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aside from the competing risks problems, this final result depends heavily on the correct specification of the parametric model forms, which are usually unknown in most epidemiologic applications, while our IPTW adjustments of confounding do not need such parametric assumptions.

5. DISCUSSION

The problem of analyzing and interpreting data concerning competing risks continues to be one of the most important and vexing in biostatistical practice. The analyses of competing risks can be made using observable population parameters. An important observable quantity is the cumulative incidence functions based on the cause-specific hazards [25–28]. Alternatively, in this paper, we presented a method for estimation and comparison of treatment group-specific marginal survival curves of time to event data in the presence of dependent competing risks. The parameter of interest in our analysis is the marginal survival distribution, which is the net probability of time to event if only one cause of event acted on a population [4, 29, 30]. The ability to isolate the effect of one risk acting a population is attractive, especially if the focus of a study is to evaluate the effect of an intervention that is targeted at reducing incidence from that specific cause. Much of the literature on competing risks approaches such a problem by assuming the existence of latent survival times for each subject, that is, the estimation of event rates for certain types of event given the removal of some or all other event types. However, the net probabilities are hypothetical quantities and not directly observable in a population. Only observable quantities are their bounds, which allow for any possible dependence structure and will often be too wide to be of value [9–11]. Therefore, it is necessary to assume some model concerning the censoring process such as (1) to identify the net probabilities from the available information on the observables including covariate histories.

The proposed method is a straightforward extension of Robins and Finkelstein [14] for settings with two or more reasons for censoring. The application of the proposed methodology to the KLIS data suggested that the IPCW marginal incidence for CHD was almost the same as the lower bound. We included as many covariates as possible to predict the conditional probabilities of having remained uncensored. This result may suggest that there was little evidence of dependent competing risks in the KLIS data. In many studies, because we cannot safely say that the dependent censorings have not occurred, it is important to conduct the analysis accounting for the dependent censorings as well as the standard one and to compare their results. When their results differ remarkably, the reasons for drop-outs are examined in detail and the effects on the final conclusion in the study should be discussed. On the other hand, when the results are nearly the same ones like the KLIS data, dependent censorings observed in the study does not cause a severe selection bias attributable to the covariates and the results from the standard analysis are robust in relation to the censorings.

It must be noted that the low incidences for the competing events do not always mean that the IPCW estimate will be close to the lower bound. The issue of interest in our analysis is whether the competing events are informative for their unobserved CHD events and whether the relation can be explained by the observed covariate histories. If they have much information on their unobserved CHD events and the covariates are available to explain the dependency, the IPCW estimate will be close to the upper bound without regard to the incidence of competing events. On the other hand, if they have little information on their unobserved CHD events, the IPCW estimate will be close to the lower bound.

For example, in a cohort of 100 subjects, suppose that at the time of 2 (years) from the start of follow-up, 5 subjects and 3 subjects experienced CHD events and non-CHD deaths, respectively, and that the remaining 92 subjects were censored at the end of study (time = 5). In this hypothetical data, the upper and lower bound

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of the incidence rate is $8/(5 \times 2 + 3 \times 2 + 92 \times 5) = 1/59.5$ and $5/(5 \times 2 + 3 \times 5 + 92 \times 5) = 1/97$, respectively. Under the independent censoring, the incidence rate is $5/(5 \times 2 + 3 \times 2 + 92 \times 5) = 1/95.2$. If a binary covariate V is available and the distribution is $V = 1$ for both 5 CHD and 3 non-CHD events and $V = 0$ for 92 censored events at time = 5, the IPCW weights are 8/5 for the former events and 1 for the latter events. In this scenario of dependent competing risks, the IPCW incidence rate is $5 \times 1.6/(5 \times 2 \times 1.6 + 3 \times 2 \times 1.6 + 92 \times 5 \times 1) = 1/60.7$, which is almost the same as the upper bound. On the other hand, if the distribution of V is $V = 1$ for 5 CHD events and $V = 0$ for the other events, the IPCW weights are 1 for the former events, and are 95/92 (time $\leqslant 2$) and 1 ($2 <$ time $\leqslant 5$) for the latter events. In this scenario of independent competing risks, the IPCW incidence rate is $5 \times 1/(5 \times 2 \times 1 + 95 \times 2 \times 95/92 + 92 \times 3 \times 1) = 1/96.4$, which is almost the same as the lower bound. For more formal explanations, see Scharfstein and Robins [15] and Scharfstein *et al.* [31], in which the relation between the IPCW estimator and the bounds is discussed. They showed that, as the censoring bias parameters α in (2) goes to $\pm\infty$ (although they consider the case where the cause-specific hazard of censoring depends also on the possibly unobserved event time T given $\overline{\mathbf{V}}_t$), the resulting IPCW estimator will converge to the bounds.

Our results are based on a nonidentifiable assumption concerning the residual dependence between time to events and competing risks due to unmeasured factors. The ordinary Kaplan–Meier estimator does not utilize recorded information on time-dependent covariates $\overline{\mathbf{V}}_t$ and assumes the independence among competing risks, while the IPCW one utilizes such information and assumes the conditional independence among them. However, because causal interpretation of IPCW estimates depends on the correctness of assumption (1), making the censoring process ignorable is more important than fitting a parsimonious model in (2). As Joffe *et al.* [32] have described in the modeling of competing causes of death, the aggregation of censoring by competing causes may obscure important differences in the effect of various predictors on each type of censoring and so lead to misspecification of the model for censoring. Therefore, we fitted separate models for each type of censoring, where the treatment group-specific baseline hazard and regression parameters were assumed for each competing risk. Furthermore, in the KLIS, many clinically important time-dependent factors were measured and all of them were used as covariates to predict the probability of remaining in the study. Therefore, there will be a certain degree of validity in our IPCW estimates.

Otherwise, it will be necessary to develop sensitivity analysis methodology to investigate the sensitivity of our inferences to the fundamental assumption (1) of no unmeasured confounders. This sensitivity analysis will be particularly important for our data, in which the IPCW marginal incidence was almost the same as the lower bound. A simple and easy sensitivity analysis is to generate a hypothetical prognostic factor both for CHD and for competing events and to include the factor in the prediction of the conditional probabilities of uncensored. In the KLIS data, a hypothetical binary time-dependent covariate with a hazard ratio of 40.0 for both CHD and competing events was randomly generated and was included in the estimation of the subject-specific weight in addition to the 17 covariates described in Section 4. The increase of the resulting IPCW incidence in each group was slight compared with the estimates (Figure 2) ignoring the effect of the hypothetical unmeasured covariates. Therefore, in the KLIS data, it is likely that the effect of unmeasured confounders on our inferences would be small. Scharfstein *et al.* [15, 31] have developed more formal sensitivity analysis. The sensitivity analysis for our data using their idea will be future work.

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Pharmaceutical STATISTICS

REFERENCES

1. The Kyushu Lipid Intervention Study Group. A coronary primary intervention study of Japanese men: study design, implementation and baseline data. *Journal of Atherosclerosis and Thrombosis* 1996; **3**:95–104.
2. The Kyushu Lipid Intervention Study Group. Pravastatin use and risk of coronary events and cerebral infarction in Japanese men with moderate hypercholesterolemia: the Kyushu Lipid Intervention Study. *Journal of Atherosclerosis and Thrombosis* 2000; **7**:110–121.
3. Gail M. A review and critique of some models used in competing risk analysis. *Biometrics* 1975; **31**: 209–222.
4. Kalbfleish JD, Prentice RL. *The Statistical Analysis of Failure Time Data* (2nd edn). Wiley: New York, 2002.
5. Tsiatis AA. A nonidentifiability aspect of the problem of competing risks. *Proceedings of the National Academy of Sciences* 1975; **72**:20–22.
6. Kaplan EL, Meier P. Nonparametric estimation from incomplete observations. *Journal of the American Statistical Association* 1958; **53**:457–481.
7. Benfante R, Yano K, Hwang LJ, Curb D, Kagan A, Ross W. Elevated serum cholesterol is a risk factor for both coronary heart disease and thromboembolic stroke in Hawaiian Japanese men: implications of shared risk. *Stroke* 1994; **25**:814–820.
8. Peterson AV. Bounds for a joint distribution function with fixed subdistribution functions: applications to competing risks. *Proceedings of the National Academy of Sciences* 1976; **73**:11–13.
9. Slud EV, Rubinstein LV. Dependent competing risks and summary survival curves. *Biometrika* 1983; **70**:643–649.
10. Klein JP, Moeschberger ML. Bounds on net survival probabilities for dependent competing risks. *Biometrics* 1988; **44**:529–538.
11. Zheng M, Klein JP. Estimates of marginal survival for dependent competing risks based on an assumed copula. *Biometrika* 1995; **82**:127–138.
12. Robins JM. Information recovery and bias adjustment in proportional hazards regression analysis of randomized trials using surrogate makers. *American Statistical Association Proceedings of the Biopharmaceutical Section* 1993; 24–33.
13. Robins JM, Rotnitzky A, Zhao LP. Analysis of semiparametric regression models for repeated outcomes in the presence of missing data. *Journal of the American Statistical Association* 1995; **90**:106–121.
14. Robins JM, Finkelstein DH. Correcting for noncompliance and dependent censoring in an AIDS clinical trial with Inverse Probability of Censoring Weighted (IPCW) log-rank tests. *Biometrics* 2000; **56**:779–788.
15. Scharfstein DO, Robins JM. Estimation of the failure time distribution in the presence of informative censoring. *Biometrika* 2002; **89**:617–634.
16. Robins JM. Causal inference from complex longitudinal data. *Latent variable modeling and application to causality*, Berkane M (ed.). Springer: New York, 1997; 69–117.
17. Rosenbaum PR, Rubin DB. The central role of the propensity score in observational studies for causal effects. *Biometrika* 1983; **70**:41–55.
18. Horvitz DG, Thompson DJ. A generalization of sampling without replacement from a finite universe. *Journal of the American Statistical Association* 1952; **47**:663–685.
19. Satten GA, Datta S. The Kaplan–Meier estimator as an inverse-probability-of-censoring weighted average. *The American Statistician* 2001; **55**: 207–210.
20. Huber PJ. The behavior of maximum likelihood estimates under nonstandard conditions. *Proceedings of the Fifth Berkeley Symposium in Mathematical Statistics and Probability*. University of California Press: Berkeley, 1976; 221–233.
21. Lin DY, Wei LJ. The robust inference for the Cox proportional hazards model. *Journal of the American Statistical Association* 1989; **84**:1074–1078.
22. Robins JM, Hernán MA, Brumback B. Marginal structural models and causal inference in epidemiology. *Epidemiology* 2000; **11**:550–560.
23. Robins JM. Marginal structural models versus structural nested models as tools for causal inference. *Statistical Models in Epidemiology, the Environment, and Clinical Trials*, Halloran ME Berry DA (eds). Springer: New York, 1999; 95–133.
24. Sato T, Matsuyama Y. Marginal structural models as a tool for standardization. *Epidemiology* 2003; **14**:680–686.
25. Gray RJ. A class of k-sample tests for comparing the cumulative incidence of a competing risk. *Annals of Statistics* 1988; **16**:1141–1154.
26. Fine JP, Gray RJ. A proportional hazards model for the subdistribution of a competing risk. *Journal of the American Statistical Association* 1999; **94**: 496–509.
27. Anderson PK, Abildstrom SZ, Rosthoj S. Competing risks as a multi-state model. *Statistical Methods in Medical Research* 2002; **11**:203–215.
28. Anderson PK, Keiding N. Multi-state models for event history analysis. *Statistical Methods in Medical Research* 2002; **11**:91–115.
29. Tsiatis AA. Competing risks. *Encyclopedia of Biostatistics*, vol. 1, Armitage M, Colton T (eds). Wiley: New York, 1997; 824–834.

