A Toxicokinetic Model of Malathion and Its Metabolites as a Tool to Assess Human Exposure and Risk through Measurements of Urinary Biomarkers

Michèle Bouchard,* Nathalie H. Gosselin,* Robert C. Brunet,† Onil Samuel,‡ Marie-Josée Dumoulin,* and Gaétan Carrier*,†

*Département de Santé Environnementale et Santé au Travail, Faculté de Médecine, Université de Montréal, C. P. 6128, Succursale Centre-ville, Montréal, Québec, Canada H3C 3J7; †Département de Mathématiques et de Statistique and Centre de Recherches Mathématiques, Faculté des Arts et des Sciences, Université de Montréal, C. P. 6128, Succursale Centre-ville, Montréal, Québec, Canada, H3C 3J7; and ‡Institut National de Santé Publique du Québec, Direction de la Toxicologie Humaine, Centre de Toxicologie, 945 Avenue Wolfe, Ste-Foy, Québec, Canada G1V 5B3

Received December 19, 2002; accepted February 10, 2003

A toxicokinetic model is proposed to predict the time evolution of malathion and its metabolites, mono- and dicarboxylic acids (MCA, DCA) and phosphoric derivatives (dimethyl dithiophosphate [DMDTP], dimethyl thiophosphate [DMTP], and dimethyl phosphate [DMP]) in the human body and excreta, under a variety of exposure routes and scenarios. The biological determinants of the kinetics were established from published data on the in vivo time profiles of malathion and its metabolites in the blood and urine of human volunteers exposed by intravenous, oral, or dermal routes. In the model, body and excreta compartments were used to represent the time varying amounts of each of the following: malathion, MCA, DCA, DMDTP, DMTP, and DMP. The dynamic of intercompartment exchanges was described mathematically by a differential equation system that ensured conservation of mass at all times. The model parameters were determined by statistically adjusting the explicit solution of the differential equations to the experimental human data. Simulations provide a close approximation to kinetic data available in the published literature. When simulating a dermal exposure to malathion, the main route of entry for workers, the model predicts that it takes an average of 11.8 h to recover half of the absorbed dose of malathion eventually excreted in urine as metabolites, compared to 3.2 h following an intravenous injection and 4.0 h after oral administration. This shows that following a dermal exposure, the absorption rate governs the urinary excretion rate of malathion metabolites because the dermal absorption rate is much slower than biotransformation and renal clearance processes. The model served to establish biological reference values for malathion metabolites in urine since it allows links to be made between the absorbed dose of malathion and the time course of cumulative amounts of metabolites excreted in urine. From the no-observed-effect level (NOEL) of 0.61 \( \mu \)mol/kg/day derived from the data of Moeller and Rider (1962), the model predicts corresponding biological reference values for MCA, DCA, and phosphoric derivatives of 44, 13, and 62 nmol/kg, respectively, in 24-h urine samples. The latter were used to assess the health risk of workers exposed to malathion in botanical greenhouses, starting from urinary measurements of MCA and DCA metabolites.

Key Words: malathion; monocarboxylic acids; dicarboxylic acids; phosphoric derivatives; toxicokinetics; risk assessment.

Malathion (O,O-dimethyl S-1,2-di(ethoxycarbonyl)ethyl phosphorodithioate) is an organophosphate (OP) insecticide widely used in agriculture and residential settings as well as in public health programs for mosquito-borne disease control (Environnement Québec, 2002; U.S. EPA, 2000). It is also used in some countries for the treatment of head lice (Roberts, 2002). Like other OP insecticides, malathion exerts its neurotoxic action in humans, as in insects, through cholinesterase (ChE) inhibition. This results in the accumulation of acetylcholine within synapses leading to over-stimulation of postsynaptic receptors (Liu and Pope, 1998). In acutely exposed individuals, clinical signs of OP intoxication usually appear at inhibition of 60–70% of acetylcholinesterase (AChE) activity in red blood cells (RBC). However, light clinical signs and symptoms were reported in subjects with 30–60% reduction in RBC–AChE activity (Sidell, 1994).

The American Conference of Governmental Industrial Hygienists proposed a biological exposure index (BEI) for AChE inhibiting pesticides to prevent cholinergic health effects (ACGIH, 2002). An RBC–AChE activity of 70% of the individual’s baseline (i.e., 30% inhibition of the activity) is proposed as a biological reference. However, a BEI based on the measurement of urinary biomarkers of malathion has not been proposed thus far, although this is desirable since it would give an earlier warning of possible effects than AChE inhibition. Indeed, it is clearly established that urinary biomarkers of exposure to malathion are more sensitive exposure indices, since they can be measured at exposure doses lower than those necessary to induce a measurable inhibition of AChE activity.

Experimental studies in animals and humans show that mal-
Malathion is oxidized by cytochrome P-450, in small amounts (4–6% in rats), to malaoxon, which is responsible for ChE inhibition (U.S. EPA, 2000). Both malathion and malaoxon are rapidly detoxified by carboxylesterases to mono-acid and di-acid derivatives, which are excreted mainly in urine; these acids can further be metabolically converted to phosphoric derivatives, also primarily excreted in urine (Ecobichon, 1992; Feldmann and Maibach, 1974; Jellinek et al., 2000; U.S. EPA, 2000).

To assess exposure to malathion in field studies, malathion mono- and dicarboxylic acids (MCA and DCA) in urine are thus used as specific biomarkers (Adgate et al., 2001; MacIntosh et al., 1999; Márquez et al., 2001) while the phosphoric derivatives dimethyl dithiophosphate (DMDTP), dimethyl thiophosphate (DMTP) and dimethyl phosphate (DMP) serve as nonspecific urinary biomarkers (Cocker et al., 2002; Coye et al., 1986; Fenske, 1988; Fenske and Leffingwell, 1989).

The use of these urinary biomarkers to assess risk is dependent on the establishment of a link between their measurements and critical biological effects. Various authors have established a relationship between the exposure dose and urinary biomarkers in controlled human studies (Feldmann and Maibach, 1974; Jellinek et al., 2000), while others have studied links between the exposure dose and the inhibition of AChE activity in volunteers (Moeller and Rider, 1962). In some reports, attempts were also made to link urinary biomarkers to the appearance of early biological effects under experimental conditions, but the administered doses were not sufficient to induce an inhibition of AChE activity (Dennis and Lee, 1999; Jellinek et al., 2000). In field studies with exposed workers, these relationships cannot be established directly on the basis of measurements of external doses (ambient air concentrations or skin deposits). In the context of malathion exposure in occupational settings, which occurs mainly through dermal contact (ACGIH, 2002; Tuomainen et al., 2002), knowledge of the kinetics is essential since dermal absorption is subject to large variations among individuals, and depending on the exposed skin site (Bronaugh and Maibach, 1999; Cohen and Rice, 2001; Feldmann and Maibach, 1974).

