Discussion

5-FU, first synthesized 40 years ago, is still one of the most widely used agents for digestive system cancers including pancreatic cancer. Since 5-FU shows a short half-life and a time-dependent effect, its continuous infusion is known to result in a better antitumor effect than bolus injection [23]. A meta-analysis of six randomized trials has demonstrated that the continuous infusion 5-FU is superior to bolus 5-FU with respect to tumor response and survival in metastatic colorectal cancer [24]. As for pancreatic cancer, a recent study by Maisey et al. [25] has reported that the continuous infusion of 5-FU for the treatment of advanced pancreatic cancer results in a response rate of 8.4% and a median survival time of 5.1 months. However, continuous infusion of 5-FU requires a catheter, and is associated with complications, such as infections, and a reduced quality of life. Moreover, patients receiving continuous infusion of 5-FU show disturbance of their circadian rhythms and intraindividual variations in plasma 5-FU levels caused by dihydropyrimidine dehydrogenase, which contribute to limiting the effect of 5-FU. In addition, continuous infusion of 5-FU may cause severe gastrointestinal toxicities such as diarrhea and stomatitis. To overcome these problems, an oral fluoropyrimidine derivative, S-1, was developed on the basis of the biochemical modulation by CDHP, a dihydropyrimidine dehydrogenase inhibitor, and Oxo, a protector against 5-FU-induced gastrointestinal toxicity. Since the antitumor effects of S-1 on various solid cancers have been reported [12–18], we considered that the efficacy of S-1 on pancreatic cancer should also be investigated.

S-1 showed a good objective response rate of 21.1% with a good tumor growth control rate (objective responses plus no change) of 73.7% for metastatic pancreatic cancer patients. In the reported phase II and III studies for pancreatic cancer, single-agent gemcitabine showed response rates ranging from 5.4 to 16.0%, mostly below 15%, and tumor growth control rates ranging from 25.1–72.0%, mostly below 50% [2–8]. Our study also demonstrated a median survival time of 5.6 months with a one-year survival rate of 15.8%, which was comparable to the results of the gemcitabine studies. S-1 was easily administered, and most patients could be treated as outpatients. These results suggest that S-1 has an antitumor effect on metastatic pancreatic cancer.

A pharmacokinetic study of S-1 has already been conducted by Hirata et al [26]. They administered S-1 twice daily at a dose of 80 $mg/m^2/day$ in 12 patients with gastric, colorectal, and breast cancer, and reported that C_{max}, T_{max}, AUC_{0-14}, and $T_{1/2}$ of 5-FU after a single administration of S-1 were 128.5 ± 41.5 ng/ml, 3.5 ± 1.7 h, 723.9 ± 272.7 ng·h/ml, and 1.9 ± 0.4 h, respectively. The pharmacokinetic parameters of 5-FU observed in our study (C_{max}, 125.7 ± 46.8 ng/ml; T_{max}, 4.0 ± 1.1 h; $AUC_{0-\infty}$, 680.5 ± 252.1 ng·h/ml; $T_{1/2}$, 1.9 ± 0.3 h) were similar to those in Hirata's study. The pharmacokinetic parameters of other compounds, FT, CDHP, and Oxo, also did not show a large difference between the two studies. Therefore, our data suggest that there were no large differences between the pharmacokinetic parameters of S-1 in patients with pancreatic cancer and those in patients with other cancers.

Toxicity of S-1 was acceptable in our study. Hematological toxicities were mild, similar to the results of clinical studies of S-1 for other cancers. However, gastrointestinal toxicities such as anorexia and vomiting tended to occur more frequently in our study. Grade ≥3 anorexia and vomiting were observed in 4.8 and 1.6% of colorectal cancer patients [12], while grade ≥3 anorexia and vomiting were seen in 15.8 and 5.3% of pancreatic cancer patients. Since the pharmacokinetic parameters of S-1 did not differ between subjects with pancreatic cancer and those with other cancers, we speculate that anorexia and vomiting were observed more frequently partly because many patients with pancreatic cancer had disease-related symptoms such as anorexia before treatment. Although phase I studies for S-1 from the Netherlands and the United States described diarrhea as a dose-limiting factor [27, 28], diarrhea was mild and low in incidence in this study, similar to the results of other cancer studies conducted in Japan. However, 3 patients in the current study required hospitalization because of ileus, an observation different from the past Japanese reports. In the United States, an 80-year-old female with gallbladder cancer was reported as developing grade 4 ileus with grade 3 diarrhea after administration of S-1 [28]. In the current study, 1 of the 3 patients had concomitant colitis, while the remaining 2 had no colitis. Although the causes of the ileus were unknown, S-1 may have been the underlying cause, because all patients recovered from ileus after cessation of S-1 with appropriate treatment. Two of the 3 patients had been put on morphine, and showed a tendency towards constipation before the onset, suggesting that the administration of S-1 requires attention to bowel movements.

In this study, since no serious adverse events occurred except the above-described ileus, most patients could be treated as outpatients. The compliance rate of the patients receiving S-1 was as good as 90%. S-1 is an oral anticancer drug, and has the advantage of being able to treat patients while maintaining their quality of life. Since the prognosis of patients with advanced pancreatic cancer is generally poor, the demonstration in this study of the effectiveness and safety of S-1 (which allows treatment on an outpatient basis) for pancreatic cancer is highly significant. As the toxicity of S-1 is relatively mild, S-1 can be used in combination with other anticancer drugs. Combination therapy with S-1 and cisplatin has already been conducted for gastric cancer, and an excellent response rate of 76% was reported in a phase II study [29], which encourages the expectation of a future combination therapy with S-1 and other anticancer drugs including gemcitabine for advanced pancreatic cancer as well.

In conclusion, although this study had a small patient population, S-1 showed a promising antitumor activity with tolerable toxicity in metastatic pancreatic cancer patients. As an oral medication, S-1 offers a potential advantage as far as patient convenience is concerned, especially in terms of the patients' quality of life. We are currently conducting a multi-institutional late phase II study of S-1 for metastatic pancreatic cancer to confirm the results in this study.

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References

1 Evans DB, Abbruzzese JL, Willett CG: Cancer of the pancreas; in De Vita VT Jr, Hellman S, Rosenberg SA (eds): Principles and Practice of Oncology, ed 6. Philadelphia, Lippincott, 2001, pp 1126–1161.
2 Burris HA 3rd, Moore MJ, Andersen J, Green MR, Rothenberg ML, Modiano MR, Cripps MC, Portenoy RK, Storniolo AM, Tarassoff P, Nelson R, Dorr FA, Stephens CD, Von Hoff DD: Improvements in survival and clinical benefit with gemcitabine as first-line therapy for patients with advanced pancreas cancer: A randomized trial. J Clin Oncol 1997;15:2403–2413.
3 Casper ES, Green MR, Kelsen DP, Heelan RT, Brown TD, Flombaum CD, Trochanowski B, Tarassoff PG: Phase II trial of gemcitabine (2,2'-difluoro-deoxycytidine) in patients with adenocarcinoma of the pancreas. Invest New Drugs 1994;12:29–34.
4 Carmichael J, Fink U, Russell RC, Spittle MF, Harris AL, Spiessi G, Blatter J: Phase II study of gemcitabine in patients with advanced pancreatic cancer. Br J Cancer 1996;73:101–105.
5 Bramhall SR, Rosemurgy A, Brown PD, Bowry C, Buckels JAC: Marimastat as first-line therapy for patients with unresectable pancreatic cancer: A randomized trial. J Clin Oncol 2001; 19:3447–3455.
6 Bramhall SR, Schulz J, Nemunaitis J, Brown PD, Baillet M, Buckels JAC: A double-blind placebo-controlled, randomised study comparing gemcitabine and marimastat with gemcitabine and placebo as first line therapy in patients with advanced pancreatic cancer. Br J Cancer 2002;87:161–167.
7 Berlin JD, Catalano P, Thomas JP, Kugler JW, Haller DG, Benson AB III: Phase III study of gemcitabine in combination with fluorouracil versus gemcitabine alone in patients with advanced pancreatic carcinoma: Eastern Cooperative Oncology Group Trial E2297. J Clin Oncol 2002;20:3270–3275.
8 Colucci G, Giuliani F, Gebbia V, Biglietto M, Rabitti P, Uomo G, Cigolari S, Testa A, Maiello E, Lopez M: Gemcitabine alone or with cisplatin for the treatment of patients with locally advanced and/or metastatic pancreatic carcinoma: A prospective, randomized phase III study of the Gruppo Oncologico dell'Italia Meridionale. Cancer 2002;94:902–910.
9 Shirasaka T, Shimamoto Y, Ohshimo H, Yamaguchi M, Kato T, Yonekura K, Fukushima M: Development of a novel form of an oral 5-fluorouracil derivative (S-1) directed to the potentiation of the tumor selective cytotoxicity of 5-fluorouracil by two biochemical modulators. Anticancer Drugs 1996;7:548–557.
10 Tatsumi K, Fukushima M, Shirasaka T, Fujii S: Inhibitory effects of pyrimidine, barbituric acid and pyridine derivatives on 5-fluorouracil degradation in rat liver extracts. Jpn J Cancer Res 1987;78:748–755.
11 Shirasaka T, Shimamoto Y, Fukushima M: Inhibition by oxonic acid of gastrointestinal toxicity of 5-fluorouracil without loss of its antitumor activity in rats. Cancer Res 1993;53:4004–4009.
12 Ohtsu A, Baba H, Sakata Y, Mitachi Y, Horikoshi N, Sugimachi K, Taguchi T: Phase II study of S-1, a novel oral fluoropyrimidine derivative, in patients with metastatic colorectal carcinoma. S-1 Cooperative Colorectal Carcinoma Study Group. Br J Cancer 2000;83: 141–145.
13 Kawahara M, Furuse K, Segawa Y, Yoshimori K, Matsui K, Kudoh S, Hasegawa K, Niitani H: Phase II study of S-1, a novel oral fluorouracil in advanced non-small-cell lung cancer. Br J Cancer 2001;85:939–943.
14 Inuyama Y, Kida A, Tsukuda M, Kohno N, Satake B: Late phase II study of S-1 in patients with advanced head and neck cancer. Gan To Kagaku Ryoho 2001;28:1381–1390.
15 Sakata Y, Ohtsu A, Horikoshi N, Sugimachi K, Mitachi Y, Taguchi T: Late phase II study of novel oral fluoropyrimidine anticancer drug S-1 (1 M tegafur-0.4 M gimestat-1 M otastat potassium) in advanced gastric cancer patients. Eur J Cancer 1998;34:1715–1720.
16 Koizumi W, Kurihara M, Nakano S, Hasegawa K: Phase II study of S-1, a novel oral derivative of 5-fluorouracil, in advanced gastric cancer. For the S-1 Cooperative Gastric Cancer Study Group. Oncology 2000;58:191–197.
17 Sugimachi K, Maehara Y, Horikoshi N, Shimada Y, Sakata Y, Mitachi Y, Taguchi T: An early phase II study of oral S-1, a newly developed 5-fluorouracil derivative for advanced and recurrent gastrointestinal cancers. The S-1 Gastrointestinal Cancer Study Group. Oncology 1999;57:202–210.
18 Furuse K, Kawahara M, Hasegawa K, Kudoh S, Takada M, Sugiura T, Ichinose Y, Fukuoka M, Ohashi Y, Niitani H: Early phase II study of S-1, a new oral fluoropyrimidine, for advanced non-small-cell lung cancer. Int J Clin Oncol 2001;6:236–241.

