Zoological Studies **38**(2): 189-195 (1999)

Zoological Studies

TBT (Tributyltin) Toxicity to the Visual and Olfactory Functions of Tigerperch (*Terapon jarbua* Forsskål)

Dar-Yi Wang and Bao-Quey Huang*

Department of Fisheries Science, National Taiwan Ocean University, Keelung, Taiwan 202, R.O.C.

(Accepted December 14, 1998)

Dar-Yi Wang and Bao-Quey Huang (1999) TBT (tributyltin) toxicity to the visual and olfactory functions of tigerperch (*Terapon jarbua* Forsskål). *Zoological Studies* **38**(2): 189-195. In order to understand tributyltin (TBT) toxicity to peripheral sensory functions, changes of visual and olfactory structures of tigerperch (*Terapon jarbua* Forsskål) were examined in juvenile and adult fish after exposure to 10, 50, and 100 μg TBT/l in juveniles and to 1 and 5 μg TBT/l in adults, respectively, for 7 d. Severe sloughing of squamous epithelial cells led to a thinning of the corneal epithelium. The retinomotor response of pigmented epithelium significantly decreased from 75.33% ± 2.88% (in the control group) to 63.33% ± 4.04% ($p < 0.05$, $n = 3$) in the 100 μg/l group. The separation and necrotic corneal epithelium may lead to partial opacity and/or irregular abrasion and thus decrease retinomotor responses. In addition, scanning electron microscopic studies revealed edema, deformity, and sloughing of the microvilli, cilia, and rods in receptor cells of the olfactory epithelium. Electroolfactogram (EOG) responses significantly decreased from −3.0 ± 0.6 mV to −1.3 ± 0.5 mV (1 μg/l) and −0.4 ± 0.1 (5 μg/l), respectively ($n = 3$).

Key words: Tributyltin, Corneal and olfactory epithelium, Retinomotor response, Electroolfactogram (EOG).

Tributyltin (TBT) has been widely used to retard fouling by marine organisms in cagenets and setnets of coastal fisheries. Nets must be cleaned periodically in order to obtain better seawater exchange. Antifoulants (such as TBT) are more economical and convenient, and therefore popularly preferred by the fisheries industry. However, they have been documented to cause cytopathological changes in invertebrates and fishes (Gibbs et al. 1988, Byrne et al. 1989, Ellis and Pattisina 1990). TBT concentrations of acute toxicity were reported to range between 3.0 and 25.9 μg/l in some studied teleosts (Bushong et al. 1988). Shorts and Thrower (1987) reported that juvenile chinook salmon were very sensitive to TBT poisoning in seawater and rapidly accumulated high concentrations of TBT in various tissues (e.g., liver, brain, and muscle) with dose- and time-dependent effects. Therefore, it is crucial that TBT-treated nets for fisheries applications be used with extreme caution.

Some studies have reported on the toxic effects of TBT on morphological and functional alterations of teleost gills in aquatic media (Byrne et al. 1989, Schwaiger et al. 1992, Tsuda et al. 1992, Wang and Huang 1998). The corneal and olfactory epithelia which are also directly exposed to the aquatic environment can be considered as potential targets. The "retinomotor response" is synchronized with the intensity of light incidence on the retina (Ali 1975). A significant decrease in retinomotor response accompanied by a thinning cornea was found in fish contaminated by a detergent (linear alkylbenzene sulfate, LAS) (Huang and Wang 1995). Organotins induced alterations in the retina and lens (Wester and Canton 1987, Fent and Meier 1992) and also induced vacuolization in the visual center of fish brain (optic tectum) (Triebskorn et al. 1994b). The corneal epithelium (the outermost structure of the eye) was selected to investigate the mode of toxic action of TBT at a directly attacked site, and the "retinomotor response" was thus chosen to examine subsequent effects of corneal alterations.

Cilia and microvilli on the olfactory epithelial surface are also directly exposed to the aquatic environ-

*To whom correspondence and reprint requests should be addressed. Tel: 886-2-24622192 ext. 5028. Fax: 886-2-24624790.

ment and are regarded as the main site of sensory transduction, thus electrical responses of the olfactory epithelium (as measured by an electroolfactogram, EOG) can be a suitable target phenomenon to reflect morphopathological damage induced by aquatic pollution (Hara 1982, Kleerekoper 1982). Therefore, EOG responses, because of their high sensitivity, can be a reliable indicator for detecting environmental contamination (Cancalon 1983). Examination of pathological alterations of the olfactory epithelium and abnormal EOGs should be a powerful tool to detect contamination of the aquatic environment.

