Toxicology and Applied Pharmacology **171,** 50–60 (2001)
doi:10.1006/taap.2000.9113, available online at http://www.idealibrary.com on IDEAL®

A Toxicokinetic Model for Predicting the Tissue Distribution and Elimination of Organic and Inorganic Mercury Following Exposure to Methyl Mercury in Animals and Humans. II. Application and Validation of the Model in Humans

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Received July 21, 2000; accepted November 29, 2000

A Toxicokinetic Model for Predicting the Tissue Distribution and Elimination of Organic and Inorganic Mercury Following Exposure to Methyl Mercury in Animals and Humans. II. Application and Validation of the Model in Humans. Carrier, G., Bouchard, M., Brunet, R. C., and Caza, M. (2001). *Toxicol. Appl. Pharmacol.* **171, 50–60.**

The objective of this study was to develop a biologically based dynamical model describing the disposition kinetics of methyl mercury and its inorganic mercury metabolites in humans following different methyl mercury exposure scenarios. The model conceptual and functional representation was similar to that used for rats but relevant data on humans served to determine the critical parameters of the kinetic behavior. It was found that the metabolic rate of methyl mercury was on average 3 to 3.5 times slower in humans than in rats. Also, excretion rates of organic mercury from the whole body into feces and hair were 100 and 40 times smaller in humans, respectively, and urinary excretion of organic mercury in humans was found to be negligible. The human transfer rate of inorganic mercury from blood to hair was found to be 5 times lower than that of rats. On the other hand, retention of inorganic mercury in the kidney appeared more important in humans than in rats: the transfer rate of inorganic mercury from blood to kidney was 19 times higher than in rats and that from kidney to blood 19 times smaller. The excretion rate of inorganic mercury from the kidney to urine in humans was found to be twice that of rats. With these model parameters, simulations accurately predicted human kinetic data available in the published literature for different exposure scenarios. The model relates quantitatively mercury species in biological matrices (blood, hair, and urine) to the absorbed dose and tissue burden at any point in time. Thus, accessible measurements on these matrices allow inferences of past, present, and future burdens. This could prove to be a useful tool in assessing the health risks associated with various circumstances of methyl mercury exposure. © 2001 Academic Press

Key Words: **methyl mercury; inorganic mercury; toxicokinetics; modeling; humans.**

Methyl mercury is a well-known neurotoxic substance in humans (Bakir *et al.*, 1973; Al-Saleem *et al.*, 1976). Exposure occurs mainly through consumption of contaminated fish and shellfish (Birke *et al.*, 1972; Weatley and Paradis, 1995; Mahaffey, 1999). Outbreaks of methyl mercury poisoning in Japan (Tsubaki and Irukayama, 1977) and Iraq (Bakir *et al.*, 1973) have led researchers to document the health risks associated with methyl mercury exposures. However, the relationship between dose, biological markers of exposure, and target tissue concentrations of mercury forms at any point in time and for different exposure scenarios needs to be further investigated.

The objective of the present study was to adapt to humans a biologically based dynamic model of methyl mercury and inorganic metabolites disposition kinetics previously developed using experimental data in rats (see companion paper, Carrier *et al.*, 2001). The model was first constructed by establishing the biological determinants of methyl mercury disposition using a set of experimental data in rats. Differential equations that describe the temporal changes in tissue or compartment uptake and loss were first derived and solved by making extensive use of the different time scales involved in the biological processes. For humans, the model parameters were determined directly by stepwise fitting to the available data on blood, hair, and excreta time-course curves. Once validated in humans, the model can be used to provide new insights into the significance of using specific biological matrices for evaluating the extent of exposure to methyl mercury.

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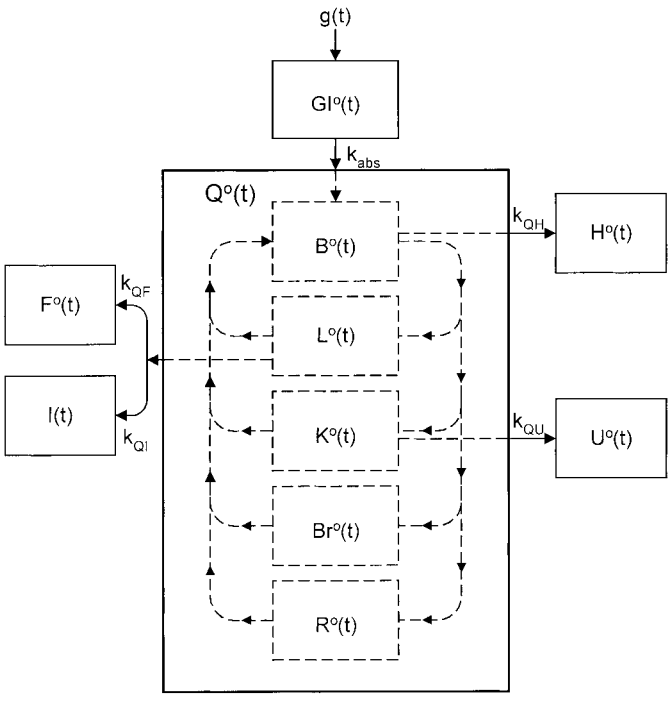

FIG. 1. Conceptual representation of organic mercury kinetics. Symbols and abbreviations are described in Table 1.

METHODS

Adaptation of the Model to Human Data

Conceptual and functional representation. The multicompartment dynamical model developed to simulate the disposition kinetics of organic and inorganic forms of mercury following exposure to methyl mercury in rats (Carrier *et al.*, 2001) was applied to human data, taking into account apparent differences in the toxicokinetics between rats and humans. For practical reasons, the model detailed in the companion paper (Carrier *et al.*, 2001) is again presented in Figs. 1 and 2. Symbols and abbreviations used in the functional representation of the model are described in Table 1. The sole difference in model representation between rats and humans is the inclusion for rats of some reabsorption of organic mercury through ingestion of hair during grooming as well as a transfer of inorganic mercury from ingested hair to feces, inorganic mercury not being easily absorbed from the gastrointestinal tract (Farris *et al.*, 1993). Otherwise, to adapt the model to human data, only the values of the intercompartment transfer rates and the tissue–blood partition coefficients needed to be modified.

Differential equations modeling the essential features of intercompartment processes are detailed in the first article of this series (Carrier *et al.*, 2001). In particular, the model is capable of relating mathematically the amounts of organic and inorganic mercury observed in hair segments to the doses absorbed and the burdens of diverse organs as they evolve with time. This is useful since for the biological monitoring of exposure to mercury in humans mercury concentrations in consecutive centimeter or half-centimeter segments of scalp hair (cut as close as possible to the scalp) are often measured.

