TABLE 1
Characteristics of Eligible Patients

	Gemcitabine	Gemcitabine+UFT	*P*
Patients, n	49	50	
Age, median (range), y	63 (38-78)	63 (38-78)	.35
Sex, women/men	31 / 18	33 / 17	.78
Period from surgery to randomization, median (IQR), d	30.0 (26.0-37.0)	26.5 (20.25-35.75)	.41
Period from surgery to start of adjuvant chemotherapy, median (IQR), d	35.0 (27.0-41.0)	30.0 (23.25-42.50)	.48
Operative procedure, PD/DP/TP	38/8/3	38/9/3	.98
UICC stage, IA/IB/IIA/IIB/III/IV	0/1/13/26/2/7	1/1/10/33/1/4	.64
JPS stage, I/II/III/IVa/IVb	0/2/16/22/9	1/2/21/17/9	.68
Primary tumor status, 1/2/3/4	0/2/45/2	2/4/23/1	.39
Nodal status, 0/1	15/34	13/37	.61
Distant metastasis, 0/1	42/7	46/4	.32
Resection status, 0/1	32/17	41/9	.06
Histology			.88
Tubular adenocarcinoma, well/mod/poor	3/34/6	6/33/7	
Papillary adenocarcinoma	1	1	
Adenosquamous carcinoma	1	1	
Anaplastic carcinoma	1	1	
Invasive carcinoma derived from intraductal tumor	3	1	

UFT indicates tegafur/uracil; IQR, interquartile range; PD, pancreaticoduodenectomy; DP, distal pancreatectomy; TP, total pancreatectomy; UICC, International Union Against Cancer; well, well-differentiated type, mod, moderately differentiated type; poor, poorly differentiated type. JPS, Japan Pancreas Society.

(undifferentiated carcinoma) determined after randomization. The remaining 99 patients were included in the analysis. The baseline characteristics of the patients in the 2 groups were comparable (Table 1). There was no statistical difference between the 2 groups in the median time from surgery to the start of chemotherapy of 35.0 days in the GEM group and 30.0 days in the GEM + UFT group.

Treatment Data

All patients received at least 1 dose of gemcitabine. The median number of the gemcitabine administrations for a patient was 12 times for the GEM group and 14 times for the GEM + UFT group. Median relative dose intensity within the first 4 cycles was 89.1% for the GEM group and 87.4% for the GEM + UFT group; there was no statistical difference between the 2 groups (Table 2). Median duration of UFT administration in the GEM + UFT group was 5 months, and the median relative dose intensity within the first 4 cycles was 100% (Table 2).

Thirty-six patients (73.5%) in the GEM group and 30 patients (60.0%) in the GEM + UFT group completed 4 or more cycles of treatment. The reasons for treatment discontinuation within 4 cycles in the GEM group were recurrent disease (10 patients, 76.9%), adverse events (2 patients, 15.4%), and patient's wish (1 patient, 7.7%). In the GEM + UFT group, the reasons were recurrent disease (5 patients, 25.0%), adverse events (11 patients, 55.0%), and patient's wish (4 patients, 20.0%).

TABLE 2
Total Dose and Relative Intensity

	Gemcitabine, median (range)	Gemcitabine+UFT, median (range)	*P*
Gemcitabine			
Total amount, g	19.2 (3.9-76.5)	19.2 (2.5-113.1)	.86
Administrations, n	12 (3-76)	14 (2-81)	.67
Relative dose intensity, %	89.1 (22.5-100)	87.4 (13.5-100)	.69
UFT			
Total amount, g	—	27.8 (1.2-158.0)	
Duration of administration, mo	—	5 (1-26)	
Relative dose intensity, %	—	100 (5.4-100)	

UFT indicates tegafur/uracil.

Toxicity

Although the majority of the patients, especially those in the GEM + UFT group, experienced minor toxicity, no grade 4 or higher toxicities were observed in either group (Table 3). Fifteen (30.6%) patients in the GEM group and 12 (24.0%) patients in the GEM + UFT group experienced grade 3 toxicity, mainly leukocytopenia. Two patients in the GEM group and 11 patients in the GEM + UFT group discontinued treatment within 4 cycles because of repeated toxicities despite dose modification. All toxicities were reversible and resolved with conservative treatment alone in all patients.

Efficacy

With a median observation period of 21 months (range, 3 months to 57 months), recurrent disease developed at comparable rates of 73.5% in the GEM group (36 of 49 patients) and 78% in the GEM + UFT group (39 of 50 patients). The sites of recurrence were similar in both groups (GEM group and GEM + UFT group); the local recurrence was observed in 75.0% and 69.2% of patients, respectively. The number of patients with local recurrence alone was 13 (36.1%) in the GEM group and 17 (43.6%) in the GEM + UFT group. The most frequent primary site of distant metastasis was the liver, with 12 (33.3%) patients of the GEM group and 13 (33.3%) patients of the GEM + UFT group. The estimated

TABLE 3
Summary of Toxicities

	Gemcitabine			Gemcitabine+UFT		
	Any Grade	Grade 3	Grade 4	Any Grade	Grade 3	Grade 4
Total	29 (59.2%)	15 (30.6%)	0 (0.0%)	45 (90.0%)	12 (24.0%)	0 (0.0%)
Hematologic						
Leukocytes	26 (53.1%)	11 (22.4%)	0 (0.0%)	36 (72.0%)	9 (18.0%)	0 (0.0%)
Hemoglobin	20 (40.8%)	4 (8.2%)	0 (0.0%)	17 (34.0%)	2 (4.0%)	0 (0.0%)
Platelets	13 (26.5%)	3 (6.1%)	0 (0.0%)	11 (22.0%)	0 (0.0%)	0 (0.0%)
Nonhematologic						
Nausea/vomiting	10 (20.4%)	0 (0.0%)	0 (0.0%)	12 (24.0%)	0 (0.0%)	0 (0.0%)
Anorexia	9 (18.4%)	1 (2.0%)	0 (0.0%)	14 (28.0%)	1 (2.0%)	0 (0.0%)
Biochemical						
AST/ALT	11 (22.4%)	0 (0.0%)	0 (0.0%)	11 (22.0%)	1 (2.0%)	0 (0.0%)
Glucose intolerance	1 (2.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	1 (2.0%)	0 (0.0%)

AST indicates aspartate aminotransferase; ALT, alanine aminotransferase.

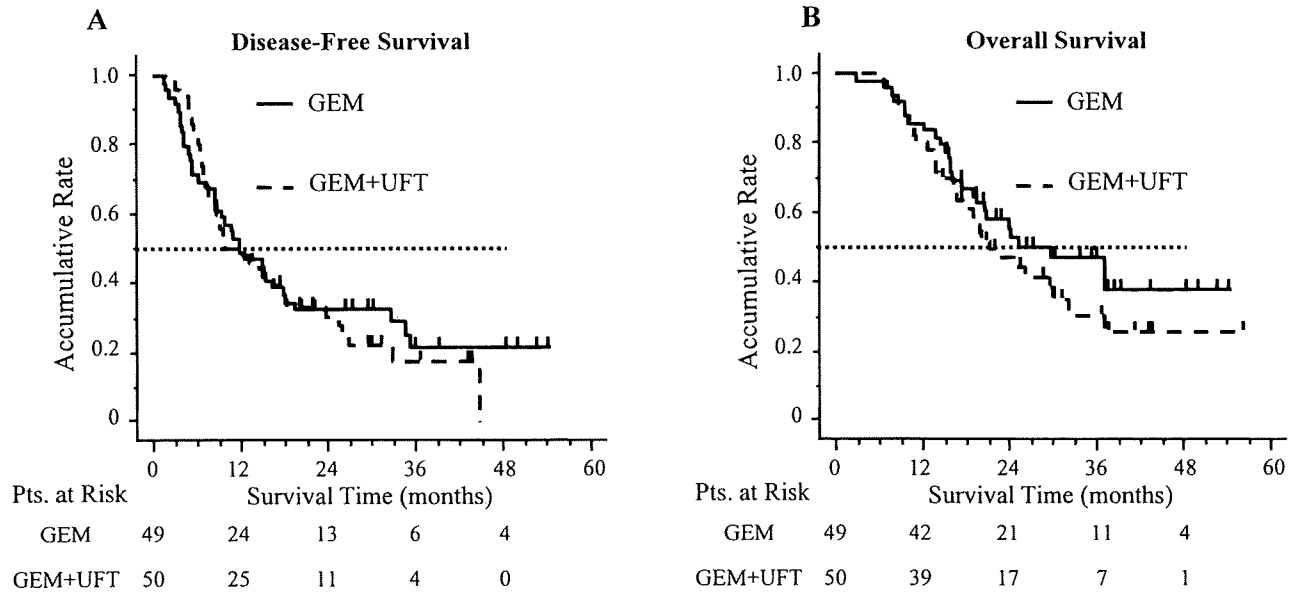

FIGURE 2. Disease-free (A) and overall (B) survival are shown. GEM indicates gemcitabine alone; Pts., patients; UFT, tegafur/uracil.