If links can be established between the cumulative amounts of urinary biomarkers and the absorbed dose of malathion, whatever the exposure route (oral, dermal, pulmonary) or scenario (single, repeated, intermittent or continuous exposure), and between the absorbed dose and the appearance of biological effects, it then becomes possible to use urinary biomarkers to assess risk in workers exposed to malathion. These links can be made through toxicokinetic modeling, provided appropriate experimental data on the metabolism and disposition of malathion and its metabolites are available. A physiologically based pharmacokinetic model has been developed to describe the human dermal absorption, metabolism, and excretion of malathion (Rabovsky and Brown, 1993). This model did not, however, seek to describe the urinary excretion kinetics of specific metabolites, and thus does not allow reconstruction of the absorbed dose starting from urinary measurements.

The objectives of this study were (i) to develop a toxicokinetic model for humans, linking the dose of malathion absorbed under different exposure routes and scenarios to the time courses of malathion metabolites in urine and (ii) to use this model as a framework for relating published human no-observed-effect level(s) (NOEL) to associated amounts of metabolites in urine, which are to be proposed as convenient biological reference values to prevent health effects.

**MATERIALS AND METHODS**

**Model development: Conceptual and functional representation.** The disposition kinetics of malathion and its metabolites, following intravenous, oral, or dermal exposure, were modeled using a multi-compartment dynamical system, described mathematically by a mass balance differential equation system (see Appendix). The model conceptual and functional representation was designed to describe the data provided by Feldmann and Maibach (1974) and Jellinek et al. (2000) on the kinetics of malathion and its metabolites in human volunteers.

The model conceptual representation is depicted in Figure 1. Symbols and abbreviations used in this study are described in Table 1. The model uses a specific body compartment for the malathion burden in blood and tissues in dynamical equilibrium with blood, i.e., tissues that rapidly reach and maintain a fixed ratio with blood (referred to later as the blood compartment B(t) for simplicity). Another compartment regroups the malathion stored in tissues S(t), i.e., malathion in lipids or bound to tissue proteins. A compartment, Mt(t), is also used to describe the whole-body burden of total metabolites. In addition, different compartments are introduced for cumulative amounts of each specific malathion metabolite in either urine or feces, that is for malathion monocarboxylic acids (MCA), dicarboxylic acids (DCA), and for the phosphoric derivatives: dimethyl dithiophosphate (DMDTP), dimethyl thiophosphate (DMTP), and dimethyl phosphate (DMP). The route-of-entry, gastro-intestinal tract or skin, is also represented as a separate input compartment. In the model, all amounts are initially expressed on a mole basis.

The compartment Mt(t) regroups unconjugated MCA, DCA, DMDTP, DMTP, and DMP metabolites, which are generated in cascade, together with their conjugates, in accordance with the metabolism described by Ecobichon (1992). Since phase I and II biotransformation processes occur on a much more rapid time scale than renal and fecal clearance of the metabolites, the fractions of total body metabolites excreted from the body as specific metabolites are mainly a function of the competition between phase II conjugation and further phase I metabolism. For instance, the amounts of MCA eliminated from the body are dependent on the competition between conjugation and biotransformation to DCA, DMDTP, and DMTP. Consequently, if 36% of an absorbed dose is recovered in excreta as MCA and 4% of malathion is metabolized to malaoxon, it indicates that 36% of the absorbed dose has been metabolized to MCA and conjugated while 60% has been biotransformed to other metabolites. Without competition between conjugation and the biotransformation cascade, unconjugated metabolites would all be metabolized to DMP.

In the model, the rates of change in the amounts of a substance in a given compartment are described mathematically as the difference between compartment rates of uptake and loss. Exchange rates between compartments represent either the physical transfer of the same substance or the transfer (on a mole to mole basis) through biotransformation of malathion to its metabolites or one metabolite to another. Solving the system of differential equations yields the mathematical function of the time courses of malathion and its metabolites in the different compartments.

**Model development: Determination of parameters.** The model parameters were determined from the in vivo time profiles provided by Feldmann and Maibach (1974) and Jellinek et al. (2000). The parameters were determined by
statistical best-fits of the explicit solutions of differential equations to experimental human time-course data, or by log-linear regression on the time-profile data (using in both cases reported average values). A professional edition of MathCad software was used for this purpose (MathSoft, Inc., Cambridge, MA).

Extensive use was made of the different time scales involved in the biological processes to simplify differential equations, using quasi-steady state approximations (QSSA) (Segel, 1988; Segel and Slemrod, 1989). This enabled no more than two model parameters to be estimated per fit. In short, QSSA predicts that compartments with rapid attrition reach a dynamic equilibrium with their slow varying “feeder” compartment. For instance, the output transfer rate, \( k_{BM}/H11001 \), of malathion in blood \( B(t) \) to total body metabolites \( M(t) \) and to storage tissues \( S(t) \) was considered large compared to the rates of change of its feeder compartments: \( R(t) \) for absorption or \( S(t) \) for storage release, because the burden of malathion in blood is quickly depleted under an oral dose (Jellinek et al., 2000).

The transfer rate constant of total metabolites in the body to urine, \( k_{MU} \), was estimated by least-square-fit adjustment of the model general solution to the data of Feldmann and Maibach (1974) on the urinary excretion time course of average total \(^{14}\text{C}\) in male human volunteers \( (n = 6) \) exposed intravenously \( (iv) \) to 1 \( \mu \text{Ci} \) of \(^{14}\text{C}\)-labeled malathion (dose in mole or g not specified). These authors reported the time course of total \(^{14}\text{C}\) urinary excretion rate (\% of the administered dose per h). For the fitting of experimental data and to determine the \( k_{MU} \) parameter, the experimental values were converted to cumulative burdens expressed as a fraction of the administered dose. Log-linear regression on the time course of average total \(^{14}\text{C}\) urinary excretion rate reported by Feldmann and Maibach (1974) during the 24- to 120-h period following the iv injection also served to determine the low transfer rate constant \( k_{SB} \) of malathion from storage tissues to blood.

