Population Pharmacokinetics of High-dose Methotrexate After Intravenous Administration in Chinese Osteosarcoma Patients from a Single Institution

Wei Zhang¹, Qing Zhang², Xiaohuang Tian³, Haitao Zhao³, Wei Lu³, Jiancun Zhen¹, Xiaohui Niu²

¹Departments of Clinical Pharmacology, Beijing Jishuitan Hospital, Beijing 100035, China
²Orthopaedic Oncology, Beijing Jishuitan Hospital, Beijing 100035, China.
³Department of Pharmaceutical Science, University Health Science Center, Beijing 100191, China

Wei Zhang and Qing Zhang contributed equally to this research.

Abstract

Background: High-dose methotrexate (HD-MTX) with folinic acid (leucovorin) rescue is the gold standard therapy in the treatment of osteosarcoma. The plasma concentration of MTX is closely related to efficacy and toxicity. There are large individual differences. Many authors have described the pharmacokinetic (PK) profile of MTX regarding osteosarcoma under a variety of circumstances. However, no data concerning Chinese osteosarcoma patient PKs using the nonlinear mixed effects models (NONMEM) have been previously reported. The goals of this study were to establish the population pharmacokinetics (PPK) of HD-MTX treatment in Chinese osteosarcoma patients, and to explore the influence of patient covariates and between-occasion variability on drug disposition.

Methods: An intravenous HD-MTX solution (10 g/m²) was given 274 times to 148 osteosarcoma patients. MTX plasma concentrations were measured at 0, 6, 12, 24, 48 and 72 h after commencement of the infusion, and the fluorescence polarization immunoassay was used to determine MTX plasma concentrations. The PPK model and parameters were estimated using NONMEM software. The effects of fixed-effect factors were evaluated, and the final regression model was obtained.

Results: The following population parameters were obtained using a two-compartment model: CL₁ (clearance of central compartment): 

\[
(CL₁) = CL_{1\text{TV}} \times [1 - \theta_{CL₁-MTXNUM} \times MTXNUM] \times [1 - \theta_{CL₁-CrCl} \times (CrCl/1 - 1.89)] \times e^{\theta_{CL₁-CrCl} \times (CrCl/1 - 1.89)} \times (L/h). V₁ (central volume): 
\]

\[
(V₁) = V_{1\text{TV}} \times e^{\theta_{V₁-CrCl} \times (CrCl/1 - 1.89)} (L). CL₂ (clearance of peripheral compartment): 
\]

\[
(CL₂) = CL_{2\text{TV}} \times [1 - \theta_{CL₂-Bodyarea} \times (bodyarea - 1.62)] \times e^{\theta_{CL₂-Bodyarea} \times (bodyarea - 1.62)} \times e^{\theta_{V₂-Bodyarea} \times (Bodyarea - 1.62)} \times (L/h). V₂ (peripheral compartment): 
\]

\[
(V₂) = V_{2\text{TV}} \times [1 - \theta_{V₂-Bodyarea} \times (bodyarea - 1.62)] \times e^{\theta_{V₂-Bodyarea} \times (bodyarea - 1.62)} \times (L). 
\]

The PPK parameters (RSD%) were CL₁, V₁, CL₂ and V₂ with values of 6.20 L/h (8.48%), 19.6 L (extremely small), 0.0172 L/h (50.9%) and 0.515 L (39.1%), respectively. Creatinine clearance and the number of methotrexate chemotherapy cycles before MTX infusion had a significant effect on the CL₁, and body surface area had a significant effect on the CL₂ and the V₂ (P < 0.01).

Conclusions: A good fit was derived for the PPK. The model could be used to provide guidance for MTX treatment and reduce adverse effects.

