A Phase I Trial of a Modified, Dose Intensive FAMTX Regimen (High Dose 5-Fluorouracil + Doxorubicin + High Dose Methotrexate + Leucovorin) with Oral Uridine Rescue

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BACKGROUND. Dose intensification of 5-fluorouracil (5-FU) is complicated by increased toxicity. 5-FU is a fluorine-substituted pyrimidine analog of uracil. In preclinical studies, administration of oral uridine (Ur) has been shown to allow for dose intensification of 5-FU with enhancement of its antitumor activity. Therefore, a Phase I trial was designed aimed at dose intensification of 5-FU as a component of a modified 5-FU-doxorubicin-methotrexate (FAMTX) regimen using oral Ur rescue.

METHODS. Methotrexate (MTX) was administered to all patients at a fixed dose of 1.5 g/m^2. MTX was followed 24 hours later by escalating doses of 5-FU starting at 800 mg/m^2 with leucovorin rescue. Cycles of 5-FU and MTX were repeated every 15 days. Every other cycle, patients received doxorubicin ("Adria cycles") at a dose of 30 mg/m^2. Oral Ur was administered at a dose of 8 gm/m^2 every 6 hours for 12 doses. In the first phase of the study, patients received Ur only if they developed Grade 3 or 4 hematologic toxicity. In the second phase, all patients received Ur 24 hours after 5-FU on all cycles.

RESULTS. Without Ur rescue, the maximum tolerated dose (MTD) of 5-FU was 900 mg/m^2 on the Adria cycles and 1.1 gm/m^2 on the non-Adria cycles. With Ur, the MTD of 5-FU increased to 1.2 gm/m^2 on the Adria cycles and to 1.6 gm/m^2 on the non-Adria cycles.

CONCLUSIONS. In this modified FAMTX regimen, oral Ur administration allowed for dose-intensification of 5-FU, with a 33% increase in the MTD of 5-FU on the Adria cycles and a 45% increase in the MTD of 5-FU dose on the non-Adria cycles. *Cancer* **1996; 78:1988–95.** © *1996 American Cancer Society.*

KEYWORDS: FAMTX, oral uridine, dose intensification, gastric carcinoma.

5-fluorouracil (5-FU) is an antimetabolite pyrimidine analogue that has been studied for many years in the treatment of patients with gastrointestinal malignancies. Evidence has accumulated in both gastric and colorectal tumors that biochemical modulation of 5-FU increases response rates and may have an impact on survival. Specifically, Klein et al. developed the combination of 5-FU, methotrexate (MTX), and doxorubicin (Adria) (FAMTX) for the treatment of patients with advanced gastric carcinoma.[1] In Phase II, single arm trials, they and the European Organization for Research and Treatment of Cancer (EORTC) demonstrated response rates of 33–50%.[1,2] Kelsen et al. performed a Phase II random assignment trial comparing FAMTX and another highly active combination of etoposide, doxorubicin, and cisplatin.[3] In this random assignment trial, FAMTX was at least as

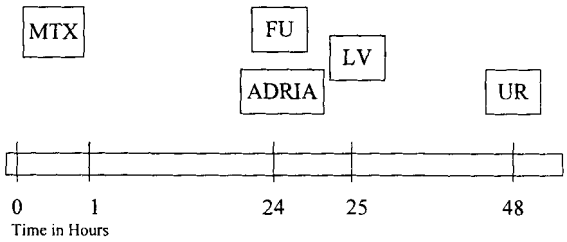
efficacious as EAP (FAMTX response rate 31%). Most important 10% of FAMTX patients had complete responses (Cr), several of which were durable. FAMTX toxicity was significantly less than that with EAP. In another Phase III study, a survival advantage for FAMTX was shown when this regimen was compared with the FU-5-containing combination of 5-FU, doxorubicin and mitomycin-C (FAM).[4]

The FAMTX combination as developed by Klein et al.[1] involves treatment with MTX and 5-FU at respective doses of 1.5 g/m^2 on Day 1 with an interval between treatments of only 1 hour and leucovorin (LV) rescue starting 24 hours after MTX. Doxorubicin is given on Day 15.[1] The treatment recycles on Day 29. Thus, on standard FAMTX patients receive high dose MTX and 5-FU only once per month. More intense therapy with 5-FU/MTX may allow further increases in the response rate. However, dose intensification of 5-FU/MTX is complicated by increased toxicity. The authors have reported that granulocyte-colony stimulating factor does not rescue patients from the hematologic toxicity associated with MTX/5-FU on the standard FAMTX regimen.[5]