19 Fukushima M, Satake H, Uchida J, Shimamoto Y, Kato T, Takechi T, Okabe H, Fujioka A, Nakano K, Ohshimo H, Takeda S, Shirasaka T: Preclinical antitumor efficacy of S-1: A new oral formulation of 5-fluorouracil on human tumor xenografts. Int J Oncol 1998;13:693–698.
20 Japan Society for Cancer Therapy: Criteria for the evaluation of the clinical effects of solid cancer chemotherapy. J Jpn Soc Cancer Ther 1993;28:101–130.
21 Aoki K, Okada S, Moriyama N, Ishii H, Nose H, Yoshimori M, Kosuge T, Ozaki H, Wakao F, Mukai K: Accuracy of computed tomography in determining pancreatic cancer tumor size. Jpn J Clin Oncol 1994;24:85–87.
22 Matsushima E, Yoshida K, Kitamura R, Yoshida K: Determination of S-1 (combined drug of tegafur, 5-chloro-2,4-dihydroxypyridine and potassium oxonate) and 5-fluorouracil in human plasma and urine using high-performance liquid chromatography and gas chromatography-negative ion chemical ionization mass spectrometry. J Chromatogr B 1997;691:95–104.
23 Lokich JJ, Ahlgren JD, Gullo JJ, Philips JA, Fryer JG: A prospective randomized comparison of continuous infusion fluorouracil with a conventional bolus schedule in metastatic colorectal carcinoma: A Mid-Atlantic Oncology Program Study. J Clin Oncol 1989;7:425–432.
24 Meta-analysis Group In Cancer: Efficacy of intravenous continuous infusion of fluorouracil compared with bolus administration in advanced colorectal cancer. J Clin Oncol 1998; 16:301–308.
25 Maisey N, Chau I, Cunningham D, Norman A, Seymour M, Hickish T, Iveson T, O'Brien M, Tebbutt N, Harrington A, Hill M: Multicenter randomized phase III trial comparing protracted venous infusion (PVI) fluorouracil (5-FU) with PVI 5-FU plus mitomycin in inoperable pancreatic cancer. J Clin Oncol 2002;20: 3130–3136.
26 Hirata K, Horikoshi N, Aiba K, Okazaki M, Denno R, Sasaki K, Nakano Y, Ishizuka H, Yamada Y, Uno S, Taguchi T, Shirasaka T: Pharmacokinetic study of S-1, a novel oral fluorouracil antitumor drug. Clin Cancer Res 1999;5:2000–2005.
27 van Groeningen CJ, Peters GJ, Schornagel JH, Gall H, Noordhuis P, de Vries MJ, Turner SL, Swart MS, Pinedo HM, Hanauske AR, Giaccone G: Phase I clinical and pharmacokinetic study of oral S-1 in patients with advanced solid tumors. J Clin Oncol 2000;18:2772–2779.
28 Cohen SJ, Leichman CG, Yeslow G, Beard M, Proefrock A, Roedig B, Damle B, Letrent SP, DeCillis AP, Meropol NJ: Phase I and pharmacokinetic study of once daily oral administration of S-1 in patients with advanced cancer. Clin Cancer Res 2002;8:2116–2122.
29 Ohtsu A, Boku N, Nagashima F, Koizumi W, Tanabe S, Saigenji K, Muro K, Matsumura K, Shirao K: A phase I/II study of S-1 plus cisplatin (CDDP) in patients (pts) with advanced gastric cancer (AGC). Proc Am Soc Clin Oncol 2001;20:656.

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A Phase I Study of Combination Chemotherapy with Gemcitabine and Oral S-1 for Advanced Pancreatic Cancer

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Key Words
Pancreatic cancer · 5-Fluorouracil · Gemcitabine · S-1

Abstract
Objective: The aim of this study was to determine the maximum-tolerated dose and dose-limiting toxicity (DLT) of combination therapy with gemcitabine and S-1 in patients with advanced pancreatic cancer. ***Methods:*** Chemotherapy-naive patients with histologically or cytologically proven unresectable or metastatic pancreatic cancer were enrolled. The patients received gemcitabine intravenously over 30 min on days 1 and 8 and S-1 orally twice daily from days 1 to 14. Cycles were repeated every 21 days until disease progression. Patients were scheduled to receive gemcitabine (mg/m^2/week) and S-1 (mg/m^2/day) at four dose levels: 800/60 (level 1), 1,000/60 (level 2), 1,000/70 (level 3) and 1,000/80 (level 4). ***Results:*** Eighteen patients were enrolled in this study. The maximum-tolerated dose was not reached even at the highest dose level (level 4) because only 2 of the 6 patients at this level experienced DLT. The DLTs were neutropenia and rash. Six (33%) of the 18 patients achieved a partial response and median overall survival time was 7.6 months. ***Conclusions:*** Combination chemotherapy with gemcitabine and S-1 was well tolerated and showed good antitumor activity in the treatment of pancreatic cancer. We recommend a gemcitabine dose of 1,000 mg/m^2/week and an S-1 dose of 80 mg/m^2/day in further studies with this schedule.

Introduction

Pancreatic cancer is a fatal disease, with a 5-year survival rate of less than 5% [1]. Surgery remains the only curative option for patients with this disease, but the vast majority of patients unfortunately present with advanced, unresectable tumors. Effective non-surgical treatment is therefore needed to improve the outcome in patients with pancreatic cancer.

A randomized controlled study demonstrated that gemcitabine, a nucleoside analogue, is effective in palliating symptoms and prolonging survival in patients with advanced pancreatic cancer: gemcitabine showed a statistically significant advantage both in clinical benefit response (23.8 vs. 4.8%, p = 0.0022) and in median survival (5.65 vs. 4.41 months, p = 0.0025) compared with weekly bolus 5-fluorouracil (5-FU) [2]. Single-agent gemcitabine is currently accepted worldwide as first-line therapy for advanced pancreatic cancer. Nevertheless, there is substantial room for improvement in chemotherapy for pancreatic cancer, because single-agent gemcitabine pro-

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vides only limited benefit, with objective response rates of less than 15% and a median survival of less than 6 months [2–5].

S-1 is an oral fluoropyrimidine derivative that combines tegafur with two modulators of 5-FU metabolism, 5-chloro-2,4-dihydroxypyridine and potassium oxonate [6]. 5-Chloro-2,4-dihydroxypyridine is a competitive inhibitor of dihydropyrimidine dehydrogenase, which is involved in the degradation of 5-FU, and acts to maintain efficacious concentrations of 5-FU in plasma and tumor tissues [7]. Potassium oxonate, a competitive inhibitor of orotate phosphoribosyltransferase, inhibits the phosphorylation of 5-FU in the gastrointestinal tract, reducing the serious gastrointestinal toxicity associated with 5-FU [8]. The efficacy of S-1 has already been demonstrated in a variety of solid tumors: the response rates for advanced gastric cancer, colorectal cancer and non-small cell lung cancer in the phase II studies conducted in Japan were 49, 35 and 22%, respectively [9–11]. Recently, the clinical efficacy of S-1 against pancreatic cancer has also been investigated. We conducted an early phase II study of S-1 for metastatic pancreatic cancer and reported that 4 (21.1%) of 19 patients achieved a partial response, with mild toxicity [12]. Hayashi et al. [13] performed a pilot study of single-agent S-1 or S-1 plus cisplatin combination therapy in patients with advanced pancreatic cancer and reported that 3 (20.0%) of the 15 patients or 8 (57.1%) of the 14 patients showed a partial response.

Since S-1 shows a favorable toxicity profile and activity in various solid tumors, including pancreatic cancer, we decided to investigate whether combination therapy with gemcitabine and S-1 is an effective chemotherapeutic regimen for pancreatic cancer. Although many clinical studies of gemcitabine in combination with fluoropyrimidines such as 5-FU, uracil/tegafur and capecitabine have been reported [14–22], little information is available on the combination of gemcitabine and S-1. Thus, we conducted a phase I study to determine the maximum-tolerated dose (MTD) and dose-limiting toxicity (DLT) of gemcitabine and S-1 combination therapy in patients with unresectable or metastatic pancreatic cancer.

Patients and Methods

Patient Selection

Patients were considered eligible if they met the following criteria: histologically or cytologically proven pancreatic adenocarcinoma, unresectable locally advanced or metastatic disease, naive to chemotherapy, Eastern Cooperative Oncology Group performance status of 0–2, age between 20 and 74 years, life expectancy of ≥8 weeks, and adequate organ function defined as white blood cell count ≥4,000/mm^3, neutrophil count ≥2,000/mm^3, platelet count ≥100,000/mm^3, hemoglobin ≥9.0 g/dl, serum creatinine ≤the upper limit of normal, serum albumin ≥3.0 g/dl, total bilirubin ≤2.0 mg/dl, and aspartate aminotansferase and alanine aminotransferase levels ≤2.5 times the upper limit of normal or ≤5 times the upper limit of normal if liver metastases or biliary drainage were present. The exclusion criteria were severe complications, such as infection, heart disease and renal disease (in this study we did not define in detail the exclusion criteria in relation to severe complications), metastasis to the central nervous system, marked pleural effusion or ascites, and watery diarrhea. Pregnant or lactating women were also excluded. Written informed consent was obtained from all patients. This study was approved by the institutional review board at the National Cancer Center and conducted in accordance with the Declaration of Helsinki.

Treatment Plan

This was an open-label, two-center, single-arm phase I study. Gemcitabine (Eli Lilly Japan K.K., Kobe, Japan) was administered as a 30-min intravenous infusion weekly for 2 weeks followed by a 1-week rest. S-1 (Taiho Pharmaceutical Co., Ltd., Tokyo, Japan) was administered orally twice daily from day 1 to day 14 followed by a 1-week rest. The treatment cycles were repeated every 3 weeks until disease progression or unacceptable toxicity occurred. If patients experienced leucopenia <2,000/mm^3, neutropenia <1,000/mm^3, thrombocytopenia <70,000/mm^3, total bilirubin >2.0 mg/dl or aspartate aminotansferase and alanine aminotransferase levels >5 times the upper limit of normal, both gemcitabine and S-1 were withheld until recovery. If patients experienced DLT, the dose of gemcitabine was reduced by 200 mg/m^2/week and the dose of S-1 was reduced by 10 mg/m^2/day in the subsequent cycle. If a rest period of more than 3 weeks was required because of toxicity, the patient was withdrawn from the study.

Patients were scheduled to receive gemcitabine and S-1 at four dose levels (table 1). At the first dose level (level 1), gemcitabine was administered at a dose of 800 mg/m^2/week and S-1 was administered at 60 mg/m^2/day. At the next dose level (level 2), gemcitabine was increased to 1,000 mg/m^2/week with S-1 kept at the same dose. At each of dose levels 3 and 4, S-1 was increased by 10 mg/m^2/day with gemcitabine kept at 1,000 mg/m^2/week. At least 3 patients were enrolled at each dose level. If DLT was observed in the initial 3 patients, a maximum of 3 additional patients was entered into the same dose level. The MTD was defined as the highest dose level that did not cause DLT in 3 of the 3 or ≥3 of the 6 patients treated at that level during the first two cycles of treatment. DLT was defined as grade 4 leucopenia or neutropenia, febrile neutropenia, grade 4 thrombocytopenia, grade 3 thrombocytopenia requiring transfusion, ≥grade 3 non-hematological toxicity excluding nausea, vomiting, anorexia and fatigue, or any toxicity that necessitated a treatment delay of more than 3 weeks. Toxicity was graded according to the National Cancer Institute Common Toxicity Criteria version 2.0.