Key words: Methotrexate; Osteosarcoma; Population Pharmacokinetics

Introduction

Osteosarcoma is the most common primary malignant bone tumor. Over the past decades, dramatic improvements have been made in the treatment of and the final clinical outcomes for this highly aggressive disease. The concept of administering chemotherapy before definitive surgery on the primary tumor was first introduced by Rosen et al.[¹,²] Since then, high-dose methotrexate (HD-MTX) with folinic acid (leucovorin) rescue is still the gold standard therapy in the treatment of osteosarcoma. Today, with the combination of preoperative or “neoadjuvant” chemotherapy including HD-MTX, cisplatin, ifosfamide or doxorubicin and radical surgery, disease-free survival rates of ≤70% can be achieved.[³]
There is a close relationship between MTX concentration, adverse reaction and treatment effect, with large individual differences. The incidence of toxicity has decreased as a result of plasma MTX concentration monitoring and appropriate leucovorin rescue, combined with adequate hydration and urine alkalinization. Effective concentration of MTX in the blood can significantly increase the clinical tumor necrosis rate.\(^{4,5}\) Recent studies have reported that a serum MTX concentration of 1000 \(\mu\)mol/L after a 4 h infusion and 700 \(\mu\)mol/L after a 6 h infusion can achieve good efficacy in the treatment of osteosarcoma. However, the higher the concentration of MTX is, the more side effects it will cause. The safe range for the area under the curve has been found to be between 4000 mM/h and 12,000 mM/h.\(^{6}\)

Therefore, it is important to obtain the individual pharmacokinetic (PK) parameters. The scattered serum drug concentration of MTX and the estimated population pharmacokinetics (PPK) parameters were analyzed in the present study using the nonlinear mixed effect model (NONMEM), the established PPK model employed for Chinese patients with osteosarcoma, and used to help obtain the final PPK model. We also examined the effects of some covariates (including height, body weight and other factors) on the PPK parameters and hence that our study could provide evidence for clinically personalized medication. In a further step, its implementation in the TDM software (Vital Scientific N.V.) that is most commonly used in the clinical setting might help optimize MTX dosing in patients with osteosarcoma and consequently, improve clinical outcome. Moreover, PPK models can also account for both the PK between-subject and between-occasion variability, allowing individual tailoring of the optimal dose.\(^{7,8}\)

The goals of the present study were: (1) To establish the PPK of HD-MTX treatment in Chinese osteosarcoma patients; and (2) to explore the influence of patient covariates and between-occasion variability on drug disposition.

## Methods

### Subjects

This study included 148 patients with osteosarcoma, who had received HD-MTX treatment for a total of 274 times (194 for males and 80 for females) from August 2009 to August 2010 in our department. Patients had the following characteristics: Age, 17.00 ± 7.06 years; height, 166.00 ± 12.44 cm; body weight, 58.00 ± 18.28 kg; and body surface area, 1.63 ± 0.27 m\(^2\).

### High-dose methotrexate administration

In the first phase (hyperhydration and urine alkalinization), a 500 ml 5% glucose injection (GDD) + a 500 ml 5% glucose and sodium chloride (GNS) injection + 10 ml of 15% KCL; (2) 200 ml 5% NaHCO\(_3\), was given at 12 h before MTX infusion.

The second phase (MTX infusion) consisted of seven groups of drugs that were consecutively administered: (1) 500 ml 5% GDD + 500 ml 5% GNS + 10 ml 15% KCL; (2) 100 ml 5% NaHCO\(_3\); (3) 2 mg vincristine + 10 ml 0.9% sodium chloride (NaCl); (4) 500 ml 5% GDD + 8–12 g/m\(^2\) MTX for 4–6 h in darkness; (5) 1000 ml 5% GDD; (6) 1000 ml 5% GNS + 10 ml 15% KCL; (7) 200 ml 5% NaHCO\(_3\); (8) 100 ml 0.9% NaCl + 5 mg Tropisetron.

In the third phase (hyperhydration and urine alkalinization) the following compounds were administered: (1) 100 ml 0.9% NaCl + 5 mg Tropisetron; (2) 1000 ml 5% GDD; (3) 1000 ml 5% GNS + 10 ml 15% KCL; and (4) 200 ml 5% NaHCO\(_3\).