DOI: 10.1002/pst

30. Crowder MJ. *Classical Competing Risks*. Chapman & Hall/CRC: London, 2001.
31. Scharfstein DO, Robins JM, Eddings W, Rotnitxky A. Inference in randomized studies with informative censoring and discrete time-to-event endpoints. *Biometrics* 2001; **57**:404–413.
32. Joffe MM, Hoover DR, Jacobson LP, Kingsley L, Chmiel JS Visscher BR, Robins JM. Estimating the effect of zidovudine on kaposi's sarcoma from observational data using a rank preserving structural failure-time model. *Statistics in Medicine* 1998; **17**:1073–1102.

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ORIGINAL ARTICLE

A phase II study of weekly irinotecan as first-line therapy for patients with metastatic pancreatic cancer

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Abstract
Purpose The aim of this study was to assess the efficacy and toxicity of weekly irinotecan in patients with metastatic pancreatic cancer.
Patients and methods Patients with histologically proven pancreatic adenocarcinoma, at least one bidimensionally measurable metastatic lesion, and no prior chemotherapy were selected. Irinotecan at a dose of 100 mg/m^2 was administered intravenously for 90 min on days 1, 8, and 15 every 4 weeks until disease progression or unacceptable toxicity. Pharmacokinetics was examined on day 1 of the first cycle of treatment.
Results Thirty-seven of 40 enrolled patients were assessable for efficacy and toxicity. A partial response was obtained in 10 patients, giving an overall response rate of 27.0% (95% confidence interval 13.8–44.1%). The median overall survival was 7.3 months with a 1-year survival rate of 29.5%. Although toxicities were generally tolerated, one patient died of disseminated intravascular coagulation syndrome induced by neutropenia with watery diarrhea. Pharmacokinetic study showed that patients with biliary drainage seemed to have higher area under the concentration versus time curve for irinotecan and its metabolites compared with patients without biliary drainage.
Conclusion Single-agent irinotecan has significant efficacy for metastatic pancreatic cancer. The toxicity with this schedule appears manageable, though it must be monitored carefully.

Keywords Irinotecan · Phase II study · Pancreatic cancer · Chemotherapy · Pharmacokinetics

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Introduction

Pancreatic cancer is a highly aggressive disease, with approximately 21,000 deaths annually in Japan [7]. While surgery remains the only potential curative option for this disease, the vast majority of patients unfortunately present with advanced, unresectable disease. Although it has been demonstrated that gemcitabine is

an effective tool for palliation of symptoms and prolonging survival in patients with advanced pancreatic cancer [2], single-agent gemcitabine has shown limited benefit, with objective response rates of less than 15% and a median overall survival of around 4–6 months [2, 4, 5]. Therefore, there is a clear need to identify a new effective chemotherapeutic regimen for pancreatic cancer.

Irinotecan is a water-soluble semisynthetic derivative of camptothecin, a plant alkaloid obtained from the *Camptotheca acuminata* tree. Irinotecan and its active metabolite, 7-ethyl-10-hydroxycamptothecin (SN-38), bind to topoisomerase I (an enzyme required for unwinding of DNA during replication), inducing double-stranded DNA breaks and consequent tumor cell death. Irinotecan is internationally approved for use in metastatic colorectal cancer, and has broad activity against other malignancies including lung cancer [6, 9, 15, 16]. Although several studies of single-agent irinotecan or irinotecan-based chemotherapy against pancreatic cancer have been reported [11, 12, 14, 18, 22], the role of irinotecan in the treatment of patients with pancreatic cancer remains unclear yet. Because there are few effective agents for pancreatic cancer to date, it is important to determine the clinical efficacy of irinotecan for this disease. We, therefore, conducted an open-label, multicenter, single-arm phase II study to evaluate the efficacy and toxicity of single-agent irinotecan in patients with pancreatic cancer. In the current study, we adopted weekly administration of irinotecan because safety of this schedule has been confirmed in other cancers in Japan [6, 9, 16]. Since patients with pancreatic cancer tend to suffer various tumor-related complications such as obstructive jaundice and impaired liver function, pharmacokinetic study was also performed.

Patients and methods

Patient selection

Patients were entered into the study if they fulfilled the following eligibility criteria: histologically or cytologically confirmed adenocarcinoma of the pancreas; at least one bidimensionally measurable metastatic lesion; no history of prior chemotherapy or radiotherapy; age 20–74 years; Karnofsky performance status (KPS) ≥ 50 points; estimated life expectancy ≥ 2 months; adequate bone marrow function (WBC count $< 12{,}000$ per mm^3, neutrophil count $\geq 2{,}000$ per mm^3, platelet count $\geq 100{,}000$ per mm^3, and hemoglobin level ≥ 10.0 g/dl), adequate renal function (serum creatinine and blood urea nitrogen level $\leq$ the institutional upper limit of normal), and adequate liver function (serum total bilirubin level ≤ 2.0 mg/dl, serum transaminases levels ≤ 2.5 times the institutional upper limit of normal); and written informed consent. Patients were excluded if there was a history of severe drug hypersensitivity; serious complications; central nervous system metastases; other concomitant malignant disease; marked pleural or peritoneal effusion; and watery diarrhea. Pregnant or lactating women were also excluded. The study was performed in accordance with the Declaration of Helsinki, approved by the institutional review board of each participating center, and conducted in accordance with Good clinical practice guideline in Japan.

Treatment plan

This study was an open-label, multicenter, single-arm phase II study. Irinotecan was supplied by Daiichi Pharmaceutical Co., Ltd. (Tokyo, Japan) and Yakult Honsha Co., Ltd. (Tokyo, Japan). Irinotecan at a dose of 100 mg/m^2 was administered intravenously for 90 min on days 1, 8, and 15 every 4 weeks until the occurrence of disease progression, unacceptable toxicity, or the patient's refusal to continue. Prophylactic administration of antiemetic agents was allowed at the investigator's discretion. Physical examination, complete blood cell counts, biochemistry tests, and urinalysis were assessed weekly during treatments. If patients experienced neutropenia of $< 1{,}500$ per mm^3, thrombocytopenia of $< 100{,}000$ per mm^3, fever (≥ 38°C) with suspected infection, grade ≥ 1 or watery diarrhea, or $\geq$ grade 3 non-hematological toxicities other than nausea, vomiting and anorexia, irinotecan administration was omitted on that day and postponed to the next scheduled treatment day. If patients experienced neutropenia of < 500 per mm^3, thrombocytopenia of $< 50{,}000$ per mm^3, fever (≥ 38°C) with suspected infection, or grade ≥ 2 or watery diarrhea at any time, the irinotecan dose of the subsequent cycle was reduced by 20 mg/m^2. Patients went off study if they required more than two dose reductions. If the next cycle could not start within 4 weeks from the scheduled day, the patient was withdrawn from the study. The toxicity of irinotecan therapy was evaluated according to the National Cancer Institute Common Toxicity criteria version 2.0.

