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The impact of organotin pollution on aquatic invertebrate communities—are molluscs the only group whose populations have been affected?

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Abstract

This review explores the impact of pollutants on aquatic wildlife populations employing the case of tributyltin (TBT), a widely used antifouling paint ingredient banned globally in 2008. Attention has focused primarily on TBT's effects on populations of mollusc species, but this review shows that many other species were affected to a greater or lesser extent, including crustaceans, tunicates and sponges. Marine invertebrate communities were seriously impoverished and are still recovering. TBT was marketed long before biocides were systematically evaluated for their adverse environmental effects, and it is concluded that modern chemical regulatory programmes would likely detect a substance with similar properties. However, environmental monitoring programmes might well miss a TBT lookalike and should be modernised to focus more on the detection of biological effects.

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Introduction

The purpose of this short, nonexhaustive, review is to consider what is known about the population-level impacts of tributyltin (TBT) on aquatic wildlife, a biocide widely used in antifouling paints until a global ban in 2008. Although the well-known endocrine disrupting effects of TBT in molluscs (imposex and related conditions) are touched on, the main focus of the article concerns other invertebrate fauna. The overall intention is to highlight a clear example where an environmental contaminant has not only damaged reproduction in exposed organisms but thereby has caused serious and widespread effects on wildlife populations and communities.

TBT began to be used in antifouling paints in the early 1960s at a time when biocides were not evaluated for their potential effects on the environment, and it dominated the market by the 1980s although the total global tonnage was small (2000–3000 t per annum) because of its high toxicity to fouling organisms. Imposex in molluscs caused by TBT was not detected by the limited routine environmental monitoring programmes that existed at the time but was initially reported as a curiosity [1] and only later linked by ecotoxicologists to TBT leaching from shipping [2]. The environmental data on TBT were thus primarily generated by one-off research programmes, although much of this research was originally stimulated by the evident damage to the mariculture industry and was ultimately funded by governments.

Impacts of organotins on mollusc populations

This topic has been thoroughly addressed in many books, papers and reviews (e.g. Refs. [3–11]), so will only be briefly summarised here. A huge body of research revealed as early as the 1970s that TBT leaching from antifouling paints on the hulls of boats and ships was damaging the reproductive capacity of many gastropods and bivalves by a form of masculinisation now thought to be caused, at least partly, by interaction with the retinoid-X receptor (RXR) [12]. Over 150 species of gastropods worldwide developed an irreversible condition known as imposex or intersex in which females developed masculine characteristics and became infertile. Breeding in many bivalves was also damaged, and the shellfish cultivation industry suffered severe losses.

A few examples of these effects are sufficient to illustrate the issue. First, the causal relationship of TBT exposure with imposex is well established in both the laboratory and the field. Thus, for example, Bettin et al. [13] showed that low levels of TBT (5–100 ng tin/L)

produced imposex in two species of neogastropods (the dogwhelk *Nucella lapillus* and netted whelk *Nassarius reticulatus*) over exposure periods of 2–5 months. Wirzinger et al. [14] demonstrated not only that imposex in free-living *N. reticulatus* declined with distance from a point source (a harbour) in Brittany but also that this was well correlated with TBT concentrations in water and whelk body tissues. *N. lapillus* populations experienced imposex around most of the European coastline, again in association with shipping activity, and have been showing clear imposex reductions since TBT levels began to decline e.g. [15–18]. Imposex led to the complete obliteration of some dogwhelk and other gastropod populations [15] and severe declines in others [e.g. 17, 18]. A related condition known as intersex was found to be damaging reproduction in the winkle *Littorina littorea* [19], and some estuaries had very low proportions of young snails at the time when TBT concentrations were high [20].