The transfer rate constants of malathion in blood to total body metabolites, \( k_{BM} \), and to storage tissues, \( k_{BS} \), could not be determined precisely for lack of available detailed time profile data of malathion in blood shortly following malathion exposure in human volunteers. However, the sum of \( k_{BM} + k_{BS} \) could be approximated from the data of Jellinek et al. (2000). These authors showed that 1 h following a single oral dose of 15 mg/kg of malathion in human volunteers, blood concentrations of malathion were below the limit of detection (102 ng/ml). This corresponds to an attrition half-life of blood malathion of at most 12 min, which is consistent with a very rapid distribution or biotransformation of malathion. These findings indicate that the rate value of the sum of \( k_{BM} + k_{BS} \) must be greater or equal to \( \ln 2/12 \text{ min} \) or 3.47. The individual values of \( k_{BM} \) and \( k_{BS} \) parameters were determined by adjustment of the numerical solution of the system of differential equations to the data of Feldmann and Maibach (1974) in volunteers exposed iv to \(^{14}\text{C}\)-malathion.
The data of Jellinek et al. (2000) were used to determine the fractions of total body metabolites recovered as each of the five metabolites: MCA, DCA, DMTP, DMDTP, and DMP. These authors reported mean urinary excretion of these metabolites (as a percentage of the administered dose) for the 0–12-, 12–24-, and 24–48-h periods following a single oral malathion dose of 0.5, 1.5, 10, and 15 mg/kg of body weight in male human volunteers as well as 15 mg/kg in female volunteers (n = 5 per group). The fractions of specific to total body metabolites were thus obtained from the molar ratio of specific metabolites excreted in urine to total metabolites in urine. This amounts to assuming that the metabolites are produced in different proportions, but are excreted in urine at the same rate. This simplification was necessary, given the lack of detailed blood and urine excretion-time profiles for each metabolite and the paucity of data on the conjugation reactions of malathion metabolites and their renal clearance. However, the data of Feldmann and Maibach (1974), from which the kabs rate was derived, together with the data of Jellinek et al. (2000) show that elimination of the metabolites to urine is almost complete 12 to 24 h following an iv or oral exposure. It is thus reasonable to assume that the renal clearance of the different conjugated malathion metabolites is more or less the same.

The oral absorption fraction used in simulations was that reported by Jellinek et al. (2000). The experimental data did not allow for the exact determination of the oral absorption rate constant; a range of values was tested,
consistent with data from studies on related compounds (Carmichael et al., 1989; Nolan et al., 1984), together with the duration of intestinal transit up to the main absorption site of malathion (jejunum). For dermal exposures, the absorption fraction and the absorption-rate constant were estimated by least-square-fit adjustment of the model general solution to the data of Feldmann and Maibach (1974) on the urinary excretion time course of average total \(^{14}\)C in male human volunteers \((n = 6)\) exposed dermally to 1 \(\mu\)Ci of \(^{14}\)C-labeled malathion on the forearms.

**Model development: Model simulations.** Once the parameters were estimated as described above, numerical solution of the complete model was performed by the Runge-Kutta method on the differential equation system. Simulations were conducted again using the professional edition of MathCad software from MathSoft, Inc. The model predicts the amounts of malathion and its metabolites in the body and in excreta at any time point postexposure for any route of exposure and scenario.

Simulations of exposure scenarios, where continuous or repeated intermittent doses are administered through time, were performed by introducing a nonhomogeneous term, \(g(t)\), describing these varying inputs (see Appendix).

**Model validation.** The model, developed using the previously mentioned data, was validated using different sets of experimental data: Maibach et al. (1971), Wester et al. (1983), and Dennis and Lee (1999). Only in the study of Dennis and Lee (1999) were the data presented in graphical form. Those graphs were thus scanned and the data were read using Sigma Plot Graphing Software (Jandel Corporation, San Rafael, CA).

**Determination of biological reference values.** Biological reference values for MCA, DCA, and phosphoric derivatives amounts in 24-h urine samples are proposed here based on model predictions and the data of Moeller and Rider (1962). These authors exposed male human volunteers repeatedly, once a day, to gelatin capsules containing malathion. The dosage regimen was 8 mg/day (0.1 mg/kg/day) for 32 days, 16 mg/day (0.2 mg/kg/day) for 47 days, or 24 mg/day (0.3 mg/kg/day) for 56 days \((n = 5\) per exposure group). Plasma and erythrocyte cholinesterase activities were measured twice weekly before, during, and after administration. No clinical signs or symptoms of toxicity were observed during the study period at any dose level. The exposure dose’s NOEL and lowest-observed-effect level (LOEL) were estimated by Moeller and Rider (1962) to be 0.2 and 0.3 mg/kg/day, respectively, based on a \(>10\%\) inhibition of both plasma and erythrocyte cholinesterases as compared to baseline values. We reanalyzed the data of Moeller and Rider (1962), considering as significant an inhibition of \(\geq 19\%\) plasma and \(\geq 12\%\) erythrocyte cholinesterases in two successive measurements, as proposed by several authors (Gage, 1967; Heath and Vale, 1992; Larsen et al., 1982). Though we used a different criterion, the NOEL and LOEL arrived at are identical to those of Moeller and Rider (1962).

From the NOEL value of 0.2 mg/kg/day, the corresponding absorbed dose was estimated, and, using the model, associated urinary amounts of MCA, DCA, and phosphoric derivatives were obtained.

To determine biological reference values starting from the amounts of MCA, DCA, and phosphoric derivatives accumulated in urine 0–24 h following the onset of exposure, the model input conditions were set to generate biological reference values with a safety margin. This was achieved with an 8-h/day absorbed NOEL dose, using the lowest absorption rate constant found compatible with the various experimental data available. The fractions of total metabolites recovered in urine as MCA, DCA, and phosphoric derivatives were also attributed the smallest individual values determined using data from the study of Krieger and Dinoff (2000), who were the only authors to report individual values of MCA, DCA, and phosphoric derivatives in urine. These input conditions ensure a “minimal” urinary excretion of the metabolites for a given dose. This provides a safety margin through a possible overestimation of the dose absorbed when starting from urinary measurements.

**Application.** The proposed biological reference values were applied to assess health risk in workers exposed to malathion. Unpublished urinary excretion data from botanical garden workers, collected by the Institut National de Santé Publique du Québec (INSPQ), Direction de la Toxicologie Humaine (Québec, Canada), were used. Twenty-four-hour urine samples were collected in male and female subjects \((n = 9\) and 2, respectively) following a 2- to 3-h work shift in greenhouses that had been sprayed with malathion 12 to 15 h earlier. The workers studied had not participated in the application of malathion.

Samples were analyzed for \(\alpha\)- and \(\beta\)-MCA and DCA metabolite contents at the INSPQ. Urinary levels were determined by capillary gas chromatography/mass spectrometry (GC/MS) after acidification, extraction on C\(_8\) micro-columns, and derivatization with diazomethane. Analysis was performed using a model 5890 gas chromatography system (Hewlett Packard), a model 5972 mass spectrometer (Hewlett Packard) and a fused silica capillary column HP-5MS \((30\,\text{m} \times 0.32\,\text{mm}, 0.25\,\mu\text{m})\). The limits of detection for MCA and DCA were 2 and 1 \(\mu\)g/l, respectively. Average recovery of urine samples spiked with 10 \(\mu\)g/l of authentic reference standards was 113 and 108% for MCA and DCA, respectively \((n = 10\) samples). Inter-day coefficient of variation for replicate analysis of the same urine sample spiked with 10 \(\mu\)g/l of reference standard was 5.6 and 4.8% for MCA and DCA, respectively \((n = 10\) days).

