Phase I and Pharmacokinetic Study of a Micronized Formulation of Carboxyamidotriazole, a Calcium Signal Transduction Inhibitor: Toxicity, Bioavailability and the Effect of Food

Jordan Berlin, Kendra D. Tutsch, Rhoda Z. Arzooomanian, Dona Alberti, Kim Binger, Chris Feierabend, Amy Dresen, Rebecca Marnocha, James Pluda, and George Wilding

University of Wisconsin Comprehensive Cancer Center, Madison, Wisconsin 53792 [J. B., K. D. T., R. Z. A., D. A., K. B., C. F., A. D., R. M., G. W.], and Medicine Branch, Division of Clinical Sciences, National Cancer Institute, NIH, Bethesda, Maryland 20892 [J. P.]

Abstract

Purpose: This Phase I study was conducted to evaluate the toxicity profile and determine the maximum tolerated dose (MTD) of an oral micronized formulation of the signal transduction inhibitor carboxyamidotriazole (CAI). Bioavailability of the micronized formulation relative to a gelatin capsule (gelcap) formulation was assessed. The effects of food intake and timing on CAI steady-state plasma concentrations (C_{ss}) were also investigated.

Experimental Design: Patients received continuous daily CAI (28-day cycles). Starting dose was 150 mg/m² daily and escalations were by 50 mg/m² increments. The first three patients enrolled were given test doses of the original gelcap formulation and two different micronized formulations to determine relative bioavailability. Toxicity and pharmacokinetic assessments were performed weekly. Additional cohorts were added after MTD determination to assess the effect of food intake and duration of fast on CAI C_{ss}.

Results: The micronized formulation was absorbed more slowly than the gelcap formulation. Twenty-nine patients were enrolled in the dose-escalation portion of the study. After dose escalation to 300 mg/m², dose-limiting neurotoxicities occurred including reversible vision loss in two patients. Other toxicities were mild. The final MTD was 150 mg/m². Pharmacokinetics appeared linear with significant inter- and intrapatient variability. Patients with C_{ss} of ≥ 4.0 mg/liter were more likely to have neurotoxicity. Nine patients with renal cell cancer and one with hepatocellular cancer had prolonged stable disease. CAI plasma concentrations were higher when taken with food.

Conclusions: Micronized CAI was well tolerated at the MTD of 150 mg/m². Higher doses were limited by significant neurotoxicity. The variability in CAI pharmacokinetics may be partially attributable to concomitant food intake and timing of the dose.

INTRODUCTION

CAI (NSC 609974) is an oral agent that has been shown in laboratory studies to inhibit tumor growth and metastasis (1–4). Its antitumor activity appears to be mediated through inhibition of calcium influx. Downstream effects of calcium influx antagonism are varied but appear to possibly include angiogenesis (5, 6). The UW reported previously a Phase I trial analyzing a PEG-400 gelcap formulation of CAI. In that study, DLT was neurocerebellar toxicity (7). Below the MTD, neurologic toxicity was minimal and was not dose limiting. Instead, the most common side effects at these levels were nausea, vomiting, and fatigue. At the MTD dose for the gelcap, 14 of 20 patients experienced grade 1–2 nausea and/or vomiting with as many as three pharmacological manipulations required for alleviation of this toxicity. One patient refused additional drug because of nausea. Fatigue occurred in a total of 19 of the 29 patients enrolled on study. In the gelcap Phase I trial, a switch to nighttime administration appeared to reduce the symptoms of nausea, vomiting, and fatigue. A second study of the gelcap formulation reported no dose-limiting cerebellar toxicity (8). In that study, chronic nausea and vomiting was “compliance-limiting” although nighttime administration was also reported helpful in controlling these symptoms. Both Phase I studies of gelcap CAI demonstrated variability in the pharmacokinetics of CAI. For patients at all levels on the UW gelcap study, the mean intrapatient variation in weekly C_{ss} was 12.4% ± 5.3%.

In an attempt to try to reduce the GI toxicity and the pk variability, newer formulations for oral CAI were developed for testing. The purpose of the study reported here was 2-fold: to evaluate the relative bioavailability of two micronized formulations of CAI compared with the gelcap formulation and to perform a Phase I trial with pharmacokinetics on one of the two
formulations. The objectives of the Phase I portion of this trial were to evaluate and observe for side effects and establish the MTD for micronized formulation CAI.

