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Mathematical Modeling — Guide to High-Dose Methotrexate Infusion Therapy

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Summary. *A mathematical model that describes methotrexate pharmacokinetics has been refined for use as a guide to dose escalation during high-dose methotrexate infusion therapy. Parameters for the model are adjusted for a patient by the SAAM computer program of Berman and Weiss, on the basis of plasma concentrations obtained during the initial course of therapy. Various dose escalations can be simulated by the computer and a print-out of predicted plasma concentrations obtained.*

The model has been used successfully to predict plasma concentrations after high-dose infusions in patients, including those with abnormal creatinine clearances. The program is designed to allow comparisons among infusions of any duration. This can be helpful when a change from a 24-hour infusion to a 6-hour infusion is contemplated for a patient.

Deviations of observed values from those predicted are used to warn of the possibility of delayed toxicity secondary to methotrexate and alert the physician that increased amounts of rescue agent may be required.

Introduction

High-dose methotrexate infusions in man are possible when normal cells are 'rescued' from the lethal effects of the drug by reduced folates [8, 18] or thymidine [10]. Several tumors are responsive to this form of therapy even when they were previously resistant to standard doses of methotrexate [7, 8, 10, 11, 16, 18, 25]. These infusions are well tolerated and relatively safe when certain guidelines are followed. The guidelines include recommendations for proper selection of patients and for insuring adequate patient hydration and urine alkalinity [22, 24, 30]. Monitoring of renal function and plasma methotrexate concentrations has been shown to add to the safety of high-dose methotrexate protocols [4, 22, 29, 30].

Two major forms of toxicity occur with high-dose methotrexate infusion therapy. One type is direct damage secondary to the physical properties of methotrexate [24]. The target organ for this type of toxicity is the kidney. When large amounts of methotrexate are delivered to the kidney tubules, crystallization of this weak acid can occur if the pH in the tubules is acidic. If urine flow is slow, blockage, with subsequent destruction of the tubules, takes place. The other type of toxicity is due to the cytotoxic effects of the drug on normal tissues with a high turnover rate [4, 13, 24, 30, 33, 34]. This toxicity is manifest in bone marrow suppression, mucositis, and dermatitis.

Since urinary excretion is the major route of elimination, renal damage by methotrexate may lead to large amounts of drug being retained in the body for prolonged periods. This retention is reflected by elevated plasma concentrations. Increased plasma concentrations are often associated with an increased frequency of complications secondary to the cytotoxicity of the drug.

Although most authors agree that determination of plasma methotrexate concentrations is essential to the management of a patient, agreement as to how they should be used has not been reached. The problem is that plasma concentrations do not directly correlate with toxicity. Concentrations of over 1×10^{-3} *M* can be obtained in patients for short periods of time without adverse reactions [13, 27], while a concentration of 1×10^{-7} *M*, or even less, can cause cell death if this concentration is maintained for several days [23]. Since time is an important factor, a methotrexate concentration obtained from a random plasma sample may not be helpful in predicting toxicity unless an appropriate value is

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known for that time point. A concentration higher than the appropriate value would suggest delayed drug clearance and the need for increased amounts of rescue agent [9].

Determining an appropriate value for a plasma concentration can be a problem, since plasma concentrations are dependent upon the dose, schedule, and route of administration of a drug as well as upon characteristics particular to an individual patient. In the case of methotrexate, these characteristics include such factors as the size of the patient, the status of renal function, urine pH, degree of protein binding, and the presence of abnormal fluid accumulations (e.g., pleural fluid or ascites), which may act as drug reservoirs [13, 24, 30]. Furthermore, there are a variety of protocols for delivering high-dose methotrexate therapy. Most involve IV infusions, but there are protocols for delivering high-dose methotrexate as multiple IM injections [31]. Dosage ranges from below 500 mg to over 12 g/m^2. Time schedules for infusions range from single, bolus injections to infusions lasting over 36 h. There also appears to be a limitless variety of rescue schedules.