During chemotherapy, 1.0 tid NaHCO\(_3\), and 200 mg tid allopurinol was taken orally daily. Urine volume was recorded over a 24 h period, and urinary pH between 7 and 9 was monitored.

### Cystic fibrosis rescue solution

At 6–8 h after the end of MTX infusion 12 mg q6h leucovorin cystic fibrosis (CF) rescue was given until the serum concentration of MTX was reduced to 0.05 \(\mu\)mol/L. If delayed excretion occurred, the dose of CF was increased according to the standard.

### Blood collection

Two millilitre of blood was drawn from the lateral vein at 0, 6, 12, 24, 48 and 72 h after MTX infusion. If the drug concentration in the blood was >0.05 \(\mu\)mol/L after 72 h, 2 ml of blood was continuously drawn every 24 h until the concentration was <0.05 \(\mu\)mol/L.

### Methotrexate assay

Blood samples were centrifuged at 3000 g for 5 min at room temperature in tubes without anticoagulant. MTX concentration was measured using fluorescence polarization immunoassay (TDX, ABBOTT T, USA) with a quantification limit of 0.01 \(\mu\)mol/L. The recovery rate of high-, medium- and low-quality control samples was 90–110%. The interday precision was <10%.

### Population pharmacokinetic analysis

Population pharmacokinetic analysis was performed using the nonlinear mixed effects model (NONMEM, Version V, Level 1.1: GloboMax, USA). A two-compartment model was established for data fitting. The nonlinear least squares principle was applied to find a group of PPK parameters to minimize the value of the objective function (OFV). The values of the OFV among the similar models approximately followed a \(\chi^2\) distribution: When \(df = 1\), \(\chi^{20.05} = 1.384, \chi^{20.01} = 10.83\). When the parameter numbers between two models differed by 1, if \(\Delta\text{OFV} > 3.84\), there was a significant difference (\(P < 0.05\)); if \(\Delta\text{OFV} > 10.83\), there was a highly significant difference (\(P < 0.001\)).

Statistical model: \(P_j = P_{ij} \cdot \text{Exp}(\eta_j)\)

where \(P_j\) is the \(i\)th individual’s \(j\)th PK parameters, \(P_{ij}\) is the typical group value of the \(j\)th PK parameters, and \(\eta_j\) represents
the inter-individual random error of the individual parameter $P_{ij}$ to the group parameter $P_{ij}^g$. The values of $\eta_i$ followed the normal distribution, zero in the center with a variance of $\omega^2$.

Residual random effect model: $C_{\text{obs}} = C_{\text{mod}} \cdot (1 + \epsilon_i) + \epsilon_2$, where and $C_{\text{obs}}$ and $C_{\text{mod}}$ are the actual observed value and the predicted value, respectively. $\epsilon_i$ and $\epsilon_2$ are inter-individual/inter-experiment random errors, which follow the normal distribution: Zero in the center with a variance of $\sigma_1^2$ and $\sigma_2^2$. $\epsilon_i$ represents the proportional error and $\epsilon_2$ represents the additive error.

Fixed effect model:

$$P_{ij} = P_{ij}^g \cdot [1 + \theta_{jk} \cdot (\text{COVR}_k - \overline{\text{COVR}_k})]$$

where $P_{ij}$ is the typical individual value of the $i$th individual’s $j$th PK parameter, $P_{ij}^g$ is the typical group value of the $j$th PK parameter, and $\text{COVR}_k$ and $\overline{\text{COVR}_k}$ are the $j$th individual’s $k$th covariate and its mean, respectively. $\theta_{jk}$ represents the degree of deviation of the individual parameter from its typical group value when the individual covariate differs from its mean by one unit.