Determination of parameters. It is important to note that all the free parameters (transfer rates) to be estimated have roles similar to those defined in the first article on rats (Carrier *et al.*, 2001) and are presented in Table 1. Contrary to the data of Farris *et al.* (1993) for rats, detailed tissue distribution and excretion kinetics of organic and inorganic forms of mercury over large periods of time were not available, to our knowledge, from the published literature. However, disposition kinetics of total mercury (the sum of organic and inorganic forms) over several weeks or months together with rough estimates of the fraction of organic and inorganic forms in human blood, hair, or excreta have been provided by several authors (Smith *et al.*, 1994; Aberg *et al.*, 1969; Miettinen *et al.*, 1971; Kershaw *et al.*, 1980). These data, when incorporated in the model, are sufficient to establish links between exposure dose, tissue burdens, and biological matrices. Furthermore, these data allowed parameters to be determined so that all the simulations are consistent with the experimental data of the various authors. Empirically, it was found that only a narrow range of values for each parameter allowed this consistency with all the data sets. The next sections describe the method used to determine the transfer parameters appropriate to human data.

Organic mercury kinetics. For the modeling of organic mercury kinetics, the free parameters were determined as follows. Absorption was considered very rapid (the time of digestion of the meal which is about 3 h) compared to the methyl mercury elimination half-life of several weeks. The absorption rate k_{abs} of 5.544 days^{-1} corresponds to an absorption half-life of 3 h. The absorption fraction (f_{abs}) was taken equal to unity since virtually 100% is absorbed according to Aberg *et al.* (1969) and Miettinen *et al.* (1971). According to Smith *et al.* (1994), it was estimated that on average 7.7% of an intravenous dose was deposited in the blood volume after a rapid tissue distribution. The proportionality constant K between whole body and blood burden of organic mercury [$K = Q°(t)/B°(t)$] was therefore estimated to be $K = 100/7.7 = 12.9870$.

No methyl mercury was detected in urine in the study of Smith *et al.* (1994), which is one of the few studies where organic and inorganic mercury forms in urine were distinguished; hence, the transfer coefficient k_{QU} was considered negligible. Again according to Smith *et al.* (1994), only a very small percentage of total mercury in feces was in the form of organic mercury. Feces contained on the average 3.5% of methyl mercury from 1 to 8 days following exposure, 2.3% from 11 to 30 days and 1.5% from 36 to 71 days. The transfer coefficient k_{QF} was therefore adjusted to obtain a visual best fit based on the available data on the kinetics of total mercury in feces (Aberg *et al.*, 1969; Miettinen *et al.*, 1971; Smith *et al.*, 1994) and the fraction of organic mercury in feces reported by Smith *et al.* (1994).

Smith *et al.* (1994) mentioned that blood contained predominantly methyl mercury; on average 98% of total mercury in blood was in the organic form from 0 to 56 h and 98.1% from 3 to 7 days. The metabolism rate constant k_{QI} was therefore adjusted to allow a good prediction of the kinetics of total mercury in human blood when compared to the data of Aberg *et al.* (1969),

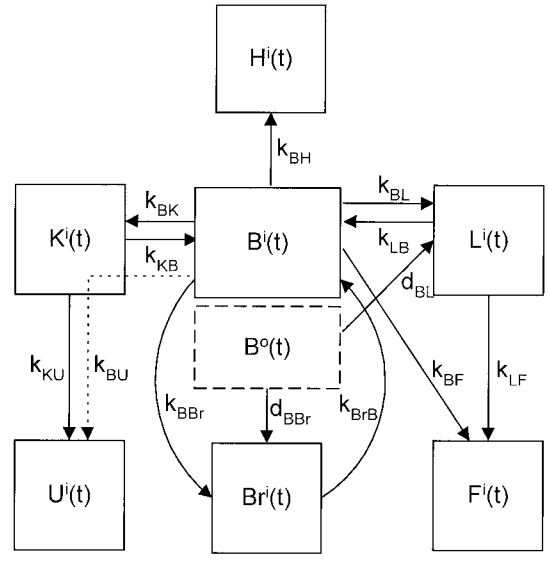

FIG. 2. Conceptual representation of inorganic mercury kinetics. Symbols and abbreviations are described in Table 1.

TABLE 1
Symbols and Abbreviations Used in the Functional Representation of the Model Adjusted to Human Data

Variables and parameters	Description
Organic mercury	
Variables	
$g(t)$	Oral dose which can describe time varying inputs
$GI^{\circ}(t)$	Burden of organic mercury in the gastrointestinal tract as a function of time
$Q^{\circ}(t)$	Whole body burden of organic mercury excluding hair and excreta as a function of time
$B^{\circ}(t)$	Burden of organic mercury in blood as a function of time
$L^{\circ}(t)$	Burden of organic mercury in liver as a function of time
$K^{\circ}(t)$	Burden of organic mercury in kidney as a function of time
$Br^{\circ}(t)$	Burden of organic mercury in brain as a function of time
$R^{\circ}(t)$	Burden of organic mercury in the rest of the body as a function of time
$H^{\circ}(t)$	Cumulative burden of organic mercury in hair as a function of time
$U^{\circ}(t)$	Cumulative burden of organic mercury in urine as a function of time
$F^{\circ}(t)$	Cumulative burden of organic mercury in feces as a function of time
$I(t)$	Whole body and excreta burden of inorganic mercury as a function of time
Constants	
K	Constant ratio $Q^{\circ}(t)/B^{\circ}(t)$
k_{abs}	Oral absorption rate constant
k_{QI}	Metabolism rate constant of organic mercury to inorganic mercury
k_{QF}	Whole body to feces transfer coefficient of organic mercury
k_{QU}	Whole body to urine transfer coefficient of organic mercury
k_{QH}	Whole body to hair transfer coefficient of organic mercury
k_{elim}	Whole body elimination rate constant of organic mercury
Inorganic mercury	
Variables	
$B^{i}(t)$	Burden of inorganic mercury in blood as a function of time
$L^{i}(t)$	Burden of inorganic mercury in liver as a function of time
$K^{i}(t)$	Burden of inorganic mercury in kidney as a function of time
$Br^{i}(t)$	Burden of inorganic mercury in brain as a function of time
$H^{i}(t)$	Cumulative burden of inorganic mercury in hair as a function of time
$U^{i}(t)$	Cumulative burden of inorganic mercury in urine as a function of time
$F^{i}(t)$	Cumulative burden of inorganic mercury in feces as a function of time
Constants	
d_{BL}	Blood to liver transfer coefficient combined with liver metabolism rate constant of organic mercury
d_{BBr}	Blood to brain transfer coefficient combined with brain metabolism rate constant of organic mercury
k_{LB}	Liver to blood transfer coefficient of inorganic mercury
k_{BK}	Blood to kidney transfer coefficient of inorganic mercury
k_{KB}	Kidney to blood transfer coefficient of inorganic mercury
k_{KU}	Kidney to urine transfer coefficient of inorganic mercury
k_{BH}	Blood to hair transfer coefficient of inorganic mercury
k_{BU}	Blood to urine transfer coefficient of inorganic mercury
k_{BF}	Blood to feces transfer coefficient of inorganic mercury
k_{LF}	Liver to feces transfer coefficient of inorganic mercury
k_{BBr}	Blood to brain transfer coefficient of inorganic mercury
k_{BrB}	Brain to blood transfer coefficient of inorganic mercury