The use of uridine (Ur) rescue to enhance the antitumor selectivity of 5-FU was first demonstrated by Martin et al.[6] using CD2F1 mice bearing COLO-26 tumors. Ur ameliorated dose-intensive 5-FU toxicity, as evidenced by increased peripheral leukocyte counts, and was associated with a marked increase in antitumor effect.[6-8] Klubes et al. showed that Ur, but not thymidine or deoxyuridine, could rescue mice from the effects of lethal doses of 5-FU.[8] The mechanism of the rescue of the 5-FU-induced toxicity by Ur is unclear. The toxic effects of 5-FU may be correlated with the incorporation of 5-fluorouridine 5′-triphosphate into RNA.[10] When given after 5-FU, Ur is converted into uridine triphosphate, which may preferentially decrease the incorporation of 5-fluorouridine 5′-triphosphate into the RNA of normal tissues as opposed to tumor tissues.[11]

In clinical trials of intravenous Ur, patients developed significant phlebitis and fever from the intravenous infusion.[12] A preferable method of Ur administration would then be the oral route. Martin et al. showed that oral Ur would safely rescue 5-FU toxicity with antitumor activity comparable to that obtained with parenteral Ur, as long as plasma Ur levels in excess of 50 μM were attained.[8]

A Phase I trial was performed aimed at a dose intensification of 5-FU as a component of the FAMTX regimen using oral Ur rescue. The activity of 5-FU against human tumors can be enhanced by pretreatment with MTX.[13,14] In vitro studies using human carcinoma lines have shown that optimal tumor toxicity is achieved with an 18–24-hour sequencing of MTX and 5-FU.[15] Therefore, the FAMTX regimen was modified to increase the interval between MTX and 5-FU from 1 hour, as originally designed, to 24 hours. Therefore, this Phase I study was designed in two phases. In the first phase, the maximally tolerated dose (MTD) of a modified, dose-intensive FAMTX regimen in which MTX and 5-FU were administered 24 hours apart, twice-monthly, without Ur rescue was defined. In the second phase, it was determined whether oral Ur ameliorated the toxicity of this modified FAMTX regimen when administered 2 hours or 24 hours after 5-FU.



Methotrexate (MTX): 1.5 gm/m2 - Day 1, 15
Fluorouracil (FU): 800 mg/m2 (escalating doses) - Day 2, 16
Leucovorin (LV): 15 mg/m2 po, q 6 hours x12 - Days 2-4, 16-18
Adriamycin (ADRIA): 30 mg/m2 - Day 16
Uridine (UR): 8.0 gm/m2, po, q 6 hours, x12 - Days 3-5, 17-19

FIGURE 1. Study design. Methotrexate (MTX) was administered on Day 1 to all patients at a fixed dose of 1.5 g/m^2. MTX was followed 24 hours later by escalating doses of 5-FU (Day 2) starting at 800 mg/m^2. Leucovorin rescue was administered 25 hours after MTX (Day 2) at a dose of 15 mg/m^2 orally every 6 hours for 12 doses. This combination was repeated every 15 days. Every other cycle patients also received 30 mg/m^2 of doxorubicin (the "Adria cycles"). Patients were administered oral uridine at a dose of 8 g/m^2 every 6 hours for 12 doses starting 24 hours after 5-FU (Days 3–5 and 17–19).

