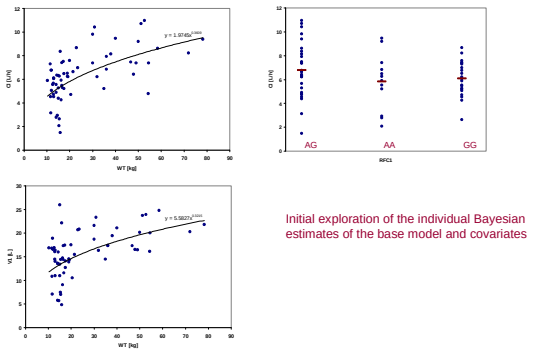
Population Pharmacokinetics of Methotrexate in Children with Lymphoid Malignancy

I. Grabnar[1], B. Faganel[2], V. Dolžan[3], A. Mrhar[1], J. Jazbec[2]

[1]Faculty of Pharmacy, University of Ljubljana
[2]Division of Oncology and Hematology, Department of Pediatrics, Medical Center Ljubljana
[3]Institute of Biochemistry, Faculty of Medicine, University of Ljubljana
Slovenia

Objectives

High dose methotrexate (MTX) regimen accompanied with folinic acid rescue is included in the treatment protocols of lymphoid malignancy (Burkitt's lymphoma, large cell lymphoma and acute lymphoblastic leukemia). Due to high inter and intra individual variability therapeutic drug monitoring of MTX is essential [1]. In children with acute lymphoblastic leukemia improvement of the treatment outcome by adjustment of the MTX dose to individual patient's clearance has been demonstrated [2]. Recently, genetic polymorphisms in the major MTX transporter - the reduced folate carrier gene 1 (RFC1) [3] and methylenetetrahydrofolate reductase (MTHFR) [4] have been described and their effects on MTX plasma levels have been shown. However, the results of these studies were not subjected to any formal pharmacokinetic analysis. Consequently, estimation of the clinical relevance of this polymorphism is difficult. Our study aims to assess population pharmacokinetics of MTX in children and to estimate the influence of RFC1 polymorphism G80A on systemic exposure.



Initial exploration of the individual Bayesian estimates of the base model and covariates

Final model

$CL\ [L/h] = 7.95 * (WT\ [kg]/25)^{0.308} * 0.919^{RFC1F}$

$V1\ [L] = 14.2 * WT\ [kg]/25$

$V2\ [L] = 3.81$

$Q\ [L/h] = 0.132$

RFC1F=0 in patients heterozygous for RFC1 (GA), or RFC1F=1 otherwise (GG and AA).

Patients and treatment

Pharmacokinetic analysis based on retrospectively collected clinical data from 62 patients, 6.6 +/- 5.0 years of age. All patients received 4 courses of MTX separated by a period of two weeks. MTX (2.0 - 5.7 g/m2) was infused over 24 h (10% of the dose in the first hour and the rest in the remaining 23 hours). Pre-hydration and urine alkalinization with sodium bicarbonate was performed. Folinic acid rescue was adjusted to MTX concentrations according to standard protocol guidelines. Venous blood samples were collected at 24, 36, 42, 48, 54, 60 and 66 hours following initiation of infusion. MTX plasma concentrations were determined by fluorescence polarisation immunoassay. A total of 881 MTX concentrations (5 - 24 per patient) were available for the analysis. 50

Pharmacokinetic analysis

Population analyses were performed using NONMEM (Version V, level 1.1, GloboMax LLC, Ellicott City, MD, USA) and Visual-NM (Version V, R.D.P.P., Montpellier, France), a Windows based interface to NONMEM. The structural model describing methotrexate pharmacokinetics was a two compartment model with first-order elimination as implemented in ADVAN3/TRANS4 PREDPP subroutine. The parameters estimated were clearance (CL), volume of distribution of the central (V1) and peripheral (V2) compartment and distribution clearance (Q). Additive, proportional, and exponential models were evaluated to describe interindividual variability of parameters (ω^2), while for residual variability of methotrexate concentration (σ^2) additive, proportional and combination error models were compared. Additionally, between-occasion variability (BOV) of clearance was estimated as there were samples from four different methotrexate courses available. First order conditional estimation (FOCE) method was used.

Modelling strategy aimed at a model with minimal structural and variability parameters needed to adequately describe the data. The model adequacy was evaluated by standard diagnostic plots. Additional criteria were convergence of minimization, number of significant digits more than 3, successful covariance step and gradients in the final iteration in the range between 10^{-3} and 10^{2}. Alternative models were compared by the likelihood ratio test. Criterion for selection of a model was a change in minimum value of objective function (ΔOBJ) of at least 3.84 per one additional parameter, corresponding to $p < 0.05$.