Evaluation

Objective tumor response was evaluated every 4 weeks according to the Japan Society for Cancer Therapy (JSCT) criteria [8], which is similar to the WHO crite-

ria. A complete response (CR) was defined as the disappearance of all evidence of cancer for at least 4 weeks. A partial response (PR) was defined as a $\geq$ 50% reduction in the sum of the products of the two longest perpendicular diameters of all measurable lesions for at least 4 weeks without any evidence of new lesions. No change (NC) was defined as a < 50% reduction or a < 25% increase in the sum of the products of the two longest perpendicular diameters of all measurable lesions for at least 4 weeks without any evidence of new lesions. Progressive disease (PD) was defined as a $\geq$ 25% increase or the appearance of new lesions. Primary pancreatic lesions were considered to be assessable but not measurable lesions, because it is difficult to measure the size of primary pancreatic lesions accurately [1]. Objective tumor response was secondarily assessed according to the response evaluation criteria in solid tumors (RECIST criteria) [20] among patients with at least one measurable metastatic lesion whose longest diameter measured by CT is no less than double the slice thickness. An external review committee confirmed objective responses and toxicities.

Clinical benefit was evaluated on the basis of established criteria [13]. Each patient was classified as a clinical benefit responder or non-responder on the basis of the change in two parameters of clinical benefit (pain and KPS). In the current study, the body weight was not used to evaluate clinical benefit response because the body weight of patients with pancreatic cancer sometimes increases due to not only improvement of their condition but also retention of malignant ascites. A positive response for pain was defined as an improved pain intensity of $\geq$ 50% from baseline for $\geq$ 4 weeks, or a decreased morphine consumption of $\geq$ 50% from baseline for $\geq$ 4 weeks. A positive response for KPS was defined as an improved KPS of $\geq$ 20 points from baseline for $\geq$ 4 weeks. To be classified as a clinical benefit responder, a patient had to achieve a positive response in at least one parameter (pain or KPS) without being negative for the other, sustained for $\geq$ 4 weeks.

Pharmacokinetics

To investigate the impact of biliary drainage on pharmacokinetics of irinotecan, we planned to recruit five patients each with and without biliary drainage. Heparinized blood samples (5 ml) for the pharmacokinetic study were obtained before infusion of irinotecan, at the end of the 90 min infusion, and 0.5, 1, 2, 4, 6, 8, 24 h after the completion of infusion on day 1 of the first cycle. Blood samples were immediately centrifuged at 3,000 rpm for 10 min to remove plasma and stored in polyethylene tubes at −20°C until analysis. Quantitative analysis of total irinotecan and its metabolites, SN-38, SN-38 glucuronide, and 7-ethyl-10-[4-*N*-(5-aminopentanoic acid)-1-piperidino] carbonyloxycamptothecin (APC) was performed by methods previously described [17, 19].

Statistical analysis

The primary goal was to evaluate the response rate (CR and PR) of irinotecan. The 95% confidence interval for response rate was calculated based on the binomial distribution. The response duration was defined as the interval from the first documentation of response to the first documentation of tumor progression. The time to progression (TTP) was calculated from the date of study enrollment to the first documentation of tumor progression; and overall survival was calculated from the date of study enrollment to the date of death or the last follow-up with censored value. Median overall survival and the median TTP were estimated by the Kaplan–Meier method and 95% confidence interval were estimated based on the Greenwood's formula. A total of 35 patients were planned to be enrolled based on the assumptions that the expected response rate of irinotecan was 15% and the threshold rate was 5%. A two-stage design was used in this study. The interim analysis was planned when 15 patients were enrolled in the first stage of the study. If the upper limit of the 90% confidence interval (one-sided) did not exceed the expected rate of 15% (no objective response in the 15 patients), irinotecan was judged to be ineffective and the study was ended. If an objective response was observed in any of the first 15 patients, additional 20 patients were enrolled in the second stage of accrual to estimate the response rate. If 6 or more out of 35 patients achieved objective response, the lower limit of the 95% confidence interval (two-sided) exceeds the threshold rate of 5%, and then the agent would be considered to be active for metastatic pancreatic cancer.

Results

Patients

Forty patients were enrolled in the study by 7 institutions between August 2001 and November 2002. Of the 40 patients, 3 patients who did not receive irinotecan because of rapid tumor progression or protocol violation were excluded from analysis. Patient characteristics of the remaining 37 patients are listed in Table 1.

All 37 patients had metastatic disease and had a good KPS of ≥ 80. Morphine was prescribed for 10 patients due to abdominal or back pain and 14 patients were assessable for clinical benefit response. Seven patients had recurrent disease after pancreatic resection. Two patients underwent percutaneous transhepatic biliary drainage for obstructive jaundice prior to study enrollment.

Treatments

Data were collected through May 4, 2004, providing 18 months of survival follow-up from the time accrual ended. Thirty-seven patients were given a total of 108 cycles of therapy, with a median of 2 cycles each (range 1–10). The administration of irinotecan on day 8 and day 15 was performed in 87 (80.6%) and 76 (70.4%) of 108 cycles, respectively. Dose reduction was required in 13 patients (35.1%), mainly due to diarrhea and fever with suspected infection. At the time of analysis, all patients had discontinued the study because of disease progression ($n = 28$), toxicity ($n = 5$), treatment-related death ($n = 1$), and withdrawal of consent due to other reasons ($n = 3$). After discontinuation of irinotecan, 26 patients received gemcitabine monotherapy or gemcitabine-based combination therapy; one patient was treated with S-1, and remaining 10 patients underwent only supportive care. Among 27 patients treated with second-line chemotherapy, 2 patients who received gemcitabine monotherapy achieved a PR.

Table 1 Patient characteristics ($n = 37$)

Characteristics		No. of patients (%)
Gender		
Male		25 (67.6)
Female		12 (32.4)
Median age, years (range)	59 (41–74)	
Karnofsky performance status, point		
100		8 (21.6)
90		25 (67.6)
80		4 (10.8)
Median body surface area (m^2) (range)	1.55 (1.31–1.85)	
History of surgical resection		7 (18.9)
PTBD		2 (5.4)
Sites of metastasis		
Liver		33 (89.2)
Lymph nodes		17 (45.9)
Lung		8 (21.6)
Others		3 (8.1)

PTBD percutaneous transhepatic biliary drainage

Efficacy

Of 37 patients, 10 patients achieved a PR according to the JSCT criteria (Table 2). The overall response rate was therefore 27.0% (95% confidence interval 13.8–44.1%) with median response duration of 4.1 months (range 0.9–7.1 months). The median TTP was 2.1 months (range 0.7–9.5 months), and the median overall survival of 7.3 months (range 0.7–25.9 months) with a 1-year survival rate of 29.5% (Fig. 1). Of 29 patients assessable for RECIST criteria, a PR was seen in 8 patients (27.6%), stable disease in 6 patients (20.7%), and PD in 12 patients (41.4%). With regard to clinical benefit, 2 of 14 evaluable patients had pain relief and were classified as a responder (Table 3).

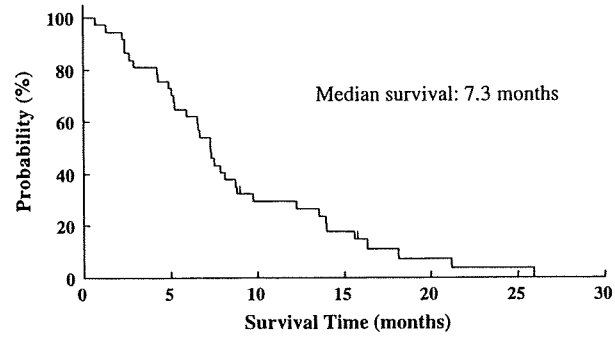

Fig. 1 Overall survival curve of all 37 patients

Table 2 Efficacy results

	No. ($N = 37$)	%
Tumor response		
Partial response	10	27.0
No change	7	18.9
Progressive disease	17	45.9
Not evaluable	3	8.1
Time to progression (months)		
Median	2.1	
Range	0.7–9.5	
Overall survival (months)		
Median	7.3	
Range	0.7–25.9	
1-year survival rate		29.5

Table 3 Clinical benefit response ($n = 14$)

		Karnofsky performance status		
		Improved	Stable	Worse
Pain	Improved	0	2	0
	Stable	0	6	1
	Worse	0	5	0

Toxicity

All 37 patients were assessable for toxicity. The major toxicities observed during the study are summarized in Table 4. The most common toxicities were hematological toxicity and gastrointestinal toxicity. Grade 3 or 4 neutropenia occurred in 10 patients (27.0%) and 5 patients received granulocyte-colony stimulating factors. The neutrophil count nadir typically occurred on day 21, and recovered to baseline values by day 28. Although nausea, vomiting, and anorexia were observed frequently, most of these toxicities recovered spontaneously or with adequate supportive treatment. Grade 3 diarrhea occurred in four patients and they were treated with loperamide. Most diarrheas appeared during the first cycle of treatment: the median time to the worst day of diarrhea was 13 days from the initiation of a cycle of therapy. Though the toxicities were mild to moderate in severity and short in duration, one patient died at day 21 of the first cycle of treatment because of disseminated intravascular coagulation syndrome and multiple organ failure induced by neutropenia and watery diarrhea due to irinotecan. The patient, a 58-year old woman with pretreatment KPS of 100, developed grade 4 neutropenia on day 12 complicated by fever (38.8°C) and grade 3 diarrhea that evolved to fatal shock despite aggressive medical management.

