Acta Oncologica Vol. 35, No. 2, pp. 207–212, 1996

DIFFERENT INTRAVENOUS ADMINISTRATION TECHNIQUES FOR 5-FLUOROURACIL

Pharmacokinetics and pharmacodynamic effects

PER-ANDERS LARSSON, GÖRAN CARLSSON, BENGT GUSTAVSSON, WILHELM GRAF and BENGT GLIMELIUS

The pharmacokinetics after 20 min intravenous infusion or a 2 min bolus (push) injection of 5-fluorouracil (500 mg/m^2) were studied in 14 colorectal cancer patients. Treatment effects and toxicity related to the administration technique of 5-fluorouracil were retrospectively analysed in 198 colorectal cancer patients. The AUC after bolus injection was 6 158 ± 874 μmol/l*min compared to 3 355 ± 428 μmol/l*min after short-time infusion of 5-fluorouracil ($p < 0.01$). The mean peak-level after bolus injection was 341 ± 34 μM versus 161 ± 17 μM after a short-time infusion ($p < 0.01$). Patients receiving bolus injections had significantly better treatment result (32% partial remission) than patients receiving infusion (10% partial remissions, $p < 0.001$). Toxic side-effects were more frequently encountered after bolus injection but subjective improvement was also more frequently experienced by these patients. Bolus 5-fluorouracil push injection rather than a short-time infusion appears to be the more efficient administration technique.

5-Flurorouracil (5-FU) remains one of the most widely used agents in pharmacotheraphy against breast and colorectal cancer (1). Basic research in 5-FU anabolism, catabolism, excretion and cytotoxicity has provided a theoretical background for its usefulness (2). However, response rates rarely exceed 20% in single-drug regimens and seldom reach 50% when combined with biochemical modulators (3). Despite extensive efforts in 5-FU pharmacokinetic research, the individual variation in clinical response and drug metabolism has been a persisting challenge. The hepatic circulation is important for 5-FU elimination, but plasma clearance rate equals or exceeds liver circulation in some patients. Extensive drug elimination has been found in the cardiopulmonary circulation and in the splanchnic area (4, 5). The plasma clearance equals or exceeds cardiac output at low-drug concentrations. A marked non-linear relationship between 5-FU dose and plasma concentrations has been found when comparing bolus injections and prolonged infusions. For standard bolus doses clearance was 0.5–1.4 l per min, whereas clearance rates after prolonged infusions were 10- to 60-fold higher. This non-linearity indicates a saturable metabolic or transport process (6, 7). Port et al. (8) have studied the elimination rate after a 10-min infusion and found a decreased plasma clearance from 1.3 l/min after infusion of 320 mg 5-FU/m^2 to 0.7 l/min after infusion of 960 mg 5-FU/m^2. All observers who have studied 5-FU pharmacokinetics have noted marked interindividual variations in plasma half-life of the drug. The characteristics of later phases of drug elimination have not been clearly elucidated (9).

In the past two decades there have essentially been two major schedules for intravenous administration of 5-FU, either bolus injection or constant infusion for usually more than 72 h (10). Despite our knowledge of 5-FU metab-

Received 22 March 1995.
Accepted 30 September 1995.
From the Department of Surgery, Östra sjukhuest, Göteborg University, Göteborg, (P-A Larsson, G. Carlsson, B. Gustavsson) and Department of Surgery (W. Graf) and Oncology (B. Glimelius), Akademiska sjukhuset, Uppsala University, Uppsala, Sweden.
Correspondence to: Dr P-A Larsson, Department of Surgery, Östra sjukhuest, S-416 85 Göteborg, Sweden.

 ISSN 0284-186X

olism, the major mechanisms for its antitumour effects have not been fully evaluated in any of these schedules (11, 12).

The technique for administration of bolus doses of 5-FU appears clear-cut, but varies between different physicians and institutions. The introduction of 'minibags' (100 ml of physiologic saline or glucose solution) has provided a convenient way for intravenous administration of drugs, but if the active drug is mixed with the solution in the minibag it has prolonged the injection time from a few minutes to time periods between 5 and 30 min. The pharmacokinetics of 5-FU has been studied extensively and dose-dependent plasma clearance has been reported for bolus puch injection and for prolonged continuous infusion (13–17), but the pharmacokinetics and pharmacodynamics of short-time infusions, less than 30–60 min, compared to bolus push injections have not yet been addressed. The influence on therapeutic efficacy of the administration technique deserves proper assessment since the use of short-time infusion with minibag systems, rather than bolus injection, is increasing.

