Bull. Environ. Contam. Toxicol. (1985) 35:620–626
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Bulletin of Environmental Contamination and Toxicology

Mercurial Induced Brain Monoamine Oxidase Inhibition in the Teleost *Channa punctatus* (Bloch)

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Neurotoxic effects of mercurials are well established in a variety of animals including man (Takeuchi 1972; Cheek 1980). In mammals, mercury is known to alter brain monoamine synthesis, high affinity uptake (Kobayashi et al. 1980; Komulainen and Tuomisto 1982; Sharma et al. 1982) and central catecholamine development (Bartolome et al. 1982). However, such investigations on the fishes are meagre. In the present communication, mercuric chloride ($HgCl_2$) and organic mercurial fungicide Emisan induced changes in the brain monoamine oxidase (MAO) content in the fish C. punctatus are described.

MATERIALS AND METHODS

Over fifty four adult C. punctatus weighing 40–45 g and measuring 10–12 cm in length used in this investigation were wild-caught and bought from the local fish market at Varanasi. They were acclimated to the laboratory conditions for 10 days prior starting the experiment. Fish were divided into three equal groups and kept in 40L glass aquaria containing well-water having pH 7.2, hardness 154 ppm (as $CaCO_3$), alkalinity 68 ppm (as $CaCO_3$), dissolved oxygen 7.2 ppm and conductivity 0.56 mMhos. The temperature of the water from March to May was 27 ± 1°C, 33 ± 2°C and 34 ± 0.5°C respectively.

Group I & II were exposed to sublethal dose 0.2 ppm of mercuric chloride ($HgCl_2$) and 0.5 ppm of Emisan (Methoxy Ethyl Mercuric chloride : MeEHgCl, bought from Excel India Ltd.), respectively. Group III served as the control. Aquaria water containing the compounds were changed every alternate day after feeding the fish with goat liver and fish

feed consisting of prawn powder and wheat flour in equal proportions. The experiment was started on 1st March, 1984, when the gonads were in the stage I condition. Six fish from each group were sacrificed at intervals of 20, 40 and 80 days and the whole brain was dissected out and frozen for the enzyme assay. MAO activity was estimated adopting the enzyme isotopic technique of Parvez and Parvez (1973). During the experiment only one fish died on the 70th day in the MeEHgCl group and the others exhibited obvious blanching of the skin.

Significance of the data was estimated using Student t-test. Correlation coefficient (r) and regression line equation ($Y = a + bX$) were also calculated. The gonadosomatic index (G.S.I.) was calculated using the formula :

$$\frac{\text{Total Gonad Weight}}{\text{Total Body Weight}} \times 100$$

RESULTS AND DISCUSSION

In the C. punctatus exposed to 0.2 ppm of $HgCl_2$ and 0.5 ppm of MeEHgCl, the changes in the brain MAO content are not significant on the 20th day (Fig. B). However, after 40 days and on 80th day when the experiment was terminated significant reduction was noticed. Those treated with MeEHgCl exhibited more significant brain MAO inhibition on the 40th and 80th ($P < 0.001$) days than that of $HgCl_2$ (40th day $P < 0.05$, 80th day $P < 0.01$). The depletion of the whole brain MAO activity has an inverse correlation with exposure time in both $HgCl_2$ ($r = -0.771$) and MeEHgCl ($r = -0.607$) exposed groups (Fig. A).

The % inhibition of MAO was more pronounced in MeEHgCl (17.23, 16.51) exposed group than $HgCl_2$ (8.23, 11.31) on the 40th and termination day (80th) of the experiment respectively. When this experiment was started in the month of March, the gonads were in the stage I condition. But when the experiment was terminated in May, the ovary of the control fish was in the III and IV stages when vitellogenesis was almost completed and the follicular epithelium was well differentiated into the thecal and granulosa layers. The testis exhibited active spermatogenesis and sperm masses were seen in the tubular lumen. But in the treated groups, the ovary was in stages I and II where vitellogenesis was not initiated and the follicular epithelium was