**Variable selection**

The sample volume independent effect size (ES, statistical effect) was used as the test indicator for the biochemical index collected from the modeling process. An ES between 0.2 and 0.8 was considered as a medium effect, and a median of 0.5 was used here as the critical value. The physiological indexes before and after drug administration were considered as being changed when the ES was $>0.5$, and were then excluded as covariates.

$$\text{ES} = \frac{S_1 - S_2}{S_d}$$

Where $S_d$ is the combined standard deviation. The equation used to calculate the $S_d$ was $S_d = \frac{S_1^2 + S_2^2}{2}$, where the upper line represents the average value, and $S$ is the standard deviation.

**Correlation of covariates**

The extent of correlation among every element could be visualized using the diagram of covariants created using the R software version 2.12.0 for Windows (Mathsoft, Halethorpe, MD, USA).

**Full regression model**

The full regression model was established using the stepwise regression method. If the OFV decreased by $>3.84$, the covariate had a significant effect ($P < 0.05$) on this model, and this covariate should be retained in the model. All covariates with no significant effect were excluded. The full model was obtained after all covariates that had a significant effect were added in.

**Final model**

All covariates involved in the full regression model were tested using a more rigorous statistical standard (when $df = 1, \chi^2_{0.001} = 10.83$). If a change in the OFV was $>10.83$, the covariates were considered to have a significant effect and were retained in the model. Otherwise, the covariates were removed from the model. The final model was obtained through reverse elimination of the full regression model.

### Results

**Base model**

The drug serum concentration-time curve is shown in Figure 1. There were 148 patients with osteosarcoma, who received HD-MTX treatment a total of 274 times. The plot indicated a two-compartment model, and NONMEM, first-order conditional estimation was applied for fitting; the PK of the two-compartment model (OFV) was 487.856.

**Covariate selection and correlation of covariates**

A summary of the selected covariates is given in Table 1. They were: Number of chemotherapy (NUM) cycles; number of methotrexate chemotherapy (MTXNUM) cycles; Volume 1, the volume of infusion 1 day before drug administration (ml); pH 1, pH 1 day before drug administration; T1, body temperature 1 day before drug administration; U1, urine volume 1 day before drug administration (ml).

In addition, we explored the correlation between these covariates. We found that the covariates with a strong correlation included MTXNUM and NUM [Figure 2]. Additionally, the three covariates—weight, body mass index (BMI) and body area—were strongly correlated with each other. In the process of modeling, special attention needed to be given to the covariates with strong correlation, and the factors with a significant effect should be selected rather than non-significant ones to prevent the development of an over-complicated model.

**Final regression model equation and parameter values**

The final model of the MTX PPK established using the NONMEM was as follows:

![Plasma Concentration -Time Curve (semilog)](image)
Table 1: Covariates investigated during the modeling process

| Covariate [1*] | Median | Minimum | Maximum | n  |
|----------------|--------|---------|---------|----|
| Number         | 8      | 1       | 28      | 272|
| Gender         | NA     | NA      | NA      | 274|
| Age (years)    | 17     | 6       | 49      | 274|
| BMI            | 19.95  | 11.32   | 40.17   | 267|
| Weight (kg)    | 58     | 20      | 97      | 268|
| Volume, (ml)   | 1520   | 710     | 4240    | 259|
| MTXNUM         | 2      | 1       | 12      | 270|
| BODYAREA (m²)  | 1.63   | 0.62    | 2.21    | 270|
| CrCl; (ml/min) | 1.88   | 0.94    | 4.64    | 245|
| pH₁            | 8      | 5       | 9       | 258|
| RBC; (1 × 10⁹/L)| 4.06  | 2.28    | 5.7     | 264|
| HCT; (%)       | 36.5   | 20.4    | 48.9    | 262|
| Cr; (mg/dl)    | 49     | 19      | 81      | 250|
| AKP; (1/U/L)   | 94     | 43      | 479     | 238|
| T₁; (°C)       | 36.3   | 35.3    | 37.9    | 231|
| Total protein; (g/L) | 67.8  | 40.9    | 87.5    | 251|
| Albumin; (g/L) | 41.2   | 24.4    | 51.7    | 250|
| Na₂; (mmol/L)  | 142.5  | 132.9   | 151.1   | 237|
| Cl; (mmol/L)   | 103    | 85.5    | 140.6   | 237|
| U₁; (ml)       | 1820   | 420     | 5670    | 222|

