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Seawater-temperature and UV-radiation interaction modifies oxygen consumption, digestive process and growth of an intertidal fish

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ABSTRACT

UV-radiation (UVR) and temperatures have increased substantially over recent decades in many regions of the world. Both stressors independently have shown to affect the metabolism and growth in fish. However, because increase of both stressors are occurring concomitantly, to better understand their influences on marine species, their combined effects were evaluated. We test the hypothesis that UVR and temperature act synergistically affecting the metabolism, digestive process and growth of an intertidal fish. Two UVR conditions (with and without UVR) and two temperature levels (20° C and 25° C) were used. UVR increase the oxygen consumption and this was associated to opaque feces production. The absorption efficiency was higher without UVR at high temperatures (25 °C) and with UVR at low temperatures (20 °C). Finally, independent of UVR treatment, fish subjected to low temperature have higher biomass than those of high temperature. The interaction between UVR and temperature may influence on the physiology and growth of animals that inhabit in extreme habitats as upper intertidal, it could pose significant functional for aquatic animal survivorship.

1. Introduction

Temperature and ultraviolet radiation (UVR) are two important global environmental drivers that can interact affecting different processes of marine animals (Day et al., 1999; Alton and Franklin, 2012; Kazerouni et al., 2016). Previous investigations have shown the effects each of these stressors (i.e. UVR and temperature increases) on marine organisms (Kazerouni et al., 2016). Investigating the responses of individual species to multi-environmental stressors provides a comprehensive framework for understanding their effects on organisms, populations, communities and ecosystems (Whitehead et al., 2004)

UVR impairs cellular function, impacting/affecting all levels of biological organization (Häder et al., 2011; Williamson et al., 2014). Increased UVR levels provoke detrimental effects on growth rate, development, locomotors performance, survival, and immune functions in both vertebrate and invertebrate species (Leech and Williamson, 2001; Jokinen et al., 2001, 2008, 2011; Macias et al., 2007; van Uitregt et al., 2007; Bancroft et al., 2008; Sharma et al., 2005, 2008; Alton et al., 2011; Cramp et al., 2014). On the other hand, the temperature is the most important abiotic parameter influencing on marine organisms (Yao and Somero, 2014). The temperature plays a key role on the physiological responses, feeding, behavior, distribution range, ecology of organisms, and resistance to diseases of many species (Cossins and Bowler, 1987; Hill et al., 2012). However, the combined effects of these environmental stressors can be complex and unknown for many species (Byrne, 2011).

The effects of UVR can be modified by the increase in temperature. For example, simultaneous exposure of amphibian embryos to low temperatures and high UVR levels caused a synergistic increase in mortality (van Uitregt et al., 2007; Searle et al., 2010). These responses have been interpreted as a great UVR damage at low temperature due to retardation of all biochemical reactions reducing the repair of UVR damage (Broomhall et al., 2000; Seebacher et al., 2016). The knowledge about effects of UVR and

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temperature has advanced, however more research will help to understand how organism of several taxonomic groups that inhabit stressful habitats as upper intertidal zone, may be globally affected by the current and potential future scenarios.

In rocky intertidal habitats, marine organisms are subject to a wide environmental variability, which affect their physiology and ecology (Helmuth et al., 2006). In this heterogeneous habitat, it has been found that the tide pools are ephemeral habitats characterized by a discrete nature in time and space, as well as by a great variability in the physical conditions (Horn et al., 1999; Pulgar et al., 2005). Tide pools physical conditions (e.g. temperature, UV-radiation, salinity) were associated to the pool position in intertidal vertical gradient, with upper tide pool being exposed to high physical variability in UVR and temperature (Pulgar et al., 2005, 2015; Carrasco-Malio et al., 2014). In tidal pools of the Chilean coast, the fish *Girella laevifrons* (Pisces: Kyphosidae) "Baunco" is one of the most abundant species. This species inhabits upper tide pools only during their juvenile stage (first two years), later migrating to the subtidal zone for reproduction (Varas and Ojeda, 1990; Muñoz and Ojeda, 1997). *G. laevifrons* is exposed to extreme physical variability in UVR and temperatures ranging from 0.2 to 9 Wcm^{-2} and from 15 to 25 °C respectively (Pulgar et al., 2005, 2015, 2017). This species is one of the most abundant fish inhabiting the rocky intertidal zone and represents an important ecological component of intertidal system (Varas and Ojeda, 1990; Pulgar et al., 2005). Therefore, the aim the present study was to investigate the impact of the combined effects of UVR and temperature increase on the physiological performance of *G. laevifrons*.