Patient Evaluation

Physical examinations, complete blood cell counts, biochemistry tests and urinalyses were performed at least once weekly. Tumor assessment with computed tomographic scan or magnetic resonance imaging and measuring of tumor marker CA 19-9 was performed every two cycles, and tumor response was evaluated by the

Table 1. Dose escalation scheme and DLT

Dose level	Gemcitabine mg/m²/week	S-1 mg/m²/day	Patients	DLT events	DLT
1	800	60	3	0	
2	1,000	60	3	0	
3	1,000	70	6	1	grade 4 neutropenia
4	1,000	80	6	2	grade 4 neutropenia grade 3 rash and grade 4 neutropenia

criteria of the Japan Society for Cancer Therapy [23], which are similar to those of the World Health Organization. Briefly, a complete response was defined as the disappearance of all clinical evidence of the tumor for a minimum of 4 weeks. A partial response was defined as a 50% or greater reduction in the sum of the products of two perpendicular diameters of all measurable lesions for 4 weeks or longer without any evidence of new lesions. No change was defined as a reduction of less than 50% or a less than 25% increase in the sum of the products of two perpendicular diameters of all lesions for a minimum of 4 weeks. Progressive disease was defined as an increase of 25% or more in the sum of the products of two perpendicular diameters of all lesions, the appearance of any new lesion, or deterioration in clinical status that was consistent with disease progression. The response duration was calculated from the day of the first sign of a response until disease progression; progression-free survival was calculated from the date of the initiation of treatment until documented disease progression or death due to any cause (whichever occurred first); overall survival time was calculated from the date of treatment initiation to the date of death or the last follow-up. The median probabilities of the progression-free or overall survival periods were estimated by the Kaplan-Meier method.

Table 2. Patient characteristics

Characteristics		Patients
Patients enrolled		18
Sex		
Male		13
Female		5
Age, years		
Median	61	
Range	43–72	
ECOG performance status		
0		10
1		8
Body surface area, m²		
Median	1.58	
Range	1.46–1.97	
Disease stage		
Locally advanced		2
Metastatic		16
Sites of metastatic disease		
Liver		13
Lung		2
Distant lymph nodes		5
Pleura		1

ECOG = Eastern Cooperative Oncology Group.

Results

Patient Characteristics

Between September 2003 and July 2004, 18 patients were enrolled in this study. All of them received at least two cycles of chemotherapy and were evaluable for toxicity and response. Patient characteristics are listed in table 2. All patients had good performance status (0 and 1). Two patients had locally advanced unresectable disease and the remaining 16 had metastatic disease. Before the start of the study, 1 patient had received surgical resection and 3 had undergone biliary drainage for obstructive jaundice. Twelve patients had abdominal and/or back pain at study entry. A total of 125 cycles of chemotherapy was administered, with a median of 6 treatment cycles per patient (range 2–22). It was possible to treat all patients as outpatients after one or two cycles of observation in hospital.

DLT and Recommended Dose

No DLT was observed at dose levels 1 or 2 (table 1). At dose level 3, 1 patient developed grade 4 neutropenia, which was considered DLT, but the remaining 5 did not develop DLT. At dose level 4, the highest dose level, 2 of the 6 patients exhibited DLTs: 1 had grade 4 neutropenia and the other had grade 3 rash concomitant with grade 4 neutropenia. All DLTs occurred in the first cycle of treatment. The MTD was not reached because only 2 of the 6 patients experienced DLT at dose level 4. Therefore, dose level 4 (gemcitabine dose of 1,000 mg/m²/week and S-1

Table 3. Toxicities across first two cycles by dose level (patient number)

Toxicity	Dose level 1 (n = 3)				Dose level 2 (n = 3)				Dose level 3 (n = 6)				Dose level 4 (n = 6)			
Grade:	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4
Leucopenia	1	2	0	0	0	2	1	0	1	4	1	0	1	2	3	0
Neutropenia	1	1	0	0	0	1	2	0	0	5	0	1	0	3	1	2
Anemia	2	0	0	0	3	0	0	0	4	1	0	0	4	2	0	0
Thrombocytopenia	2	0	0	0	1	2	0	0	4	1	0	0	3	0	0	0
Nausea	2	0	0	0	1	0	1	0	2	2	0	0	2	0	0	0
Vomiting	0	0	0	0	1	0	0	0	0	1	0	0	0	0	0	0
Anorexia	1	0	0	0	0	0	1	0	2	1	0	0	2	1	0	0
Diarrhea	1	0	0	0	1	0	0	0	0	0	0	0	2	0	0	0
Stomatitis	2	0	0	0	1	0	0	0	1	0	0	0	0	0	0	0
Rash	0	1	0	0	1	2	0	0	2	0	0	0	3	1	1	0
ALT elevation	1	0	0	0	2	0	0	0	3	2	0	0	1	0	0	0
Creatinine elevation	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Fever	0	0	0	0	0	1	0	0	0	0	0	0	3	0	0	0
Fatigue	1	0	0	0	1	0	0	0	1	1	0	0	2	1	0	0

Toxicity was graded according to the National Cancer Institute Common Toxicity Criteria version 2.0. ALT = Alanine aminotransferase.

dose of 80 mg/m^2/day) was considered the recommended dose in further studies with this schedule.

Toxicity

All 18 patients were assessable for toxicity. The major toxicities observed during the first two cycles are summarized in table 3. Hematological toxicity, particularly neutropenia, was the most pronounced toxicity of gemcitabine and S-1 with this schedule of administration. Although 3 patients experienced grade 4 neutropenia during the first two cycles of treatment, all of them recovered quickly without any severe complications. The neutrophil nadir typically occurred on day 15, and neutrophil counts recovered to baseline values by day 22. The non-hematological toxicities commonly observed with our regimen were gastrointestinal toxicities, such as nausea (≥grade 1; 55.6%) and anorexia (≥grade 1; 44.4%), although most of them were mild and transient. Although 1 patient at dose level 2 experienced grade 3 anorexia and grade 3 nausea in the first cycle, he recovered from the toxicities with the use of antiemetic agents and could continue treatment without reducing the doses of gemcitabine and S-1. Skin rash was also frequently seen in the current study (≥grade 1; 61.1%). The rash typically appeared on the arms and legs and spread to the trunk within 10 days of the initiation of chemotherapy. Most rashes were mild and resolved promptly with appropriate medical treatment such as antihistamines and steroids, although 1 patient at dose level 4 exhibited grade 3 rash that required temporary treatment discontinuation and dose reduction in the next cycle. Although 125 cycles of chemotherapy have been administered, there was no indication of cumulative toxicity.

Table 4. Objective tumor response

Dose level	Patients	Response				Response rate, %
		CR	PR	NC	PD	
1	3	0	2	1	0	66.7
2	3	0	0	1	2	0
3	6	0	3	3	0	50
4	6	0	1	4	1	16.7
Total	18	0	6	9	3	33.3

CR = Complete response; PR = partial response; NC = no change; PD = progressive disease.

Efficacy

The objective tumor responses at each dose level are shown in table 4. A partial response was seen even at the lowest dose level, and across all dose levels, 6 of the 18 patients achieved a partial response, resulting in an overall response rate of 33.3 (95% confidence interval, 13.3–59.0%). No change was noted in 9 patients (50%) and progressive disease in 3 patients (16.7%). The mean response duration was 4.8 months (range 2.8–15.9). The serum CA 19-9 level was reduced to less than half from baseline values in 8 (61.5%) of the 13 patients who had a pretreatment level greater than the upper limit of normal (37 U/ml). At the time of analysis, 9 patients had died because of disease progression. The median progression-free and the median overall survival times were 5.0 and 7.6 months, respectively.

Discussion

To improve the prognosis of patients with advanced pancreatic cancer, gemcitabine-based combination chemotherapy has been actively investigated, although many phase III trials have failed to demonstrate any survival benefit of combination chemotherapy in comparison with gemcitabine as a single agent. 5-FU has been selected as a candidate to be investigated in combination with gemcitabine in patients with pancreatic cancer because of its favorable toxicity profile and modest but substantial activity in this disease. Gemcitabine is considered to enhance the effect of the 5-FU metabolite 5-FdUMP by reducing the concentration of its physiological competitor via inhibition of ribonucleotide reductase [24]. Preclinical studies have demonstrated synergy between gemcitabine and 5-FU in tumor cell lines, including pancreatic cancer cells [25, 26]. Clinical studies have reported activity of gemcitabine in pancreatic cancer patients with refractoriness to 5-FU [27], suggesting the lack of cross-resistance between the two agents. Several phase I and II studies of combination therapy with gemcitabine and 5-FU for advanced pancreatic cancer have demonstrated relatively good response rates of around 20% with acceptable toxicity profiles [14–18]. A phase III study comparing gemcitabine alone with gemcitabine plus weekly bolus 5-FU showed that median progression-free survival was significantly longer in the combination arm compared with gemcitabine alone (3.4 vs. 2.2 months, $p = 0.022$); however, median overall survival was not significantly prolonged (6.7 vs. 5.4 months, $p = 0.09$) [5].

The novel oral anticancer agent S-1 was developed to improve the tumor-selective toxicity of 5-FU and has shown efficacy in a variety of solid tumors, including pancreatic cancer [9–13]. With the aim of developing a more effective chemotherapeutic regimen for pancreatic cancer, we decided to conduct a clinical study of combination therapy with gemcitabine and S-1. Since this combination has not previously been investigated, a phase I study was carried out to determine MTD and DLT.

In the present study, MTD was not reached because only 2 of the 6 patients experienced DLT at the highest dose, level 4. Although the 6 patients at level 4 have received a total of 34 cycles of treatment (average 5.7, range 2–12), there was no indication of cumulative toxicity. Therefore, dose level 4 (gemcitabine 1,000 mg/m^2/week, S-1 80 mg/m^2/day) was considered the recommended dose in further studies of this combination regimen. Because 2 of the 6 patients experienced DLT at this level, it goes without saying that more large-scale studies will be necessary to confirm the safety of our recommended dose. The overall toxicity of this regimen was mild, and neither unexpected nor life-threatening toxicities were observed during the study, indicating that S-1, like other fluoropyrimidines, can be safely combined with gemcitabine.

Neutropenia was the major DLT of this combination regimen: 1 of the 6 patients at dose level 3, and 2 of the 6 patients at dose level 4, experienced grade 4 neutropenia. Neutropenia as the DLT was to be expected because myelosuppression, especially neutropenia, is one of the most common toxicities of each individual drug. The neutrophil nadir typically occurred on day 15, but in most cases, the neutrophil count spontaneously recovered to baseline values within a week. Furthermore, no febrile neutropenia was observed during any of the 125 cycles of treatment, suggesting that the myelosuppression caused by this combination regimen is manageable on an outpatient basis.

The non-hematological toxicities commonly observed with our regimen were gastrointestinal toxicities such as nausea and anorexia. Although 1 patient at dose level 2 experienced transient grade 3 nausea and grade 3 anorexia, no DLTs associated with gastrointestinal toxicities were observed. Diarrhea was also mild and rare in the current study, similar to previous reports from Japanese studies of single-agent S-1; however, relatively severe diarrhea induced by S-1 has been reported in studies from Europe and the United States [28–30]. For example, Hoff et al. [28] reported that severe diarrhea occurred in all of the 3 patients who received S-1 at a dose of 40 mg/m^2 b.i.d. It is not clear why the toxicity profile and MTD of S-1 in Western studies differ from those in studies with Japanese populations, although a pharmacokinetic study suggested that the conversion of tegafur to 5-FU may occur more slowly in Japanese patients than in patients from other ethnic groups [31]. In any event, it may be dangerous to apply the results of our study directly to treatment of Western patients, particularly from the viewpoint of gastrointestinal toxicity.