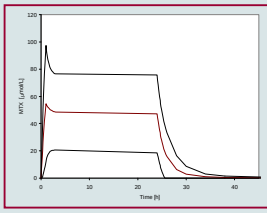
In the first step base model was derived, while in the second step covariates were included into the base model one at a time and their effect was evaluated using the likelihood ratio test. Other diagnostic criteria for inclusion of covariate were reduction of interindividual variability, closer association between the observed and model predicted methotrexate concentrations and that the 95% confidence interval for the structural parameters did not include zero. Covariates investigated were patient weight (WT), body surface area (BSA), age and RFC1 polymorphism. Significant covariates according to the likelihood ratio test were rank-ordered and introduced into the full model. The final model was determined by backward elimination of covariates one by one from the full model to see if they should remain using a likelihood ratio test.

Results

Measured methotrexate concentrations were best described with a two-compartment model with exponential error model for interindividual variability and combination model, including proportional and additive components for residual variability. Scatter plots obtained with the base model suggested a strong association between CL and V1 and covariates related to body size: WT, BSA and age, and weak effect of RFC1 polymorphism on CL. Contrary to the results of Laverdiere, et al [3] who found that MTX plasma levels were higher in the AA group than in AG and GG groups of patients, in the present study MTX clearance was higher in patients heterozygous for G80A compared to patients homozygous for G80A for both alleles. As all covariates associated with body size strongly correlated, only the effect of WT was considered for inclusion into the full model. From the plot of CL and V1 versus WT nonlinear relationship was observed and consequently power model was used to evaluate this effect. However, only the exponent for the effect of WT on CL was retained in the backward elimination step. In patients, heterozygous for G80A clearance was 8% higher (95%CI: 5-12%) compared to patients homozygous for G80A for both alleles. Although minor, the effect of RFC1 polymorphism was marginally significant ($p<0.05$) and was included in the final model because the 95% confidence interval excluded the value of 1.

Parameters of the final model

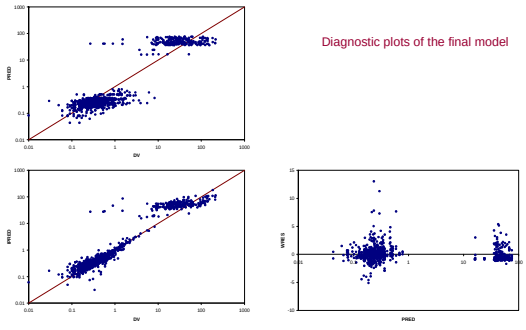
Parameter	Mean [95% CI]	% IIV (SE)
Coefficient of CL (L/h)	7.95 [6.51, 9.39]	31.9 (22.3)
Exponent for WT on CL	0.308 [0.217, 0.399]	
RFC1 on CL	0.919 [0.883, 0.955]	
V_1 (L/25 kg)	14.2 [10.1, 18.3]	49.2 (32.9)
V_2 (L)	3.81 [2.91, 4.71]	45.9 (39.1)
Q (L/h)	0.132 [0.104, 0.160]	64.2 (46.4)
Between-occasion variability		
κ_{CL} (L/h)		0.97 (0.48)
Residual variability		
σ_1 (%CV)		67.9 (34.5)
σ_2 (μmol/L)		0.065 (0.048)



Expected time course of MTX concentration (mean, 95% CI) in an average child (25 kg, heterozygous for RFC1) following a dose of 5 g/m^2

Conclusions

Developed population pharmacokinetic model is suitable for individualization of MTX dosing in children. Significant effect of RFC1 polymorphism on MTX clearance was observed, however this effect is hard to explain. Additional study including P-glycoprotein and methylenetetrahydrofolate reductase genotype is underway.



Diagnostic plots of the final model

References

[1] Aumente D, Buelga DS, Lukas JC, Gomez P, Torres A, Garcia MJ. Population pharmacokinetics of high-dose methotrexate in children with acute lymphoblastic leukaemia. Clin Pharmacokinet. 2006;45(12):1227-38.

[2] Evans WE, Relling MV, Rodman JH, Crom WR, Boyett JM, Pui CH. Conventional compared with individualized chemotherapy for childhood acute lymphoblastic leukemia. N Engl J Med. 1998 Feb 19;338(8):499-505.

[3] Laverdiere C, Chiasson S, Costea I, Moghrabi A, Krajinovic M. Polymorphism G80A in the reduced folate carrier gene and its relationship to methotrexate plasma levels and outcome of childhood acute lymphoblastic leukemia. Blood. 2002 Nov 15;100(10):3832-4.

[4] Imanishi H, Okamura N, Yagi M, Noro Y, Moriya Y, Nakamura T, Hayakawa A, Takeshima Y, Sakaeda T, Matsuo M, Okumura K. Genetic polymorphisms associated with adverse events and elimination of methotrexate in childhood acute lymphoblastic leukemia and malignant lymphoma. J Hum Genet. 2007;52(2):166-71.

iztok.grabnar@ffa.uni-lj.si