Tigerperch (*Terapon jarbua* Forsskål) is one of the most common euryhaline teleosts in Taiwanese waters. Juvenile fish usually live in estuaries, and adults also inhabit coastal and fresh waters. In addition, these wild-captured fish are commonly kept in netcages at local fish ports before being sold. Therefore, tigerperch face impacts of TBT leaking from nets and/or boats at various life stages. Hence they are valuable animal indicators for investigating TBT toxicity. Compared to other studied fishes at early stages, the 24 h LC50 in the early life stage of tigerperch ranged between 19.7 and 135.0 μg/l, and this species showed a relatively higher tolerance (Bushong et al. 1988, Wang and Huang 1998). The present study was carried out to evaluate toxicity of TBT on the structure and function of peripheral sensory (visual and olfactory) organs by applying scanning electron microscopic (SEM) and light microscopic (LM) investigations. Furthermore comparisons were made of retinomotor responses and electroolfactographic (EOG) responses were recorded.

MATERIALS AND METHODS

Juvenile and adult tigerperch (*Terapon jarbua* Forsskål), ranging from 21.9 to 54.3 and 179.0 to 239.9 mm in body length, were obtained from the northeastern coast of Taiwan by hand nets, and kept in seawater (pH 7.93-8.04, salinity 35‰, and temperature 27.5 to 28.2 °C) under a 12L/12D photoperiod. A stock solution of tributyltin chloride (Merck, Darmstadt, Germany) was prepared by diluting 0.02 g TBT in 1 ml acetone before use. Acetone used at this experiment (0.05 to 5 μl/l) is known to be nontoxic as judged by histopathological effects (Fent and Meier 1992). All experiments were conducted by using a static system. TBT solutions in each experimental tank were renewed once per day.

The cornea and retinae were dissected from excised eyes of fully light-adapted juvenile fish which were exposed to nominal concentrations of TBT (10, 50, 100 μg/l, and a control group, n = 3 per group) for 7 d. After Bouin's fixation, samples were processed by a routine procedure for paraffin sections and staining by haematoxylin-eosin. Retinomotor responses (mean thickness of pigment epithelium/mean thickness of photoreceptor layer) were measured by the use of a microimage processing system (Hamamatsu Photonic C2400, Tokyo, Japan and JAVA soft, Corte Madera, Ca, USA). Means and standard deviations of each group of measurements were obtained from 3 fish (50 data points for each individual retina measured from random areas), and the effects of TBT concentrations were evaluated by ANOVA (Duncan multiple comparison).

Additional measurements were made of the morphological changes of the epithelial surface of the cornea. Additional 3 individuals from each group were also studied by SEM. Sampled tissues were first fixed in 2% glutaraldehyde (in cocadylate buffer, pH 7.4) followed by fixation in 4% osmium (mixed with 3% lantanium nitrite, 0.2 M s-collidine solution). After being dehydrated in an ethanol series (50% to absolute) and dried by the critical point method (liquid CO_2), specimens were coated with gold (E101 Hitachi, Tokyo, Japan) and examined by SEM (C2400 Hitachi, Tokyo, Japan).

To study the sensitivity of the olfactory system to TBT, fish were placed in concentrations that had no observed effects on cornea epithelium (below 10 μg/l TBT, according to the above experiment). In each experimental tank (0, 1, and 5 μg/l TBT), 3 juvenile tigerperch were exposed to TBT for 7 d. After this period, SEM examination of the surface of the olfactory epithelia was performed.

Prior to recording EOG responses, adult tigerperch were initially acclimated in freshwater 2 mo before testing because high conductivity of seawater may cause the shunting of electrical signals (Silver 1976). Throughout the experiment, fish were kept in freshwater (29-30 °C, pH 7.6-7.9). Adult tigerperch were exposed to 0 (as control), 1, and 5 μg/l TBT (nominal concentrations) solutions for 7 d. Then the fish were paralyzed with an intramuscular injection of gallamine triethiodide (4 mg/100 g wt) and the nasal cavities were surgically exposed under a stereomicroscope. EOG was elicited by perfusing (M312 Gilson, Villers-le-Bel, France) the nasal cavity with 10^{-2} M L-alanine (Sigma, Louis, Mo, USA) (perfusion speed: 0.4 ml/min; stimulation time: 10 s). Signals were recorded by placing a 3 M KCl-filled glass capillary electrode (outer diameter, o.d. 1 mm, fiber-filled, borosilicate, AM System Co., Everett, Wa, USA) which was held by a micromanipulator

