Tohoku J. Exp. Med., 2002, **196**, 71-77

Review

Modification of Mercury Toxicity by Selenium: Practical Importance?

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WATANABE, C. *Modification of Mercury Toxicity by Selenium: Practical Importance?* Tohoku J. Exp. Med., 2002, **196** (2), 71-77 —— The interaction between mercury and selenium may involve a variety of toxicologically and biochemically distinct processes. In this paper, the interaction between inorganic mercury and sodium selenite, the interaction most extensively studied, as well as the interaction between methylmercury (MeHg) and selenium, the interaction perhaps most significant for non-occupational human populations, will be discussed. It has been shown that the former interaction can be understood as a modification of the kinetic behavior of inorganic mercury by selenite, but this interaction may occur only under very limited conditions. On the other hand, the mechanism of the latter interaction is largely unknown, and kinetic modification appears to play only a minor role. An interaction between MeHg and selenoproteins or a possible interaction between the inorganic mercury, resulting from the demethylation of MeHg, and the selenium may be important. Compared to the experimental findings, little evidence of the toxicological modification of MeHg by selenium was obtained in epidemiological studies. ——— selenium; inorganic mercury; methylmercury; selenoproteins; interaction

Research on the interaction between selenium (Se) and mercury (Hg) has a relatively long history. Parizek and Ostadalova (1967) reported on the alleviation of the lethal toxicity of mercuric chloride by sodium selenite (SS) simultaneously administered to rats. While this report is recognized as the first on this subject, there are some observations reported prior to Parizek and Ostadalova's paper (1967) that might have led to the earlier commencement of research in this area. For example, it was reported that in the tissues of Minamata victims, unusually high concentrations of not only Hg but also Se were found. It was twelve years after this report that Ganther et al. (1972) showed the mitigating effect of SS on the tox-

Received January 15, 2002; revision accepted for publication February 28, 2002.
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This paper was presented at the Sendai Symposium: Paradigm Shift in Heavy Metal Toxicology held on November 21-22, 2001.

icity of methylmercury. Another important earlier finding is that both Hg and Se co-accumulated in the autopsied tissues (including brain) of mercury miners (Kosta et al. 1975). As a result of these early studies, much research has been carried out regarding the mutual interaction of these two elements.

It should be noted here that many heterogeneous phenomena could be classified as interactions between Se and Hg. There are three types of Hg that have toxicological relevance to humans: methylmercury (MeHg), elemental mercury ("mercury vapor"; Hg^0), and inorganic mercury salts (I-Hg). Risk-associated exposure to Hg^0 or I-Hg is largely confined to occupational settings, while non-occupational exposure to MeHg occurs in the general population through dietary consumption of fish. On the other hand, Se is an essential micronutrient existing in the mammalian body in association with a variety of selenoproteins. In addition, some of the low-molecular-weight (LMW) Se compounds have been shown to possess biological activities that might explain the anticancer property of Se. The mode of interaction between Hg and Se would thus vary depending on the particular combination of the chemical forms of both elements. In this paper, the interaction between I-Hg and Se, the interaction most extensively studied, and the interaction between MeHg and Se, the interaction most relevant to human health, are discussed.

Interactions between I-Hg and Se

As stated above, alleviation of I-Hg toxicity by SS (a LMW Se compound) was reported as early as 1967. Since then, many studies dealing with this phenomenon have been published. Among such studies, Imura and Naganuma's group (Imura and Naganuma 1991) conducted a series of experiments to elucidate how and why these two compounds interact with each other. As summarized in their review, when I-Hg and SS are co-administered, i) SS forms a high-molecular-weight (HMW) complex with Hg and a plasma protein, thereby substantially reducing the accumulation of Hg in the kidney, the target organ; ii) it also forms an inert HMW complex in the RBC and stays there without doing harm to other tissues; and iii) if forms other stable complexes in such organs as kidney or liver. Recently, the plasma protein to which the Hg-Se complex attaches has been identified as selenoprotein P, the major plasma selenoprotein (Yoneda and Suzuki 1997).

While the toxicity alleviation is distinct in the case of the co-administration of I-Hg and SS, as mentioned above, for this interaction to occur, precise timing of the dosing is required. The alleviation of I-Hg toxicity could be observed when both compounds were administered simultaneously; however, when the interval between I-Hg injection and the subsequent SS injection was even as short as one hour, the toxicity-alleviating effects disappeared (Naganuma et al. 1984) and in its place, an aggravating effect was observed. In addition, one requirement for the alleviation to occur is that both chemicals should be given i.v. at (near) equimolar doses.

Magos et al. (1984) examined the effects of the chemical forms of Se on this interaction. When the SS was replaced by "biological Se", the powdered liver of the rat administered SS, the formation of the Hg-Se complex was significantly limited and the marked reduction in the renal accumulation of Hg observed upon co-administration of SS was not seen.

Thus, the interaction between I-Hg and SS as depicted above appears to occur only in very limited situations, which considerably lessens the potential of SS as an antidote for I-Hg poisoning (Magos 1991). Nevertheless the interaction may be important for human health, as will be discussed later.