Miettinen *et al.* (1971), Kershaw *et al.* (1980), and Smith *et al.* (1994) and of the fraction of organic mercury in blood reported by Smith *et al.* (1994).

The transfer constant of organic mercury from body burden to hair, k_{QH}, was adjusted to fit the data of Kershaw *et al.* (1980) on the time course of total mercury in hair multiplied by 0.80. This latter value corresponds to the fraction of organic mercury in hair that was found by several authors, whatever the exposure scenario (acute, subchronic, or chronic exposure). They reported that organic mercury is usually the predominant form of mercury in hair samples of individuals exposed mainly to the organic form of mercury. In fact, in most circumstances, organic mercury represents more than 70–80% of total mercury in scalp hair (Bakir *et al.*, 1973; Phelps *et al.*, 1980; Lee and Lee, 1999).

Whereas for rats, detailed tissue concentration–time profile data allowed determination of tissue-blood partition coefficients, this was not possible in humans for lack of similar data sets.

Inorganic mercury kinetics. For the modeling of inorganic mercury kinetics, the transfer parameters were determined as follows. The rate constant k_{BH} was adjusted to fit the data of Kershaw *et al.* (1980) on the time course of total mercury in hair multiplied by a fraction such that inorganic mercury in hair was less than 20% of total mercury as found by Bakir *et al.* (1973), Phelps *et al.* (1980), and Lee and Lee (1999).

Differences in mercury kinetics between rats and humans can also be attributed to variations in the renal handling of inorganic forms of mercury. In particular, there seems to be a higher retention of inorganic mercury in the human kidney due to binding to metallothionein compared to rats (Zalups *et*

al., 1993; Hellemans *et al.*, 1999; Yoshiba *et al.*, 1999). Renal constants k_{BK}, k_{KB}, and k_{KU} and fecal constants k_{LF} and k_{BF} were therefore adjusted to fit the data of Miettinen *et al.* (1971) on the daily urinary and fecal excretion of total mercury and on the time course of the whole-body fraction of total mercury in blood as well as the data of Aberg *et al.* (1969) on the time courses of total mercury cumulative urinary and fecal excretion.

A coherence test was carried out by verifying that the previously estimated values of the renal and fecal parameters provided a good visual fit to the time profile of total mercury in blood established by Kershaw *et al.* (1980). Data from Smith *et al.* (1994) on the cumulative urinary and fecal excretion of total mercury as well as on the blood–time profile were used to corroborate values estimated for the renal and fecal constants. It was also verified that the parameter values yielded the correct fraction of organic mercury in blood and feces described by Smith *et al.* (1994), hence that of inorganic mercury by difference.

Blood–brain exchange parameters for inorganic mercury and brain metabolism rate constant $\{k_{BBr}, k_{BrB}, d_{BBr}\}$ could not be determined specifically for humans for lack of time profile data. Since the amount of inorganic mercury in the brain is very small compared to the total inorganic mercury burden (in the rat at most 0.011%), precise knowledge of its value was not necessary to determine the mercury kinetics in other organs, blood, hair, and excreta. By default, the exchange parameters used were the same as those for rats (Carrier *et al.*, 2000). Also, varying the liver to blood transfer rate k_{LB} and the blood to urine secretion rate k_{BU} had no significant impact on the kinetics of inorganic mercury in humans and therefore, values were kept as determined using the detailed data on rats provided by Farris *et al.* (1993).

Model simulation. Mathematical resolution of the complete model, as represented by the system of differential equations (see the first article of the series, Carrier *et al.*, 2001), was carried out using the numerical Runge–Kutta method. A professional edition of Mathcad PLUS Software (MathSoft, Inc., Cambridge, MA) was used for this purpose and to provide model simulations. As mentioned in the article on rats, this model can predict the burdens of organic and inorganic mercury in tissues, blood, hair, or excreta at any point in time after a variety of exposure scenarios to methyl mercury: single, intermittent, or continuous.

Model Validation

Once the parameters were determined using the previously mentioned data, the model was validated using the data of Sherlock *et al.* (1984) and Birke *et al.* (1972).

RESULTS

Model Parameters Adjusted to Human Data

Model parameters adjusted to the available human data are provided in Table 2 (see Table 1 for description of symbols and abbreviations). These values can be compared to those determined using data on rats (see companion paper, Carrier *et al.*, 2001). As observed in rats, absorption of methyl mercury was very rapid (2–3 h) compared to its whole-body elimination rate k_{elim}, which represents the sum of excretion rates of the organic form from the body together with the metabolism rate to inorganic mercury ($k_{elim} = k_{QF} + k_{QU} + k_{QH} + k_{QI}$). Organic mercury in feces, urine, and hair was found in humans to be in much smaller amounts than in rats. The human transfer rate constant k_{QF} of organic mercury from the whole-body burden to feces was 100 times lower than in rats. The transfer rate constant k_{QU} from the whole-body burden to urine was found to be negligible. The value of the human transfer coefficient k_{QH} from the whole-body burden to hair was 40 times less than that obtained for rats.

