Contents lists available at ScienceDirect



Journal of Experimental Marine Biology and Ecology

journal homepage: www.elsevier.com/locate/jembe



# The susceptibility of spores and propagules of Antarctic seaweeds to UV and photosynthetically active radiation — Field versus laboratory experiments

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#### ARTICLE INFO

Article history: Received 28 November 2013 Received in revised form 8 May 2014 Accepted 9 May 2014 Available online 24 May 2014

Keywords: Adenocystis utricularis Glacial discharge Global warming Himantothallus grandifolius Iridaea cordata Light climate

# ABSTRACT

The Western Antarctic Peninsula is strongly affected by stratospheric ozone depletion, leading to higher UVB radiation (UVBR) on the Earth surface. It is furthermore experiencing the fastest rates of global warming worldwide, resulting in an increased sediment run-off from glacial melting, altering the underwater light climate. Very little is known of how Antarctic organisms can cope with this rapidly changing environment. Seaweeds play an essential role within the Antarctic coastal ecosystems, building highly complex and productive underwater communities. The unicellular spores are the most sensitive stage in their life-cycle, forming the bottle-neck for successful recruitment. To supplement the very rare field experiments on seaweed propagules, three ecologically important Antarctic seaweeds (Adenocystis utricularis, Himantothallus grandifolius, Iridaea cordata) were investigated. The germination of spores after exposure in the field to different water depths to three light treatments (PAR; PAR + UVA; PAR + UVA + UVB) was recorded. In parallel, spores were exposed to the same treatments under artificial radiation in the laboratory for different periods. Germination of the intertidal species A. utricularis was not affected by the treatments. In spores of I. cordata and H. grandifolius depth was a major factor for successful germination. High PAR fluxes at 1 and 2 m water depth inhibited germination significantly. UVR further lowered germination in H. grandifolius while in I. cordata UVBR had a negative impact only in the laboratory experiment. The results show that already the unicellular life stage expresses strong species-specific susceptibility to changes in the radiation climate. Not only UVR but also the high PAR fluxes in the field are important factors in determining the upper distribution limit of Antarctic seaweeds and laboratory experiments show stronger UVB effects as studies under natural radiation.

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# 1. Introduction

Ecological effects of global environmental changes on Antarctic organisms, particularly the increase in ultraviolet-B radiation (UVBR, 280–315 nm) caused by stratospheric ozone depletion (Farman et al., 1985; Weatherhead and Andersen, 2006), and rising temperatures due to global warming (IPCC, 2001) are of considerable concern (Roleda et al., 2007a). The Western Antarctic Peninsula (WAP) belongs to the most rapidly warming regions on earth, with a rise of surface air temperature of 3.4 °C (Vaughan et al., 2003) per century, compared to a global mean increase of 0.6 °C (Turner et al., 2007). One of the consequences of the local warming trend is a significant increase in land and sea ice melting, which intensifies the sediment run-off from the glaciers into the oceanic system during the melting season (spring-summer; Cook et al., 2005; Turner et al., 2007), affecting the underwater light climate in the proximity to the glaciers with unknown consequences for the primary producers (Schloss et al., 2002).

At the WAP, seaweeds constitute a year-round essential carbon sink through production of high amounts of biomass with maximum wet biomass in the sublittoral of over 10 kg fresh weight  $m^{-2}$  (Gómez et al., 2009; Quartino and Boraso de Zaixso, 2008). In some Antarctic areas the phytoplankton productivity is very low and it is postulated that the benthic primary producers (seaweeds and benthic microalgae) form an important food source for the heterotrophic community (Schloss et al., 2002). In this way, seaweeds are crucial for the diversity and stability of the polar coastal ecosystems. Due to retreating glaciers, new areas in the upper subtidal and intertidal will be accessible for seaweed colonization in the future, thereby altering the oceanic food web (Quartino et al., 2013).