In an experiment in which the tissue concentrations of total Se are monitored after I-Hg injection in mice, a transient increase in Se concentration in liver and kidney was observed

one to three hours after the injection. Since the molecular ratio of Se : Hg far exceeded 1 : 1, and since the increases in the Hg concentrations in these organs were not transient, the increase in the Se concentration could not be explained by the formation of the Hg : Se complex. This result suggests a possible interaction of I-Hg with pre-existing Se in the mammalian body.

Interaction between MeHg and Se: toxicity

Ganther et al. (1972) were the first to report the interaction of MeHg and Se. Their results clearly showed the alleviating effects of SS on MeHg-induced mortality and the suppression of weight gain in rats. The alleviating effects of SS can also be observed in terms of neurotoxicological endpoints; mice depleted of tissue Se following consumption of a low-Se diet exhibited enhanced neurotoxicity in response to MeHg exposure, as evidenced by hind-leg crossing, a typical symptom of MeHg poisoning in rodents, compared to Se-sufficient mice (Imura 1986). The mechanisms underlying this enhanced neurotoxicity, however, are not clear.

Since mammals are most susceptible to MeHg exposure during fetal development, the effects of the supplementation or depletion of Se during pregnancy on MeHg toxicity were also investigated. In an earlier work, mice were administered MeHg and/or SS before and during pregnancy and throughout the suckling period. Depending on the combination of the doses of the two chemicals, both enhancement and suppression of fetotoxicity were observed, and when the fetotoxicity increased, increased teratogenicity was also noted (Nobunaga et al. 1979). Deficiency of Se in mice enhanced fetolethality of maternally administered MeHg compared with the Se-sufficient group (Nishikido et al. 1987). Using a similar experimental protocol, the functional development of MeHg-exposed rodents was reported. The offspring of mice co-injected with MeHg and SS during pregnancy showed slightly improved behavioral development compared to the offspring of the MeHg-only group in the very early postnatal period (Satoh et al. 1985). When rats were supplied with SS through diet, the MeHg-induced behavioral effects in their offspring were alleviated compared to the offspring MeHg-only group (Fredriksson et al. 1993). Another study showed that in Se-deficient mice, development of the walking behavior and open-field activity were affected by MeHg exposure to a greater degree than in Se-sufficient mice (Watanabe et al. 1999a). The interpretation of the results was, however, complicated because the Se depletion per se induced some behavioral effects.

Thus, the results of these studies suggest that when SS is co-administered with MeHg, the fetotoxicity, neurotoxicity, or developmental toxicity of MeHg is alleviated, and the toxicity is enhanced in Se-deficient animals, although an exception to this finding exists (e.g., Nobunaga et al. 1979).

Interaction between MeHg and Se: kinetic studies

In studies on the kinetics of the interaction, the kinetic modification of Hg per se appears to be of minor importance, particularly regarding prenatal exposure. Two of the reports mentioned above (Fredriksson et al. 1993; Watanabe et al. 1999a) showed that the distribution of Hg to the fetal/neonatal brain was essentially unchanged when the dietary level of Se was reduced or increased.

Similar to I-Hg, MeHg reacts with co-administered SS to form a complex, bis (methylmercuric) selenide (BMS), both in the bloodstream and in tissues such as the brain, liver, and kidney. BMS, in contrast to the Hg-Se complex mentioned above, unstable and rapidly eliminated from tissues. The toxicological significance of the formation of BMS has not been elucidated thus far.

Other than reports on the formation of the BMS complex, there have been few reports on the kinetic interaction between MeHg and Se.

It is known that SS reduces the biliary excretion of MeHg, an important pathway in enterohepatic circulation. Recently, it was shown that this reduced biliary excretion is not due to the formation of a complex between MeHg and SS (Urano et al. 1997). In their experimental system, however, the hepatic Hg level was not modified by the presence of SS. Interestingly, MeHg suppresses the biliary secretion of exogenous Se (administered as SS), while it increases the exhalation of methylated Se compounds. S-adenosyl methionine (SAM), the common methyl donor used for methylating selenide to the volatile (exhalable) form, is required at relatively lower concentrations in SS+MeHg treatment than in SS-only treatment. Although the underlying mechanism is unknown, the authors suggested that MeHg might be acting as the methyl donor, thereby reducing the concentration of SAM required for methylation. The net result of this, if true, would be the demethylation of MeHg by selenide, which then could lead to other interactions between selenide molecules and newly formed I-Hg (Gregus et al. 2001).

Interaction between MeHg and endogenous Se

Almost all of the Se in the mammalian body is present in the form of selenoproteins. There are more than 10 selenoproteins with or without known function(s), the levels of which are influenced by the dietary intake of Se to various extents; i.e., some respond very quickly to dietary Se, while others hardly respond at all. It was shown in earlier studies that the activity of glutathione peroxidase (GPx), a selenoprotein acting as an antioxidant, can be inhibited by MeHg. However, the dose of MeHg required to inhibit the activity of this selenoenzyme is relatively high.

