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Methotrexate administered by 6-h and 24-h infusion: a pharmacokinetic comparison*

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Summary. The pharmacokinetics of 8 g/m^2 methotrexate (MTX) was compared following short (6 h) and long (24 h) infusions of the drug to 11 children with osteogenic sarcoma (OS; 42 infusion) and 28 children with acute lymphoblastic leukemia (ALL: 118 infusions), respectively. No difference was observed in the first-phase half-life, in systemic clearance or in the volume of distribution of the drug ($P > 0.05$). The concentration of MTX at the end of the infusion was ~4-fold higher when the drug was given over only 6 h. However, patients receiving 24-h infusions had ~9-fold higher levels by 24 h after the beginning of the infusion. The area under the data curve from start of the MTX infusion until the beginning of folinic acid rescue administration was significantly higher in patients with osteogenic sarcoma (6-h infusions), while the area under the log-data curve was significantly longer in the ALL group (24-h infusions) for the same period. The latter parameter is considered to be characteristic for the concentration-time-effect relationship. The longer duration of MTX administration (with delayed rescue) is thought to be more beneficial from the pharmacokinetic aspect. Patients with osteogenic sarcoma had significantly lower concentrations of MTX at the end of their last treatment with MTX than at the end of the first infusion. Patients developing MTX toxicity had shorter half-lives of MTX in the beta phase. It is suggested that cisplatin induced tubular loss of MTX and folinic acid is responsible for these observations. A wider application of clinical pharmacologic findings in the practice of the administration of cytostatics is indicated.

Introduction

The pharmacokinetics of methotrexate (MTX) has been extensively studied during the last 39 years: a Medline search provided 1883 titles for the period of 1966–1986 under the keywords methotrexate, pharmacodynamics. However, in spite of the large number of publications in this field, most of the protocols for the administration of intermediate or high-dose MTX with folinic acid rescue have empiric rather than pharmacokinetic bases. Therefore, the generally used regimens vary widely in dosage (200–33 600 mg/m^2), duration of administration (1–42 h), and exposure time (3–48 h). The dose- and time-schedules of folinic acid rescue administration also differ. We have compared the pharmacokinetic parameters of MTX following short (6 h) and long (24 h) infusions of 8000 mg/m^2 of the drug.

Patients and methods

Data were obtained during and after 42 infusions of 8 g/m^2 over 6 h to 11 children age: 11.8±3 years) with osteogenic sarcoma (OS) and 118 infusions of 8 g/m^2 MTX in 24 h to 28 children (age: 8.1±4.9 years) with acute lymphoblastic leukemia (ALL). MTX was administered as a part of the EORTC/SIOP/MRC 80831 protocol for OS and as a part of the Norwegian Pilot Study for ALL [10]. No loading dose was given to patients with OS, while a loading dose of 800 mg/m^2 was administered during 1 h to patients with ALL. All patients received 3000 ml/m^2 per 24 h i.v. hydration and urinary alkalinization. The administration of folinic acid rescue started 24 h (OS) or 36 h (ALL) after the start of the MTX infusion. None of the patients had evidence of renal or hepatic impairment at the time of treatment. No significant toxicity due to MTX developed after the treatments in the ALL group, whereas one severe (bone marrow, gastrointestinal, renal and hepatic disfunction) and four milder (mucosal ulcerations, fever, emesis) toxic reactions occurred in the OS group.

The patients with ALL were treated in the Department of Paediatrics, University of Trondheim, and the treatment of patients with OS was conducted in the 2nd Department of Paediatrics, Semmelweis Medical School, Budapest.

Venous blood samples were drawn 4–6, 24, 48 and 72 h after the start of the MTX infusion in the OS study and 22–26, 38–44, 66–78, and 88–110 h after the start of the MTX infusions in the ALL-study. All blood samples were subsequently centrifuged and analysed. The determination of MTX concentration was performed by radioassay (Trondheim) or enzyme-inhibition assay [3, 12]. Results of these two methods are known to correlate well enough (11) to allow comparison of the two data groups.

Compartment model-independent pharmacokinetic parameters of MTX were calculated with a PC program

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Table 1. Comparison of pharmacokinetic parameters of MTX, 8 g/m^2, administered over 6 h (OS) or over 24 h (ALL)