The aims of the present study were to analyse the plasma peak levels and AUC after bolus push injection compared to short-time infusion in cancer patients treated with 5-FU and to evaluate the clinical response rates and frequency of adverse side-effects in patients with advanced colorectal cancer with regard to these administration techniques.

Material and Methods

Pharmacokinetic study

Patients, treatment and sample collection. A series of 14 patients (8 men and 6 women, average age 70 ± 5 years) with surgically non-curable colorectal cancer were investigated after informed consent. All patients had liver metastases and all were treated with weekly 5-FU/ leucovorin at the outpatient chemotherapy office, Östra sjukhuset, Göteborg, Sweden. The treatment schedule and drug doses used in this pharmacokinetic study did not differ from the standard palliative treatment for advanced colorectal cancer at the clinic. All patients were treated with a weekly regimen of 5-FU 500 mg/m^2, in combination with leucovorin, 50 mg. 5-FU was either added to 100 ml NaCl 9 mg/ml and infused at a constant rate in a forearm vein during 20 min or push injected in the forearm vein at a concentration of 50 mg/ml (injection time 2 min) followed by an injection of 20 ml isotonic saline. Leucovorin was given in the same vein 30–40 min after 5-FU. Blood samples were collected from the other arm immediately before the bolus injection or at the start of the infusion. Additional samples were collected at 5, 10, 20, 30 and 60 min after the start of the bolus injection or the infusion. After centrifugation of the samples, 2 ml plasma was aspirated and stored at −20°C for drug analysis. Each patient was treated three times by the bolus injection technique and three times with the short-time infusion technique. The study was approved by the committee for ethical supervision of research, Göteborg University, Sweden.

Drug assay. The plasma concentrations were determined according to earlier described methods with some modifications (18, 19). Briefly 1.0 ml plasma samples, adjusted to pH 11 with KOH, mixed with 4.0 ml 1% picric acid and 100 µl 1.0 mM thymine as internal standard, were applied to an ion exchange AG-1-X8 resin column (BioRad, Richmond, CA) and eluated with 20.0 ml 0.1 M formic acid. The eluate was lyophilised and subsequently redissolved in 15 mM KH_2PO_4/K_2HPO_4 buffer. The aqueeous extract was chromatographed on C_{18}-resolve column (Waters Associates, Milford, Mass) with 0.0015-M KH_2PO_4 as eluent. The UV absorption at 254 nm of the eluate was monitored and compared with the external standard curves from HPLC-buffer, spiked with known amounts of 5-FU and 100 µl 1.0 mM thymine. Typically we have 75% recovery of 5-FU by this technique. The intra-/inter-assay coefficients of variation were 3.0% and 3.8% respectively.

Mathematical and statistical analysis. The non-compartmental area under the plasma concentration curve (AUC) was determined from the complete time concentration curves by using the linear trapezoidal rule before, and a logarithmic trapezoidal rule beyond the peak concentration. Peak value and AUC was recorded for every treatment. The half-life ($t_{1/2}$) was determined according to the trapezoidal rule. The interindividual variation and the intraindividual variation are presented in Table 1. Plasma peak concentrations, AUC values and patient group characteristics are presented as mean ± standard error of the mean (SEM).

Restrospectrive analysis of treatment efficacy

Analysis of treatment and side-effects of FUra in patients. In a retrospective follow-up study, 198 eligible patients with advanced colorectal cancer, included in a

Table 1

Coefficients of variations due to assay and between and within individual variations for peak and AUC-levels

	Overall %	Bolus %	Infusion %
AUC-levels			
Assay	4.9		
Within object	28.9	26.4	32.2
Between objects	37.0	19.2	52.1
Peak-levels			
Assay	2.2		
Within object	41.1	34.1	25.2
Between objects	44.1	35.9	39.2