After completion and MTD determination of the standard Phase I trial of micronized CAI, two supplemental studies were also done to additionally investigate the pharmacokinetics and potential dosing regimens for micronized CAI. The first supplemental study was designed to evaluate the effects of a bedtime snack on plasma $C_{ss}$ of CAI. The dose of CAI chosen for this study was the MTD from our Phase I study as discussed below. A second supplemental study was designed to look at the feasibility of a fixed-dose for CAI rather than dosing based on body surface area. At the same time, because preliminary data indicated that food intake affects CAI steady-state levels (9), the fixed-dose study was also designed to evaluate the effects of duration of fast on CAI $C_{ss}$ levels and toxicity profile. The dose chosen for this trial was 400 mg/day, which approximated 225 mg/m$^2$/day in an average patient. This is midway between the MTD established by this UW Phase I trial and that established by the NCI trial for micronized CAI (10).

**PATIENTS AND METHODS**

**Patient Selection.** After approval by the UW Human Subjects Committee (Institutional Review Board), eligible patients were enrolled in the study. No patient was allowed to enroll in more than one portion of the study. No intrapatient dose escalations were allowed.

Eligible patients included age ≥ 18 years with Eastern Cooperative Oncology Group performance status of 0, 1, or 2 and ability to provide informed consent. Patients must have had advanced malignancy (solid tumor or lymphoma) refractory to known forms of effective therapy but without history of brain metastases. Patients were to have had adequate organ function including WBC ≥ 4,000/mm$^3$, absolute neutrophil count ≥ 1,500/mm$^3$, platelet ≥ 100,000/mm$^3$, bilirubin ≤ 1.5 mg/dl, aspartate aminotransferase ≤ 2 times normal, serum creatinine ≤1.5 mg/dl for females, creatinine ≤ 1.8 mg/dl for males, and serum calcium ≤ 11.0 mg/dl. Because of neurotoxicity observed previously, patients with history of peripheral neuropathy were excluded. In addition, patients taking calcium channel blocking antihypertensives unable to be switched to alternate medications were also excluded from study.

**Drug Formulation.** The NCI, Division of Cancer Treatment and Diagnosis, provided all of the formulations of CAI. Micronized CAI formulations were supplied in a capsule containing 50 mg of CAI and microcrystalline cellulose. Gelcap CAI used for the bioavailability study was provided in 50-mg capsules containing CAI dissolved in PEG-400.

**Treatment Plan.** Because this was an oral agent available as 50-mg capsules, rounding of dosages was required. Therefore, for all portions of this study patients had their doses rounded down to the next lowest 50-mg increment. Actual, rather than ideal, body weight was used to calculate body surface area. All of the patients were asked to keep an administration diary, and pill counts were performed by the nurses to monitor compliance. Patients self-administered CAI during the evening except as noted below.

There were two groups of patients initially enrolled for evaluation of relative bioavailability of micronized CAI. Group 1 consisted of six patients given a single dose (75 mg/m$^2$) of a micronized formulation designated as “A,” then after a 1-week washout period started on daily dosing of the gelcap formulation. The toxicity information for this group was presented in our previous paper on the chronic administration of gelcap CAI (7), and bioavailability data only on these patients is included in this report. Group 2 consisted of three patients each administered single doses of gelcap formulation, micronized “A” formulation, and a second micronized formulation designated as “B,” 1 week apart, all at a dose of 75 mg/m$^2$. Subsequently, the members of group 2 were started on daily administration of micronized B formulation of CAI at the designated starting daily dose for the Phase I trial, 150 mg/m$^2$. These three patients served as the members of the first dose level for the present Phase I study.

The Phase I portion of this study consisted of daily administration of micronized B CAI at doses of 150, 200, 250, and 300 mg/m$^2$. Dose escalation was conducted on a standard design in which three patients were enrolled per cohort. If no DLTs were seen then the next cohort would be entered at the next higher level. If one of three patients experienced DLT, a second cohort of three patients was enrolled. If at any dose level, two or more DLTs were observed, then the MTD was exceeded, and the previous dose level was considered the MTD. Study design called for additional enrollment at the MTD to obtain pk and toxicity data.

**Evaluation of the Effect of Food.** After determination of the MTD for micronized CAI as discussed below, patients were enrolled in one of two subsequent supplemental studies conducted in series. The first, termed the “snack study,” was designed to assess the effects of food versus fasting on pharmacokinetic parameters of oral CAI. Two cohorts of six evaluable patients were enrolled in a nonrandomized fashion, all receiving CAI at a dose of 150 mg/m$^2$. Patients in the first cohort of six were asked to administer their CAI pills for the first 28 days at bedtime along with a bedtime snack of ~300 calories and ~50% fat composition. Patients could not have any food or liquid other than water in the 2 h before the snack and no food after the prescribed snack. Several examples of snacks that met these criteria were provided to the patients before enrolling on study. During the second 28-day cycle, patients were required to fast for at least 2 h before bedtime administration of CAI with no food or liquid except water. The second cohort of six patients was scheduled to reverse the order of the 2 cycles, fasting for the first cycle and coadministering a snack with the CAI for the second cycle. Because of dietary requirements in this study, patients unable to tolerate fasting periods required by the study (such as brittle diabetics) were excluded.