In the present study, 11 (61.1%) of the 18 patients experienced grade 1 or greater rash. This toxicity was mild and manageable, although 1 patient at dose level 4 developed grade 3 rash, requiring temporary treatment discontinuation. The reason for the enhanced cutaneous toxicity during combination therapy with gemcitabine and S-1 is unknown, although cutaneous toxicity has already been reported in patients receiving gemcitabine and 5-FU combination regimens. Hidalgo et al. [14] reported grade 1 or greater cutaneous toxicity in 11 (42.3%) of the 26 patients in a phase I–II study with gemcitabine and 5-FU. One of these patients developed a severe cutaneous reaction, manifested as generalized exfoliative dermatitis, after the first cycle of chemotherapy.

Combination therapy with gemcitabine and S-1 was associated with promising activity in advanced pancreatic cancer. Six (33.3%) of the 18 patients achieved an objective response. Of the 13 patients who had a pretreatment serum CA 19-9 level greater than 37 U/ml, the CA 19-9 level decreased more than 50% in 8 patients (61.5%). In addition, the median progression-free survival time of 5.0 months and the median overall survival time of 7.6 months are encouraging. These efficacy data in this study, which compare favorably with those reported for single-agent gemcitabine, support further studies of this regimen.

In conclusion, our combination regimen of gemcitabine and S-1 was well tolerated up to dose level 4. The major toxicities were myelosuppression, gastrointestinal toxicity and skin rash, although most of these toxicities were mild and reversible. Six of the 18 patients showed a partial response, suggesting a promising antitumor activity of this regimen against pancreatic cancer. A multicenter phase II study of this regimen, 1,000 mg/m^2/week gemcitabine on days 1 and 8 and 80 mg/m^2/day S-1 from days 1 to 14 every 3 weeks, is under way in patients with metastatic pancreatic cancer.

References

1 Evans DB, Abbruzzese JL, Willett CG: Cancer of the pancreas; in De Vita VT Jr, Hellman S, Rosenberg SA (eds): Cancer: Principles and Practice of Oncology. Philadelphia, Lippincott-Raven, 2001, pp 1126–1161.

2 Burris HA 3rd, Moore MJ, Andersen J, Green MR, Rothenberg ML, Modiano MR, Cripps MC, Portenoy RK, Storniolo AM, Tarassoff P, Nelson R, Dorr FA, Stephens CD, Von Hoff DD: Improvements in survival and clinical benefit with gemcitabine as first-line therapy for patients with advanced pancreas cancer: a randomized trial. J Clin Oncol 1997;15:2403–2413.

3 Casper ES, Green MR, Kelsen DP, Heelan RT, Brown TD, Flombaum CD, Trochanowski B, Tarassoff PG: Phase II trial of gemcitabine (2,2′-difluorodeoxycytidine) in patients with adenocarcinoma of the pancreas. Invest New Drugs 1994;12:29–34.

4 Carmichael J, Fink U, Russell RC, Spittle MF, Harris AL, Spiessi G, Blatter J: Phase II study of gemcitabine in patients with advanced pancreatic cancer. Br J Cancer 1996;73:101–105.

5 Berlin JD, Catalano P, Thomas JP, Kugler JW, Haller DG, Benson AB 3rd: Phase III study of gemcitabine in combination with fluorouracil versus gemcitabine alone in patients with advanced pancreatic carcinoma: Eastern Cooperative Oncology Group Trial E2297. J Clin Oncol 2002;20:3270–3275.

6 Shirasaka T, Shimamato Y, Ohshimo H, Yamaguchi M, Kato T, Yonekura K, Fukushima M: Development of a novel form of an oral 5-fluorouracil derivative (S-1) directed to the potentiation of the tumor selective cytotoxicity of 5-fluorouracil by two biochemical modulators. Anticancer Drugs 1996;7:548–557.

7 Tatsumi K, Fukushima M, Shirasaka T, Fujii S: Inhibitory effects of pyrimidine, barbituric acid and pyridine derivatives on 5-fluorouracil degradation in rat liver extracts. Jpn J Cancer Res 1987;78:748–755.

8 Shirasaka T, Shimamoto Y, Fukushima M: Inhibition by oxonic acid of gastrointestinal toxicity of 5-fluorouracil without loss of its antitumor activity in rats. Cancer Res 1993;53:4004–4009.

9 Sakata Y, Ohtsu A, Horikoshi N, Sugimachi K, Mitachi Y, Taguchi T: Late phase II study of novel oral fluoropyrimidine anticancer drug S-1 (1 *M* tegafur-0.4 *M* gimestat-1 *M* otastat potassium) in advanced gastric cancer patients. Eur J Cancer 1998;34:1715–1720.

10 Ohtsu A, Baba H, Sakata Y, Mitachi Y, Horikoshi N, Sugimachi K, Taguchi T: Phase II study of S-1, a novel oral fluorophyrimidine derivative, in patients with metastatic colorectal carcinoma. S-1 Cooperative Colorectal Carcinoma Study Group. Br J Cancer 2000;83:141–145.

11 Kawahara M, Furuse K, Segawa Y, Yoshimori K, Matsui K, Kudoh S, Hasegawa K, Niitani H: Phase II study of S-1, a novel oral fluorouracil, in advanced non-small-cell lung cancer. Br J Cancer 2001;85:939–943.

12 Ueno H, Okusaka T, Ikeda M, Takezako Y, Morizane C: An early phase II study of S-1 in patients with metastatic pancreatic cancer. Oncology 2005;68:171–178.

13 Hayashi K, Imaizumi T, Uchida K, Kuramochi H, Takasaki K: High response rates in patients with pancreatic cancer using the novel oral fluoropyrimidine S-1. Oncol Rep 2002;9:1355–1361.

14 Hidalgo M, Castellano D, Paz-Ares L, Gravalos C, Diaz-Puente M, Hitt R, Alonso S, Cortes-Funes H: Phase I-II study of gemcitabine and fluorouracil as a continuous infusion in patients with pancreatic cancer. J Clin Oncol 1999;17:585–592.

15 Cascinu S, Silva RR, Barni S, Labianca R, Frontini L, Piazza E, Pancera G, Giordani P, Giuliodori L, Pessi MA, Fusco V, Luporini G, Cellerino R, Catalano G: A combination of gemcitabine and 5-fluorouracil in advanced pancreatic cancer, a report from the Italian Group for the Study of Digestive Tract Cancer (GISCAD). Br J Cancer 1999;80:1595–1598.

16 Matano E, Tagliaferri P, Libroia A, Damiano V, Fabbrocini A, De Lorenzo S, Bianco AR: Gemcitabine combined with continuous infusion 5-fluorouracil in advanced and symptomatic pancreatic cancer: a clinical benefit-oriented phase II study. Br J Cancer 2000;82:1772–1775.

17 Oettle H, Arning M, Pelzer U, Arnold D, Stroszczynski C, Langrehr J, Reitzig P, Kindler M, Herrenberger J, Musch R, Korsten EW, Huhn D, Riess H: A phase II trial of gemcitabine in combination with 5-fluorouracil (24-hour) and folinic acid in patients with chemonaive advanced pancreatic cancer. Ann Oncol 2000;11:1267–1272.
18 Rauch DP, Maurer CA, Aebi S, Pampallona S, Friess H, Ludwig CU, Buchler MW, Borner MM: Activity of gemcitabine and continuous infusion fluorouracil in advanced pancreatic cancer. Oncology 2001;60:43–48.
19 Feliu J, Lopez Alvarez MP, Jaraiz MA, Constenla M, Vicent JM, Belon J, Lopez Gomez L, de Castro J, Dorta J, Gonzalez Baron M: Phase II trial of gemcitabine and UFT modulated by leucovorin in patients with advanced pancreatic carcinoma. The ONCOPAZ Cooperative Group. Cancer 2000;89:1706–1713.
20 Feliu J, Mel R, Borrega P, Lopez Gomez L, Escudero P, Dorta J, Castro J, Vazquez-Estevez SE, Bolanos M, Espinosa E, Gonzalez Baron M: Phase II study of a fixed dose-rate infusion of gemcitabine associated with uracil/tegafur in advanced carcinoma of the pancreas. Ann Oncol 2002;13:1756–1762.
21 Hess V, Salzberg M, Borner M, Morant R, Roth AD, Ludwig C, Herrmann R: Combining capecitabine and gemcitabine in patients with advanced pancreatic carcinoma: a phase I/II trial. J Clin Oncol 2003;21:66–68.
22 Scheithauer W, Schull B, Ulrich-Pur H, Schmid K, Raderer M, Haider K, Kwasny W, Depisch D, Schneeweiss B, Lang F, Kornek GV: Biweekly high-dose gemcitabine alone or in combination with capecitabine in patients with metastatic pancreatic adenocarcinoma: a randomized phase II trial. Ann Oncol 2003;14:97–104.
23 Japan Society for Cancer Therapy: Criteria for the evaluation of the clinical effects of solid cancer chemotherapy. J Jpn Soc Cancer Ther 1993;28:101–130.
24 Heinemann V, Xu YZ, Chubb S, Sen A, Hertel LW, Grindey GB, Plunkett W: Inhibition of ribonucleotide reduction in CCRF-CEM cells by 2′,2′-difluorodeoxycytidine. Mol Pharmacol 1990;38:567–572.
25 Ren Q, Kao V, Grem JL: Cytotoxicity and DNA fragmentation associated with sequential gemcitabine and 5-fluoro-2′-deoxyuridine in HT-29 colon cancer cells. Clin Cancer Res 1998;4:2811–2818.
26 Bruckner H, Zhou G, Haenel P, Szrajer L, Greenspan E, Kurbacher C: Ex vivo ATP tumor testing of gemcitabine for combination chemotherapy and biochemical modulation (abstract 2116). Proc Am Assoc Cancer Res 1998;39:310a.
27 Rothenberg ML, Moore MJ, Cripps MC, Andersen JS, Portenoy RK, Burris HA 3rd, Green MR, Tarassoff PG, Brown TD, Casper ES, Storniolo AM, Von Hoff DD: A phase II trial of gemcitabine in patients with 5-FU-refractory pancreas cancer. Ann Oncol 1996;7:347–353.
28 Hoff PM, Saad ED, Ajani JA, Lassere Y, Wenske C, Medgyesy D, Dwivedy S, Russo M, Pazdur R: Phase I study with pharmacokinetics of S-1 on an oral daily schedule for 28 days in patients with solid tumors. Clin Cancer Res 2003;9:134–142.
29 van Groeningen CJ, Peters GJ, Schornagel JH, Gall H, Noordhuis P, de Vries MJ, Turner SL, Swart MS, Pinedo HM, Hanauske AR, Giaccone G: Phase I clinical and pharmacokinetic study of oral S-1 in patients with advanced solid tumors. J Clin Oncol 2000;18:2772–2779.
30 Cohen SJ, Leichman CG, Yeslow G, Beard M, Proefrock A, Roedig B, Damle B, Letrent SP, DeCillis AP, Meropol NJ: Phase I and pharmacokinetic study of once daily oral administration of S-1 in patients with advanced cancer. Clin Cancer Res 2002;8:2116–2122.
31 Comets E, Ikeda K, Hoff P, Fumoleau P, Wanders J, Tanigawara Y: Comparison of the pharmacokinetics of S-1, an oral anticancer agent, in Western and Japanese patients. J Pharmacokinet Pharmacodyn 2003;30:257–283.