Nordic multicenter trial comparing 5-FU and leucovorin (n = 100) versus sequential methotrexate, 5-FU and leucovorin (n = 98), monitored by the Nordic Gastrointestinal Tumor Adjuvant Therapy Group (20) were analysed for response and adverse effects of the treatment protocol. These patients were treated at 19 different hospitals of which 7 (82 included patients) administered 5-FU as a bolus injection in less than 5 min (median 2 min) and 12 (116 patients) administered 5-FU as a short-time infusion (5–30 min) using a minibag as described above. The median injection time was 20 min in the second group. All patients at every single hospital were treated in accordance with the same administration routines, and the categorization into the two groups was done prior to data analysis. The proportions of patients receiving bolus injections and short-time infusions in the different treatment arms are presented in Table 2. The 5-FU dose was 500 mg/m^2 body surface area, either combined with leucovorin, 60 mg/m^2, or with methotrexate, 250 mg/m^2, followed by leucovorin rescue (15 mg × 8). No differences in treatment outcome were seen between two regimens (20). Fifty-seven per cent of the patients were men and 43% women with an average age of 63 years. All patients had a Karnofsky performance index of 60 or more at the onset of the study. Neither of these parameters nor treatment arm differed significantly between the group treated with 5-FU bolus push injections and that treated with short-time infusions. The patients were interviewed regarding subjective treatment effects, and objective response was recorded, usually by CT-scans 2 and 4 months after onset of treatment. The minimum duration of any response or disease stabilisation was 4 months. The leukocyte and platelet counts were recorded prior to each treatment course. At 8 hospitals (70 patients) participating in an associated quality of life study, subjective side-effects such as nausea, vomiting, diarrhoea, stomatitis and conjunctivitis were recorded by means of questionnaires (21). Statistical evaluation of the clinical data was performed using standard analyses, i.e. the χ^2-test for comparing proportions and Student's t-test for comparing means. Surivial was evaluated with the log-rank test. P-values < 0.05 were regarded as statistically significant.

Table 2

Response and changes in quality of life among patients given intravenous injection or infusion. Figures are number of patients (%)

	Intravenous injection	Intravenous infusion
	< 5 min (n = 82)	⩾ 5 min (n = 116)
Treatment arm+		
MFL (n = 98)	40 (49)	58 (50) n.s.
FLv (n = 100)	42 (51)	58 (50) n.s.
Objective response		
Partial remission	26 (32)	12 (10)***
Stable disease	15 (18)	32 (28)
Progressive disease	41 (50)	72 (62)
Subjective response		
Improved	46 (56)	34 (29)**
Stable/worse	36 (44)	82 (70)
Quality of life (n = 70)	(n = 33)	(n = 37)
Improved	15 (45)	10 (27) n.s.

+MFL = sequential methotrexate, 5-fluorouracil and leucovorin
FLv = 5-fluorouracil and leucovorin
*** $p < 0.001$. ** $p < 0.01$, n.s = not significant.

Results

Time concentration curves after bolus injection and intravenous infusion are presented in the Figure. As shown, the peak level after bolus injection and after the start of infusion was reached after 5 min. The half-life ($t_{1/2}$) varied between 9 and 17 min with an average of 15 min for both administration techniques. Average distribution volume after bolus injection was 26% of body weight or 18 046 ml and clearance was 1 203 ml/min/m^2.

Average AUC after bolus injection was 6 158 ± 874 μmol/l*min and after infusion 3 355 ± 428 μmol/l*min ($p < 0.01$). Peak levels after bolus injection were 341 ± 34 μM, and after infusion 161 ± 17 μM ($p < 0.01$). During the short-time infusions, steady state conditions were not obtained (Figure). Plasma clearance during a 20-min infusion was 1 851 ml/min/m^2 and average distribution volume was the same as after bolus injection. The shape of the plasma concentration curve during the short time infusion (Figure) is unusual for a one-compartment model with declining concentrations during ongoing infusion but results from the plasma clearance which, at a concentration of 120 μM, considerably exceeds the infu-

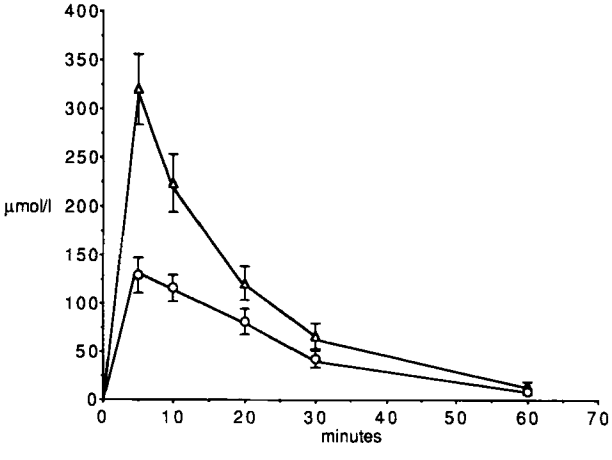

Figure. 5-FU Plasma concentration curves after bolus (push) injections (2 min). Means and S.E.M. from 14 patients. △ = Bolus injections; ○ = Intravenous infusions.

sion rate of 45 mg 5-FUra/min. The calculated plasma clearance is in accordance with previously published pharmacokinetic data after continous infusion (for review see (9)).

