# EFFECT OF EXPOSURE TO HARBOUR DREDGE SPOILS ON IMMUNE CAPABILITY IN THE COMMON SHRIMP, CRANGON CRANGON

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## Introduction

Dredge spoil disposal constitutes a significant pathway through which industrially-derived contaminants may be mobilized into aquatic environments. Traditionally, the procedures used to evaluate the effect of sediment contamination depend upon observation of a variety of responses in a range of indicator species. To date, these tests have not included evaluation of immunocompetence, despite the importance of immune capability as a determinant of species health and, hence, ecosystem stability. The shrimp, Crangon crangon, is a common epibenthic scavenger with a wide geographical distribution and is a key link in the food chain of temperate waters. Previous work has established that sub-acute exposure of this animal to PCB congeners results in a reduction in the number of circulating haemocytes, an increase in recoverable haemolymph volume (indicative of impaired clotting) and a decline in phenoloxidase activity within the haemocytes (Smith & Johnston, 1992). These haematological parameters therefore represent sensitive indicators of environmentally-induced immunosuppression in marine crustaceans. The present study was directed at examining the effect of exposure to contaminated harbour dredge spoils on immune capability in C. crangon.

## Methods

Specimens of the common shrimp, C.crangon were collected by beam trawl from the Dutch Wadden Sea and exposed to dredge spoils for 12 days at 16± 2°C. Each tank contained 0.3m$^3$ of sediment comprising either clean sand (the reference tank, designated tank A), dredged spoils from Rotterdam harbour (tank B) or a mixture of 95% sand plus 5% harbour dredgings (tank C). The sediment in each tank was overlaid with water from the main mesocosm ponds at IBN-DLO, Texel (Vethaak, 1993). Duplicate tanks were set up for each treatment. Groups of ca 20 C. crangon were sampled, and the recoverable haemolymph volume (RHV), total haemocyte count (THC) and blood cell phenoloxidase activity (PO) determined as described in Smith & Johnston (1992). As a further measure of defence capability, antibacterial activity towards the marine bacterium, Psychrobacter immobilis (NCIMB 308), was also assessed in vitro using the procedure of Chisholm & Smith (1992). Antibacterial activity was calculated as the percentage killing of bacteria over nine hours. To ensure that variations in these parameters were not due to diurnal or other endogenous rhythms, sampling was repeated at least three times on different occasions using specimens taken from each of the duplicate treatment tanks.

## Results

Table 1 shows that while the mean RHV from animals housed in the control tank A was similar to that of animals incubated in tank B, specimens incubated in the dredgings-sand mix (tank C) yielded a higher RHV. A corresponding reduction in the mean THC was observed following exposure of the host animals to the dredge spoils, with again strongest effect seen in tank C animals. These changes were observed consistently irrespective of the time of day. Regarding PO activation, enzyme activity was found to be depressed in samples prepared from animals in tank B compared to both controls (tank A) and intermediate animals (tank C). This indicates a reduction either in the size of the granular population containing PO or a reduction in the amount of enzyme per cell. The increase in RHV, coupled with the decline in the THC and change in PO activity in the tank B animals, is interpreted as indicative of impaired clotting.

Since the circulating blood cells are the main mediators of host defence in crustaceans and play a central role in processes such as phagocytosis, encapsulation, clotting and the release of antimicrobial factors/agents (Smith & Chisholm, 1991; Smith, 1991), a reduction in the cell count would severely impair immune capability of the host animal. Surprisingly, no differences were seen in the ability of the various HLS samples to reduce the viable count of P. immobilis. In every

case, the HLS samples significantly reduced the viable count over nine hours incubation (Table 1). While this indicates that antibacterial vigour (per unit protein) by C. crangon haemocytes is unaffected by exposure to harbour dredge spoils, the reduction in the number of circulating haemocytes which contain the antibacterial agents means that, overall, antibacterial capability is impaired in the experimental animals.

**Table 1. Haemotoxic effect of exposure to harbour dredge spoils in C. crangon**

| | control (tank A) | 100% dredge spoil (tank B) | 5% dredge spoil + 95% sand (tank C) | [1]n |
|---|---|---|---|---|
| Recoverable haemolymph vol (RHV) ($\mu$l) | [2]26.7 ± 1.4 | 28.3 ± 1.4 | 34.2 ± 1.9 | 60 |
| Total haemocyte count (THC) ($x10^6$ $ml^{-1}$) | 2.5 ± 0.2 | 1.7 ± 0.2 | 0.9 ± 0.1 | 60 |
| [3]Blood cell phenoloxidase activity | 16.6 ± 4.3 | 8.8 ± 2.4 | 14.1 ± 4.6 | 5 |
| % bacterial killing after 9h | 79.0 ± 6.0 | 73.0 ± 17 | 70.0 ± 13 | 5 |

[1] number of samples examined
[2] all values are means ± SE
[3] change in absorbance ($\Delta A_{490}$) $min^{-1}$ mg $protein^{-1}$ after trypsin treatment

## Conclusions

This study has revealed that subtle changes in haemocytic parameters in C. crangon are brought about by short term exposure to contaminated sediments. In particular, with respect to THC and clotting, a conventional exposure/response function was not found in relation to the contamination gradient used. The reason for this is unclear, although, possibly, the nature of the sediment in the experimental tanks may have influenced the amount or composition of contaminants experienced by the host animals. Alternatively, there may have been higher rates of mortality in the containing 100% dredge spoils, leading to pre-selection of a more robust population of C. crangon in this tank than in tank C. The finding that sediments containing as little as 5% dredge spoils have a marked effect on immune capability in this crustacean could have serious implications for the dumping of toxic wastes at sea.

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