With the aid of clinical pharmacokinetics we can predict plasma concentrations. A mathematical model allows computer-assisted adjustments of dose, schedule, and route of administration of methotrexate, as well as allowing for adjustments for certain characteristics of an individual patient. Predicting plasma concentrations of drug in individual patients is proposed as the first step in predicting which patients will develop methotrexate toxicity.

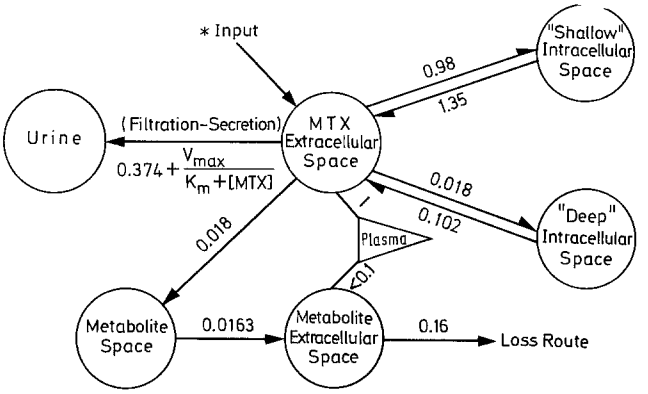

Fig. 1. Nonlinear compartmental model for methotrexate (MTX). *Arrows* represent drug transfer between compartments (*circles*). *Numbers* associated with *arrows* are rate constants (h^{-1}). The *triangle* is an operational unit representing plasma sampling as a linear combination of material in the two extracellular spaces, MTX and metabolite. The *number* associated with the *line* to the *triangle* is the fraction of material sampled from either the MTX extracellular space (always 100%) or the metabolite extracellular space (< 10%). The input function is dependent upon the amount of drug infused and the duration of the infusion

Materials and Methods

The mathematical model used to predict plasma methotrexate concentrations is a modification of two earlier models that use nonlinear kinetics to describe intercompartmental transport. The original model was based on methotrexate concentrations obtained from patients who received single, IV injections of tritium-labeled methotrexate at doses that did not require rescue drugs [19]. The next model accounted for methotrexate pharmacokinetics when high-dose infusions with rescue were given and plasma concentrations were obtained by radioimmunosassay for methotrexate [27]. For both these models, somewhat arbitrary mathematical expressions were used to generate continuous changes in the nonlinear rate parameters.

The current model (Fig. 1) is similar to the second, except that the urinary excretion parameter is expressed in terms of Michaelis-Menten kinetics. The differential equations for the model are solved by a Control Data Corporation 6400 computer using the Simulation, Analysis, and Modeling (SAAM) program of Berman and Weiss [1–3].

To simulate IV infusion methotrexate is introduced at a constant concentration per unit time into the central, extracellular fluid compartment. The size (in liters) of this compartment is estimated to be 18% of body weight. The computer program allows the dose and the length of time for the infusion to be specified.

Methotrexate in the central extracellular fluid compartment is in rapid equilibrium with the so-called 'shallow' peripheral compartment. This compartment has a plasma equivalent volume of about 14% of body weight. The compartment may represent intracellular methotrexate, which is loosely bound to intracellular proteins. Methotrexate in the central, extracellular fluid compartment is also in slower equilibrium with the so-called deep peripheral compartment. This compartment has a relatively small plasma equivalent volume and represents about 3% of body weight. Methotrexate that is tightly, but reversibly, bound to intracellular proteins may account for this compartment. A transport route leads from the central extracellular fluid compartment to a compartment where metabolites are formed (metabolite space), and from this space metabolites are returned to the extracellular fluid. The two compartments labeled "extracellular space" represent the same extracellular volume, but they are separated because one compartment contains methotrexate and the other metabolites. Although a radioimmunoassay for methotrexate (Diagnostic Biochemistry, San Diego, CA) is used to measure plasma concentrations, we assume that a certain fraction of these metabolites are measured as methotrexate. We therefore have a plasma sample space that sums the concentration of methotrexate in the extracellular fluid and a small fraction (< 10%) of metabolites in the extracellular space that are measured as methotrexate. The fraction is determined from observed data points and varies from patient to patient. Without this correction, data points after about 72 h will be underestimated by the model. There are other ways to fit the 'tail' of the curve, but we have chosen this particular method so we can include a metabolic pathway, since methotrexate is metabolized in humans [12, 15]. Metabolites are cleared from the extracellular space of the model by an unknown loss route.