PATIENTS AND METHODS

Study Design

The study design shown in Figure 1 was utilized. Prehydration began the evening before treatment with 5% dextrose in water with 100 mEq sodium bicarbonate ($NaHCO_3$)/L at 150 mL per hour. Alternatively, oral hydration was performed. Two days prior to MTX therapy, patients were instructed to drink 2 L of fluid each day. Sodium bicarbonate ($NaHCO_3$, 3 tablets, 324 mEq/tablet) was given every 6 hours starting the day prior to treatment. MTX, 1.5 gm/m^2, was infused on Day 1 over 1 hour in 1 L of 5% dextrose in water with 50 mEq $NaHCO_3$/L. Prehydration fluids continued until 24 hours after MTX treatment. MTX was followed 24 hours later by escalating doses of bolus 5-FU (Day 2) starting at 800 mg/m^2. LV rescue was begun 25

hours after MTX (Day 2) at a dose of 15 mg/m^2 orally every 6 hours for 12 doses. This cycle was repeated every 15 days. However, to approximate the original FAMTX regimen of Klein et al., patients also received a 30 mg/m^2-bolus of doxorubicin (e.g., the "Adria cycles") with every other cycle. In this manner, doxorubicin was administered only with the second of the two monthly MTX and 5-FU therapies. Antiemetics, including lorazepam and prochlorperazine, were administered as needed.

Oral Ur tablets were supplied by the National Cancer Institute in 1-g tablets. Patients were administered oral Ur at a dose of 8 g/m^2 every 6 hours for 12 doses. To not break the tablets, the total Ur dose was rounded to the nearest whole number. There were three cohorts of patients. In the first cohort, patients did not receive Ur on their initial cycles. Toxicity was graded according to NCI cotoxicity criteria. If they developed Grade 3 or greater hematologic toxicity, they received Ur on all subsequent cycles starting 24 hours after 5-FU. In the second cohort, all patients received Ur 2 hours after 5-FU on all cycles on Days 2–4 and 16–18. An early analysis of the data with the 2-hour Ur rescue suggested that the Ur might be having an adverse effect on the anticipated response rate. In a patient population that predominantly included gastric carcinoma patients, a third cohort was opened. In the third cohort, all patients received Ur 24 hours after 5-FU on all cycles on Days 3–5 and 17–19. Five patients who were still being maintained on the 2-hour rescue were switched to the 24-hour rescue. These patients remained evaluable for response but not for toxicity.

At least three patients were treated at each dose level. The dose of 5-FU was escalated by 100 mg/m^2-increments in the phase of the study without Ur rescue and in 200 mg/m^2-increments with Ur rescue until toxicity was reached. If one-third of the patients experienced Grade 4 leukopenia, thrombocytopenia, or nonhematologic toxicity of any kind, three additional patients were treated at that level. The MTD was defined as the 5-FU dose level that produced Grade 3 toxicity or greater in two-third of the patients.

A complete response CR was defined as complete disappearance of all clinically evident disease for a minimum of 8 weeks. A partial remission (PR) was defined as decrease of 50% or greater in the sum of the product of the greatest dimensions of the measured lesions without simultaneous increase in the size or any lesion or appearance of new lesions. This regression had to exist for at least 1 month.

Patient Eligibility

Patients treated as part of this trial had to meet a series of eligibility criteria. All patients had to have been $\geq$18 years of age with histologically confirmed carcinoma by the Pathology Department of Memorial Hospital. Patients must have had an incurable cancer and a Karnofsky performance status (KPS) $\geq$ 60% with a life expectancy of at least 8 weeks. All patients must have had a leukocyte count $\geq$ 4000/μL and a platelet count $\geq$ 150,000/μL prior to starting therapy, serum bilirubin $\leq$ 1.5 mg/dL, serum creatinine $\leq$ 1.5 mg/dL, and normal heart function (New York Heart Association Class I–II with no prior history of congestive heart failure). Patients without measurable disease were allowed entry on this Phase I protocol although attempts were made to define measurable disease in all patients so that the therapeutic efficacy of the combination could be evaluated. Previously treated patients were accepted as long as they had received myelosuppressive chemotherapy within the previous 4 weeks (6 weeks for prior nitrosurea or mitomycin C). However, no patient who entered this study had received any prior treatment. The total dose of prior doxorubicin had to be less than 200 mg/m^2. They may not have received radiation therapy to major bone marrow-containing areas within the prior 4 weeks. Patients with large pleural effusions or ascites were excluded from the study. Informed consent indicating that patients were aware of the investigational nature of the treatment had to be obtained.

The pretreatment evaluation included a complete medical history and physical examination including documentation of all measurable disease. Pretreatment laboratory studies included a complete blood count (CBC), platelet count, differential, biochemical screening profile, serum creatinine, and chest X-ray. Radiologic tests were performed as clinically indicated. Pretreatment gated heart scan (MUGA) was indicated if there was a prior history of serious cardiac disease (e.g., myocardial infarction) and the ejection fraction had to be $\geq$45%.