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Chemoradiotherapy for Locally Advanced Pancreatic Carcinoma in Elderly Patients

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Key Words
Chemoradiotherapy · Pancreatic carcinoma · Elderly

Abstract
Objectives: Chemoradiotherapy, which is one of the standard treatments for locally advanced pancreatic carcinoma, is considered a high-risk procedure in elderly patients. This study investigated the outcome and tolerability of this treatment in elderly patients. ***Methods:*** We reviewed our database from November 1993 to March 2003 and retrospectively examined the clinical data of patients with histologically confirmed exocrine pancreatic carcinomas that were nonresectable but confined to the pancreatic region, who were treated with protracted 5-fluorouracil infusion (200 mg/m^2/day) and concurrent radiotherapy (50.4 Gy in 28 fractions over 5.5 weeks). We evaluated the outcome of patients ≥70 years and those <70 years. ***Results:*** There were 19 patients ≥70 and 39 patients <70. On pretreatment evaluation, the elderly patients showed lower serum albumin levels, lower transaminase levels, better ECOG performance status, more frequent body weight loss and less frequent abdominal and/or back pain with the administration of morphine than the younger patients. There were no significant differences in the frequency of severe toxicity. Neither the response rate nor the incidence of treatment discontinuation differed significantly between the two groups. The median survival time was longer in the elderly patients than in the younger patients (11.3 vs. 9.5 months, $p = 0.04$). ***Conclusions:*** With careful patient selection, chemoradiotherapy can be one of the treatment options for locally advanced pancreatic carcinoma in elderly patients.

Introduction

The prognosis of patients with pancreatic carcinoma is extremely poor because of difficulty in the early detection of this disease and the ineffectiveness of nonsurgical treatments. For patients with locally nonresectable disease, the results of previous randomized trials indicated that concurrent external beam radiation therapy (EBRT) and 5-fluorouracil (5-FU) therapy resulted in significantly better survival compared with EBRT alone [1, 2] or chemotherapy alone [3]. However, this combination treatment sometimes induces intolerable toxic effects, and approximately 10–20% of patients cannot complete the scheduled course of treatment [4, 5]. Consequently, this treatment is considered to be frequently contraindicated in elderly patients, who are thought to be less likely to tolerate its potential toxicity than younger patients.

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Furthermore, many physicians believe that pancreatic carcinoma is less treatable in the elderly because of the presence of comorbid illnesses. On the other hand, it was reported that elderly patients often tolerate aggressive chemotherapy or radiotherapy for other carcinomas as well as their younger counterparts [6–16].

Some studies have shown that for resectable pancreatic carcinoma, pancreatic resections can be performed for the elderly with acceptable morbidity and mortality rates and possible long-term outcome [17–25]. However, in locally advanced pancreatic carcinoma treated with chemoradiotherapy, the tolerability, efficacy of treatment and long-term outcome have not been discussed extensively.

The current retrospective analysis examines the outcome and tolerability of elderly patients (i.e. those aged ≥70 years) within our database. The main purposes of this examination were to determine if the outcome for elderly patients was different from that for younger patients and to characterize the toxicity experienced by the elderly patients.

Methods

We reviewed the database of the Hepatobiliary and Pancreatic Oncology Division of the National Cancer Center Hospital from November 1993 to March 2003. In this retrospective analysis, we examined the clinical data of all patients who met the following requirements: (1) histological diagnosis of exocrine pancreatic carcinoma, (2) nonresectable disease confined to the pancreatic region, (3) treatment with protracted 5-FU infusion and concurrent radiotherapy, and (4) absence of prior treatment for pancreatic carcinoma. We divided the patients into two groups according to age, those ≥70 years and those <70 years. We evaluated the patient characteristics, toxicities, efficacies and survival in both groups.

Treatment was performed according to the treatment protocol of our division; radiotherapy was delivered via a microtron (MM22, Scanditronix, Upsala, Sweden) with 10- or 14-MV X-rays or a racetrack microtron (MM50, Scanditronix) with 25-MV X-rays. A total dose of 50.4 Gy was delivered in 28 fractions over 5.5 weeks. All patients had treatment planning computed tomography (CT) scans (X-vision, Toshiba, Tokyo, Japan), and FOCUS (Computerized Medical Systems, St. Louis, Mo., USA) was used as a radiotherapy treatment planning system. The clinical target volume included the primary tumor, nodal involvement detected by CT scan, and regional draining and para-aortic lymph nodes, which included the peripancreatic nodes, celiac and superior mesenteric axes. The planning target volume was defined as the clinical target volume plus a 10-mm margin. Four field techniques (anterior, posterior and opposed lateral fields) were used. The spinal cord dose was maintained below 45 Gy, ≥50% of the liver was limited to ≤30 Gy, and ≥50% of both kidneys was limited to ≤20 Gy. 5-FU was given from the first day of radiation and continued through the entire course of radiation at a dose of 200 mg/m^2/day through a central venous catheter. Patients were admitted to the hospital during chemoradiotherapy. Within 8 weeks after the completion of chemoradiotherapy, maintenance chemotherapy was delivered on an outpatient basis and continued until disease progression. For the maintenance chemotherapy, we used a weekly administration of 5-FU (500 mg/m^2, 30-min infusion) before the approval of gemcitabine for pancreatic carcinoma in Japan (April 2001), and thereafter, we used weekly administration of gemcitabine (1,000 mg/m^2, 30-min infusion) 3 times every 4 weeks.

During chemoradiotherapy, the toxicity of the treatment was scored weekly according to the World Health Organization criteria [26]. Both radiotherapy and chemotherapy were suspended when ≥grade 3 toxicities other than anorexia, fatigue, nausea/vomiting, constipation and hyperglycemia occurred and were resumed when recovery to grade 2 toxicity levels was achieved. If there was a total delay of 2 weeks due to toxicity for any reason, the combined treatment was discontinued. In this retrospective analysis, we obtained the information regarding adverse events about the subjective symptoms from the doctor's record in as much detail as possible. As a rule, follow-up CT was performed within 1 week after the completion of chemoradiotherapy and every 2 months thereafter to evaluate the objective tumor response with reference to the World Health Organization criteria.

Statistics

Frequencies in 2 × 2 and larger contingency tables of the patient characteristics, response rates and toxicities were compared with the χ^2 or Fisher's exact test. Distributions of continuous variables were compared with the Mann-Whitney test. Overall survival was measured from the first day of treatment, and the survival curves were calculated according to the Kaplan-Meier method. The log rank test was used to detect differences between the curves. All p values in this study were of the two-tailed type. Significance was defined as a p value of 0.05 or less. Statistical analyses were performed with Stat View version 5.0.

Results

One hundred and ninety-nine patients with locally advanced pancreatic carcinoma admitted to the Hepatobiliary and Pancreatic Oncology Division of the National Cancer Center Hospital from November 1993 to March 2003. Thirty-nine patients were ≥70 years and 160 were <70 years. Nineteen (49%) of the 39 patients ≥70 and 39 (24%) of the 160 of those <70 met the above-mentioned conditions. The remaining 141 patients were excluded from this analysis. One hundred and thirty-eight received other anticancer treatments including chemoradiotherapy using other regimens (130), systemic chemotherapy (7) and radiotherapy alone (1). Three patients underwent only the best supportive care. The patient characteristics are shown in table 1 and the pretreatment laboratory data are shown in table 2. The male-to-female ratio was 1.7:1 in the elderly patients and 1.4:1 in the younger patients.

Table 1. Patient characteristics

	≥70 years	<70 years	p
Patients	19	39	
Age			
Median	75	60	
Range	70–86	35–69	
Sex			0.78
Male	12 (63)	23 (59)	
Female	7 (37)	16 (41)	
ECOG PS			0.004
0	6 (32)	1 (3)	
1	11 (58)	36 (92)	
2	2 (11)	2 (5)	
Diabetes mellitus	9 (47)	10 (26)	0.14
Abdominal and/or back pain[a]	3 (16)	19 (49)	0.02
Biliary drainage	4 (21)	8 (21)	>0.99
Regional lymph node	11 (58)	22 (56)	>0.99
Body weight loss[b]	14 (74)	24 (62)	0.20
Tumor location			0.42
Uncus	1 (5)	5 (13)	
Head	12 (63)	25 (64)	
Body	5 (26)	9 (23)	
Tail	1 (5)	0 (0)	
Treatment start			>0.99
Before April 2001[c]	10 (53)	21 (54)	
After April 2001[c]	9 (47)	18 (46)	

Figures in parentheses are percentages. ECOG = Eastern Cooperative Oncology Group.

[a] Abdominal and/or back pain: with consumption of morphine.

[b] Body weight loss: more than 7% of previous body weight within 6 months.

[c] April 2001: approval of gemcitabine.

Table 2. Pretreatment laboratory data

	≥70 years	<70 years	p
Albumin, g/dl	3.6 (3.0–4.3)	3.8 (3.1–4.5)	0.002
AST, IU/l	19 (11–66)	23 (10–274)	0.04
ALT, IU/l	17 (9–136)	32 (6–332)	0.01
Total bilirubin, mg/dl	0.7 (0.3–1.3)	0.6 (0.2–3.7)	0.20
CA19-9, U/ml	769.5 (3–27,000)	624.0 (4–6,310)	0.06
CEA, ng/ml	6.9 (2.1–76.4)	4.9 (0.7–1,620)	0.11

AST = Aspartate aminotransferase; ALT = alanine aminotransferase; CA19-9 = carbohydrate antigen 19-9; CEA = carcinoembryonic antigen.

Table 3. Response to chemoradiotherapy

	≥70 years	<70 years	p
Complete response	0 (0)	0 (0)	
Partial response	2 (11)	2 (5)	
No change	14 (74)	28 (72)	
Progressive disease	3 (16)	7 (18)	
Not evaluable	0 (0)	2 (5)	0.60

Figures in parentheses are percentages.

In the elderly patients, there were 6 patients (32%) who had an ECOG performance status (PS) of 0, but there was only 1 such patient (3%) among the younger patients ($p = 0.004$). The incidence of patients who had abdominal or back pain with consumption of morphine was smaller in the elderly patients ($p = 0.02$). There was no significant difference between the younger and elderly patients with regard to the period prior to treatment initiation (before or after the gemcitabine approval) ($p > 0.99$). The serum albumin level and transaminase levels were lower in the elderly patients. The other patient characteristics of those ≥70 years were generally similar to those of the younger patients.

The results of the treatment outcome are shown in table 3. Even though this study was conducted retrospectively, the antitumor response in CT was obtained in all but 2 younger patients who were transferred to another hospital before the completion of treatment. The laboratory data were also maintained for all patients, whose blood examinations were performed at least weekly. Four subjects among the elderly patients (21%) suspended the chemoradiotherapy during the schedule, as did 11 (28%) among the younger patients. One elderly patient (5%) discontinued chemoradiotherapy, as did 5 (13%) of the younger patients. Chemoradiotherapy was discontinued because of patient request due to unacceptable toxicities such as fatigue (1 younger patient), nausea/vomiting (3 younger patients and 1 elderly patient) and patient refusal (1 younger patient). A partial response was obtained in 2 (11%) elderly and 2 (5%) younger patients. Fourteen (74%) elderly patients and 28 (72%) younger patients showed no change. The survival curves are shown in figure 1. The median survival time was longer for the elderly patients than for younger patients (11.3 months in the elderly patients, 9.5 months in the younger patients, $p = 0.04$). The longest survivor in both groups was a 71-year-old male who survived 60.1 months (5.0 years) after the initiation of treatment.

