Contamination of organotin compounds and endocrine disruption in gastropods

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the coastal waters. At the end of 20th century humankind consider that their impacts on the hydrosphere are greater than its capacity. In the 21st century, they must preserve the environment of hydrosphere from their industrial and agricultural activities and live together with the hydrosphere free from pollution for the healthful co-existence.

The word “pollution” generally means the environmental damage caused by wastes discharged into the hydrosphere, the occurrence of wastes in the hydrosphere or the wastes themselves. Therefore, it is necessary to define it correctly for scientific discussion. The “inputs” are used for wastes in the sea (hydrosphere), the “contamination” is for the occurrence of wastes in the sea and the “pollution” is for the damaging effects of the wastes.

Classification of pollution is very useful for understanding degree of impacts by the pollution. It can be classified into four categories on a basis of duration of impact according to Prof. A. Meinez at the Université de Nice-Sophia Antipolis.

■ Short-term pollution that disappears after stopping causes. It corresponds to a period within ten years.
■ Middle-term pollution that is caused by most of chemicals non-biodegradable and active on fauna and flora. It corresponds to a period from several decades. The author classifies non-biodegradable chemicals to irreversible pollution due to persistancy in the hydrosphere.
■ Long-term pollution that are related to ecosystem destruction which needs long time for its reconstruction. It corresponds to a period of a hundred years.
■ Point of no return, irreversible pollution, that is definitive destruction of species or ecosystem. It corresponds to a period more than several centuries.

When the causes of pollution are not excluded, even short-term pollution continues as middle-term pollution. The concept is based on the duration after stopping or eliminating causes of pollution. Short-term to long-term pollution are reversible (non-conservative), and the other is irreversible (conservative) pollution. Irreversible pollution means that pollutants are accumulated and not decomposed for several hundred years or more. Prof. A. Meinez proposed that reclamation is irreversible pollution because it is very difficult to recover reclaimed coast to original one and the reclaimed area exits quasi-perpetually. The irreversible pollution must be avoided for sustainable development. On the other hand, reversible pollution means that pollutants can be decomposed or diluted by natural force. A representative example is oil spill pollution. Pollution can disappear in ten or twenty years after the oil spill accident. This kind pollution can be controlled by appropriate policy such as removing source of inputs or improving treatment of discharged water. In our presentation, I introduce some examples of these classification.

To understand present situation of coastal waters, monitoring is important. Obtained knowledge serve to ameliorate its environment and predict future conditions. Since pollution is transboundary, monitoring must be conducted in international frames. Most of pollution is caused by human activities on land. Water and ecosystem quality in the hydrosphere depends on comprehensive management along course of water flow from precipitation to the sea through mountains, rivers and lakes. I present some results of monitoring coastal waters including seagrass beds which are very important for marine ecosystem and environments.

Contamination of organotin compounds and endocrine disruption in gastropods

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Imposen, a superimposition of male genital organs (penis and vas deferens) on female gastropods, occurs cause specific and at low concentrations of certain organotins such as tributyltin (TBT) and triphenyltin (TPhT) [1–6]. Reproductive failure is known at severely affected stages [7, 8]. Since the first report from Plymouth, England in 1969, imposex have been reported from all over the world involving more than 140 species [9–11]. Among them, the main cause of population declines in at least seven species is considered to be reproductive failure related to imposex [12]. In this paper, we will report the present status of imposex of gastropods and organotin pollution in Japan, together with endocrine disruption observed in abalones.

Imposen in the rock shell (Thais clavigeru) and organotin pollution in Japan

Among rock shell samples collected between September 1996 and January 1999 from 93 locations along the Japanese coast, imposex were observed in 86 locations whereas no or rare cases were found in the remaining seven locations. The percentages occurrence of imposex were as high as or close to 100% at 73 locations. It is expected that spawning obstruction occurs in more than half the population of females when the Relative Penis Length (RPL) index exceeds 40, based on the relationships of RPL index, the Vas Deferens Sequence (VDS) index and the percentage occurrence of oviduct (vulva) blockage in females. Among the 93 locations, RPL index values exceeded 40 were found at 40 locations. Compared to the results of the survey conducted last time, the index values
slightly decreased or remained almost unchanged in some locations. TPhT concentrations in tissue of the rock shell showed decrease with time but varied distinctively with locations, and relatively high pollution remained in a few locations was detected. Decreases in TBT concentrations were not so distinctive and decrease degrees were low compared to TPhT. Changes with time were not observed in some locations near marinas and dockyards.

