Bioavailability of Higher Dose Methotrexate Comparing Oral and Subcutaneous Administration in Patients with Rheumatoid Arthritis

MONIQUE HOEKSTRA, CEES HAAGSMA, CEES NEEF, JOHANNES PROOST, ANTONIUS KNUIF, and MART van de LAAR

ABSTRACT. ***Objective.*** To determine the bioavailability of higher oral doses of methotrexate (MTX) in adult patients with rheumatoid arthritis (RA).

Methods. A pharmacokinetic analysis was performed in 15 patients with RA taking a stable dose of MTX (≥ 25 mg weekly). Separated by 2 weeks, a pharmacokinetic analysis was performed in each patient after oral and subcutaneous administration of the same dose of MTX. MTX serum concentrations were measured by a fluorescence polarization immunoassay. Pharmacokinetic analysis was performed with an iterative 2-stage Bayesian population procedure, obtaining population and individual pharmacokinetic parameters.

Results. The median MTX dose was 30 mg weekly (range 25–40 mg). A 2-compartment model best described the serum MTX concentration versus time curves. The mean bioavailability after oral MTX was 0.64 (range 0.21–0.96) compared to subcutaneous administration. There was a statistically significant difference in the bioavailability of the 2 administration regimens.

Conclusion. Bioavailability of a higher oral dose of MTX in adult patients with RA is highly variable, and on average two-thirds that of the subcutaneous administration. To improve efficacy of MTX at dosages of 25 mg weekly or more, a change to parenteral administration should be considered. (J Rheumatol 2004;31:645–8)

Key Indexing Terms:
METHOTREXATE PHARMACOKINETICS RHEUMATOID ARTHRITIS

Methotrexate (MTX) is commonly used in weekly single-dose regimens in the treatment of rheumatoid arthritis (RA). A dose-effect relation was established for doses of 7.5–25 mg per week[1,2]. In clinical trials in RA, the MTX dose is increased up to 25 mg weekly, until efficacy is reached. It is not clear whether even higher oral doses of MTX are more effective. Efficacy of high intravenous doses of MTX (40–500 mg/m^2), in patients with refractory RA, was described in several studies[3-5].

The bioavailability of oral MTX could be a limiting factor for its efficacy. Oral MTX is absorbed in the proximal intestine by a specific transport mechanism, and a relation between dose and absorption of oral MTX was observed in 2 clinical studies[6,7]. Pharmacokinetic studies in adult patients with RA show comparable bioavailability of oral and parenteral MTX in doses up to 25 mg weekly[8-11]. In these studies the mean relative bioavailability of oral MTX, compared to intramuscular administration, ranged from 0.85 to 1.0. In other studies, using 15 mg MTX and 10 mg/m^2 MTX, bioavailability of oral compared to intravenous MTX was 0.67 and 0.70, respectively[12,13]. In a comparison of 25 mg MTX, the mean bioavailability after oral administration was 73% compared to the intravenous route[14]. Despite the impression given by a few studies[8,15], it is not certain that the bioavailability of intravenous, intramuscular, and subcutaneous MTX is strictly comparable.

Pharmacokinetic studies in patients with malignant diseases have shown that the absorption of higher doses of MTX (≥ 25 mg weekly) is incomplete[6,16-19]. The relative bioavailability of 40 mg/m^2 oral MTX in a study in children with acute lymphoblastic leukemia was 42%; in adult patients with solid tumors using 15 mg/m^2 MTX this was 57%[18,17]. Another study in 15 children[19] showed a decreased absorption of oral MTX at doses > 12 mg/m^2. However, the results of pharmacokinetic studies in disorders other than RA, and even more so in children, cannot be extrapolated to adult patients with RA.

Although a clear relation between pharmacokinetic parameters and efficacy has not been demonstrated in RA, it seems likely that improvement of the bioavailability of MTX will lead to better efficacy, given the dose-effect rela-

From the Department of Rheumatology, and the Department of Clinical Pharmacology, Medisch Spectrum Twente, Enschede; and the Department of Pharmacokinetics and Drug Delivery, University Centre for Pharmacy, Groningen, The Netherlands.