TABLE 2
Numerical Values of Constant Parameters Used in the Model Adjusted to Human Data

Constant parameters	Values (days^{-1})[a]
Organic mercury	
K	12.9870
k_{abs}	5.5440
k_{QI}	0.01347[b]
k_{QF}	9.0668×10^{-5}
k_{QU}	≈ 0
k_{QH}	2.3825×10^{-4}
k_{elim}	0.01380
Inorganic mercury	
d_{BL}	0.1750
d_{BBr}[c]	$\ll d_{BL}$
k_{LB}[d]	0.8940
k_{BK}	17.1234
k_{KB}	0.0010
k_{KU}	0.006949
k_{BH}	0.1400
k_{BU}[d]	0.06994
k_{BF}[d]	3.9917
k_{LF}[d]	1.5476
k_{BBr}[d]	0.0028
k_{BrB}[d]	0.0520

[a] Except K which is a ratio and not a rate as are the other parameters.
[b] Average value.
[c] The value of d_{BBr} was considered very small compared to that of d_{BL}.
[d] The value was kept as in rats.

The human metabolism rate constant of organic mercury into inorganic mercury k_{QI} was on average 3 to 3.5 times lower than that of rats. This parameter is likely subject to interindividual variations. To best fit the data of various authors, the k_{QI} value was adjusted to 0.01437, 0.01347, 0.01232, 0.01306, and 0.005672 days^{-1} to obtain a whole-body elimination half-life of organic mercury ($0.693/k_{elim}$) corresponding to 47.1 days to fit data of Aberg *et al.* (1969), 50.2 days for those of both Smith *et al.* (1994) and Miettinen *et al.* (1971), 54.8 days for those of Kershaw *et al.* (1980), 51.7 days for those of Sherlock *et al.* (1984), and 115.5 days for those of Birke *et al.* (1972), respectively.

The human transfer rate coefficient k_{BH} of inorganic mercury from blood to hair was found to be 5 times lower than in rats. On the other hand, kidney retention was much more important in humans than in rats. This brought about a human transfer rate constant k_{BK} of inorganic mercury from blood to kidney 19 times higher than in rats and a human transfer coefficient k_{KB} of inorganic mercury from kidney to blood 19 times smaller. Furthermore, the transfer rate k_{KU} of inorganic mercury from human kidney to urine was twice that of rats. As mentioned previously, the other parameters for inorganic mercury kinetics did not appear to be significant determinants of the disposition

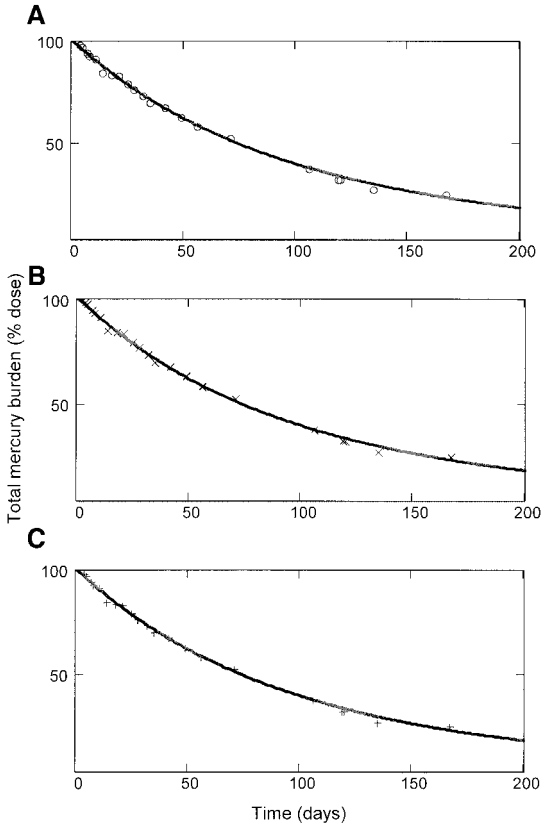

FIG. 3. Comparison of model simulations (lines) with experimental data (symbols which represent values from a single individual) of Aberg *et al.* (1969) on the time course of total mercury body burden over close to 200 days in three volunteers (A, B, C) exposed orally to 11 μg of methyl mercuric nitrate.

of inorganic mercury in humans and were thus left as determined in rats.

Simulation of the Time Course of Mercury Disposition after Acute Exposure in Humans

Figure 3 shows that the model simulates the data obtained by Aberg *et al.* (1969) on the time course of total mercury body burden in three male volunteers exposed orally to a single dose of 11 μg of methyl mercuric nitrate. It is interesting to note that whole-body elimination of total mercury appears log-linear over more than 100 days (see Fig. 4). However, when simulating the profile over a larger time span, total mercury elimination from the body is shown to be multiphasic. Indeed, during the first 100 days postexposure, both organic and inorganic forms contribute to total mercury body burden in such a way as to indicate a quasi log–linear feature. However, after 300 days, mostly inorganic mercury remains in the body and its elimination does not follow a log–linear pattern. With time, the kinetics of total mercury body burden approaches that of inorganic mercury whose elimination is multiexponential.

Figure 5 compares model simulations to the experimentally

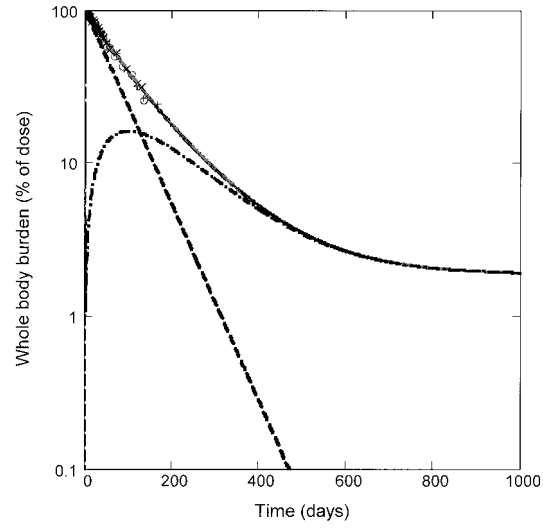

FIG. 4. Comparison of model simulations of the whole-body burden time profiles of total (—), organic (- - -), and inorganic (− −●− −) mercury over 1000 days with the corresponding time courses of total mercury experimentally determined by Aberg *et al.* (1969) in three volunteers (symbols ○, ×, and +) following an acute oral exposure to 11 μg of methyl mercuric nitrate.

observed time course of total mercury cumulative excretion in urine and feces as determined by Aberg *et al.* (1969). Predictions were in the same value range as those observed experimentally, although the model slightly underestimated the urinary and fecal excretion. Figure 5 also shows that feces contains mainly (>98%) inorganic mercury (total mercury and