The major loss route of methotrexate from the system occurs through urinary excretion by a concentration-dependent transport mechanism. We assume that methotrexate excretion is due to glomerular filtration and tubular secretion and that the tubular secretion mechanisms are saturable by the high concentrations of methotrexate obtainable in extracellular fluid, since this type of excretion was shown to occur in monkeys [5].

The rate of tubular secretion can be described by Michaelis-Menten kinetics. Currently we find values for the maximum velocity of secretion (V_{max}) and the Michaelis-Menten constant (K_m) by

curve-fitting techniques incorporated into the SAAM program by means of observed methotrexate plasma concentrations. The values for V_{max} and K_m therefore may not be directly related to the V_{max} and K_m that actually control renal secretion at the level of the kidney tubule. Once values for K_m and V_{max} are determined for an individual patient, these values are fixed and are used to predict or simulate concentrations for subsequent doses of methotrexate.

Renal dysfunction is simulated by multiplying the urinary excretion parameter by a fraction obtained from the ratio of observed creatinine clearance to expected normal creatinine clearance. We assume that the Michaelis-Menten constant remains unchanged with renal damage. This is compatible with the 'intact nephron' hypothesis [6].

We do not routinely collect urine for methotrexate assay. It should be noted, however, that urinary excretion as predicted by the model is comparable to that reported in the literature for patients with either normal or impaired renal function [26, 28].

Patient Population

Patients reported in this study had advanced cancer. All had been treated previously with radiation therapy, surgery, chemotherapy, or a combined-modality approach. All received methotrexate as the only antitumor drug during the study. Dose and infusion time are variable, since different protocols were used. Because the number of patients who have had multiple courses of methotrexate therapy with computer-assisted evaluation is small, our data are as yet too limited to correlate clinical toxicity and plasma disappearance curves. The usefulness of the kinetic model for predicting plasma concentrations will be shown by the following examples.

Results

Patient 1 had metastatic Ewing's sarcoma resistant to chemotherapy with dactinomycin and with cytoxan and vincristine. He received a course of therapy with methotrexate at 750 mg/m² given over 24 h, with leucovorin rescue starting at 30 h. Plasma samples were obtained and the model parameters that best fit these data were determined. Figure 2a shows the observed and calculated plasma concentrations. Parameter values are given in the legend to the figure. These parameters were fixed for subsequent simulations. Three weeks later this patient received 1000 mg/m² as a 24-h infusion and the observed and predicted values are shown in Fig. 2b. Because this patient had progression of his tumor, he was placed on a protocol that involved weekly 6-h infusions of methotrexate with rescue starting 2 h after the finish of the infusion. A family of computer-generated curves showing the disappearance of methotrexate from plasma as predicted by our model, taking into account the planned dosage increments, is shown in Fig. 3. The observed and predicted plasma concentrations for the patient's first 6-h infusion are shown in Fig. 4a. One week later he received his second infusion, with the observed and predicted values shown in Fig. 4b. Within 36 h from the start of the infusion, before any overt toxic manifestation, the wide deviation from expected values was noted, and we prescribed prolonged rescue with leucovorin. Even with increased doses of rescue factor the patient required hospitalization for fever with pancytopenia. Renal dysfunction appeared to play a major role in this patient's toxicity, since his plasma creatinine rose from a preinfusion value of 0.9 mg/dl to 2.85 mg/dl by 60 h after the infusion.