Evaluations during the study included a CBC and platelet count weekly during the course of therapy. A history, physical exam, CBC, and serum creatinine were required prior to each treatment and a chest X-ray was obtained each month. Gated heart scan was required at 200 mg/m^2 and 400/mg/m^2 total doxorubicin dosages. Appropriate radiologic studies were repeated every 6 weeks to evaluate response. Patients were followed on study until disease progression.

Plasma Uridine Pharmacokinetics

In 7 patients who received Ur starting 24 hours after 5-FU, plasma samples were obtained for Ur levels. Samples were obtained pretreatment, 30 minutes after the first Ur dose, and then 1, 2, 3, 4, 6, 8, and 24 hours after the first Ur dose. Samples were stored at -70 °C

TABLE 1
Patient Characteristics on the Modified FAMTX Protocol with Uridine Rescue

Patients entered	57
Median age (yrs) (range)	58 (35–74)
Median KPS (range)	80 (70–100)
Male:female	40:17
Prior chemotherapy	0
Primary sites of disease	Stomach: 38
	Pancreas: 6
	Unknown primary: 6
	Gallbladder: 3
	Duodenum: 2
	Bile duct: 1
	Esophagus: 1

FAMTX: high dose 5-fluorouracil, doxorubicin, high dose methotrexate, and leucovorin, KPS: Karnofsky performance status.

until time of analysis. Samples were thawed to 37 °C in a water bath and 0.5 mL of plasma was added to 0.8 mL of 0.8 N $HClO_4$ (or 10% trichloracetic acid) in a microfuge tube. The sample was vortexed for 10 seconds and placed on ice for 10 minutes. The tube was centrifuged for 2 minutes (1200 × g) to pellet the precipitated protein. Approximately 0.4 mL of the supernatant fluid was removed and to this was added to 2 volumes of Freon/tri-n-octyl amine solution. This was centrifuged for 2 minutes and the top aqueous layer was withdrawn for high performance liquid chromatography (HPLC) analysis. HPLC was performed with Beckman Ultrasphere columns (Beckman, Irvine, CA) at 20 °C using a ultraviolet detection system at 260 nanometers and a gradient system of 100 nM ammonium acetate, pH 6.5, in 50% acetonitrile.

RESULTS

Patient Characteristics

As shown in Table 1 57 patients entered the study; 40 were male and 17 female. The median age of this group was 58 years (range, 35–74 years) and the median KPS was 80 (range, 70–100). In view of the known activity of standard FAMTX in gastric carcinoma, there was a preponderance of patients with gastric carcinoma entered into the study (38). Other primary sites of disease represented in this phase I trial included pancreas (six patients), unknown primary (six patients), gallbladder (three patients), duodenum (two patients), bile duct (one patient), and esophagus (one patient). No patient entered into this study had received prior chemotherapy.

Modified FAMTX Toxicity without Ur Rescue

The hematologic toxicity encountered in the first cohort of patients is summarized in Table 2. Two different nadirs are shown. On the non-Adria cycles with a 5-FU dose of 900 mg/m^2, the mean leukocyte count nadir was 4.4 × $10^3/\mu$L and the mean absolute neutrophil count (ANC) nadir was 2.4 × $10^3/\mu$L. However, on the midcycle treatments that included doxorubicin the mean leukocyte count nadir was 3.3 × $10^3/\mu$L and the mean ANC nadir was 1.2 × $10^3/\mu$L. With 900 mg/m^2 of 5-FU on the Adria-cycles, 2 patients experienced Grade 3 and 4 hematologic toxicity. For this reason, the MTD for 5-FU on the Adria cycles in the absence of Ur was defined as 900 mg/m^2.