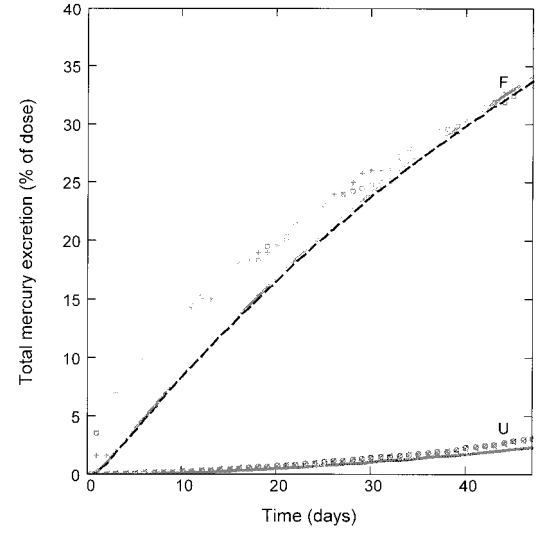

FIG. 5. Comparison of model simulations of the time-dependent cumulative urinary (U) and fecal (F) excretion profiles of total (—) and inorganic (- - -) mercury (total and inorganic mercury curves overlap), over 50 days approximately, with the corresponding time courses of total mercury experimentally determined by Aberg *et al.* (1969) in two volunteers (symbols × and ○ for urine; + and □ for feces) exposed orally to 11 μg of methyl mercuric nitrate.

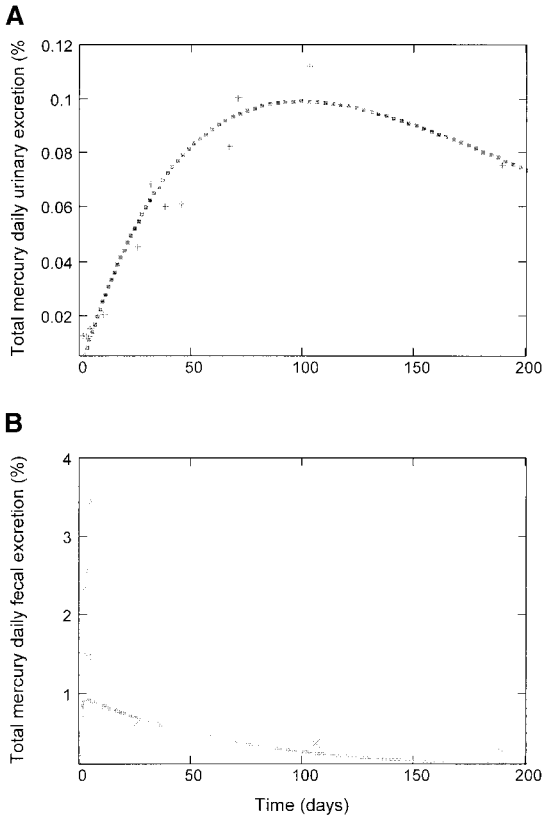

FIG. 6. Comparison of model simulations (lines) with experimental data (symbols which represent mean values) of Miettinen *et al.* (1971) on the time courses of daily urinary (A) and fecal (B) excretion of total mercury over approximately 200 days in volunteers following an acute oral exposure to 22 μg of methyl mercuric nitrate through fish consumption.

inorganic mercury curves almost overlap). In urine, only the inorganic form of mercury is found.

The model also predicts the time course of daily urinary and fecal excretion of total mercury in volunteers exposed orally to an acute dose of 22 μg of methyl mercury nitrate through fish consumption when compared to the data of Miettinen *et al.* (1971) (see Fig. 6). Only in the first few days postexposure are observed fecal excretion values of total mercury higher than predicted values.

Figure 7 shows that the model provides a close approximation to the concentration–time profile of total mercury in blood and hair as determined by Kershaw *et al.* (1980) in male volunteers exposed orally to a single dose of 20 μg of methyl mercury per kilogram of body weight through fish consumption. It is also apparent that blood contained essentially the organic form of mercury (organic and total mercury kinetics overlap) over 200 days postexposure and that organic mercury accounted for more than 80% of total mercury in hair. The model, naturally enough, also provided a good fit to the data of Smith *et al.* (1994), which were mainly used for the estimation of model parameters (Fig. 8).

Validation of the Model

Comparison of model simulations to the available data in volunteers subchronically or chronically exposed to methyl mercury shows that the model applies equally well to multiple exposure scenarios. Indeed, simulations were in close agreement with the data of Sherlock *et al.* (1984) on the blood concentration–time profile of total mercury in volunteers exposed to either 42, 77, 101, or 226 μg per day of methyl mercury through fish consumption over 3 months as presented in Figs. 9A–9D.

The model also simulates the data of Birke *et al.* (1972) on the elimination kinetics of total mercury concentrations in red blood cells and hair after a chronic exposure to methyl mercury through fish consumption (see Fig. 10). According to the model predictions, because of regular intake, organic mercury represents a larger fraction of total mercury concentrations in blood and hair over the respective 770 and 945 days experimental

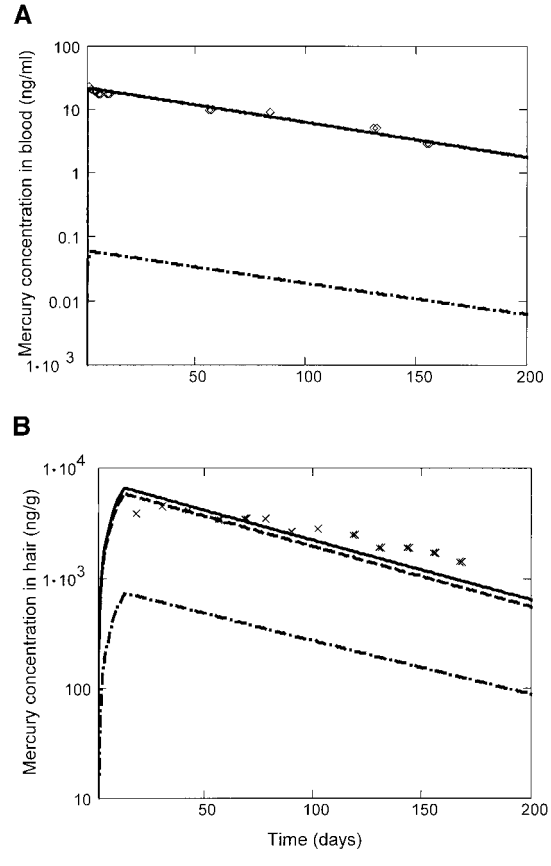

FIG. 7. Comparison of model simulations of the concentration–time profiles of total (—), organic (- - -), and inorganic (− −●− −) mercury in blood (A) (total and organic mercury blood curves practically overlap) and hair (B), with the corresponding time courses of total mercury experimentally determined by Kershaw *et al.* (1980) in blood, over approximately 165 days, and in 13 consecutive 0.5 cm hair strands of volunteers (symbols which represent mean values) exposed orally to an acute dose of ≈20 μg of methyl mercury per kg of body weight through fish consumption.

