Methylmercury: Significance of Intrauterine and Postnatal Exposures

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Outbreaks of methylmercury poisoning in Japan and Iraq have demonstrated the sensitivity of the fetus to neurotoxic effects. Based on toxicokinetics and considerations of practicability, the optimal biomarker of methylmercury exposure is the hair concentration, but whole-blood measurements of mercury are also useful. Dose–response relations are still incompletely known, especially concerning developmental neurotoxicity under conditions of chronic exposure. Available evidence indicates that neurobehavioral dysfunction in children may occur if the maternal mercury concentration in hair is >6 μg/g (30 nmol/g). This value corresponds to a blood mercury concentration of ~24 μg/L (120 nmol/L). The period of maximum sensitivity of the nervous system to methylmercury toxicity is unknown, but the transfer of mercury to the newborn through human milk may represent an additional risk. In view of the wide occurrence of mercury contamination in developing countries, increased use of the exposure biomarkers is encouraged.

Indexing Terms: environmental exposure/hair analysis/neurotoxicity/pregnancy/toxicology

Since 1955, a severe neurological disease has occurred in a large number of people living at Minamata Bay, Kyushu, Japan. Although cases were seen in all age groups, infants appeared to be the most severely affected; they showed a syndrome of cerebral paresis, which at a later stage was associated with ataxia, mental retardation, dysarthria, and hypersalivation. By 1958, it was clear that the disease was caused by chemical pollution of Minamata Bay by methylmercury (1).

In infants the disease was often not recognized until at least 3 months after birth, at a time when the child was still being nursed. However, a surprising observation was that many of the mothers appeared healthy (1). Could toxic amounts of a chemical be transferred across the placenta or into human milk? These were controversial notions in the 1950s.

Other research has documented that the nervous system is particularly vulnerable to toxic effects from environmental toxicants during the last two trimesters of pregnancy and during early postnatal life (2). Experimental studies have also shown that methylmercury can pass the placental barrier and result in even higher concentrations in the fetus than in the mother (3).

Serious poisonings have also occurred as a result of the accidental use of methylmercury-treated seed to make bread, e.g., in Iraq (4). In addition, methylmercury is a widespread pollutant that is a health hazard to large population groups that rely on seafood. The clinical and epidemiological information from the unfortunate poisoning episodes (1, 4) should therefore be utilized to design strategies to prevent toxic exposures to this compound. In this regard, biomarkers of methylmercury exposure are likely to play a key role.

Choice of Exposure Markers

Several detection methods are available for measuring mercury and its compounds in biological media (5). Therefore, the optimal exposure marker can be chosen on the basis of toxicokinetics and consideration of practicality.

Methylmercury becomes distributed relatively evenly in the body, and short-term elimination of the compound approximates to simple first-order kinetics (6). While urinary mercury excretion is a good indicator of exposure to inorganic mercury, it does not reflect exposure to methylmercury. In this case, the most useful samples for analysis are likely to be hair and blood.

Laboratory animal studies involving tracer techniques have shown that, following acute dosage with methylmercury, blood mercury concentrations will initially reflect organ concentrations reasonably well, but, with time, an increasing fraction of the body burden will be in the brain, muscles, and kidney. Also, the concentration of total mercury in the blood, and especially in serum, is affected by exposure to elemental mercury and inorganic mercury compounds. In populations exposed mainly to methylmercury, account must be taken of the fact that umbilical cord blood contains 20%–30% higher concentrations of mercury than does maternal blood (6).

Sex differences in whole-body clearance and tissue distribution of intestinally absorbed methylmercury have been demonstrated in rodents (7, 8). In particular, a sex-related difference has been observed in the ratio between concentrations of mercury in blood and in potential target organs. Unless this factor is taken into account, the proportionately lower blood concentrations observed in male animals will cause an underestimation of the true body burden. Interestingly, a recent study of blood mercury concentrations in a random population sample showed significantly higher values in women than in men, even when exposure factors were considered (9). Accordingly, attention must be paid to a possible sex-related difference in the toxicokinetics of mercury in humans.