**RESULTS**

**Model Development**

Table 2 presents parameter values of the model determined using the data of Feldmann and Maibach (1974) and Jellinek et al. (2000). Figure 2 shows that with these parameter values, the model reproduces closely the data of Feldmann and Maibach.

### TABLE 2

**Numerical Values of Parameters Used in the Model**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Absorption fractions</td>
<td></td>
</tr>
<tr>
<td>( f_{\text{oral}} )</td>
<td>0.738</td>
</tr>
<tr>
<td>( f_{\text{dermal}} )</td>
<td>0.0705</td>
</tr>
<tr>
<td>Transfer rates</td>
<td></td>
</tr>
<tr>
<td>( k_{\text{abs-oral}} )</td>
<td>0.345–1.373</td>
</tr>
<tr>
<td>( k_{\text{abs-dermal}} )</td>
<td>0.103</td>
</tr>
<tr>
<td>Metabolite fractions</td>
<td></td>
</tr>
<tr>
<td>( f_{\text{MCA}} )</td>
<td>0.544</td>
</tr>
<tr>
<td>( f_{\text{DCA}} )</td>
<td>0.131</td>
</tr>
<tr>
<td>( f_{\text{DMTP}} )</td>
<td>0.019</td>
</tr>
<tr>
<td>( f_{\text{MSP}} )</td>
<td>0.182</td>
</tr>
<tr>
<td>( f_{\text{MFP}} )</td>
<td>0.124</td>
</tr>
</tbody>
</table>

**Note.** The description of constant parameters is given in Table 1. Absorption fractions and transfer rates were determined in this study by statistical best-fits; metabolite fractions were obtained from the literature.

*These values were determined from the data of Feldmann and Maibach (1974). However, the dermal absorption fraction and absorption rate constants are known to be subject to variations among individuals and depending on the exposed skin region. To simulate the various data presented in this paper, the value range for \( f_{\text{abs-oral}} \) was found to be 0.04–0.25 and for \( k_{\text{abs-dermal}} \) 0.05–0.123.

*Different values for \( k_{\text{oral}} \) were used, corresponding to absorption half-lives between 30 min and 2 h.*

Using the \( k_{\text{oral}} \) value of 0.215 determined from the data of Feldmann and Maibach (1974), the best fit was obtained for a \( k_{\text{oral}}//k_{\text{oral}} \) ratio equal to 25 and a rate value for the sum of \( k_{\text{oral}} + k_{\text{oral}} \) equal to 6.24, indicating that \( k_{\text{oral}} + k_{\text{oral}} \approx k_{\text{oral}} \) and malathion biotransformation is a very rapid process \((k_{\text{oral}} = 6)\).
(1974) on the cumulative urinary excretion time course of total $^{14}$C in human volunteers following an intravenous (A) and dermal (B) exposure to $1 \mu$Ci of $^{14}$C-malathion. Each point represents mean value of experimental data ($n = 6$ per exposure group).

FIG. 2. Model simulations (lines) compared with experimental data of Feldmann and Maibach (1974) (symbols) on the cumulative urinary-excretion time course of total $^{14}$C (% of administered dose) in male human volunteers following an intravenous (A) and dermal (B) exposure to $1 \mu$Ci of $^{14}$C-malathion. Each point represents mean value of experimental data ($n = 6$ per exposure group).

The model was also validated through simulations of the data of Dennis and Lee (1999) on the urinary excretion time course of phosphoric derivative metabolites (the sum of DMDTP, DMTP, and DMP) in volunteers exposed to an aqueous- or alcohol-based head lice formulation containing malathion (Fig. 4). The model provided a close approximation to the data presented by these authors using the model parameter values in Table 2, except for the dermal absorption fraction and rate constant. For the simulation of exposure to the aqueous-based formulation, the absorption fraction was set equal to 0.075; the value of the absorption rate constant was left as that reported in Table 2. For the simulation of exposure to the alcohol-based solution, the absorption fraction was set equal to 0.068 and the absorption rate constant to 0.123 h$^{-1}$.

Inferences of the model: Prediction of the time course of malathion and its metabolites in the body and excreta under different exposure scenarios. The model was used to predict the time course of malathion and its metabolites in the body and in accessible biological matrices (blood, urine) under different exposure scenarios likely to occur in practice. Figures 5 and 6 show model predictions of the time profiles of malathion and its metabolites in the body and urine that would result from a single oral or dermal exposure to malathion. Following oral exposure, with an absorption fraction of 0.738 and absorption rate constant of 1.373 h$^{-1}$ (half-life of 30 min) as reported in Table 2, the predicted time course of malathion in blood $B(t)$ shows maximum level 19-min postexposure, which represents 10.6% of the exposure dose (14.4% of the absorbed dose). Because of an early partial storage in tissues and a slow return from these tissues to blood, elimination of malathion from blood follows a bi-exponential pattern with a more rapid phase followed by a slower phase (Fig. 5A). Model simulations predict a low transfer of blood malathion to storage tissues $S(t)$, where the maximum storage level represents 2.6% of the exposure dose (3.5% of the absorbed dose) (Fig. 5A).

Model Validation

The proposed model was applied to the data collected by Maibach et al. (1971) (Fig. 3) and Wester et al. (1983) (data not shown) on the urinary excretion time profile of average total $^{14}$C in male human volunteers following a single dermal application of $^{14}$C-labeled malathion at different anatomical regions of the body. The parameter values in Table 2 were used for these model simulations, except for the dermal absorption fraction and rate constant, which are known to vary largely according to the exposed region of the skin as well as from one individual to another. To simulate the experimental data of Maibach et al. (1971), the best-fit values for the dermal absorption fraction were 0.075, 0.062, 0.134, and 0.25 for applications to the forearm, the palm of the hand, the hand dorsum and the forehead, respectively. The corresponding values for the absorption rate constant were: 0.103, 0.05, 0.11, and 0.11 h$^{-1}$. To simulate the data of Wester et al. (1983) in volunteers exposed dermally to 6.25 $\mu$Ci of $^{14}$C-malathion on the forearm, the dermal absorption fraction was found to be 0.04 and the absorption rate constant 0.05 h$^{-1}$.

