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Clinical report

A phase I trial of 5-fluorouracil, leucovorin and interferon-α2b administered by 24 h infusion in metastatic colorectal carcinoma

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A phase I trial of 5-fluorouracil (5-FU), leucovorin (LV) and interferon (IFN) was conducted in 15 advanced colorectal cancer patients refractory to a bolus regimen of 5-FU/LV. Therapy consisted of a weekly i.v. infusion of 5-FU at 2600 mg/m^2 administered concomitantly with LV at 500 mg/m^2 over a 24 h period. IFN-α2b was administered by 24 h infusion from the second cycle at escalating dose (4.5, 9, 18 and 27 MU/m^2). The maximum tolerated dose of IFN was 18 MU/m^2. At 27 MU/m^2 two patients complained of diarrhea grade 3, so that the escalation of IFN dose was stopped. Two patients achieved a partial response (IFN level dose 9–18 MU/m^2). Eight patients had stable disease. Pharmacokinetics of 5-FU were not influenced by IFN at any level dose. Our results show that doses of IFN of 18 MU/m^2 given by a 24 h infusion can be administered safely to an established and active schedule of weekly 24 h infusion of 5-FU and LV. A phase II study has been planned to define the level of activity of this regimen.

Key words: Continuous infusion, 5-fluorouracil, interferon, leucovorin.

Introduction

In preclinical models interferons (IFN) have been shown to synergistically enhance the cytotoxic effects of 5-fluorouracil (5-FU). The locus and the mechanism of interactions between 5-FU and IFN seem to be different from those between 5-FU and leucovorin (LV).[1–5] On the basis of these data several clinical trials with 5-FU/LV and IFN were initiated. Although preliminary studies resulted in high objective response rate,[6,7] in further trials the activity of this combination has not been confirmed to be higher than 5-FU/LV alone.[8–10]

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These negative results can reflect the lack of potentiation among 5-FU, IFN and LV, achievable in the clinical setting, or simply the fact that almost all these regimens were designed empirically and the schedules used did not fit well with the available data in preclinical models. Some experimental and clinical data seem to suggest that a schedule of cyclic i.v. infusion, allowing higher doses to be administered concomitantly to 5-FU and LV, could determine an optimal modulation with the minimum of side effects.

Recently, Ardalan *et al.* demonstrated that high doses of 5-FU and LV could be given by short protracted infusion on a weekly basis, inducing a significant antitumor activity without severe toxicity.[11] This regimen appeared particularly suitable to verify the feasibility of a short infusion of IFN and to define its maximum tolerated dose (MTD), when given by this schedule together with 5-FU and LV.

Patients and methods

Fifteen patients with histologically documented advanced colorectal carcinoma entered the study. All patients had received 5-FU (370 mg/m^2, i.v., days 1–5) and 6S-LV (100 or 10 mg/m^2), i.v., days 1–5, every 28 days. Patients whose tumor progression was documented under the treatment were enrolled in this study. Further entry criteria required: ECOG performance status 0–2, life expectancy of at least 3 months, WBC count above 3000/μl, platelet count above 100 000 μl, serum bilirubin below 2 mg/dl, serum creatinine below 2 mg/dl and a treatment-free interval of at least 3 weeks from any prior chemotherapy.

The major objectives of this study were to establish the feasibility of adding i.v. IFN to an active schedule of 5-FU/LV and to define the MTD for IFN.

Table 1. Patient characteristics

Age (years)	
median	58
range	44–63
Sex	
M/F	10/5
Performance status (ECOG)	
0	6
I	7
II	2
Prior surgery	
none	1
curative	14
Sites of primary tumor	
colon	10
rectum	5
Sites of metastases	
liver	10
lung	3
abdomen/peritoneum	2
lymph nodes	3

Therapy consisted of IV infusion of LV at a dose of 500 mg/m^2 administered concurrently with 5-FU at 2600 mg/m^2 over a 24 h period. IFN-α2b was administered from the second cycle at escalating doses as a 24 h infusion with 5-FU and LV. It was studied in a phase I fashion. The starting dose was 4.5 MU/m^2. Dose in cohorts of three patients was escalated to 9, 18 and 27 MU/m^2. Dosages were not escalated over successive treatment courses for individual patients.

