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Survival of the Hermit Crab, *Clibanarius vittatus*, Exposed to Selenium and Other Environmental Factors

Virginia Wolfenberger

Texas Chiropractic College, 5912 Spencer Highway, Pasadena, TX 77505

Recent investigations of water quality criteria have frequently examined the effects of a pollutant; however, a more realistic investigation would consider effects of multiple environmental factors and their interactions with the pollutant. Awareness of selenium as a pollutant is increasing. The growing sulfur and petroleum industries are only two of the potential sources of the element on the Texas coast. Geologically, selenium occurs widely and can enter the environment via the weathering of rocks and soils (NAS 1976). The element may also enter the environment as a result of the combustion of fossil fuels (Fleming et at. 1979; Bertine & Goldberg 1971; Gutenmann and Bache 1976). It can also be a runoff contaminant from soil dressing containing selenium in selenium deficient areas (Gissel-Nielson and Gissel-Nielson 1973). Since selenium tends to accumulate in sediments (Sidelnikova 1970), dredging operations might also increase available selenium in the marine environment.

Chemically, selenium is a metalloid, element number thirty-four, whose behavior resembles that of sulfur, with which it is often associated. Of the common oxidation states, 0,+4, and +6, Chau and Riley (1965) found Se^{+4} to be the most probable ion in the marine environment.

Physiologically, selenium is widely acknowledged as an essential micro-nutrient for most animals (Rosenfeld & Beath 1964). It is a component of the ubiquitous enzyme glutathione peroxidase (Flohe et al. 1973), and has been implicated in non-heme iron proteins (Lucy et al. 1970) and the electron transport chain (Levander et al. 1973).

This study examined the toxicity of selenium to the hermit crab *Clibanarius vittatus* (Bosc) under twelve different combinations of temperature and salinity. Additionally, the impact of the organisms' original environment was considered as an environmental factor.

MATERIALS AND METHODS

Clibanarius vittatus were collected from two sites near Port Aransas, Texas. One site, Steadman's Island, is a bay environment to the south of the causeway connecting Aransas Pass and Port Aransas. The second site is on the southwestern shore of Corpus Christi Pass through Mustang Island into Corpus Christi Bay. This area is exposed to Gulf waters, temperatures and salinities.

Experimental conditions included four salinities, three temperatures, and the presence or absence of a pollutant, selenium (Table 1). Preparation of animals from each site for experimentation included 96 h acclimation to salinities of 10, 20, 30 and 40 parts per thousand (ppt) with six to ten animals per 4L aquarium. An airstone was placed in each container. These aquaria were kept in an incubator at 16°C, 20°C, or 24°C (all temperatures are ± 1°C) for at least 96 h prior to having any experimental tests performed. These temperatures and salinities are within the ranges for the Texas coast. A 12-12 h light-dark cycle was maintained. Animals subjected to similar conditions were exposed to the pollutant selenium in the form of sodium selenite. To yield the 100 parts per million (ppm) exposure concentration of selenium, 0.65714 grams (g) of the compound were added to 3L of sea water at the appropriate salinity. The pollutant was added by dissolving the salt in the 3L of sea water 72 h after acclimation was begun so that the exposure time to the pollutant was 24 h. Sodium selenite was selected as the pollutant compound because it is more likely to be found in the marine environment than is a selenate (Chau & Riley 1965). The loss of selenium from the system was assumed to be negligible (Glickstein 1979). However, that assumption was verified using ^{75}Se. All temperatures and salinities as well as aquaria that contained animals and aquaria that did not were tested for loss of selenium. To 3L of sea water, 0.65714 g of Na_2SeO_3 were added. It was agitated and 1milliliter (mL) of $Na_2{}^{75}SeO_3$ solution added. Immediately five 1-mL samples were taken from each aquarium and stored in air tight vials. 24 h later a second set of 1-mL samples were taken. Both sets were then counted using a Beckman Gamma 4000 gamma spectrometer. The difference in the two counts was determined and percentage loss to the systems calculated.