Patient 2, with metastatic squamous-cell carcinoma of the larynx, had a creatinine clearance prior to chemotherapy of 55 ml/min, or approximately one-half the normal clearance rate. The cause for his compromised

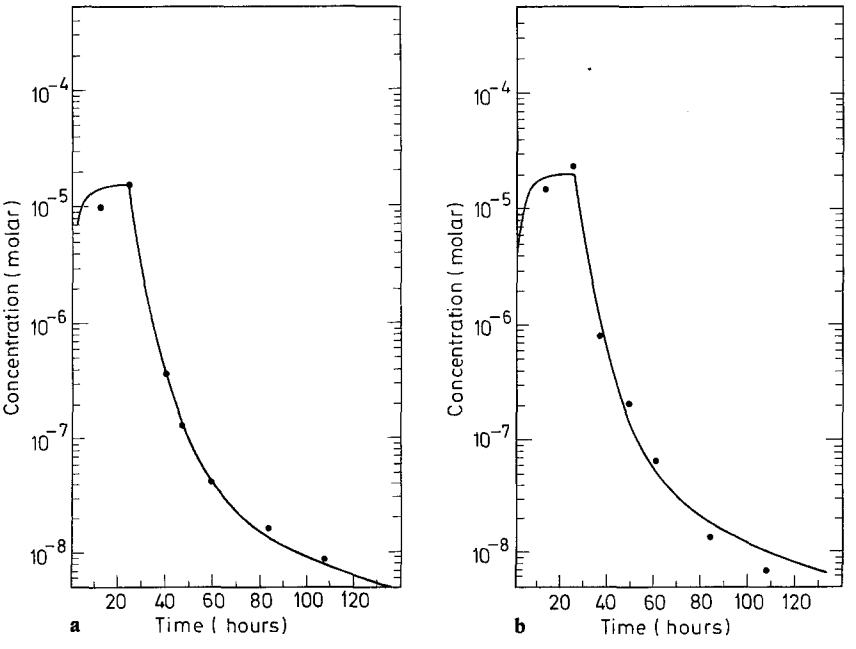

Fig. 2a and b. Observed (*circles*) and predicted (*line*) plasma methotrexate concentrations for Patient 1. $V_{max} = 3.0 \times 10^{-5}$ mol/liter/h; $K_m = 1.0 \times 10^{-4}$ *M*. Metabolite contribution to plasma concentrations = 4.5% (other parameters are as given in Fig. 1). **a** Infusion of 750 mg/m² over 24 h; **b** Infusion of 1000 mg/m² over 24 h

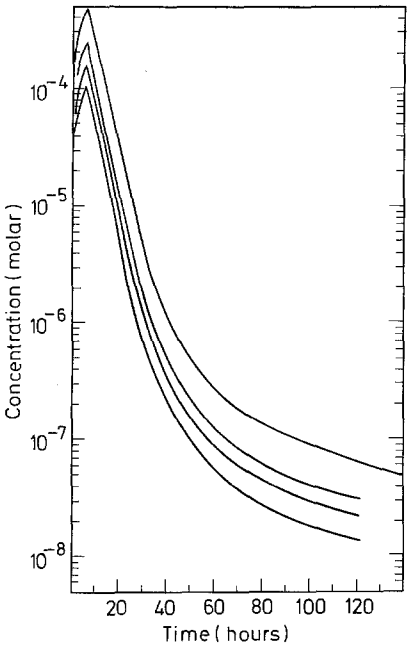

Fig. 3. Predicted plasma methotrexate concentrations for various dose levels from top: 6000; 3000, 2250, and 1500 mg/m^2) given in 6 h infusions. Model parameters are the same as given for Fig. 2

renal function was unknown. The patient received 500 mg methotrexate/m^2 over 24 h. Since a prior course of therapy had not been given, values for K_{max}, V_{max}, and metabolite contribution were not available, so the values derived for patient 1 were used. However, the transfer rate from the central compartment into urine was reduced to one-half. A reasonably close prediction of observed plasma concentrations was made, which is shown with observed data in Fig. 5.