Table 4 Treatment-related adverse events ($n = 37$): worst grade reported during treatment period

Toxicity	Grade				Grade 1–4 (%)	Grade 3–4 (%)
	1	2	3	4		
Hematologic						
Leukopenia	15	6	8	1	81.1	24.3
Neutropenia	5	11	8	2	70.3	27.0
Anemia	0	14	3	0	45.9	8.1
Thrombocytopenia	1	1	1	1	10.8	5.4
Non-hematologic						
Nausea	7	12	15	–	91.9	40.5
Vomiting	7	14	5	0	70.3	13.5
Diarrhea	15	8	4	0	73.0	10.8
Constipation	1	8	2	0	29.7	5.4
Anorexia	4	7	14	1	70.3	40.5
Stomatitis	2	0	0	0	5.4	0
Rash	1	0	0	0	2.7	0
Alopecia	24	1	–	–	67.6	–
Fatigue	3	8	1	1	35.1	5.4
Fever	3	1	0	0	10.8	0
Infection	2	1	4	1	21.6	13.5
Total bilirubin	4	1	1	0	16.2	2.7
AST	5	5	2	0	32.4	5.4
ALT	4	4	3	0	29.7	8.1
Hyponatremia	6	0	3	0	24.3	8.1
Creatinine	0	0	2	0	5.4	5.4

AST aspartate aminotransferase, *ALT* alanine aminotransferase

Pharmacokinetics

A pharmacokinetic analysis was performed in five patients without biliary drainage and in two patients who underwent percutaneous transhepatic biliary drainage (Planned five patients could not be enrolled in drainage group because only two patients had biliary drainage in the current study). Table 5 and Fig. 2 show the pharmacokinetic parameters for irinotecan and its three major metabolites in patients with and without biliary drainage. Although it was difficult to assess the influence of biliary drainage in this study because of the small number of subjects analyzed, patients with biliary drainage seemed to have higher area under the concentration versus time curve for irinotecan and its metabolites compared with patients without biliary drainage.

Discussion

The prognosis of the patients with pancreatic cancer remains poor even after a randomized study demonstrated survival advantage of gemcitabine against advanced pancreatic cancer, indicating necessity of new effective agents or combination regimens for this dismal disease. Irinotecan, which has a quite different mechanism from gemcitabine, has been considered one of the attractive agents for pancreatic cancer, since this agent has demonstrated substantial activity in various types of malignant tumor [6, 9, 15, 16]. The current multicenter phase II study was, therefore, conducted to evaluate the efficacy and toxicity of single-agent irinotecan in patients with metastatic pancreatic cancer.

In this study, we found that weekly irinotecan demonstrated a good overall response rate of 27.0% in 37 patients with metastatic pancreatic cancer. In addition, a relatively long median overall survival of 7.3 months was shown, though all patients in our study had metastatic disease. As to clinical benefit response, 2 of 14 patients achieved clinical benefit response. These results indicate that irinotecan has a substantial antitumor effect on pancreatic cancer.

The major toxicities of irinotecan that were seen in the study were myelosuppression and gastrointestinal toxicities, similar to the previous observation of irinotecan monotherapy in other cancers [6, 9, 16]. Most toxicity was mild to moderate, and manageable with conservative treatment. However, one patient died of disseminated intravascular coagulation syndrome and multiple organ failure induced by neutropenia and diarrhea. Pretreatment condition of this patient was good (KPS = 100), and it was difficult to predict these

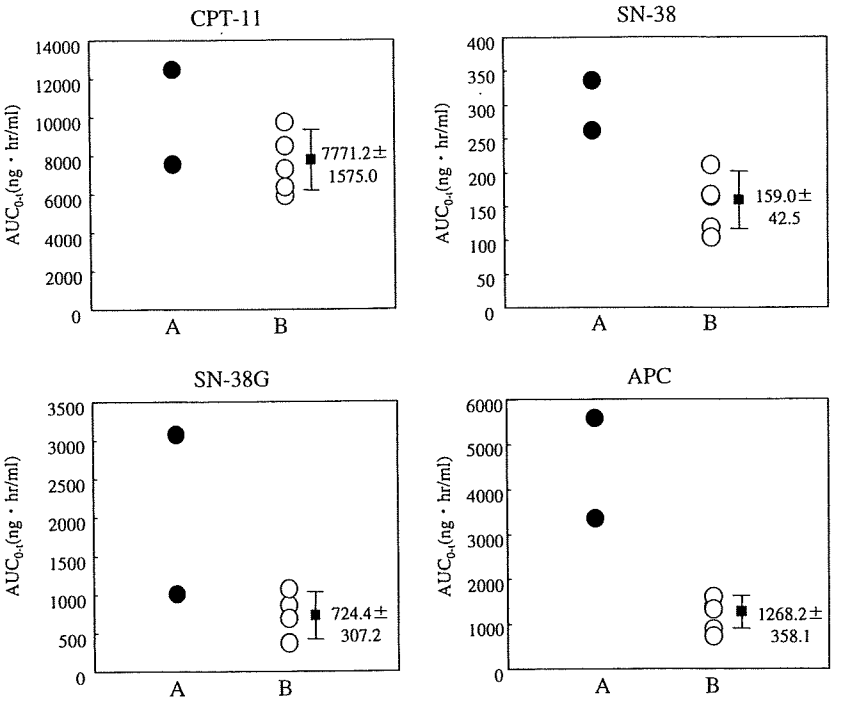

Fig. 2 Area under the concentration versus time curve for irinotecan and metabolites in patients with biliary drainage (A, $n = 2$) and without drainage (B, $n = 5$). The values are expressed as the mean ± SD

Table 5 Pharmacokinetic parameters after single administration of irinotecan at a dose of 100 mg/m^2 ($n = 7$)

		C_{max} (ng/ml)	T_{max} (h)	$T_{1/2}$ (h)	AUC_{0-t} (ngh/ml)	CL (l/h m^2)
Irinotecan	A	1,188.5, 1,997.6	1.6, 1.5	7.8, 8.2	7,762, 12,692	11.8, 7.1
	B	1,701.0 ± 348.3	1.5 ± 0.1	7.7 ± 0.9	7,771.2 ± 1,575.0	12.4 ± 2.5
SN-38	A	25.5, 26.2	2.1, 1.5	14.7, 9.9	268, 342	–
	B	17.5 ± 3.8	2.3 ± 0.8	30.2 ± 27.6	159.0 ± 42.5	
SN-38G	A	81.3, 207.2	3.6, 2.0	10.8, 12.5	1,063, 3,130	–
	B	78.8 ± 34.1	2.2 ± 0.2	21.6 ± 13.2	724.4 ± 307.2	
APC	A	309.2, 359.3	2.6, 5.5	7.0, 9.5	3,441, 5,673	–
	B	116.6 ± 39.7	3.0 ± 0.6	8.8 ± 0.7	1,268.2 ± 358.1	

A Patients with biliary drainage $n = 2$

B Patients without biliary drainage (parameters are represented as the mean ± SD) $n = 5$

episodes before onset. Although our study indicated that weekly irinotecan administration would be tolerable in patients with metastatic pancreatic cancer, careful observation is required during the treatment period, since pancreatic cancer patients tend to suffer various tumor-related complications and easily take a turn to the worse because of tumor progression.

There are two studies of single-agent irinotecan that assessed efficacy and toxicity against pancreatic cancer [14, 22]. Sakata et al. [14] studied irinotecan at a dose of 100 or 150 mg/m^2 administered weekly or bi-weekly to previously treated or untreated patients with pancreatic cancer in Japan. Although 57 of 61 enrolled patients were assessable, only 4 patients (7.0%) showed a PR. This study included 28 patients (49.1%) with poor performance status of 2–3 and 22 patients (38.6%) with prior chemotherapy, and no patient showed a PR in these patients with poor performance status or prior chemotherapy. Wagener et al. [22] demonstrated that irinotecan at a dose of 350 mg/m^2 administered every 3 weeks to chemo-naïve pancreatic cancer patients with performance status of ≤ 2, achieved a PR in 3 of 32 assessable patients (9.4%) with an median overall survival of 5.2 months. Although precise reason for the discrepant response rates between our study and the other two studies is unclear, patient background may be one possible explanation because only chemo-naïve patients with good performance status were entered into our study (89.2% of our patients had good KPS of ≥ 90).

For the purpose of the improvement on response rate and prognosis, several studies of combination therapy have been conducted in patients with pancreatic cancer. With regard to irinotecan with gemcitabine, an

encouraging activity, response rates between 20.0 and 24.7% and median overall survival between 5.7 and 7 months, have been reported in two phase II studies [11, 18]. However, survival benefit of this combination therapy was not shown in a phase III study [12], in which, 360 patients were randomized to treatment with a combination of gemcitabine 1,000 mg/m^2 followed by irinotecan 100 mg/m^2 given on days 1 and 8 of a 3-week cycle versus gemcitabine monotherapy. The response rate for the combination therapy was higher at 16.1% compared with 4.4% for gemcitabine alone, but there was no difference in median overall survival (6.3 vs. 6.6 months). However, several clinical studies have recently indicated that irinotecan-based chemotherapy seemed to be an effective treatment for advanced pancreatic cancer after gemcitabine failure: irinotecan–raltitrexed combination demonstrated overall response rate of 16% (3/19) in patients with gemcitabine-pretreated pancreatic cancer [21], and Cantore et al. [3] reported that irinotecan plus oxaliplatin showed response rate of 10% (3/30) with a clinical benefit response of 20% (6/30) for patients with advanced pancreatic cancer after gemcitabine failure.