Because the mean leukocyte count nadir at 900 mg/m^2 on the non-Adria cycles in the absence of Ur was 4.3 × $10^3/\mu$L, it was decided to continue the dose escalation of 5-FU on the non-Adria cycles only, whereas on the Adria cycles, 5-FU was fixed at 800 mg/m^2 without Ur. As shown in Table 2, in the absence of Ur, dose-limiting hematologic toxicity occurred at a 5-FU dose of 1.1 g/m^2. At this dose level, the mean leukocyte count nadir was 1.8 × $10^3/\mu$L, the mean ANC nadir was 0.4 × $10^3/\mu$L, and the mean platelet count nadir was 158 × $10^3/\mu$L. Two patients experienced Grade 4 and two patients experienced Grade 3 neutropenia, with two admissions for neutropenic fever. There was no dose-limiting nonhematologic toxicity. Two patients developed Grade 3 stomatitis. This was observed at both the 800 mg/m^2 and the 1100 mg/m^2 5-FU doses. One patient developed MTX toxicity with reversible Grade 4 nephrotoxicity. There was also one episode of Grade 3 fatigue. No patient developed hand-and-foot syndrome.

Table 3 summarizes the nadir counts of patients initially treated at the 1.1 g/m^2 5-FU level without Ur who experienced Grade 3 or greater hematologic toxicity and went on to receive Ur 24 hours after 5-FU in subsequent cycles. At this level, the 5-FU dose with the Adria cycles was fixed at 800 mg/m^2. As shown, with subsequent administration of oral Ur for individual patients, there was a dramatic increase in both the nadir leukocyte count and ANC. For example, patient 2, when treated with 1.1 g/m^2 of 5-FU without Ur, had leukocyte count and ANC nadirs of 2.6 × $10^3/\mu$L and 0.5 × $10^3/\mu$L, respectively. With the addition of Ur, this increased to 3.8 × $10^3/\mu$L and 1.7 × $10^3/\mu$L, respectively. Similar results were obtained for Patients 5 and 7. There was only a marginal benefit with Ur on the nadir counts of Patient 6. After their initial nadirs, Patients 3 and 4 elected to be taken off study and were not retreated with Ur. Therefore, a comparative analysis is not possible. Because of the significant leukocyte count nadirs with the first non-Adria cycle, almost all the patients were receiving Ur by the time they reached the first Adria cycle. Only two patients (1 and 2) had nadir counts that allowed them to pro-

TABLE 2
Hematologic and Nonhematologic Toxicity with Modified FAMTX without Uridine Mean Nadir Counts

5-FU mg/m^2	Adria, Yes/no	No. of patients	Leukocyte count (range) $\times 10^3/\mu$L	ANC (range) $\times 10^3/\mu$L	Platelets (range) $\times 10^3/\mu$L	% admissions for nadir fever	Grade 3/4 nonhematologic toxicity
800	No	4	4.9 (1-6.6)	3 (0.4-5.2)	147 (150-182)	25	0
800	Yes	3	3.9 (0.9-11.3)	1.3 (0.0-9.4)	154 (85-267)	33	0
900	No	5	4.4 (3.3-10.8)	2.4 (1.6-8.4)	179 (120-366)	20	0
900[a]	Yes	5	1.6 (0.3-5.7)	1.2 (0.1-5.2)	111 (74-234)	20	0
1000	No	4	3.3 (2-3.6)	1.5 (0.3-2.6)	120 (33-127)	0	Stomatitis: 1
1100[b]	No	7	1.8 (0.3-2.7)	0.4 (0-2.6)	158 (18-210)	28	Renal: 1

FAMTX: high dose 5-fluorouracil, doxorubicin, high dose methotrexate, and leucovorin; 5-FU: 5-fluorouracil; Adria: doxorubicin (Adriamycin) cycles; ANC: absolute neutrophil count.
[a] Maximally tolerated dose of 5-fluorouracil (5-FU) on Adria cycles; 5-FU dose fixed at 800 mg/m^2 in all subsequent Adria cycles.
[b] Maximally tolerated dose of 5-FU without Adria.