Table 4. Toxicity in patients receiving chemoradiotherapy

	Grades 1–4		p	Grades 3 and 4		p
	≥70 years	<70 years		≥70 years	<70 years	
Leukocytes	9 (47)	20 (51)	>0.99	1 (5)	2 (5)	>0.99
Hemoglobin	8 (42)	16 (41)	>0.99	0 (0)	0 (0)	–
Neutrophils	3 (16)	12 (31)	0.37	0 (0)	0 (0)	–
Platelets	4 (21)	4 (10)	0.48	0 (0)	0 (0)	–
Albumin	10 (53)	16 (41)	0.58	0 (0)	0 (0)	–
AST	4 (21)	8 (21)	>0.99	0 (0)	2 (5)	0.81
ALT	3 (16)	15 (38)	0.15	0 (0)	3 (8)	0.54
Total bilirubin	2 (11)	3 (8)	>0.99	0 (0)	1 (3)	>0.99
Creatinine	2 (11)	0 (0)	0.2	0 (0)	0 (0)	–
Nausea	11 (58)	34 (87)	0.03	2 (11)	13 (33)	0.12
Vomiting	4 (21)	19 (49)	0.07	0 (0)	1 (3)	>0.99
Anorexia	16 (84)	35 (90)	0.9	6 (32)	22 (56)	0.13
Stomatitis	3 (16)	2 (5)	0.85	1 (5)	0 (0)	0.71
Diarrhea	4 (21)	13 (33)	0.47	0 (0)	2 (5)	0.81
Fatigue	3 (16)	13 (33)	0.28	0 (0)	1 (3)	>0.99

Figures in parentheses are percentages. AST = Aspartate aminotransferase; ALT = alanine aminotransferase.

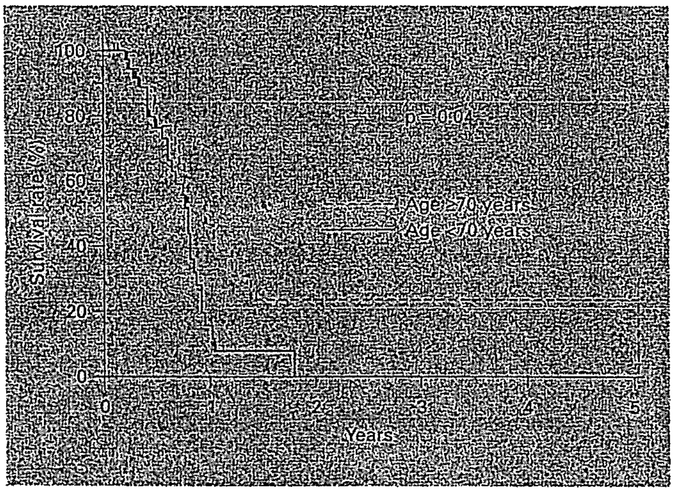

Fig. 1. Overall survival curves for patients ≥70 years (n = 19) and those for patients <70 years (n = 38).

The percentages of overall toxicities (grades 1–4) and severe toxicities (grades 3 and 4) are listed in table 4. Although the incidence of nausea (grades 1–4) was significantly higher in the younger patients, there were no significant differences in the incidence of other overall toxicities or all severe toxicities. The toxicities of both groups were generally mild and reversible. One younger patient died from a fungal infection of the lung due to pneumothorax which occurred as a complication of the insertion of a central venous catheter. There was no conspicuous late toxicity in either group.

Discussion

Based on previous randomized trials [1–3], concurrent EBRT and 5-FU result in significantly better survival compared with EBRT alone or chemotherapy alone and are generally accepted as the standard treatment for locally advanced pancreatic carcinoma. However, this treatment restrains patients for more than 1.5 months during treatment. Furthermore, the life expectancy for the majority of these patients is still short, with a median survival of approximately 10–11 months. The poor prognosis and long duration of treatment makes us hesitant to indicate chemoradiotherapy for patients with locally advanced pancreatic carcinoma, especially for patients at high risk for complications. Elderly patients have been generally considered a high-risk population for chemoradiotherapy due to a number of physiological and pharmacological reasons. For example, diminished bone marrow cellularity can potentially result in decreased tolerance to myelosuppressive therapies. In addition, a decrease in hepatic and renal function may reduce the efficiency of drug metabolism and excretion, resulting in greater toxic potential.

However, in this study, no differences were found in the response rate, incidence of treatment discontinuation and toxicity profile, except for nausea, between the two groups. The median survival time was significantly longer in the elderly patients than in the younger patients. The most important reason for the favorable results of the elderly patients may be the careful selection of patients. Ikeda et al. [27] reported that a good PS was one of the independent favorable prognostic factors in patients with locally advanced pancreatic carcinoma receiving chemoradiotherapy. In our study, 32% of the patients ≥70 had an ECOG PS of 0, as opposed to 3% of those <70. Since this was a retrospective analysis, indication according to a physician's decision might have been different for younger and for elderly patients, only allowing the elderly patients in very good condition to receive chemoradiotherapy. As a result, this may be a comparison of elderly patients with a very good PS and younger patients with a less good or average PS.

An imbalance in the incidence of patients with abdominal pain between the two groups might also have affected the treatment outcome in our study. According to the report of Kelsen et al. [28], unresectable pancreatic carcinoma patients with abdominal pain had a median survival of 4.7 months, whereas the median survival among patients without such pain was 8.3 months.

In this study, there was no significant difference between the younger patients and the elderly patients with regard to the ratio of the patients who received maintenance chemotherapy using gemcitabine. Although it is possible that maintenance therapy had some effect on survival, the survival time did not differ significantly between the gemcitabine maintenance chemotherapy group and the 5-FU maintenance chemotherapy group in this study (data not shown).

The mild toxicity of this treatment may be another favorable factor for elderly patients [4]. This study showed that severe toxicities except anorexia were observed infrequently in both groups and that discontinuation of the treatment was required in only 1 elderly patient. Protracted 5-FU infusion with concurrent radiotherapy, which is considered a less toxic treatment than radiotherapy and bolus 5-FU [29, 30], is feasible even in elderly patients.

Krzyzanowska et al. [31] reported an attractive retrospective cohort study in 1,696 patients diagnosed with locally advanced pancreatic carcinoma. According to the report, older age was associated with a lower likelihood of receiving carcinoma-directed therapy, much less of a combined therapy such as chemoradiotherapy. However, Cox proportional hazard models showed that carcinoma-directed therapy, including chemoradiotherapy, has the potential to prolong the survival of elderly patients with locally advanced pancreatic carcinoma. These findings, which suggest that chemoradiotherapy can be an optimal treatment option for locally advanced pancreatic carcinoma in elderly patients, are supported by the results of our study.

Since this study was conducted retrospectively, the results do nothing more than suggest possibilities of the efficacy of the 5-FU-based chemoradiotherapy for selected elderly patients with locally advanced pancreatic carcinoma. To identify the benefit of the treatment in elderly patients, we must design a large prospective study. In summary, this study demonstrates that chemoradiotherapy for locally advanced pancreatic carcinoma is well tolerated and does not lead to an increase in treatment interruption or discontinuation in elderly patients. We conclude that, with careful patient selection, chemoradiotherapy can be considered an appropriate treatment for elderly patients.

References

1 Moertel CG, Childs DS Jr, Reitemeier RJ, et al: Combined 5-fluorouracil and supervoltage radiation therapy of locally unresectable gastrointestinal cancer. Lancet 1969;ii:865–867.

2 Moertel CG, Frytak S, Hahn RG, et al: Therapy of locally unresectable pancreatic carcinoma: A randomized comparison of high dose (6000 rads) radiation alone, moderate dose radiation (4000 rads + 5-fluorouracil), and high dose radiation + 5-fluorouracil: The Gastrointestinal Tumor Study Group. Cancer 1981;48:1705–1710.

3 Treatment of locally unresectable carcinoma of the pancreas: Comparison of combined-modality therapy (chemotherapy plus radiotherapy) to chemotherapy alone. Gastrointestinal Tumor Study Group. J Natl Cancer Inst 1988;80:751–755.

4 Ishii H, Okada S, Tokuuye K, et al: Protracted 5-fluorouracil infusion with concurrent radiotherapy as a treatment for locally advanced pancreatic carcinoma. Cancer 1997;79:1516–1520.

5 Okusaka T, Okada S, Tokuuye K, et al: Lack of effectiveness of radiotherapy combined with cisplatin in patients with locally advanced pancreatic carcinoma. Cancer 2001;91:1384–1389.

6 Ludbrook JJ, Truong PT, MacNeil MV, et al: Do age and comorbidity impact treatment allocation and outcomes in limited stage small-cell lung cancer? A community-based population analysis. Int J Radiat Oncol Biol Phys 2003;55:1321–1330.