The MTD was defined to be one level below the dose producing unacceptable toxicity. The occurrence of grade 3–4 toxicity of any type in two or more patients at a given dose was considered unacceptable. A minimum of six patients were treated at the dose determined to be the MTD. The National Cancer Institute's common clinical toxicity criteria were used to grade the side effects.[12]

Patients who received 8 weeks of therapy were evaluated for antitumor response. Response was assessed as recommended by the WHO criteria.[13] Patients with responding or stable disease and no toxicity from a prior cycle continued to receive treatment with this regimen. Patients with unacceptable toxicity or evidence of progressive disease received no further therapy.

5-FU serum levels were determined in all the patients during the 24 h infusion in the first (without IFN) and the second cycle (with IFN). 5-FU was assessed by HPLC using the technique described previously.[14] Student's *t*-test was performed to assess potentially significant differences between the pharmacokinetic data obtained without and with IFN.

Results

Fifteen patients were enrolled in this study. Ten patients were males and five females. Median age was 62 years (range 45–68 years). All patients had measurable disease. Patient characteristics are summarized in Table 1.

Toxicity is reported in Table 2. At 27 MU/m^2 two patients complained of diarrhea grade 3, so that the escalation of IFN dose was stopped.

Two patients achieved a partial response (IFN dose level 9–18 MU/m^2); six patients had stable disease; five progressed on therapy and four were not evaluable because after the first cycle with IFN at the dose of 27 MU/m^2 they were withdrawn from the study because of toxicity. The two responses lasted 2 and 4 months. Median survival time for all the patients was 7 months.

5-FU AUC_{0-24} and C_{max} were not significantly influenced by IFN (Table 3).

Table 2. Toxic effects encountered according to IFN dose levels

Toxic effects	WHO grade	No. of patients with toxicity at IFN dose (MU/m^2)			
		4.5	9	18	27
Leukopenia	1–2	1	1	1	2
	3–4	–	–	–	–
Thrombocytopenia	1–2	–	–	1	–
	3–4	–	–	–	–
Diarrhea	1–2	1	1	1	1
	3–4	–	–	–	2
Stomatitis	1–2	1	1	1	1
	3–4	–	–	–	1

Table 3. 5-FU pharmacokinetics with and without IFN

Patient	IFN dose (MU/m^2)	AUC		C_{max}	
		with IFN	without IFN	with IFN	without IFN
1	4.5	167.37	122.72	6.4	6.2
2	4.5	86.50	141.61	5.9	7.3
3	4.5	132.13	132.16	7.1	7.1
4	9	174.93	155.95	9.7	9.8
5	9	226.66	272.52	9.8	9.1
6	9	197.44	231.20	7.2	7.1
7	18	128.83	143.88	7.5	7.2
8	18	132.25	144.49	9.9	6.9
9	18	203.76	144.17	10.4	10.4
10	18	235.79	318.35	8.9	11.6
11	18	231.06	240.37	10.6	11.2
12	18	125.89	229.98	9.9	8.4
13	27	179.62	161.59	8.8	8.9
14	27	227.81	199.49	11.7	10.8
15	27	163.33	160.24	10.1	11.7