The second supplemental study, termed the “fixed-dose study,” was designed to enroll a single cohort of six patients to receive micronized CAI at a fixed dose of 400 mg not based on body surface area. This dose was chosen after discussion with the NCI and evaluation of the results of this Phase I trial as well as the NCI Phase I trial (10). Although this dose exceeded the MTD proposed by the UW, it was well below that proposed by the NCI trial. Patients enrolled in this trial were assigned to one of two administration schedules for the first cycle, and then they were to be crossed over to the other schedule for the second
cycle. This study was designed to assess the feasibility of this fixed dose and the effects of duration of fast on the pharmacokinetics of oral CAI. The first administration schedule was an overnight fast before CAI administration on an empty stomach. The other administration schedule was a 2-h fast after the evening meal with administration of CAI also on an empty stomach. No food or liquid except water was allowed for at least 1 h after CAI administration.

**Pharmacokinetic Assessment.** For the patients enrolled in the bioavailability studies, plasma samples were obtained over a 48-h period after morning administration of each formulation of CAI. Relative bioavailability of the micronized formulation compared with the gelcap formulation of CAI was determined by comparison of the 24-h AUC values.

All of the patients subsequently receiving daily dosing of the micronized B formulation on the Phase I trial had the single-dose AUC determined after the first dose and plasma concentrations assessed weekly for the first month and then monthly. These samples were drawn ~12 h after CAI administration. Cohorts in the subsequent snack study and fixed-dose study had plasma samples drawn twice weekly after day 8. CAI assay was by reverse Phase high-performance liquid chromatography and has been described previously (7). All of the samples were assayed in duplicate. Intraday variability of the assay was <5%, and interday variability of a control patient sample was 2.6% over 6 weeks. Standard CAI for the assay was supplied by the NCI Drug Synthesis and Chemistry Branch.

As in the previous study (7) the single-dose parameters, $C_{max}$ and $t_{max}$ were model-independent and determined by inspection. AUC was determined by linear trapezoidal rule, and the $t_{1/2}$ was assessed by linear regression of the terminal portion of the concentration-time curve. $C_{ss}$ values reported here represent the mean of all concentrations obtained after day 8. $C_{ss}$ at steady-state was calculated as dose/24 h/$C_{ss}$. Terminal $t_{1/2}$ was also determined for patients who had samples drawn after CAI administration was discontinued.

**RESULTS**

**Bioavailability Study.** The micronized formulation of CAI did alter the absorption characteristics of the drug with a relatively lower bioavailability and slower absorption than the gelcap formulation studied previously (7). The nine patients enrolled in groups 1 and 2 for the bioavailability study demonstrated that the relative bioavailability for a single dose of the micronized A formulation compared with gelcap was 23.9% ± 13%. The single dose $C_{max}$ for micronized CAI was 20.6% ± 8.5% of the comparable gelcap dose. Additionally, the data from the three patients in group 2 who received both a single dose of micronized A and a single dose of micronized B indicated that the bioavailability of B was similar to that of A (90% ± 21%). Micronized B was used for the Phase I trial and the subsequent supplemental studies. Overall, the $t_{max}$ for a single-dose of the micronized formulation was 10.8 ± 8.0 h, compared with 2.4 ± 1.5 h for the gelcap formulation.

**Phase I Trial.** There were 29 patients enrolled in the Phase I portion of this study, including the three members of group 2 of the bioavailability study who received daily micronized CAI dosing at 150 mg/m². Of the 29 patients, 1 patient was not evaluable for either toxicity or MTD determination because of intracranial hemorrhage secondary to newly diagnosed brain metastases. This was determined within days of starting CAI and was not drug-related; therefore, this patient was not included in the analysis. Demographics for the remaining 28 patients are listed in Table 1. Of the remaining 28 patients, 3 were not considered evaluable for MTD determination because they discontinued drug before completing the first 28-day cycle without experiencing DLT; however, their data are included in the toxicity analysis. Two of the 3 patients were removed from study because of disease progression, and the third had rapid disease progression with death attributed to disease while on study. The 28 patients included in this analysis received ≥ 76 full or partial 28-day cycles of CAI with a median of two cycles administered.