Many bivalve species also experienced TBT-related reproductive problems which affected their populations. For example, Langston et al. [18] showed that TBT-contaminated populations of the peppery furrow shell *Scrobicularia plana* were severely depressed or eliminated in several UK estuaries, some only beginning to recover from about 1994 or even later. A semicontrolled field study in the Crouch estuary [21–24] found that several bivalve species including *Ostrea edulis* and *Mytilus edulis* became more abundant as TBT concentrations declined after a local ban. In addition, TBT-related shell thickening in farmed Pacific oysters *Crassostrea gigas* e.g. [25] made it impossible for the mariculture industry to function in many estuaries, and *C. gigas* population abundances were affected in France [6].

These impacts on mollusc populations were some of the most devastating effects of chemical pollution ever seen in aquatic wildlife and drew much of the scientific attention. However, the next section will show that other invertebrate populations were also being damaged.

Possible impacts of organotins on other aquatic invertebrates

A brief summary of the main field studies of TBT's impact on aquatic invertebrate populations is given in Table 1.

TBT, tributyltin

Semicontrolled field studies

Using the concept of pollution-induced community tolerance (PICT), Blanck and Dahl [26] showed that TBT had exerted strong selection pressure on periphyton communities near a marina before a local ban on TBT-based paints. There has also been a microcosm experiment in which an artificial seagrass community dosed weekly with a high concentration of TBT (initially 22.2 μg TBT/L) for 6 weeks showed declines in several invertebrate population abundances, including two mollusc species, an amphipod crustacean and a polychaete annelid [27]. However, the most valuable data emerged from two so-called natural experiments.

The first of these took place in San Diego Bay, California, in 1987, when the use of TBT was widespread [28]. This study compared the hard-bottom invertebrate communities living on the pontoons of floating docks located in areas where either many or few small boats were moored. Respective boat densities were 2025–3580/km^2 and 0–904/km^2. It was found that the invertebrate communities living in areas with many boats were extremely impoverished, being dominated largely by serpulid polychaetes and a solitary tunicate (*Ciona intestinalis*), plus large amounts of filamentous algae. In areas with few boats, the encrusting communities were much more diverse, with large numbers of mussels, sponges, bryozoans, crustaceans and tunicates. Total numbers of motile species were 18–23 where boats were sparse, and only 6–16 where boats were numerous. Respective numbers of sessile species were 14–19 and 3–9.

These major community differences could not be explained in terms of hydrographic factors, but areas of densely moored boats in so-called back bay sites contained much more TBT in the water (160–400 μg/L) than similar areas with few boats (16–86 μg TBT/L). It was tentatively concluded that TBT may have been a causative factor, but it is possible that other contaminants could have been responsible.

The second natural experiment took place in the Crouch estuary, UK, from 1987 to 2005, a period when TBT contamination was steeply declining because of a local ban on the use of TBT-based antifoulants on small vessels [21–24]. There were few other TBT inputs during this period as large vessels rarely used the estuary, and other pollutant inputs were absent except for some small sewage treatment works discharges. TBT concentrations in sediment at the most contaminated (upstream) station were 0.16 mg/kg dry wt. (1987), 0.12 mg/kg (1988), 0.1 mg/kg (1990), 0.04 mg/kg (1991), <0.01 mg/kg (1997) and 0.022 mg/kg (2005). Both benthic in-faunal and epi-faunal communities were surveyed at each of five stations along the estuary in 6 years between 1987 and 2005.