Ovarian dysmaturity accompanied with imposex may have brought about the decreased catches in the ivory shell (Babylonia japonica). Occurrence of imposex, decreases in catches, and in the amount of spawning from parent snails at seed producing facilities have been recognized in the ivory shell. In contrast to the rock shell, however, oviduct blockage by vas deferens formation is not observed in the ivory shell, so physical suppression of spawning is unlikely. It was suspected that decreases in catches were caused by ovarian insufficiency associated with imposex. Gonad tissue preparations were obtained by a usual method from a total of 135 individuals of ivory shells sampled every month between December 1988 and November 1989 in Prefecture A. The results of microscopy showed a distinctive reproductive cycle (peak maturity in summer) in males (43 specimens) but not in females (92 specimens). Females did not show prominent maturity and reproductive cycle like males even in summer. In addition to the suppressed ovarian maturity, spermatogenesis in ovaries was observed in six female specimens (five imposex and one normal). It can be inferred that suppressed ovarian maturity included spermatogenesis in some cases appeared as dysfunction of ovaries and this might cause the reduction of spawning volume. Interestingly, remarkable accumulation of TPhT was observed in ovary, and the penis length was positively correlated with triorganotin concentration (total concentration of TBT and TPhT) in ovary of imposex-exhibited females.

**Occurrence of imposex in other marine mesogastropods and neogastropods in Japan**

Imposon was also observed in Alabaster False Tun, Galeocerds leucodoma, (Order Mesogastropoda, Family Ooctrythidae) trawled from 200–250 m in depth off the Atsumi Peninsula in 1999 [12]. Among the 69 species of Japanese marine gastropods studied including the rock shell and the ivory shell, a total of 39 species (7 from Mesogastropoda and 32 from Neogastropoda) were observed to be affected by imposex. Although imposex has been observed mostly in shallow-water species in the previous surveys, we need to conduct detailed studies on species living at depths of 200 m or more because of our latest finding of imposex in Alabaster False Tun.

**Endocrine disruption in abalones**

The catches of abalones in Japan have been decreasing since 1970s. Various studies have been conducted to pursue the cause of the decrease but still unresolved. With a hypothesis that organotins (TBT and TPhT) cause reproductive insufficiency in abalones, we have studied this subject continuously since 1994. Our findings up to now are outlined here [13]. Based on the results of a preliminary survey started in 1994, the gonadal maturity of the giant abalone (Haliotis madaka) was histologically studied, with area B selected as a reference area, which was considered to be normal in view of catches and other characteristics, and area C selected as an affected area, where decreases in catches were prominent and the ratio of artificially raised and released individuals among the total catch was very high (i.e. reproductibility was very low in the natural abalone population). The results showed that both sexes of giant abalones in area B became sexually mature between the late fall and the early winter whereas those in area C did not become sexually mature at the same time. In area C, the males showed several peaks of sexual maturity in a year but the sexual maturity of females varied widely among individuals and the average level of maturity in the female population was suppressed. While few hermaphrodites, evidenced by ovarian spermatogenesis, were found in area B, 11 (20.4%) hermaphrodites out of 54 specimens were observed in area C [13].

Because abalones fertilize externally by releasing sperm and eggs in seawater, it is very important for both sexes to become sexually mature at the same time in order to heighten fertility. Their fertility may drop if there are many immature individuals in either sex or if the peaks of sexual maturity do not coincide between both sexes. Because abalones are known to be dioecious, hermaphrodites (masculinization of females) of about 20% found in area C is abnormal. This is considered to be a masculinization phenomenon similar to the imposex observed in the rock shell and the ivory shell. Interestingly, determined values of organotins in tissues of the giant abalone from both areas revealed significantly higher concentrations in area C than area B [13].

