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Protective Effect of Selenium on Methylmercury Toxicity: A Possible Mechanism

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The mutual antagonism of mercury and selenium is one of the strongest and most general examples of interactions in the trace element field, a field where such interactions are recognized among nutritionists as the rule rather than the exception. The interaction of mercury and selenium, resulting in decreased toxicity of mercury and/or selenium, has been observed in both acute and chronic studies, with earlier inorganic mercury or methylmercury salts, and with both inorganic and organic forms of adverse interactions being reported (HUCKABEE & GRIFFITH 1974). The underlying basis for this interaction undoubtedly lies in the chemical nature of the elements. Interesting scientific or technological applications of this interaction have appeared from time to time (NORDLANDER 1927; TEINEIRA *et al.* 1970).

An interaction of mercury with selenium in animals was noted over 30 years ago (GUSBERG *et al.* 1941). Not until 1967 was the effectiveness of selenium against acute poisoning with inorganic mercury discovered (PARIZEK & OSTADALOVA 1967). The effectiveness of dietary selenium against chronic toxicity of methylmercury (GANTHER *et al.* 1972; STILLINGS *et al.* 1974; POTTER & MATRONE 1974), the major form of mercury in fish, or mercuric chloride (POTTER & MATRONE 1974) was discovered independently in three different laboratories at about the same time, the first published in March, 1972 (GANTHER *et al.* 1972). This study demonstrated that 0.5 ppm of selenium as sodium selenite markedly improved growth and prolonged survival in rats given much larger amounts of methylmercury. This has subsequently been confirmed in the rat (GANTHER *et al.* 1973) and Japanese quail (GANTHER & SUNDE 1974). By means of both light and electron microscopy, this protective effect of selenium against methylmercury toxicity in both central and peripheral nervous systems has been shown in cats and in rodents (CHANG *et al.* 1977; CHANG 1982).

It is well known now that selenium is an intrinsic component of glutathione peroxidase, an antioxidase enzyme (ROTRUCK *et al.* 1973; OH *et al.* 1974) which may behave like other antioxidants, such as vitamin E and N,N'-diphenyl-p-phenylenediamine (DPPD) (WELSH 1979; CHANG *et al.* 1978; YIP & CHANG 1982) in providing protection to the neuronal tissues against methylmercury toxicity via antioxidative actions. Indeed, in methylmercury poisoned animals, the activity of glutathione peroxidase was found to be significantly suppressed (GANTHER *et al.* 1975; HIROTA *et al.* 1980).

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Our present study is designed to examine the state of glutathione peroxidase activity in methylmercury intoxicated rats with or without the influence of selenium.

METHODS

Young adult male Charles River rats were used in this experiment. Animals, in groups of 10, were divided into four groups: Group I - slaine control; Group II - injected (i.p.) with 2.0 mg/kg b.w. methylmercury chloride (MeHg); Group III - injected (i.p.) with 2.0 mg/kg b.w. sodium selenite (Se); Group IV - treated with both 2.0 mg/kg b.w. MeHg and 2.0 mg/kg b.w. Se. All animals were injected daily and were sacrificed at the 8th week of intoxication.

Blood samples were collected via orbital sinus venipuncture at weeks 3, 6 and 8 after dosing in 3.8% buffered sodium citrate tubes (Vacutainer #4857). Erythrocytic glutathione peroxidase activity was determined according to the method described by MARAL *et al.* (1977). Erythrocytes were washed three times in 0.9% saline and lysed by freeze-thaw. The rate of oxidation of NADPH with t-butylhydroperoxide as a substrate was followed at 340 nm and 25°C. The glutathione peroxidase activity was recorded as μmoles NADPH consumed/min/g hemoglobin.

RESULTS

Toxic signs (crossing reflex of the hind limbs) were displayed by MeHg-treated animals by the 6th week of intoxication. By 8 weeks of the experiment, overt neurological signs (crossing reflex, ataxic gait, and weight loss) were observed in MeHg-treated animals. No observable toxic signs or symptoms were evident in the control animals (saline or Se-treated) and in the MeHg/Se treated rats (Table 1).

Table 1. Observable Toxic Signs in Animals

	3 wk	6 wk	8 wk
Saline	-	-	-
Se	-	-	-
MeHg	-	+	++
MeHg/Se	-	-	-

Analysis of glutathione peroxidase (GSH-Px) activity in blood showed no significant change in any group of the animals after 3 weeks of intoxication. While there was still no significant deviation of GSH-Px activity in animals treated with saline, Se alone, or MeHg/Se, there was a marked reduction (35%) of this antioxidative enzyme in MeHg-treated animals after 6 weeks of poisoning (Figure 1).