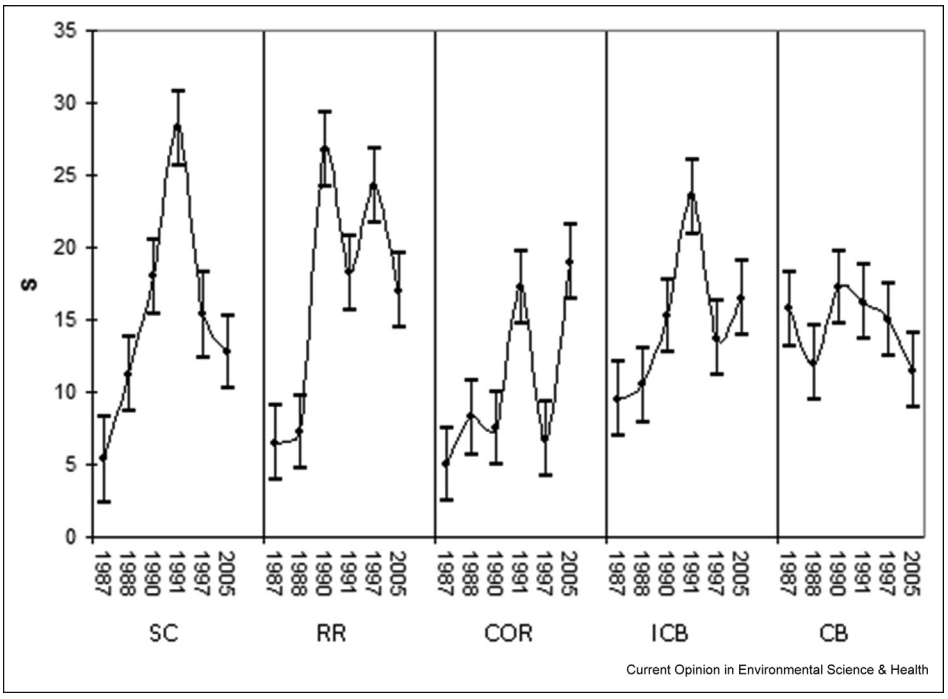
Total numbers of epifaunal taxa per trawl at the 3 most contaminated upstream stations increased from 21–30 (mean = 25.2), in 1987/88, to 27–44 (mean = 32.8) in 1992/97, an average 30% increase. Mean numbers of infaunal species at the same stations increased from 5–6 in 1987 to 13–19 in 2005 (Figure 1). In both cases, the

Table 1

A summary of the main field studies of TBT's impact on aquatic invertebrate populations, including a subjective assessment of the strength of the cause–effect relationships.

Effects studied	Summary of main observations	Strength of cause–effect relationship	References
Marine mollusc populations worldwide	• Imposex or intersex observed in >150 marine gastropod species, leading to collapse of some populations • Population declines and collapses in some marine bivalves • Severe shell thickening and poor reproduction in farmed bivalves	High: The association of these effects with TBT is firmly established and replicated	• *e.g.* [14–20] • *e.g.* [6; 25] • *e.g.* [18; 21–24]
Invertebrate populations in San Diego Bay, US	• Hard-bottom invertebrate communities were impoverished in areas of dense boating (2025–3580 boats/km^2) compared with sparse boating reference areas (0–904 boats/km^2) • Motile species numbers were 6–16 in dense boating areas and 18–23 in the reference areas • Sessile species numbers were 3–9 and 14–19, respectively • The main affected groups were bivalve molluscs, sponges, bryozoans, crustaceans and tunicates	Medium: A single year's study with no follow-up. Some potential for other causes.	[28]
Invertebrate populations in the Crouch estuary, UK	• Describes increases in numbers of benthic invertebrate species after a local TBT ban on small boats in 1987 • TBT concentrations at the most contaminated, upstream, station decreased from 0.16 mg/kg dry wt. in 1987 to <0.01–0.022 mg/kg in 1997–2005 • At the 3 most contaminated stations, mean numbers of epi-faunal taxa increased from 25.2 to 32.8 per trawl between 1987/88 and 1992/97 • At the same 3 stations, mean numbers of in-faunal species increased from 5 to 6 in 1987 to 13–19 in 2005 • The main affected groups were molluscs, annelids, crustaceans and tunicates	High: There were almost no continued TBT inputs from large vessels, and other polluting inputs were small. A consistent time trend of recovery occurred simultaneously with TBT declines	[21–24]
Brown shrimp (*Crangon crangon*) fishery in the Southern Bight, North Sea	• TBT levels in *C. crangon* tail muscle decreased approximately 10-fold to 4–124 μg/kg dry wt. in the 6 years after the international TBT ban in 2003 • Mean shrimp landings per unit effort declined from 0.14 kg wet wt. × trawler horsepower^{-1} × fishing hours^{-1} in 1973, to 0.06 in 2002, and then recovered to 0.18 in 2010	Low: There were several potential influences on the shrimp population in addition to TBT, including weather and disease, although changes in predatory fish species were ruled out as a cause	[29–31]
Macrophyte populations in the Norfolk Broads, UK	• Extensive loss of macrophytes was coincident in time with high levels of TBT in the water (0.9 μg/L) • Microfossils from that period suggest that TBT reduced populations of gastropods grazing on periphyton and zooplankton grazing on phytoplankton • It is postulated that this would have severely limited light influx, causing the macrophytes to die off	Medium: There are no direct measurements of the affected invertebrate populations, or of changes in light levels, but the indirect evidence is persuasive	[40]