Inferences of the model: Prediction of the time course of malathion and its metabolites in the body and excreta under different exposure scenarios. The model was used to predict the time course of malathion and its metabolites in the body and in accessible biological matrices (blood, urine) under different exposure scenarios likely to occur in practice. Figures 5 and 6 show model predictions of the time profiles of malathion and its metabolites in the body and urine that would result from a single oral or dermal exposure to malathion. Following oral exposure, with an absorption fraction of 0.738 and absorption rate constant of 1.373 h$^{-1}$ (half-life of 30 min) as reported in Table 2, the predicted time course of malathion in blood $B(t)$ shows maximum level 19-min postexposure, which represents 10.6% of the exposure dose (14.4% of the absorbed dose). Because of an early partial storage in tissues and a slow return from these tissues to blood, elimination of malathion from blood follows a bi-exponential pattern with a more rapid phase followed by a slower phase (Fig. 5A). Model simulations predict a low transfer of blood malathion to storage tissues $S(t)$, where the maximum storage level represents 2.6% of the exposure dose (3.5% of the absorbed dose) (Fig. 5A). As mentioned above, return to blood from $S(t)$ is, however, relatively slow; the predicted half-life for this process is 19.8 h. Malathion is also readily and extensively metabolized in the
The body and its metabolites are rapidly eliminated once formed; the maximum level of total metabolites M(t) is reached 1.7 h postexposure and represents 48.9% of the exposure dose (66.3% of the absorbed dose) (Fig. 5A). The cumulative urinary excretion time courses of malathion metabolites show that the different metabolites are rapidly eliminated (Fig. 5B); the model predicts that 79, 90, and 98% of total amounts recovered in urine are excreted during the first 8, 12, and 24 h postexposure, respectively. Asymptotically, urinary MCA, DCA, and phosphoric derivatives represent, respectively, 36.1, 8.7, and 21.6% of the exposure dose (48.9, 11.8, and 29.2% of the absorbed dose).

Following a single dermal exposure, using an absorption fraction of 0.070 and an absorption rate constant of 0.103 h⁻¹.

### Table 3

Comparison of Model Simulations with the Experimental Data of Jellinek et al. (2000) on the Cumulative Urinary Excretion of Malathion Metabolites 0–12, 0–24, and 0–48 h following a Single Oral Exposure to 0.5, 1.5, 10, or 15 mg/kg of Malathion in Male and Female Human Volunteers

<table>
<thead>
<tr>
<th>Malathion metabolite</th>
<th>Urine collection period (h)</th>
<th>0.5 mg/kg (male)</th>
<th>1.5 mg/kg (male)</th>
<th>10.0 mg/kg (male)</th>
<th>15.0 mg/kg (male)</th>
<th>15.0 mg/kg (female)</th>
<th>Mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>MCA</td>
<td>0–12</td>
<td>39.5</td>
<td>33.6</td>
<td>30.7</td>
<td>41.3</td>
<td>29.6</td>
<td>35.0</td>
</tr>
<tr>
<td></td>
<td>0–24</td>
<td>39.7</td>
<td>34.0</td>
<td>31.5</td>
<td>42.6</td>
<td>30.1</td>
<td>35.6</td>
</tr>
<tr>
<td></td>
<td>0–48</td>
<td>39.9</td>
<td>34.0</td>
<td>31.6</td>
<td>42.7</td>
<td>30.3</td>
<td>35.7</td>
</tr>
<tr>
<td>DCA</td>
<td>0–12</td>
<td>11.4</td>
<td>3.7</td>
<td>3.4</td>
<td>10.0</td>
<td>3.9</td>
<td>6.5</td>
</tr>
<tr>
<td></td>
<td>0–24</td>
<td>13.1</td>
<td>7.1</td>
<td>4.8</td>
<td>11.9</td>
<td>4.4</td>
<td>8.3</td>
</tr>
<tr>
<td></td>
<td>0–48</td>
<td>15.9</td>
<td>7.3</td>
<td>5.0</td>
<td>12.1</td>
<td>4.6</td>
<td>9.0</td>
</tr>
<tr>
<td>Phosphoric derivatives</td>
<td>0–12</td>
<td>15.0</td>
<td>21.7</td>
<td>18.0</td>
<td>24.8</td>
<td>15.9</td>
<td>20.1</td>
</tr>
<tr>
<td></td>
<td>0–24</td>
<td>15.9</td>
<td>24.4</td>
<td>19.5</td>
<td>26.6</td>
<td>16.9</td>
<td>20.6</td>
</tr>
<tr>
<td></td>
<td>0–48</td>
<td>17.2</td>
<td>24.7</td>
<td>20.0</td>
<td>26.9</td>
<td>17.5</td>
<td>21.3</td>
</tr>
</tbody>
</table>

Note. MCA, malathion mono-carboxylic acids; DCA, malathion dicarboxylic acids; phosphoric derivatives, the sum of dimethyl dithiophosphate (DMDTP), dimethyl thio phosphate (DMTP), and dimethyl phosphate (DMP). The predicted values reported in this table were obtained using an oral absorption rate of 1.373.

**FIG. 3.** Model simulations (lines) compared with experimental data of Maibach et al. (1971) (symbols) on the cumulative urinary excretion time course of total ¹⁴C (% of administered dose) in male human volunteers following a dermal exposure to ¹⁴C-malathion (4 μg/cm²) on the forearm (×), the palm of the hand (+), the hand dorsum (open square) and the forehead (open diamond). Each point represents the mean value of experimental data (n = 6 per exposure group).

**FIG. 4.** Model simulations (lines) compared with experimental data of Dennis and Lee (1999) (symbols) on the cumulative urinary excretion time course of phosphoric derivative metabolites (the sum of DMDTP, DMTP, and DMP) (nmol) in human volunteers following a dermal exposure (on an intact skin) to an aqueous-based head-lice formulation containing on average 0.126 g of malathion (open circle) and an alcohol-based head-lice formulation containing on average 0.089 g of malathion (open square). Each point represents mean value of experimental data (n = 5 to 10 volunteers per treatment group).
As reported in Table 2, the predicted time courses of malathion in blood $B(t)$ and in storage tissues $S(t)$, and the total metabolites in the body $M(t)$ show respective peak levels of 0.67, 16.1, and 6.5 h postexposure (Fig. 6A). Since the dermal absorption of malathion is small and relatively slow compared to metabolism and elimination, maximum values for $B(t)$, $S(t)$, and $M(t)$ only amount to 0.11, 0.16, and 1.55% of the exposure dose, respectively (1.6, 2.3, and 22.0% of the absorbed dose). Additionally, because dermal absorption of malathion is slow compared to its biotransformation, a dynamic equilibrium is quickly reached between the skin compartment $R(t)$ and blood compartment $B(t)$. Consequently, $B(t)$ begins its attrition at the rate of the absorption rate constant of 0.103 h$^{-1}$ (half-life of 6.7 h), which is not the case following an oral exposure, where absorption is more rapid than elimination processes (Fig. 5A).