Dose escalation on this trial initially went well with cohorts non-experiencing DLT at the 150, 200, and 250 mg/m² levels. However, at 300 mg/m², dose-limiting neurocortical toxicity was seen in 2 of 6 patients. Subsequent dose de-escalation ensued. Table 2 lists the progression of study drug dose escalation and de-escalation, and this illustrates how this study arrived at an MTD of 150 mg/m² for the micronized formulation of CAI. All of the DLTs listed in Table 2 were neurological. Table 3 lists all of the neurological toxicity observed in this trial. Although no dose-limiting cerebellar toxicity was seen in this study, visual changes occurred in 2 patients. The first patient

**Table 1** Phase I study: demographic data on 28 patients evaluable for toxicity data

<table>
<thead>
<tr>
<th>Data</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>Median age (range)</td>
<td>61.5 years (23–81)</td>
</tr>
<tr>
<td>No. of patients</td>
<td>28</td>
</tr>
<tr>
<td>Performance status</td>
<td>0/10</td>
</tr>
<tr>
<td></td>
<td>1/14</td>
</tr>
<tr>
<td></td>
<td>2/4</td>
</tr>
<tr>
<td>Male:Female</td>
<td>17:11</td>
</tr>
<tr>
<td>Primary tumor type</td>
<td></td>
</tr>
<tr>
<td>Colorectal</td>
<td>9</td>
</tr>
<tr>
<td>Kidney</td>
<td>8</td>
</tr>
<tr>
<td>Sarcoma</td>
<td>2</td>
</tr>
<tr>
<td>Ovarian</td>
<td>2</td>
</tr>
<tr>
<td>Prostate</td>
<td>2</td>
</tr>
<tr>
<td>Thyroid</td>
<td>1</td>
</tr>
<tr>
<td>Head and neck</td>
<td>1</td>
</tr>
<tr>
<td>Lung</td>
<td>1</td>
</tr>
<tr>
<td>Liver</td>
<td>1</td>
</tr>
<tr>
<td>Unknown primary</td>
<td>1</td>
</tr>
<tr>
<td>No. of patients with</td>
<td></td>
</tr>
<tr>
<td>previous exposure</td>
<td></td>
</tr>
<tr>
<td>to taxane, vinca</td>
<td></td>
</tr>
<tr>
<td>alkaloid, or platinum</td>
<td></td>
</tr>
<tr>
<td>regimen</td>
<td></td>
</tr>
<tr>
<td>150 mg/m² ($n = 6$)</td>
<td>0</td>
</tr>
<tr>
<td>200 mg/m² ($n = 8$)</td>
<td>3</td>
</tr>
<tr>
<td>250 mg/m² ($n = 6$)</td>
<td>1</td>
</tr>
<tr>
<td>300 mg/m² ($n = 8$)</td>
<td>3</td>
</tr>
</tbody>
</table>
had been on CAI for 194 days when a near complete loss of bilateral vision was noted. This patient had a recent eye exam in preparation for a left cataract removal performed 1 week before developing toxicity. Loss of visual acuity on the right from 20/25 to 20/250 was confirmed by a second ophthalmologist. MRI of the head was normal, but electroretinogram showed a central visual defect on the left. Western blots of serum and retinal homogenate were tested against the patient’s serum and did not demonstrate a paraneoplastic process. CAI was discontinued, and a course of prednisone was initiated. Eleven days after discontinuing CAI, visual acuity on the right had returned to 20/40. Rechallenge was not performed, and the patient returned to baseline without recurrence of symptoms after discontinuation of prednisone. A second patient at the same dose level developed neurovision toxicity on day 41 of CAI administration. At that time, the patient reported blurring of vision over the last “several” days before evaluation until outlines and shapes were visible but no details. There was no recent eye examination available for baseline comparison, but on day 41, acuity was recorded at 20/400 on the right and 20/200 on the left. CAI was discontinued, and 14 days later visual acuity was noted to be 20/30 and 20/20 (right and left, respectively). At that time, color vision was tested, and the patient missed 6 of 14 plates. Two weeks later, the patient was able to successfully identify 14 of 14 plates. With the patient’s consent she was rechallenged on two subsequent occasions at successively lower doses (200 mg/m² and 150 mg/m², respectively). Both attempts resulted in bilateral reduced visual acuity and color vision with complete resolution on withdrawal from CAI. Subsequent episodes were not as severe as the first.