Figure 1



Temporal trends in in-faunal invertebrate species numbers (S) at five stations on the Crouch estuary from 1987 to 2005 (±least significant intervals; on the basis of pooled variance). TBT concentrations became progressively lower on the transect from the upper estuary at Stow Creek (SC) to the outer estuary at the Crouch Buoy (CB). RR, Raypits Reach; COR, Corinthian Yacht Club; ICB, Inner Crouch Buoy. [24].

biggest increases in species numbers occurred between 1987 and 1991, when TBT levels in sediment were decreasing most rapidly. As well as the expected resurgences in mollusc populations, numbers of polychaete annelid, crustacean and tunicate species also increased significantly.

It seems likely that these large increases in invertebrate species numbers in the Crouch estuary were at least partly associated with the decreases in TBT contamination as the two variables simultaneously altered rapidly, and no other major polluting inputs or habitat alterations were known to occur.

Brown shrimp fisheries

Verhaegen et al. [29] advanced the hypothesis that the decline in North Sea brown shrimp (*Crangon crangon*) landings between 1973 and 2002 was at least partly because of organotins. Mean landings per unit effort (LPUE – kg wet wt. × trawler horsepower^{-1} × fishing hours^{-1}) in the Southern Bight declined from 0.14 in 1973 to 0.06 in 2002, and then bounced back to 0.18 in 2010. They showed that shrimp were able to accumulate TBT from contaminated sediments because of high biota–sediment bioaccumulation factors (BSAF), leading to TBT levels in tail muscle of 4–-124 µg/kg dry weight in southern North Sea estuaries such as the Scheldt in 2009.Six years earlier, TBT concentrations in shrimp from the same area were approximately 10-fold higher, so the international ban on new TBT-based paint applications which began in 2003 had clearly led to a big reduction in the contamination of shrimp.

This, of course, is not proof that TBT was responsible for the shrimp population crash, although the coincidence in time with the TBT ban may be significant, and alternative explanations such as variations in predatory fish populations or weather were ruled out by Siegel et al.[30]. The hypothesis that TBT was responsible is reinforced by the fact that eventual recovery of shrimp stocks in the highly TBT-contaminated Scheldt estuary lagged about 10 years behind that in the less-contaminated Waddenzee [31]. The possibility that TBT had indeed interfered with brown shrimp breeding is perhaps given additional weight by the work of Verhaegen et al. [32] who showed that female brown shrimp exposed in the laboratory to acutely toxic levels of TBT for 96 h experienced strongly reduced expression of ecdysteroid and retinoid-X receptors in their ovaries which could in turn damage reproduction. However, no data are available on whether observed environmental levels of TBT could also cause such effects.

There are also some doubts raised about the putative linkage between TBT and *C. crangon* population decline

in the southern North Sea when one considers the long-term study of the Crouch estuary, UK, between 1987 (when TBT contamination in sediments was high) and 2005 (when TBT had declined below detection limits) [33]. This showed that *C. crangon* populations at the originally most contaminated stations *declined* over the study period by factors of 35–56.

In summary, the hypothesis that TBT contributed to a population decline in the North Sea brown shrimp population is not proven, although it remains a possibility due to the known sensitivity of larval crustaceans to TBT (see below).