From the cumulative urinary-excretion time courses of malathion metabolites (Fig. 6B), the model predicts that 32, 51, and 83% of total amounts recovered in urine are excreted during the first 8, 12, and 24 h, respectively, following a dermal exposure. Asymptotically, urinary MCA, DCA, and phosphoric derivatives represent 3.45, 0.83, and 2.06% of exposure dose (48.9, 11.8, and 29.2% of the absorbed dose; these latter excretions are identical to those after oral exposure, as should be). These simulations show that following a dermal exposure
to malathion, the absorption-rate constant governs the overall urinary excretion rate of the metabolites, because the dermal absorption rate, $k_{abs-dermal}$, is small compared to the biotransformation rate and renal clearance (represented in the model by $k_{BM}$ and $k_{MU}$, respectively). According to model predictions, to recover half of the absorbed dose of malathion eventually excreted in urine as metabolites takes 11.8 h following a single dermal application, compared to 3.2 h following an intravenous injection and 4.0 h after oral exposure. Furthermore, as mentioned previously, the dermal absorption rate constant varies among individuals and depends on the skin region exposed to malathion. Table 4 shows that, for dermal exposure to malathion, varying the absorption rate constant markedly affects the cumulative urinary excretion time course of total metabolites (the sum of MCA, DCA, and phosphoric derivatives).

The proposed model can also predict the time evolution of malathion and its metabolites in the body and excreta following repeated exposures to malathion. Simulations of an 8-h-per-day dermal exposure, 5 days a week for 4 consecutive weeks, are shown in Figure 7. With the parameter values given in Table 2, during a week of exposure, there is a slight increase from day to day in peak burdens of malathion in blood $B(t)$ and in storage tissues $S(t)$ and total body metabolites $M(t)$ (measured as a fraction of the absorbed daily dose). However, there is no significant week-to-week increase in daily maximum levels, because, at the end of a 2-day break in exposure, the elimination is nearly complete.

Reconstruction of the absorbed dose of malathion starting from urinary measurements of the metabolites. The model can help estimate, through back calculations, the absorbed dose of malathion starting from urinary measurements of the metabolites, provided the periods of exposure and urine collection are known. Table 5 shows simulations of the cumulative urinary excretion of total metabolites, expressed as a fraction of the absorbed dose, at different time periods following the onset of an 8-h dermal exposure to malathion.

### Table 4
Effect of Variations in the Absorption Rate Constant on the Simulated Cumulative Urinary Excretion of Total Metabolites over Different Time Periods following a Single Dermal Exposure to Malathion

<table>
<thead>
<tr>
<th>Urine collection period postexposure (h)</th>
<th>Cumulative urinary excretion of total metabolites (fraction of the absorbed dose)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>$k_{abs} = 0.12$ $k_{abs} = 0.10$ $k_{abs} = 0.07$ $k_{abs} = 0.05$</td>
</tr>
<tr>
<td>0–4</td>
<td>0.09 0.08 0.06 0.04</td>
</tr>
<tr>
<td>0–8</td>
<td>0.30 0.26 0.20 0.15</td>
</tr>
<tr>
<td>0–12</td>
<td>0.48 0.43 0.34 0.26</td>
</tr>
<tr>
<td>0–16</td>
<td>0.62 0.56 0.46 0.37</td>
</tr>
<tr>
<td>0–24</td>
<td>0.77 0.73 0.64 0.53</td>
</tr>
<tr>
<td>0–36</td>
<td>0.86 0.84 0.78 0.69</td>
</tr>
<tr>
<td>0–48</td>
<td>0.88 0.87 0.84 0.78</td>
</tr>
<tr>
<td>0–200</td>
<td>0.90 0.90 0.90 0.90</td>
</tr>
</tbody>
</table>

*Note.* $k_{abs}$, dermal absorption rate constant values, which are in the range of those used for simulations of the various dermal data presented in this article.

### Table 5
Predicted Cumulative Urinary Excretion of Total Metabolites (the Sum of Mono- and Dicarboxylic Acids and Phosphoric Derivatives) at Different Time Periods following the Onset of an 8-h Dermal Exposure to Malathion

<table>
<thead>
<tr>
<th>Urine collection period following the onset of exposure (h)</th>
<th>Cumulative urinary excretion of total metabolites (fraction of the absorbed dose)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0–4</td>
<td>0.02 0.01</td>
</tr>
<tr>
<td>0–6</td>
<td>0.06 0.03</td>
</tr>
<tr>
<td>0–8</td>
<td>0.13 0.06</td>
</tr>
<tr>
<td>0–12</td>
<td>0.32 0.16</td>
</tr>
<tr>
<td>0–16</td>
<td>0.49 0.27</td>
</tr>
<tr>
<td>0–20</td>
<td>0.62 0.38</td>
</tr>
<tr>
<td>0–24</td>
<td>0.72 0.46</td>
</tr>
<tr>
<td>0–36</td>
<td>0.84 0.65</td>
</tr>
<tr>
<td>0–48</td>
<td>0.88 0.76</td>
</tr>
<tr>
<td>0–60</td>
<td>0.89 0.82</td>
</tr>
<tr>
<td>0–72</td>
<td>0.89 0.85</td>
</tr>
</tbody>
</table>

*Note.* The predicted cumulative urinary excretion of total metabolites over different time periods is given considering two dermal exposure scenarios, with relatively high and low absorption rate constants (0.12 and 0.05, respectively). Background urinary excretion is considered negligible. Also note that, eventually, no more than 90% of absorbed dose will be excreted in urine; $k_{abs}$, dermal absorption rate constant.
of an 8-h work shift and assuming zero background levels. Simulations are performed under two dermal-exposure scenarios, one with a high value for the absorption rate constant (0.12 h⁻¹) and another with a low value (0.05 h⁻¹). Given the importance of the absorption rate constant in determining dermal exposure, the excretion values reported in Table 5 can be viewed as ‘best’ and ‘worst’ case scenarios. From Table 5, the dose absorbed by workers can be inferred from measurements of the amounts of metabolites in urine, when the urine collection time period following the beginning of a work shift is known as well as the work-shift duration.