Methylmercury is apparently incorporated into hair that is being formed in the follicle. On the basis of the growth rate of hair, the concentrations of methylmer-
cury in the hair strands in each 1-cm segment of scalp represents exposure for \(-1\) month (6). However, hair mercury concentrations may be augmented by the binding of exogenous mercury to the surface, and very high concentrations of total mercury in hair have been documented in workers exposed to mercury vapor (10). Currently, there is no way of reliably separating this contamination from the proportion of mercury that was originally incorporated in the follicle.

Under reasonably constant conditions of exposure, hair mercury concentrations in humans average \(-250\)-fold the whole-blood mercury concentration (6). This ratio varies, probably in part due to individual differences in hair structure and growth rate, and because concomitant oral exposure to inorganic mercury adds more to the concentration in blood than in hair. Also, as discussed above, the ratio could vary between men and women. Mercury concentrations in cord blood and in maternal scalp hair correlate quite well (Fig. 1); the single point in the upper left quadrant could very well represent a case of exogenous contamination of the hair.

The elimination half-life of methylmercury has been determined from blood concentrations in humans following accidental intake of high doses or after experimental exposure to tracer doses. The average half-life is \(-60\) days (11), but it decreases to almost one-third during lactation, possibly due to the excretion of mercury in the milk (12). Analyses of sequential segments of hair have shown similar results (4, 12).

Mercury concentrations in other biological samples are rarely of interest as biomarkers of exposure. However, due to the particular susceptibility of the fetus and the newborn to methylmercury toxicity, samples of placenta, umbilical cord, or milk may occasionally be analyzed to obtain an estimate of the absorbed dose.

When the disease occurred in Minamata, the cause was not known, and nobody knew which types of samples would be needed for later analysis. However, in this particular area of Japan, an ancient custom was to dry and preserve the umbilical cord and keep it in a box. When methylmercury eventually appeared as a likely cause of the disease, umbilical cords were collected from 12 patients who had been born with congenital Minamata disease, from 16 children with other mental disturbances, and from 64 other children of the same area who served as controls (1). The methylmercury concentrations were generally much higher in the cords of Minamata disease patients, but the distributions overlapped considerably between the three groups. Because the diagnostic criteria were not described in the published paper, the extent of possible misclassification is difficult to assess.

To relate these cord concentrations to more useful hair mercury concentrations, a correlation has been established between the total mercury concentrations in the two types of samples obtained from stratified sampling of 50 births in the Faroe Islands (13). From the equation for the regression line, the hair mercury concentration (in \(\mu g/g\)) could be calculated as \(19.5 \times \text{cord mercury (in } \mu g/g \text{ dry weight}) + 3.6\). (The concentration in \(\mu g/g\) can be converted to nmol/g by multiplying the result by 5.0.) Using the graph published by Harada (1), one finds that the median mercury concentration in the umbilical cords from Minamata would correspond to \(-22.8 \mu g/g\) (114 nmol/g) maternal hair (Fig. 2). However, this value may not necessarily represent the true median in mothers of patients who had congenital Minamata disease because the group studied by Harada may have included patients who did not actually have the disease. Also, the mercury concentration in the umbilical cord at birth may not accurately reflect the exposure that caused the disease. Further, Dalgård et al. (13) measured total mercury, rather than methylmercury, and any significant occurrence of inorganic mercury in the cord tissue would therefore result in an underestimation of the hair mercury concentration in the Japanese mothers.

Human milk contains trace amounts of mercury, partly in the form of methylmercury. Mercury concentrations in human milk correspond to \(-8\%) of the concentration in whole blood (4, 14), but some of the mercury occurs in the form of mercuric ion, in particular when exposure to inorganic mercury has occurred (15). In Iraq human milk was the suspected source of methylmercury exposure of several infants who developed symptoms of methylmercury poisoning during the nursing period (14).

### Early Effects

For preventive purposes, documentation should be obtained to identify the methylmercury exposure concentration that causes the earliest adverse effects. By keeping human exposures below that threshold, the population (or some statistically defined large proportion) will be protected against toxicity. With methylmercury, the difficult task is to determine the exact character of such early dysfunctions and then to assess
their dependence on methylmercury exposure prenatally and postnatally.