M. Hoekstra, MD, Rheumatologist; C.J. Haagsma, MD, Rheumatologist; M.A.F.J. van de Laar, MD, Rheumatologist, Department of Rheumatology, Medisch Spectrum Twente; C. Neef, PhD, Hospital Pharmacist; A.J.H. Knuif, Pharmaceutical Analyst, Department of Clinical Pharmacology, Medisch Spectrum Twente; J.H. Proost, PhD, Pharmacist, Department of Pharmacokinetics and Drug Delivery, University Centre for Pharmacy.

Address reprint requests to Dr. M. Hoekstra, Department of Rheumatology, Medisch Spectrum Twente, PO Box 50000, 7500 KA Enschede, The Netherlands. E-mail: m.hoekstra@ziekenhuis-mst.nl

Submitted May 22, 2003; revision accepted October 27, 2003.

tion[1,2]. This idea is supported by a study in patients with psoriasis in which a relation was found between the area under the curve of the time versus MTX concentration and a decrease in the Psoriasis Activity and Severity Index (PASI)[20].

The bioavailability of higher MTX doses can be improved by parenteral administration. To study this option, we performed a crossover pharmacokinetic study in adult patients with RA, comparing the bioavailability of oral and subcutaneous MTX at doses ≥ 25 mg weekly.

MATERIALS AND METHODS

Patients and MTX administration. Patients with RA, who were treated with MTX in a stable (≥ 3 months) dose of ≥ 25 mg weekly, oral or parenteral, were studied. Consecutive outpatients fulfilling these inclusion criteria were invited to participate. The local ethics committee approved the study and written informed consent was obtained from each patient.

Baseline data were gathered on diagnosis, age, sex, disease duration, dose, serum creatinine, folic acid supplementation, and use of disease modifying antirheumatic drugs (DMARD), nonsteroidal antiinflammatory drugs (NSAID), and prednisolone. Pharmacokinetics were studied twice in each patient with a 2-week interval: once with their regular MTX dose by oral route of administration, and once with the same dose of MTX by subcutaneous administration in random order. Folic acid supplementation was allowed, but not on the day of MTX intake. Leukopenia, thrombocytopenia, and transaminase elevations were reasons for exclusion.

Patients were admitted in the hospital in the morning. They were allowed to have breakfast at home, at least 1.5 hour before MTX intake. Comedication was continued during both sampling episodes. Other DMARD and prednisone were allowed, with stable doses throughout the study. The concurrent medication was taken at least 1.5 hour before and more than 2 hours after MTX intake. Oral MTX was administered with water. MTX was injected subcutaneously in the upper leg in all patients by the examiner. Blood samples were drawn from an indwelling catheter at Time 0 (preadministration) and at 0.25, 0.5, 0.75, 1.0, 1.25, 1.5, 2, 4, 6, 8, 12, 24, and 48 h after administration of MTX. The blood samples were centrifuged and the serum stored at –20°C until analysis.

MTX assay. MTX serum concentrations were determined using a fluorescence polarization immunoassay technique (MTXII: list no. 7A12, TDX-Abbott Diagnostics, North Chicago, IL, USA)[21]. The lower detection limit was 10 µg/l. At 10 µg/l the coefficient of variation of the test is 15%. The standard deviation (SD) of the test is described by the formula: SD = 4.76 + 0.05*C, where C = concentration.

Pharmacokinetic analysis. The MTX concentration data of both administrations from all patients were analyzed simultaneously by an iterative 2-stage Bayesian analysis using the program MW\Pharm, version 3.54[22,23]. The pharmacokinetic model was a one-compartment (parameters k_e, V_1) or a 2-compartment model (parameters k_e, V_1, k_{12}, k_{21}), with first-order absorption with a lag-time for oral and subcutaneous administration, with parameters F (bioavailability), k_a (absorption rate constant), and T_{lag} (lag-time) for each route of administration. Since absolute bioavailability cannot be assessed without an intravenous reference administration, the analysis was performed assuming that bioavailability of the subcutaneous administration was 100%. Measurement data were weighted according to the reciprocal of their variance ($1/SD^2$). A log-normal distribution for the pharmacokinetic population parameters was assumed. Goodness-of-fit was evaluated from visual inspection of the measured and calculated data points. The choice between a one- and 2-compartment model was based on Akaike's Information Criterion (AIC)[24].

MTX clearance (CL), volume of distribution (V), elimination half-life ($t_{1/2}$), and for each route of administration the area under the concentration-time profile (AUC), time to maximum concentration (T_{max}), and maximum concentration (C_{max}) were calculated from the model parameters for each patient.