An analysis of variance was performed using the following first order model:

$$Y = \mu + A + B + C + AB + AC + BC + ABC + E_{ij}$$

where Y is the survival time, A, B and C are the linear effects of salinity, temperature and site respectively; AB, AC and BC are the linear interactions of salinity and temperature, salinity and site, temperature and site; ABC is the linear by linear by linear interaction of temperature, site and salinity. μ is the intercept and E_{ij} is the error term (Ott 1977).

RESULTS AND DISCUSSION

After 24 h there was no significant loss of selenium from the systems employed in this study under any of the combinations of temperature and salinity as determined using both an F-test and a student's t-test.

The means of the survival times of Clibanarius vittatus exposed to 100 ppm selenium are graphed by site and temperature at the four salinities tested (Fig. 1). At 16°C, the animals from Corpus Christi Pass survived longer in 20 ppt salinity than at 10, 30 and 40 ppt, the salinity of the shortest survival time being 40 ppt. The animals from Steadman's Island at 16°C also survived longest in 20 ppt with survival at 10 ppt being only slightly lower; the mean survival times in 30 and 40 ppt were the same (104 h).

Table 1. Summation of treatments to which whole animals and animals from which tissues were taken were subjected. Salinities are in parts per thousand. Temperatures are in degrees Centigrade, selenium concentration is either 100 parts per million or 0 parts per million.

	STEADMAN'S ISLAND				CORPUS CHRISTI PASS			
Salinity / Temp.	10	20	30	40	10	20	30	40
16	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm
20	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm
24	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm	100 ppm / 0 ppm

At 20°C, the patterns of survival for crabs from the two sites were similar at the three lower salinities, the longevity of the animals being greater in 10 ppt, decreasing in 20 ppt and increasing in 30 ppt. However, in 40 ppt, the survival time of the animals from Corpus Christi Pass increased while it decreased for those from Steadman's Island. In general, animals from Steadman's Island survived longer at 16 and 20°C and for shorter times at 24°C than did those from Corpus Christi Pass.

When the survival times were divided by either wet or dry animal weights, the magnitude of the longevity patterns changed, but the patterns themselves were virtually unaltered.

An analysis of variance indicated that all single factors and their interactions, except that of salinity and site, were significantly influential at the $\alpha = .05$ level on the survival times in the experiment.

The lack of loss of selenium from the systems is substantiated by Glickstein (1979), who found no loss using similar methods. The large quantity of added selenium made detection of the loss of such small amounts as might be incorporated into animal tissue impossible, since the percentage of selenium lost from the system to animal tissue would be smaller than the percentage error using the technique employed.

The toxicity of selenium was diminished at 20°C and 24°C in 10 ppt salinity for both sites. The lower osmotic pressure inside the cells possibly enabled the hermit crabs to exclude the pollutant-solute from their bodies at the low salinity. The lower temperature, 16°C, may have caused the loss of this protective mechanism, at least for the smaller animals from Corpus Christi Pass. That there was a difference attributable to temperature is further supported by the decrease in longevity of animals from both sites at 20°C and 20 ppt salinity.