**Biological reference values.** The model also served to establish biological reference values for malathion metabolites in urine since it links the absorbed dose to the amounts of metabolites excreted in urine over different time periods. From the data of Moeller and Rider (1962), a NOEL oral exposure dose of 0.61 μmol/kg/day (0.2 mg/kg/day) was derived. With the estimated oral absorption fraction (see Table 2), this corresponds to an absorbed dose NOEL of 0.45 μmol/kg/day (0.15 mg/kg/day). The corresponding biological reference values for MCA, DCA phosphoric derivatives are then 44, 13, and 62 nmol/kg, respectively, in 24-h urine samples. The values for the sum of acids or total metabolites are thus 57 and 119 nmol/kg, respectively.

**Application**

The model was used to assess the health risk related to malathion exposure in botanical-garden workers, starting from measurements of the amounts of MCA and DCA metabolites in 24-h urine samples (as described in the Materials and Methods section). The workers exhibited excretion values of MCA and DCA in their 24-h urine samples corresponding to a range of 0.017 to 0.210 times the biological reference value proposed for MCA and a range of 0.011 to 0.324 times the value proposed for DCA, indicating a negligible health risk for those workers (Table 6).

**DISCUSSION**

A heuristic toxicokinetic model was developed, which integrates a wealth of experimental *in vivo* time-profile data, to uncover the critical biological determinants of the kinetics of malathion and its metabolites in humans. The model provides a good understanding of the time evolution of malathion and its metabolites in the body and in urine, under different exposure scenarios (single, repeated, continuous, or intermittent exposure). With this model, it is also possible to assess the effect of the route of entry on time-course data. In the context of biological monitoring, the model facilitates the interpretation of urinary measurements collected over different time periods. It can also be used to propose optimum sampling strategies for routine biological monitoring.

The model showed that, upon absorption, malathion in blood is either readily biotransformed or transferred to storage tissues, though only a small fraction of the absorbed dose of malathion is in fact transferred to storage tissues. Metabolites are also rapidly excreted in urine and feces. Consequently, the exposure route determines the time-dependent urinary excretion rate of malathion metabolites, because different routes-of-entry imply different absorption rates. For example, when simulating a single oral exposure, 80–89% of the absorbed dose is eliminated from the body within 12 h, whereas, after a single dermal application, only between 29–53% of the absorbed dose is excreted during the same time period (these value ranges are obtained by considering the lowest and highest absorption rate constant used to simulate the various available data). In workers who are typically exposed to malathion mainly through the skin, the absorption rate thus limits the output rates of metabolites in urine, because dermal absorption is much slower than biotransformation rate and renal clearance. This is not the case in individuals exposed by the oral or pulmonary routes, where the absorption rate is not a limiting step in the excretion kinetics.

Considering a continuous 8-h dermal exposure during a workday, the model predicts that 52–80% of the absorbed dose will be eliminated from the body within the 24-h period following the onset of exposure, as compared to 84–98% within 48 h. Consequently, for the biological monitoring of worker exposure to malathion through measurements of the amounts of urinary metabolites, collection of 24-h urine samples, start-

---

**TABLE 6**

<table>
<thead>
<tr>
<th>Subjects</th>
<th>Sex*</th>
<th>MCA</th>
<th>DCA</th>
<th>MCA</th>
<th>DCA</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>F</td>
<td>47.3</td>
<td>7.9</td>
<td>0.020</td>
<td>0.011</td>
</tr>
<tr>
<td>2</td>
<td>F</td>
<td>159.4</td>
<td>69.4</td>
<td>0.066</td>
<td>0.097</td>
</tr>
<tr>
<td>3</td>
<td>F</td>
<td>205.0</td>
<td>75.5</td>
<td>0.085</td>
<td>0.106</td>
</tr>
<tr>
<td>4</td>
<td>F</td>
<td>80.8</td>
<td>12.9</td>
<td>0.033</td>
<td>0.018</td>
</tr>
<tr>
<td>5</td>
<td>F</td>
<td>42.2</td>
<td>10.3</td>
<td>0.017</td>
<td>0.014</td>
</tr>
<tr>
<td>6</td>
<td>F</td>
<td>113.4</td>
<td>23.8</td>
<td>0.047</td>
<td>0.033</td>
</tr>
<tr>
<td>7</td>
<td>M</td>
<td>51.4</td>
<td>31.5</td>
<td>0.017</td>
<td>0.035</td>
</tr>
<tr>
<td>8</td>
<td>F</td>
<td>98.5</td>
<td>45.5</td>
<td>0.041</td>
<td>0.064</td>
</tr>
<tr>
<td>9</td>
<td>M</td>
<td>228.7</td>
<td>105.9</td>
<td>0.074</td>
<td>0.116</td>
</tr>
<tr>
<td>10</td>
<td>F</td>
<td>62.4</td>
<td>15.5</td>
<td>0.026</td>
<td>0.022</td>
</tr>
<tr>
<td>11</td>
<td>F</td>
<td>509.0</td>
<td>231.7</td>
<td>0.210</td>
<td>0.324</td>
</tr>
</tbody>
</table>

*Note. MCA, malathion mono-carboxylic acids; DCA, malathion dicarboxylic acids. Cumulative 24-h urinary excretion is given in nmol. F, female and M, male, which are considered to weigh, as default values, 55 and 70 kg of body weight, respectively.

*Biological reference values proposed in this study for MCA and DCA are, respectively, 44 and 13 nmol/kg in 24-h urine samples.*
ing at the beginning of the work shift, appears as an adequate sampling strategy. In the case of workers subjected to exposure day after day, malathion is predicted by the model to build up in storage tissues during the course of a workweek, thus resulting in a progressive increase in total body burden. Repeating the exposure from week to week does not, however, cause any significant increase in maximum and minimum body burdens. In this situation, it is best to collect the 24-h urine samples on the last day of a workweek. Measurements should be repeated periodically when exposure levels are suspected to vary significantly with time.

It is interesting to note that, based on the data of Jellinek et al. (2000), cumulative amounts of metabolites in urine (expressed as a percentage of the malathion exposure dose) appear in the following order: MCA > phosphoric derivatives > DCA. These findings, combined with the fact that the MCA metabolite is specific to malathion exposure (Márquez et al., 2001; Tuomainen et al., 2002), suggest that MCA in urine is the most useful individual biological indicator of exposure to malathion. Of course, measurements of phosphoric derivatives can also be of interest as nonspecific bio-indicators of exposure to organophosphate compounds (Cocker et al., 2002; Coye et al., 1986).

The model enables links to be established at all times between the dose, the body burden of malathion and that of its metabolites, and the amounts of specific metabolites excreted in urine. The model can thus be used to reconstruct, through back calculations, the absorbed dose of malathion following oral or dermal exposure, starting from measurements of cumulative amounts of a specific metabolite excreted in urine over a given period of time. Reconstructing the absorbed dose from metabolites in urine avoids unnecessary assumptions or approximations about the absorption fraction, which is known to be subject to large inter-individual variations and, in the case of dermal exposure, to vary according to anatomical skin regions (ACGIH, 2002; Cohen and Rice, 2001; Feldmann and Maibach, 1974).