Patient 3 had metastatic disease. She was treated with a methotrexate infusion (1.5 gm/m^2) over 24 h, with rescue starting at 30 h. When the values for the model parameters from patient 1 were used a poor fit was obtained. However, when the Michaelis-Menten constant was decreased by one-half and then fixed a reasonable fit could be obtained, as shown in Fig. 6a. The model then predicted plasma concentrations as the doses were escalated, as shown in Fig. 6b and c.

Discussion

Several approaches to mathematical modeling have been made to predict plasma concentrations for methotrexate [12, 14, 19, 27, 28, 35]. The simplest model assumes that the drug is distributed into two body compartments and that there is a route of elimination from the system [28]. This can be expressed as a mathematical equation, which is the sum of two exponentials with the form:

$$C = Ae^{-\alpha t} + Be^{-\beta t}$$

where C is the concentration at time t and A, B, α, and β are constants determined from observed plasma concentrations. A computer is used to find the best values for the constants. If a three-compartment, open model is used with elaborate statistical analysis, then the variance of an individually measured plasma methotrexate concentration from the mean expected value for the study population can be derived [14]. Toxicity can be predicted on the basis of this variance. Unfortunately, this

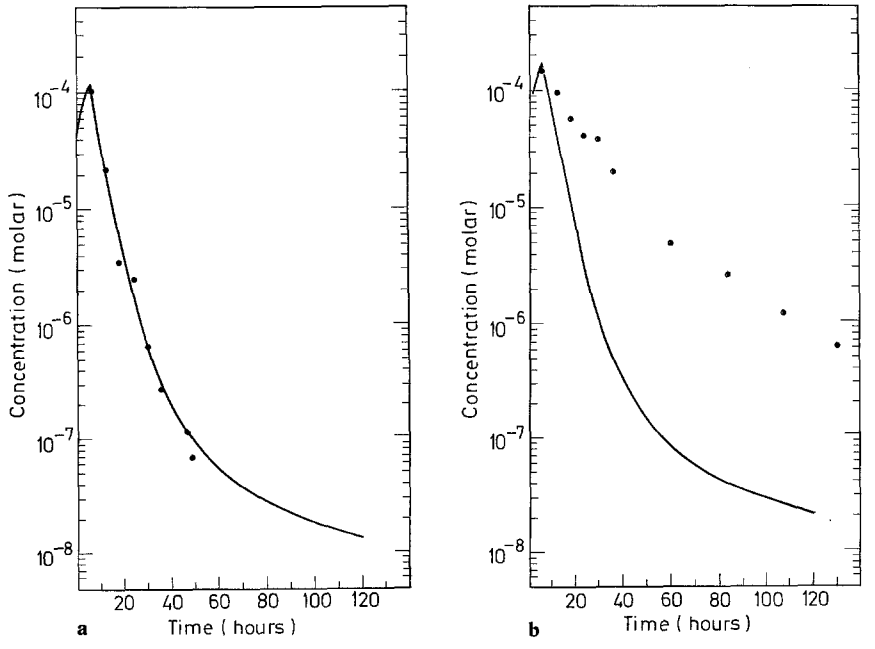

Fig. 4a and b. Observed (*circles*) and predicted (*line*) plasma methotrexate concentrations for Patient 1. Model parameters are the same as given for Fig. 2. **a** Infusion of 1500 mg/m^2 over 6 h. No toxicity; **b** Infusion of 2250 mg/m^2 over 6 h. Toxicity developed

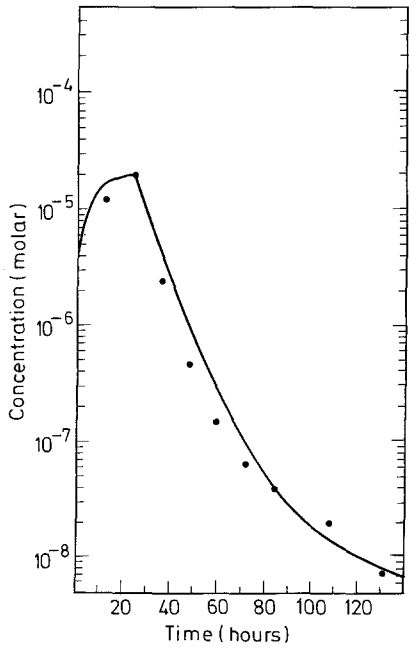

Fig. 5. Observed (*circles*) and predicted (*line*) plasma methotrexate concentrations for Patient 2. Model parameters are the same as given in Fig. 2 except that urinary excretion parameter (filtration + secretion) is multiplied by 0.5 to compensate for renal dysfunction

approach requires large numbers of plasma determinations to define the constants, the population mean, and the variance. If any other protocol is used, the constants may not be applicable.