1- and 3-year disease-free survival rates were 49.0% and 21.6% in the GEM group and 50.0% and 17.7% in the GEM + UFT group, respectively. The median disease-free survival time was also comparable to 12.0 months in the GEM group and 12.3 months in the GEM + UFT group (log-rank, $P = .67$, Fig. 2A).

In the randomized patients, 57 patients (26 in the GEM group and 31 in the GEM + UFT group) died because of recurrent disease; there were no deaths attributed to any other causes in the observation period. The median overall survival time was 29.8 months in the GEM group and 21.2 months in the GEM + UFT group. The estimated survival rates at 1 and 3 years were 85.7% and 46.9% in the GEM group and 80.0% and 30.4% in the GEM + UFT group, respectively. There was no statistical difference between the overall survival times of the GEM group and the GEM + UFT group (log-rank, $P = .28$, Fig. 2B).

Prognostic Factors for Patients With Adjuvant Chemotherapy

We analyzed the clinical outcomes of all patients in this study to estimate the efficacy of adjuvant chemotherapy using gemcitabine for patients with resected pancreatic cancer. The median disease-free

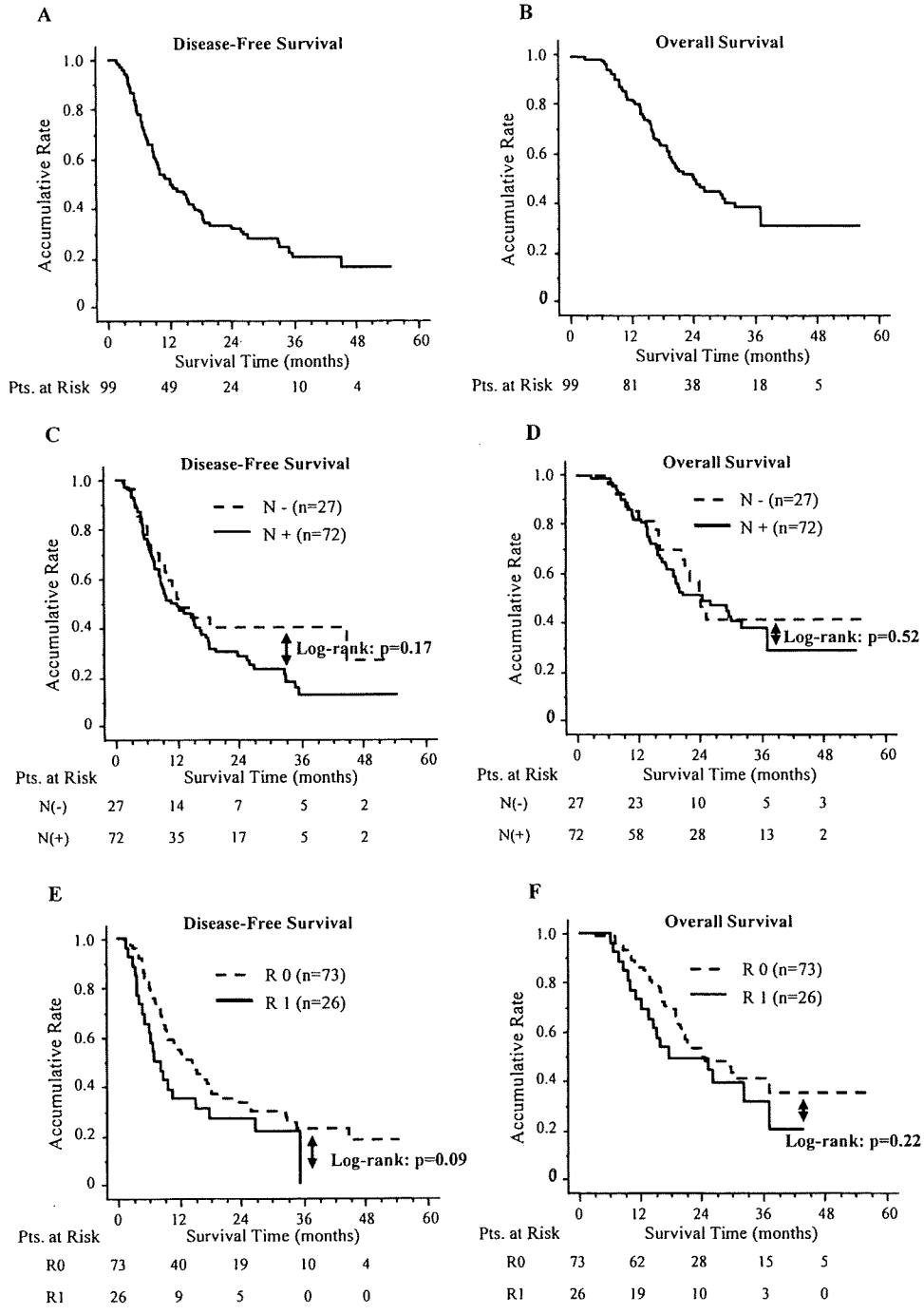

FIGURE 3. Disease-free and overall survival of all patients (Pts.) are shown: (A) disease-free survival; (B) overall survival; (C, D) disease-free (C) and overall (D) survival of the patients categorized by nodal status (solid line indicates lymph node positive [N+]; dotted line: lymph node negative [N−]); (E, F) disease-free (E) and overall (F) survival of the patients categorized by resection status (R) (solid line indicates R1; dotted line, R0).

survival time of all 99 patients in this study was 12.0 months, and the estimated 1- and 3-year disease-free survival rates were 49.5% and 19.5%, respectively (Fig. 3A). The median overall survival time of those patients was 24.1 months, and the estimated 1- and 3-year survival rates were 82.8% and 38.8%, respectively (Fig. 3B).

To assess the influence of prognostic factors, the relationships between the survival outcomes and the following variables were investigated: sex, age (≤63

years/>63 years), tumor location (head/body or tail), International Union Against Cancer stage (IA-IIA/IIB-IV), Japan Pancreas Society (JPS) stage (I-III/IVa, b), operation time (≤444 minutes/>444 minutes), blood loss during operation (≤920 mL/>920 mL), tumor size (≤2 cm/>2 cm), nodal status (negative/positive), para-aorta lymph node metastasis (negative/positive), tumor histology (poorly differentiated tubular adenocarcinoma/other), and resection status (R0/R1). We performed univariate analysis of these factors for disease-free and overall survival times. Among these factors, only the JPS stage showed a significant value for disease-free survival time (hazard ratio, 0.473; 95% confidence interval; 0.288-0.775; $P = .003$), and no factors exerted a significant influence on overall survival time.

Because factors that are known to have prognostic value for survival such as nodal status and resection status did not show significant values on univariate analysis, we performed a Kaplan-Meier analysis of the disease-free and overall survival times for all 99 patients, and categorized the outcome by these factors. As shown in univariate analysis, there were no significant differences between patients with and without lymph node metastasis with respect to disease-free and overall survival times (Fig. 3C, D). Moreover, there were no significant differences between patients with R0 and R1 resection with respect to disease-free and overall survival times (Fig. 3E, F).

DISCUSSION

For the treatment of patients with pancreatic cancer, even with advanced cancer, the best survival rates are achieved after surgical resection.[3,4] However, because of the high recurrence rate, the prognosis of the patient remains poor even after curative surgery. This indicates that it is important to establish an effective multidiscipline therapy for pancreatic cancer. In this study, we aimed to estimate the efficacy of adjuvant chemotherapy using gemcitabine and UFT for patients with resected pancreatic cancer.

Burris et al first reported in 1997 that gemcitabine improved the survival of patients with advanced pancreatic cancer, with a median survival time of 5.65 months compared with 4.41 months for patients treated with 5-FU.[19] Since then, gemcitabine has become the first-line chemotherapy for patients with pancreatic cancer. Meanwhile, several randomized studies have shown that 5-FU-based adjuvant chemotherapy can improve the survival of patients with resected pancreatic cancer.[13,27] Therefore, we strongly expected that adjuvant chemotherapy using gemcitabine would improve the survival of our patients. We thus attempted in our phase II study to optimize the efficacy of adjuvant chemotherapy by combination with gemcitabine and other agents.