Statistical analysis. To compare the values of the pharmacokinetic parameters of the oral and subcutaneous route of administration, a signed-rank test was employed. A 2-sided p value < 0.05 was considered significant.

RESULTS

Fifteen patients with RA were studied. Patient characteristics are presented in Table 1. All patients received folic acid supplementation in varying doses (5–25 mg weekly), but not on the day of MTX intake. Three patients concurrently used hydroxychloroquine, one chloroquine, one sulfasalazine, and one aurothiomalate. Low dose prednisolone (≤ 10 mg daily) was used by 8 patients, and NSAID by 11 patients.

A 2-compartment model fitted significantly better to the data than a one-compartment model (AIC value –250 and –956 for the one- and 2-compartment model, respectively). The mean bioavailability (F) was 0.64, with a rather large range from 0.21 to 0.96. The pharmacokinetic parameters with paired statistical analysis are shown in Table 2. The AUC of oral MTX was significantly lower than the AUC of the subcutaneous route of administration ($p < 0.001$). The fitted mean time-concentration curves of oral and subcutaneous administration are presented in Figure 1.

DISCUSSION

The bioavailability of oral MTX (≥ 25 mg weekly) was highly variable, and was significantly less compared to subcutaneously administered MTX in patients with RA. It varied between 0.21 and 0.96, with a mean of 0.64.

In the design of our study comedication was continued, and patients were allowed to have breakfast at home before coming to the hospital. Because of the time between comedication, breakfast, and MTX administration, an effect on MTX absorption is unlikely. Further, the effect of food has been extensively studied and no effect on MTX absorption was found[9,13].

The majority of pharmacokinetic studies in adult patients with RA have used low doses of MTX. In studies using MTX doses of 7.5 to 20 mg weekly, bioavailability after oral compared to parenteral administration ranged from 0.67 to 1.0[8-13]. Only one study compared 25 mg oral and intravenous MTX, in 18 patients with rheumatic diseases[14]. The bioavailability of oral MTX was 0.73, somewhat higher than what we found, but in our study most patients used higher

Table 1. Patient characteristics (n = 15: 11 women, 4 men).

	Median	Range
Age, yrs	61	31–72
Disease duration, yrs	7	2–32
Weight, kg	76	63–110
Creatinine clearance, ml/min	80	57–124
Dose, mg weekly	30	25–40
Dose, mg/kg	0.40	0.27–0.57

Table 2. Pharmacokinetic parameters of oral and subcutaneous route of administration (n = 15). Signed-rank test, p value < 0.05 is significant.

	AUC	Lag-time	ka	T_{max}	C_{max}	V1	V	k12	k21	ke	CL	t½ el
Oral	2466	0.36	0.87	1.2	594	9.6	34.5	0.81	0.55	0.88	8.4	2.9
	(785)	(0.18)	(0.29)	(0.3)	(208)	(2.0)	(8.1)	(0.31)	(0.04)	(0.11)	(2.2)	(0.5)
Subcutaneous	3786	0.06	0.36	1.7	519							
	(873)	(0.05)	(0.10)	(0.3)	(142)							
p	0.001	< 0.001	< 0.001	0.001	0.30							

Values are mean (standard deviation). AUC: area under curve (0–48 hours) in h·µg/l; lag-time in hours; ka: absorption rate constant; T_{max}: time to maximum concentration (hours); C_{max}: maximum concentration (µg/l); V1: volume of distribution of first compartment; V: volume of distribution (liter); k12: rate constant of transport between compartment 1 and 2; k21: rate constant of transport between compartment 2 and 1; ke: elimination rate constant; CL: total body clearance (liter/hour); t½ el: half-life of elimination (hours).

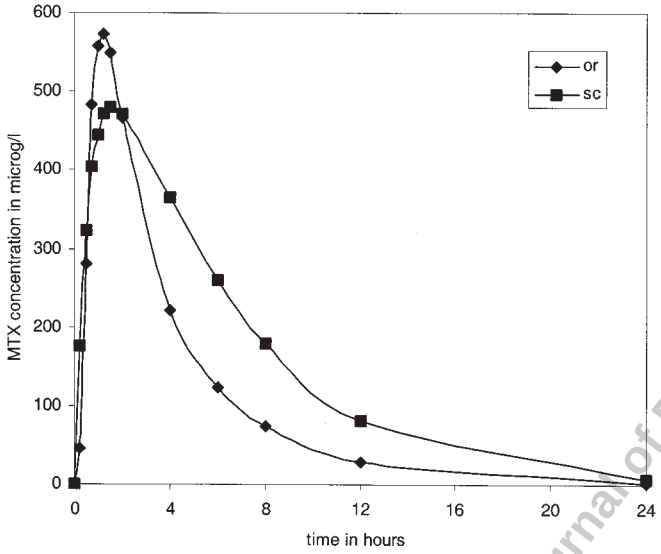

Figure 1. Plasma concentration-time curves of oral (or) and subcutaneous (sc) methotrexate. Values are means.

