



Carryover effects of short-term UV-B radiation on fitness related traits of the sea urchin *Strongylocentrotus intermedius*

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ARTICLE INFO

Keywords:

Sea urchin
UV-B
Carryover effect
Fitness

ABSTRACT

Carryover effects of UV-B radiation are largely unknown in marine invertebrates, despite the ecological importance. For the first time, we investigated fitness related traits of the sea urchin *Strongylocentrotus intermedius* 8 weeks after short-term (1 h) UV-B radiations (0, 10 and 20 $\mu\text{W cm}^{-2}$). Short-term UV-B radiations had significant negative effects on survival, food consumption, test diameter, test height, test height: test diameter, gonad weight and crude protein of gonads of *S. intermedius*, despite of the absence of UV-B radiation for 8 weeks. Survival, food consumption and crude protein of gonads were significantly lowest in *S. intermedius* exposed to UV-B radiation at 20 $\mu\text{W cm}^{-2}$, highlighting that 20 $\mu\text{W cm}^{-2}$ is a dangerous UV-B radiation intensity for the fitness of sea urchins (at least *S. intermedius*). Gonads were significantly more sensitive to UV-B radiation than the gut. The present study increases our understanding of carryover effects of UV-B radiations on sea urchins and provides valuable information into marine environmental safety.

1. Introduction

Anthropogenic gases induced ozone depletion greatly increased the transmission of solar ultraviolet-B radiation (UV-B, 280–315 nm) through the atmosphere (Day and Neale, 2002). Empirical evidence indicates the impacts of UV-B radiation on marine organisms in intertidal and shallow seas (reviewed by Lamare et al., 2011). Most studies investigated real-time effects of UV-B on behaviors (for example, Sigg et al., 2007), cellular responses (for example, Lu and Wu, 2005), DNA damage (for example, Mitchell et al., 2009), reproduction (for example, Kane and Pomory, 2001) and development (for example, Bonaventura et al., 2006) of marine organisms. However, marine organisms which exposed to UV-B radiation cannot recover immediately from the negative impacts (for example, DNA damage) (Mitchell et al., 2009; Newman et al., 2018). This indicates that UV-B radiation probably has carryover effects on the fitness of marine organisms despite of the absence of UV-B radiation for a period of time. To our knowledge, however, few investigations have documented this. This lack probably leads to an underestimate on the adverse effects and/or an acclimation of UV-B radiation on marine invertebrates in intertidal and shallow seas. Full understanding of carryover effects of UV-B radiation on marine invertebrates can provide valuable information into marine environmental safety.

The sea urchin *Strongylocentrotus intermedius* is an ecologically important marine invertebrate in structuring marine benthic communities in intertidal and shallow seas (Agatsuma, 2013; Zhadan et al., 2017). Further, *S. intermedius* has a number of fitness related traits that can be investigated. Thus, *S. intermedius* is an ideal research model to investigate effects of short-term UV-B radiation on fitness of marine invertebrates. We previously studied behavioral responses of *S. intermedius* to short-term UV-B radiation and found that *S. intermedius* were behaviorally sensitive to short-term UV-B radiation (Shi et al., in press). Consequently, we were encouraged to investigate whether short-term UV-B radiation affects other fitness related traits of sea urchin (for example, survival, growth and food consumption), despite of the absence of UV-B radiation for a long-term period (for example, 8 weeks).

The main aim of the present study is to investigate fitness related traits of *S. intermedius* 8 weeks after exposure to short-term (1 h) UV-B radiation. We asked 1) whether short-term UV-B radiations have significant carryover effects on survival, behavior, food consumption, growth and tissue development of *S. intermedius*; 2) whether different intensities of short-term UV-B radiation have significantly different carryover effects on fitness related traits of *S. intermedius*; 3) which organs are especially sensitive to short-term exposure to UV-B radiations and which are not.

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<https://doi.org/10.1016/j.ecoenv.2018.08.078>

Received 14 June 2018; Received in revised form 18 August 2018; Accepted 21 August 2018

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2. Materials and methods

2.1. Sea urchins and experimental design

The present study is an extension of our previous study (Shi et al., in press). The source of sea urchins and experimental design were fully described in Shi et al. (in press) and briefly explained as follows:

Juvenile *S. intermedius* were transported from the hatchery of Dalian Haibao Fishery Company to the Key Laboratory of Mariculture and Stock Enhancement in North China's Sea, Ministry of Agriculture and Rural Affairs at Dalian Ocean University. The sea urchins were cultured at the density of $\sim 5 \text{ kg m}^{-3}$ in a tank (length \times width \times height: $180 \times 100 \times 80 \text{ cm}$) with aerated seawater until the experiment. They were fed wild fresh *Saccharina japonica* and *Ulva lactuca ad libitum* under natural photoperiod. The seawater was changed every three days.