We selected UFT as the agent to use in combination with gemcitabine. There were several reasons. First, in vitro analysis showed that pretreatment of pancreatic cancer cells with 5-FU increased the intracellular concentration of gemcitabine, suggesting that UFT, a prodrug of 5-FU, might have the potential to supplement the therapeutic benefits of gemcitabine.[22] Second, UFT, an oral fluoropyrimidine, might be more convenient to administer than continuous 5-FU infusion, especially in an adjuvant setting. In addition, several groups had shown favorable results from the use of a combination of gemcitabine and UFT as a treatment for advanced pancreatic cancer.[28,29]

Our study showed that adjuvant chemotherapy with gemcitabine, with or without UFT, could be carried out with acceptable safety. No grade 4 toxicities were observed in any patients in either group, and no patient died because of toxic events related to adjuvant therapy. Grade 3 hematologic toxicities were observed in about 30% of the patients in both groups. Leukocytopenia was most frequently observed, as shown in the CONKO-001 study.[20] Although a high incidence of leukocytopenia from gemcitabine has also been reported in the treatment of nonresected pancreatic cancer with 9.7% grade 3 leukocytopenia,[19] the frequency in this study was relatively high. Onoue et al have reported as well that severe leukocytopenia induced by gemcitabine administration developed readily in patients who had undergone surgical resection.[30] This suggests that it is very important to observe patients closely, especially in adjuvant chemotherapy, to avoid fatal toxicities. Nevertheless, as no serious adverse events were observed in this study, we concluded that the adjuvant chemotherapy using gemcitabine with or without UFT can be carried out safely.

Unfortunately, this study failed to show any additional benefit in using UFT in concert with gemcitabine for patients with resected pancreatic cancer. Disease-free survival was similar in both groups, with a 1-year disease-free survival rate of 49.0% in the GEM group and a 50.0% rate in the GEM + UFT group. Moreover, the overall survival rate was slightly worse among the patients of the GEM + UFT group than among those of the GEM group, with median survival time of 21.2 months and 29.8 months, respectively. Although the observation period was short, we concluded from our data that other combinations with gemcitabine must be considered as

future trials for adjuvant chemotherapy for resected pancreatic cancer.

Although UFT did not induce any survival benefits, the patients of both groups who received gemcitabine adjuvant chemotherapy after surgery experienced relatively longer survival times. The median disease-free survival time of the total of 99 patients in this study was 12.0 months, and the median overall survival time was 24.1 months. The overall median survival time is usually reported as about 10 months to 20 months for patients with resected pancreatic cancer who did not received adjuvant chemotherapy.[12-17,20] In addition, survival in this study was favorable compared with studies of adjuvant therapy for pancreatic cancer. The median overall survival time of patients with adjuvant therapy was reported as 20.0 months in the GITSG study[12] and 20.1 months in the ESPAC-1 study.[17] Also, as expected, our data were almost equivalent with the CONKO-001 study, in which the median survival time of patients with gemcitabine adjuvant chemotherapy was 22.1 months. These results support the use of gemcitabine as adjuvant chemotherapy in resectable pancreatic cancer.

The involvement of radiation as the adjuvant therapy for patients with resected pancreatic cancer has been discussed. The GITSG study showed the efficacy of chemoradiation as the adjuvant therapy.[12] Conversely, the recent ESPAC-1 study failed to show the efficacy of this therapy on postoperative survival, in contrast to chemotherapy alone.[17] In this study, although the local recurrence was most frequently observed, many of these patients also showed other types of recurrences at the same time. This may indicate that the effect of radiation therapy as the adjuvant therapy might be limited regarding the survival of patients with resected pancreatic cancer.

Interestingly, in this study there were no significant differences in disease-free and overall survival rates between N− and N+ patients and between R0 and R1 patients. Nodal status (N) and resection status (R) are usually considered prognostic factors for pancreatic cancer after resection.[31-33] However, the ESPAC-1 study also reported that patients with R1 resection also benefited from adjuvant chemotherapy.[31] These results may indicate that patients with N+ or R1 status benefit more from adjuvant chemotherapy using gemcitabine.

In conclusion, the present study did not demonstrate the efficacy of a UFT and gemcitabine combination as an adjuvant therapy for patients with resected pancreatic cancer compared with gemcitabine alone. It did, however, add further evidence that an adjuvant therapy using gemcitabine can produce favorable effects on the prognosis without any severe toxicity. This strongly suggests that further clinical trial resources for adjuvant chemotherapy should be addressed through the use of other combinations of agents with gemcitabine.

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ORIGINAL ARTICLE

High intratumoral dihydropyrimidine dehydrogenase mRNA levels in pancreatic cancer associated with a high rate of response to S-1

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Abstract
Purpose Although the prognosis in patients with pancreatic cancer has been poor, we recently reported unusually high response rate and survival benefit of S-1 treatment in patients with pancreatic cancer. The aim of this study was to reveal genetic background of this unique activity of S-1 against pancreatic cancer. S-1 is a novel oral fluoropyrimidine derivative consisting of Tegafur (FT) and dihydropyrimidine dehydrogenase (DPD) inhibitor (5-chloro-2,4-dihydroxypyridine; CDHP). Accordingly, intratumoral DPD mRNA expression level was measured to reveal whether the level in pancreatic cancer was different from other GI cancer and whether it was relevant to chemosensitivity.
Methods Thirty-three recurrent pancreatic cancer patients treated with S-1 were studied. We obtained 15 responders and 13 non-responders according to the change of serum CA19-9. The mRNA was extracted from paraffin-embedded surgical specimens using laser captured microdissection, and relative expression levels of each DPD/β-actin were measured using a quantitative reverse transcription polymerase chain reaction (RT-PCR) (Taqman) system. Forty-four colorectal cancer patients and 20 gastric cancer patients treated with S-1 were enrolled as control groups. Thymidylate synthase (TS) mRNA expression levels were also measured.
Results Intratumoral DPD mRNA expression level was significantly higher in pancreatic cancer than that in colorectal cancer ($P = 0.0003$; median level, 1.38 vs. 0.44) and gastric cancer ($P = 0.0061$; 1.38 vs. 0.82). No difference in TS mRNA expression levels was observed among cancer types. DPD expression among responded pancreatic cancer was significantly lower than non-responded. ($P = 0.012$, Mann–Whitney U test).
Conclusions Intratumoral DPD mRNA expression level in pancreatic cancer was significantly higher than the other malignancies. This result may elucidate possible reasons for the high effectiveness of S-1 in pancreatic cancer.

Keywords Pancreatic cancer · DPD · S-1 · Gene expression

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Introduction

The outcomes of patients with pancreatic cancer remain very poor. Even after curative resection, the 5-year survival rate is only 7% [19]. 5-Fluorouracil (5-FU) had been the mainstay of treatment for pancreatic cancer, although, its response rate as a single agent is less than 20% [9]. Clinical trials have shown that 5-FU-based combination chemotherapy is no more effective than single-agent treatment, with greater toxicity [4, 5, 7]. In recent studies, gemcitabine showed a survival benefit over 5-FU [3] and is now used as a standard treatment for pancreatic cancer.

S-1 is a novel oral fluoropyrimidine derivative consisting of tegafur (FT) and two modulators, 5-chloro-2,4-dihydoxypyridine (CDHP) and potassium oxonate (Oxo) [24]. Antitumor effect is provided by the 5-FU prodrug FT. CDHP competitively inhibits the 5-FU degradative enzyme dihydropyrimidine dehydrogenase (DPD), resulting in prolonged active concentrations of 5-FU in blood [12, 26]. Nowadays, S-1 is widely used to treat many types of cancer, including gastric cancer [23], colorectal cancer [20], breast cancer [22], and head and neck cancer [18].

We have used S-1 in patients with pancreatic cancer and obtained a high response rate and prolonged survival [10]. Although, our previous study was small and retrospective, the response rate was 20% in patients given S-1 alone, as compared with 57.1% in those given S-1 plus cisplatin. To elucidate possible reasons for the high effectiveness of S-1 in pancreatic cancer, which is generally resistant to 5-FU-based therapy, we studied intratumoral DPD expression. We hypothesized that DPD expression levels were higher in pancreatic cancer than in other types of gastrointestinal cancers. We measured the DPD gene expression levels of pancreatic cancers by real-time reverse-transcriptase polymerase chain reaction (RT-PCR) and compared the results with the levels in colorectal cancer and gastric cancer. Since thymidylate synthase (TS) is the other key enzyme for 5-FU metabolism, TS gene expression levels were also measured in these samples.