Much information has been gathered from the episodes of methylmercury poisoning in Japan and Iraq. In the patients with congenital Minamata disease, motor functions have improved with time, whereas mental retardation has emerged as the main sign of the disease in older children. In less seriously exposed children, cognitive functions may have been impaired by methylmercury. Thus, a study conducted in 1962 reported that, of 72 children born in Minamata in 1953–54 and 1959 (i.e., just before and just after the highest pollution period), 21 children (29%) had an IQ <70 (1). Nine years later, school children born in the Minamata area during 1955–59 were examined, and high prevalences of mental retardation, sensory disturbance, mild dysarthria, and adiadochokinesia were observed (1). Unfortunately, the epidemiological designs do not appear to be ideal, and detailed information on the methods used is not available. However, the data seem to present qualitative evidence that methylmercury exposure in the Minamata area also caused more nonspecific neurobehavioral dysfunction in children.

From data collected in Iraq (16), a dose–response relation was established between the peak mercury concentrations in maternal hair during pregnancy, and the prevalence of psychomotor retardation (delayed walking and talking), as well as an increased number of neurological signs in children, particularly in boys (6, 17). In Fig. 2, mercury concentrations in maternal hair are given for children with suspected methylmercury toxicity and for control children. It is noteworthy that the mercury concentrations measured in Iraq are much higher than those calculated for the Minamata cases. Only part of this difference can be explained by the fact that the data from Iraq represent peak values, whereas those calculated for Minamata would reflect means over time. The signs identified in Iraqi patients were partly recognized by interviewing the mothers, and, although these effects may not be considered the earliest indications of methylmercury neurotoxicity, the Iraqi patients were probably, on average, less severely affected than the group of patients from Minamata. Again, the epidemiological design of this study was not ideal, and the limited number of observations results in wide calculated confidence limits for the dose-response relation.

In most cases of congenital methylmercury poisoning, the diagnosis was not made at birth. That the problem was not recognized until months later may not necessarily mean that postnatal exposures played a significant causative role. The nervous system is not fully developed at birth, and only a crude examination of its integrity can be made at that time. Effects incurred prenatally may not become apparent until the nervous system has matured sufficiently to express the dysfunctions. Indeed, some neurobehavioral functions may only be assessed if the child is able to cooperate with the examiner. Accordingly, the evidence available so far does not allow any detailed characterization of the more subtle forms of neurotoxicity caused by methylmercury.

The degree of sensitivity during the early postnatal period is also unclear. Methylmercury has a particularly long elimination half-time in infants (12, 18), and considerable amounts of mercury can be transferred through breastfeeding (18). Surprisingly, mercury from human milk gave rise to blood mercury concentrations >1000 μg/L (5000 nmol/L) in five infants during the episode in Iraq, but none of them showed immediate signs of poisoning (12). Concentrations of that magnitude have been associated with methylmercury poisoning in adults (4). Unfortunately, no long-term follow-up of these subjects is available.

Methylmercury poisoning in adults has been well described from Japan and Iraq (6). However, studies in fish-eating populations elsewhere (19–21) have failed to identify any individual with a clear-cut case of methylmercury poisoning, despite blood mercury concentrations within the lower range seen in adult patients in the Iraqi outbreak (4). The reason for this absence of apparent toxicity is unknown.
Among blood whether Acceptable Concentrations

The result of a biomarker assay for methylmercury must be viewed in a toxicokinetic perspective, i.e., whether the exposure was acute or chronic, the route of exposure, and the speciation of mercury. For long-term methylmercury exposures, mercury concentrations in blood or hair should be evaluated on the basis of epidemiological evidence of dose-response relationships for neurotoxicity.

The data from the outbreaks in Japan and Iraq have some limitations, in particular because of the nonspecific character of the first signs of insidious toxicity, and may for that reason overestimate the threshold for early neurotoxicity. Nonetheless, a detailed evaluation of the information from Iraq (6, 17) suggests that neurological dysfunction may develop when maternal peak hair mercury concentrations are >10–20 μg/g (50–100 nmol/g). Some supporting evidence is available from two other studies of children prenatally exposed to methylmercury (22, 23).

In a study of 234 Cree Indian children, ages 12–30 months (22), the mother's hair was analyzed in 1-cm segments; the peak mercury concentration during the pregnancy period was used as the exposure index. Among several neurological measures, supplemented by the Denver developmental scale, only abnormal muscle tone or reflexes in boys was related to the mercury exposure index, though not in a clear dose-dependent way (22).