The intraindividual and the between object variation of AUC values were more pronounced after short-time infusion than after bolus injection (Table 1).

In the retrospective study of treatment results from the Nordic trial, 32% (26/82) of the patients treated at hospitals giving bolus injection obtained partial remission, compared to 10% (12/116) of the patients receiving short-time infusion ($p < 0.001$, Table 2). The median survival of the patients treated with 5-FU bolus push injections was 247 days (range 5-889) compared with 237 days (range 9–919) in those treated with short-time infusion (not significant). When the patients were interviewed for subjective improvement (Table 2), 46 of the 82 patients (56%) given bolus injections reported subjective improvement compared with 34 of the 116 patients (29%) given intravenous infusion ($p < 0.01$). The distribution of adverse reactions, both with respect to laboratory tests and subjective toxicity, are presented in Table 3. It can be seen that myelosuppression, diarrhoea, and stomatitis were significantly more common after bolus injection. Fifteen of 33 (45%) of the patients receiving bolus injections but only 10 of 37 (27%) of the patients receiving short-time intravenous infusion reported an improved quality of life (not significant).

Table 3

Laboratory and subjective side-effects of the treatment in patients receiving bolus injection or intra-venous infusion in the retrospective follow-up study. Figures are number of patients (%)

	Intravenous injection	Intravenous infusion
	<5 min (n = 82)	≥5 min (n = 116)
Granulocyte count	15 (18)	3 (3)***
Grade+ II	14	1
Grade III	1	
Grade IV		2
Nausa/Vomiting	29 (35)	58 (50) n.s.
Grade I	19	36
Grade II	5	15
Grade III	5	6
Grade IV		1
Diarrhoea	32 (39)	19 (16)***
Grade I	11	11
Grade II	15	5
Grade III	5	3
Grade IV	1	
Stomatitis	23 (28)	12 (10)**
Grade I	4	4
Grade II	11	7
Grade III	6	
Grade IV	2	1
Cutaneous reactions	4 (5)	10 (9) n.s.
Grade II	2	7
Grade III	1	1
Grade IV	1	2
Conjunctivitis	18 (22)	13 (11) n.s.
Grade I	14	9
Grade II	3	2
Grade III	1	2

+WHO: Recommendations for grading
*** $p < 0.001$. ** $p < 0.01$, * n.s. = not significant.

Discussion

Two main treatment protocols were found to be used in the retrospective follow-up study, i.e. bolus (push) injection with a syringe and short-time infusion using minibags. The median infusion times with these two techniques were 2 and 20 mins respectively and thus formed the rationale for chosing these infusion times for the prospective pharmacokinetic study.

Bolus (push) injection compared to an infusion during 20 mins of 500 FUra/m^2 results in higher peak levels and AUC of the drug, most probably explained by saturation of the metabolic degradation pathway during a significant part of the 2-min injection time. Since the first plasma concentration levels were obtained 5 min after start of the injections (3 min after termination of the push injection), peak concentrations are probably underestimated and may be closer to 500 μM, which would further enhance the difference between these two administration techniques. The infusion of the same amount of 5-FU is not sufficient for achieving steady state conditions, as the infusion time 20 min represents about 1.5 $t_{1/2}$ and achievement of steady state levels requires infusion times of 4–5 $t_{1/2}$ (5).

The subgroup analysis from the Nordic trial (20) indicates that an intravenous push injection rather than a short-time infusion results in a significantly increased response rate. In vitro studies have shown that 1 μM of 5-FU is required for cytotoxicity, but unequivocal data for plasma concentration limits to obtain an antitumor effect are not prevailing. Previous reports have stated a significant correlation between AUC and clinical toxicity (16). The results in this pharmacokinetic study are not sufficient for establishing concentration limits for cytotoxicity but our data provide evidence that either the increased AUC values obtained due to the higher peak levels or the peak levels per se, being significantly higher after push injections, are of importance for the efficacy of 5-FU therapy.