Another type of pharmacokinetic model employs compartments with a priori physiologic and anatomic identification [35]. Volumes, blood flows, membrane permeabilities, and even enzyme kinetics are independently measured for tissues or organs, and these are incorporated into the model. This is a very sophisticated method, but the data necessary to adjust the parameters are often lacking for a species or an individual patient. Blood flow to the liver, for example, may be quite different for a patient with lung carcinoma and for one having a hepatoma. Since blood flow may not be a measurable entity for these patients, only educated guesses can be made as to how parameters should be adjusted.

A third type of mathematical model [19, 27] divides the body into compartments which behave similarly, and uses broad classifications such as 'central' or 'peripheral' or 'extracellular' compartments. This type uses an operational approach and employs the fewest compartments necessary to fit the experimental data. In most cases these data consist of plasma and urine con-

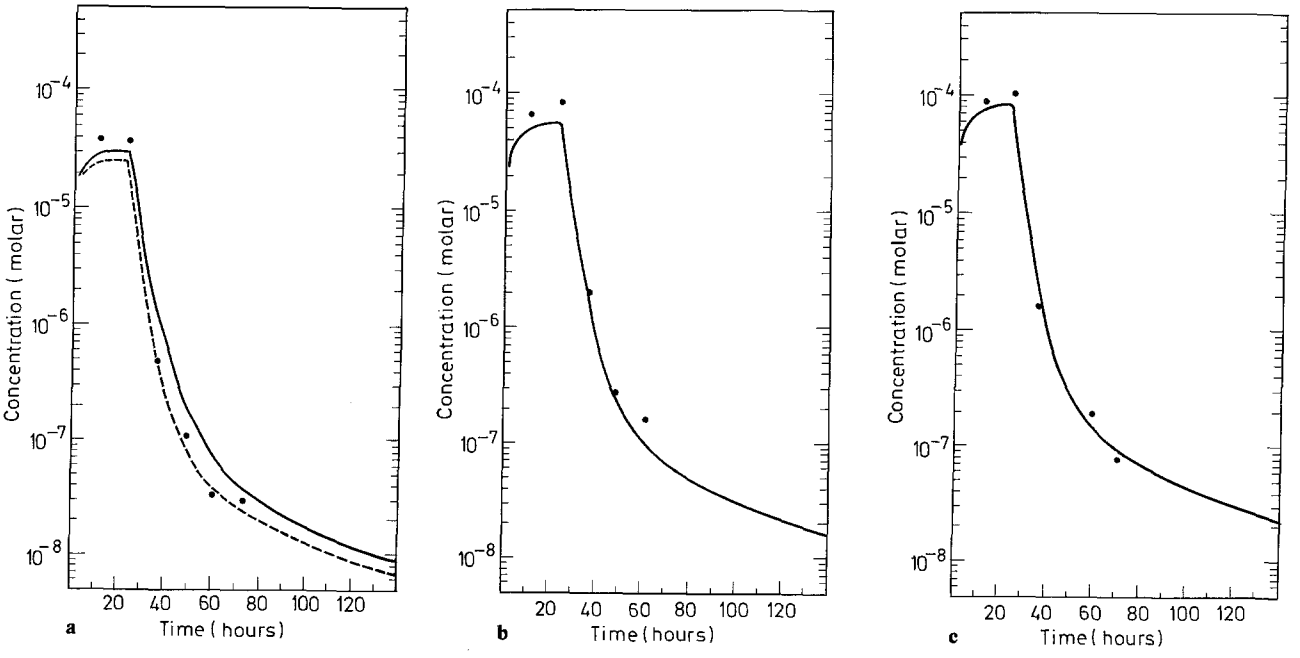

Fig. 6 a–c. Observed (*circles*) and predicted (*lines*) plasma methotrexate concentrations for Patient 3. Except for those noted below, all model parameters are the same as given in Fig. 2. **a** Infusion of 1500 mg/m^2 over 24 h: *solid line*, prediction with $K_m = 1.0 \times 10^{-4}$ *M*; *dashed line*, prediction with $K_m = 5.0 \times 10^{-5}$ *M*. **b** Infusion of 3000 mg/m^2 over 24 h: prediction with $K_m = 5.0 \times 10^{-5}$ *M*. **c** Infusion of 4000 mg/m^2 infusion over 24 h: prediction with $K_m = 5.0 \times 10^{-5}$ *M*

centrations. Certain constraints can be placed on the model based on assumptions about known aspects of the physiology of the system or pharmacokinetics of the drug. It may be possible to add physiologic significance to a compartment retrospectively after the size and the kinetics of a compartment have been determined experimentally. Parameters for this model can be adjusted with the aid of data observed in an individual patient. Provisions can be made for simulating different doses, schedules, and routes of drug administration.

Although these types of models do not account for all possible pharmacokinetic models, they are the ones currently used to describe methotrexate pharmacokinetics. They are similar in that they represent the solution of simultaneous differential equations. They differ in their complexity and in the number of assumptions one is willing to make about the absorption, distribution, biotransformation, and excretion of the drug.

