ARTICLE IN PRESS

Marine Environmental Research xxx (xxxx) xxx–xxx

Contents lists available at ScienceDirect

Marine Environmental Research

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

Physiological responses to variations in grazing and light conditions in native and invasive fucoids

Celia Olabarria[a,*], Francisco Arenas[b], Ángela Fernández[b], Jesús S. Troncoso[a], Brezo Martínez[c]

[a] Departamento de Ecoloxía e Bioloxía Animal, Facultade de Ciencias Experimentais, Campus Lagoas-Marcosende, 36310, Vigo, Pontevedra, Spain
[b] Aquatic Ecology & Evolution Group, Interdisciplinary Centre for Marine and Environmental Research (CIIMAR), University of Porto, Terminal Cruzeiros Porto Leixões, 4450-208 Matosinhos, Portugal
[c] Area de Biodiversidad y Conservación, Rey Juan Carlos University, 28933 Móstoles, Madrid, Spain

ARTICLE INFO

Keywords:
Fucoids
Ecophysiological responses
Grazing
Light quality and intensity
Additive effects

ABSTRACT

Poor physiological acclimatization to climate change has led to shifts in the distributional ranges of various species and to biodiversity loss. However, evidence also suggests the relevance of non-climatic physical factors, such as light, and biotic factors, which may act in interactive or additive way. We used a mechanistic approach to evaluate the ecophysiological responses of four seaweed species (three dominant intertidal fucoids, *Fucus serratus*, *Ascophyllum nodosum*, *Bifurcaria bifurcata,* and the invasive *Sargassum muticum*) to different conditions of grazing, light irradiance and ultraviolet (UV) radiation. We performed a large-scale mesocosm experiment with a total of 800 individual thalli of macroalgae. The factorial experimental design included major algal traits, photoacclimation, nutrient stoichiometry and chemical defence as response variables. Few significant effects of the factors acting alone or in combination were observed, suggesting a good capacity for acclimatization in all four species. The significant effects were generally additive and there were no potentially deleterious synergistic effects between factors. *Fucus serratus*, a species currently undergoing a drastic contraction of its southern distribution limit in Europe, was the most strongly affected species, showing overall lower photosynthetic efficiency than the other species. The growth rate of *F. serratus* decreased when UV radiation was filtered out, but only in the presence of grazers. Moreover, more individuals of this species tended to reach maturity in the absence of grazers, and the nitrogen content of tissues decreased under full-spectrum light. Only the phlorotannin content of tissues of *B. bifurcata* and of exudates of *A. nodosum*, both slow-growing species, were positively affected by respectively removal of UVB radiation and the presence of grazers. The findings for *S. muticum*, a well-established invasive seaweed across European coasts, suggested similar physiological response of this fast-growing species to different levels of grazing activity and light quality/intensity. As expected, this species grew faster than the other species. *Bifurcaria bifurcata* and *A. nodosum* only showed minor effects of light quality and grazing on phlorotannins content, which suggests good resistance of these two long-lived species to the experimental conditions. Mechanistic approaches that are designed to analyse interactive effects of physical and biotic factors provide an understanding of physiological responses of species and help to improve the confidence of predictive distribution models.

1. Introduction

Intertidal species, which inhabit the interface between land and sea, endure highly variable oceanic and atmospheric environmental conditions (e.g. Helmuth et al., 2002) such as changing levels of light, salinity and temperature (reviewed in Lobban and Harrison, 1994; Hurd et al., 2014). Canopy-forming macroalgae are the most important structural engineers of temperate intertidal ecosystems and play important biotic roles such as providing habitat, shelter and food for the accompanying flora and fauna (Wikström and Kautsky, 2007).