Other crustaceans

Both of the natural experiments described above suggested that some crustacean populations were probably affected by TBT. Smith et al. [33] reported that *Palaemon elegans* and *Crangon allmanni* in the Crouch estuary became relatively abundant by 2005 compared with earlier years, although the reverse was the case for *Pandalus montegui* and *C. crangon*. *Macropodia rostrata* also showed a clear increase in abundance over this period. The numbers of in-faunal crustacean species at the most contaminated upstream station increased from 0 in 1987 to 6 in 1990/1991, whereas equivalent numbers of epifaunal species doubled from 5 to 10. However, it should be noted that species numbers had declined somewhat by 2005. Likewise, Lenihan et al. [28] showed that the numbers of crustacean species per scrape sample increased from 3 to 9 in areas of San Diego Bay with dense boating activity to 11–13 where boats were rare.

Some crustaceans are known to be almost as sensitive to TBT as molluscs. For example, larval development of the copepod *Acartia tonsa* is chronically affected at only 1 ng TBT/L [34]. More recently, Kim et al. [35] have shown that newly hatched juveniles of a marine mysid experience reductions in 20-hydroxyecdysone titres and growth at a 28 d lowest observed effect concentration (LOEC) of 5 ng TBT/L, thus corroborating the findings of Verhaegen et al. [32] about a possible toxic mechanism in crustaceans.

Although the field data could potentially be explained in other ways, the high sensitivity of crustaceans in the laboratory combined with a plausible mechanism of action suggests that TBT was probably responsible for at least some of these population impacts.

Tunicates, sponges and bryozoans

Lenihan et al. [28] reported that percentage cover of the substrate by sponges and bryozoans in San Diego Bay was significantly greater in areas where boats were numerous and TBT concentrations high in 1987. Percentage cover by tunicates was not affected, but the biomass of solitary tunicates increased from 0.6 to 8.2 g/sample in boating areas to 15.9–36.1 g/sample in the relative absence of boats. Overall, the numbers of sessile invertebrate species rose from 3–9 to 14–19 when boats were few. Smith et al. [33] did not find significant effects among the bryozoans or sponges in the recovering Crouch estuary, but there were major recoveries among the tunicates. At the most TBT-contaminated upstream site, numbers of tunicate species increased from 2–3 in 1987/88 to 5–6 in the period 1989–2005. The coverage of two tunicates (*Ascidiella aspersa* and *Ascidia conchilega*) increased from 0 and 14–31 individuals, respectively, per 250 m beam trawl tow in 1987, to 4024–7806 and 824–2454, respectively in 1997, although numbers fell back somewhat in 2005. This was a large change to the sessile invertebrate community, implying a substantial increase in the community's filtration capacity.

As with all field observations, it is impossible to be sure that these results simply represent the impacts of TBT alone, and there are some clear differences in response between the two marine ecosystems. However, the data on tunicates are worth serious consideration given that similar effects consistently occurred in both locations. There are relatively few toxicity data on tunicates, but Bellas et al. [36] reported a 20 h EC50 of 7.1 µg TBT/L (LOEC = 4 µg TBT/L) for abnormal embryonic development in *C. intestinalis*, whereas Pellerito et al. [37] observed damaged hatching and swimming in embryos and larvae of the same species after short exposures to 35 µg TBT/L. Higher TBT concentrations are needed to cause similar effects in larvae of *Styela plicata* [38]. These toxicities, if representative of tunicates in general, are at least an order of magnitude lower than in crustaceans but are nevertheless sufficiently high to explain the observed effects in San Diego Bay (range of seawater concentrations 89–400 µg TBT/L; [28]), if not the Crouch (highest mean 0.05 µg TBT/L in 1986; [39]).

Indirect population impacts

Given the far-reaching nature of the changes observed in marine invertebrate communities [24,28], it seems likely that some were indirect effects rather than primary toxicity. Furthermore, the large reductions in filter-feeding capacity because of the near-elimination of sessile organisms in some areas would almost certainly have had consequential effects on estuarine plankton populations. Unfortunately, data on this subject are not available.