Other than neurotoxicity, side effects were generally mild. GI toxicity consisting of nausea and/or vomiting and hematological toxicity were tolerable (Tables 4 and 5). No patients were withdrawn from the study because of GI intolerance of CAI. Fatigue was also seen at all of the dose levels but does not have a grade on the version of the common toxicity criteria used for this study. Mild to moderate fatigue was reported in 3 of 6 patients at 150 mg/m², 3 of 8 patients at 200 mg/m², 5 of 6 patients at the 250 mg/m², and 2 of 8 patients at the 300 mg/m². An additional 2 of the 8 patients treated at 300 mg/m² had severe fatigue, both falling asleep during work (painting a home and standing at a sales counter). Other clinical toxicities were mild including grade 1 alopecia in 1 patient, grade 1 constipation in 2 patients, grade 1 diarrhea in 1 patient, and grade 1 gastritis in 1 patient. Mild laboratory toxicities occurred in a few patients, including grade 1 bilirubin in 1 patient, grade 1 alkaline phosphatase in 1 patient, and grade 1 creatinine in 1 patient.

### Pharmacokinetics

Table 6 highlights the pk measurements from this trial. Terminal $t_{1/2}$ of CAI after cessation of daily dosing was $64 \pm 19$ h as derived from 14 patients from which the samples were drawn after discontinuing CAI. There was significant variability in the $C_{ss}$ values on this study with mean intrapatient coefficient of variation of $14 \pm 8\%$ and mean interpatient coefficient of variation of $33 \pm 10\%$. There was also substantial variability in CI/F with an overall mean of $5.45 \pm 2.7$ liter/hr. Fig. 1 shows the pattern of $C_{ss}$ for one patient receiving micronized CAI for 14 cycles. Clearance of CAI did not change significantly over time. Ten patients received the same dose of CAI for at least 3 months, and there was no statistical difference in the day 15 versus the month 3 $C_{ss}$ values ($P = 0.402$ by paired $t$ test). Day 1 AUC increased with increasing CAI dose to a limited extent over the small dose range studied ($P = 0.048$). Eventual $C_{ss}$ increased significantly with dose ($P = 0.0049$; Fig. 2). Neither AUC nor $C_{ss}$ on day 1 predicted for eventual $C_{ss}$.

Evaluating all of the patients in our trial with neurotoxicity, there is no relationship between AUC, single dose $C_{ss}$, or duration of therapy with the appearance of neurotoxicity. There was an apparent increase in the frequency of neurological side effects in patients with mean $C_{ss} \geq 4.0$ mg/liter (Fig. 3). Six of 9 patients with $C_{ss} > 4$ mg/liter had grade 2 or 3 neurotoxicity, whereas only 3 of 18 patients with $C_{ss}$ below 4 had more than grade 1 neurotoxicity. One of the 28 patients was not evaluable for toxicity.

**Snack Study.** In comparing the data from this Phase I trial to the NCI micronized trial, pk data were fairly similar, but the MTD was different (10). The major difference in the conduct of the two studies rested in the timing of meals before administration of oral CAI. Whereas this Phase I study did not control timing of administration in regard to meals or snacks, the NCI trial administered CAI in the morning with a previous overnight fast. CAI is a lipophilic compound, and theoretically absorption may be affected by dietary intake. This supplemental study was designed to evaluate the effects of a bedtime snack on $C_{ss}$ levels of CAI using the MTD from this trial.

A total of 17 patients were enrolled. Two patients were not evaluable and are not included in the analysis. The first developed an ileus, grade 2 neurocortical changes, and grade 1 neuromotor changes while on study. The symptoms of the patient did not resolve with discontinuation of CAI and CT scan revealed numerous brain metastases making the etiology of symptoms unclear. The second also had neurotoxicity with grade 3 neurovision changes, grade 1 neurocerebellar and grade 1 neuromotor toxicity, but had received the wrong formulation of CAI (gelcap formulation); therefore, this patient was not evaluable for purposes of this trial. Toxicities in both patients reversed to baseline after withdrawal from CAI.

The demographic data for the remaining 15 patients are listed in Table 7. Four patients did not complete two cycles at full dose. One each discontinued CAI during cycle 1 because of nausea and neuromod toxicity, respectively. The third had grade 2 neurosensory toxicity necessitating temporary discon-
continuation of CAI followed by dose reduction. A fourth went off study after one course because of progressive disease. Table 8 lists the toxicities observed in the snack study during cycles 1 and 2 only. It shows a fairly similar distribution of the toxicities seen when patients administered CAI either fasting or with snack. Toxicity after cycle 2 was mild. There was 1 additional patient who developed grade 2 neurosensory toxicity and another patient with grade 1 neurosensory. Other side effects included mild to moderate fatigue in 7 patients.