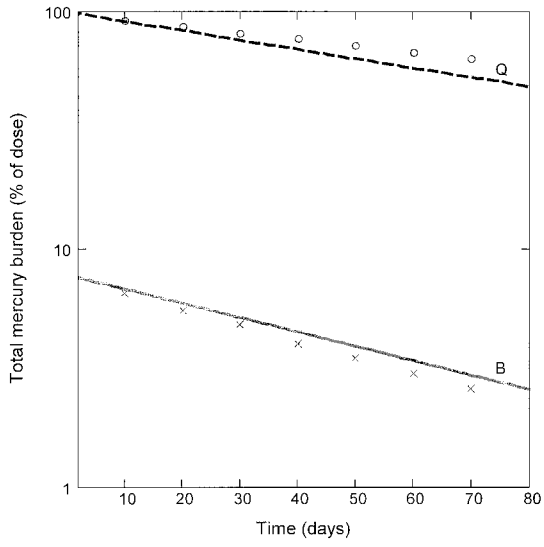

FIG. 8. Comparison of model simulations (lines) with experimental data (symbols which represent mean values) of Smith *et al.* (1994) on the whole-body (Q) and blood (B) burden–time profiles of total mercury over 70 days in volunteers exposed intravenously to approximately 3.85 μg of methyl mercury.

sampling period, compared to the situation after a single exposure.

The model allows as well the prediction of the time-dependent disposition of organic and inorganic mercury (expressed as a fraction of absorbed daily unit dose) during a chronic continuous oral exposure to methyl mercury. These simulations show that, after 1 to 2 years of exposure, near steady state levels are reached for organic and inorganic mercury burdens in blood, as well as for inorganic mercury levels in liver and brain. The values predicted are, respectively, 5.6 and 0.02 times the daily unit dose for organic and inorganic mercury in blood, and 0.4 and 10^{-3} times the daily unit dose for inorganic mercury in liver and brain (simulations not shown). On the other hand, equilibrium between tissue uptake and elimination is reached only after 5 years for inorganic mercury in kidney, with a steady state value of 40 times the absorbed daily unit dose of methyl mercury (simulations not shown).

Unfortunately, the data available in humans do not allow the validation of the kinetics of organic mercury in liver, kidney, and brain. Nonetheless, the model predictions provide an approximation of liver and kidney asymptotic values which are indirectly adjusted to ensure congruence with the blood concentration–time profile and the urinary and fecal excretion time courses. As for the predicted brain asymptotic values, they are tentative and can be viewed as giving an order of magnitude for this ratio since they are not directly or indirectly determined from human data, but are rather merely an extrapolation from a combination of human and rat constants.

Following a chronic continuous exposure to methyl mercury, it is further interesting to note that the model predicts that the ratios of daily excretion rate of organic to inorganic mercury in hair and in feces become constant after 1 or 2 years of continuous exposure with values of about 6.6 for hair (simulations not shown) and 1/105 for feces (simulations not shown). The ratio of fecal-to-urine daily excretion rate of inorganic mercury becomes constant after 5 years with a value of 2.5 (simulations not shown).

Chronic exposure scenario simulations also provide the tissue–blood concentration partition coefficients of organic and inorganic mercury at steady state, as listed in Table 3. These values clearly indicate an important bioaccumulation of both organic and inorganic mercury in hair as well as inorganic mercury in kidney.

DISCUSSION

A biologically based dynamical model of the uptake and disposition of methyl mercury in animals and humans has been developed. This model is a refinement of conventional data-based models which allows animal-to-human, route-to-route comparisons for various exposure scenarios. The main requirement for the development of such a model is the availability of extensive amounts of *in vivo* experimental data in animals and humans. An important feature of the current model lies with its few parameters compared to physiologically based pharmacokinetic (PBPK) models.

Differences in Mercury Kinetics between Rats and Humans

For animal-to-human extrapolation of mercury kinetics, critical biological determinants of species differences were determined. The most obvious difference is the blood concentration–time profile of organic mercury which, after a single dose, exhibits a monoexponential decrease in humans (Smith *et al.*, 1994), whereas elimination is biexponential in rats since there is a feedback loop resulting from the ingestion of hair during grooming (Farris *et al.*, 1993). In adult monkeys and cats, postdistributive elimination kinetics of organic mercury in blood appears similar to that of humans over 100 to 150 days following a single oral methyl mercury exposure (Hollins *et al.*, 1975; Evans *et al.*, 1977; Rice, 1989).

It is also noteworthy that after a single dose, the internal distribution quickly settles to a percentage of organic mercury body burden in blood that is different in rats and humans: 7.7% in humans and 30% in rats. This is built in the model to agree with the observed data of Smith *et al.* (1994) in humans and Farris *et al.* (1993) in rats. Conversely, the model simulations predict a maximum total inorganic mercury burden of the body is 16.10% in humans and 5.60% in rats of the administered methyl mercury dose. This might account for a higher toxic potential from methyl mercury exposure in humans (per kilogram of body weight) since inorganic metabolites are thought to be responsible for much of the neurotoxic effects induced by methyl mercury (Friberg and Mottet, 1989; Charleston *et al.*, 1994; Vahter *et al.*, 1994). However, for humans, the high load