TABLE 3
Mean Nadir Counts in the Same Patient Treated with or without Uridine on Subsequent Cycles at the 1100 mg/m^2 5-FU Level of Modified FAMTX

Patient 5-FU	(mg/m^2)	Uridine (Yes/no)	Leukocyte count $\times 10^3/\mu$L	ANC $\times 10^3/\mu$L	Platelets $\times 10^3/\mu$L
A[a]	800	No	1.8	0.2	190
A	800	Yes	11.4	7.4	262
B	800	No	3.9	1.4	213
B	800	Yes	9.6	6.2	119
B	1100	No	2.6	0.5	127
B	1100	Yes	3.8	1.7	173
E[b,c]	1100	No	1	0.2	190
E	1100	Yes	2.7	0.7	210
F[c]	1100	No	2	0.5	207
F	1100	Yes	2.5	0.6	172
G[c]	1100	No	1.6	0.2	167
G	1100	Yes	5	2.5	190

5-FU: 5-fluorouracil: FAMTX: high dose 5-fluorouracil, doxorubicin, high dose methotrexate, and leucovorin; ANC: absolute neutrophil count.
[a] Pre- and posturidine comparison only possible for 800 mg/m^2 5-fluorouracil dose, which was fixed at this level on the Adria cycles.
[b] Patients C and D were never treated with uridine.
[c] Pre- and posturidine comparison only possible for 1100 mg/m^2 5-fluorouracil dose.

ceed to the first cycle without concomitant Ur. Both appeared to benefit from the addition of Ur when treated with 800 mg/m^2 5-FU dose on the Adria cycles.

Modified FAMTX Toxicity with Ur Rescue

The hematologic toxicity with Ur starting 24 hours after 5-FU is shown in Table 4. In this phase of the study, there were two different nadirs. With the escalation of 5-FU to 1.2 g/m^2 on the non-Adria cycles, the mean leukocyte count nadir was 2.9 $\times 10^3/\mu$L with a mean ANC nadir of 0.8 $\times 10^3/\mu$L. On the Adria cycles, the mean leukocyte count nadir was 1.4 $\times 10^3/\mu$L with a mean ANC nadir of 0.2 $\times 10^3/\mu$L. This included two patients with Grade 4 leukopenia. There was also an effect on the platelets, especially on the Adria cycles, with a mean platelet nadir of 88 $\times 10^3/\mu$L. Therefore, the MTD of 5-FU on the Adria cycles with Ur administered 24 hours after 5-FU was defined as 1.2 g/m^2. Identical results were obtained with Ur rescue starting 2 hours after FU (data not shown).

On subsequent cycles, it was decided to keep the 5-FU dose fixed at 1 g/m^2 on Adria cycles, but to continue the dose escalation with 5-FU on the non-Adria cycles. As shown in Table 4, at a 5-FU dose of 1.4 g/m^2 the mean leukocyte count nadir was 1.7 $\times 10^3/\mu$L and the mean ANC nadir was 0.3 $\times 10^3/\mu$L. Because the admission rate for neutropenic fever at this level was only 14% and only 1 patient experienced Grade 4 leukopenia, it was decided to escalate the 5-FU dose level of 1.6 g/m^2. At this dose, the mean leukocyte count nadir was 1.1 $\times 10^3/\mu$L and the mean ANC nadir was 0.2 $\times 10^3/\mu$L. Three of the 6 patients treated at

TABLE 4
Hematologic and Non-Hematologic Toxicity with Modified FAMTX and 24-Hour Uridine Rescue (Mean Nadir Counts)

5-FU g/m^2	Adria Yes/no	No. of patients	Leukocyte count (range) $\times 10^3/\mu L$	ANC (range) $\times 10^3/\mu L$	Platelets (range) $\times 10^3/\mu L$	% admissions for nadir fever	Grade 3/4 non-hematologic toxicity
1	No	4	3.9 (1.2-5.3)	2.3 (0.1-3.9)	197 (134-384)	25	0
1	Yes	4	1.6 (0.5-3.6)	0.2 (0-1.6)	136 (54-293)	0	0
1.2	No	7	2.9 (1.7-4)	0.8 (0.1-1.5)	136 (59-547)	14	Alopecia: 1
1.2[a]	Yes	7	1.4 (0.3-2)	0.2 (0-0.6)	88 (14-199)	14	0
1.4	No	7	1.7 (0.8-6.7)	0.3 (0-3.4)	157 (29-243)	14	Renal: 1
1.6[b]	No	6	1.1 (1-3.9)	0.2 (0-1.2)	110 (35-250)	33	Stomatitis: 1