Number: Number of chemotherapy cycles; Gender: Sex; BMI: Body mass index; Volume: The amount of fluid the day before administration; MTXNUM: Number of MTX chemotherapy cycles; BODYAREA: Body surface area; CrCl: Creatinine clearance before administration; PH₁: Urinary pH before dosing day; RBC: Erythrocyte count before administration; HCT: Hematocrit level before administration; Cr₁: Serum creatinine level before administration; AKP: Alkaline phosphatase before administration; T₁: Temperature before administration; Total protein: Total protein level before administration; Albumin: Albumin level before administration; Na₂: Serum sodium level; Cl: Serum chloride; U₁: Urine volume before administration; RBC: Red blood cell; NA: Not available.

\[(CL₁) = CL_{1,TF} × [1−θ_{c₁−MTXNUM} × MTXNUM] × [1−θ_{c₁−CII} × (CrCl₁−1.89)] × e^{\eta_{2,1} (L/h)}\]

\[(CL₂) = CL_{2,TF} × [1−θ_{CL2−BODYAREA}] × (BODYAREA−1.62) × e^{\eta_{2,2} (L/h)}\]

\[(V₁) = V_{1,TF} × e^{\eta_{1} (L)}\]

\[(V₂) = V_{2,TF} × [1−θ_{V₂−BODYAREA}] × (BODYAREA−1.62) × e^{\eta_{2} (L)}\]

where CL₁, V₁, CL₂ and V₂ are the individual central compartment clearance rate, the central compartment distribution volume, the peripheral compartment clearance rate and the peripheral compartment distribution volume in the population, respectively. \(\eta\) is the inter-individual variation among the various parameters. MTXNUM was the time of chemotherapy using MTX before chemotherapy, body area is the body surface area, and Cr is the serum creatinine clearance rate. The PKs parameters of the basic model and final model are listed in Table 2.

**Internal validation method**

This model was validated using the Bootstrap method. [10] Sampling was normally repeated 200–1000 times, building 200–1000 random sets of validation data. It was reported in the literature that the error would be <5% if the sampling frequency was >50. [11] In our study, sampling was carried out 1000 times for validation of the final distribution [Figure 3].

**Evaluation of the diagnosis from the base model and the final model**

The correlation between the observed value, the population predicted value and time

The goodness of fit between the predicted and observed values of serum drug concentration could be evaluated overall by plotting these two values against time. The half-logarithm coordinate diagram of the observed value, population predicted value, individual predicted value and time is shown in Figure 4. Figure 4a shows the basic model while Figure 4b shows the final model; the population predicted values from final model were closer to the observed values than was the case for the basic model, indicating that final model was an improvement on the basic model.

**The correlation between the population predicted value and the observed value**

We could observe the quality of the curve fitting approximately by plotting the predicted values (individual, group) as abscissa and observed values as vertical coordinates. The fitting quality was better when the data points were uniformly distributed on both sides of the line with a zero intercept and a slope of 1. The closer to the line the data points were, the better the precision of their fitting. The relationship between the population predicted value, the individual predicted value and the observed value are shown in Figure 5. Figure 5a shows the basic model and Figure 5b shows the final model; I was the population predicted value and 2 was individual predicted value. It was found that the individual predicted value was better fitted than the population predicted value in both the basic model and the final model. In addition, the final model proved to be better than the basic model.