Because biliary excretion is a major elimination pathway for irinotecan and its metabolites, we investigated the impact of biliary drainage on the pharmacokinetics for this agent. Our results suggested that patients with biliary drainage tended to have higher area under the concentration versus time curve of irinotecan and metabolites compared with patients without biliary drainage. Meyerhardt et al. [10] reported that modest elevation of bilirubin (1.0–1.5 mg/dl) is associated with increased grade 3 to 4 neutropenia in patients treated with irinotecan. The fact that the two patients with biliary drainage in the current study had slight elevation of baseline serum bilirubin level (1.4 and 1.7 mg/dl) might influence pharmacokinetics for irinotecan. Although no severe hematological or non-hematologic toxicities appeared in these two patients, careful observation may be required when treating patients with biliary drainage.

In conclusion, single-agent irinotecan showed a substantial antitumor activity for patients with metastatic pancreatic cancer, rendering a 27.0% response rate. The toxicity with this schedule appears manageable, though it must be monitored carefully.

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References

1. Aoki K, Okada S, Moriyama N, Ishii H, Nose H, Yoshimori M, Kosuge T, Ozaki H, Wakao F, Takayasu K, Mukai K (1994) Accuracy of computed tomography in determining pancreatic cancer tumor size. Jpn J Clin Oncol 24:85–87
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 (1997) Improvements in survival and clinical benefit with gemcitabine as first-line therapy for patients with advanced pancreas cancer: a randomized trial. J Clin Oncol 15:2403–2413
3. Cantore M, Rabbi C, Fiorentini G, Oliani C, Zamagni D, Iacono C, Mambrini A, Del Freo A, Manni A (2004) Combined irinotecan and oxaliplatin in patients with advanced pretreated pancreatic cancer. Oncology 67:93–97
4. Carmichael J, Fink U, Russell RG, Spittle MF, Harris AL, Spiessi G, Blatter J (1996) Phase II study of gemcitabine in patients with advanced pancreatic cancer. Br J Cancer 73:101–105
5. Casper ES, Green MR, Kelsen DP, Heelan RT, Brown TD, Flombaum CD, Trochanowski B, Tarassoff PG (1994) Phase II trial of gemcitabine (2,2′-difluorodeoxycytidine) in patients with adenocarcinoma of the pancreas. Invest New Drugs 12:29–34
6. Fukuoka M, Niitani H, Suzuki A, Motomiya M, Hasegawa K, Nishiwaki Y, Kuriyama T, Ariyoshi Y, Negoro S, Masuda N, Nakajima S, Taguchi T (1992) A phase II study of CPT-11, a new derivative of camptothecin, for previously untreated non-small-cell lung cancer. CPT-11 Lung Cancer Study Group. J Clin Oncol 10:16–20
7. http://www.mhlw.go.jp/toukei/saikin/hw/jinkou/kakutei03/hyo5.html (accessed August 18, 2005)
8. Japan Society for Cancer Therapy: Criteria for the evaluation of the clinical effects of solid cancer chemotherapy (1993) J Jpn Soc Cancer Ther 28:101–130
9. Masuda N, Fukuoka M, Kusunoki Y, Matsui K, Takifuji N, Kudoh S, Negoro S, Nishioka M, Nakagawa K, Takada M (1992) CPT-11: a new derivative of camptothecin for the treatment of refractory or relapsed small-cell lung cancer. J Clin Oncol 10:1225–1229
10. Meyerhardt JA, Kwok A, Ratain MJ, McGovren JP, Fuchs CS (2004) Relationship of baseline serum bilirubin to efficacy and toxicity of single-agent irinotecan in patients with metastatic colorectal cancer. J Clin Oncol 22:1439–1446
11. Rocha Lima CM, Savarese D, Bruckner H, Dudek A, Eckardt J, Hainsworth J, Yunus F, Lester E, Miller W, Saville W, Elfring GL, Locker PK, Compton LD, Miller LL, Green MR (2002) Irinotecan plus gemcitabine induces both radiographic and CA 19-9 tumor marker responses in patients with previously untreated advanced pancreatic cancer. J Clin Oncol 20:1182–1191
12. Rocha Lima CM, Green MR, Rotche R, Miller WH Jr, Jeffrey GM, Cisar LA, Morganti A, Orlando N, Gruia G, Miller LL (2004) Irinotecan plus gemcitabine results in no survival advantage compared with gemcitabine monotherapy in patients with locally advanced or metastatic pancreatic cancer despite increased tumor response rate. J Clin Oncol 22:3776–3783
13. 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 (1996) A phase II trial of gemcitabine in patients with 5-FU-refractory pancreas cancer. Ann Oncol 7:347–353

14. Sakata Y, Shimada Y, Yoshino M, Kambe M, Futatsuki K, Nakao I, Ogawa N, Wakui A, Taguchi T (1994) A late phase II study of CPT-11, irinotecan hydrochloride, in patients with advanced pancreatic cancer (in Japanese). CPT-11 study group on gastrointestinal cancer. Jpn J Cancer Chemother 21:1039–1046
15. Saltz LB, Cox JV, Blanke C, Rosen LS, Fehrenbacher L, Moore MJ, Maroun JA, Ackland SP, Locker PK, Pirotta N, Elfring GL, Miller LL (2000) Irinotecan plus fluorouracil and leucovorin for metastatic colorectal cancer. N Engl J Med 343:905–914
16. Shimada Y, Yoshino M, Wakui A, Nakao I, Futatsuki K, Sakata Y, Kambe M, Taguchi T, Ogawa N (1993) Phase II study of CPT-11, a new camptothecin derivative, in metastatic colorectal cancer. J Clin Oncol 11:909–913
17. Sparreboom A, de Bruijn P, de Jonge MJA, Loos WJ, Stoter G, Verweij J, Nooter K (1998) Liquid chromatographic determination of irinotecan and three major metabolites in human plasma, urine and feces. J Chromatogr B Biomed Sci Appl 712:225–235
18. Stathopoulos GP, Rigatos SK, Dimopoulos MA, Giannakakis T, Foutzilas G, Kouroussis C, Janninis D, Aravantinos G, Androulakis N, Agelaki S, Stathopoulos JG, Georgoulias V (2003) Treatment of pancreatic cancer with a combination of irinotecan (CPT-11) and gemcitabine: a multicenter phase II study by the Greek Cooperative Group for Pancreatic Cancer. Ann Oncol 14:388–394
19. Tanaka S, Yoshida Y, Suzuki W, Sudo K, Hakusui H (1994) A new HPLC method for the determination of irinotecan hydrochloride (CPT-11) and its metabolite, SN-38 in human plasma and urine (in Japanese). Jpn Pharmacol Ther 22:163–172
20. Therasse P, Arbuck SG, Eisenhauer EA, Wanders J, Kaplan RS, Rubinstein L, Verweij J, Van Glabbeke M, van Oosterom AT, Christian MC, Gwyther SG (2000) New guidelines to evaluate the response to treatment in solid tumors. European organization for research and treatment of cancer, National Cancer Institute of the United States, National Cancer Institute of Canada. J Natl Cancer Inst 92:205–216
21. Ulrich-Pur H, Raderer M, Verena Kornek G, Schull B, Schmid K, Haider K, Kwasny W, Depisch D, Schneeweiss B, Lang F, Scheithauer W (2003) Irinotecan plus raltitrexed vs raltitrexed alone in patients with gemcitabine-pretreated advanced pancreatic adenocarcinoma. Br J Cancer 88:1180–1184
22. Wagener DJ, Verdonk HE, Dirix LY, Catimel G, Siegenthaler P, Buitenhuis M, Mathieu-Boue A, Verweij J (1995) Phase II trial of CPT-11 in patients with advanced pancreatic cancer, an EORTC early clinical trials group study. Ann Oncol 6:129–132

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Phase II Study of Combination Chemotherapy with Gemcitabine and Cisplatin for Patients with Metastatic Pancreatic Cancer

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Objective: The objectives of this study were to evaluate the efficacy and toxicity of combination chemotherapy with gemcitabine and cisplatin in patients with metastatic pancreatic cancer.
Methods: Patients naïve to chemotherapy who had histologically or cytologically confirmed metastatic pancreatic adenocarcinoma were entered. Gemcitabine was given at a dose of 1000 mg/m^2 over 30 min on days 1, 8 and 15, and cisplatin was given at a dose of 80 mg/m^2 over 150 min on day 1, in 28-day cycles.
Results: A total of 38 patients were enrolled in this study between August 2001 and December 2003. There were no complete responses and 10 partial responses, resulting in an overall response rate of 26% (95% CI: 13.4–43.1%]. Twenty-one patients (55%) had stable disease, whereas 7 (18%) had progressive disease. The median time to progression was 4.2 months and the median overall survival was 7.5 months with a 1-year survival rate of 24%. Grade 3–4 toxicities included neutropenia in 26 patients (68%), thrombocytopenia in 19 (50%), anorexia in 15 (39%) and nausea in nine (24%). There was only one episode of neutropenic fever and there were no significant bleeding episodes or treatment-related deaths.
Conclusion: The combination of gemcitabine and cisplatin administered by this schedule produced a good response rate associated with moderate but manageable toxicities in patients with metastatic pancreatic cancer.

Key words: gemcitabine – cisplatin – phase II study – chemotherapy – pancreatic cancer

INTRODUCTION

Pancreatic cancer currently represents the fifth leading cause of cancer-related mortality in Japan, with an estimated 22 260 deaths attributable to the disease in 2004 (1). Most patients with pancreatic cancer have advanced, unresectable disease at the time of diagnosis and their prognosis is extremely poor. Since a randomized study by Burris et al. in 1997 demonstrated that gemcitabine had a survival benefit versus fluorouracil (2), gemcitabine has been accepted as the standard treatment for advanced pancreatic cancer. However, the median survival of patients with advanced pancreatic cancer treated with single-agent gemcitabine has been only about 6 months (2–4), indicating the pressing need for development of novel treatment strategies.

