

**LABORATORY AND FIELD INVESTIGATIONS ON THE EFFECTS OF TRIBUTYL TIN ON THE OYSTER, *OSTREA EDULIS***

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Tributyltin (TBT) is the main active ingredient in organotin-based antifouling paints. It is possibly the most toxic substance that is intentionally introduced into the marine environment and its ecological impact at extremely low levels, particularly on molluscs, has been confirmed through a number of laboratory and field investigations (AXIAK *et al.*, in press). The present work reports on cytopathological effects in the adult oysters *Ostrea edulis* on laboratory exposure to environmentally realistic levels of TBT, with special reference to digestive cell atrophy. Moreover, the likely ecological significance of this effect is discussed and evaluated through field studies. These investigations form part of a programme of evaluation of biological impact of TBT in the Mediterranean.

Adult specimens of *Ostrea edulis* were exposed to nominal concentrations of TBT chloride (dissolved in ethanol) of 100 and 10 ng l<sup>-1</sup> in unfiltered sea water. Exposure was conducted for 96 hours with test mixtures being kept aerated throughout the whole period and renewed every 24 hours. Oysters were left unfed during the experiment, which was conducted at 17°C with a photoperiod of 12:12 dark to light hours. After exposure, surviving animals were fixed in Bouin's reagent, dehydrated in ethanol, cleared in xylene and embedded in paraffin wax. 7µm thick sections were then stained with Erlich Haematoxylin and Eosin. The effect of TBT on the digestive cells (Type I) of the diverticula of the digestive gland was assessed by measuring the heights of such cells under a magnification of X 1000.

The mean height of digestive cells decreased from 13.44 µm (sd: 2.25) in controls to 11.43 µm (sd: 2.41) in animals exposed to 10 ng l<sup>-1</sup> of TBT, with effects being found to be statistically significant at P < 0.001 with Tukey's multiple comparison tests at this and all other levels of exposure.

In another experiment, shell thickening was evaluated using a shell thickness index (STI) in *Ostrea* collected from five coastal sites around the major harbour area (Grand Harbour and Marsamxett) in Malta (Central Mediterranean). Malta's major yacht marinas and ship-repairing yards are located there. Oysters were also collected from a clean reference site which is only exposed to limited boating activities. These sites differed markedly in the levels of organotins in sediments as well as in the water column as measured by GC-FPD (AXIAK *et al.*, in press). Mean seasonal levels of TBT in superficial sediments at the different sites, expressed as ng Sn per g dry weight ranged from 18 to 210 for Grand Harbour; from 18 to 410 for Marsamxett; and below detection limit at a reference site outside the harbours. Levels of TBT at 1m depth in the water column ranged from 8 to 120 for Grand Harbour; and from 8 to 40 ng Sn /g DW for Marsamxett.

STI which is the ratio between valve length to valve thickness, has been used as an index of abnormal shell growth in bivalves. Low STI values are indicative of shell abnormalities which are usually due to the formation of various types of minute chambers within the shell matrix (BRYAN and GIBBS, 1991). The mean STI value for the lower valve of animals collected from the reference site was found to be 21.65, while mean STI values (for the lower valve) for animals collected from Grand Harbour and Marsamxett ranged from 9.9 to 10.5 and from 8.5 to 11.8, respectively. STI for both upper and lower valves indicated that shell abnormalities were significantly higher in areas within the Grand Harbour and Marsamxett Harbour than in the reference site.

Digestive cells of bivalves are known to undergo atrophy on exposure to a wide range of contaminants. This cell atrophy is normally correlated with catabolic metabolism and reduced bioenergetic balances of bivalves. However such cytological stress in *Ostrea* as reported here, has never been recorded for any contaminant at such low environmental concentrations. It is here proposed that reduced bioenergetic balance and the consequent reduction in body weight may lead to a shell chambering effect in this test species as shown in the field experiment. Shell abnormalities in *Ostrea*, as indicated by STI were partly due to the formation of minute water-filled chambers within the shell. New shell deposition by an animal with reduced somatic growth may lead to the formation of such chambers. Such shell abnormalities may be part of a general stress syndrome which may be elicited by a range of contaminants and not just TBT. Nonetheless, the laboratory experiments reported here, provide evidence that this species is particularly sensitive to low levels of TBT (10 ng/l) in sea water, exhibiting significant atrophy in digestive cells.