Sea urchins were exposed to UV-B exposure for one hour at 0, 10 and $20 \mu\text{W cm}^{-2}$ using a UV-B lamp (280–315 nm, TL 40 W/12 RS, Philips Co., Germany). The intensities of UV-B radiation (10 and $20 \mu\text{W cm}^{-2}$) were set by regulating the distance between UV-B lamp and the surface of the water (Adams, 2001). The control group was set as $0 \mu\text{W cm}^{-2}$. Environmental UV-B radiation was not involved, because the UV-B exposures were carried out in an isolated box with a UV-B lamp.

Strongylocentrotus intermedius were put into three randomly distributed cages (length \times width \times height: $75 \times 43 \times 43 \text{ cm}$) in a tank (1 t in volume) after each UV-B radiation (0, 10 and $20 \mu\text{W cm}^{-2}$), with 20 individuals in each cage. All sea urchins were cultured in a consistent environment at laboratory temperature with very little UV-B radiation from solar radiation ($0\text{--}0.5 \mu\text{W cm}^{-2}$) for 8 weeks. The seawater was changed every three days. Water temperature ranged from 21.4°C to 25.8°C .

2.2. Survival

The number of dead urchins was recorded during the experiment in each cage for all the three groups. Number of survivors was calculated by the number of sea urchins minus the number of dead individuals in each cage ($N = 3$).

2.3. Food consumption

Strongylocentrotus intermedius were fed *ad libitum* with locally collected fresh *S. japonica*. Food consumption was measured for the first 12 days after UV-B exposure. It was calculated as the amount of food provided minus the uneaten food amount in each cage ($N = 3$) (Zhao et al., 2013).

2.4. Righting response time

Righting response time of ten *S. intermedius* was individually measured after the 8 weeks experiment in a tank (length \times width \times height: $60 \times 35 \times 20 \text{ cm}$). Sea urchins were gently placed onto the bottom of the tank with their aboral side down. Righting response time was the time required for *S. intermedius* to right themselves with aboral side up within 10 min. The righting response time was recorded as 600 s if *S. intermedius* did not right themselves within 10 min.

The average of the ten individuals was considered as the righting response time for the cage ($N = 3$).

2.5. Body size and tissue measurements

Body size and tissue measurements were made at the end of the experiment after the methods of our previous study (Zhao et al., 2016a), which were briefly described as follows:

Test diameter, test height and lantern length were measured by a

digital vernier calipers (Mahr Co., Germany). Body, test, lantern, gonads and gut were weighed using an electric balance (G&G Co., USA). Test height:test diameter ratio was calculated. Test thickness was measured using a digital vernier caliper according to the method of Byrne et al. (2014).

All live *S. intermedius* were crushed to measure the crushing force value using a pressure testing instrument (Shijin Co. Jinan, China). The crushing force value is the pressure necessary to break tested objects, which was recorded by the load measurement system of the instrument. Tests were placed with the oral surface down between the two steel plates. The speed of the plate was set as 50 mm min^{-1} . One of the two steel plates automatically applied pressure to the tests until they broke. The average value of 15 individuals was considered as the trait value for the cage ($N = 3$).

2.6. Crude protein of gonads and gut

Crude protein of gonads and gut were measured using the method of semi-micro Kjeldahl nitrogen (Chen et al., 2008; Zhao et al., 2016b). Gonads and gut of 15 individuals were mixed as one sample for the cage, respectively ($N = 3$).

2.7. Statistical analysis

The data were analyzed for homogeneity of variance and normal distribution. One-way ANOVA was used to analyze all experimental traits except food consumption. Food consumption was analyzed using one-way repeated measured ANOVA. Pairwise multiple comparisons were carried out using LSD test when significant difference was found in ANOVAs. All statistical analysis was performed using SPSS 16.0 statistical software. A probability level of $P < 0.05$ was considered as significant.

3. Results

3.1. Survival

UV-B radiation at $20 \mu\text{W cm}^{-2}$ significantly reduced survival of *S. intermedius* compared to those exposed to UV-B radiation at 0 and $10 \mu\text{W cm}^{-2}$ ($P = 0.04$, Fig. 1). However, there was no significant difference in number of surviving sea urchins between the individuals exposed to 0 and $10 \mu\text{W cm}^{-2}$ ($P = 0.482$).

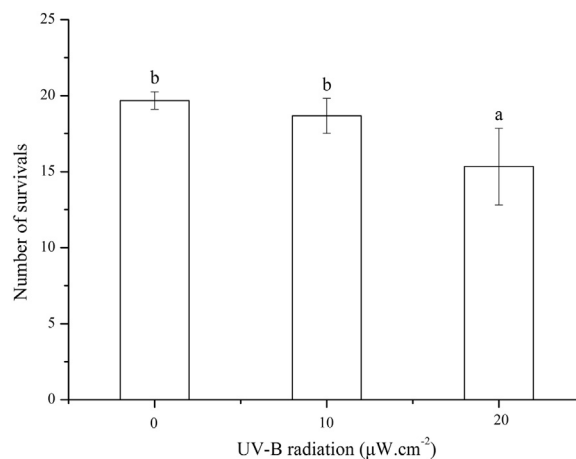


Fig. 1. Number of surviving sea urchins at different intensities of UV-B radiation ($N = 3$, mean \pm SD). Letters above the bars represent significant difference.

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