SHORT-TERM RESPONSES OF SOME PLANKTONIC CRUSTACEA EXPOSED TO ENHANCED UV-B RADIATION

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ABSTRACT

With shrimp and crab larvae, several-day exposures to UV-B radiation below lethal threshold levels of dose-rate and total dose had no significant effects on either activity or development (molting). Above those UV levels, activity, development, and survival rapidly declined. The specimens in those experiments were held in flow-through seawater containers of 8 cm depth. Perhaps these near-surface zooplankton in nature could increase their depth slightly, if they could sense fatal dose-rates or doses of UV-B. With such a short-term response they might completely avoid damaging solar UV.

Experiments in paired 2-1 glass cylinders (38 cm water depth) suggested that there is no difference in behavior between irradiated and non-irradiated shrimp larvae <u>(Pandalus platyceros)</u> or copepods <u>(Epilabidocera longipedata)</u>. The irradiated specimens maintained their near-surface positions (as did the controls) until the time of decreased activity and death at about 4 days. The progress and outcome were similar in experiments using a 44-1 glass cylinder (140 cm water depth).

The zoea stage of the shore crab <u>Hemigrapsus nudus</u> is extremely attracted to strong visible light. No differences in response-time (seconds) was noted between control larvae and larvae receiving lethal UV-B doses until decreased activity and death of the irradiated larvae at about 10 days. There was no apparent reluctance of the crab larvae to swim toward and to hold themselves within lethal doses of UV-B. Investigations of the biological effects of UV-B radiation on aquatic organisms have included a variety of approaches, ranging from histological analyses to simple survival studies. Most of this work has been concerned with the direct effects of natural or enhanced UV-B radiation. The assumption implicit in some of the conclusions is that the organisms which live in the near-surface layer will be passive recipients of ambient UV-B radiation, regardless of its intensity.

Because some of these near-surface organisms appear to be living near their UV tolerance limits (Damkaer et al., 1980), it is possible they have evolved a sensitivity to fluctuations in UV-B radiation. In addition to photorepair mechanisms they may possess an ability to avoid harmful levels of UV-B by appropriate defensive behavior. In moderately productive ocean waters (0.5 mg Chl a m^{-3}), the DNA-weighted irradiance at 1 m is only about 40% of incident (Smith and Baker, 1979). Under these conditions, the capacity to sense harmful UV-B and to avoid it by simply swimming or sinking a meter below the surface would provide considerable protection.

Only recently have attempts been made to determine the behavioral response of aquatic organisms to UV-B. For small, primarily benthic crustaceans, there is some evidence that correlates horizontal positioning behavior with resistance to UV-B (Barcelo and Calkins, 1979). That is, sensitive organisms (shown by low survival) tended to avoid UV radiation more than did the more-tolerant organisms.

However, even if sensory mechanisms exist to warn an organism of dangerous UV-B radiation, the particular stimulus which elicits the avoidance response may not be effective under a new spectral distribution caused by ozone depletion. With a reduction in the ozone layer the UV-B band would

increase disproportionately to UV-A and visible light, and an avoidance response linked to high levels of visible irradiance, for example, might not be activated quickly enough. It seems likely that only a sensory system which directly detects irradiance in the UV-B range and does so before irreparable damage occurs will afford a surface-living organism a safety potential.

The present short-term response experiments supplement the previous observations of UV-effects on shrimp and crab larvae (Damkaer et al., 1980). The 8 cm depth **of** the flow-through water-table containers could have restricted vertical movement if test organisms attempted to avoid exposure to the UV-B irradiation. However, examination of the containers during exposures always showed the healthy shrimp and crab larvae near the surface until UV-B damage (when it occurred) curtailed their activity. Afforded a greater vertical range in the present study, these larval forms still behaved in a similar fashion.