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Combination of gemcitabine with other agents would be one promising avenue for improving the effect of treatment for advanced pancreatic cancer. In fact, a few recent randomized phase III studies of combinations such as gemcitabine/erlotinib (5) and gemcitabine/capecitabine (6) have demonstrated statistically significant survival benefit in comparison with gemcitabine alone in patients with advanced pancreatic cancer, although there is still no worldwide consensus about the results. As well as these combinations, gemcitabine plus cisplatin has been considered an attractive regimen for pancreatic cancer for several reasons: (i) single-agent cisplatin shows modest activity against pancreatic cancer (7), (ii) preclinical *in vitro* and *in vivo* studies have demonstrated synergistic effects between gemcitabine and cisplatin (8), (iii) the two drugs have non-overlapping, dose-limiting toxicities, and (iv) this combination has demonstrated activity against various malignancies, and is accepted as one of the standard therapies for non-small-cell

lung cancer and urothelial cancer based on large randomized studies (9,10). Several phase II studies of gemcitabine plus cisplatin for advanced pancreatic cancer have been published to date, most of which have shown that this combination seems to be effective, with response rates of 9–31%, and median overall survivals of 5.6–9.6 months (11–16). However, because there have been few studies of Asians receiving gemcitabine and cisplatin for treatment of pancreatic cancer, we conducted the present phase II study to evaluate the efficacy and toxicity of this combination therapy in Japanese patients with metastatic pancreatic cancer. Although various schedules for the combination of gemcitabine and cisplatin have been reported in previous studies, we administered gemcitabine at a dose of 1000 mg/m^2 on days 1, 8 and 15 and cisplatin at a dose of 80 mg/m^2 on day 1 of a 28-day cycle, based on the results of a phase I study conducted in Japanese patients with non-small-cell lung cancer (17).

PATIENTS AND METHODS

Patient Selection

Patients with histologically or cytologically proven pancreatic adenocarcinoma with at least one bidimensionally measurable metastatic lesion were eligible for the study. Other eligibility criteria included: no previous treatment for pancreatic cancer except surgery; age $\geq$20 and $\leq$74 years, Karnofsky performance status (KPS) $\geq$50, life expectancy $\geq$8 weeks, adequate bone marrow function (white blood cell count $\geq$4000/mm^3, neutrophil count $\geq$2000/mm^3, platelet count $\geq$100 000/mm^3 and hemoglobin level $\geq$10.0 g/dl), adequate renal function (serum creatinine concentration $\leq$ upper limit of normal and creatinine clearance $\geq$60 ml/min), adequate hepatic function (serum bilirubin level $\leq$2.0 mg/ml, serum aspartate and alanine transaminase (AST and ALT) levels $\leq$2.5 times upper normal limit or $\leq$5 times upper normal limit if liver metastases or biliary drainage were present) and adequate pulmonary function (PaO_2 $\geq$70 mmHg). Exclusion criteria were as follows: symptomatic pulmonary fibrosis or interstitial pneumonia, marked pleural effusion or ascites, central nervous system metastasis, active concomitant malignancy, severe mental disorder, serious complications such as active infection, active gastrointestinal ulcer, or cardiac disease and pregnant or lactating women. 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, single-center, single-arm phase II study. The patients received gemcitabine at a dose of 1000 mg/m^2 intravenously over 30 min on days 1, 8 and 15, and cisplatin at a dose of 80 mg/m^2 just after gemcitabine administration over 150 min on day 1. The treatment cycles were repeated every 4 weeks for a maximum of six cycles unless disease progression or unacceptable toxicity occurred. If patients completed the planned six cycles of treatment without disease progression, then they received gemcitabine monotherapy until disease progression. If patients developed leukopenia of <2000/mm^3, neutropenia of <1000/mm^3, or thrombocytopenia of <75 000/mm^3 during the cycle, gemcitabine administration was skipped. If patients developed leukopenia of <3000/mm^3, neutropenia of <1500/mm^3, thrombocytopenia of <100 000/mm^3, total bilirubin of >2.0 mg/dl, or creatinine clearance of <50 mil/min, initiation of the next cycle was prolonged until recovery. Dose reduction of gemcitabine from 1000 to 800 mg/m^2 was allowed when patients experienced (i) grade 4 leukopenia or neutropenia, (ii) febrile neutropenia, (iii) grade 4 thrombocytopenia or grade 3 thrombocytopenia requiring blood transfusion, or (iv) grade 3 or greater non-hematological toxicities other than nausea, vomiting, anorexia and hyperglycemia. Patients were dropped from the study if they required more than two dose reductions, or if they were unable to start the next cycle within 4 weeks from the scheduled day.

Clinical Assessments

Physical examination, complete blood cell counts, serum chemistry and urinalysis were performed at the baseline and at least once weekly after the start of treatment. All patients who received at least one dose of gemcitabine were evaluable for safety. Toxicities were graded according to the National Cancer Institute common toxicity criteria version 2.0. Tumor assessment with computed tomographic scan or magnetic resonance imaging and measurement of the tumor marker CA 19-9 was performed every 4 weeks, and tumor response was evaluated using the criteria of the Japan Society for Cancer Therapy (18), which are similar to those of the World Health Organization. Briefly, a complete response (CR) was defined as the disappearance of all clinical evidence of the tumor for a minimum of 4 weeks. A partial response (PR) 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 (NC) 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 (PD) 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 of clinical status that was consistent with disease progression. Primary pancreatic lesions were considered to be assessable but not measurable lesions, because it is difficult to measure the size of primary pancreatic lesions accurately. Time to tumor progression (TTP) was calculated from the date of the start of therapy until

documented PD or death owing to any cause, whichever occurred first. For patients still alive at the time of analysis and who did not have disease progression, TTP was censored at the date of the last follow-up visit. Overall survival was calculated from the date of the start of therapy to the date of death owing to any cause. Patients alive on the date of the last follow-up visit were censored on that date. Median probability of survival and the median TTP were estimated by the Kaplan–Meier method. A total of 38 patients were scheduled for enrollment based on assumptions that the expected response rate of this regimen was 20%, the threshold rate was 5%, the α error was 5% (one-sided), and the β error was 10%.

RESULTS

Patients

Thirty-eight patients with metastatic pancreatic cancer were enrolled in this study between August 2001 and December 2003 at the National Cancer Center Hospital, Tokyo, Japan. All of them received at least one cycle of chemotherapy and were evaluable for toxicity and response. The patient characteristics are shown in Table 1. Before the start of the study, six patients had received surgical resection and 10 had undergone biliary drainage for obstructive jaundice. The KPS was $\geq$80 in all patients. Twenty-eight patients had abdominal and/or back pain before treatment, and morphine had been prescribed for 18 of them.

Table 1. Patient characteristics ($n = 38$)

Characteristics	No. of patients (%)
Gender	
Male	24 (63)
Female	14 (37)
Median age, years (range)	
58 (45–73)	
Karnofsky performance status, point	
100	12 (32)
90	24 (63)
80	2 (5)
History of surgical resection	6 (16)
History of biliary drainage	10 (26)
Sites of metastasis	
Liver	28 (74)
Lung	9 (24)
Lymph node	8 (21)
Peritoneum	3 (8)
Others*	4 (11)

*Spleen 2; local recurrence 1; abdominal wall 1.

Treatments

A total of 107 cycles were administered to the 38 patients with a median of 2 cycles per patient (range 1–6). Gemcitabine was administered on day 8 and day 15 in 93 (87%) and 63 (59%) of the 107 cycles, respectively. Mean dose intensity for gemcitabine and cisplatin was 557 mg/m^2/week (range 368–750) and 18.6 mg/m^2/week (range 17–20), corresponding to 74 and 93% of the planned protocol dose, respectively. Gemcitabine dose reduction was required in 10 patients owing to hematological toxicity. After completion or discontinuation of the protocol study, 20 patients received subsequent chemotherapy (19 patients received gemcitabine monotherapy and one patient received fluorouracil and cisplatin combination therapy), and the remaining 18 patients received only supportive care.

Response and Survival

There were no complete responses and 10 partial responses, giving an overall response rate of 26% (95% CI: 13.4–43.1%). NC was noted in 21 patients (55%), and PD in seven (18%). The serum CA 19-9 level was reduced to less than half in 10 of 32 patients (31%) in whom the pretreatment level of CA 19-9 had been elevated to above the upper normal limit (37 U/ml). At the time of analysis, all the patients were confirmed to have died, except for one who was lost to follow-up. The cause of death was disease progression in all cases. The median TTP was 4.2 months and the median overall median survival was 7.5 months with a 1-year survival rate of 24% (Fig. 1).

Toxicity

All 38 patients were assessed for toxicities, which are listed in Table 2. The most common toxicities were myelosuppression, especially neutropenia and thrombocytopenia. Grade 3–4 neutropenia and thrombocytopenia occurred in 68 and 50% of the patients, respectively. The neutrophil and

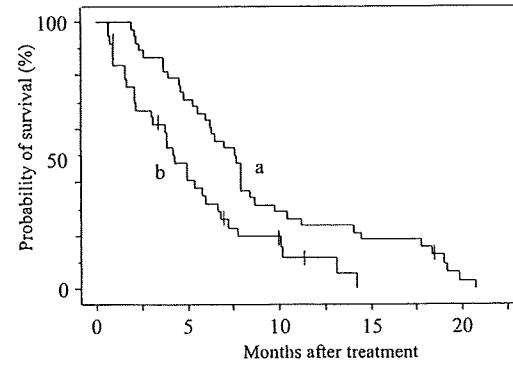

Figure 1. Overall survival curve (a) and time to progression (b) for all 38 patients.