**The correlation between weighted residual and population predicted value**

The variance in weighted residual (WRES) under different concentrations could be estimated by plotting the population predicted value as abscissa and the WRES as vertical coordinates. The WRES made it easier to compare the results from different models. With better fitting, data points should be randomly and uniformly distributed on both sides of the zero line (−4, 4). The correlation between the WRES and the population predicted value is shown in Figure 6, where Figure 6a shows the basic model and Figure 6b shows the final model. A portion of the WRESs was beyond the −4–4 range, with a few >20. When compared with the distribution of WRES in the basic model, the WRES values in the final model were more uniformly distributed, which was an improvement to a certain extent.
Figure 2: Correlation charts for some covariates, VOLUME, the amount of fluid the day before administration; NUM, number of chemotherapy cycles; Number of MTX chemotherapy cycles, number of MTX chemotherapy cycles; GEND, sex; BMI: Body mass index; BODYAREA, body surface area; pH1, urinary pH before the dosing day; Cr1, serum creatinine level before administration; CrCl1, creatinine clearance before administration; MTX: Methotrexate.

Table 2: Estimates of pharmacokinetic parameters regarding the base and final model

| PPK parameter | Basic model standard value | Final model standard value |
|---------------|-----------------------------|-----------------------------|
|               | Standard value | RSE % | Inter-individual RSD % | Standard value | RSE % | Inter-individual RSD % |
| CL1           | 5.81          | 2.07  | 8.93               | 6.20          | 4.87  | 8.48                |
| V1            | 19.2          | 2.34  | -                  | 19.6          | 4.39  | -                   |
| CL2           | 0.0154        | 5.61  | 55.0               | 0.0172        | 14.9  | 50.9                |
| V2            | 0.471         | 4.88  | 47.3               | 0.515         | 9.92  | 39.1                |
| θCL1-MTXNUM   |               |       |                    | 0.0183        | 35.6  |                    |
| θCL1-CrCl     |               |       |                    | 0.0416        | 32.2  |                    |
| θV2-BODYAREA  |               |       |                    | 0.8800        | 28.3  |                    |
| θV2-BODYAREA  | 0.874         | 21.2  |                    |               |       |                    |

CL1: Clearance of the central compartment; V1: The apparent distribution volume of the central compartment; CL2: Clearance of the peripheral compartment; V2: The apparent distribution volume of the peripheral compartment; θCL1-MTXNUM: The correction factor for MTXNUM regarding the parameter CL1; θCL1-CrCl: The correction factor of CrCl regarding the parameter CL1; θV2-BODYAREA: The correction factor for BODYAREA regarding the parameter V2; PPK: Population pharmacokinetics; RSE: Relative standard error; RSD: Relative standard deviation.
The correlation between residuals, weighted residual and time

Residuals (RES) and WRES were plotted against time to determine their change over time. In a better fitting model, all the data points should be uniformly distributed on both sides of line zero. The correlation between RES and time is shown in Figure 7, and between WRES and time in Figure 8, where A is the basic model and B is the final model in both figures.

**DISCUSSION**

The PPK model of high-dose MTX in osteosarcoma patients was established in the present study. In the final model, OFV was 373.294, a decrease of 114.562 when compared to the initial model, indicating a significant improvement. In addition, this model provided a very good fit for the predicted and observed concentrations with the exception of C0 [Figure 5]. The RSEs% of two variables were greater than 30%, but were still acceptable. In addition, the calculated

---

*Figure 3: Output of model evaluation running 1000 times: Theta Parameters Bootstrap Analysis Run mtx 1204.*

*Figure 4: Observed concentration (DV; semilog), individual predicted concentration (IPRED; semilog) and population predicted concentration (PRED; semilog) versus TIME in the base model (a) and the final model (b). The blue crosses represent the DV, the red blank circles represent the IPRED and the yellow triangles represent the PRED.*
results were verified by means of Bootstrap and thus were reliable with a small RSE%.