<u>Methods</u>

Experiments and measurements were conducted at the National Marine Fisheries Service/NOAA Aquaculture Laboratory at Clam Bay, Manchester, Washington, directly across Puget Sound from Seattle. Seawater for all experiments was pumped directly from the bay, sand-filtered, and partially sterilized with germicidal UV. Simulation of ambient and enhanced solar irradiance, measurements of spectral irradiance, and the water tables used in response-time studies were as described by Damkaer et al. (1980). Control organisms in all experiments were irradiated by one FS-40 sunlamp and one cool-white lamp, both filtered by a clear Mylar © plastic sheet (0.25 mm thickness). The duration of all light exposures was 3 h each day, centered around solar noon.

Adult copepods <u>Epilabidocera longipedata</u> were collected at mid-day from surface swarms in Clam Bay. Egg-bearing females of the shore crab <u>Hemigrapsus</u> <u>nudus</u> and the shrimp <u>Pandalus platyceros</u> were held in tanks until the eggs hatched. All shrimp and crab larvae were less than 24 h old at the beginning of experiments.

One set of experiments consisted of paired 2-1 glass cylinders each filled to a depth of 38 cm and containing 10 specimens. The cylinders were nearly submerged in an aquarium with continuously flowing seawater. The bottom, sides, and top of the aquarium were covered with opaque black plastic sheeting, except for an opening directly above each cylinder. The light sources were placed directly above the cylinders. A glass side of the tank could be uncovered at intervals during experiments, permitting direct observation of the cylinders with the test organisms. Visible markings divided the cylinders vertically into thirds, and the number of organisms within each division (upper, middle, lower) was recorded at five times: in relative darkness (before turning on lamps) and at 30, 60, 120, and 180 min after lamps had been turned on. Seawater in the cylinders was changed daily.

A dose-rate of 0.018 $_{Wm-2[DNA]}$ was administered for 3 h·d-¹ to shrimp larvae <u>(Pandalus platyceros)</u> and adult copepods <u>(Epilabidocera longipedata)</u> in the 2-1 glass cylinders. This dose-rate is comparable to the noon irradiance of a clear summer day and is well above irradiance levels shrimp larvae are likely to encounter during their season of surface occurrence in spring. Moreover, this dose-rate as well as the daily dose far exceed the threshold values established for shrimp larvae (Damkaer et al., in press). The threshold values for <u>Epilabidocera longipedata</u> have yet to be determined. This copepod is often found at the surface in full spring and summer sunlight.

To further investigate behavioral responses to potentially damaging UV-B and to complement initial findings, a larger cylinder was constructed which allowed even greater freedom of movement of the test organisms. This clear plastic cylinder, 165 cm high and 20 cm in diameter, was filled to a depth of 140 cm, at which level it contained about 44 1 of seawater. Because only one large cylinder was available, UV-B treatment and control phases of these experiments were consecutive, with different sets of organisms, rather than concurrent. The cylinder, containing 50 specimens, was in a darkened room where the only source of light was the UV or control lamp-combinations suspended above the opening at the top of the cylinder. The cylinder was marked at depths of 30 cm, 60 cm, and 90 cm, and the vertical distributions of test organisms were recorded before turning on the lamps and at 30, 60, 120, and 180 min after UV exposure began. Seawater in this cylinder was slowly drained and replenished once each day.

We also considered response-time as a possibly sensitive index of exposure to UV radiation. Newly hatched zoea larvae of the shore crab <u>Hemi-</u><u>Grapsus nudus</u> were placed in 1,000-ml beakers which were held in plastic baskets in the water table. Ten specimens were placed in each of ten beakers, with five beakers receiving UV-B and five beakers as controls receiving no UV-B. Daily 3 h light exposures were administered while the animals were in the water table. Each day specimens were removed from the beakers and placed in a small petri dish (6 cm diameter) which was covered to eliminate light for 5 minutes. In a darkened room, the cover was removed, a 60-W incandescent light 30 cm to one side of the dish was turned on and a stop-watch activated. When 60% of the specimens had actively responded by swimming to the light edge of the dish, the watch was stopped and the elapsed time recorded. The animals were returned to the beakers which had been cleaned and

refilled with seawater, and the beakers were again placed in the water-table baskets.