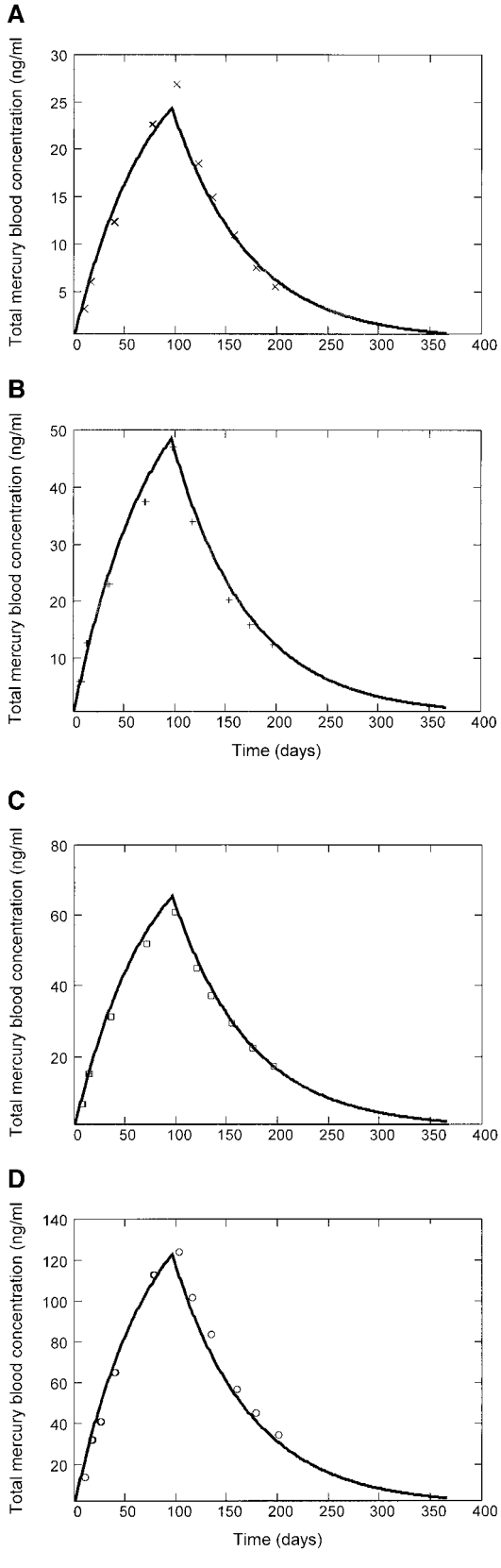

FIG. 9. Comparison of model simulations (lines) with experimental data (symbols which represent mean values) of Sherlock *et al.* (1984) on the blood concentration–time profile of total mercury in four groups of volunteers during and following approximately 96 days of exposure to 42 (A), 77 (B), 101 (C), or 226 (D) μg per day of methyl mercury through fish consumption.

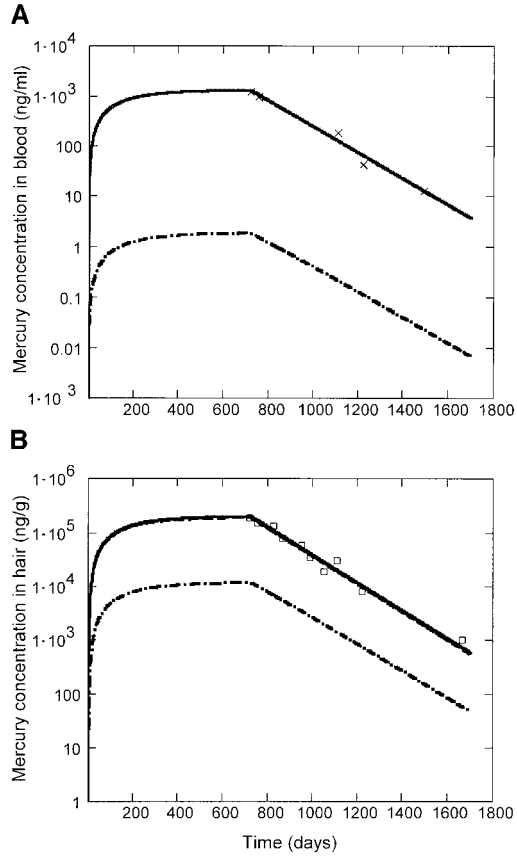

FIG. 10. Comparison of model simulations of the concentration–time profiles of total (—), organic (- - -) (total and organic mercury curves practically overlap), and inorganic (– –●– –) mercury in red blood cells (A) and hair (B) with the corresponding time courses of total mercury experimentally determined by Birke *et al.* (1972) over approximately 1000 days following several years of exposure to 800 μg of methyl mercury per day through fish consumption in a volunteer (symbols).

of inorganic mercury in the kidney ($K^i(t)$), compared to blood ($B^i(t)$), might leave less inorganic mercury circulating in blood and available for transfer to the brain.

In addition, when comparing model simulations of methyl mercury kinetics between animals and humans, as well as literature data, it is evident that elimination kinetics is slower in humans. These findings can be explained in part by the usually slower metabolic rate in humans compared to rats. In the current study, a single set of values for model parameters has been found to apply to all human subjects studied except for the noteworthy metabolism rate constant. This parameter can vary substantially from one individual to another as a result of differences in the rate of demethylation and conjugation (Al-Shahristani and Shihab, 1974; Al-Shahristani *et al.*, 1976).

TABLE 3

Estimated Human Tissue–Blood Concentration Partition Coefficients for Organic, Inorganic and Total Mercury at Near Equilibrium Determined After Simulation of a Chronic Continuous Exposure Over 70 Years

	Concentration partition coefficients[a]		
Tissues	Organic mercury	Inorganic mercury	Total mercury
Hair	291	13,164	333
Kidney	—	38,761	—
Liver	—	64	—

[a] Tissue and blood concentrations were estimated by dividing predicted tissue or blood burdens at steady state by weights or volumes reported by Sumino *et al.* (1975).

Variability in the biological half-life of methyl mercury in humans is quite substantial as shown by Al-Shahristani *et al.* (1976) ranging from 35 to 120 days. According to model simulations of kinetic profiles described in various studies, most half-life values required by the model to fit the data ranged between 45 and 55 days except for the data of Birke *et al.* (1972), where elimination half-life in one volunteer was 120 days.

Another important aspect of species differences in elimination kinetics is the greater tissue blood flow rate in smaller species than in larger species (Boxenbaum, 1980), resulting in chemicals being more rapidly carried to organs of clearance in smaller mammals. Indeed, in the current study, to adequately simulate data in humans starting from the model for rats, transfer rates of organic mercury to excretory compartments, namely feces, urine, and hair, had to be reduced. However, the smaller fecal excretion of organic mercury in humans could also result from species differences in the biliary excretion of organic mercury, which is partly eliminated in feces through this pathway after conjugation to glutathione and its derivatives (Ballatori and Clarkson, 1983). Small rodents such as rats and mice excrete chemical substances to a greater extent into bile than larger species (Klaassen and Watkins, 1984). This phenomenon is explained by the higher molecular weight threshold for biliary excretion in humans (475 ± 50) compared to rats (325 ± 50) (Smith, 1973).