Environmental stress associated with climatic and non-climatic factors and biotic stressors can affect macroalgae at biochemical, ecophysiological, morphological and population levels (Weidner et al., 2004; Martínez et al., 2012a; Celis-Plá et al., 2014; Fernández et al., 2015). Although light promotes photosynthetic activity, seaweeds may be physiologically damaged by excessive solar irradiance, including increased levels of UV radiation, particularly during low tide (Figueroa and Viñegla, 2001; Gao and Xu, 2010; Figueroa et al., 2014). The increased levels of solar radiation associated with weaker radiative effects of clouds and aerosols in temperate latitudes, already observed in

* Corresponding author.
E-mail address: colabarria@uvigo.es (C. Olabarria).

https://doi.org/10.1016/j.marenvres.2018.05.016
Received 16 March 2018; Received in revised form 19 April 2018; Accepted 15 May 2018

temperate latitudes (Wild, 2012; Sanchez-Lorenzo et al., 2013), may compromise the performance and/or survival of seaweeds (Quintano et al., 2013), especially in intertidal habitats. Regarding biotic constraints, the actions of grazers may compromise growth, photosynthesis and reproduction of macroalgae (Svensson et al., 2009; Rothäusler et al., 2009; Kraufvelin, 2017) and thus strongly affect the distribution and biomass of the latter. In some areas, grazers can remove up to 100% of macroalgae present (e.g. Weidner et al., 2004). Not only direct grazing, but also release of chemical cues by grazers may have strong effect on population dynamics and physiological performance of macroalgae (Rohde and Wahl, 2008), although responses may vary depending on the identity of grazers (Moore et al., 2007).

Macroalgae show different levels of sensitivity to stress depending on species, morpho-functional group, ecological strategy and life cycle (Figueroa et al., 2014; Hurd et al., 2014). Overall, the physiological vulnerability and acclimatization of seaweeds to UV radiation will depend on the balance between photoprotective mechanisms, accumulation of antioxidants and activation of antioxidant enzymes, as well as the accumulation of UV-screening photoprotective compounds, such as phlorotannins in brown macroalgae (Abdala-Díaz et al., 2006; Figueroa et al., 2014). These compounds have been associated with various cell functions and in particular, as deterrents to grazers (e.g. Pavia and Toth, 2000). The protective role of phlorotannins may be important both intracellularly and extracellularly, mainly *via* exudation (Koivikko et al., 2005). Nevertheless, the role of phlorotannins in inducible or constitutive defence against grazers is highly controversial (e.g. Kubanek et al., 2004; Toth et al., 2005; Long et al., 2013), and it is possible that factors such as phlorotannin structure, grazer gut environment or geographical variation may mediate the effects (Kubanek et al., 2004).

Although intertidal macroalgae are characterized by a plastic ecophysiology, responses to stress are species-specific and vary depending on the environmental context (Einav et al., 1995; Figueroa et al., 2014). This is of particular importance when comparing native and invasive species because the latter are expected to show greater physiological plasticity and a broader physiological tolerance to abiotic and biotic factors (reviewed in Schaffelke et al., 2006; Pansch et al., 2008; Engelen et al., 2011).

A mechanistic understanding of the factors that determine the performance of different species is essential for studying their resilience to environmental changes (Fernández et al., 2015; Martínez et al., 2015). Poor physiological acclimatization in response to climate change has resulted in the decline of populations in numerous species worldwide, in association with shifts and retreats in the distribution ranges of marine and terrestrial species and the spread of invasive species of warm affinity (Hickling et al., 2006; Perry et al., 2005; Mieszkowska et al., 2006; Lima et al., 2007; Tylianakis et al., 2008). However, nonclimatic physical and biotic factors also play a fundamental role in shaping the distribution of species (Martínez et al., 2012a; Hansen and Stone, 2015; Godsoe et al., 2017).

Interactive effects among environmental and biotic factors on physiological performance of species can cause unexpected changes in species distribution (Darling and Côté, 2008; Godsoe et al., 2017). Although it has been often assumed the prevalence of synergistic interactive effects, additive and antagonistic effects are more common than expected, which suggests highly idiosyncratic responses (Crain et al., 2008; Darling and Côté, 2008). Potentially synergistic effects between physical and biotic factors in the intertidal zone have been reported (Harley et al., 2006; Kubicek et al., 2011). However, scant evidence of non-additive effects between physical factors involved in the ongoing decline of canopy-forming species at their geographic distributional limits has been obtained in several previous experimental studies (Martínez et al., 2012a; Ferreira et al., 2014; Celis-Plá et al., 2014; Fernández et al., 2015). Previous research on the interactive effects of solar radiation and grazing activity on brown macroalgae did not yield conclusive results (Lotze et al., 2002; Macaya et al., 2005; Kubicek et al., 2011; Rothäusler et al., 2011). While both factors may severely affect the productivity of early recruits (Lotze et al., 2002), adults can show efficient photoacclimation and be less affected by grazers (Macaya et al., 2005; Rothäusler et al., 2011).