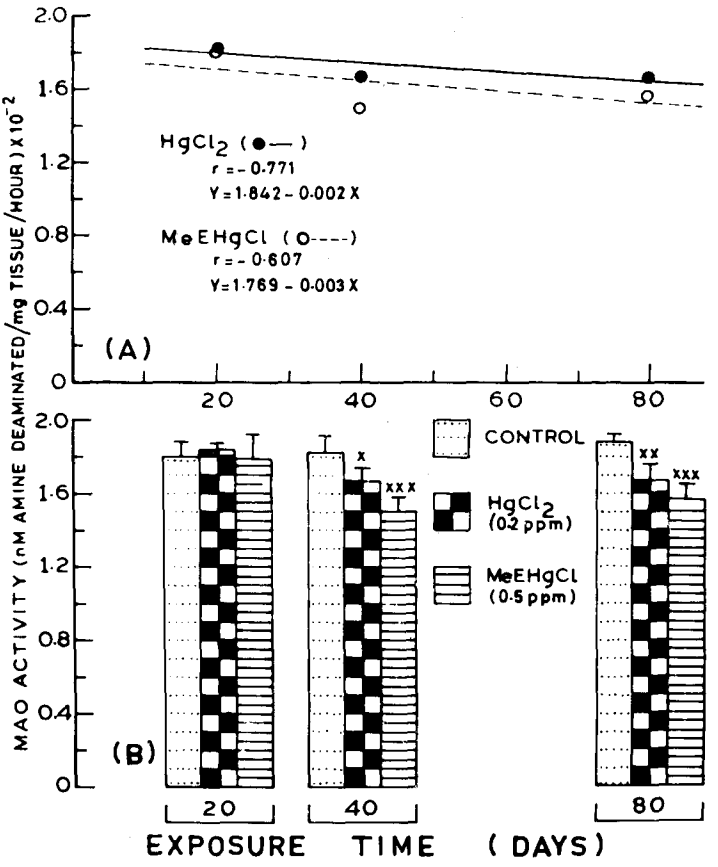

Fig. (A). Showing correlation coefficient (r) value and regression line equation between the brain MAO activity (Y) and exposure time (X).

Fig. (B). Showing changes in the brain MAO activity of mercuric chloride ($HgCl_2$) and Emisan (MeEHgCl) treated fish at different exposure intervals. Values are the mean of six fish $\pm$ SD in all the groups. (x $P < 0.05$, xx $P < 0.01$, xxx $P < 0.001$).

undifferentiated. The testis had large number of spermatogonia and spermatocytes and the spermatogenesis did not progress beyond the spermatid stage. The retarded gonadal growth was also evident by the significant reduction in G.S.I. value of treated groups. The G.S.I. values (mean ± SD) of the ovary of the control, $HgCl_2$ and MeEHgCl groups are 1.78 ± 0.077, 1.61 ± 0.065 ($P < 0.01$) and 1.56 ± 0.066 ($P < 0.002$) whereas that of the testes are 0.17 ± 0.017, 0.13 ± 0.023 ($P < 0.05$) and 0.14 ± 0.025 ($P < 0.05$), respectively.

MAO is a FSH dependent (Urry et al. 1974) mitochondrial enzyme responsible for oxidative deamination of monoamines. In the rat, based on the inverse correlation between gonadotrophins release and dopamine levels, Srivastava et al. (1980) observed that MAO may be playing an important role in the dopaminergic control of gonadotrophin release. $HgCl_2$ is suggested to block the dopaminergic, cholinergic, α-adrenergic receptor sites and also inhibit dopamine stimulated adenylcyclase activity in rat (Bondy and Agrawal 1980). Intraocular injection of $HgCl_2$ in the rat caused marked inverse dose dependent degeneration of the sympathetic adrenergic nerve plexus (Björklund 1981) and disappearance of nerve terminals in the iris, which are comparable to the action of the neurotoxic drug, 6-hydroxydopamine (Jonsson and Sachs 1969). Mercury is also known to inhibit axonal flow (Wakabayashi et al. 1976) and depolymerize microtubules (Abe et al. 1975). Neonatal exposure of rat to methylmercury caused both acute and long lasting effects on maturation of central catecholamine system which may be transmitter specific (Bartolome et al. 1982). Postnatal exposure is accompanied by permanent increase in brain seratonin level (Taylor and DiStefano 1976), decreased activity of catecholamine synthesising enzyme tryptophan hydroxylase, and breakdown of MAO and Catechol-O-methyl transferase (Tsuzuki 1981). Mercury is known to retard gonadal growth in fishes (Kihlstrom 1971; McIntyre 1973; Ram and Sathyanesan 1983). In C. punctatus significant depletion of the brain MAO is obvious after 40th day of exposure to both $HgCl_2$ and MeEHgCl. However, the later seems to be more potent which may be due to increased accumulation of organic mercury than inorganic one. Berlin et al. (1969) and Magos (1968) reported that biological cell membrane and the blood-brain-barrier discriminate against ionic and inorganic

mercury, but permit the passage of only organic mercury due to its high lipid solubility (Clarkson 1972; Vallee and Ulmer 1972).

Reduction of brain MAO in C. punctatus induced by mercurials is an indirect evidence of impairment of aminergic system. The inhibition of gonadal growth may be at least in part due to the impaired monoaminergic system responsible for modulating the hypothalamo-hypophysial gonadal axis.

Acknowledgment. The senior author is grateful to the Council of Scientific and Industrial Research of India for the award of a Senior Research Fellowship, which made this work possible.

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Received October 10,1984; accepted November 29,1984