As the penetration of light into the water column is altered by an accelerated glacial melting due to global warming, both, the upper and lower depth distribution limit of seaweeds might be modified. At the study site King George Island, Antarctica, the number of annual fresh water discharge days and daily discharge volume has doubled within the period from 2002 to 2006 (Eraso and Dominguez, 2007). While the lower depth distribution limit of the seaweeds depends on the capacity of the species to maintain a positive carbon balance (Gómez

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et al., 1997), the upper limit is determined by the capability to cope with excessive photosynthetically active radiation (PAR 400–700 nm) and the tolerance to UV radiation (UVR 280–400 nm, Bischof et al., 2006; Hanelt, 1998; Wiencke et al., 2004, 2006). Negative effects of UVR on seaweeds are, among others, the inhibition of photosynthesis or even photodamage, protein breakdown and the damage of the DNA resulting in lower germination and growth rates (reviewed in Bischof et al., 2006; Karsten et al., 2009; Roleda et al., 2007a).

The upper depth distribution of seaweeds can be determined by the susceptibility of their early life history stages (unicellular spores and propagules) to environmental perturbations (Swanson and Druehl, 2000; Wiencke et al., 2000). These stages are known to be the bottle neck for the successful recruitment of the species because they are more vulnerable to changes in the abiotic environment compared to the mature sporophytes in their life history (Agrawal, 2009; Coelho et al., 2000; Cordi et al., 2001; Roleda et al., 2007a; Véliz et al., 2006; Wiencke et al., 2006). It is therefore necessary to understand the physiological limits of the early developmental stages of the important seaweed species to be able to identify the main factors leading to a successful recruitment.

Most of the studies on young developmental stages have been performed under laboratory conditions with unnatural ratios between PAR, UVA and UVB radiation. Experiments on young developmental algal stages under ambient solar radiation in Antarctica are very rare (but see Zacher et al., 2007a; Zacher and Campana, 2008 working on early successional communities). In laboratory experiments UVR was shown to negatively affect the photosynthetic efficiency of Adenocystis utricularis zoospores, additionally damaging the DNA by forming cyclobutane pyrimidine dimers (CPDs). However, a full recovery is observed after 48 h under low light (Zacher et al., 2007a, 2007b). The subtidal red alga Gigartina skottsbergii and the endemic brown algae Ascoseira mirabilis were not able to repair their DNA damage completely after 8 h of exposure to 0.4 W m<sup>-2</sup> UVBR but fully recovered after shorter times of exposure (Roleda et al., 2007b, 2008). In other regions of the Earth some experiments on the high PAR and UV tolerance of young developmental stages have been performed with inconsistent results (Hanelt et al., 1997; Jiang and Gao, 2008; Steinhoff et al., 2011; Wiencke et al., 2006). While Wiencke et al. (2006) and Steinhoff et al. (2011) found a decreased spore germination of subtidal species due to UVR and not under high PAR, Hanelt et al. (1997) and Jiang and Gao (2008) found a strong photoinhibition of photosynthesis due to high PAR and a weaker additional inhibition due to UVR.

To our knowledge, no data have been published so far on the germination of Antarctic seaweeds under ambient solar radiation, even though this region exhibits the strongest degree of stratospheric ozone depletion worldwide. In order to get deeper insights in the high PAR and UV tolerance of Antarctic seaweeds, the spore germination of the inter- to subtidal Iridaea cordata (Rhodophyta), the inter- to upper subtidal Adenocystis utricularis and the Antarctic endemic subtidal Himantothallus grandifolius (both Phaeophyceae) was investigated in Potter Cove, King George Island, Antarctica. Spores were exposed for ~24 h to i) PAR (P), ii) PAR + UVAR (PA) and iii) PAR + UVAR + UVBR (PAB) in different water depths (1, 2, 4 and 8 m) in the field and germination rates were subsequently determined after exposure to low light in the laboratory. For comparative reasons the same species were simultaneously exposed under laboratory conditions, where the different depths were simulated by different periods of exposure (1, 2, 4 and 8 h).

The study aimed at answering several questions:

- 1. Which wavelength range exerts the strongest effects on the germination of Antarctic propagules?
- 2. Is there a possible alteration in the upper vertical distribution limit of the subtidal species due to an increased sediment inflow during summer?