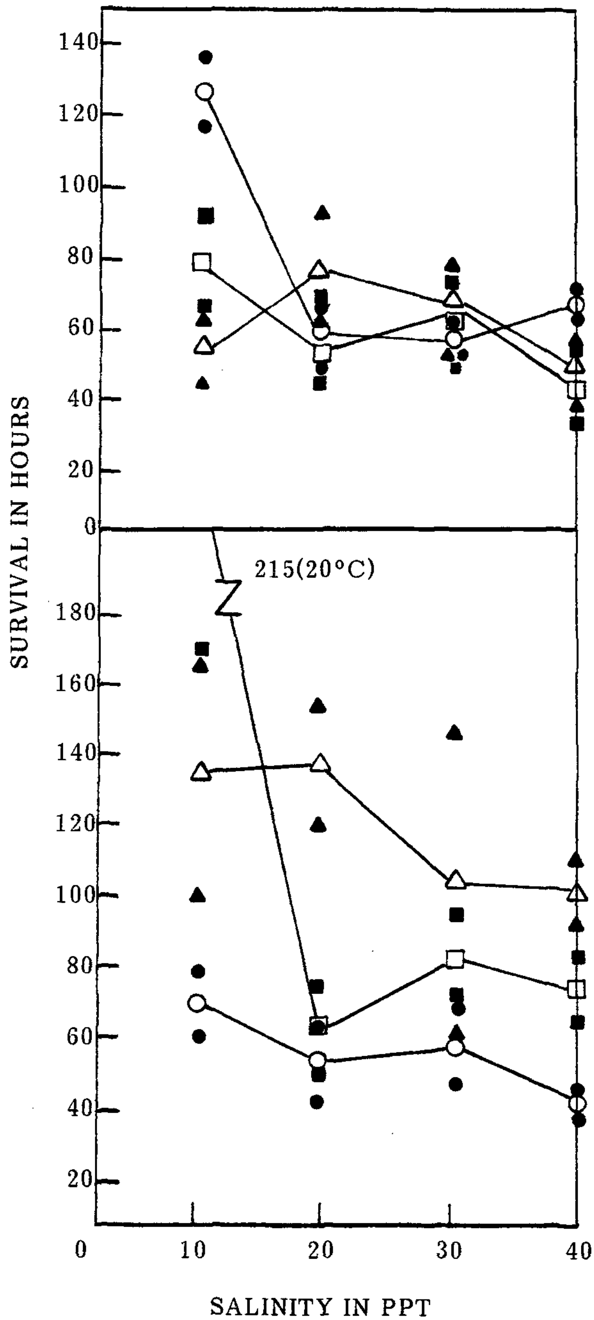

Figure 1. Survival time of Clibanarius vittatus from Corpus Christi Pass (top) and Steadman's Island (bottom) after exposure to 100 ppm selenium. Temperatures are 16°C,△; 20°C, □ ; and 24°C, ○ ; standard errors are represented by ▲ , ■ , and ● respectively.

For both sites, however, 16°C and 20 ppt salinity were the conditions of maximum survival. Perhaps this was due to a lower metabolic rate and, also, a slower rate of selenium accumulation at the lower temperature. There is apparently an interactive effect of temperature with salinity and selenium in the 30 ppt salinity experimental conditions; at the two higher temperatures, survival times were longer than at 16°C. There were some differences that apparently were attributable to the effect of site.

Throughout the experiment, dead animals frequently were noted to have orange-colored material deposited in blood vessels, in the gills, in the pericardial sinus and other less consistent locations in the body. Since reduced selenium is orange, it seems probable that the animals were reducing the Se^{+4} and depositing the element in some manner in the circulatory system.

The interactions of temperature with salinity were noticeable. There is only one instance in which 20°C and 24°C did not follow the same trend in survival times, with 16°C taking the opposite tack. It seems then that, generally, there was an interaction of temperature and salinity. The fact that the survival times of animals from Steadman's Island were noticeably longer than for animals from Corpus Christi Pass was probably because the Island animals were larger. When the hours survived during exposure to selenium were divided by the wet weights, the survival times for the animals from the two sites were not noticeably different, and when the survival times were divided by dry weights, the animals from Corpus Christi Pass were shown to live longer per unit weight. The decrease in the surface area to volume ratio would allow for less accumulation of selenium in the body per unit of weight in a given period of time. This would account for the greater longevity of larger animals.

Interaction of environmental parameters with a pollutant should definitely be considered in establishing pollution guidelines. Further evaluation of interactions and synergistic effects is needed.

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