Population pharmacokinetics studies outside of China regarding MTX are currently focused on blood tumors; there have been two PPK studies on osteosarcoma. One involving adult patients, concluded that the three-compartment model provided a good fit. The parameter estimates for the final model were $CL_1 = 6.57 L/h$ and $V_1 = 42 L$. In our study, $CL_1 = 6.20 L/h$ and $V_1 = 19.6 L$. The difference in V1 between the two studies may be related to the age of the patients. The average age in the previous study was 26.7 years, while the average age in our study was 17 years. In another PPK study of MTX in osteosarcoma patients at average age of 15 years, the obtained parameter values were $CL_1 = 4.79 L/h$, $V_1 = 16.7 L$, $CL_2 = 0.019 L/h$ and $V_2 = 0.464 L$, close to those obtained in the present study. In another PPK study of MTX in osteosarcoma patients at average age of 15 years, the obtained parameter values were $CL_1 = 6.20 L/h$ and $V_1 = 19.6 L$. The difference in V1 between the two studies may be related to the age of the patients. The average age in the previous study was 26.7 years, while the average age in our study was 17 years. In another PPK study of MTX in osteosarcoma patients at average age of 15 years, the obtained parameter values were $CL_1 = 4.79 L/h$, $V_1 = 16.7 L$, $CL_2 = 0.019 L/h$ and $V_2 = 0.464 L$, close to those obtained in the present study. In another PPK study of MTX in osteosarcoma patients at average age of 15 years, the obtained parameter values were $CL_1 = 4.79 L/h$, $V_1 = 16.7 L$, $CL_2 = 0.019 L/h$ and $V_2 = 0.464 L$, close to those obtained in the present study. In another PPK study of MTX in osteosarcoma patients at average age of 15 years, the obtained parameter values were $CL_1 = 4.79 L/h$, $V_1 = 16.7 L$, $CL_2 = 0.019 L/h$ and $V_2 = 0.464 L$, close to those obtained in the present study.
Other covariates reported in literature include hydration\(^{16}\) and the pH of urine.\(^ {17}\) However, these two covariates had no significant effect in our model; the reason for urine pH having no significant effect was possibly the result of poor recording, because patients self-measured the pH using pH test paper, with a precision of only one digit and the frequent appearance of recordings of 7–8 or 6–9. Regarding hydration, different doses of drug were administered in the present study as compared with other previous studies. Moreover, it has been reported\(^ {7}\) that differences in serum drug concentration were not detectable at high infusion volume, which might have also happened in our study.

There were some patients who received HD-MTX multiple times, and we treated them as totally separate individuals in the modeling process to gather more data, but that also meant that we ignored the internal correlation within patients. This may partially explain the existing difference.

Through the analysis of the data from 274 cases with high-dose MTX chemotherapy, we considered that the PK characteristics followed the two-compartment model; the elimination of MTX was influenced by the timing of MTX chemotherapy, the clearance rate of creatinine and body surface area. The clearance rate of MTX decreased with increased times of MTX chemotherapy or a decreased creatinine clearance rate, while body surface area had a positive correlation with the peripheral clearance rate and the apparent volume of distribution of the peripheral compartment. The results indicate that it is important to strengthen the pharmaceutical care of patients receiving multiple MTX chemotherapy treatments, paying attention to the dosage calculations in amputee patients and obtaining renal creatinine clearance data prior to chemotherapy. The objective of this study was to improve the clinical effectiveness of chemotherapy and reduce the serious risks associated with this treatment. The findings provide an important theoretical basis for the proposed clinical MTX chemotherapy and the critical monitoring points for reducing the risks. An in-depth PPK study of individual doses should be undertaken in the future in a large osteosarcoma patient population involving HD-MTX to develop software for individualized dose calculation.

**References**

1. Rosen G, Marcove RC, Caparros B, Nirenberg A, Kosloff C, Huvos AG. Primary osteogenic sarcoma: The rationale for preoperative chemotherapy and delayed surgery. Cancer 1979;43:2163-77.

2. Winkler K, Beron G, Kotz R, Salzer-Kuntschik M, Beck I, Beck W, et al. Neoadjuvant chemotherapy for osteogenic sarcoma: Results of a Cooperative German/Austrian study. J Clin Oncol 1984;2:617-24.