Patients and samples

Thirty-three patients with recurrent pancreatic cancer were studied (21 men and 11 women; median age 61.5 years, range 37–80). All patients had undergone surgical resection between 1998 and 2001 at the Department of Gastroenterology, Tokyo Women's Medical University, Tokyo, Japan. Diagnoses were individually confirmed by histopathological examination. All patients received S-1 and cisplatin after confirmation of recurrence. S-1 was given orally twice daily for 21 days, and cisplatin 30 mg/m^2 was given on days 1 and 8, followed by a 2 week period of no treatment. The dose of S-1 was based on body surface area (BSA) as follows: BSA < 1.25 m^2, 40 mg; BSA > 1.25 but < 1.5 m^2, 50 mg; and BSA > 1.5 m^2, 60 mg. All of the patients were Japanese, and written informed consent was obtained from each patient according to institutional regulations. No patient had preoperatively received neoadjuvant chemotherapy. Serum CA19–9 tumor marker levels were measured every 2 weeks during chemotherapy. Forty-four patients with advanced colorectal cancer and 20 with advanced gastric cancer were studied as controls.

Microdissection

FFPE tumor specimens were cut into serial sections 10 μm in thickness. For pathological diagnosis, one slide was stained with hematoxylin and eosin and evaluated by a pathologist. Other sections were stained with nuclear fast red (NFR, American MasterTech Scientific Inc., Lodi, CA) to facilitate visualization of histologic features. All tumor samples underwent laser capture microdissection (P.A.L.M. Microlaser Technologies AG, Munich, Germany) to ensure that only tumor cells were dissected.

RNA isolation and cDNA synthesis

RNA was extracted and cDNA was prepared from each sample as described previously [15, 16].

Reverse transcription-PCR

Quantification of TS, DPD and an internal reference gene (β-actin) was done using a fluorescence-based real-time detection method (ABI PRISM 7900 Sequence Detection System [Taqman]; Applied Biosystems, Foster City, CA) as described previously [11]. The primers and probe sequences used are listed in Table 1. The PCR reaction mixture consisted of 1,200 nM of each primer, 200 nM probe, 0.4 U of AmpliTaq Gold Polymerase, 200 nM each of dATP, dCTP, dGTP, and dTTP, 3.5 mM $MgCl_2$ and 1× Taqman Buffer A containing a reference dye, to a final volume of 20 μl (all reagents from PE Applied Biosystems, Foster City, CA, USA). Cycling conditions were 50°C for 2 min, 95°C for 10 min, followed by 46 cycles at 95°C for 15 s and 60°C for 1 min. Gene expression values (relative mRNA levels) are expressed as ratios (differences between the Ct values) between the gene of interest (TS, DPD) and an internal reference gene (β-actin), which provides a normalization factor for the amount of RNA isolated from a specimen.

Statistical analysis

Median DPD mRNA levels were compared among pancreatic, colorectal, and gastric cancers by the Mann–Whitney *U* test, and multiple testing was corrected using the Benjamini and Hochberg False Discovery Rate. Median DPD mRNA levels were compared between responders and non-responders with the use of Mann–Whitney's *U* test. TS mRNA levels were compared by the Kruskal–Wallis test. *P*-values of less than 0.05 were considered to indicate statistical significance. All values were two-sided.

Table 1 Primers and probes

Thymidylate synthase (TS)		
Gen bank accession: NM_001071		
Forward primer	TS-764F	5′-GCCTCGGTGTGCCTTTCA-3′
Reverse primer	TS-830R	5′-CCCGTGATGTGCGCAAT-3′
Probe	TS-785T	5′-TCGCCAGCTACGCCCTGCTCA-3′
Dihydropyrimidine dehydrogenase (DPD)		
Gen bank accession: NM_000110		
Forward primer	DPD-51F	5′-AGGACGCAAGGAGGGTTTG-3′
Reverse primer	DPD-134R	5′-GTCCGCCGAGTCCTTACTGA-3′
Probe	DPD-71Tc	5′-CAGTGCCTACAGTCTCGAGTCTGCCAGTG-3′
β-actin		
Gen bank accession: NM_001101		
Forward primer	β-actin-592F	5′-TGAGCGCGGCTACAGCTT-3′
Reverse primer	β-actin-651R	5′-TCCTTAATGTCACGCACGATTT-3′
Probe	β-actin-611T	5′-ACCACCACGGCCGAGCGG-3′

Results

DPD mRNA expression levels in pancreatic, gastric, and colorectal cancers are shown in Table 2 and Fig. 1. The median DPD mRNA level in pancreatic cancer was significantly higher than the levels in colorectal cancer ($P = 0.0003$; median level, 1.38 vs. 0.44) and gastric cancer ($P = 0.0061$; 1.38 vs. 0.82). The median DPD mRNA level also significantly differed between gastric cancer and colorectal cancer ($P = 0.0025$; 0.82 vs. 0.44). No significant difference in TS mRNA levels was observed among pancreatic, colorectal, and gastric cancer ($P = 0.10$) (Table 2).

The serum CA19-9 tumor marker level was above the upper limit of normal (37 U/ml) in 28 of the 33 patients with pancreatic cancer. During chemotherapy the serum CA19-9 level fell by at least 50% in 18 patients (64.3%, responders) and either decreased by less than 50% or increased in 10 (35.7%, non-responders). The median DPD mRNA level in the responders was significantly lower than that in the non-responders ($P = 0.02$; 1.25 vs. 2.20) (Fig. 2).

None of the demographic and clinicopathological variables were significantly associated with the tumor response to S-1 chemotherapy at a P-value of 0.10 on Fisher's exact test (data not shown).

Table 2 Intratumoral DPD mRNA levels in various types of cancer

	Cancer type		
	Gastric	Pancreatic	Colorectal
Number of samples	20	33	44
DPD mRNA levels (median)	0.82	1.38	0.44
Range	0.17–2.21	0.21–4.16	0–2.64
TS mRNA levels (median)	3.26	2.38	2.47
Range	1.69–10.99	0.04–6.68	0.49–19.23

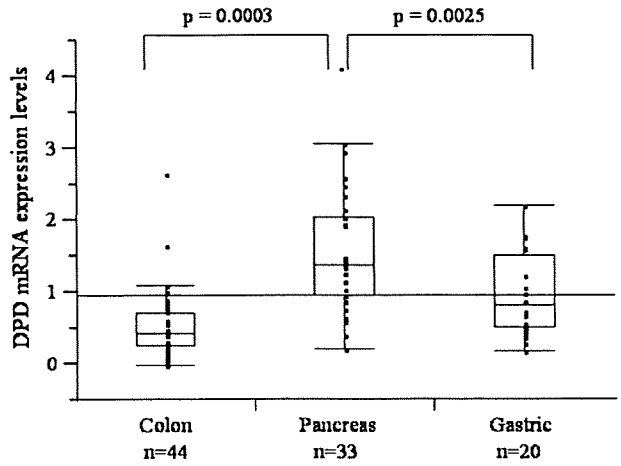

Fig. 1 DPD mRNA expression levels in pancreatic, colorectal, and gastric cancer. DPD expression in pancreatic cancer was significantly higher that that in colorectal [16] and gastric cancer [3]

Discussion

In our study, the DPD mRNA expression levels was significantly higher in pancreatic cancer than in colorectal cancer or gastric cancer, although no difference was seen in TS mRNA expression levels. This difference in DPD expression may explain the discrepancy between the high resistance to 5-fluorouracil and the high sensitivity to S-1 in patients with pancreatic cancer. In patients with colorectal cancer, treatment with 5-fluorouracil alone or a combination of 5-fluorouracil and leucovorin is somewhat effective [1, 6, 8, 21] and had been used as standard therapy before the advent of irinotecan. The response rate of pancreatic cancer is only 7% with 5-fluorouracil alone [5] and is not much higher with 5-fluorouracil plus leucovorin [4]. Ohtsu et al. [20] reported that S-1 had a response rate of 35% in