Of these 15 patients, 4 did not have enough time points on both cycles to evaluate the effects of snack on the $C_{SS}$ of CAI. Pharmacokinetic analyses in the 11 remaining patients demonstrated that $C_{SS}$ levels were higher when patients took the drug with food than without food. With a snack, $C_{SS}$ was $4.51 \pm 2.21$ mg/liter versus $3.72 \pm 2.21$ mg/liter without a snack ($P = 0.021$). Thus, $C_{SS}$ levels with snack were 128% of those without a snack. However, when these $C_{SS}$ values are compared with those for the initial cohort at 150 mg/m$^2$ (Table 6) there is no statistical difference because of the wide intrapatient variability. These results are similar to those seen in another recent study of the effect of food on CAI pharmacokinetics (9).

Table 3  Phase I study: neurotoxicity by grade and dose level

<table>
<thead>
<tr>
<th>Toxicity dose level</th>
<th>Cerebellar grade</th>
<th>Cortical grade</th>
<th>Headache grade</th>
<th>Mood grade</th>
<th>Sensory grade</th>
<th>Vision grade</th>
</tr>
</thead>
<tbody>
<tr>
<td>150 mg/m$^2$</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>1</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>200 mg/m$^2$</td>
<td>2</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>2</td>
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<tr>
<td>250 mg/m$^2$</td>
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<tr>
<td>300 mg/m$^2$</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>1</td>
</tr>
</tbody>
</table>

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Fixed-Dose Study. Both this trial and the NCI trial (9) demonstrated variability in pk parameters. CAI is p.o. administered with a limited number of dose sizes available requiring “rounding off” of the actual dose. Coupling these two facts, it would be rational to consider a fixed-dose of CAI rather than dosing based on body surface area. This supplemental study was designed to look at the feasibility of such a dose. At the same
time, because the snack study data indicated that food intake affects the C_{ss} of CAI, the fixed-dose study also was designed to evaluate the effects of duration of fast on CAI C_{ss} levels and toxicity profile. The dose chosen for this trial was 400 mg/day, which approximated 225 mg/m²/day in an average patient. This is midway between the MTD established by this Phase I trial and that established by the NCI trial.

A total of 6 patients were enrolled on this portion of the trial. Demographic data are listed in Table 7. One patient discontinued drug during the first cycle because of elevated bilirubin secondary to progressive disease. The remaining 5 patients either discontinued drug or required dose reduction during the first cycle because of toxicity. Median number of days on the 400 mg/day dose was 11 with a range of 2–27 days. Two patients discontinued because of nausea and vomiting, 1 because of neurocortical toxicity, and 1 because of blurred vision (the version of the common toxicity criteria used for this study had no grade 1 or 2 vision toxicity options). Three of the patients resumed CAI at a dose of 300 mg/day, 1 each of whom required additional dose modification to 250 mg and 200 mg/day, respectively. A fourth patient resumed CAI at 250 mg/day and maintained that dosage for 4 cycles of therapy. After the first 6 patients were treated, it was determined that 400 mg/day was not a feasible dose for CAI, and no additional patients were enrolled.

Only 1 patient had pk data available for both the a.m. and p.m. dose, but this was done at 300 mg/day. The CAI plasma concentrations of the patient were higher with a shorter 2-h fast than with the longer overnight fast (3.5 ± 0.4 mg/liter versus 2.4 ± 0.3 mg/liter; \( P = 0.001 \)). However, this patient received a dose equivalent to 150 mg/m², and these C_{ss} values all fell within the range observed at that dose.

Toxicities seen in this cohort at 400 mg/day and at all of the subsequent doses are listed in Table 9. Toxicities listed in the first column are included in the second. The only new side effect seen in this cohort was a single patient with extensive hepatic metastases attributable to renal cell carcinoma who developed erectile dysfunction of uncertain relationship with the CAI. This patient was started on antidepressants at the same time, had intermittent hypercalcemia and borderline testosterone levels, and may have had some level of symptomatology before initiating CAI.

**Response.** No partial or complete remissions of tumor were seen. On the original micronized Phase I study, there were
4 patients with prolonged stable disease resulting in administration of CAI for > 6 cycles. Two patients with renal cell cancer received 7 and 12 cycles of CAI with stable disease. The former had drug discontinued because of loss of visual acuity mentioned previously. One patient with thyroid carcinoma reported improvements in disease symptomatology and received 7 cycles of CAI, but this patient had multiple prolonged delays in administration of drug because of vision toxicity, and stability of disease is uncertain. One patient with hepatocellular carcinoma that was reported to have been rapidly progressive had stable to minimally progressive disease for 14 cycles.