Table 2. Treatment-related adverse events: worst grade reported during treatment period

Toxicity	Grade (No. of patients)					
	1	2	3	4	1–4 (%)	3–4 (%)
Hematological						
Leucopenia	10	11	13	4	100	45
Neutropenia	2	9	15	11	97	68
Anemia	7	15	13	2	97	39
Thrombocytopenia	10	8	18	1	97	50
Non-hematological						
Nausea	12	10	9	–	82	24
Vomiting	15	9	1	0	66	3
Diarrhea	8	0	2	0	26	5
Anorexia	9	10	15	0	89	39
Stomatitis	2	0	1	1	11	5
Rash	0	5	1	0	16	3
Alopecia	7	2	–	–	24	–
Fatigue	16	11	2	0	76	5
Fever	8	1	0	0	24	0
Peripheral neuropathy	3	0	0	0	8	0
Total bilirubin	13	5	1	0	50	3
AST	8	6	3	0	45	8
ALT	10	8	4	0	58	11
Creatinine	11	9	0	0	53	0

AST, aspartate aminotransferase; ALT, alanine aminotransferase.

platelet count nadirs typically occurred on day 15. Although most of these hematologic toxicities were transient and reversible, one patient, a 45-year-old man, required hospitalization as a result of severe myelosuppression (grade 4 neutropenia and grade 4 thrombocytopenia) accompanied by severe non-hematological toxicities (grade 4 stomatitis, grade 3 rash, grade 3 fatigue and grade 3 febrile neutropenia) in the middle of the first cycle of treatment. After intensive medical therapies including antibiotics, granulocyte colony-stimulating factor and platelet transfusion, he recovered from these toxicities. No other unexpected severe toxicities were observed during the study and there were no treatment-related deaths. Although gastrointestinal toxicities such as nausea, vomiting and anorexia were frequently observed after cisplatin administration, most of them were manageable with appropriate medical treatment (all of the study patients received cisplatin on day 1 on an inpatient basis). There were no cumulative toxicities except for renal toxicity: six patients discontinued the protocol study because their creatinine clearance decreased to 50 ml/min or less after several cycles of treatment (median 4 cycles, range 1–5), although the serum creatinine level was within 2.0 mg/dl in all patients.

DISCUSSION

We conducted the present study to evaluate the efficacy and toxicity of gemcitabine and cisplatin combination therapy in 38 Japanese patients with metastatic pancreatic cancer. This combination therapy produced a relatively good response rate of 26%. In addition, the median TTP of 4.2 months and median overall survival of 7.5 months were better than those reported in most studies of gemcitabine monotherapy for advanced pancreatic cancer (TTP 2–3 months, overall survival about 6 months) (2–4). To date, several phase II studies of this combination for advanced pancreatic cancer have been published (Table 3) (11–16). Although those studies used various schedules of gemcitabine and cisplatin administration, most of them demonstrated promising efficacy of this combination, with a response rate of around 20% or higher and/or a median survival of >7 months.

The major toxicity of the gemcitabine and cisplatin combination is myelosuppression. In many studies of this combination, more than half of the patients were reported to suffer grade 3–4 neutropenia and/or thrombocytopenia during the study period (Table 3). Among these studies, hematological toxicity in our study was strong, with a 68% incidence of grade 3–4 neutropenia and a 50% incidence of thrombocytopenia. The schedule adopted in our study, in which cisplatin was administered as an undivided dose on day 1, might have enhanced these toxicities. Although the incidences of G3–4 neutropenia and thrombocytopenia in our study were high, most of such episodes were transient and resolved spontaneously. There was only one episode of neutropenic fever, no significant bleeding episodes and no treatment-related deaths. Furthermore, non-hematological toxicities including nausea and anorexia were manageable, and no unexpected ones occurred. Therefore, we conclude that the gemcitabine and cisplatin combination used according to our schedule is tolerable in patients with advanced pancreatic cancer. However, since the incidences of G3–4 hematological toxicity are high, caution will be required when using this regimen for patients with poor performance status.

Recently, Heinemann et al. (19) conducted a randomized phase III study comparing the gemcitabine plus cisplatin combination with gemcitabine alone. The combination regimen included gemcitabine 1000 mg/m^2 with cisplatin 50 mg/m^2 given on days 1 and 15 of a 28-day cycle. They reported that progression-free survival was improved in the combination arm (5.3 months versus 3.1 months, $P = 0.053$), although overall survival showed only a non-significant tendency for improvement (7.5 months versus 6.0 months, $P = 0.15$). Another randomized study performed by the Italian Group (20) also failed to demonstrate a survival benefit of combination treatment, although marked improvements in the response rate (26.4% versus 9.2%, $P = 0.02$) and TTP (20 weeks versus 8 weeks, $P = 0.048$) were demonstrated. Combination therapy with oxaliplatin, another platinum analog, has also failed to demonstrate a statistically

Table 3. Phase II studies of gemcitabine–cisplatin chemotherapy for advanced pancreatic cancer

Author	Gemcitabine (mg/m^2)	Cisplatin (mg/m^2)	Cycle (day)	No. of patients	RR (%)	Median TTP (month)	Median OS (month)	Grade 3/4 neutropenia (%)	Grade 3/4 thrombocsytopenia (%)
Brodowicz et al. (11)	1000, days 1, 8, 15	35, days 1, 8, 15	28	16	31	7.4	9.6	31	63
Clayton et al. (12)	1000, days 1, 8, 15	25, days 1, 8, 15	28	36	9	5.8	9.5	60	60
Heinemann et al. (13)	1000, days 1, 8, 15	50, days 1, 15	28	41	11	4.3	8.2	34	29
Philip et al. (14)	1000, days 1, 8, 15	50, days 1, 15	28	42	26	5.4	7.1	64	62
Cascinu et al. (15)	1000, days 1, 8	35, days 1, 8	21	45	9	3.6	5.6	6	11
Ko et al. (16)	1000[a], days 1, 8	20, days 1, 8	21	51	19	3.9	7.1	53	16
Current study	1000, days 1, 8, 15	80, day 1	28	38	26	4.2	7.5	68	50

RR, Response rate; TTP, Time to tumor progression; OS, Overall survival.
*Fixed-dose-rate infusion of 10 mg/m^2/min.

significant survival benefit in comparison with gemcitabine alone in two randomized phase III studies (21,22). Therefore, although many phase II studies including ours have shown promising efficacy for the gemcitabine plus platinum combination, the results of the phase III studies did not support the clinical use of this combination as a first-line therapy for advanced pancreatic cancer.

However, some recent studies have suggested potential activity of platinum-containing chemotherapy for advanced pancreatic cancer. Reni et al. (23) conducted a randomized study of a four-drug regimen including cisplatin, epirubicin, fluorouracil and gemcitabine (PEFG) in patients with advanced pancreatic cancer, and reported that patients allocated the PEFG regimen showed a small but significant improvement in overall survival: 1-year survival rate was 38.5% in the PEFG group and 21.3% in the gemcitabine group. Oettle et al. (24) performed a randomized study of second-line therapy for gemcitabine-refractory advanced pancreatic cancer and reported that the median survival time from the start of second-line therapy in the oxaliplatin/folinic acid/fluorouracil group was significantly longer than that in best supportive care group (21 weeks versus 10 weeks, $P = 0.0077$). Although the numbers of patients recruited in these studies were small, the results suggested that there is still room for assessing the value of platinum agents for treatment of pancreatic cancer.

In conclusion, our phase II study of gemcitabine plus cisplatin combination therapy demonstrated a good response rate of 26% in patients with metastatic pancreatic cancer with moderate toxicities. However, since all phase III studies reported so far have failed to demonstrate a survival benefit of adding platinum to gemcitabine for advanced pancreatic cancer, other strategies should be considered in further studies.

Conflict of interest statement

None declared.