In the present study, we aimed to investigate the interactive effects of solar radiation and grazing activity on the ecophysiological responses of intertidal fucoids, including a well-known invasive species that dominate European coasts. We focused on four habitat-forming fucoid species: the cold-temperate *Ascophyllum nodosum* (Linnaeus) and *Fucus serratus* Linnaeus, the southern lusitanic *Bifurcaria bifurcata* R. Ross, and the invasive Asiatic species *Sargassum muticum* (Yendo) Fensholt *Ascophyllum nodosum* has a scattered but persistent geographic distribution in southern Europe, although some populations have recently retreated, at least in the Bay of Biscay and on the coast of Asturias, N Spain (Viana et al., 2014). *Fucus serratus* has been undergoing a drastic contraction of its distribution range on the northern Spanish coast during the last decade (Viejo et al., 2011; Duarte et al., 2013), whereas the distribution of *B. bifurcata* and *S. muticum* are currently expanding towards southern Portugal and Morocco (Lima et al., 2007; Martínez et al., 2012b; Sabour et al., 2013). The latter species is now successfully established on European coasts (Farnham et al., 1973). Overall, these distributional trends suggest that these species show different levels of sensitivity to climatic and non-climatic physical factors (Svensson et al., 2009; Martínez et al., 2012a; b; Fernández et al., 2015; Martínez et al., 2015).

We performed a factorial mesocosm experiment in order to evaluate the ecophysiological response of the above-mentioned macroalgae to the combined effects of light irradiance, UV radiation and grazing by intertidal invertebrates. Specifically, we assessed major vital traits (growth rate and reproductive output), photoacclimation (by PAM fluorometry), nutrient stoichiometry (total C and N), and chemical defence (phlorotannin contents of tissues and exudates). These variables were considered functional indicators of the physiological responses to stress conditions and consequently of the capacity for acclimatization (see Figueroa et al., 2014). We expected that the ecophysiological performance of the four species, in relation to their current distributional trends, would be affected differently and that the successful invader *S. muticum* would show an enhanced acclimatization capacity. On the basis of our previous findings about physiological responses of fucoids to diverse environmental factors (Martínez et al., 2012a; Ferreira et al., 2014; Fernández et al., 2015), we also expected additive effects to be of greater importance than interactive effects.

2. Material and methods

2.1. Collection sites and organisms

One week before the start of the experiment (31 August 2012), vegetative fronds of *B. bifurcata* and *S. muticum* were collected from intertidal shores at Cabo Estai (42°11′ N, 8° 48′ W), whereas *A. nodosum* was collected at Rande (42°17′ N, 8°39″ W), both in the Ria de Vigo, NW Spain. Vegetative fronds of *F. serratus* were collected at Amorosa (41° 38′ N, 8°49′ W), N Portugal. The fronds were immediately transported to the laboratory in a cool icebox and submerged in seawater to a depth of about 8 cm in a 300 L aerated tank outdoors until required. Before being weighed, all experimental fronds were carefully cleaned to remove grazers and epiphytes, and excess water was removed with absorbent paper.

Two very abundant mesograzers, the snails *Gibbula umbilicalis* (da Costa) and *Littorina obtusata* (Linnaeus), which are naturally associated with the selected macroalgae, were chosen for this study. The snails were collected by hand from the intertidal zone of Cabo Estai and Rande (both in the Ría de Vigo. In the laboratory, the gastropods were maintained outdoors in a 30 L aerated tank and fed ad libitum with the four macroalgae until the start of the experiment. Snails of similar size were used to minimize size-specific differences in feeding behaviour