(Campden Instrument, London) to the midline of the olfactory lamella. A silver electrode was positioned on the surface of the head skin as the reference electrode. Both electrodes were connected to an amplifier (MEZ 8201 Nihon Kohden, Tokyo, Japan) with the signals displayed on an oscilloscope (VC-9, Nihon Kohden, Tokyo, Japan) and recorded on a thermal array recorder (RTA-1100M, Nihon Kohden, Tokyo, Japan).

RESULTS

TBT exposure caused dose-dependent damage to corneal and olfactory epithelia (Figs. 1, 2) as revealed by both LM and SEM. Histological and scanning electron microscopic examinations of the cornea revealed significant differences in both thickness and superficial appearance of the outermost epithelial structure of the corneas. In the control group

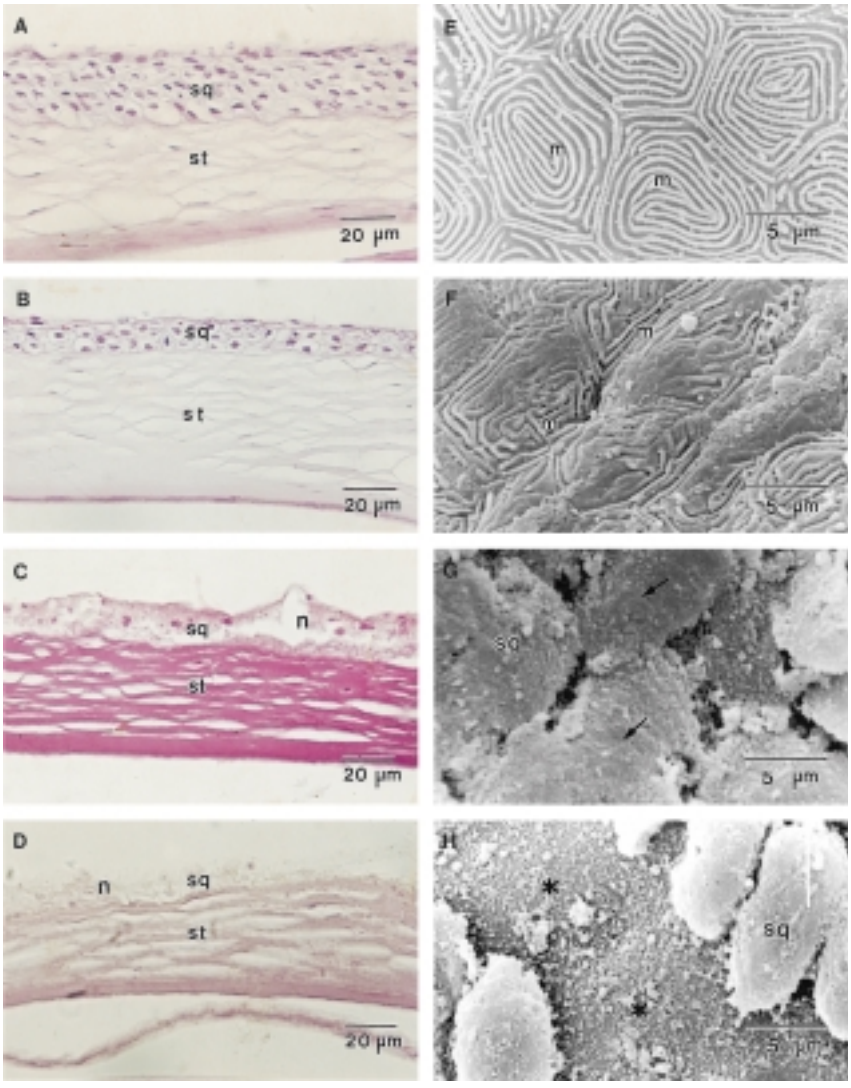

Fig. 1. Pathologic effects of TBT on the corneal epithelium by LM (A-D) and SEM (E-H) studies.
A. and E. Control group showing corneal epithelium consisting of multiple layers of squamous (sq) epithelial cells, stroma (st), and healthy corneal epithelial cells with clear microridges (m) on the surface.
B. and F. Effects of 10 µg/l TBT on the cornea showing fewer layers of squamous (sq) epithelium. Microridges (m) of epithelial cells were not so clear and regular as in controls.
C. and G. Effects of 50 µg/l TBT showing thinning and necrosis (n) of the squamous epithelial layer (sq); microridges of epithelial cells have disappeared (arrows).
D. and H. Effects of 100 µg/l TBT showing necrosis (n) and loss of the squamous epithelial layer (sq); lifting and sloughing of squamous epithelial cells are obvious (*).
A-D: x400. E-H: x3000.