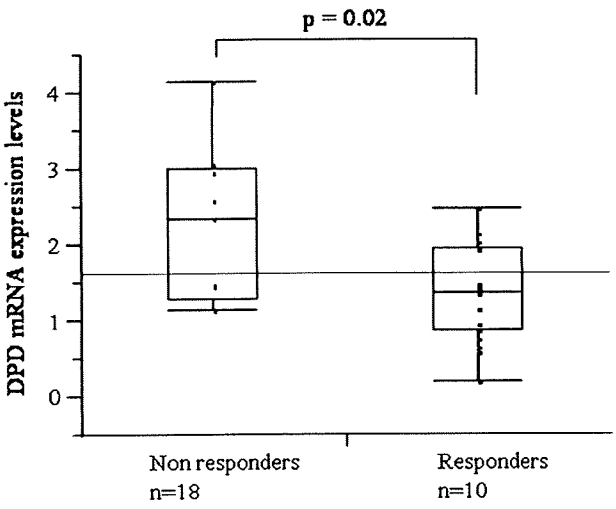

Fig. 2 DPD mRNA expression levels between responders and non-responders. Responders are defined as the serum CA19-9 level fell by at least 50% (18 patients), and non-responders are defined as their serum CA19-9 either decreased by less than 50% or increased (10 patients). DPD expression levels in responders were significantly lower than those in non-responders

phase II clinical trials of patients with colorectal cancer. This rate was not appreciably higher than the response rate with conventional 5-fluorouracil-based combination therapy. In pancreatic cancer, however, we previously obtained a response rate of higher than 50% with a combination of S-1 and cisplatin, which was considerably better than the response rate with conventional 5-fluorouracil treatment. Takechi et al. demonstrated that the in vitro antitumor activity of 5-FU against tumor cells with low DPD expression levels is not appreciably affected by the addition of CDHP. Against tumor cells with high DPD expression, however, the 50% inhibitory concentration differed by about twofold [25]. In tumors with low DPD expression such as colorectal cancer, the presence of CDHP provides no benefit, and the response to S-1 is similar to that with 5-fluorouracil. In tumors with high DPD expression such as pancreatic cancer, S-1 may be more effective than 5-fluorouracil.

Studies of DPD expression in patients with pancreatic cancer are scarce. Mori et al. used enzyme-linked immunosorbent assay [17] to measure DPD protein expression in various types of tumors and reported that pancreatic cancer had high DPD expression levels, similar to cancer of the neck, liver, esophagus, and breast [17]. Kamoshida et al. [14] used immunostaining to measure DPD levels in various types of tumors. They found that DPD expression was high in pancreatic cancer and low in colorectal cancer. To our knowledge, expression of DPD mRNA has not been assessed previously in pancreatic cancer. Uetake et al. reported that DPD gene expression correlated with DPD enzyme activity in colorectal cancer [27]. Mori et al. [17] showed that DPD enzyme activity strongly correlated with DPD protein expression as measured by ELISA. They found that the level of DPD protein expression was about threefold higher in pancreatic cancer and twofold higher in gastric cancer than in colorectal cancer, consistent with the mRNA levels measured by RT-PCR. These results suggest that the mRNA, protein, and activity levels of the DPD gene strongly correlate. DPD mRNA levels can be measured even in small specimens obtained by endoscopy or needle biopsy. The concomitant use of laser-captured microdissection allows DPD mRNA expression of tumors to be assessed quantitatively and objectively, providing clinically significant advantages over conventional procedures.

In our study, DPD levels were significantly lower in the responders to S-1 as assessed by the change in tumor marker levels than in the non-responders (1.25 vs. 2.20; $P = 0.02$). The median DPD level in the non-responders was 2.20, equivalent to about twofold that in the responders and fourfold that in colorectal cancer. Ichikawa et al. measured DPD mRNA levels in patients with gastric cancer and reported that the anticancer activity of S-1 does not depend on the level of DPD expression because CDHP inhibits DPD [13]. Although, the median DPD level did not differ significantly between responders and non-responders to S-1 in their study, patients in whom DPD expression exceeded a certain level did not respond to S-1. The inhibitory activity of CDHP against DPD is about 130-fold higher than that of uracil. However, in patients whose tumors have very high levels of DPD, the inhibitory effect of CDHP on DPD may be limited. In patients with such tumors, more potent DPD inhibitors should be used, or drugs with other mechanisms of action, such as gemcitabine, should be used instead of fluoropyrimidines. As for the former, eniluracil, a new, more potent DPD inhibitor, was developed and evaluated in clinical trials. Eniluracil decreases DPD enzyme activity by more than 99% in the liver and reduces 5-fluorouracil clearance by more than 18-fold [2]. In a phase II study of eniluracil combined with 5-fluorouracil in patients with pancreatic cancer, the median survival was only 3.6 months, indicating an unsatisfactory outcome. Moreover, 68% of the patients had grade 3 or higher toxicity. Subsequent clinical development was therefore terminated Rothenberg, 2002. Because eniluracil itself is free of serious toxicity, the toxic effects in clinical trials were attributed to prolonged, high serum concentrations of 5-fluorouracil. These results suggested the limitations of combination therapy with 5-fluorouracil and DPD inhibitors.

Our data may provide a molecular biologic basis for the high antitumor activity of S-1 in patients with pancreatic cancer. Gemcitabine has been shown to be somewhat effective against pancreatic cancer [3], but its anticancer activity

is far from satisfactory. Further studies of S-1, in patients with pancreatic cancer are awaited.

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Intrahepatic cholangiocarcinoma: analysis of 44 consecutive resected cases including 5 cases with repeat resections

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KEYWORDS:
Intrahepatic cholangiocarcinoma;
Liver resection;
Pulmonary resection

Abstract

BACKGROUND: Prognosis after resection for intrahepatic cholangiocarcinoma (ICC) remains unsatisfactory. There remains no effective therapy after recurrent ICC.

OBJECTIVE: The current study sought to evaluate risk factors associated with recurrent ICC and possible therapies after resection.

METHOD: A review of data from patients who underwent potentially curative resection for ICC was performed.

RESULTS: A total of 44 potentially curative resections were performed from 1995 to 2008. Mortality was 0% and morbidity was 35%. The 5-year overall and recurrence-free survival rates were 43% and 39%, respectively. Multivariate analysis identified the presence of multiple nodules and poor histologic grade as independent negative prognostic factors for overall and recurrent-free survival. Postoperative recurrence occurred in 25 patients (57%). Solitary recurrence occurred in 5 patients (liver, n = 4; lung, n = 1), all of who had undergone surgical resection. Three of the 5 patients survived for more than 5 years after 2 resections.

CONCLUSION: Prognosis after curative resection of solitary ICC appears favorable. In selected patients with sequential single hepatic or pulmonary recurrence, repeat resection may prolong survival.

Intrahepatic cholangiocarcinoma (ICC) is the second most common primary liver cancer, accounting for 10% of primary hepatic cancers.[1] In recent years, the incidence of ICC has been increasing worldwide, and the tumor is gaining attention.[1–4] In contrast to hepatocellular carcinoma, no strong high-risk groups have been identified for ICC. Therapeutic outcomes for patients with ICC are poor due to the highly malignant nature of the cancer. Surgical resection is currently the only curative option, but 5-year survival rates following curative resection range from 23% to 40%, with a median survival time of 18 to 37 months.[5–9] Furthermore, prognosis for patients with unresectable ICC is extremely poor, at less than 1 year.[4] No treatments have been established for unresectable or recurrent ICC. Although local chemotherapy, systemic chemotherapy, or a second surgical

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resection has been effective in selected cases, the number of patients involved is small and thus the efficacy of these therapies remains unclear. The present study retrospectively reviewed outcomes for ICC following resection in a single cancer hospital.

Patients and Methods

We retrospectively examined consecutive ICC cases in our institution. From January 1995 to February 2008, a total of 60 patients underwent exploratory surgery with the prospect of curative resection for ICC. Cases with concomitant hepatocellular carcinoma were excluded from this study and 8 patients displayed unresectable lesions, giving an overall resectability rate of 87% (52 of 60). Of these 52 resected cases, 44 ICC patients (15 women and 29 men) who underwent potentially curative resection were analyzed in this study. Eight cases of palliative resection (R2) were excluded for the following reasons: residual para-aortic lymph node metastases (n = 2), gross residual tumor at the resection margin (n = 4), and residual liver metastases in the residual liver (n = 2).