However, one example of probable indirect effects of TBT has been reported from the freshwater Norfolk Broads in the UK [40]. This study showed that there was extensive macrophyte loss in these waters associated with a peak of high TBT contamination (up to

a maximum of 0.9 μg TBT/L in water and 750 mg TBT/kg in sediment) between 1962 and 1987. On the basis of microfossil and TBT-analytical data from sediment cores, the authors postulated that macrophyte loss was owing to TBT toxicity on grazing organisms such as gastropods and zooplankton. This would have led to excessive periphyton and phytoplankton growth, which in turn would have restricted growth of macrophytes.

Discussion and conclusions

This brief review shows that the now-banned antifouling paint ingredient, TBT, had far-reaching effects on invertebrate populations and communities from which some are still recovering. It is clear that many mollusc populations were severely damaged or eliminated by this substance, partly through its effects on the retinoid-X hormonal system and consequent masculinisation, leading to reduced reproductive success. What is less well known and understood are the impacts on other invertebrates. The available evidence strongly suggests that a number of crustacean populations were directly damaged by TBT in estuarine and coastal areas, possibly through activity via the ecdysteroid or retinoid-X systems. There are also reasonably compelling data to suggest that estuarine tunicate populations were adversely affected, and some evidence for impacts on bryozoans and sponges in highly contaminated areas.

It is to be expected that some impacts were indirect in nature, although few data on this point are available. However, there is some evidence that impacts on freshwater grazing organisms had adverse indirect effects on aquatic macrophytes.

The interesting question arises as to whether current biocide regulatory programmes would be able to detect a substance with properties similar to TBT and thereby prevent its use. In the European Union, antifouling paint ingredients are stringently regulated by the Biocidal Products Directive (BPD) (Directive 98/8/EC—see www.echa.europa.eu), but similar controls apply in many other jurisdictions. Among a long list of data, the BPD requires information on the biodegradation of a new marine antifouling substance in both the pelagic and sediment compartments (generated using methods such as ISO/DIS Standards 14592-1 and 2, and Organisation for Economic Cooperation and Development (OECD) Test Guideline 308), and substances with the very long aquatic half-life of TBT (many years in anaerobic sediments) would certainly be detected. Furthermore, a wide range of sensitive aquatic toxicity tests is now available, including reproduction tests for toxicity to molluscs (e.g. OECD Test Guidelines 242 and 243). The assessment factors applied to laboratory toxicity data to derive predicted safe concentrations are large (50−10,000) unless the available toxicity data include long-term no-observed effect-concentrations (NOECs) for species from three trophic levels (algae/crustaceans/fish) plus long-term NOECs for an additional two marine groups such as echinoderms or molluscs. As with persistent chemicals, a substance with the high toxicity of TBT to aquatic organisms should therefore be detected before it enters the market, thus allowing adequate risk assessment.

No chemical regulatory scheme is infallible, so the back-up provided by environmental monitoring programmes is essential. However, such programmes tend to be focused at present on the detection of a relatively short list of target pollutants and not so much on adverse biological effects. It is arguable that the types of impacts caused by TBT in freshwater and especially marine ecosystems might well not be detected by routine monitoring programmes such as those operated in support of the European Union Water Framework and Marine Strategy Framework Directives (EU Directives 2000/60/EC and 2008/56/EC, respectively), and a case can therefore be made to modify these programmes to make them more focused on biological effects. Modern techniques based, *inter alia*, on the use of bioassays, biomarkers and genomics are increasingly allowing the early diagnosis of unexpected environmental problems.

In summary, TBT-based antifoulants caused serious damage to entire aquatic invertebrate communities, as might have been expected of a broad-spectrum biocide of such potency. What is more surprising is that TBT continued in use for about 20 years after most of the evidence of these impacts was available. Some lessons concerning the need for environmental risk assessment of new biocides have now been learned, but approaches to monitoring aquatic ecosystems need updating to limit the scale of catastrophes resulting from underpredicted environmental damage.

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Conflict of interest

Nothing declared.

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