FAMTX: high dose 5-fluorouracil, doxorubicin, high dose methotrexate, and leucovorin; 5-FU: 5-fluorouracil; Adria: doxorubicin (Adriamycin) cycles; ANC: absolute neutrophil count.
[a] Maximally tolerated dose of 5-fluorouracil (5-FU) with Adria, 5-FU dose fixed at 1 g/m^2 in all subsequent Adria cycles.
[b] Maximally tolerated dose of 5-fluorouracil without Adria.

this 5-FU level had leukocyte count nadirs of 1 and 2 of these 3 were admitted for neutropenic fever for an overall neutropenic admission rate of 33%. Therefore, a 5-FU dose of 1.6 g/m^2 was determined to be the MTD of 5-FU when administered 24 hours after 1.5 g/m^2 of MTX and with 8 g/m^2 of Ur administered every 6 hours starting 24 hours after 5-FU.

The nonhematologic toxicities for the 24-hour Ur rescue are divided into 5-FU dose levels observed on either the non-Adria or Adria cycles. As shown in Table 4, except for infrequent Grade 3 toxicity for stomatitis, there was no dose-limiting nonhematologic toxicity. The nonhematologic toxicities were generally mild (Grade 2 toxicity or less) and most of these were gastrointestinal in origin (i.e., diarrhea, nausea, and vomitting). One patient developed MTX toxicity associated with reversible renal failure. No patient developed significant skin toxicity.

Ur Pharmacokinetics

To demonstrate adequate Ur levels, Ur pharmacokinetics were obtained on 7 patients for 8 g/m^2 of oral Ur starting 24 hours after 5-FU. The mean peak concentration after the first dose of Ur was 54.2 $\pm$ 9.2 μmol. This was achieved 2 to 3 hours after administration. The mean through concentration, which was obtained immediately prior to the second Ur dose or 6 hours after the first, was 42.9 $\pm$ 18.7 μmol.

Clinical Response

The response data by dose level for the 24-hour rescue schedule is summarized in Table 5. In this phase of the study, there were 20 evaluable patients with 5 PRs and 2 CRs (overall response rate of 35%). Six of these patients had metastatic gastric carcinoma. One of the two CR patients had metastatic adenocarcinoma of unknown primary with a large peripancreatic mass. This CR was surgically confirmed and the patient remained disease free 2 years after completion of chemotherapy. The other CR lasted 11.9 months in a patient with gastric carcinoma who had multiple lung metastases.

TABLE 5
Response on the Modified FAMTX Protocol with 24-Hour Uridine Rescue

5-FU (g/m^2)	No. of evaluable patients	Response (tumor type)
1	3	1 CR[a] (gastric) 2 PR (gastric)
1.2	6	1 PR (gastric) 1 CR (gastric)
1.4	5	1 PR (gastric)
1.6	6	1 PR (gastric) 1 CR (unknown primary)

FAMTX: high dose 5-fluorouracil, doxorubicin, high dose methotrexate, and leucovorin; 5-FU: 5-fluorouracil; CR: complete response; PR: partial response.
[a] Patient initially treated with 2-hour uridine rescue without a major response. When switched to 24-hour uridine rescue, the patient achieved a complete response.

When Ur was administered 2 hours after 5-FU, there were no major responses in 9 evaluable patients. However, 1 patient with gastric carcinoma who started on the 2-hour rescue converted from stable disease to a CR that was durable for 9 months when changed to the 24-hour rescue schedule.

CONCLUSIONS

The goal of this Phase I study was to intensify the dose of 5-FU each month by administering bimonthly MTX and 5-FU with oral Ur rescue as part of a modified FAMTX regimen. In the original FAMTX, MTX and 5-FU were administered 1 hour apart.[1] The FAMTX regimen was modified in accordance with in vitro data indicating that a 24-hour interval between MTX and

5-FU produced a superior antitumor effect.[15] Clinical trials that have employed high dose MTX followed by LV rescue with a 20–24 hour interval between MTX and 5-FU had shown acceptable toxicity.[16–18] Because this combination on this schedule had never been tested in patients before, the first phase of this study was designed to test the impact of the 24-hour dosing interval on the toxicity of the FAMTX regimen when given bimonthly. The second phase of the study was to determine whether oral Ur would ameliorate the toxicity of the modified FAMTX regimen when given either 2 hours or 24 hours after 5-FU administration.