Group	OS: 6-h infusion		ALL: 24-h infusion
Number of infusions	42		118
Concentration at end of infusion	1.1 (± 0.7) × 10^{-3} mol l^{-1}	*	2.9 (± 4.6) × 10^{-4} mol l^{-1}
Concentration 24 h after start of infusion	3.3 (± 3.0) × 10^{-5} mol l^{-1}	*	as above
Systemic clearance ml/min/per m^2	56.4 (±29.8)	**	66.3 (±36.4)
Elimination half-life (α) (h)	2.45(± 1.43)	**	2.21(± 0.4)
Elimination half-life (β) (h)	6.1 (± 2.1)	*	9.4 (± 4.4)
V_d (at end of infusion) l/m^2	11.8 (± 8.1)	**	13.6 (± 6.6)
AUDC mg ml^{-1} × min	361.5 (±162.2) (0–24 h)	*	234 (±140.9) (0–36 h)
AULDC	1.11(±0.2) (0–24 h)	*	3.26(±0.32) (0–36 h)

V_d, Volume of distribution; AUDC, area under the data curve; AULDC, area under the log-data curve
* $P < 0.01$; ** $P > 0.05$

PharmCalc [4]. Student's *t*-test was used to test significant differences between mean pharmacokinetic parameters of grouped data. Statistical calculations were performed with STATMED PC program [8] kindly provided by Nycomed Scandinavia.

Results

The pharmacokinetic parameters examined are summarized in Table 1. The comparison of concentrations of MTX in the serum at the end of the infusion of the drug revealed that the level of MTX was almost 4-fold higher when the drug was given over 6 h. The difference is statistically significant ($P < 0.01$). However, 24 h after the start of the infusion OS patients had 9-fold lower concentrations than patients with ALL.

OS patients had significantly higher levels of MTX at the end of their first MTX treatment than at the end of their last infusion of MTX: 1.7 (±0.4) × 10^{-3} mol/l and 8.4 (±1.6) × 10^{-4} mol/l, respectively ($P < 0.01$).

No significant difference was found between the values for systemic clearance of MTX in the OS and ALL groups. The half-life of MTX was found to be about the same during the first elimination phase in the two groups ($P > 0.05$), but the half-life during the second 24 h of the postinfusion period was significantly shorter in the OS group ($P < 0.01$). It is of interest that the five "toxic" infusions in the OS group were characterized by significantly shorter second-phase half-life values than the infusions without toxicity (4.2 ± .04 h versus 6.9 ± 2.2 h: $P < 0.01$).

There was no significant difference in the volume of distribution of MTX in the two groups of patients.

Area under the data curves (AUDC) and area under the log concentration-data curves (AULDC) for the periods 0–24 h and 0–36 h were calculated for the OS and ALL groups, respectively. These periods were chosen instead of total AUC because they reflect the actual duration of drug exposure (before the rescue administration has been started). Comparison revealed an inverse relationship: AUDC (0–24 h) was significantly higher in the OS group than was AUDC (0–36 h) in the ALL group, but AULDC (0–36 h) of the ALL patients was significantly higher than AULDC (0–24 h) in the OS group.

Discussion

We have analysed a number of the pharmacokinetic parameters of MTX in two groups of patients receiving the same dose (8 g/m^2) during a short (6 h) and a longer term (24 h) infusion. No significant difference was observed in the mean values for systemic clearance, first-phase elimination half-life, or volume of distribution, but the mean values for concentration achieved at the end of the infusion, half-life during the second 24 h of the postinfusion period, and AUDC and AULDC (from the start of the infusion until the beginning of the rescue administration) differed significantly.

However, we were concerned to find whether or not the above-mentioned differences in the pharmacokinetic parameters have any clinical relevance. In OS patients, concentrations at the end of the infusion are peak concentrations (infusion time did not last for 3.3 half-lives [15], but 24-h concentrations of ALL patients are representative of steady-state levels. Although peak levels in the OS group were more than 4 times those of the ALL group, by about 5 h after the end of the infusion (estimated value), as a result of the fast postinfusion decrease, concentrations were around the same, and 24 h after the start of the treatment levels of MTX were about 9 times higher in the ALL group. Moreover, MTX had a further 12 h to act in ALL patients. By 36 h after the start of treatment the concentrations of MTX in the ALL group had decreased to around the same level as was present in OS patients 24 h after the beginning of the infusion (data not shown). Although threshold MTX levels have been reported for a variety of normal and tumorous tissues in vitro [1, 6, 13, 16], the authors do not know of any such threshold concentration of MTX for OS cells. Our data suggest that OS patients may benefit more from the short infusion only if the level of sensitivity of the tumor is above 8–10 × 10^{-4} mol/l, but the very short duration of such high levels makes even that uncertain. However, if this is so, the administration of a higher dose in 24-h infusions would be advisable.

As an explanation for the difference in the half-lives during the second 24 h of the postinfusion period it is suggested that the distribution phase might not have been complete during the first 24 h in the OS group, but our sampling times did not allow a more specific analysis. The

shorter half-life of MTX observed in OS patients developing toxicity, evaluated together with the sequential decrease in the peak concentrations of MTX during the consecutive treatments in the OS group, might be a result of the administration of cisplatin in this protocol. Hypothetically, the lack of tubular reabsorption of MTX and folinic acid due to cisplatin tubulopathy could be responsible for our findings. These observations indicate another mechanism of the effect of cisplatin on the pharmacokinetics of MTX [7]. However, the amount of MTX excreted in the urine was not determined in our study.