2. Material and methods

2.1. Study animals and treatments

Juvenile individuals of *Girella laevifrons* (n = 36, standard length = 4.07 ± 0.13 cm and mean weight = 0.95 ± 0.08 g) were captured at Isla Negra (33°26′32″S 71°41′07″W), central Chile during summer 2015 (seawater temperature: 18 °C, salinity: 34.2 PSU, dissolved oxygen: 6.32 mgO_2/l, pH: 8.04). All fish were obtained from upper intertidal pools using BZ-20 anesthetic (dose 15 mL L^{-1}) and were immediately introduced into seawater with constant aeration and transported to the laboratory. At the laboratory, all specimens were maintained for seven days in a recirculation system with filtered seawater at a controlled temperature between 17 and 18° C and salinity of 34‰. During experimental period all fish were fed *ad libitum* with individuals of the mussel *Mytilus chilensis*.

After acclimation period the fish were assigned to one of four treatment during a period of six days: i) with UVR and 20 °C, ii) without UVR and 20 °C, iii) with UVR and 25 °C and iv) without UVR and 25 °C. Water temperature in this setup was in agreement with the highest values registered by in the upper intertidal pools from central Chile (Pulgar et al., 2005). Each experimental treatment consisted of three replicates with three individuals each (Fig. 1). UV light was provided by a 40W fluorescent tube (wavelength 350–400 nm, Phillips Actinic BL), and a 100W Luminaria tube (wavelength of 290–315 nm, ZINGG ILUMINA) that emits an irradiance of 0.3 Wcm^{-2}. Individuals who did not receive UV irradiance, received Photosynthetic Active Radiation (PAR), emitted by 2 Phillips daylight tubes (wavelength >400 nm) that emitted an irradiance of 0.0002 Wcm^{-2} (all measurements were made at 30 cm from tubes). The photoperiod 12:12 h was used; in UVR treatment 4 h of UV light was used after feed. The 4 h used in trials in UVR, was due to the time of exposure to which the animals are subjected under natural conditions (Pulgar et al., 2005, 2017). Two Thermo-heating Atman (200 W) was used to maintain a constant temperature for all treatments (Fig. 1).

2.2. Oxygen consumption

The oxygen consumption was determined through a metabolic chamber (Chapelle and Peck, 1995). In order to avoid metabolic costs of digestion and the endogenous cycles of intertidal fish (Horn et al., 1999), all fish were starved 48 h before oxygen consumption measurements. Flasks of 800 mL were filed with seawater and fully saturated with oxygen by constant bubbling. Once seawater saturated with oxygen, the dissolved oxygen concentration (mg L^{-1}) was measured with an oxygen-meter (OXI-Check, HI9147-04, Hanna Instruments, EEUU). Each individual was placed in a metabolic chamber without leaving bubbles inside, where individuals remained for 120 min. Then, each metabolic chamber was carefully opened and the dissolved oxygen was measured (Peck and Veal, 2001). After each oxygen consumption measure, the standard length (cm) and total body weight (g) were estimated. Finally, oxygen consumption was calculated as the difference between the final and initial concentration of dissolved oxygen in each test, and was expressed as mg L^{-1} O_2*g^{-1}*min^{-1}.

2.3. Absorption efficiency and fecal analysis

On the fifth day of experimentation, the absorption efficiency was measured using Conover, 1966 method, based on the relationship between organic and inorganic matter values of ingested food and fecal material. This method assumes that the absorption affects only the organic portion of the food. Absorption efficiency was calculated according to the following equation:

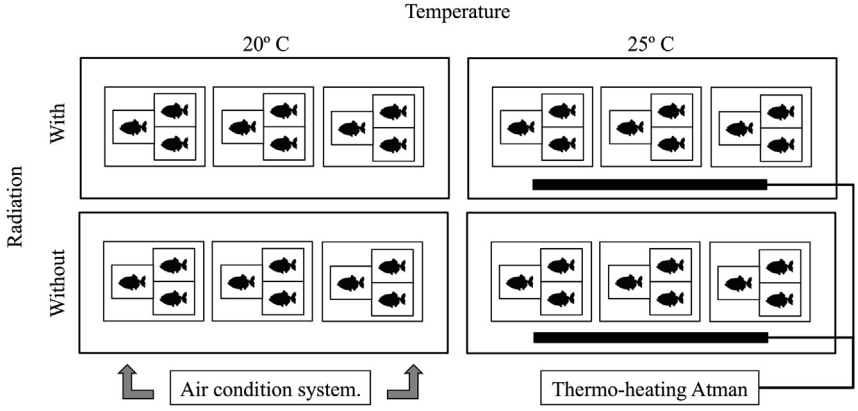

Fig. 1. Schematic diagram of experimental design. Aquariums and replicates for each treatment are shown.