(Fig. 1A), the corneal epithelium consisted of stratified layers of squamous epithelial cells, forming the outermost layer and overlaying the neighboring the corneal stroma. Further reduction in layer and necrosis of the surface squamous epithelium became more obvious as TBT concentrations increased (Fig. 1B-D). The SEM examination showed that the normal superficial cornea is paved with flat, regularly arranged epithelial cells, each with a clear concentric microridge pattern (Fig. 1E). At higher TBT exposure concentrations, the irregularity and erosion at the surface resulted in a smoothing of the microridges and, finally, their loss as (Fig. 1F-H) cellular necrosis advanced (Fig. 1H).

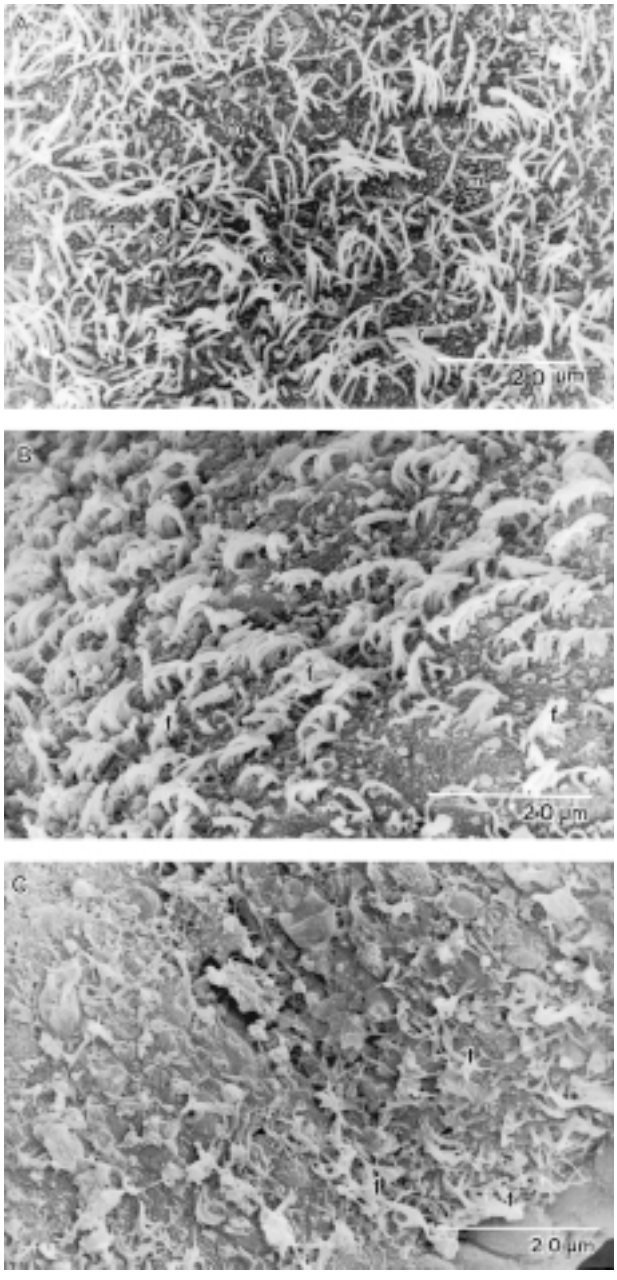

Fig. 2. Pathologic effects of TBT on the olfactory epithelium. Control group (A) shows the ciliated (c) and microvillous (m) receptor cells and rods (r) of rod cells. The 1 and 5 μg/l TBT groups (B, C) show bending (arrows), irregular lying (arrowheads), or distal fusion (f) of the cilia on the superficial epithelium. In the 5 μg/l TBT group (C), distal fusion (f) of cilia has become obvious and the microvilli and rods are not found in the sensory area. A-C: x1500.