In a population-based study in New Zealand (23, 24), ~1000 mothers consumed fish for dinner >three times per week during pregnancy, and 73 of them had hair mercury concentrations >6 μg/g (30 nmol/g). At 4 years of age, 31 of their children were examined along with matched controls who had low levels of prenatal mercury exposure; in the Denver test, abnormal or questionable results occurred three times more often in the exposed group (23). At 6–7 years of age, 61 of the children and three control groups were tested with the Wechsler Intelligence Scale for Children; an average maternal hair mercury concentration of ~15 μg/g (75 nmol/g) was associated with decreased test performance (24).

When evaluating all the available evidence, one must take account of several factors that could potentially bias the results toward the null hypothesis (25). Thus, ingestion of alcohol results in a decrease of the blood mercury concentration while at the same time causing a risk of neurological damage to the fetus. Ingestion of fatty fish that contain essential polyunsaturated fatty acids seems to cause an increased gestation time and a higher birth weight—two factors that are associated with a decreased risk of neurological dysfunction in the infant. The hair mercury concentration as a marker of seafood intake also serves effectively as an indicator of exposure to other neurotoxic contaminants, such as polychlorinated biphenyls, that are likely to occur in marine biota. In addition, ethnic differences and factors related to the subjects' socioeconomic situations could cause potential confounding.

On the basis of the early outbreaks of methylmercury-related disease, a Provisional Tolerable Weekly Intake (PTWI) was determined by an FAO/WHO expert group; a PTWI of 0.3 mg of mercury (of which no more than 0.2 mg may be methylmercury) for adults is estimated to protect against toxic effects (6). According to data obtained by Sherlock et al. (26), 0.2 mg of methylmercury per week, i.e., ~30 μg (150 nmol) per day, would, at steady state, result in a blood mercury concentration of 24 μg/L (120 nmol/L) or a hair mercury concentration of ~6 μg/g (30 nmol/g). These values are well below the concentrations associated with clinical effects in adults. However, considering the aforementioned uncertainties and the fact that, for example, the high exposure group in the New Zealand study had maternal hair mercury concentrations >6 μg/g (30 nmol/g) (23, 24), the safety margin offered by the PTWI for prenatal exposures could be very slim.

While recognizing the uncertainties, the PTWI can also be translated into an approximate limit for mercury in human milk. If the standard adult person weighs 70 kg, the PTWI corresponds to a daily dose of ~0.6 μg (3 nmol) mercury per kg body weight. For infants, the average milk intake during the nursing period is ~125 ml/kg body weight per day (27). An intake corresponding to the PTWI on a per kilogram basis would then occur if the milk mercury concentration were 4.8 μg/L (24 nmol/L).

Current Extent of the Problem

Mercury is a natural constituent of the environment and is continuously released from many sources, e.g., volcanos. Anthropogenic releases originate from chloralkali plants, paper mills, and gold-mining operations. Occupational exposures to methylmercury per se are rare, and its use in antifungal treatment of grain is nowadays quite limited. Thus, environmental sources are most important for the population at large. In the aquatic environment, mercuric ions are methylated, and the methylmercury ion is accumulated in fish (6). Populations dependent on fish are therefore likely to absorb increased amounts of this form of mercury. In Sweden, erythrocyte mercury concentrations up to 1100 μg/L (5500 nmol/L) have been reported in adults who frequently ate freshwater fish from contaminated lakes (19). Similarly, highly increased mercury concentrations have been documented in other fish-eating populations, e.g., from Peru (20).

Methylmercury shows some degree of biomagnification so that species close to the top of the food chain have relatively high body burdens. Far from sources of pollution where marine mammals form part of the human diet, population groups from Alaska (28), northern Canada (29), Faroe Islands (30), and Greenland (31) show very high concentrations of mercury in blood, hair, and milk.

Mercury deposited in lakes is apparently quite persistent, and a decreasing water pH may even accelerate the release of mercury from the sediments. Releases are also seen in reservoirs created by hydroelectric power.
plants. As mercury concentrations in fish may remain high for a very long time, nutritional counseling offers the major avenue for preventive efforts in these instances. However, consumers may not necessarily be able to distinguish fish with high contaminant concentrations from those that are acceptable. Also, such advice may not be appropriate in the Arctic where other sources of food are not readily available, and where local populations have relied on a seafood diet for centuries.