Tumors were staged according to the International Union Against Cancer (UICC) tumor-node-metastasis (TNM) classification system (6th ed).[10] Overall and disease-free survival rates were analyzed. The following clinicopathological features were analyzed: age; sex; primary site (colon/rectum); pStage (UICC); macroscopic type; preoperative serum carbohydrate antigen (CA) 19-9 level; preoperative serum carcinoembryonic antigen (CEA) level; bile duct invasion; vascular invasion; serosal invasion; number of nodules; lymph node metastases; tumor size; histologic grade; background liver status; lymph node dissection; and transfusion status. At our institution, ICC is generally treated by hemihepatectomy or extended hemihepatectomy. Systematic lymphadenectomy is not performed in the absence of metastasis to regional lymph nodes (hepatoduodenal nodes). Systemic lymphadenectomy along the common hepatic arteries and the hepatoduodenal ligament is per-

Table 1 Patient characteristics

Sex (M/F)	29/15	
Age (y) (range)	65.0 (41–85)	
pT stage (%)		
Stage I	14	32%
Stage II	8	18%
Stage IIIa	4	9%
Stage IIIb	0	0%
Stage IIIc	8	18%
Stage IV	10	23%
Macroscopic classification (%)		
Mass-forming type	41	93%
Intraductal type	2	5%
Infiltrating type	1	2%
Tumor size (cm) (median; range)	5.7 (2.0–12.0)	
Tumor number		
Solitary (%)	29	66%
Multiple (%)	15	34%
Background liver		
Normal liver	39	89%
Chronic hepatitis or liver fibrosis	4	9%
Cirrhosis	1	2%
Viral infection		
None	39	89%
Hepatitis B	1	2%
Hepatitic C	2	5%
HBC double-positive	2	5%

Table 2 Surgical procedures and results for 44 patients with intrahepatic cholangiocarcinoma

Surgical procedure	No.	%
Mortality	0	0%
Morbidity	13	30%
Transfusion	11	25%
Operation time (min)	435 (225–850)	
Blood loss (mL)	710 (260–3,440)	
Postoperative hospital stay (d)	21 (9–85)	
Type of hepatectomy		
Left hemihepatectomy	12	27%
Extended right hemihepatectomy	12	27%
Extended left hemihepatectomy	8	18%
Right hemihepatectomy	3	7%
Left trisectionectomy	3	7%
Central bisectionectomy	2	5%
Right trisectionectomy	2	5%
Limited resection	1	2%
Extended right lateral sectionectomy	1	2%
Combined resection		
Lymph node dissection	24	55%
Extrahepatic bile duct	12	27%
Stomach	1	2%
Pancreas	1	2%
Inferior vena cava	1	2%

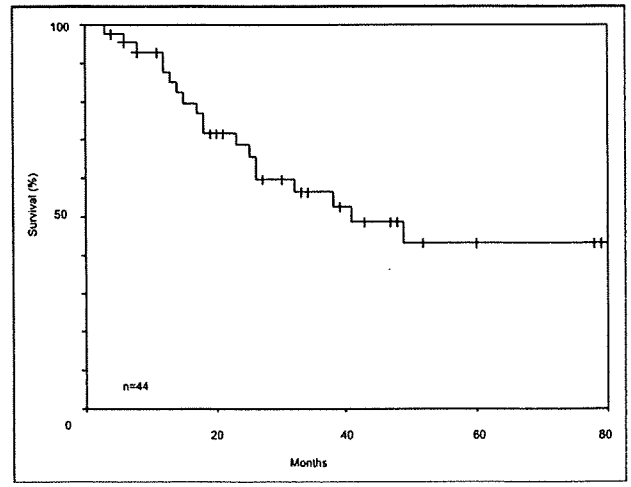

Figure 1 Kaplan-Meier overall survival for 44 patients who underwent curative resection for intrahepatic cholangiocarcinoma.

Table 3 Univariate analysis of risk factors associated with overall and recurrence-free survival for 44 patients who underwent curative resection for intrahepatic cholangiocarcinoma

Characteristic	n	5-year survival (%)	Median survival (mo)	P*	5-year disease-free (%)	Median disease-free (months)	P*
Overall	44	43	41		39	34	
Age (y)							
<70	31	44	32		35	15	
≥70	13	0	49	0.8140	28	41	0.309
Sex							
Male	29	44	49		35	18	
Female	15	43	41	0.8020	40	37	0.928
UICC stage							
1	14	79			83	74	
2	8	45	49	0.1560	31	34	.026*
3a	4	33	26	0.1090	0	4	.0014*
3c	8	0	41	.0141*	0	15	0.011
4	10	30	17	.0133*	20	11	.0063*
Macroscopic type							
Mass-forming type	41	42	41		37	34	
Intraductal type	2	100		0.1550	100		0.319
Infiltrating type	1	0		0.3060	0	11	0.196
Residual tumor							
R0	39	42	49		37	34	
R1	5	53		0.5470	60	0	0.908
Marginal width							
≥1 mm	27	60	82		50	67	
<1 mm	17	18	23	.0106*	21	12	.0359*
CA19-9							
<100 U/mL	34	47	49		48	41	
≥100 U/mL	10	27	17	.0215*	0	5	.002*
CEA							
<5	32	48	49		41	37	
≥5	8	32	23	0.1430	30	17	0.236
Bile duct invasion							
Absent	34	38	41		35	34	
Present	5	100		0.6090	80	67	0.792
Vascular invasion							
Absent	29	35	38		41	37	
Present	13	61	82	0.4050	23	13	0.424
Serosal invasion							
Absent	25	52	82		54	67	
Present	19	36	26	0.4700	26	14	0.193
No. of nodules							
Solitary	30	65	82		52	67	
Multiple	14	0	25	.0007*	0	6	.0022*
Lymph node metastases							
Absent	26	55			53	67	
Present	18	24	23	.0223*	15	13	.057*
Extrahepatic bile duct resection							
Absent	32	44	49		36	34	
Present	12	40	41	0.9840	41	37	0.887
Tumor size							
<5 cm	18	45	41		43	21	
≥5 cm	26	42	38	0.6480	34	18	0.359
Histological grading							
Well	14	62			53		
Mod	17	51	82	0.1490	45	17	0.161
Poor	9	12	15	.0001*	16	5	.0017*
Background liver							
Normal	39	42	38		36	18	
Injured	5	50	41	0.5540	50	37	0.217
Lymph node dissection							
Absent	20	44	49		48	41	
Present	24	41	32	0.3240	28	17	0.123
Transfusion							
Absent	33	47	41		43	17	
Present	11	38	49	0.7390	28	34	0.984

Well = well-differentiated adenocarcinoma; Mod = moderately differentiated adenocarcinoma; Poor = poorly differentiated adenocarcinoma.
*Log-rank test.

Table 4 Multivariate analysis of factors associated with overall and recurrence-free survival for 44 patients who underwent curative resection for intrahepatic cholangiocarcinoma

	Overall survival			Disease-free survival		
Risk factors	HR	95% CI	*P*	HR	95% CI	*P*
No. of tumors						
Solitary	1	—	—	1	—	—
Multiple	3.50	1.06–11.4	.039	2.98	1.15–7.71	.028
Histological grade						
Well or Mod	1	—	—	1	—	—
Poor	2.22	1.08–4.59	.030	2.01	1.07–3.73	.024

Well = well-differentiated adenocarcinoma; Mod = moderately differentiated adenocarcinoma; Poor = poorly differentiated adenocarcinoma.

formed if regional lymph nodes show metastasis, excluding para-aortic lymph nodes.

Postoperative monitoring comprised monthly blood biochemistry testing and diagnostic imaging such as computed tomography (CT) every 6 months. The therapeutic plan for recurrent cancer at the hospital is described. Surgical resection of the recurrent disease was performed for hepatic and pulmonary metastases if certain conditions were met, as follows: (1) hepatic and extrapulmonary lesions were solitary; and (2) surgery could be safely performed. However, in the case of ICC, the following conditions were added: (1) solitary lesion at any site, and (2) metachronous use of degradable starch microsphere transhepatic arterial chemoembolization (DSM-TACE) or hepatic arterial infusion (HAI) if the patient had only hepatic metastasis or if the hepatic metastasis was critical.

Systemic chemotherapy using gemcitabine or S-1 (TS-1; tegafur, gimeracil, oteracil, and potassium), an oral fluoropyrimidine, was performed on performance status 0/1 patients with recurrence in multiple organs after 2003.

Statistical analysis

Cumulative overall and disease-free survival rates were estimated according to Kaplan-Meier methods. The log-rank test was used to compare significant differences. Values of $P < .05$ were considered statistically significant. Parameters identified by univariate analysis of overall survival with $P < .05$ were entered into a Cox proportional hazard regression model to identify independent predictors of survival. All statistical analyses were conducted using SPSS version 9.0 software (SPSS, Chicago, IL).

Results

Patient characteristics are listed in Table 1. Mean duration of follow-up was 34 months (range 3–137 months; median 25.5 months). Surgical procedures and outcomes are listed in Table 2.

Eighteen of the 44 patients died of carcinoma progression, but no patient died of other disease. The cumulative overall survival rate was 87% at 1 year, 56% at 3 years, and 43% at 5 years (Figure 1). Cumulative recurrence-free survival rate was 64% at 1 year, 47% at 3 years, and 39% at 5 years. The median survival time for all patients was 41 months (95% confidence interval [CI] 18–63 months). Using univariate analysis, we found that 6 of 19 variables for overall or recurrence-free survival provided a significant estimate of prognosis (Table 3). In this study, all 10 patients with stage IV disease had lymph node metastases along the lesser curvatures and/or common hepatic arteries. UICC stage, multiple nodules, serum CA19-9 >100 U/mL, marginal width <1 mm, presence of lymph node metastasis, and poor histologic grade indicated significantly poor overall and recurrence-free survival. Multivariate analysis of the 5 factors other than UICC stage identified the presence of multiple nodules or poor histologic grade as independent prognostic factors (Table 4).