UV-B irradiance (285-315 nm) is reported in DNA-weighted effective doses. These values were calculated using the analytical representation of Green and Miller (1975) which is based on the DNA action spectrum (Setlow, 1974).

<u>Results</u>

Throughout the first day's exposure the control copepods and the copepods receiving a high daily dose of UV-B (194 at a high dose-rate Jm-2[DNA]) (0.018)tended to swarm at the surface of the small cylinders Wm-2[DNA]) (Fig. 1). During the next two days the general condition of the copepods receiving the UV-B deteriorated (including mortalities), although specimens which remained active continued to move toward the surface during the UV-B (with visible light) exposure. All control organisms were active and 40-50% were always in the top third of the cylinder during light treatments. Even control organisms which were at times in the bottom third of the cylinder remained active and mobile. During the final light treatment on the fourth day, the effects of the UV-B radiation were apparent. All of the irradiated copepods were either moribund (50%) or dead (50%) and thus were on the cylinder bottom. The control copepods, however, continued to be active and 30-50% were within the upper two-thirds of the cylinder at all times (Fig. 1).

An experiment of similar design with <u>Pandalus platyceros</u> led to equivalent results. Before and during the first day's 3 h exposure in the small cylinders there was no apparent difference between the vertical positioning of the shrimp larvae receiving damaging doses of UV-B irradiance and the response of the control shrimp. The vertical distributions were almost uniform with depth in both cylinders before the lights were turned on. After 30 min of exposure





Fig. 1. Vertical distributions of the copepod <u>E ilabidocera</u> <u>longipedata</u> in paired glass cylinders (water depth 38 cm), during 3-h exposures with and without UV-B radiation. and when observations were made at 60, 120, and 180 min the greatest number of shrimp larvae were consistently found in the upper third of each cylinder.

Before the second day's 3 h exposure all shrimp larvae were in the bottom third of each cylinder. When the lamps were turned on some of the larvae in each cylinder immediately swam to the surface. Throughout the remainder of that light exposure the vertical distributions of the larvae in the two treatments were similar, with the larvae most often congregating in the top third of the cylinders.

By the third day, shrimp larvae receiving the high doses of UV-B became far less active than the control organisms, with at least 80% of the UV-B irradiated larvae inactive and on the bottom of the cylinder. In contrast, some control larvae were always in the upper third, with at least 50% within the upper two-thirds during exposure. During the fourth and last day of exposure, the UV-B irradiated larvae were lying moribund on the bottom of the cylinder. In the control cylinder, all larvae were still active and 40-50% were in the upper two-thirds of the cylinder during light treatment.

In the first of two experiments with the large cylinder, shrimp larvae were irradiated with 0.0047 for 3 h. This dose-rate is more than $_{Wm-2[DNA]}$ for 3 h. This dose-rate is more than twice the dose-rate threshold for shrimp larvae (Damkaer et al., in press). In both the UV-B treatment and the control phases the shrimp larvae were concentrated in the upper half of the cylinder throughout the 3 h exposure, with the maximum abundance generally in the upper 30 cm (Fig. 2).

Adult Epilabidocera longipedata were also placed in the large cylinder and exposed to UV-B (0.0047 Wm-2[DNA]). As in the other large-cylinder experiments, the control phase without UV-B was run afterwards with a second group of copepods. The response during the first day's 3 h exposure was nearly identical for each phase (Fig. 3). No avoidance of this harmful level



Larval Panda/us p/atyceros

Fig. 2. Vertical distributions of shrimp larvae <u>Pandalus platyceros</u> in a plastic cylinder (water depth 140 cm), during the first day's 3-h exposure with and without UV-B radiation.