An in situ exposure test was conducted, using caged abalone (Haliotis gigantea), transplanted from area B to a location close to a dockyard in area C (giant abalones could not be used for the test because of difficulty in obtaining a sufficient number of individuals). The test was carried out for seven months from June 1998 and we collected about 40 specimens in January 1999. Gonad tissue preparations of individuals were observed under a microscope, and masculinization, such as ovarian spermatogenesis, was observed in 15 female specimens (88.2%) out of 17. Also, significant accumulation of organotins during the test period was observed. No ovarian spermatogenesis was observed in the control specimens from area B. The microscopic features of the observed ovarian spermatogenesis are the same as those found in the ivory shell suffering from imposex. Ovarian spermatogenesis and testicular development associated with imposex have been found also in the rock shell and other neogastropod species. The findings in our study indicate that masculinization (ovarian spermatogenesis) essentially equal to imposex occurs in abalones as an endocrine disruption phenomenon, without development of external genital organs such as a penis. We infer that some environmental factor, most probably organotins, caused ovarian spermatogenesis in abalones around the dockyard in area C [13].

References

Endocrine disrupting chemicals in the Canadian aquatic environment

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The presence of endocrine disrupting chemicals (EDCs) in the environment has recently become a high profile international issue. Field studies have shown that the growth, reproduction and development of many species including invertebrates, fish, birds and mammals, may have been altered by chemicals that interact with the endocrine system. Many of these chemicals have shown the ability to cause adverse biological effects at very low levels commonly found in the environment that may have been previously considered safe. Although reproduction and development have been, and continue to be, a major endpoint for assessment of chemicals by Environment Canada, the EDC issue has heightened our concerns for detecting effects mediated through the endocrine system. This presentation will review the scientific evidence for endocrine related alterations in wildlife in the Canadian aquatic environment. A few of the conclusive studies will be examined in detail demonstrating their direct links between chemical exposure and alterations in endocrine function. Our studies of the impacts of exposure of fish to pulp and paper mill effluents will be examined, as over the last ten years we have developed a number of endocrine techniques in order to evaluate the mechanisms responsible for the reproductive effects demonstrated in wild fish. A description of studies examining the correlation between declines in Lake Trout populations in Lake Ontario and sediment levels of 2,3,7,8 tetrachlorodibenzo-p-dioxin will also be made. Other Canadian studies of contaminant effects where direct links to endocrine dysfunction have yet to be demonstrated will also be discussed including studies examining the effects of municipal sewage wastes, heavy metals, pesticides, insecticides and other forms of agricultural runoff, refinery discharges, steel mill effluents and tributyltin.

Following this presentation of examples of endocrine disruption in the Canadian aquatic environment, Environment Canada's national EDC strategy will be discussed. It focuses the activities and research initiatives of the Department on determining the extent to which the Canadian environment is impacted by endocrine disrupting chemicals. Environment Canada is giving priority to assessing those sites and sectors, which have been identified as having the highest potential for adverse effects on growth, reproduction or development. These include both those sites previously identified, such as pulp and paper mill effluents, and those emerging issues such as intensive agriculture (pesticides and animal wastes), urban effluents (especially sewage) and priority substances which have been shown or are suspected of having effects on the endocrine system. A new Federal Government initiative has earmarked research funds for studying the effects of endocrine disruptors. A summary of the research being conducted through the Toxic Substances Research Initiative will also be made.

A description of Canada's new Environmental Effects Monitoring Program (EEM) will also be discussed. The EEM program was first initiated in 1992 for the pulp and paper industry in Canada, which requires industry to monitor the effects of their effluents on the receiving environment including those on fish and benthic invertebrates. This program is cyclical in nature (every three years) and is designed to evaluate whether the existing effluent regulations are protecting the Canadian receiving environment. The EEM program has been expanded into the metal mining industry, which will conduct its first cycle in the year of 2002. A brief description of my studies at Environment Canada relating to both endocrine disruptors and the EEM program will also be presented.