Several ways to overcome the catabolic pathways and increase the intratumour bioavailability of 5-FU have been proposed. Recently 5-ethynyluracil has been shown to be a promising agent increasing 5-FU efficacy against experimental tumours in rats (22) and 5-FU in combination with leucovorin, methotrexate or levamisole have been successful in clinical trials. Thus 5-FU is today rarely given as a single-drug regimen which further complicates analysis and optimisation of dosage and schedule (3, 23, 24).

Low 5-FU concentrations have been reported to produce a thymidine-reversible toxicity in cell cultures, whereas toxic effects from high 5-FU concentrations are not reversible by thymidine (25). From these early studies it was also reported that human cells in vitro were not affected by low concentrations of 5-FU (5–20 μM) due to their higher innate thymidylate synthase activity and their more developed ability to synthesise dUMP. The human cell lines were, however, sensitive to inhibition by 5-FU at concentrations 50–200 μM. Controversy exists in the literature whether the relatively good clinical responses achieved by continuos infusion of 5-FU are related to the steady state concentrations or to the AUC (17). Some studies state that the toxicity related to continuos infusion of 5-FU is proportional to the height of the time concentration curve in contrast to the therapeutic efficacy which is maximal at lower plasma concentrations. However, the mechanism of the 5-FU cytotoxicity in these schedules may be different from the major effect mechanism after bolus injection/short-time infusion. 5-FU is a cytotoxically inactive parent compund which after metabolisation to FUTP, FdUPT or FdUMP excerts its effects by a) incorporation of FUTP into RNA, b) FdUPT incorporation into DNA and c) association of FdUMP with TS resulting in inhibition of dTTP formation. Although the relative importance of these mechanisms are still controversial the concentrations of the active metabolites are related to the achieved concentration of the parental 5-FU.

The frequencies of leukocyte depletion and gastrointestinal reactions were significantly higher in the patients receiving bolus push injections. Earlier reports have stated that granulo- and trombocytopenia are more common after bolus injection, whereas stomatitis and gastroenteritis are more common after prolonged continuos infusion. Thus, prolongation of the administration time for non-continuos 5-FU treatment could be a way of minimizing side-effects, but the greater proportion of patients reporting subjective improvement and improved quality of life after bolus push injection disproves such benefits.

Since curative therapy against metastatic colorectal cancer is not available today, the main goal for treatment of these patients must be to improve their quality of life and to prolong survival without adversely affecting their well-being. Various 5-FU based schedules have a proven beneficial effect for some of these patients (20, 21). As long as a better drug is not available, it is of utmost importance for the responsible physician to provide the patient an optimized treatment. Although a short-time intravenous infusion is a simple and convenient method for administration of 5-FU, the bolus push injection technique seems to be superior for these patients. A pharmacokinetic rationale for this difference in treatment effect has been documented in the present study. Since the data from the 'Nordic Trial' were based on a retrospective analysis, and thus only can be hypothesis-generating, a prospective study, focusing on the therapeutic efficacy with regard to the different administration techniques is now conveyed, monitored by the Nordic Gastrointestinal Tumor Adjuvant Therapy Group.

ACKNOWLEDGEMENTS

This study was supported by grants from the Swedish Cancer Society (0512-B93-06XCC) and the Swedish Medical Research Council (B94-04X 07155-10C).