Retinomotor responses of retinal pigmented epithelium decreased from 75.33% ± 2.88% (0 μg/l TBT, control group) to 71.33% ± 4.50% (10 μg/l TBT), 69.50% ± 2.12% (50 μg/l TBT) and, 63.33% ± 4.04% (100 μg/l TBT). No significant differences could be found between the control and low-dose groups (10 and 50 μg/l). A significant difference could be found only between the control and the 100 μg/l groups (Table 1, $p < 0.05$).

In the olfactory epithelium of the control group, ciliated and microvillous receptor cells were associated with rod cells randomly protruding above the surface (Fig. 2A). After exposure to 1 and 5 μg/l TBT, the cilia began bending, shortening and/or irregularly lying on the superficial epithelium, and the microvilli and rods gradually disappeared (Fig. 2B, C).

The electroolfactogram of tigerperch in the control group showed a monophasic response, rising from the baseline to its peak (ranging between –2.3 and –4.0 mV) when the stimulant (10^{-2} M L-alanine) began to perfuse the nasal cavity, and the amplitude of the response dropped sharply to the baseline when the stimulation stopped (Fig. 3A). After exposure to 1 μg/l TBT, the EOG response to L-alanine stimulation showed a significant reduction of the initial fast peak from –3.0 ± 0.6 mV (control group) to –1.3 ± 0.5 mV (Fig. 3B). In the 5 μg/l TBT group, the EOG again showed a further significant decrease to an almost non-observable level (–0.4 ± 0.1 mV) (Fig. 3C). EOG records were statistically analyzed by ANOVA (Duncan multiple comparison) and are shown in table 2.

Table 1. Means and standard deviations of retinomotor responses (%) at various concentrations of TBT exposure ($n = 3$)

TBT concentration	Retinomotor response (%)	Significance
Control	75.33 ± 2.88	a
10 μg/l	71.33 ± 4.50	a
50 μg/l	69.50 ± 2.12	a
100 μg/l	63.33 ± 4.04	b

The notes a and b indicate significant differences as determined by ANOVA (Duncan multiple comparison) ($p < 0.05$).

DISCUSSION

Toxic effects of TBT on peripheral sensory functions (corneal and olfactory epithelium) were characterized in tigerperch by light microscopic, scanning electron microscopic, and electrophysiological studies. In the present investigation, the outermost epithelial layer of the cornea became thinner, and progressed to death and severe sloughing of squamous epithelial cells. It is clear that toxicity of TBT directly damages the cornea and cause edematous changes in the stroma, thereby disrupting the fibrillar array (Wester and Canton 1987, Bruno and Ellis 1988, Fent and Meier 1992). In rabbits, remarkable corneal necrosis and sclera edema were also found after dropping TBT directly on to their eyes (Pelikan 1969). Yoshizuka et al. (1991) also found corneal edema and ultrastructural changes in the mitochondria of corneal endothelial cells within hours after applying an intramuscular injection of TBT to rats. Therefore, the toxic mechanism resulting from direct contact or indirect transport through the circulatory system may be similar, but the toxicity mechanism may be different because of the different pathways toward the affected target tissues.

Studies on the toxic effects of TBT in teleosts have indicated that TBT penetrates the corneal epithelium to a small extent, and induces minor injury to the retinal layer possibly through the circulatory system (Wester and Canton 1987, Fent and Meier 1992). Fent and Meier (1992) reported that long-term exposure to TBT might further lead to opacity of the cornea and lens, and even cause blindness in fish (Bruno and Ellis 1988). Yoshizuka et al. (1991) reported that intramuscular injection of TBTO in rats caused edema and necrosis of the corneal endothelium, but they did not mention damage to the retina. In the present study, a decrease in the retinomotor responses of retinal pigmented epithelium was noted when fish were exposed to increasing concentrations of TBT. This is not sufficient evidence to explain that exposure to TBT decrease retinomotor response, although Huang and Wang (1995) found that the toxicity of the detergent LAS causes a thinning of the corneal epithelium and a decrease of retinomotor responses, and they suggested that the necrotic and ruptured cells of the superficial corneal epithelium might lead to abrasion, and thus cause roughness of the cornea. According to Bennett and Mattson's (1989) function $\delta = \lambda(TIS/4\pi)^{1/2}$ (δ: roughness, λ: wavelength of light, TIS: total integrated scattering), the emerging beam is increasingly diffused as the light is increasingly scattered, therefore the intensity of light reaching the retina may be reduced. The present SEM examination confirms that TBT causes cellular necrosis, lifting, and sloughing of the corneal epithelium. Thus the sloughing cells of superficial tissue may cause an increase of roughness and a concomitant increase of TIS from the corneal epithelium. Therefore, a reduction in light transparency of the cornea may lead to inhibition of retinomotor responses.