7 Kim HK, Silver B, Li S, et al: Hodgkin's disease in elderly patients (> or = 60): Clinical outcome and treatment strategies. Int J Radiat Oncol Biol Phys 2003;56:556–560.
8 Begg CB, Carbone PP: Clinical trials and drug toxicity in the elderly. The experience of the Eastern Cooperative Oncology Group. Cancer 1983;52:1986–1992.
9 Arias F, Duenas M, Martinez E, et al: Radical chemoradiotherapy for elderly patients with bladder carcinoma invading muscle. Cancer 1997;80:115–120.
10 Popescu RA, Norman A, Ross PJ, et al: Adjuvant or palliative chemotherapy for colorectal cancer in patients 70 years or older. J Clin Oncol 1999;17:2412–2418.
11 Pignon T, Gregor A, Schaake Koning C, et al: Age has no impact on acute and late toxicity of curative thoracic radiotherapy. Radiother Oncol 1998;46:239–248.
12 Sengelov L, Klintorp S, Havsteen H, et al: Treatment outcome following radiotherapy in elderly patients with bladder cancer. Radiother Oncol 1997;44:53–58.
13 Huguenin PU, Glanzmann C, Hammer F, Lutolf UM: Endometrial carcinoma in patients aged 75 years or older: Outcome and complications after postoperative radiotherapy or radiotherapy alone. Strahlenther Onkol 1992;168:567–572.
14 Zachariah B, Balducci L: Radiation therapy of the older patient. Hematol Oncol Clin North Am 2000;14:131–167.
15 Pignon T, Horiot JC, Van den Bogaert W, et al: No age limit for radical radiotherapy in head and neck tumours. Eur J Cancer 1996;32A:2075–2081.
16 Pignon T, Horiot JC, Bolla M, et al: Age is not a limiting factor for radical radiotherapy in pelvic malignancies. Radiother Oncol 1997;42:107–120.
17 DiCarlo V, Balzano G, Zerbi A, Villa E: Pancreatic cancer resection in elderly patients. Br J Surg 1998;85:607–610.
18 Fong Y, Blumgart LH, Fortner JG, Brennan MF: Pancreatic or liver resection for malignancy is safe and effective for the elderly. Ann Surg 1995;222:426–434.
19 Hannoun L, Christophe M, Ribeiro J, et al: A report of forty-four instances of pancreaticoduodenal resection in patients more than seventy years of age. Surg Gynecol Obstet 1993;177:556–560.
20 Hodul P, Tansey J, Golts E, et al: Age is not a contraindication to pancreaticoduodenectomy. Am Surg 2001;67:270–275; discussion 275–276.
21 Kayahara M, Nagakawa T, Ueno K, et al: Pancreatic resection for periampullary carcinoma in the elderly. Surg Today 1994;24:229–233.
22 Richter A, Schwab M, Lorenz D, et al: Surgical therapy of pancreatic carcinoma in elderly patients over 70. Langenbecks Arch Chir Suppl Kongressbd 1996;113:492–494.
23 Magistrelli P, Masetti R, Coppola R, et al: Pancreatic resection for periampullary cancer in elderly patients. Hepatogastroenterology 1998;45:242–247.
24 Spencer MP, Sarr MG, Nagorney DM: Radical pancreatectomy for pancreatic cancer in the elderly. Is it safe and justified? Ann Surg 1990;212:140–143.
25 Bathe OF, Levi D, Caldera H, et al: Radical resection of periampullary tumors in the elderly: Evaluation of long-term results. World J Surg 2000;24:353–358.
26 Miller AB, Hoogstraten B, Staquet M, Winkler A: Reporting results of cancer treatment. Cancer 1981;47:207–214.
27 Ikeda M, Okada S, Tokuuye K, et al: Prognostic factors in patients with locally advanced pancreatic carcinoma receiving chemoradiotherapy. Cancer 2001;91:490–495.
28 Kelsen DP, Portenoy R, Thaler H, et al: Pain as a predictor of outcome in patients with operable pancreatic carcinoma. Surgery 1997;122:53–59.
29 Leichman CG: Prolonged infusion of fluorinated pyrimidines in gastrointestinal malignancies: A review of recent clinical trials. Cancer Invest 1994;12:166–175.
30 Poen JC, Collins HL, Niederhuber JE, et al: Chemo-radiotherapy for localized pancreatic cancer: Increased dose intensity and reduced acute toxicity with concomitant radiotherapy and protracted venous infusion 5-fluorouracil. Int J Radiat Oncol Biol Phys 1998;40:93–99.
31 Krzyzanowska MK, Weeks JC, Earle CC: Treatment of locally advanced pancreatic cancer in the real world: Population-based practices and effectiveness. J Clin Oncol 2003;21:3409–3414.

Severe Drug Toxicity Associated with a Single-Nucleotide Polymorphism of the *Cytidine Deaminase* Gene in a Japanese Cancer Patient Treated with Gemcitabine plus Cisplatin

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Abstract **Purpose:** We investigated single-nucleotide polymorphisms of the cytidine deaminase gene (*CDA*), which encodes an enzyme that metabolizes gemcitabine, to clarify the relationship between the single-nucleotide polymorphism 208G>A and the pharmacokinetics and toxicity of gemcitabine in cancer patients treated with gemcitabine plus cisplatin.

Experimental Design: Six Japanese cancer patients treated with gemcitabine plus cisplatin were examined. Plasma gemcitabine and its metabolite 2′,2′-difluorodeoxyuridine were measured using an high-performance liquid chromatography method, and the *CDA* genotypes were determined with DNA sequencing.

Results: One patient, a 45-year-old man with pancreatic carcinoma, showed severe hematologic and nonhematologic toxicities during the first course of chemotherapy with gemcitabine and cisplatin. The area under the concentration-time curve value of gemcitabine in this patient (54.54 μg hour/mL) was five times higher than the average value for five other patients (10.88 μg hour/mL) treated with gemcitabine plus cisplatin. The area under the concentration-time curve of 2′,2′-difluorodeoxyuridine in this patient (41.58 μg hour/mL) was less than the half of the average value of the five patients (106.13 μg hour/mL). This patient was found to be homozygous for 208A (Thr^{70}) in the *CDA* gene, whereas the other patients were homozygous for 208G (Ala^{70}).

Conclusion: Homozygous 208G>A alteration in *CDA* might have caused the severe drug toxicity experienced by a Japanese cancer patient treated with gemcitabine plus cisplatin.

Gemcitabine (2′,2′-difluorodeoxycytidine) is a deoxycytidine analogue that is efficacious against non-small cell lung cancer and pancreatic carcinoma, as a single agent or in platinum combination therapy (1, 2). Its major adverse effects are hematologic toxicity, weakness, and emesis, and its dose-limiting toxicity is hematologic toxicity, including leukocytopenia, anemia, and thrombocytopenia (1). Single-agent and platinum combination gemcitabine therapy is relatively well tolerated, but hospitalization is occasionally required due to significant hematologic toxicity (1, 2), and it has been difficult to predict the toxicity.

Gemcitabine is activated by intracellular phosphorylation to gemcitabine monophosphate by deoxycytidine kinase, which is subsequently phosphorylated to the higher-order phosphates, gemcitabine diphosphate followed by gemcitabine triphosphate. Gemcitabine triphosphate can be incorporated into DNA followed by one more deoxynucleotide, after which DNA polymerization stops. This process is referred to as "masked chain termination" (3, 4).

Gemcitabine and gemcitabine monophosphate are deaminated to the inactive metabolite 2′,2′-difluorodeoxyuridine (dFdU) and 2′,2′-difluorodeoxyuridine monophosphate by cytidine deaminase (CDA) and dCMP deaminase, respectively. Multiple mechanisms potentiate the activity of gemcitabine both by increased formation of active gemcitabine diphosphate and gemcitabine triphosphate and decreased elimination of gemcitabine, as follows: (a) gemcitabine diphosphate, through its inhibition of ribonucleotide reductase, depletes the deoxyribonucleotide pool available for DNA synthesis and repair; (b) the decreased concentration of dCTP activates deoxycytidine kinase, which accelerates phosphorylation of gemcitabine;

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and (c) an inactivating enzyme, dCMP deaminase, is inhibited by the decreased concentration of intracellular dCTP and increased concentration of gemcitabine triphosphate (5-7). Polymorphisms of the DNAs encoding the above enzymes may influence the pharmacokinetics and pharmacodynamics of gemcitabine.

To establish the medical guidelines for treatment based on individual genetic polymorphisms, we have launched multicenter, prospective, pharmacogenomic trials (as the Millenium Genome Project) of antineoplastic agents, such as gemcitabine, paclitaxel, irinotecan, and other commonly used drugs.

At the time point when 97 gemcitabine-treated patients had been recruited, we experienced extremely severe toxicities in one patient. Because this patient was coadministered cisplatin in addition to gemcitabine, we compared the clinical data, pharmacokinetics and *CDA* genotype between this patient and the other five control patients, who were also coadministered the two drugs.

Patients and Methods

Selection of patients and treatment schedule. Patients being treated with gemcitabine plus cisplatin were eligible for the trial if they met all of the following inclusion criteria: histologically or cytologically proven carcinoma, no prior treatment with gemcitabine, age above 20 years, Eastern Cooperative Oncology Group performance status between 0 and 2, absence of severe infectious or neurologic disease, and no evidence of heart or interstitial lung disease. Other requirements included adequate bone marrow function (WBC ≥3,000/μL, neutrophils ≥1,500/μL, and platelets ≥75,000/μL), hepatic function (serum total bilirubin ≤3 mg/d, aspartate aminotransferase and alanine aminotransferase less than five times the upper limit of normal), and renal function (serum creatinine within the upper limit of normal). The trial was approved by the Ethics Review Committees of the National Cancer Center Hospital and NIH Sciences, and oral and written informed consent was obtained from all patients before entering.

Gemcitabine was given to all patients at a dose of 1,000 mg/m^2 (30-minute infusion) on days 1, 8, and 15 and followed by 1 week of rest. If adequate bone marrow function (WBC ≥2,000/μL, neutrophils ≥1,000/μL, and platelets ≥70,000/μL) was confirmed, gemcitabine was given on days 8 and 15.

Cisplatin was given at a dose of 80 mg/m^2 (150-minute infusion) on day 1, immediately after gemcitabine. All patients received antiemetic prophylaxis with granisetron plus dexamethasone. Granulocyte-colony stimulating factor was not given routinely. The treatment schedule was repeated every 28 days until disease progression or unacceptable side effects occurred.

Toxicity was scored according to the National Cancer Institute Common Toxicity Criteria ver 2.0. A complete blood cell count and serum chemistry were repeated weekly. At the start of every new course, the dose was reevaluated according to toxicity. If the WBC count was <2,000/μL and the platelet count was <70,000/μL, then treatment was delayed until the recovery of bone marrow function. If grade 4 leukocytopenia, neutrocytopenia, or thrombocytopenia was observed in the previous course, the gemcitabine dose was reduced to 800 mg/m^2 in subsequent courses.

Blood sampling. Before the start of the treatment, a 5-mL heparinized blood sample was collected to measure CDA activity, and a 14-mL blood sample, to which EDTA was added, was collected to extract leukocyte DNA for genetic analysis. On day 1 of the first course, a 5-mL heparinized blood sample for gemcitabine and metabolite analysis in plasma was collected from the opposite arm before the infusion, at 3 minutes before the end of the infusion, and 15, 30, 60, 90, 120, and 240 minutes after the end of the infusion, and 50 μL of 10 mg/mL tetrahydrouridine (Wako Junyaku, Co., Ltd., Osaka, Japan) was immediately added to each of the samples. The samples were centrifuged at 3,000 × *g* for 5 minutes at 4°C, and the plasma was collected and stored at −70°C until analyzed.

Analysis of gemcitabine and its metabolite, 2′,2′-difluorodeoxyuridine. The concentrations of gemcitabine and dFdU in the plasma were determined by the method of Venook et al. with slight modifications (8). A 25 μL volume of 25 mg/mL 3′-deoxy-3′-fluoro-thymidine (Aldrich Chem. Co., St. Louis, MO) was added to an 0.25-mL aliquot of plasma sample containing 0.1 mg/mL tetrahydrouridine as an internal standard. After adding 1 mL of acetonitrile, the mixtures were centrifuged at 12,000 × *g* for 5 minutes, and the supernatant was evaporated to dryness under a nitrogen stream. The residue was dissolved in 0.25 mL of 15 mmol/L ammonium acetate buffer (pH 5.0), and the solution was filtered twice through Ultrafree-MC (0.45 μm; Millipore Corp., Billerica, MA) and Microcon YM-10 (10,000 MW; Amicon). Twenty microliters of sample were loaded into a high-performance liquid chromatography system (HP 1100 model) with diode array detection and electrospray-mass spectrometry detection. The chromatographic conditions were as follows: column, CAPCELL PACK C18 MG column (5 μm, 2.0 × 150 mm; Shiseido Co., Ltd., Tokyo, Japan) with a CAPCELL C18 MG S-5 guard cartridge (4.6 mm i.d. × 10 mm; Shiseido); column temperature, 40°C; mobile phase, 15 mmol/L ammonium acetate (pH 5.0)/methanol; running program of the mobile phase: 95:5 (0 minute), −75:25 (10-15 minutes), −60:40 (20-25 minutes), − 95:5 (30-40 minutes); flow rate: 0.3 mL/min; diode array detection: 268 nm for gemcitabine, 258 nm for dFdU, and 266 nm for 3′-deoxy-3′-fluoro-thymidine; electrospray-mass spectrometry: *m/z* 264 for gemcitabine, *m/z* 265 for dFdU, and *m/z* 245 for 3′-deoxy-3′-fluoro-thymidine. Detection and integration of chromatographic peaks were done by the HP Chemistation data analysis system (Hewlett-Packard, Les Ulis, France).