Fig. 3. Vertical distributions of the copepod <u>Epilabidocera longipedata</u> in a plastic cylinder (water depth 140 cm)j during 3-h exposures with and without UV-B radiation.

of UV-B irradiance was observed. During the second 3 h exposure (Day 2), the vertical distributions for the UV-B treatment and the control were again similar, and while the concentration of copepods in the upper 30 cm of the cylinder was somewhat reduced from the previous day, the majority of the organisms remained in the upper half of the cylinder throughout the 3 h period. On Day 3 the damaging effect of UV-B became evident. While the vertical distribution of the control animals was similar to that of the previous day and all of the control animals were active, the copepods receiving a third exposure of UV-B were now concentrated and inactive in the bottom half of the cylinder.

The zoea stage of the shore crab <u>Hemigrapsus</u> <u>nudus</u> was selected as an experimental organism because of its rapid positive response to strong visible light. Zoeae were first timed within hours of hatching and then before each day's exposure. The test organisms received $0.0073_{Wm-2[DNA]}$ for 3 h per day, which is more than twice the biologically effective threshold dose-rate for the zoea stage of the crab <u>Cancer magister</u> (Damkaer et al., in press).

There was very little difference between the mean response times of the UV-B irradiated test organisms and the control organisms for the first 5 days (Fig. 4). However, by Day 9, the mean response time for the control groups was almost half that of the test organisms. The physical condition of the test organisms deteriorated rapidly from this point in the experiment, and by Day 12 the UV-B irradiated larvae were far less responsive than the control larvae. After Day 12, the UV-B irradiated larvae were virtually dead, while the controls remained active with excellent survival.

In this experiment the <u>Hemigrapsus</u> <u>nuclus</u>larvae continued to seek out strong visible light (while being timed) long after receiving lethal total doses of UV-B irradiance. The stimulus which elicited the timed response included no UV-B, but observations of the larvae while being irradiated



Fig. 4. Visible-light response time in seconds for zoea larvae of the shore crab <u>Hemigrapsus nudus</u>; closed circles are UV-B irradiated groups, open circles are controls without UV-B exposure; large circles are means, small circles are ranges.

with UV-B in the water table indicate that as long as the larvae are active they mostly remain at or near the surface.

<u>Discussion</u>

Each of these experiments suggests that there were no differences in behavior between UV-B irradiated organisms and the control organisms until lethal doses of radiation reduced their activity. This is not considered to be an active response to UV, and it would have no value in avoidance. The apparent inability to perceive potential danger from UV-B occurred at doserates well above established laboratory thresholds and at exposures to total doses which, in most experiments, surpassed lethal total dose thresholds for similar animals (Damkaer et al., in press). If the animals tested in this study possess a behavioral mechanism for protection from dangerous levels of irradiation it seems unlikely that it could be based on the direct sensing of UV-B intensity. The shrimp and crab larvae and the adult copepods tested here seemed to be attracted to wavelengths longer than in the UV-B range, and the additional exposure to high levels of UV-B irradiance did not alter their short-term behavior. Within the limits examined, these animals generally positioned themselves as near the light source as possible. That they continue to seek out a strong light source even while doomed from past UV-B exposures demonstrates not only the strength of this photo-positive response but also, probably, their inability to independently discriminate between safe and dangerous levels of UV-B irradiance.

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Figure Captions

- Fig. 1. Vertical distributions of the copepod <u>Epilabidocera longipedata</u> in paired glass cylinders (water depth 38 cm), during 3-h exposures with and without UV-B radiation.
- Fig. 2. Vertical distributions of shrimp larvae <u>Pandalus platyceros</u>in a plastic cylinder (water depth 140 cm), during the first day's 3-h exposure with and without UV-B radiation.
- Fig. 3. Vertical distributions of the copepod <u>Epilabidocera longipedata</u> in a plastic cylinder (water depth 140 cm), during 3-h exposures with and without UV-B radiation.
- Fig. 4. Visible-light response time in seconds for zoea larvae of the shore crab <u>Hemigrapsus nudus;</u> closed circles are UV-B irradiated groups, open circles are controls without UV-B exposure; large circles are means, small circles are ranges.