Although the release of anthropogenic mercury to the environment is being controlled in most industrialized countries, serious contamination occurs in other parts of the world. Many of these problems are local in scale, but one major problem is currently developing in the Amazon basin. Mercury is used in the mining and extraction of gold, and during the 1980s at least 1000 tons of mercury were lost from the sluices or released from the burning of amalgam. High mercury concentrations in fish have already been discovered (32), and the hair mercury concentration increases steeply with the number of fish dinners per week (33). A large part of the Amazon basin may now be seriously polluted with this metal. Such situations obviously call for a systematic use of exposure biomarkers to monitor the extent and temporal development of the problems as a basis for decisions on intervention.

Strategy for Use of Biomarkers

In each situation, the proper choice of an exposure biomarker must be based on all available information about the sources of exposure, the route of entry, the mercury speciation, and the population at risk. Practical considerations, cost, and ethical constraints must be considered as well.

Often, scalp hair samples will be the preferred type of specimen, in so far as the collection procedure is noninvasive and storage and transportation present limited problems. These advantages are especially important in developing countries where other biological samples may be subject to decomposition before they reach the analytical laboratory. Following oral exposure, the hair mercury concentration is generally taken to reflect the body burden of methylmercury (6). However, as exogenous contamination of the hair may occur, e.g., from mercury vapor, and as scalp hair is not always available from the subjects, other samples may need to be considered. (In some cases, pubic or other body hair has been used for analysis.) A particular disadvantage is that a hair sample will not reflect a recent increased exposure.

Hair samples should include full-length hair strands cut with a pair of sharp scissors close to the root in the occipital area of the scalp. For most methods, the samples should preferably weigh at least 100 mg; this size would also allow segmental analysis by some methods. The hair should be tied close to the root with a cotton string or plastic clamp and saved in a marked plastic bag. The subject's normal hair treatment should be recorded.

Methylmercury absorption leads to increased concentrations of mercury in erythrocytes; inorganic mercury compounds mainly cause an increase in the serum concentrations. Although the erythrocyte mercury concentration is more specific for methylmercury exposure, whole blood is generally used for practical reasons. If exposure to mercury vapor or inorganic mercury compounds is suspected, then a serum sample should also be analyzed. All blood samples should be collected by venipuncture into evacuated anticoagulant-containing vials that are certified to be free of mercury. Likewise, other biological samples must be collected under controlled conditions with proper prevention of contamination.

For many years, a commonly used analytical method was the one first described by Magos and Clarkson (34), which allows discrimination between total mercury and inorganic mercury by addition to the reaction vessel of both CdCl₂ and SnCl₂, or the latter only. The elemental mercury so generated is aspirated through the detection cell of an ultraviolet absorbometer. Other methods based on similar principles have utilized cold-vapor atomic absorption instrumentation (35). Better detection limits can be obtained by plating mercury vapor onto a gold filter, from which it is then released by heating (36). The organomercury component can be quantified by several means, including radiochemistry, and accurate results have been produced in experienced laboratories (37).

The technique of standard additions may be necessary with methods where interference occurs. All samples should preferably be measured at least twice. Blanks and working standards should be determined at the beginning and at the end of each analytical run. Quality-assurance samples at relevant concentrations must also be included to monitor precision and accuracy. Suitable reference materials may be obtained from the National Institute of Standards and Technology (Gaithersburg, MD), e.g., freeze-dried urine (#2672a) and bovine liver (#1577a), or from Behringwerke (Marburg, Germany), i.e., Control Blood for Metals. Samples of powdered hair were at one point available from the International Atomic Energy Agency (Vienna, Austria), but reference materials for hair mercury at relevant concentrations do not seem to exist at present. Participation in interlaboratory comparison programs, such as the routine distribution of blood samples organized by Le Centre de Toxicologie du Québec (Montréal, Canada), is highly recommended. Informal quality-control programs for hair mercury (38) have been very useful and ought to be organized on a more permanent basis.

References


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