Pollutants may also damage the sensory surface of the olfactory epithelium by loss and removal of microvilli and cilia of sensory cells (Brown et al. 1982, Cancalon 1983). In the present study, the olfactory cilia showed degenerative morphological alterations. The cilia of the sensory cells were also found to become shorter, which implies that TBT lipophilic characteristics might affect the ciliary membrane and interrupt binding capacity. Therefore 2 hypotheses for neurotoxicity (changes of membrane permeability and lesions in the membrane) (Song et al. 1987, Oyama et al. 1993, Virkki and Nikinmaa 1993, Triebskorn et al. 1994a,b) may serve as possible explanations for the effects within the olfactory epithelium.

Several studies have indicated that TBT toxicity relates to changes in mitochondrial membranes (Krigman and Silverman 1984, Mercier et al. 1994) and acts as an inhibitor of oxidative phosphorylation in mitochondrial function (Wester et al. 1990, Yoshizuka et al. 1991, Fent and Meier 1992). The loss of energy supply in cells may thus cause edema and cell death (Song et al. 1987, Lee 1991, Schwaiger et al. 1992). In this study, pathological and degenerative alterations in corneal epithelial cells may be at

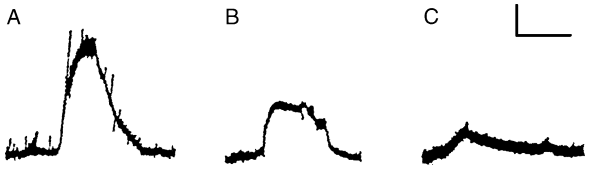

Fig. 3. Changes in EOG after treatment with TBT concentrations for 7 d. A: control, B: 1 μg/l, C: 5 μg/l. Scale: 10 s, 1 mV.

Table 2. Means and standard deviations of EOG potentials (mV) after exposure to TBT (n = 3)

TBT concentration	EOG (mV)	Significance
Control	– 3.0 ± 0.6	a
1 μg/l	– 1.3 ± 0.5	b
5 μg/l	– 0.4 ± 0.1	c

The notes a, b, and c indicate significance as analyzed by ANOVA (Duncan multiple comparison) ($p < 0.05$).

least in part initiated by damage to mitochondrial functions and thus a shortage of energy supply. TBT may also increase intracellular Ca^{2+} concentrations (Oyama et al. 1993) by increasing specific membrane permeability. TBT induces intramyelin vacuolization in the optic tectum and optic nerve of rainbow trout (*Oncorhynchus mykiss*) (Triebskorn et al. 1994a,b) which was explained by the high lipid content of the myelin interacting with lipophilic TBT molecule (Triebskorn et al. 1994a). Increasing electron densities of injured nerve fibers and cells in the optic tectum by increasingly abnormal levels of $[Ca^{2+}]$ were thought to disturb intracellular signal transductions and perhaps lead to pathological states (Oyama et al. 1993, Triebskorn et al. 1994b). In the present study, the inhibited EOG responses could be related to changes in cellular permeability induced by TBT toxicity.

To date, the specific neurotoxicity of TBT to retinal and olfactory functions has rarely been discussed (Cancalon 1983, Wester and Canton 1987). Only a few pathomorphological descriptions and ethological alterations have been demonstrated (Triebskorn et al. 1994a,b). However, the present study clearly reveals that TBT toxicity severely alters morphological structures (SEM) and thus impacts neurophysiological functions (retinomotor responses in the visual system and EOG responses in the olfactory system). However, further detailed studies by transmission electron microscopy together with biochemical investigations may be useful for further assessing the mechanisms of TBT toxicity. The calculated exposure concentrations used in this study were probably higher than those actually in the ambient media, because of rapid degradation resulting from tank adsorption, evaporation, and incorporation by the fish (65%-87% in 24 h) was considered (Fent and Meier 1992 1994). Thus, better assessment of standard toxic concentrations will be necessary in future investigations.

Acknowledgments: This work was partially supported by the National Science Council, R.O.C. (NSC 88-2313-B-019-039). We are also grateful to anonymous reviewers and English editors for critically reviewing the manuscript and offering valuable suggestions.

