

ABSTRACT

Cellular, histopathological and molecular responses of *Corbula caribea* (Mollusca, Bivalvia) from a natural oil seep, to field and/or experimental exposure to polycyclic aromatic hydrocarbons

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Corbula caribea (Mollusca, Bivalvia) is the most common non-polychaete macrofaunal organism identified at a large natural oil seep at La Brea in south Trinidad. It is hypothesized that chronic exposure to potentially toxic contaminants at the seep has stimulated these organisms to develop adaptive mechanisms that allow them to tolerate these conditions.

The concentration of polycyclic aromatic hydrocarbons (PAH) was determined in water and sediment samples collected from 10 stations in the vicinity of the seep. Eleven PAHs were identified in water samples during the November 1995 sampling period. The total concentration of PAH ranged between 517 and 2301 $\mu\text{g/L}$ [API (American Petroleum Institute) #34 Trinidad crude oil equivalents]. In sediment samples the concentrations of total PAH ranged from 18.5 to 101 $\mu\text{g/g}$ (API #34 Trinidad crude oil equivalents) dry weight of sediments. The PAH levels in water were significantly higher than the maximum of 300 ng/L recommended by the USEPA (1992) for the protection of marine life.

Animals from the seep were assayed in a closed continuous flow system with each of four PAH, typical of the seep site (naphthalene, acenaphthene,

phenanthrene and dibenzothiophene), at concentrations up to 10 times the levels detected in sediment samples. No mortality was detected following 72-hour exposures. Two of the main histopathological changes evident following exposure to toxicant concentrations greater than the levels detected at the seep were metaplasia of stomach epithelia and atrophy of intestinal epithelia. However, these responses were not evident in animals examined from the field. Using stereological methods, it was possible to quantify cellular responses at different toxicant concentrations. Within the stomach, normalized metaplastic responses showed an exponential increase with increasing toxicant concentration. Within the gills and palps mobilization of adipogranular cells increased with increasing toxicant concentration. Animals from the seep also showed high concentrations of adipogranular cells in the palps and gills. A metric which we have termed the 'Cellular Response Index' (the ratio of spaces to adipogranular cells within the vascular junction of the palps), enabled us to relate the degree of cellular mobilization to the ambient toxicant concentration.

It is possible that these animals may possess innate homeostatic mechanisms that allow them to ameliorate the potentially deleterious effects of contaminants at the seep and at exposure concentrations that reflect field levels. However, at concentrations that exceed these levels the neutralizing capacity of these mechanisms is exceeded and toxicant induced responses are manifest.

Subsequently, investigations were conducted to determine if molecular responses might be responsible for the apparently enhanced tolerance of *Corbula caribea* within its natural environment. Investigations focused on components of

the mixed function oxygenase (MFO) system (cytochrome P-450, cytochrome b₅ and NADPH-ferrihemoprotein reductase) which are reportedly involved in the metabolic detoxification of xenobiotics. Microsomal extracts prepared from animals collected at the seep showed no cytochrome P-450 or cytochrome b₅ activity. However, NADPH-ferrihemoprotein reductase activity was demonstrated using serial cryostat sections. In seep animals this activity proved to be significantly higher than in animals from two non-seep reference sites. At the latter sites, total PAH levels were significantly lower than at the seep site. Hence, it is likely that the ability of *Corbula caribea* from the seep to tolerate high levels of PAH may be due to the presence of this adaptive mechanism.

Key words: Natural oil seep; histopathology; stereology; polycyclic aromatic hydrocarbons; NADPH-ferrihemoprotein reductase.