REFERENCES

1. Heidelberger C. Fluorinated pyrimidines and their nucleosides. In Sartorelli AC, Johns DG, eds. Handbook of experimental pharmacology, antineoplastic and immunosuppressive agents. New York: Springer Verlag, 1975.
2. Moertel CG. Chemotherapy for colorectal cancer. Engl J Med 1994; 330: 1136–42.
3. Einhorn LH. Improvements in fluorouracil chemotherapy? J Clin Oncol 1989; 7: 1377–9.
4. Andersson M, Billström Å, Domellöf L, et al. Cardio-pulmonary elimination of 5-fluorouracil after bolus injection in the hepatic artery. Eur J Surg Oncol 1987; 13: 35–9.
5. Gustavsson BG, Brandberg Å, Regårdh CG, et al. Regional and systemic serum concentrations of 5-fluorouracil after portal and intravenous infusion: an experimental study in dogs. J Pharmacokinet Biopharm 1979; 7: 665–73.
6. Diasio RB, Harris BE. Clinical pharmacology of 5-fluorouracil. Clin Pharmacokinet 1989; 16: 215–37.
7. Santini J, Milano G, Thyss A, et al. 5-FU therapeutic monitoring with dose adjustment leads to an improved therapeutic index in head and neck cancer. Br J Cancer 1989; 59: 287–90.
8. Port RE, Daniel B, Ding RW, et al. Relative importance of dose, body surface area, sex, and age of 5-fluorouracil clearance. Oncology 1991; 48: 177–81.
9. Grem JL. Fluorinated pyrimidines in cancer chemotherapy. In: Chabner BA, Collins JM, eds. Priniciples and practice. Philadelphia: J.B. Lippincott Company, 1990.
10. Anderson N, Lokich J. Controversial issues in 5-fluorouracil infusion use. Dose intensity, treatment duration and cost comparisons. Cancer 1992; 70: 998–1002.
11. Chu E, Johnston P, Takimoto C, et al. Antimetabolites. Cancer Chemother Biol Response Modif 1993; 14: 1–25.
12. Aschele C, Sobrero A, Faderan MA, et al. Novel mechanism(s) of resistance to 5-fluorouracil in human colon cancer (HCT–8) sublines following exposure to two different clinically relevant dose schedules. Cancer Res 1992; 52: 1855–64.
13. Port RE, Edler L, Herrmann R, et al. Pharmacokinetics of 5-fluorouracil after short systemic infusion: plasma level at the end of the distribution phase as an indicator of the total area under the plasma concentration-time curve. Ther Drug Monit 1991; 13: 96–102.
14. Payet B, Fabre G, Tubiana N, et al. Plasma kinetic study of folinic acid and 5-methyltetrahydrofolate in healthy volunteers and cancer patients by high-performance liquid chromatogrraphy. Cancer Chemother Pharmacol 1987; 19: 319–25.
15. Heggie GD, Sommadossi J-P, Cross DS, et al. Clinical pharmacokinetics of 5-fluorouracil and its metabolites in plasma, urine, and bile. Cancer Res 1987; 47: 2203–6.
16. van Groeningen CJ, Pinedo HM, Heddes J, et al. Pharmacokinetics of 5-fluorouracil assessed with a sensitive mass spectrometric method in patients on a dose escalation schedule. Cancer Res 1988; 48: 6956–61.
17. Yoshida T, Araki E, Iigo M, et al. Clinical significance of

monitoring serum levels of 5-fluorouracil by continuous infusion in the patients with advanced colonic cancer. Cancer Chemother Pharmacol 1990; 26: 352–4.
18. Buckpitt AR, Boyd MR. A sensitive method for determination of 5-fluorouracil and 5-fluorodeoxyuridine in human plasma by high-pressure liquid chromatography. Anal Biochem 1980; 106: 432–7.
19. Gustavsson B, Baldesten A, Hasselgren P-O, et al. New assay of 5-fluorouracil in serum by isotachophoresis. Chromatogr 1979; 179: 151–9.
20. The Nordic Gastrointestinal Tumor Adjuvant Therapy Group. Biochemical modulation of 5-fluorouracil: A randomized comparison of sequential methotrexate, 5-fluorouracil and leucovorin versus sequential 5-fluorouracil and leucovorin in patients with advanced symptomatic colorectal cancer. Ann Oncol 1993; 4: 235–40.
21. Glimenlius B, Hoffman K, Graf W, et al. Quality of life during chemotherapy in patients with symptomatic advanced colorectal cancer. Cancer 1994; 73: 556–62.
22. Cao S, Rustum YM, Spector T. 5-Ethynyluracil (776C85): modulation of 5-fluorouracil efficacy and therapeutic index in rats bearing advanced colorectal carcinoma. Cancer Res 1994; 54: 1507–10.
23. Advanced Colorectal Cancer Meta-Analysis Project. Modulation of fluorouracil by leucovorin in patients with advanced colorectal cancer: evidence in terms of response rate. J Clin Oncol 1992; 10: 896–903.
24. Advanced Colorectal Cancer Meta-Analysis Project. Meta-analysis of randomized trials testing the biochemical modulation of fluorouracil by methotrexate in metastatic colorectal cancer. J Clin Oncol 1994; 12: 960–9.
25. Evans RM, Laskin JD, Hakala MT. Effects of excess folates and deoxyinosine on the activity and site of action of 5-fluorouracil. Cancer Res 1981; 41: 3288–95.