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Exposure and response of aquacultured oysters, *Crassostrea gigas*, to marine contaminants

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Abstract

Comparative growth rates were monitored in the Pacific oyster, *Crassostrea gigas*, at two sites in Singapore, one uncontaminated and the other contaminated with respect to ambient seawater quality. Growth rates differed significantly at the two sites, revealing that marine water quality can have potentially adverse effects for the oyster aquaculture industry in Singapore. Shell abnormalities (chambering) were observed for juvenile and mature oysters at the contaminated site. Water quality parameters including temperature, salinity, dissolved oxygen, total organic carbon and chlorophyll-*a* were essentially similar at both sites. Differences in the levels of tributyl tin detected in soft tissues were not observed, but significant differences in the burden of persistent organic pollutants (POPs) existed between the two sites. On a positive note, the effects of pollution on oysters were found to be reversible, where transplantation of individuals to the uncontaminated site resulted in the ability of *C. gigas* to recover in terms of growth rate and the burden of bioaccumulated POPs.

Keywords: Oysters; Aquaculture; Persistent organic pollutants; Chambering effect; South-east Asia

1. Introduction

Due to the increasing global human population and widespread depletion of natural fish stocks, the world is placing increasingly dependence on aquaculture to meet its food requirements. However, marine pollution represents a potential risk to the aquaculture industry in nearshore coastal waters as seafood exposed to prevailing contaminants, even at trace levels, may be rendered as unfit for human consumption. Recent scientific reports on contamination of salmon tissue with a range of persistent organic pollutants (POPs) have highlighted the human health risks associated with the consumption of tainted aquaculture products (Hites et al., 2004). Potential human health effects resulting from exposure to POPs include developmental and neural effects, immunotoxicity and teratogenicity (UNEP, 2003; Vallack et al., 1998). Such studies have generated a vociferous debate, and precipitated in an immediate, negative impact on the farmed salmon aquaculture business. Additionally, marine contaminants can present a threat to aquaculture productivity by impairing growth yields and the health of organisms (McDowell Capuzzo, 1996; Sugawara and Okoshi, 1993). Although scientific studies have been conducted under controlled laboratory conditions to evaluate the uptake of pollutants, there is very limited information on the accumulation of contaminants and their impact in nearshore aquaculture systems.

The Pacific oyster, *Crassostrea gigas*, is cultured in the Asia-Pacific Region, but also in Europe, America and Africa. World production of *C. gigas* reached 4.1 millions of tons in 2001, with 85% of the total produced in China (FAO, 2001). As a filter feeder, *C. gigas* has been used as a bioindicator for anthropogenic contaminants (Gunther et al., 1999; Hunter et al., 1995; McDowell Capuzzo, 1996) and is known to accumulate POPs to levels similar, or greater, than mussels (Gunther et al., 1999). However,

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high mortality rates have often been observed in bioaccumulation monitoring studies using transplanted organisms, which undermines the value of the data collected with respect to consumption-related health risks (Gunther et al., 1999). Bioassays using Pacific oyster embryo larvae are routinely used to monitor the occurrence of water-borne pollutants (Nice et al., 2000). Recently, *C. gigas* has been proposed as a model organism to study the effect of pollutants, such as PCBs, on the immunological system of bivalves (Gagnaire et al., 2006).

Total aquaculture production in Singapore has a value of approximately US$74 million (2004 data) (http://www.ava.gov.sg). There remains considerable potential for further development of the country's aquaculture industry, but this aspiration must be balanced with worldwide concerns over food security and product quality. In a previous study, POPs, including polychlorinated biphenyls (PCBs), polybrominated diphenyl ethers (PBDEs) and organochlorine pesticides (OCPs) were detected in Singapore's natural marine biota (Bayen et al., 2003, 2005a). A recent assessment of the mean daily intake of POPs from seafood amongst the general population in Singapore has also revealed potential health risks (Bayen et al., 2005b, c). On this basis, there is a justifiable need for enhanced monitoring and investigation of the fate and uptake of POPs in nearshore aquaculture systems.

In this study, the comparative growth rates of *C. gigas* was measured at two sites in Singapore, one deemed as 'uncontaminated' and the other 'contaminated' with respect to ambient seawater quality. POPs bioaccumulation in *C. gigas* was monitored continuously from the juvenile stage until organisms reached their market size in order to evaluate potential health risks of shellfish consumption. POPs analysed included the PCBs, DDT pesticides, chlordanes and the PBDEs. The study also investigated the effect of transplanting mature organisms between the uncontaminated and contaminated sites to determine the response of oysters to contaminant exposure and their ability to recover. Information on the reversibility of adverse effects induced by anthropogenic contaminant bioaccumulation is lacking (McDowell Capuzzo, 1996) and this study represents, to our knowledge, the first of its kind based on oysters cultivated for human consumption.

2. Experimental

2.1. Experimental design

In this study, the growth of oysters was compared at two sites in Singapore's marine environment between July 2003 and May 2004. The first site is located near busy shipping lanes, and the second site on a more open, coastal site. The sites were chosen for their marked difference in prevailing levels of POP contamination, as described in previous studies (Bayen et al., 2003, 2004a, b).

C. gigas 'seeds' were imported from Australia at an average size of 6 ± 1 mm and acclimatized for 3 weeks at the uncontaminated site. At the end of this period, the survival rate was $>95\%$, and 1000 oysters were then transferred to the contaminated site (day 0); whilst another 1000 oysters remained in the uncontaminated site. Oysters were grown in cages suspended at a constant depth (1 m) for the duration of the experiment, i.e. 230 days. The deployment site remained constant all over the experiment. Maintenance included monthly cleaning of the cage to remove fouling organisms and size-grading of the oysters. At the initial stage of growth (size <20 mm), oysters seeds were grown in nylon bags (2 mm mesh) within the cage. As the shell size increased, oysters were transferred to bags of a larger mesh size (8 mm) and finally to the cage itself (15 mm mesh). The number of oysters per cage was gradually reduced over the remaining period of the experiment, in accordance with aquaculture practices, so to avoid overcrowding and excessive competition between individuals. The number of surviving oysters, and their size, at each site was recorded monthly. As explained later, strong differences were observed between the two sites in terms of oyster growth and POPs contaminant load. As a result, after 126 days, half of the remaining oysters from each site were transferred to the other site, and were then compared to the remaining half, in order to study the organism's response to a changed environment with respect to growth rate and tissue contaminant burden.

2.2. Water quality

Temperature, salinity, dissolved oxygen and total organic carbon are monitored monthly at both sites by the Marine Port Authority of Singapore (private communication). Levels of POPs in seawater have also been monitored monthly at the same sites by co-workers (Wurl and Obbard, 2005).

2.3. Sample collection and preparation

Each month, between October 2003 (day 0) and May 2004 (day 230), 100 oysters were collected from each site for analysis. Samples were separated into distinct class sizes for analysis (i.e. <25, 25–45, 45–65 and >65 mm) and transported to the laboratory in polyethylene bags in ice-boxes. To prepare samples for chemical analysis, soft tissues were removed from the shell and homogenized in a stainless steel blender. The size, total weight and weight of the soft tissues were recorded for each organism. Homogenized samples were then frozen at $-20\,^{\circ}$C prior to analysis.

2.4. Chemical analysis

Oyster samples were analysed for POPs at day 0, 126 and 230. All organic solvents used were of pesticide residue analytical grade. PBDE congeners 47, 99 and 100