In the first cohort of patients, it was shown that when the interval between 5-FU and MTX increased to 24 hours in the absence of Ur rescue, the MTD of FU was 1.1 g/m^2 on non-Adria cycles and 900 mg/m^2 on Adria cycles. This represents a total 5-FU dose of 2 g/m^2/month. Most of the patients on Cohort 1 who subsequently received oral Ur 24 hours after MTX and 5-FU exhibited a substantial increase in their subsequent leukocyte count nadirs.

For patients in the second phase of the study, an increase in the MTD for the non-Adria cycles with 24-hour Ur rescue was demonstrated. On non-Adria cycles, the MTD of 5-FU is 1.6 g/m^2, whereas on the Adria cycles the MTD of 5-FU is 1.2 g/m^2. Therefore, the total 5-FU dose is 2.8 g/m^2/month with Ur rescue. This represents a 40% increase in the MTD for the total dose of monthly 5-FU administered when compared with the same schedule without Ur. A Phase II 5-FU dose of 1.4 g/m^2 on the non-Adria cycles and 1 g/m^2 on the Adria cycles when administered in combination with oral Ur rescue can be recommended.

The total oral Ur dose, as well as the timing of Ur administration relative to 5-FU, appear to be critical determinants in terms of rescuing effects. The 8 g/m^2 oral Ur dose was selected based on clinical studies performed by van Groeningen et al.[10] This group showed that absorption of Ur from the gastrointestinal tract is saturated at dose levels of 8 g/m^2 and higher doses do not result in increased peak levels. The 8 g/m^2 dose, even with multiple dosing, was associated with only minimal diarrhea.[10] From the in vivo studies, it was predicted that Ur administration starting 2 hours after 5-FU would protect the host but not protect the tumor.[8] Eleven of the 12 patients treated with Ur rescue 2 hours after 5-FU had gastric carcinoma (0 of 11 responses, 95% confidence interval response rate, 0–22%). From published studies of standard FAMTX, a major response rate of at least 30% was anticipated.[3] However, on the 2-hour rescue schedule there were no major responses up to an 5-FU dose of 1.2 g/m^2. It was hypothesized that Ur, when administered 2 hours after 5-FU, was blocking the therapeutic effect of the 5-FU against the tumor, as well as its systemic cytotoxicity.

When Ur was subsequently administered at the same dose (8 g/m^2 every 6 hours for 12 total doses) starting 24 hours after the modified FAMTX, a change in response rate was noted with the first cohort of patients at the 5-FU dose of 1 g/m^2. At this dose level, all three evaluable patients had gastric carcinoma and two of these three patients had PRs. Therefore, it was suspected that the 2-hour interval between 5-FU and oral Ur blocks 5-FU's anticancer effect and that a 24-hour interval is at least sufficient to ensure rescue from 5-FU-induced hematologic toxicity without adversely impacting on tumor response.

The majority of patients who were analyzed for Ur levels after their first dose of 8 g/m^2 oral Ur were able to achieve peak Ur levels 2 to 3 hours later of approximately 50 μM. These serum levels are comparable to those reported by van Groeningen et al. with an identical 8 g/m^2 dose.[10] From the preclinical studies, the 50 μM concentration appeared to be a serum threshold that was necessary to obtain protection from 5-FU toxicity.[8] However, the trough Ur levels, which were obtained immediately prior to the second Ur dose at 6 hours, generally fell below the 50 μM level. This would indicate that 8 g/m^2 of oral Ur on an every-6-hour schedule may not be sufficient to maintain serum Ur levels that will provide sustained protection from 5-FU. Alternative approaches may be to administer the Ur tablets more frequently (i.e., every 4 hours) or to use derivatives of Ur (i.e., triacetyluridine) that have been shown to provide levels of more than 50 μM for up to 6 hours.[19]

When tested with a larger cohort of patients, Ur administration allowed for an approximately 40% increase in the MTD for the total amount of 5-FU administered each month when compared with the same 5-FU schedule without Ur. The administration of doxorubicin with 5-FU and MTX negatively impacted on the dose intensification of 5-FU with Ur and removal of doxorubicin from this bimonthly program is being considered in future clinical trials. These studies indicate the use of oral Ur will result in a significant increase in the amount of 5-FU that can be administered as part of this modified, dose-intensive FAMTX regimen.

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