Similarly, inorganic mercury is partly eliminated in feces through biliary excretion, although transfer from blood to intestinal lumen (i.e., intestinal secretion) also appears to be a significant excretion route according to animal studies (Zalups, 1998; Zalups *et al.*, 1999). The previously mentioned mechanism would help to understand the relatively higher inorganic mercury excretion in rat feces compared to humans. The alternate route of elimination through urine would thus be proportionally more important in humans. This is observed as a higher urinary excretion rate of conjugated inorganic mercury in humans compared to rats. Furthermore, according to the model simulations and available literature data, there seems to be differences in the renal handling of inorganic mercury between rats and humans; in particular, there is evidence for an increased retention of inorganic mercury in the human kidney due to binding to metallothionein compared to rats (Zalups *et al.*, 1993; Hellemans *et al.*, 1999; Yoshiba *et al.*, 1999).

On the other hand, in both rats and humans, blood contains predominantly the organic form of mercury which reversibly associates with proteins or thiol-containing compounds because of its high affinity for sulfhydryl groups (Cember *et al.*, 1968; Sundberg *et al.*, 1999). Nonetheless, red blood cells (RBCs) to plasma concentration ratio of methyl mercury varies between rodents and humans (U.S. EPA, 1997) and could result in differences in tissue distribution. The RBCs to plasma concentration ratio of methyl mercury has been reported to be about 9–10:1 in humans and 100–200:1 in rats following methyl mercury exposure (Suzuki *et al.*, 1971; Magos, 1987; U.S. EPA, 1997). There are also differences in the affinity of organic and inorganic mercury for blood proteins (Suzuki *et al.*, 1971; Hall *et al.*, 1994). Contrary to organic mercury, inorganic mercury is equally distributed between erythrocytes and plasma in human blood (ratio 1:1) following inorganic mercury exposure (Hall *et al.*, 1994). Obviously, amounts of body lipids can also affect tissue distribution, methyl mercury being lipophilic.

Finally, for humans and rats, the model considers that methyl mercury either crosses the blood–brain barrier and is demethylated in brain tissues as suggested by some authors (Lind *et al.*, 1988; Friberg and Mottet, 1989), or alternatively it is the inorganic mercury in blood that directly enters the brain to induce its toxic effects (Berlin *et al.*, 1975; Berlin, 1986) (see article on rats for more details, Carrier *et al.*, 2001). Clearly, additional work on this issue is needed to elucidate the mechanism of brain toxicity induced by mercury compounds.

Model Predictions and Human Data

Taking into consideration all the critical determinants of animal-to-human differences in the kinetics of mercury, the model predicted adequately the available literature data. The model did however slightly underestimate the initial fecal excretion of total mercury measured by Aberg *et al.* (1969) and Miettinen *et al.* (1971). This is likely due to the fact that virtually all of the orally administered methyl mercury dose was assumed here to be absorbed as reported by Aberg *et al.* (1969) and Falk *et al.* (1970). If rather a 97% absorption ratio of ingested methyl mercury is applied, a perfect fit is obtained. These figures were not represented in the current article because model parameters were determined assuming a 100% absorption ratio. Another explanation provided by Smith and Farris (1996) is that the high initial levels of fecal mercury result from the presence of inorganic mercury in the dosing materials for both the studies of Aberg *et al.* (1969) and Miettinen *et al.* (1971).

The model also provided a good fit to the data of Miettinen

et al. (1971) on the daily urinary excretion of total mercury and those of Smith *et al.* (1994) on the cumulative urinary excretion of total mercury, although it slightly underestimated the cumulative urinary excretion of total mercury observed by Aberg *et al.* (1969). Differences between model predictions and the experimental data of the latter authors possibly stem from the substantial recorded variations in the daily urinary excretion of total mercury. This causes an increased uncertainty when expressing values as a cumulative percentage of dose. Increased renal accumulation of inorganic mercury could also result in an enhanced urinary excretion of the metabolite at the expense of fecal excretion.

Importance of Time of Sampling and Metal Speciation

Model simulations of the time course of organic and inorganic mercury in the whole body as well as in blood and hair further illustrate the importance of the time of sampling relative to the time of exposure for the biological monitoring of exposure to methyl mercury. For example, since the urinary excretion levels of inorganic mercury, the main form of mercury in urine (Smith *et al.*, 1994), is strongly influenced by the history of past exposure, it is important to determine judiciously the best sampling strategy. Indeed, data analysis (Aberg *et al.*, 1969) and model predictions show that, although inorganic mercury excretion in urine is a minor route of methyl mercury elimination, it increases with time after a chronic exposure. This is due to the long retention of inorganic mercury in kidney. It is corroborated by data of Cappon and Smith (1981) on deceased Canadian Indians who chronically consumed fish with high levels of methyl mercury ($>$0.5 ppm) where the mercury accumulated in the kidney was mostly in the inorganic form, that is, approximately 94%.

Furthermore, model simulations emphasize as well the need for metal speciation to better predict toxic outcomes. In particular, blood and hair are often used as biological matrices for exposure estimates. According to model simulations, the organic form of mercury in blood and hair is the predominant mercury species (more than 90 and 80%, respectively) during a constant exposure to methyl mercury and during the first two years following cessation of exposure. A significant increase in the fraction of inorganic mercury in these biological matrices, compared to the above, during the previously mentioned time period following cessation of exposure would suggest a concomitant exposure to metallic mercury by dental amalgams or industrial emissions.

Steady State Levels and Tissue–Blood Partition Coefficients

The proposed model also estimates the time needed to reach effective steady state levels in blood and tissues (measured as a fraction of daily dose) given a chronic continuous exposure to methyl mercury and thus provides tissue–blood concentration partition coefficients at near equilibrium. The model clearly supports the hypothesis that kidney accumulates inorganic mercury as proposed by some authors (Farris *et al.*, 1993; Sallsten *et al.*, 1994; Smith *et al.*, 1994). Indeed, steady state levels of inorganic mercury in the kidney are reached only after five years of continuous exposure, whereas those of blood, liver, and brain are reached after one or two years. At that time, kidney–blood concentration partition coefficient of inorganic mercury burden is about 600 times the liver–blood concentration partition coefficient.

The model also predicts a hair-to-blood concentration ratio of total mercury in humans of 333 on average at near equilibrium (i.e., following a year or more of constant exposure). This compares well with the mean value of 292 reported by Kershaw *et al.* (1980) and the 200:1–300:1 range of ratios reviewed by Katz and Katz (1992).

Overall, the current model succeeded in integrating various experimental data to uncover critical determinants of methyl mercury kinetics in animals as well as in humans. The model predicts the time courses of various tissue burdens for different dose regimens and exposure scenarios. It also generates hypothesis as to the experimental uncertainties that should be addressed.

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