There were no partial or complete responders on either of the supplemental studies. Four patients on the snack study with renal cell carcinoma had stable disease for 4, 5, 8, and 43 cycles of CAI therapy. All 3 of the renal cell carcinoma patients from the fixed-dose study remained on CAI with stable disease for 5 cycles. One of the 3 discontinued CAI because of toxicity, whereas the other 2 had progressive disease.

**DISCUSSION**

The MTD for the Phase I portion of the study is 150 mg/m^2 of CAI administered daily. A Phase I study of the same micronized formulation conducted at the NCI reported an MTD of 300 mg/m^2 daily (10). At first glance there appears to be a significant disparity between the two trials. How can we explain this apparent difference?

First, of significant importance is the fact that the UW trial initially escalated to a dose of 300 mg/m^2 daily without any difficulty. The toxicities then began to occur, necessitating a reversal of our escalation until an MTD exactly equal to the starting dose was achieved, as outlined in Table 2. Phase I trials are often criticized for sample size, and concern exists over exceeding the MTD (11). Nonetheless, Phase I trials have a history of being generally safe and predictive of a safe dose for Phase II and III study (12). Although the NCI and this study each had 6 patients treated with a dose of 300 mg/m^2, small sample size may still play a role in the disparity of results. Despite the differences in the MTD for the two studies, each reported a single patient treated at 300 mg/m^2 as receiving 12 cycles of CAI without any significant problems (10).

In addition to a disparity in MTD results between the UW and NCI trials, there may be a difference in the toxicity profiles of the two studies (11). The DLT for this Phase I trial was neurotoxicity. A diverse spectrum of significant neurotoxicity was seen including visual toxicity. Of 21 patients treated at the NCI with the micronized formulation, only 3 had neurotoxicity of any form (10). Nonetheless, neurotoxicity was the DLT in the NCI trial. Thus, it appears there was a difference in the frequency of neurological side effects that occurred at the two institutions but not in the fact that neurotoxicity precluded additional dose escalation. Evaluating all of the patients in our trial with neurotoxicity, there is no relationship between AUC,
single dose \( C_{\text{max}} \), or duration of therapy with the appearance of neurotoxicity. There was an apparent increase in the frequency of neurological side effects in patients with mean \( C_{\text{ss}} \geq 4 \) mg/liter (Fig. 3). That raises the question of whether or not there are differences in the pk data between this trial and the NCI trial, and, if so, why.

Both studies reported comparable inter- and intrapatient variability in pk parameters for the micronized formulation (10). The \( t_{1/2} \) values on the two trials are similar. Although the NCI reported an overall \( C/F \) (8.7 ± 9.5 liter/hr) higher than that reported in this paper (5.45 ± 2.7 liter/hr), the confidence intervals overlap. The mean \( C_{\text{ss}} \) values for the NCI study ranged from 0.56 ± 0.1 to 3.18 ± 1.7 mg/liter and appear to be slightly lower than the \( C_{\text{ss}} \) values for the UW study (3.2 ± 0.8 to 4.2 ± 1.5) for doses over a similar range. It is possible that the higher steady-state levels at the UW may explain the increased toxicity.

If the UW study patients were maintaining higher plasma levels of CAI was it attributable to absorption or elimination? Factors that may affect absorption characteristics include coadministered medications, level of bowel edema, bowel motility rates, mesenteric blood flow, and food intake including fatty composition of food. Of these factors, only food intake and medication coadministration can be controlled. Our initial micronized Phase I used a nighttime administration schedule with no controls of food intake, whereas the NCI administered the CAI in the morning after an overnight fast (10). Because CAI is a lipophilic drug, it may be hypothesized that this difference in study design may explain the difference in the pk results. Therefore, we undertook a supplemental study evaluating the effects of bedtime snacks and duration of fast on CAI pharmacokinetics. On the snack study, the effect of taking CAI with a snack did reach the level of statistical significance, increasing plasma concentrations of CAI by an average of 29%. On the fixed-dose study not enough data points were determined to show a definite effect of the duration of fast before CAI administration. A previous study at the NCI had also shown increased bioavailability when CAI was taken with a high fat meal (9). However, the overall variability in \( C_{\text{ss}} \) appears to overshadow the effects of food on CAI \( C_{\text{ss}} \) levels.

Although the data shows that food intake does increase plasma concentrations of CAI, it does not prove that this factor is enough to explain the difference in toxicity results in these small cohorts of patients (11). If food restriction prevented or ameliorated side effects from CAI then this would be a good rationale for requiring fasting administration of drug. However, there were no significant differences in the toxicity profiles for fasting administration versus coadministration with a nighttime snack. In the supplemental studies, patients met with nurses weekly to assure compliance with food restriction and drug administration. Although this method appears to have been effective in maintaining the compliance during this trial, it is not practical in an off-study setting. Because the UW Phase I trial established an MTD with no food restrictions, and the impact from food restrictions does not explain the full spectrum of pk variability and toxicity, we do not recommend fasting requirements in future Phase II and III trials.