Discussion

The preclinical and clinical rationale for combining LV with 5-FU is well defined and, over the past years, several methods of administration of 5-FU and LV have been popularized.[3] However, the results obtained by these regimens in the different schedules are modest with an overall response rate of 15–20% and no advantage in survival.[15] Among the attempts to increase 5-FU/LV activity, the use of a double biochemical modulation with other modulating agents, as well as prolonged infusion of 5-FU, were proposed.[6,7,16] Because of its properties, IFN appeared to be an interesting modulatory agent of fluoropyrimidine to be used together with LV. However, despite the preclinical data[4,5] and the encouraging results of the first clinical trials, the advantage of a double biochemical modulation by LV and IFN did not seem to be confirmed.[6–10] The very complex and not well defined interactions between 5-FU and IFN may explain the difficulty in selecting a clinically promising regimen. Some available experimental data can help to design a clinical regimen. The most important aspect is that 5-FU activity can be potentiated by IFN in a dose- and schedule-dependent manner.[1,2,4] In fact, a major critical point can be the proper timing and the duration of IFN administration. In preclinical models it has been demonstrated that if IFN was given after 5-FU, the cytotoxicity of the fluoropyrimidine was not potentiated while it was in the reverse sequence or when both agents were administered concomitantly.[2] Taking into account the pharmacokinetics of bolus 5-FU and the side-effects of IFN, it is reasonable to conclude that prolonged administration of IFN even when 5-FU is not administered cannot be useful. Furthermore, the administration of IFN three times a week could reduce the S phase fraction of tumor cells, thus reducing sensitivity to 5-FU.[17]

Another important aspect is IFN dose. It was demonstrated to modulate the cytotoxic effects of 5-FU in a dose-dependent fashion.[2] Moreover, only higher doses of IFN ($10\ MU/m^2$) were found responsible for changes in 5-FU pharmacokinetics.[16,18] The most relevant problem in the administration of high doses of IFN is the associated toxicity. In a phase I study of 5-FU/IFN combination, the MTD of IFN, s.c., was $15–18\ MU/m^2$ daily.[18] These doses, however, were tolerated poorly and treatment could not be sustained with the inability to deliver 5-FU for extended periods. Intravenous administration seems to allow higher IFN doses to be given with lower toxicity.[19] Because the pharmacokinetics of IFN given by i.v. push is characterized by a brief peak and a fast disappearance, a short-term infusion, concomitantly to 5-FU, could represent an optimal schedule.[20]

Even 5-FU seems to be preferable given by infusion rather than bolus to meet the situation of experimental models.[21] The concentrations of 5-FU used by Wadler *et al.* in their preclinical model were 10- to 40-fold lower than the peak concentration clinically achievable after bolus administration of 5-FU at a dose of $720\ mg/m^2$, but the $C \times T$

was higher. This latter experimental situation can be obtained clinically with a prolonged infusion of 5-FU.[2]

On the basis of these premises, the weekly 24 h infusion of the 5-FU and LV regimen as proposed by Ardalan *et al.*[11] appeared particularly suitable to associate with a 24 h infusion of IFN, in order to assess the feasibility and the MTD of IFN. The MTD of IFN when given by this schedule was found to be 18 MU/m^2. This dose was optimally tolerated and treatment could be sustained for a long time. It did not cause a reduction of 5-FU dose or delays in weekly administration. In our opinion the route and the timing of IFN administration could explain the good tolerability of such a relatively high dose. A similar regimen but with IFN given s.c. three times a week at a dose of 10 MU gave severe toxicity in about 50% of patients.[22]

In a phase I trial, it was possible to associate to a short infusion of 5-FU (500 mg/m^2 daily for 7 days) and a bolus LV (200 mg/m^2 daily for 7 days), IFN at a dose of only 5 MU daily for 7 days.[23]

Bukowsky *et al.* found that the MTD of s.c. IFN was 9 MU/m^2 when associated to a 5 days bolus of 5-FU (430 mg/m^2) and LV (200 mg/m^2).[24]

In our study, 5-FU pharmacokinetics were not influenced by IFN (Table 2). These data are similar to those obtained by Kreuser and Sparano with a short and prolonged infusion of 5-FU.[23,25]

In conclusion, our results show that doses of IFN (18 MU/m^2) given by a 24 h infusion can be administered safely together with an active schedule of weekly 24 h infusion of 5-FU and LV. A phase II study has been planned in order to define the activity of this regimen.

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