The descriptive parameter of concentration-exposure time, AUDC was higher in OS patients (0–24 h) than in ALL patients (0–36 h), but this parameter cannot be connected directly with the efficiency of the drug [14]. Although the cytotoxic effects of anticancer agents are considered to be irreversible, the mechanism of action and the clinical working concept of high-dose MTX with leucovorin rescue is based on the reversible binding of MTX to the target enzyme dihydrofolate reductase. Therefore, the effect of the drug theoretically depends on the exposure time and the logarithm of the amount of the drug in the body [15]. The area under the log-concentration curve was significantly higher (~3 times) in the ALL group. It is interesting that when this method is applied for the equitoxic concentration-time data reported by Pinedo and Chabner [14] it also results in rather similar values. Clinical observations also support our assumption that a longer infusion time is more effective: good results have been reported with long, 36-42 h infusions of MTX to patients with ALL, lymphoma and rhabdomyosarcoma [2, 5, 9]. Moreover, this treatment was also effective in those patients who did not respond to short infusions of the drug.

It is beyond the scope of this study to analyse therapeutic results with either of the protocols used here. Although therapeutic response depends on a number of factors other than the pharmacologic effect of the drug, the authors believe that a more thorough consideration of clinical pharmacokinetic findings in the planning of a protocol could improve the therapeutic results.

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References

1. Bender RA (1975) Membrane transport of methotrexate (NSC-740) in human neoplastic cells. Cancer Chemother Rep 6: 73
2. Bode U (1986) Methotrexate as relapse therapy for rhabdomyosarcoma. Am J Pediatr Hematol Oncol 8: 70
3. Borsi JD (1984) Simple assay of methotrexate for the practice of clinical oncology. Orv Hetil 125: 2629
4. Borsi JD, Klepp O, Moe PJ (1986) PharmCalc: Program for the calculation of clinical pharmacokinetic parameters of methotrexate. Cancer Chemother Pharmacol (submitted for publication)
5. Buchmann S, Fengler R, Hartmann R, Laskari J, Wolfrom C, Ebell W, Gutjahr P, Havers W, Janka G, Neuhaus C, Suder J, Henze G (1987) Comparison of intermediate versus high dose methotrexate for relapsed childhood acute lymphoblastic leukemia. Fourth International Symposium on Therapy of Acute Leukemias, Rome, abstract book, p 405
6. Chabner BA, Young RC (1973) Threshold methotrexate concentration for in vivo inhibition of DNA synthesis in normal and tumorous target tissues. J Clin Invest 52: 1804
7. Jaffe N, Keifer R III, Robertson R, Takue Y (1987) Renal toxicity with cumulative doses of *cis*-diamminedichloroplatinum-II in pediatric patients with osteosarcoma. Cancer 59: 1577
8. Klepp O (1986) Statmed program in GWBASIC. Nycomed, Oslo
9. Magrath IT, Spiegel RJ, Edwards BK, et al. (1980) Altered relapse pattern of Burkitt lymphoma in America. Proc Am Soc Clin Oncol 21: 348
10. Moe PJ, Kolmannskog S, Finne PH, Seip M, Wesenberg F (1987) Methotrexate infusions in childhood acute lymphocytic leukemia. Fourth International Symposium on Therapy of Acute Leukemias Rome, abstract book, p 188
11. Monjanel S, Rigault JP, Cano JP (1979) High-dose methotrexate: preliminary evaluation of a pharmacokinetic approach. Cancer Chemother Pharmacol 3: 189
12. Myers CE, Lippman ME, Eliot HM, Chabner BA (1975) Competitive protein binding assay for methotrexate. Proc Natl Acad Sci USA 72: 3683
13. Ohnuma T, Lo RJ, Scanlon KJ, Kamen BA, Ohnoshi T, Wolman SR, Holland JF (1985) Evolution of methotrexate resistance of human acute lymphoblastic leukemia cells in vitro Cancer Res 45: 1815
14. Pinedo HM, Chabner BA (1977) Role of drug concentration, duration of exposure, and endogenous metabolites in determining methotrexate cytotoxicity. Cancer Treat Rep 61: 709
15. Rowland M, Tozer TN (1980) Clinical pharmacokinetics: concepts and applications. Lea and Febiger, Philadelphia
16. Sirotnak FM, Donsbach RC (1974) The intracellular concentration dependence of antifolate inhibition of DNA synthesis in L1210 leukemia cells. Cancer Res 34: 3332

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