doses than 25 mg.

When we compare our data to other studies using higher dose MTX (> 25 mg), only pharmacokinetic studies in patients with malignancies are available. In these studies wide variability in MTX absorption was observed, and therefore split-dose regimens have been tried to improve bioavailability[25]. A comparable investigation is the study by Freeman-Narrod, *et al*. Doses of 15 mg/m^2 (25–35 mg) were used in adults with solid tumors. Eighteen patients received this dose by both oral and intramuscular administration. The mean cumulative AUC up to 24 h was higher with the intramuscular route, and the mean oral bioavailability was 0.57[17].

We analyzed the data assuming first-order absorption after a lag-time, which may be a simplification of the true absorption kinetics. In general a difference in AUC between oral and subcutaneous administration of medication could be due to either an absorption limitation or a first-pass effect. Decreasing bioavailability with an increasing dose favors an absorption limitation. The number of patients in our study with different MTX doses was too small to draw conclusions about a dose-bioavailability relation. However, there is a positive relation between the subcutaneous AUC and the dose of MTX (linear regression; $R^2 = 0.33$, $p = 0.03$), whereas the oral AUC does not increase with an increasing dose. Hamilton, *et al* studied 21 RA patients on more occasions. They found a decreasing bioavailability with an increasing oral dose, mean maximum dose being 17 mg/week[7]. These results support the idea of an absorption limitation of oral MTX with an increasing dose. The finding of higher bioavailability of oral split high dose MTX, compared to a single dose, in patients with solid tumors[25] supports a reduced bioavailability due to an absorption limitation. However, to pursue this question for the MTX doses we use in RA, an additional study is needed that directly compares a single-dose with a split-dose regimen.

Although controlled trials studying the effect of higher doses of MTX are lacking, higher dosing of MTX may be clinically useful. A dose escalation study in 54 patients with RA concluded that increasing the intramuscular MTX dose from 15 to 45 mg weekly did not result in improved disease control[26]. However, the number of patients was small, and detailed data about baseline disease activity scores were not provided. In our opinion, additional controlled trials are needed to evaluate the effect of higher doses of MTX, which are in fact widely used in rheumatology practice. In our observational study of MTX use in 1022 RA patients, we found that 12% of the patients reached a maximum dose of ≥ 25 mg weekly (maximum 40 mg weekly)[27].

Our data suggest that doses between 25 and 40 mg MTX per week, administered orally, result in limited bioavailability. Bioavailability is enhanced by the subcutaneous route of administration, and this may increase efficacy.

REFERENCES

1. Furst DE, Koehnke R, Burmeister LF, Kohler J, Cargill I. Increasing methotrexate effect with increasing dose in the treatment of resistant rheumatoid arthritis. J Rheumatol 1989;16:313-20.
2. Seideman P. Methotrexate — the relationship between dose and