We use the operational approach for our computer-solved mathematical model. We describe the plasma clearance of methotrexate as a function of dose, schedule of administration, and time with respect to an individual patient. Its major advantage is that minor adjustments in only a few parameters will allow simulation of most patients' plasma methotrexate concentrations. Although our interpretation of the physiological significance of these parameters or of other compartments may be inaccurate, we can at least test our interpretation experimentally. We can, for example, modify the urinary transport parameters as we gain information about the renal excretion of methotrexate from either animal or patient studies. In addition, as methotrexate metabolites are isolated and identified and their plasma and urine kinetics determined we can make improvements in the metabolic pathway of the model.

The model is helpful is allowing us to visualize the processes that determine the plasma kinetics of methotrexate. We are now in a position to unify the various clinical studies taking place. We can, for example, use the area under the predicted curve to determine drug exposure (concentration $\times$ time, or C $\times$ T) [17, 20]. We can determine the drug exposure for individuals and attempt to correlate this with toxicity or efficacy. If we assume that most damage to normal tissues takes place prior to administration of the rescue drug, we can determine drug exposure during the period without rescue. Perhaps the optimum schedule of methotrexate and rescue drug exposure can be determined once optimal drug exposure is determined.

Although drug exposure (C $\times$ T) may be an important determinant of toxicity, it is not the only factor to consider. Cytokinetics of normal and tumor tissue may play a large role in determining methotrexate toxicity and efficiency. However, computer-solved models of cytokinetics have also been developed [21, 32]. It may be possible to overlap both pharmacokinetic and cytokinetic models to help us understand how best to use methotrexate.

The theoretical implications of mathematical models may increase our understanding of methotrexate pharmacology. In addition, clinical pharmacokinetics can be of use at present. If we are able to predict plasma concentrations of methotrexate that are appropriate for a particular individual under a specific set of circumstances, such as dose of methotrexate, length of infusion, and state of renal function, then we can determine whether a plasma concentration observed for a patient is following the prediction. If it is not, we would be aware that some aspect of drug disposition has changed and we would be aware that some aspect of drug disposition has changed and we would be alert early in the course of treatment to the possibility of delayed toxicity. The cost of computer calculations is minimal compared with the large cost of the drug and the investment of time and effort in the treatment of adverse reactions. We hope that the model will improve patient therapy as well as help us to understand the pharmacology of this active antineoplastic agent.

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