Postoperative recurrence occurred in 25 patients, with a median postoperative period of 23 months before recurrence (range 2–74 months; Table 5). Initial cancer-directed therapies after recurrence were surgical resection (N = 4: 3 liver, 1 lung), TACE, or HAI (n = 7, all liver), systemic chemotherapy (N = 6: 4 gemcitabine, 2 S-1) and best-practice supportive care (n = 4). One patient underwent liver resection following 3 courses of DSM-TACE. Table 6 provides data on 5 patients who underwent repeated resec-

Table 5 Site of relapse

	No. of patients (n = 44)	Percent
No. of relapses	25	57%
Site of first recurrence		
Liver	9	36%
Lymph nodes	3	12%
Lung	1	4%
Local	1	4%
Peritoneum	2	8%
Multiple sites	9	36%

Table 6 Course of five patients with repeat resection of recurrent ICC

Patient	Age (y)	Sex	Macroscopic type	Maximum tumor size (cm)	Number of tumours	Histology	Vascular invasion	Lymph node metastasis	1st recurrence Site	1st recurrence Therapy	1st recurrence Duration after 1st resection (mo)	2nd recurrence Site	2nd recurrence Therapy	2nd recurrence Duration after 1st resection (mo)	3rd recurrence Site	3rd recurrence Therapy	3rd recurrence Duration after 1st resection (mo)	Survival (mo)	Outcome
1	65	Male	MF	6	1	Well	P	A	Liver (solitary)	Resection*	4							33	NED
2	63	Male	MF	12	2	Well	A	A	Liver (solitary)	Resection	12	Local	Radiation	27				38	DFD
3	67	Male	MF	5	1	Mod	P	P	Liver (solitary)	Resection	13	Lung (solitary)	Resection	44				130	NED
4	59	Male	MF	9	1	Well	P	A	Liver (solitary)	Resection	34	Local	Chemo	42				79	AWD
5	44	Male	MF	8	1	Mod	P	A	Lung (solitary)	Resection	74	Adrenal	Resection	88	Lung (solitary)	Resection	107	137	NED

MF = mass-forming type; Well = well-differentiated adenocarcinoma; Mod = moderately differentiated adenocarcinoma; P = present; A = absent; Chemo = chemotherapy; NED = no evidence of disease; AWD = alive with disease; DFD = died from disease; DSM-TACE = degradable starch microsphere transhepatic arterial chemoembolization.
*Hepatectomy was performed after 4 courses of DSM-TACE.

tion for recurrent ICC. A second resection was performed for 5 patients with solitary recurrent cancer (liver, n = 4; lung, n = 1), and 2 of these 5 patients underwent a third resection for second recurrence (lung, n = 1; adrenal gland, n = 1). One patient (no. 5) underwent a fourth resection for recurrent lung metastasis and survived 137 months after the first resection (30 months after the fourth resection).

Median durations of survival for patients with recurrent ICC who received DSM-TACE and systemic chemotherapy were 14 months and 8 months, respectively.

Comments

This study analyzed 44 consecutive patients who received curative resection of ICC in a single institution, including 5 patients who underwent repeat resections for a solitary recurrence over 13 years.

The overall 5-year survival rate was 43% and 7 patients survived more than 5 years. Given the aggressive nature of ICC, extended resection is necessary for a curative outcome. Others have found that surgical results are more favorable with extended liver resection in patients with ICC.[4,8] In our series, 40 of the 44 patients underwent hemihepatectomy or extended hemihepatectomy, with favorable short-term outcomes and no hospital deaths. Extended hemihepatectomy therefore seems to represent a valid therapeutic option for ICC. Conversely, most previous reports state that survival data may be adversely affected by mortality rates of 1% to 7%.[4,7,8] Significant advances over recent decades in imaging modalities, surgical technique, anesthesia, and critical care medicine have greatly improved the safety of major hepatic surgery. The current study may thus more accurately reflect clinical outcomes to be expected from treatment in the era of advanced surgical techniques.

Many reports have described favorable prognostic factors after resection of ICC.[6,8,9,11] These include absence of tumor at resection margins, elevated serum CA19-9 levels, solitary lesion, absence of lymph node involvement, presence of well-differentiated adenocarcinoma, and absence of vascular invasion. In our study, multiple tumors were identified as an independent poor prognostic factor, showing a 0% survival rate after 5 years for such cases compared to 64% for patients with solitary ICC. Previous studies gave a dismal prognosis even after curative resection for patients with node-positive ICC. The 5-year survival rate for patients with node-positive ICC in this study was 24%. In the present study, some patients with lymph node metastasis lived for a long time, and lymph node metastasis was not identified by our analysis as a factor associated with poor prognosis. This is probably due to the limited number of patients. Nevertheless, no consensus has yet been reached regarding lymph node dissection, and there are several reports of dire outcomes in patients with node-positive ICC even after lymph node dissection.[12] Chou et al[13] reported that the survival rate with node-positive ICC was almost the

same as with noncurative resection even after lymph node dissection, while Inoue et al[14] reported similar results of lymph node dissection not prolonging survival. Conversely, others have reported 5-year survival rates of 23% to 34% after curative resection for node-positive ICC. The outcome of hepatectomy in patients with lymph node metastasis is poor; however, our study found no desperate need for hepatectomy in a case with regional lymph node involvement. Theoretically, adjuvant chemotherapy should be considered following resection and may prolong survival, particularly in patients with poor prognostic factors. However, no standard protocol exists to extend survival in patients with ICC and further studies are clearly needed.

Recurrence rates following curative resection remain high, with 50% to 80% recurrence reported even after curative resection.[9] In the present study of 25 patients with tumor recurrence (57%), a second resection was performed on 5 patients with solitary hepatic or pulmonary metastasis, with favorable results. The liver is the most frequent site of recurrence, followed by bone, peritoneal dissemination, and then lymph nodes.[8,12] No specific therapy has been recommended for recurrent ICC, but this study presented promise that repeated resection may improve overall survival. The efficacy of surgery for hepatic and pulmonary metastases of colon cancer is well documented, but the efficacy of repeated resection for recurrent ICC remains unclear, despite several small studies.[4,15,16] The present findings indicate that some patients with ICC have no more than a few resectable lesions, as is the case for hepatic and pulmonary metastasis of colon cancer. Indications for repeated resection were not conclusive due to the small number of patients in the present study. This is a common problem because ICC is a rare disease, and most previous studies involved only a few dozen cases from a single institution. Future analyses must comprise many more ICC patients across multiple institutions.

Conclusion

Prognosis after curative resection is poor in ICC patients with multiple nodules. In selected patients with solitary hepatic or pulmonary recurrence, repeated resection may offer long-term survival.

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Preoperative endoscopic pancreatic stenting: a novel prophylactic measure against pancreatic fistula after distal pancreatectomy

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Abstract

Background/Purpose. The prevention of pancreatic fistula is still a major problem in distal pancreatectomy (DP). We have recently adopted preoperative endoscopic pancreatic stenting with the aim of preventing the leakage of pancreatic juice from the resection plane of the remnant pancreas after DP. We reviewed ten patients who underwent this intervention.

Methods. One to 6 days before surgery, the patients underwent an endoscopic transpapillary pancreatic stent (7 Fr., 3 cm) placement. The perioperative short-term outcomes were assessed.

Results. Preoperative endoscopic pancreatic stenting was successfully performed in all ten patients. Two (20%) patients, both with intraductal papillary mucinous tumor, developed mild acute pancreatitis after the stent placement. None of the ten patients developed pancreatic fistula. The pancreatic stent was removed 8–28 days (mean, 11 days) postoperatively.

Conclusions. Preoperative endoscopic pancreatic stenting may be an effective prophylactic measure against pancreatic fistula development following DP.