The spectrum of side effects seen in the trials reported here were similar to what was seen in other trials of CAI (7, 8, 10). Neurotoxicity and GI side effects limited the ability to administer the drug. Five of the 6 patients treated with the fixed 400 mg/day dose after an overnight fast experienced administration-limiting side effects within the first cycle of therapy, including the vision toxicity discussed below. The sixth patient had minor side effects before having to stop drug because of biliary dysfunction from his disease. Thus, we can conclude that the 400-mg daily dose of CAI, which exceeds the UW MTD dose, is too high a dose for future trials. The snack study reaffirms that the toxicity profile of a 150 mg/m\(^2\) daily dose of CAI is tolerable and should remain the MTD for dosing by body surface area. In laboratory studies, antiproliferative and antimetastatic activity were seen at concentrations of CAI as low as 0.8 mg/liter (1, 13). Patients on this trial treated at 150 mg/m\(^2\) had serum \( C_{\text{ss}} \) levels of 1.9 mg/liter to 4.5 mg/liter, well within this range. These same patients all received either 250 mg or 300 mg of CAI daily with a greater number receiving the lower dose. The concept of a fixed-dose administration schedule for CAI remains viable and practical. In light of these facts, an alternative to our MTD of 150 mg/m\(^2\) of daily CAI may be a fixed-dose of 250 mg of daily CAI. This may be administered with no dietary restrictions and is well tolerated with nighttime administration. In another approach to CAI dosing, a recent study used a targeted dosing scheme to achieve \( C_{\text{ss}} \) levels of 2–5 mg/liter (14). However, 5 of 15 patients experienced grade 2 or 3 neurotoxicity, and there was a weak association between plasma concentration and this toxicity, indicating that perhaps this was too high a targeted \( C_{\text{ss}} \) range.

The visual acuity changes were the only new toxicity seen on this Phase I trial. There is no clear predictor for this toxicity. All of the patients were receiving CAI at dose equivalents higher than the MTD level recommended by our Phase I study. Two patients were treated at the same dose level (250 mg/m\(^2\)), but patients at the higher 300 mg/m\(^2\) level did not develop visual changes. One patient (receiving 400 mg/day on the fixed-dose study) who experienced this did not have an ophthalmological examination but reported significant blurring of vision. A fourth patient was receiving 150 mg/m\(^2\) of a gelcap formulation, which has a higher relative bioavailability than the micronized formulation he was intended to receive. Other patients without vision changes had higher AUC levels, higher \( C_{\text{max}} \) and higher \( C_{\text{ss}} \) levels. Because this toxicity was seen after several weeks to months of treatment, a candidate factor might be total CAI dose administered. However, several patients had significantly higher cumulative CAI doses including a patient receiving 14 cycles of therapy at 300 mg/m\(^2\) and a patient who received the gelcap formulation for nearly 3 years (7). Visual toxicity is a concern in any study. Other agents have been associated with loss of visual acuity. One of the investigators on this trial has reported very similar symptoms and findings with cisplatin therapy (15). As with our CAI patients, the return of visual acuity after cisplatin appeared to precede the return of color vision; however, the cisplatin patients took significantly longer to achieve full recovery (if they recovered) than our patients. Because only a few episodes of each type of neurotoxicity, including visual toxicity, occurred with CAI administration, future studies with more patients will be needed to better clarify the true incidence and spectrum of these side effects.

As expected, this Phase I trial and the supplemental studies did not result in any clinical or radiographic responses. How-
ever, of 19 patients with renal cell carcinoma treated overall, 9 had stable disease for as long as 40 months. In the previous Phase I trial of gelcap CAI, 2 of the 6 patients with renal cell carcinoma had prolonged stable disease for > 1 year (7). One patient was on gelcap CAI for nearly 3 years before discontinuing for progressive disease. Approved therapies for renal cell carcinoma are limited to interleukin 2 and IFN therapy both with a response rates of 15–20% (16). Because this was a highly selected group of patients treated at multiple dose levels of CAI with two different formulations, little can be said comparing the numbers to those of the cytokines. However, the fact that prolonged stabilization was seen in Phase I trials warrants additional investigation into this disease site. Thus, an Eastern Cooperative Group Phase II trial for patients with renal cell carcinoma who had failed biotherapy was opened using a fixed daily dose of 250 mg of micronized CAI administered in the evening.

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