In mice prenatally exposed to MeHg, the hepatic concentration of Se was increased, while the activity of GPx was inhibited, suggesting that the availability of Se in this organ was decreased for unknown reasons (Nishikido et al. 1987). Using a similar experimental design, it was shown that maternal exposure to MeHg decreased both Se concentration and GPx activity in the brain of neonatal mice. In a subsequent study, it was shown that prenatal exposure to MeHg affected the activities of the iodothyronine deiodinases (DIs), another class of selenoproteins, in the mother-fetus complex. The effects are somewhat complex; in the MeHg-treated group, while the 5-DI activity in the placenta was enhanced, that in the fetal brain was suppressed and the 5′-DI activity was marginally enhanced. Since 5-DI inactivates and 5′-DI activates thyroid hormones, the entire pattern of the enzymatic changes mimicks those observed in hypothyroid animals. Since the T4 level was normal in the fetal plasma, these enzymatic changes might lead to an abnormal and presumably harmful excess of thyroid hormones in the fetal brain (Watanabe et al. 1999b) It should also be noted that relatively small changes in these enzyme activities could be associated with the change of thyroid hormone levels in the brain (Campos-Barros et al. 1997). Although the doses used in these studies were relatively high and administered using a small number of injections, we have observed, in a preliminary experiment, that GPx activity in the fetal brain was suppressed by 0.1 mg/kg/day of MeHg given to the pregnant animals from GD 8 to 17 (Watanabe et al., unpublished data).

Thus, the effects of MeHg on "endogenous" Se might contribute to the neurotoxicity of MeHg in fetus/neonates. The dose-response relationships, the mechanisms and consequences of each enzyme activity change, as well as the effects on the other selenoproteins found in the brain, such as selenoprotein P and the thioredoxin reductases, should be further investigated.

Relevance of the I-Hg-SS interaction to MeHg toxicity

It is known that some of the MeHg in the

mammalian body undergoes demethylation to form I-Hg. Although the exact mechanism of the demethylation is unclear, it has been suggested that oxygen radicals might play a role in this process (Hirayama and Yasutake 2001). In the brains of monkeys chronically fed (6–18 mos) MeHg, high concentrations of I-Hg were detected, the I-Hg presumably being formed as a result of demethylation (Bjorkman et al. 1995). Interestingly, there was a positive correlation between I-Hg level and Se level, but only a negative correlation was obtained between MeHg and Se levels. This suggested the formation of an inert Hg-Se complex in the brain, which may be regarded as a process of detoxification. Since the only source of Se was dietary in these MeHg-dosed monkeys, the interaction might occur between dietary Se and MeHg. Furthermore, although the formation of Hg–Se complex has not been experimentally determined, if this were the case, selenide, produced as a result of the seleno-amino acid metabolism, could interact with I-Hg resulting from the demethylation of MeHg in the brain. Both selenide and mercuric ions are highly reactive and do not exist in the free form for very long. Therefore, it may be necessary that these compounds co-exist in a relatively limited compartment so that both can achieve significant concentrations to facilitate their interaction with each other. A significant proportion of the Hg present in the body was found in the autopsied brains of humans without known occupational exposure to Hg (Matsuo et al. 1989).

Interaction between MeHg and Se: human populations

The impact of fish consumption by pregnant women on the fetus is currently a public health concern. The results of the two largest prospective cohort studies have led to disagreement regarding whether or not fish consumption by pregnant mothers adversely affects their babies. Since these studies, although similar in many respects, differ in terms of subject population, age at examination, neurobehaviors studied, and pattern of fish eating, their conflicting results should not be taken as an indication that either study was defective. Rather, it has been suggested that confounding factors including nutrition should be assessed more precisely to account for the differences (NIEHS 1999).

The intake of Se could be among the nutritional factors that modify the toxicity of MeHg. The practical significance of Se in the chronically MeHg-exposed human population, however, has not been established. Among the Faroe islanders, who take in substantial quantities of MeHg as a result of consumption of whale meat, a positive correlation was observed between blood Hg and blood Se levels (Grandjean et al. 1995). In a subsequent report (Steuerwald et al. 2000), the modifying effect of Se on the neurological effects of prenatal MeHg exposure was suggested to be minimal although supporting statistical evidence was weak. Another ongoing large-scale study being conducted in the Seychelles has not considered the nutritional status of Se. It is noteworthy that in a Latvian fish-consuming population, the plasma TSH (thyroid stimulating hormone) level was negatively correlated with the plasma Se level was level, while the plasma Se correlated with the frequency of fish consumption. In this population, the level of plasma Se was low, and the authors suggested a higher rate of TSH secretion at low plasma Se levels. Unfortunately, Hg levels were not measured in this population (Hagmar et al. 1998).

Future studies

As outlined above, the interactions between Se and Hg may include several different phenomena. From the perspective of environmental health, priority should be given to the interaction between MeHg and Se. To elucidate the mechanism and significance of the interaction, several issues need to be addressed. First, the

chemical form(s) of Se in fish should be identified. Several recent studies have suggested that Se in fish muscle is protein-bound and in the form of selenocystein. Second, the relative importance of the demethylation of MeHg with respect to both its toxicity and interaction should be quantitatively determined. Finally, the possible interaction with Hg involving one or several selenoproteins should also be evaluated.

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