**REFERENCES**

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**COASTAL MARINE LITTER IN THE CENTRAL MEDITERRANEAN : BASELINE INFORMATION ON BEACH STRANDING, COASTAL DENSITIES AND RATES OF PHOTODEGRADATION**

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Floating marine debris is considered to be a potential environmental hazard especially in semi-enclosed waters such as the Mediterranean. The Maltese Islands lie at the junction between the two major Mediterranean basins, and therefore are ideally situated to provide data on the background levels of the densities of marine litter of the whole region as well as on exchanges of floating debris between the Western and Eastern Mediterranean.

The present paper reports on a study undertaken in 1991-92. It presents baseline qualitative and quantitative data on marine litter within the Central Mediterranean coastal areas, as found stranded on beaches, or floating at sea. It also reports on the rate of degradation of certain types of plastics. The potential economic impact of litter on sea crafts was also assessed.

Marine litter can originate from two sources : ship traffic or land-based sources. Two beaches located near Fomm ir-Rih (North-East, Malta) were surveyed in August 1991 and April 1992 in order to assess qualitatively and quantitatively the extent of pollution by marine debris originating from sources at sea. These beaches were remote from built-up areas and with very limited accessibility, so that most of the litter found stranded on their shoreline must have originated from the sea. The extent of litter on the beaches was investigated using belt transects. The litter was counted, weighed, and its age, fabrication material and previous contents recorded. Litter densities, percentages by weight and number of the various components were calculated. Frequency distributions showing the geographical origins of containers in the study areas were tabulated and correlated with shore profiles and degree of beach exposure.

The slopes of the beaches under study were generally found to be low and litter tends to be easily trapped on this type of shores. This study revealed a wide range of fabrication materials and contents for beach-stranded marine litter. The mean density of shore-stranded litter ranged from 60 to 650 g/m<sup>2</sup> according to position on the beach away from the waterline. This was in general comparable to that reported in other regions in the Mediterranean. Plastic litter occupied the highest percentage by number of items, with wood being also predominant. Litter was found to be both of local and foreign origin.

Litter density distribution down-shore, generally showed a bimodal distribution. Containers marketed or manufactured locally accounted for the modal classes of both beaches. Twelve different countries of origin were identified in all. Most containers were current production types but one was manufactured in 1986. Litter accumulation was found to be influenced by the extent of exposure of beach localities as well as by local patterns of wind and sea currents.

Sea surveys in inshore waters around Malta were carried out during winter 1991. Floating megalitter was counted and recorded from a boat moving in a straight course or along the shore at distance of up to 3 km offshore. Only litter which was observed within 10 m on either side of the boat was recorded. The area of water observed in a single trip was therefore equal to the distance covered by the boat multiplied by 20 m. The density (frequency) of litter was then expressed as the number of items observed per unit area.

An overall mean density of 41 items per km<sup>2</sup> of floating megalitter was recorded for the coastal areas around Malta. In 55% of the surveys, the highest densities were recorded for plastic debris. The highest mean density for plastics in fact was 158 items per km<sup>2</sup>. The next predominant type of litter at sea was found to be wood at 21.6%, with nylon occurring at 5.1%.

A survey amongst boat owners indicated that the negative economic implications of such marine litter may be considerable. A number of cases were reported in which water intakes of yachts were clogged by floating litter, or litter got entangled with craft propellers or drive shaft. Cases of floating debris interfering with radar signals were also reported.

The rates of photodegradation of plastics exposed to different environmental conditions were measured by tensile testing of standard test strips. The rates of photodegradation were assumed to be negatively correlated with the exposure time required by the test strips to reach 5% elongation before they break (ie. time required by plastic to turn brittle). Two types of plastics were utilized: low density polyethylene (LDPE) and enhanced photodegradable ethylene-carbon monoxide copolymer (E/CO). Test strips were exposed to sunlight either in dry conditions (on a roof top) or while continuously washed by seawater, being attached to floating platforms at sea. Exposure experiments were simultaneously undertaken in Malta and U.K. (Farnborough College of Technology). Data for E/CO test strips is tabulated below :

	Mean Exposure days required by test strips to reach 5% elongation	
	Jul-Sep.	Oct-Feb.
Wet Exposure in Malta	21	25
Dry Exposure in Malta	13	22
Wet Exposure in U.K.	71	74
Dry Exposure in U.K.	65	60

These data indicate that rates of degradation were substantially lower at sea and under U.K. prevalent climatic conditions. Other data on the tensile properties of the exposed plastics, will be published elsewhere.

