29] J. Marcenaro, D. Gallegos, J. Gobbi, Relevamiento Ecológico Costero—Informe final, Servicio de Hidrografía Naval Argentina, Biblioteca Centro Austral de Investigaciones Científicas, Ushuaia, 1984.
- [30] M.B. Collantes, J. Anchorena, J. Ontiver, O. Bianciotto, Mapa de vegetación del Norte de Tierra del Fuego XX Jornadas Argentinas de Botánica, Sociedad Argentina De Botánica, Salta, 1985.
- [31] M.B. Collantes, A.M. Faggi, Los humedales del Sur de Sudamérica. En Ana Inés Malvarez, in: *Tópicos sobre humedales subtropicales y templados de Sudamérica*, UNESCO-MAB, 1999, pp. 15–25.
- [32] D.M. Moore, in: A. Nelson (Ed.), *Flora of Tierra del Fuego*, Missouri Botanical Garden, USA, 1983, p. 396.
- [33] P.S. Searles, S.D. Flint, S.B. Diaz, M.C. Rousseaux, C.L. Ballaré, M.M. Caldwell, Solar ultraviolet-B radiation influence on *Sphagnum* bog and *Carex* fen ecosystems: first field season findings in Tierra del Fuego, Argentina, *Global Change Biol.* 5 (1999) 225–234.
- [34] S.B. Diaz, G. Deferrari, D. Martinioni, A. Oberto, Regression analysis of biologically effective integrated irradiances versus ozone, clouds and geometric factors, *J. Atmos. Solar-Terrestrial Phys.* 62 (2000) 629–638.

- [36] M.C. Rousseaux, A.L. Scopel, P.S. Searles, M.M. Caldwell, O.E. Sala, C.L. Ballare, Responses to solar ultraviolet-B radiation in a shrub-dominated natural ecosystem of Tierra del Fuego (southern Argentina), *Global Change Biol.* 7 (2001) 467–478.
- [37] M.M. Caldwell, Solar ultraviolet radiation as an ecological factor for alpine plants, *Ecol. Monogr.* 38 (1968) 243–268.
- [38] J.H. Sullivan, A.H. Teramura, The effects of ultraviolet-B radiation on loblolly pines. 2. Growth of field-grown seedlings, *Trees* 6 (1992) 115–120.
- [39] E.W. Koch, A. Klueberg, P. Neale, J. Sullivan, C. Costa, T. Gonzalez, M. Bertiller, A.M. Beeskow, C.L. Sain, W. Helbling, V. Duro, F. Coronato, M. Tourn, O. Bianciotto, L. Pinedo, N. San Roman, A. Blessio, L. Orce, M. Collantes, 2000. Final report, in Workshop 'The impact of UV-B radiation on salt-marsh vegetation along a latitudinal gradient', August 31st–September 4th 2000, Horn Point, MD, USA. Unpublished manuscript, pp. 26–28.
- [40] T.A. Day, C.T. Ruhland, F.S. Xiong, Influence of solar ultraviolet-B radiation on Antarctic Terrestrial plants: result from a 4-year field study, *J. Photochem. Photobiol. B: Biol.* 62 (2001) 78–87.
- [41] M.B. Collantes, J. Anchorena, G. Korembli, A soil nutrient gradient in magellanic *Empetrum* heathlands, *Vegetatio* 80 (1989) 183–193.
- [42] R. Mendoza, M.B. Collantes, J. Anchorena, S.M. Cainzos, Efectos del encalado y de la fertilización con nitrógeno y fósforo en suelos ácidos de la Isla Grande de Tierra del Fuego, in: XIII Congreso Argentino de las Ciencias del Suelo, S.C. de Bariloche, Libro de resúmenes, 1991.
- [43] M.M. Caldwell, A.H. Teramura, M. Tevini, J.F. Bornman, L.O. Bjorn, G. Kulandaivelu, Effects of increased solar ultraviolet radiation on terrestrial plants, *Ambio* 24 (3) (1995) 166–173.
- [44] A.H. Teramura, H.J. Sullivan, Effects of UV-B radiation on photosynthesis and growth of terrestrial plants, *Photosynth. Res.* 39 (1994) 463–473.
- [45] T.A. Day, G. Martin, T.C. Vogelmann, Penetration of UV-B radiation in foliage: evidence that the epidermis behaves as a non-uniform filter, *Plant Cell Environ.* 16 (1993) 735–774.
- [46] J. Li, T.M. Ou Lee, R. Raba, R.G. Amundson, R.L. Last, Arabidopsis flavonoids mutants are hypersensitive to UV-B irradiation, *Plant Cell* 5 (1993) 171–179.
- [47] E.H. De Lucía, T.A. Day, T.C. Vogelmann, Ultraviolet-B radiation and the Rocky Mountain environment: measurement of incident light and penetration into foliage, *Curr. Top. Plant Biochem. Physiol.* 10 (1991) 32–48.
- [48] U. Johansson, C. Gehrke, L.O. Bjorn, T.V. Callaghan, The effects of enhanced UV-B radiation on the growth of dwarf shrubs in a subarctic heathland, *Funct. Ecol.* 9 (1995) 713–719.
- [49] L.O. Bjorn, T.V. Callaghan, I. Johnsen, J.A. Lee, Y. Manetas, N.D. Paul, M. Sonesson, A.R. Wellburn, D. Coop, H.S. Heide-Jorgensen, C. Gehrke, D. Gwynn-Jones, U. Johansson, A. Kypris, E. Levizou, D. Nikolopoulos, Y. Petropoulou, M. Stephanou, The effects of UV-B radiation on European heathland species, *Plant Ecol.* 128 (1997) 252–264.
- [50] P.S. Searles, B.R. Kropp, S.D. Flint, M.M. Caldwell, Influence of solar UV-B radiation on peatland microbial communities of southern Argentina, *New Phytol.* 152 (2001) 213–221.