REFERENCES

Ali MA. 1975. Retinomotor responses of the goldfish (*Carassius auratus*) to unilateral photic stimulation. Rev. Can. Biol. **23:** 45-53.

Bennett JM, L Mattson. 1989. Introduction to surface roughness and scattering. Washington, D.C.: Optical Soc. Am.

Brown SB, RE Evans, BE Thompson, TJ Hara. 1982. Chemoreception and aquatic pollutants. *In* TJ Hara, ed. Chemoreception in fish. New York: Elsevier Press, pp. 363-394.

Bruno DW, AE Ellis. 1988. Histopathological effects in Atlantic salmon, *Salmo salar* L., attributed to the use of tributyltin antifoulant. Aquaculture **72:** 15-20.

Bushong SJ, LW Hall Jr., WS Hall, WE Johnson, RL Herman. 1988. Acute toxicity of tributyltin to selected Chesapeake Bay fish and invertebrates. Water Res. **22:** 1027-1032.

Byrne P, D Speare, HW Ferguson. 1989. Effects of a cationic detergent on the gills and blood chemistry of rainbow trout *Salmo gairdneri*. Dis. Aquat. Org. **6:** 195-196.

Cancalon P. 1983. Influence of a detergent on the catfish olfactory mucosa. Tissue Cell **15:** 245-258.

Ellis DV, A Pattisina. 1990. Widespread neogastropod imposex: a biological indicator of global TBT contamination. Mar. Pollut. Bull. **21:** 248-253.

Fent K, W Meier. 1992. Tributyltin-induced effects on early life stages of minnows *Phoxinus phoxinus*. Arch. Environ. Contam. Toxicol. **22:** 429-438.

Fent K, W Meier. 1994. Effects of tributyltin on fish early life stages. Arch. Environ. Contam. Toxicol. **27:** 224-231.

Gibbs PE, PL Pascoe, GR Burt. 1988. Sex change in the female dog-whelk, *Nucella lapillus*, induced by tributyltin from antifouling paint. J. Mar. Biol. Assoc. **68:** 715-731.

Hara TJ. 1982. Structure-activity relationships of amino acids as olfactory stimuli. *In* TJ Hara, ed. Chemoreception in fish. New York: Elsevier Press, pp. 135-158.

Huang BQ, DY Wang. 1995. Corneal damage in young tigerperch (*Terapon jarbua*) exposed to the surfactant linear alkylbenzene sulfonate (LAS). Zool. Stud. **34:** 41-46.

Kleerekoper H. 1982. The role of olfaction in the orientation of fishes. *In* TJ Hara, ed. Chemoreception in fish. New York: Elsevier Press, pp. 201-226.

Krigman MR, AP Silverman. 1984. General toxicology of tin and its organic compounds. Neurol. Toxicol. **5:** 129-140.

Lee RF. 1991. Metabolism of tributyltin oxide by marine animals and possible linkages to effects. Mar. Environ. Res. **32:** 39-35.

Mercier A, E Pelletier, JF Hamel. 1994. Metabolism and subtle toxic effects of butyltin compounds in starfish. Aquat. Toxicol. **28:** 259-273.

Oyama Y, T Ueha, A Hayashi. 1993. Effects of tri-n-butyltin on intracellular Ca^{2+} concentration of rat cerebellar neurons. Euro. J. Pharmacol. **248:** 89-93.

Pelikan Z. 1969. Effects of bis (tri-n-butyltin) oxide on the eyes of rabbits. Brit. J. Industr. Med. **26:** 165-170.

Schwaiger J, F Bucher, H Ferling. 1992. A prolonged toxicity study on the effects of sublethal concentration of bis (tri-*n*-butyltin) oxide (TBTO): histopathological and histochemical findings in rainbow trout (*Oncorhynchus mykiss*). Aquat. Toxicol. **23:** 31-48.

Short JW, FP Thrower. 1987. Toxicity of tri-n-butyl-tin to chinook salmon, *Oncorhynchus tshawytscha*, adapted to seawater. Aquaculture **61:** 193-200.

Silver WL, J Caprio, JF Blackwell, D Tucker. 1976. The underwater electro-olfactogram: a tool for the study of the sense of smell of marine fishes. Experimentia **32:** 1216-1217.

Song Y, M Xu, J Xie. 1987. Studies of the effects of tributyltin fluoride on the *Hydroides ezoensis*. Studia Marina Sinica 2**8:** 109-114.