3. Are laboratory experiments suitable to provide results which can be used to predict the performance in the field?

## 2. Materials and methods

# 2.1. Algal material

Fertile specimens of the brown algae *A. utricularis* (Bory) Skottsberg, *H. grandifolius* (A.Gepp and E.S.Gepp) Zinova and the red alga *I. cordata* (Turner) Bory de Saint-Vincent were collected in November 2008 (*A. utricularis* and *I. cordata*) and February 2010 (*H. grandifolius*) at Potter Cove (King George Island, South Shetland Islands, 62°14.80′S, 58°41.26′W) during two expeditions. *Adenocystis utricularis* and *I. cordata* were collected in the intertidal, whereas *H. grandifolius* grows in the subtidal and was collected by SCUBA diving at approx. 10 m depth. After collection, the specimens were brought immediately to the nearby laboratory and kept at ~2 °C under low light conditions until further processing.

# 2.2. Spore release

Numerous individuals of each species were divided randomly in 5 replicates and prepared for spore release by blotting with tissue paper and treating the different species in the following ways: I. cordata tetrasporophytes were cut into smaller pieces and put into glass flasks with filtered seawater  $(0.2 \ \mu m)$  for collection of spores after a few days. Complete thalli of A. utricularis and fertile tissue of H. grandifolius (cut with a razor blade) were kept in darkness in moist chambers overnight or a few days at <5 °C. Spore release was obtained by flooding the algae with filtered, slightly warmer seawater in photo-dishes according to Clayton and Wiencke (1986). The initial spore density of brown algae was counted by the use of a Neubauer-chamber (Brand, Wertheim, Germany) and of the red alga by a Rafter chamber (Sedgewick-Rafter Cell S50 spore counter, Graticules Ltd., Tonbridge, UK), respectively. Initial spore densities for A. utricularis spore suspension were (zoospore length around 4  $\mu$ m) approx. 1.8  $\times$  10<sup>5</sup> spores ml<sup>-1</sup>, for *H. grandifolius* spore suspension (zoospore length around 4  $\mu m)$  approx. 1.42  $\times$ 10<sup>5</sup> spores ml<sup>-1</sup> and for *I. cordata* tetraspore suspension (mean diameter  $20 \,\mu\text{m}$ ) approx. 6000 spores ml<sup>-1</sup>. Spore solutions were then divided and diluted between the field and the laboratory approach. The spore solution of the same species was exposed in the field and under laboratory conditions at the same date, whereas spores of different species were exposed at different dates due to different times of fertility. Himantothallus grandifolius e.g. gets fertile in austral summer to autumn, and was investigated in 2010 whereas fertile I. cordata and A. utricularis were collected in spring 2008.

#### 2.3. Field experiment

The field experimental units consisted of an aluminum frame (~1 m  $\times$ 1 m) with a black plastic bottom and a top of UV-transparent Plexiglas (GS 2458, Röhm, Darmstadt, Germany). It contained 16 Petri dishes  $(53 \times 12 \text{ mm})$  arranged in a  $4 \times 4$  grid. Petri dishes were filled with the spore solutions and exposed to three different light treatments and four depths (1, 2, 4 and 8 m) in a two-factorial design (n = 5, *I. cordata* and *A. utricularis* and n = 4, *H. grandifolius*). Three kinds of filter foils were used to obtain the different light treatments (see Bischof et al. 2002 for details): 1. Ultraphan transparent (Digefra GmbH, Germany), 2. Folanorm 320 (Folex GmbH, Germany), and 3. Ultraphan URUV farblos, corresponding to the PAR + UVAR + UVBR (PAB, 280 to 700 nm), PAR + UVAR (PA, 320 to 700 nm) and PAR (P, 400 to 700 nm) treatments, respectively. The cut-off wavelengths of the available filter-material were slightly different from the definition of CIE (Commission Internationale De l'Éclairage, UVB = 280–315 nm, UVA = 315-400 nm) but are for practical reasons commonly used in environmental science (Franklin et al., 2003). Experimental units were

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