3. Crews KR, Liu T, Rodriguez-Galindo C, Tan M, Meyer WH, Panetta JC, et al. High-dose methotrexate pharmacokinetics and outcome of children and young adults with osteosarcoma. Cancer 2004;100:1724-33.

4. Bacci G, Ferrari S, Delepine N, Bertoni F, Picci P, Mercuri M, et al. Predictive factors of histologic response to primary chemotherapy in osteosarcoma of the extremity: Study of 272 patients preoperatively treated with high-dose methotrexate, doxorubicin, and cisplatin. J Clin Oncol 1998;16:658-63.

5. Graf N, Winkler K, Betlemovic M, Fuchs N, Bode U. Methotrexate pharmacokinetics and prognosis in osteosarcoma. J Clin Oncol 1994;12:1443-51.

6. Comandone A, Passera R, Boglione A, Tagini V, Ferrari S, Cattel L. High dose methotrexate in adult patients with osteosarcoma: Clinical and pharmacokinetic results. Acta Oncol 2005;44:406-11.

7. Ette EI, Williams PJ, Lane JR. Population pharmacokinetics III: Design, analysis, and application of population pharmacokinetic Studies. Ann Pharmacother 2004;38:2136-44.

8. Karlsson MO, Sheiner LB. The importance of modeling interoccasion variability in population pharmacokinetic analyses. J Pharmacokinet Biopharm 1993;21:735-50.

9. Chen GX, Rong D. Estimating methods and applications of statistical effectiveness and effect. J Enterp Sci Technol Dev 2010;22:132-3.

10. Ding JJ, Jiao Z, Li ZD, Shi XQ, Zhong MK. Validation to multi-regression model of limited sampling strategy by Bootstrap method. J Chin J Health Stat 2004;5:289-92.

11. Ette EI. Stability and performance of a population pharmacokinetic model. J Clin Pharmacol 1997;37:486-95.

12. Dupuis C, Mercier C, Yang C, Monjanel-Mouterde S, Ciccolini J, Fanciullino R, et al. High-dose methotrexate in adults with osteosarcoma: A population pharmacokinetics study and validation of a new limited sampling strategy. Anticancer Drugs 2008;19:267-73.

13. Colom H, Farré R, Soy D, Peraire C, Cendros JM, Pardo N, et al. Population pharmacokinetics of high-dose methotrexate after intravenous administration in pediatric patients with osteosarcoma. Ther Drug Monit 2009;31:76-85.

14. Rousseau A, Sabot C, Delepine N, Delepine G, Debord J, Lachâtre G, et al. Bayesian estimation of methotrexate pharmacokinetic parameters and area under the curve in children and young adults with localised osteosarcoma. Clin Pharmacokinet 2002;41:1095-104.

15. Aquerreta I, Aldaz A, Martínez V, SierraSéguimaga L, Giráldez J. Predicción del retraso en la eliminación de metotrexato mediante métodos bayesianos. Farm Hosp 2002;26:90-5.

16. Li HY, Zheng Y, Yu JN, Zheng CJ. Relationship between difference volume hydration and serum concentration in chemotherapy of high-dose methotrexate. J Appl Clin Pediatr 2010;25:216-8.

17. Sand TE, Jacobsen S. Effect of urine pH and flow on renal clearance of methotrexate. Eur J Clin Pharmacol 1981;19:453-6.

**Received**: 17-10-2014 **Edited by**: Xiuyuan Hao

**How to cite this article**: Zhang W, Zhang Q, Tian X, Zhao H, Lu W, Zhen J, et al. Population Pharmacokinetics of High-dose Methotrexate After Intravenous Administration in Chinese Osteosarcoma Patients from a Single Institution. Chin Med J 2015;128:111-8.

**Source of Support**: Nil. **Conflict of Interest**: None declared.