clinical effect. Br J Rheumatol 1993;32:751-3.
3. Gabriel S, Creagan E, O'Fallon WM, Jaquith J, Bunch T. Treatment of rheumatoid arthritis with higher dose intravenous methotrexate. J Rheumatol 1990;17:460-5.
4. Shiroky J, Allegra C, Inghirami G, Chabner B, Yarboro C, Klippel JH. High dose intravenous methotrexate with leucovorin rescue in rheumatoid arthritis. J Rheumatol 1988;15:251-5.
5. Shiroky JB, Neville C, Skelton JD. High dose intravenous methotrexate for refractory rheumatoid arthritis. J Rheumatol 1992;19:247-51.
6. Henderson ES, Adamson RH, Oliverio VT. The metabolic fate of tritiated methotrexate. II. Absorption and excretion in man. Cancer Res 1965;25:1018-24.
7. Hamilton RA, Kremer JM. Why intramuscular methotrexate may be more efficacious than oral dosing in patients with rheumatoid arthritis. Br J Rheumatol 1997;36:86-90.
8. Seideman P, Beck O, Eksborg S, Wennberg M. The pharmacokinetics of methotrexate and its 7-hydroxy metabolite in patients with rheumatoid arthritis. Br J Clin Pharmacol 1993;35:409-12.
9. Hamilton RA, Kremer JM. The effects of food on methotrexate absorption. J Rheumatol 1995;22:630-2.
10. Godfrey C, Sweeney K, Miller K, Hamilton R, Kremer J. The population pharmacokinetics of long-term methotrexate in rheumatoid arthritis. Br J Clin Pharmacol 1998;46:369-76.
11. Jundt JW, Browne BA, Fiocco GP, Steele D, Mock D. A comparison of low dose methotrexate bioavailability: oral solution, oral tablet, subcutaneous and intramuscular dosing. J Rheumatol 1993;20:1845-9.
12. Herman RA, Veng-Pedersen P, Hoffman J, Koehnke R, Furst DE. Pharmacokinetics of low dose methotrexate in rheumatoid arthritis patients. J Pharmacol Sci 1989;78:165-71.
13. Oguey D, Kölliker F, Gerber NJ, Reichen J. Effect of food on the bioavailability of low-dose methotrexate in patients with rheumatoid arthritis. Arthritis Rheum 1992;35:611-4.
14. Korber H, Iven H, Gross WL. Bioavailability and pharmacokinetics of methotrexate (MTX) and its metabolite 7-hydroxy-MTX (7-OH-MTX) after low-dose MTX (25 mg) in patients with chronic rheumatic diseases [abstract]. Arthritis Rheum 1992;35 Suppl:S142.
15. Brooks PJ, Spruill WJ, Parish RC, Birchmore DA. Pharmacokinetics of methotrexate administered by intramuscular and subcutaneous injections in patients with rheumatoid arthritis. Arthritis Rheum 1990;33:91-4.
16. Ballis FM, Savitch JL, Bleyer WA. Pharmacokinetics of oral methotrexate in children. Cancer Res 1983;43:2342-5.
17. Freeman-Narrod M, Gerstley BJ, Engstrom PF, Bornstein RS. Comparison of serum concentrations of methotrexate after various routes of administration. Cancer 1975;36:1619-24.
18. Teresi ME, Crom WR, Choi KE, Mirro J, Evans WE. Methotrexate bioavailability after oral and intramuscular administration in children. J Pediatr 1987;110:788-92.
19. Balis FM, Mirro J, Reaman GH, et al. Pharmacokinetics of subcutaneous methotrexate. J Clin Oncol 1988;6:1882-6.
20. Chladek J, Grim J, Martinkova, et al. Pharmacokinetics and pharmacodynamics of low-dose methotrexate in the treatment of psoriasis. Br J Clin Pharmacol 2002;57:147-56.
21. Abbott Laboratories Diagnostics Division. TDX system assays manual. North Chicago, IL: Abbott Laboratories; 1984.
22. Steimer JL, Mallet A, Golmard JL, Boisvieux JF. Alternative approaches to estimation of population pharmacokinetic parameters: comparison with the nonlinear mixed-effect model. Drug Metab Rev 1984;15:265-92.
23. Proost JH, Meijer DKF. MW/Pharm, an integrated software package for drug dosage regimen and therapeutic drug monitoring. Comput Biol Med 1992;22:155-63.
24. Akaike H. An information criterion. Math Sci 1976;14:5-9.
25. Steele WH, Stuart JFB, Lawrence JR, et al. Enhancement of methotrexate absorption by subdivision of dose. Cancer Chemother Pharmacol 1979;3:235-7.
26. Lambert CM, Sandhu S, Lochhead A, Hurst NP, McRorie E, Dhillon VB. Dose escalation of intramuscular (im) methotrexate (MTX) for patients with active rheumatoid arthritis (RA) does not improve disease control — a randomized placebo controlled trial [abstract]. Arthritis Rheum 2002;46 Suppl:S204.
27. Hoekstra M, van de Laar MAFJ, Bernelot Moens HJ, Kruijsen MWM, Haagsma CJ. Longterm observational study of methotrexate use in a Dutch cohort of 1022 rheumatoid arthritis patients. J Rheumatol 2003;30:2325-9.