It was also possible, with the model, to propose biological reference values with a margin of safety by simulating a dermal exposure scenario typical of a daily worker exposure, and using a combination of kinetic parameters that overestimates the reconstructed absorbed dose, starting from urinary measurements. The establishment of biological reference values, based on a dermal-exposure scenario rather than an oral or pulmonary exposure, contributes to safe estimates whatever the exposure route. Indeed, in an individual exposed to malathion mainly by inhalation or ingestion, the urinary excretion of metabolites is more rapid than following a dermal exposure and hence urinary excretion values in 24-h urine samples collected at the beginning of an exposure period are more important. It should also be reminded that the biological reference values were determined with the model, using the slowest absorption rate found compatible with the available literature data on excretions, together with the lowest individual values found for the fractions of total metabolites recovered in urine as MCA, DCA, or phosphoric derivatives. This was to consider the variations in the absorption rate among individuals and depending on the exposed skin regions (Feldmann and Maibach, 1974) as well as the reported variations from one study to another in the fractions of total metabolites recovered in urine as MCA, DCA, and phosphoric derivatives (Bradway and Shafik, 1977; Jellinek et al., 2000; Krieger and Dinoff, 2000). However, the study of Jellinek et al. (2000) showed that over the 0.5 to 15 mg/kg oral dose range, the fraction of total metabolites recovered in urine as MCA, DCA, and phosphoric derivatives was independent of the dose.

To assess the risk of exposure to malathion, the amounts of MCA, DCA, and phosphoric derivatives in the 24-h urine samples of exposed subjects can be compared to the proposed reference values. Ideally, it is best to establish the risk from measurements of the sum of acids rather than MCA or DCA individually, for a specific but more precise estimate. Otherwise, the risk can be estimated from measurements of the sum of all metabolites in urine, which, despite not being entirely specific to malathion exposure, allows the bypass of uncertainties about the relative weights of each metabolite in total excretion. Also, when 24-h urine samples cannot be collected for practical reasons, collection periods can be shortened; Table 5 can be used to estimate the 24-h urinary excretion in exposed subjects starting from measurements over other urine collection periods. For a “safe-side” estimation, excretion values in Table 5 obtained from the slowest absorption rate should be used. However, to minimize the effects of inter-individual and inter-site variations of absorption rates on urinary outputs, it is best to collect urine samples over the longest possible time period.

The model and proposed biological reference values were used in this study to assess the risk for workers exposed to malathion in a botanical garden. The risk was predicted to be negligible given that the measurements of MCA and DCA in 24-h urine samples were lower than the proposed biological reference values. In general, these workers appear to be less exposed than the greenhouse workers of a recent Spanish report (Márquez et al., 2001) where MCA was measured in the urine of three individuals following applications of malathion. The total amounts of MCA excreted during the 24-h period following applications were calculated to be 134, 182, and 671 µg, corresponding to 31.7, 8.6, and 6.3 nmol/kg, assuming a body weight of 70 kg. These values represent respectively 0.72, 0.20, and 0.14 of the proposed biological reference value.

In summary, a toxicokinetic model was developed that accounts for the constraints related to significant variations in some of the parameters: (i) variations in the dermal absorption fraction and the rate of absorption among individuals and according to the exposed skin regions and (ii) variations from one report to another in the fractions of total metabolites recovered in urine as MCA, DCA, and phosphoric derivatives. In this study, the model was used to reconstruct the absorbed
dose starting from measurements of urinary biomarkers; the effect of varying absorption fractions was thus bypassed. Variations in the absorption rate and relative proportions of the different metabolites in urine can impair accurate estimation of the absorbed dose starting from amounts of biomarkers in urine. Under these conditions, to calculate biological reference values with a margin of safety, the smallest values for the latter parameters afforded by the literature were used. This leads to a possible overestimation of the corresponding value for the reconstructed dose. The model also assumes the absence of saturation in the metabolism and clearance processes; over the exposure dose range modeled in this study, the available data in volunteers and workers were accurately predicted without having to introduce saturation. The model cannot, however, be used to predict the kinetics in the saturating exposure dose ranges, as expected in the case of intoxicated subjects. The available literature data remained sufficient to build a robust model to better understand the kinetics of malathion and its metabolites and to propose convenient biological reference values together with sampling strategies. These can be of use immediately for the biological monitoring of malathion exposure through measurements of metabolites in urine.

APPENDIX

First order linear differential equations for each compartment. From Figure 1, the following differential equations are obtained (see Table 1 for definitions of symbols and abbreviations):

\[
\frac{dR(t)}{dt} = g(t) - k_{\text{abs}} \times R(t)
\]

where \( g(t) \) is the absorbed oral or dermal dose \( D_{\text{abs}} \) per unit of time.

\[
D_{\text{abs}} = D_{\text{exp}} \times f_{\text{abs}}
\]

where \( f_{\text{abs}} \) is the absorption fraction and \( D_{\text{exp}} \) is the exposure dose.

For intravenous injection, \( g(t) = 0 \) for \( t > 0 \) and at time \( t = 0: B(0) = D_{\text{abs}}. \)

\[
\frac{dB(t)}{dt} = k_{\text{abs}} \times R(t) + k_{\text{ex}} \times S(t) - (k_{\text{abs}} + k_{\text{ex}}) \times B(t)
\]

\[
\frac{dS(t)}{dt} = k_{\text{ex}} \times B(t) - k_{\text{in}} \times S(t)
\]

\[
\frac{dM(t)}{dt} = k_{\text{in}} \times B(t) - (k_{\text{su}} + k_{\text{up}}) \times M(t)
\]

\[
\frac{dF(t)}{dt} = k_{\text{up}} \times M(t)
\]

\[
\frac{dM_{i}(t)}{dt} = f_i \times k_{\text{in}} \times B(t) - (k_{\text{su}} + k_{\text{up}}) \times M_i(t)
\]

\[
\frac{dU_i(t)}{dt} = k_{\text{su}} \times M_i(t)
\]

where: \( i \in \{ \text{MCA-C, DCA-C, DMDTP-C, DMTP-C, DMP-C} \} \), and \( M(t) = M_i(t) \).

ACKNOWLEDGMENTS

This study was funded by the Institut de Recherche en Santé et Sécurité du Travail du Québec. Authors wish to thank Pierre Dumas of the Institut national de Santé Publique du Québec for the analysis of malathion metabolites in urine.

REFERENCES

ACGIH (2002). Documentation of the Threshold Limit Values and Biological Exposure Indices, 7th ed. American Conference of Governmental Industrial Hygienists, Cincinnati, OH.