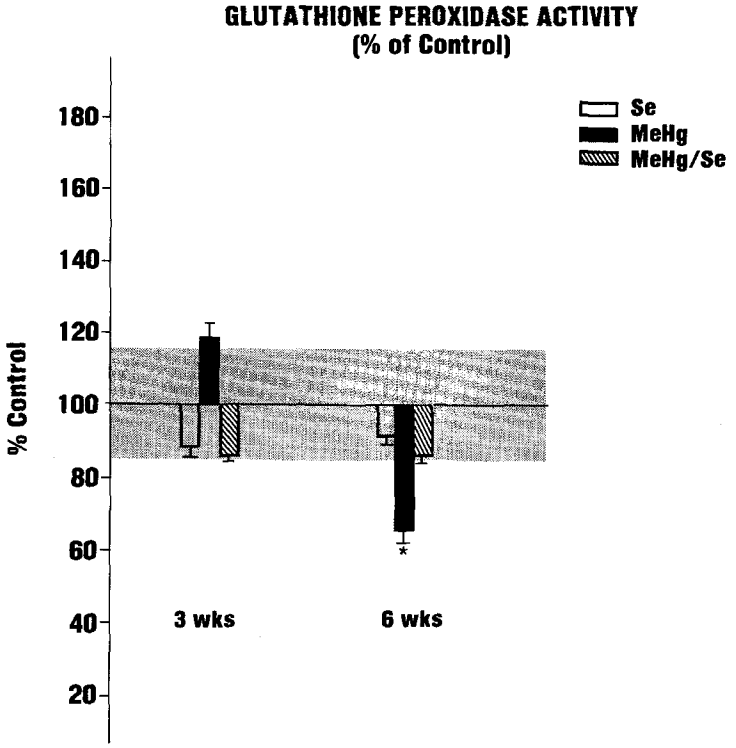

Figure 1. Blood glutathione peroxidase (GSH-Px) activity in methylmercury (MeHg)-intoxicated animals with or without the influence of selenium (Se). Although there was no significant change in GSH-Px activity after 3 weeks of exposure, a marked suppression of GSH-Px was noted in MeHg-treated animals after 6 weeks of intoxication. Animals exposed to both MeHg and Se, however, showed no significant alteration in the GSH-Px activity.

Significant change in the GSH-Px activity was still observed in the MeHg-treated animals through the 8th week of the experiment while no apparent changes were observed in the control (saline or Se) and MeHg/Se-treated animals. However, because of the poor nutritional and health status of the MeHg-treated animals at the end of the experiment, it is difficult to discern MeHg effects from other influences, and are therefore not included in our presentation.

DISCUSSION

Since the reports denoting administration of selenite decreased toxicity of methylmercury (GANTHER *et al.* 1972; STILLINGS *et al.* 1974; POTTER & MATRONE 1974; OHI *et al.* 1975; SUMINO *et al.* 1977; CHANG *et al.* 1977), these studies have also attempted to explore the

underlying mechanism for such protective phenomenon. Our present study investigates the possible role of selenium in securing the antioxidative enzyme glutathione peroxidase (GSH-Px) which is known to be suppressed in methylmercury poisoning and is important in the maintenance of cellular integrity.

In our experiment, we have confirmed that exposure to methylmercury suppressed the activity of GSH-Px as reported by previous investigators (GANTHER *et al.* 1975; HIROTA *et al.* 1980). Furthermore, we have demonstrated that with co-administration of selenium (sodium selenite), the inhibitory effect of MeHg on GSH-Px was totally alleviated. These findings provided a direct correlation on the health effects (symptomology and pathology) of methylmercury on similarly treated animals (CHANG 1982) suggesting the level of GSH-Px level is important in influencing the toxic consequences in MeHg-intoxicated animals and may be useful as a predictive indicator for methylmercury toxic conditions of the animals.

Glutathione peroxidase, a selenoprotein, may decrease the toxic effects of methylmercury by securing the integrity of the biological components of cells and tissues via antioxidation. Similar protective effects by vitamin E, another antioxidant, against methylmercury toxicity has also been reported (CHANG *et al.* 1978; WELSH 1979; YIP & CHANG 1982). These findings tend to provide some support to the proposal that radical production by methylmercury in the biological system may constitute, at least in part, the pathogenetic mechanisms of methylmercury intoxication (GANTHER 1978).

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