Triebskorn R, HR Kohler, J Flemming, T Braunbeck, RD Negele, H Rahmann. 1994a. Evaluation of bis(tri-n-butyltin) oxide (TBTO) neurotoxicity in rainbow trout (*Oncorhynchus*

mykiss). I. Behaviour, weight increase, and tin content. Aquat. Toxicol. **30:** 189-197.

Triebskorn R, HR Kohler, KH Kortje, RD Negele, H Rahmann, T Braunbeck. 1994b. Evaluation of bis(tri-n-butyltin) oxide (TBTO) neurotoxicity in rainbow trout (*Oncorhynchus mykiss*). II. Ultrastructural diagnosis and tin localization by energy filtering transmission electron microscopy (EFTEM). Aquat. Toxicol. **30:** 199-213.

Tsuda T, S Aoki, M Kojima, T Fujita. 1992. Accumulation and excretion of tri-*n*-butyltin chloride and tributyltin chloride by willow shiner. Comp. Biochem. Physiol. **101C:** 67-70.

Virkki L, M Nikinmaa. 1993. Tributyltin inhibition of an adrenergically activated sodium/proton exchange in erythrocytes of rainbow trout (*Oncorhynchus mykiss*). Aquat. Toxicol. **25:** 139-146.

Wang DY, BQ Huang. 1998. Toxic effects of tributyltin (TBT) on early life stages of thornfish (*Terapon jarbua* Forsskål). J. Fish. Soc. Taiwan **25:** 15-25.

Wester PW, JH Canton. 1987. Histopathological study of *Poecilia reticulata* (guppy) after long-term exposure to bis (tri-*n*-butyltin) oxide (TBTO) and di-n-butyltindichloride (DTBC). Aquat. Toxicol. **10:** 143-165.

Wester PW, JH Canton, AAJ Van Iersel, EI Krajnc, HAMG Vaessen. 1990. The toxicity of bis(tri-*n*-butyltin) oxide (TBTO) and di-*n*-butyltindichloride (DBTC) in the small fish species *Oryzias latipes* (medaka) and *Poecilia reticulata* (guppy). Aquat. Toxicol. **16:** 53-72.

Yoshizuka M, N Haramaki, M Yokoyama, K Hara, A Kwahara, Y Umezu, H Araki, N Mori, S Fujimoto. 1991. Corneal edema induced by bis (tributyltin) oxide. Arch. Toxicol. **65:** 651-655.

三丁基錫 (Tributyltin, TBT) 對花身雞魚 (*Terapon jarbua* Forsskål) 視覺及嗅覺功能之毒性作用

王達益[1] 黃寶貴[1]

為瞭解抗附著劑(antifoulant)三丁基錫(tributyltin, TBT)對花身雞魚(*Terapon jarbua* Forsskål)視覺及嗅覺之毒性作用，分別將幼魚及成魚於不同濃度 TBT 溶液處理 7 天，再以光學石蠟切片、電顯(SEM)及電生理技術進行形態及生理功能之病變研究。結果顯示處理組之角膜上皮細胞呈顯著之剝離及壞死現象，且其視網膜黑色素移動反應(retinomotor response)由控制組的 75.33% ± 2.88% 降至 100 µg/l 組之 63.33% ± 4.04% ($p < 0.05$, $n = 3$)。推測角膜剝離及壞死所形成的粗糙表面可能造成入射至視網膜的光線減少，因而造成黑色素移動反應受抑制。由 SEM 的結果也發現嗅皮膜表面的纖毛 (cilia)、微絨毛 (microvilli) 及桿狀細胞 (rod) 均呈現退化及剝離現象，其嗅上皮電位記錄 (electroolfactogram,EOG) 也顯示嗅覺功能受到抑制，分別由 –3.0 ± 0.6 mV 降至 –1.3 ± 0.5 mV (1 µg/l) 及 –0.4 ± 0.1 mV (5 µg/l) ($n = 3$)，各組間也呈現顯著差異。花身雞魚自仔稚魚至成魚均普遍出現在本省河口及沿岸，沿岸漁業設施使用之抗附著劑對本魚種之毒性作用極具生態指標意義。

關鍵詞：三丁基錫, 抗附著劑, 角膜及嗅上皮, 黑色素移動反應, 味覺電位記錄(EOG)。

[1] 國立臺灣海洋大學漁業科學研究所