Pharmacokinetic analysis. Compartment model independent pharmacokinetic variables were calculated using WinNonlin software, ver. 4.1 (Pharsight Co., Mountain View, CA). The values are expressed as means ± SD, except for those of the patient with severe toxicity.

DNA sequencing. DNA used for sequencing was extracted from peripheral blood. All of the four exons of *CDA* were amplified from 100 ng of genomic DNA using multiplex primers listed in Table 1 (PCR). The PCR conditions have been described previously (9). After the second amplification for each exon, the PCR products were purified and directly sequenced on both strands with the sequencing primers listed in Table 1 (sequencing), as described previously (9). All variations were confirmed by repeating the sequence analysis from the first-round PCR with DNA. National Center for Biotechnology Information accession no. NT_004610.16 was used for the reference sequence.

Table 1. Primer sequences used for the analysis of the human *CDA* gene

Region	Forward primer (5'-3')	Reverse primer (5'-3')
PCR		
Exon 1	TCCACCCTCCAATTGAGATA	AGTCGGCAGGGTAGGAACATTC
Exon 2	TTGATGGGACACATTCAGACCA	CCGCTTTATGTTTCAATGCTGC
Exon 3	CTCTTTGACCTTTGTATTCCC	TTGACTCAGAAACGCCACTGTT
Exon 4	GCACTATGATCCAGGTACAA	TCAGCTCTCCACACCATAAGG
Sequencing		
Exon 1	TGAGACAGGGTCTGGCTCTCTGT	GTGCTTCACACTCTCCCTTA
	CAGTAGCGTGGCACCACCTTCT	CGCCTCTTCCTGTACATCTT
	ATGGCCCAGAAGCGTCCT	GGCCCCAGACACGATTGC
Exon 2	CCACCTTGTTTGGAGTAACC	CTGGCACATAGGAAGTCCAC
	TGGGATGAGTGCTGAGGATA	TGTGTAAGGAAGATGTTGGC
Exon 3	CTTCAGGACACAGTGGATCT	TTCCAGTGACTCATGCAAGC
Exon 4	ATGGTCATTCCCCTTTTACA	GTCCCTCCTAAGAGCTGCAA
		AGGCTGGAGTGTAATCTGGA

Results

We encountered a patient treated with gemcitabine and cisplatin who developed extremely severe toxicities (grade 4 neutropenia, thrombocytopenia, and stomatitis and grade 3 rash, fatigue, and febrile neutropenia). To clarify the cause of these life-threatening toxicities, we determined the plasma levels of gemcitabine and its metabolite, dFdU, and the genotypes of *CDA* encoding a major gemcitabine-metabolizing enzyme, cytidine deaminase, of this patient (patient 1) and the other five gemcitabine/cisplatin-administered patients (patients 2-6).

Pharmacokinetics. Plasma concentration-time profiles of gemcitabine and dFdU are shown in Figs. 1 and 2, and pharmacokinetic variables are summarized in Table 2. The maximum plasma gemcitabine concentration (C_{max}) and area under the concentration-time curve of patient 1 were about twice and five times higher, respectively, than the average values of patients 2 to 6. In patient 1, gemcitabine clearance was decreased to one fifth of the average value of the other five cases, and the terminal phase half-life ($T_{1/2}$) of gemcitabine was four times longer than the average value in patients 2 to 6. The C_{max} and area under the concentration-time curve of dFdU in patient 1 were one third and one half, respectively, of the average values of patients 2 to 6. The area under the concentration-time curve ratio (dFdU/gemcitabine) of patient 1 was about one tenth of the average value in patients 2 to 6.

Genotypes. The results of *CDA* genotyping analysis are shown in Table 3. We only found three known single nucleotide polymorphisms (SNP) in the coding regions in these patients. Patient 1 was homozygous for 208G>A (Ala^{70}Thr) in exon 2 (10), but had homozygous wild-type alleles for the other SNPs in exons 1 and 4. All of the other patients carried the homozygous wild-type alleles in exon 2. Thus, it was assumed that the increased plasma gemcitabine levels in patient 1 might have been caused by the Ala^{70}Thr substitution in cytidine deaminase.

Discussion

There was no nephrotoxicity or neurotoxicity in patient 1, which is specifically associated with cisplatin (11). In addition,

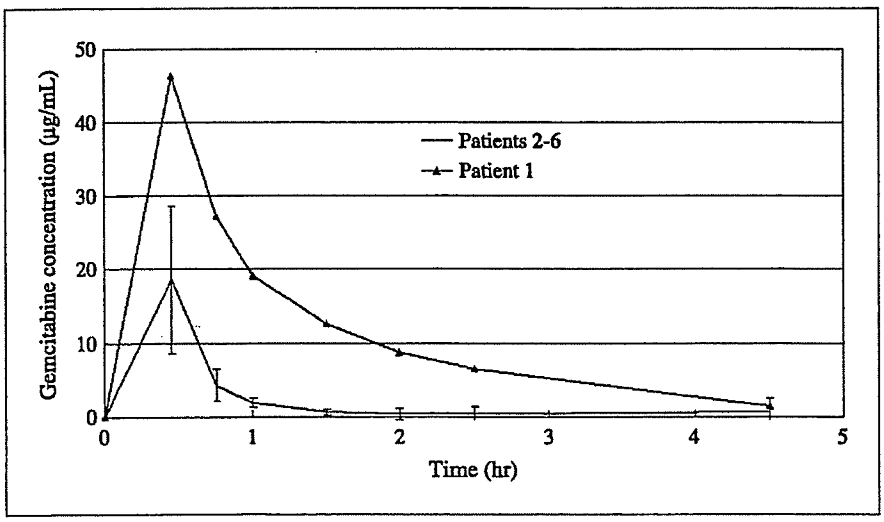

Fig. 1. Plasma disposition curve for gemcitabine in patient 1 (▲) and mean curve for patients 2 to 6 (*solid line*). Bars, SD.

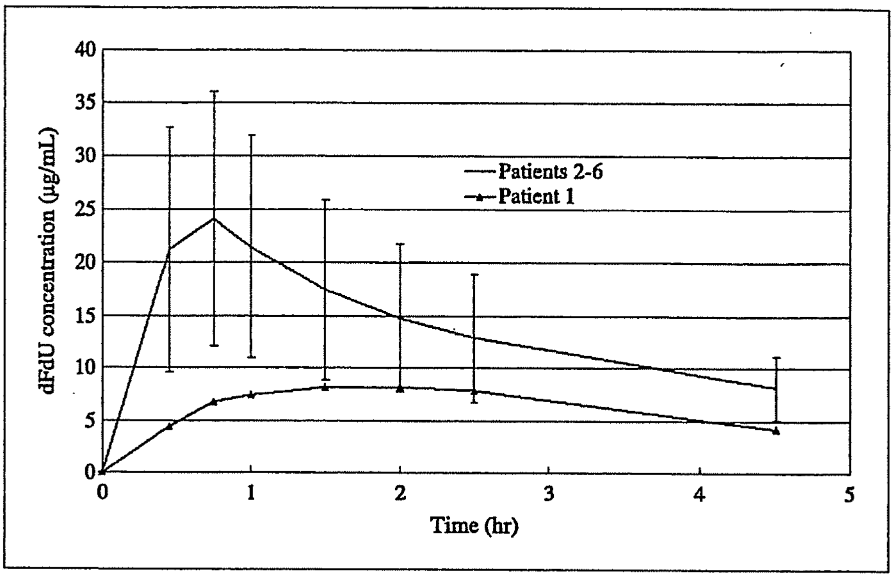

Fig. 2. Plasma disposition curve for dFdU in patient 1 (▲) and mean curve for patients 2 to 6 (*solid line*). Bars, SD.

the skin rash observed in patient 1 is common (with a reported 30.1% occurrence) in patients treated with gemcitabine in single-agent therapy (1). Therefore, we considered that the severe toxicity profile observed in patient 1 was mainly caused by the administration of gemcitabine.

Because the average pharmacokinetic profiles of gemcitabine and dFdU in patients 2 to 6 were almost the same as the population pharmacokinetic profiles in phase I and late phase II trials in Japan (12-14), the pharmacokinetic profiles of patients 2 to 6 can be regarded as standard for a Japanese population. Therefore, the plasma gemcitabine levels of patient 1 were remarkably high. Because the DLST of gemcitabine and cisplatin were negative in patient 1, the toxicities, especially a severe systemic rash including stomatitis and purpura, were unlikely to have been caused by drug allergies, such as Stevens-Johnson syndrome. Thus, the exposure to increased levels of gemcitabine is most likely responsible for the severe toxicities experienced in patient 1.

The patient backgrounds showed no major difference in age, body surface area, and performance status among patients with and without severe toxicities; age ranged from 45 to 69 years, the bovine serum albumin ranged from 1.42 to 1.78 m^2, and Eastern Cooperative Oncology Group performance status ranged from 0 to 1 (patient 1: 45 years, 1.78 m^2, performance status 0). None of the patients had received any prior chemotherapy or radiotherapy. It was unlikely that the patient backgrounds other than the *CDA* genotype caused the abnormal pharmacokinetics observed in patient 1.

Patient 1 was homozygous for the SNP 208G>A ($Ala^{70}Thr$), and all of the other patients carried the homozygous wild-type allele. Patient 1 carried no other known nonsynonymous and synonymous *CDA* polymorphisms (79A>C and 435C>T, respectively). The variant CDA enzyme with Thr^{70} was reported to show 40% and 32% of the activity of the wild-type for cytidine and 1-β-D-arabinofuranosylcytosine substrates in an *in vitro* experiment, respectively (10). Thus, the

Table 2. Compartment-independent pharmacokinetic variables of gemcitabine and its metabolite, dFdU

	Patient 1	Patients 2-6 (mean ± SD)
Gemcitabine		
C_{max} (µg/mL)	46.42	22.28 ± 5.08
AUC_∞ (µg hour/mL)	54.54	10.88 ± 1.64
Cl (L per h per m²)	18.34	93.17 ± 15.61
$T_{1/2}$ (h)	0.97	0.26 ± 0.03
V_z (L/m²)	25.62	35.2 ± 7.47
dFdU		
C_{max} (µg/mL)	8.19	28.75 ± 4.09
AUC_∞ (µg h/mL)	41.58	106.13 ± 31.44
Cl/F (L per h per m²)	24.05	10.04 ± 2.98
$T_{1/2}$ (h)	2.17	2.46 ± 0.52
V_z (L/m²)	75.4	34.29 ± 6.6
AUC ratio (dFdU/gemcitabine)	0.76	9.68 ± 2.05

Abbreviations: C_{max}, maximum plasma concentration; AUC_∞, area under the concentration-time curve; Cl, clearance; $T_{1/2}$, terminal-phase half-life; V_z = Dose / (λ_z × AUC); λ_z, elimination rate constant at terminal phase; *F*, metabolite fraction (*F* can be assumed to lie between 0.90 and 0.95).

Table 3. Genotypes of the three known polymorphic loci in exons of the *CDA* gene

Patient	Exon 1, 79A>C, K27Q	Exon 2, 208G>A, A70T	Exon 4, 435C>T, T145T (silent)
1	A/A	A/A	C/C
2	A/C	G/G	C/T
3	A/A	G/G	C/C
4	A/A	G/G	C/T
5	A/A	G/G	C/C
6	C/C	G/G	C/T