References

1. Ministry of Health, Labour and Welfare. The dynamic statistics of the population in 2004. http://www.mhlw.go.jp/toukei/saikin/hw/jinkou/kakutei04/hyo7.html (accessed 1 June 2006).
2. Burris HA 3rd, Moore MJ, Andersen J, Green MR, Rothenberg ML, Modiano MR, et al. 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;15:2403–13.
3. Bramhall SR, Rosemurgy A, Brown PD, Bowry C, Buckels JA. Marimastat Pancreatic Cancer Study Group. Marimastat as first-line therapy for patients with unresectable pancreatic cancer: a randomized trial. *J Clin Oncol* 2001;19:3447–55.
4. 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;20:3270–5.
5. Moore MJ, Goldstein D, Hamm J, Figer A, Hecht JR, Gallinger S, et al. Erlotinib plus gemcitabine compared to gemcitabine alone in patients with advanced pancreatic cancer. A phase III trial of the National Cancer Institute of Canada Clinical Trials Group [NCIC-CTG]. *Proc Am Soc Clin Oncol* 2005;24:(abstr 1).
6. Cunningham D, Chau I, Stocken D, Davies C, Dunn J, Valle J, et al. Phase III randomised comparison of gemcitabine (GEM) versus gemcitabine plus capecitabine (GEM-CAP) in patients with advanced pancreatic cancer. Presented at 13th European Cancer Conference (ECCO) 2005:(abstr 617).
7. Wils JA, Kok T, Wagener DJ, Selleslags J, Duez N. Activity of cisplatin in adenocarcinoma of the pancreas. *Eur J Cancer* 1993;29A:203–4.
8. Peters GJ, Bergman AM, Ruiz van Haperen VW, Veerman G, Kuiper CM, Braakhuis BJ. Interaction between cisplatin and gemcitabine *in vitro* and *in vivo*. *Semin Oncol* 1995;22:72–9.
9. Sandler AB, Nemunaitis J, Denham C, von Pawel J, Cormier Y, Gatzemeier U, et al. Phase III trial of gemcitabine plus cisplatin versus cisplatin alone in patients with locally advanced or metastatic non-small-cell lung cancer. *J Clin Oncol* 2000;18:122–30.
10. von der Maase H, Hansen SW, Roberts JT, Dogliotti L, Oliver T, Moore MJ, et al. Gemcitabine and cisplatin versus methotrexate, vinblastine, doxorubicin and cisplatin in advanced or metastatic bladder cancer: results of a large, randomized, multinational, multicenter, phase III study. *J Clin Oncol* 2000;18:3068–77.
11. Brodowicz T, Wolfram RM, Kostler WJ, Tomek S, Vaclavik I, Steger GG, et al. Phase II study of gemcitabine in combination with cisplatin in patients with locally advanced and/or metastatic pancreatic cancer. *Anticancer Drugs* 2000;11:623–8.

12. Clayton AJ, Mansoor AW, Jones ET, Hawkins RE, Saunders MP, Swindell R, et al. A phase II study of weekly cisplatin and gemcitabine in patients with advanced pancreatic cancer: is this a strategy still worth pursuing? *Pancreas* 2006;32:51–7.
13. Heinemann V, Wilke H, Mergenthaler HG, Clemens M, Konig H, Illiger HJ, et al. Gemcitabine and cisplatin in the treatment of advanced or metastatic pancreatic cancer. *Ann Oncol* 2000;11:1399–403.
14. Philip PA, Zalupski MM, Vaitkevicius VK, Arlauskas P, Chaplen R, Heilbrun LK, et al. Phase II study of gemcitabine and cisplatin in the treatment of patients with advanced pancreatic carcinoma. *Cancer* 2001;92:569–77.
15. Cascinu S, Labianca R, Catalano V, Barni S, Ferrau F, Beretta GD, et al. Weekly gemcitabine and cisplatin chemotherapy: a well-tolerated but ineffective chemotherapeutic regimen in advanced pancreatic cancer patients. A report from the Italian Group for the Study of Digestive Tract Cancer (GISCAD). *Ann Oncol* 2003;14:205–8.
16. Ko AH, Dito E, Schillinger B, Venook AP, Bergsland EK, Tempero MA. Phase II study of fixed dose rate gemcitabine with cisplatin for metastatic adenocarcinoma of the pancreas. *J Clin Oncol* 2006;24:379–85.
17. Kurita Y, Yokoyama A, Matsui K, Hara N, Nakai Y, Ohhashi Y, et al. Phase I study of gemcitabine hydrochloride (LY 188011) combination therapy with cisplatin in the patients with non-small cell lung cancer [in Japanese]. *Gan To Kagaku Ryoho* 1999;26:898–907.
18. Japan Society for Cancer Therapy. Criteria for the evaluation of the clinical effects of solid cancer chemotherapy. *J Jpn Soc Cancer Ther* 1993;28:101–30.
19. Heinemann V, Quietzsch D, Gieseler F, Gonnermann M, Schonekas H, Rost A, et al. Randomized phase III trial of gemcitabine plus cisplatin compared with gemcitabine alone in advanced pancreatic cancer. *J Clin Oncol* 2006;24:3946–52.
20. Colucci G, Giuliani F, Gebbia V, Biglietto M, Rabitti P, Uomo G, et al. 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 Oncologia dell'Italia Meridionale. *Cancer* 2002;94:902–10.
21. Louvet C, Labianca R, Hammel P, Lledo G, Zampino MG, Andre T, et al. Gemcitabine in combination with oxaliplatin compared with gemcitabine alone in locally advanced or metastatic pancreatic cancer: results of a GERCOR and GISCAD phase III trial. *J Clin Oncol* 2005;23:3509–16.
22. Poplin E, Levy DE, Berlin J, Rothenberg M, Cella D, Mitchell E, et al. Phase III trial of gemcitabine (30-minute infusion) versus gemcitabine (fixed-dose-rate infusion) versus gemcitabine + oxaliplatin in patients with advanced pancreatic cancer. *Proc Am Soc Clin Oncol* 2006;25: (abstr 4004).
23. Reni M, Cordio S, Milandri C, Passoni P, Bonetto E, Oliani C, et al. Gemcitabine versus cisplatin, epirubicin, fluorouracil, and gemcitabine in advanced pancreatic cancer: a randomised controlled multicentre phase III trial. *Lancet Oncol* 2005;6:369–76.
24. Oettle H, Pelzer U, Stieler J, Hilbig A, Roll L, Schwaner I, et al. Oxaliplatin/folinic acid/5-fluorouracil [24 h] (OFF) plus best supportive care versus best supportive care alone (BCC) in second-line therapy of gemcitabine-refracitory advanced pancreatic cancer (CONKO 003). *Proc Am Soc Clin Oncol* 2005;24:(abstr 4031).

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Minireview

Pharmacogenomics of gemcitabine: can genetic studies lead to tailor-made therapy?

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Gemcitabine is a deoxycytidine analogue that has a broad spectrum of antitumour activity in many solid tumours including pancreatic cancer. We have recently carried out a pharmacogenomic study in cancer patients treated with gemcitabine, and found that one genetic polymorphism of an enzyme involved in gemcitabine metabolism can cause interindividual variations in the pharmacokinetics and toxicity of this agent. In this paper, we review recent genetic studies of gemcitabine, and discuss the possibility of individualised cancer chemotherapy based on a pharmacogenomic approach.
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With progress in the development of anticancer agents, many cancer patients now benefit from chemotherapy. Before treatment, however, it is difficult to predict whether the selected chemotherapy will be really effective and tolerable to the patient. Therefore, considerable effort has been made to obtain information that could be used to devise tailor-made therapy. Recent progress in molecular biology has revealed that genetic factors can at least partly explain interindividual variations in the efficacy and toxicity of anticancer agents. We have recently carried out a prospective pharmacogenomic study in cancer patients treated with gemcitabine (2′,2′-difluorodeoxycytidine, dFdC), and found that one of the single-nucleotide polymorphisms (SNPs) in the cytidine deaminase gene influences the pharmacokinetics and toxicities of this agent (Sugiyama *et al*, 2007). Gemcitabine is a deoxycytidine analogue that demonstrates broad anticancer activity in various solid tumours, including pancreatic cancer and non-small-cell lung cancer (NSCLC). Because of the widespread use of gemcitabine, a better understanding of the mechanisms determining its activation, and development of resistance against it has been needed, and this has prompted active genetic studies in relation to this agent. In this review, therefore, we focus on genetic studies of gemcitabine that have yielded data potentially useful for the establishment of individualised cancer chemotherapy.

GEMCITABINE METABOLISM AND MECHANISM OF ACTION

Like cytarabine, another widely used nucleoside analogue, gemcitabine is a prodrug that requires cellular uptake and intracellular phosphorylation in order to exert its action (Figure 1) (Fukunaga *et al*, 2004; Mini *et al*, 2006). Once administered, gemcitabine is transported into cells by nucleoside transporters. Gemcitabine is then phosphorylated into gemcitabine monophosphate (dFdCMP) by deoxycytidine kinase (DCK), and dFdCMP is subsequently phosphorylated to gemcitabine diphosphate (dFdCDP) and gemcitabine triphosphate (dFdCTP) by nucleoside monophosphate (UMP/CMP) and diphosphate kinase. Gemcitabine exerts its cytotoxic effect mainly through inhibition of DNA synthesis by being incorporated into the DNA strand as the active dFdCTP. It is known that gemcitabine has a unique mechanism of action known as 'self-potentiation' (Heinemann *et al*, 1992). For example, dFdCDP potently inhibits ribonucleotide reductase, resulting in a decrease of competing deoxyribonucleotide pools necessary for DNA synthesis. Again, dFdCTP suppresses inactivation of dFdCMP by inhibiting deoxycytidine monophosphate deaminase (DCTD). On the other hand, more than 90% of administered gemcitabine is converted, and thus inactivated, by cytidine deaminase (CDA) into 2′-deoxy-2′,2′-difluorouridine (dFdU). Phosphorylated metabolites of gemcitabine are reduced by cellular 5′-nucleotidase (5′-NT), and dFdCMP is also converted, and inactivated, by DCTD into 2′-deoxy-2′,2′-difluorouridine monophosphate (dFdUMP).

This paper discusses these various metabolic pathways related to gemcitabine cellular pharmacology and DNA repair. In Table 1, we summarise the genetic polymorphisms related to gemcitabine pathways, their allele frequencies in different ethnic groups, and the resulting functional changes. In this paper, A of the translation initiation codon ATG is numbered 1 and the first methionine of a protein is numbered 1.

NUCLEOSIDE TRANSPORTERS

Gemcitabine is transported into cells by five nucleoside transporters, two equilibrative nucleoside transporters (ENTs; ENT1

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