Key words Pancreatic fistula · Distal pancreatectomy · Endoscopic pancreatic stenting

Introduction

Pancreatic fistula is a common complication of distal pancreatectomy (DP).[1] The incidence of pancreatic fistula after DP ranges from 5% to 60%.[2–15] Recent advances in medical and surgical care in DP cannot completely eliminate the possibility of pancreatic fistula development. Most pancreatic fistulas usually resolve spontaneously, albeit over a lengthy period, after conservative treatment including pancreatic rest (no oral intake with total parenteral nutrition or octreotide administration) and adequate drainage (drains placed during the initial operation or postoperatively via the percutaneous approach).[16–18] However, some patients with pancreatic fistula develop complications, such as intraabdominal abscess, sepsis, and lethal hemorrhage. For pancreatic fistulas refractory to conservative treatment, surgical intervention is indicated, but this is often technically difficult and associated with a high morbidity rate (13%).[19] Therefore, prophylactic strategies can be employed to decrease the incidence of pancreatic fistula after DP.[20]

We previously reported the case of a patient in whom preoperative endoscopic pancreatic stenting prevented pancreatic fistula development following local pancreatic resection,[21] and another case of a patient in whom endoscopic pancreatic stenting was effective for a pancreatic pseudocyst that developed after DP.[22] Taken together, these experiences suggested that preoperative endoscopic pancreatic stenting might be envisaged as a prophylactic measure against pancreatic fistula after DP. Thus, we have recently adopted this intervention with the aim of preventing the leakage of pancreatic juice from the resection plane of the remnant pancreas after DP.[23] This preoperative endoscopic intervention is simple and easy and appears to minimize the incidence of pancreatic fistula after DP.[24] In this article, we present the short-term outcomes of ten patients who underwent preoperative endoscopic pancreatic stenting for the prophylaxis of pancreatic fistula after DP.

Patients and methods

Ten patients underwent preoperative endoscopic pancreatic stenting for the prophylaxis of pancreatic fistula after DP. The patients were five men and five women with a mean age of 58 years (range, 27–80 years). One to 6 days (mean, 5 days) before surgery, the patients

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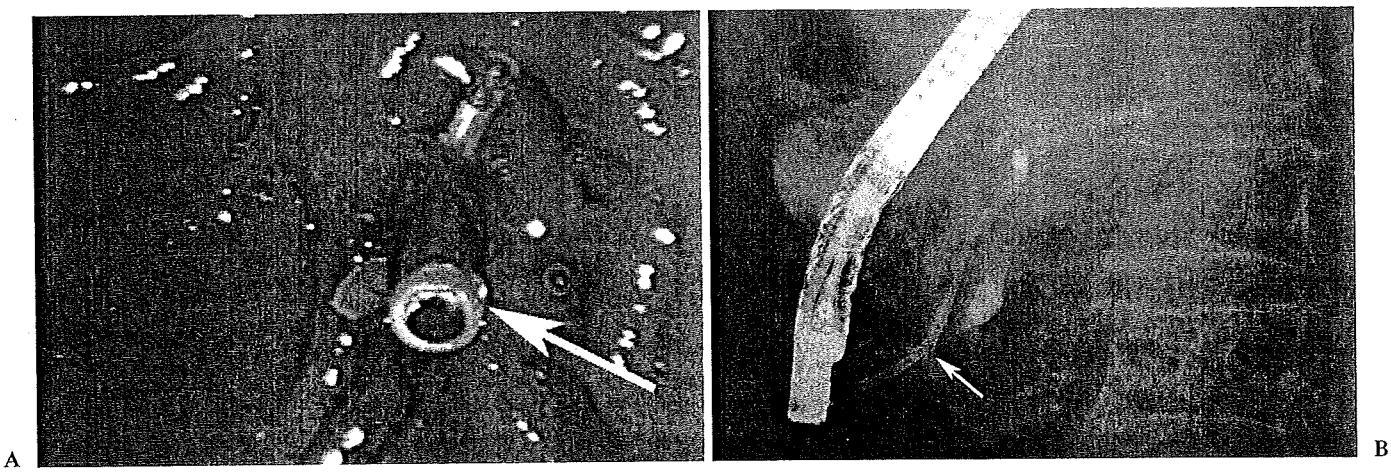

Fig. 1. A Duodenoscopic view of the papilla of Vater after pancreatic stent (*arrow*) placement. **B** Retrograde pancreatogram after endoscopic pancreatic stent (*arrow*) placement

underwent an endoscopic transpapillary pancreatic stent placement (Geenen, 7 Fr., 3 cm; Wilson-Cook Medical, Winston-Salem, NC, USA) (Fig. 1A,B). Before applying pancreatic stenting, informed consent was obtained from each patient.

The indications for DP included ductal adenocarcinoma (n = 4), intraductal papillary mucinous tumor (n = 3), ductal adenocarcinoma concomitant with intraductal papillary-mucinous tumor (n = 1), and mucinous cystic tumor (n = 2) of the pancreas. A concomitant splenectomy was carried out in all ten patients. During the pancreatic transection, the pancreatic parenchyma was divided using an electrocautery and/or a surgical scalpel (electrocautery, 4; scalpel, 2; both, 2), and the main pancreatic duct was ligated with a nonabsorbable suture. Pancreatic ducts ranged from 2 to 4 cm (mean, 3 cm) in diameter. The stump of the pancreas was not closed and was left open. Fibrin glue was not applied to the stump. One or two closed suction drains were placed in the splenic fossa adjacent to the pancreatic stump. Octreotide was not administered postoperatively.

The definition of pancreatic fistula was a drain output of any measurable volume of fluid on or after postoperative day 3 with an amylase content greater than three times the serum amylase level.[24] Other potential major complications after DP were categorized and defined as follows: intraabdominal infection (drainage of purulent but amylase-poor fluid); intraabdominal abscess (need for percutaneous or surgical drainage)[25]; and late postpancreatectomy hemorrhage (intraabdominal hemorrhage 24 h or more after operation).[26]

Results

Preoperative endoscopic pancreatic stenting was successfully performed in all ten patients. Endoscopic sphincterotomy or balloon dilatation was not required in any of the patients. The proximal tip of the stent was placed in the main pancreatic duct of the pancreatic head. Two patients, both with intraductal papillary mucinous tumor, developed mild acute pancreatitis the day after the stent placement. Both patients underwent endoscopic pancreatic stenting 6 days before the surgery. These patients were treated conservatively by pancreatic rest and protease inhibitor administration.

Postoperatively, none of ten patients developed pancreatic fistula. Other major complications including intraabdominal infection or abscess, and late postpancreatectomy hemorrhage did not occur in these ten patients. The pancreatic stent was removed 8–28 days postoperatively (mean, 10 days). No pancreatic stent occlusion was found by macroscopic observation of the stent lumen.

Discussion

The standard surgical technique for preventing the leakage of pancreatic juice from the resection plane of the remnant pancreas after DP is reportedly the ligation of the main pancreatic duct and closure by suturing of the resection plane of the remnant pancreas.[27] However, the resection plane has several stumps of the branch pancreatic ducts communicating with the main duct.[4] Konishi et al.[4] reported a mean number of two such branches, ranging from zero to five branches. These small ducts cannot all be identified and ligated during transection because they have small diameters. Furthermore, the closure of the resection plane of the remnant pancreas by suturing may not occlude such small ducts. These may lead to postoperative pancreatic fistula.[21] The prevention of pancreatic fistula is therefore still a major problem in DP.

Endoscopic pancreatic stenting has been used to treat pancreatic ductal stricture, pancreatolithiasis, pancreas divism, and pancreatic duct disruption (fistula and pseudocyst development secondary to acute or chronic pancreatitis, or pancreatic trauma).[28–30] However, only a few reports have described endoscopic pancreatic stenting for the treatment or prophylaxis of pancreatic fistula development after a pancreatic surgery.[21–23,29,31] In this article, we have presented the short-term outcomes of preoperative endoscopic pancreatic stenting as a prophylactic measure against pancreatic fistula after DP. The rationale for this new approach was based on previous results showing that (1) endoscopic treatment (pancreatic sphincterotomy and/or pancreatic stenting) is effective for treating pancreatic fistula[29,31] or pseudocyst[22] development after DP; and (2) preoperative endoscopic pancreatic stenting prevented pancreatic fistula after local pancreatic resection.[21] The results reported herein show that endoscopic pancreatic stenting is a minimally invasive technique that may prevent pancreatic fistula after DP, because 100% of patients who underwent this procedure actually did not develop pancreatic fistula. A pancreatic stent seems to allow the site of pancreatic leakage to seal via a pancreatic decompression effect by abolishing the pressure gradient between the pancreatic duct and the duodenum.[21,22,28,29] However, it could not be confirmed whether the pancreatic stent itself prevented pancreatic fistula. Further investigation of more cases is required to determine the value of this approach, and the randomization of patients to undergo either DP with versus without pancreatic stenting will be warranted to determine the actual efficacy of this approach.

The early complications associated with the stent placement are noteworthy. Two of four (50%) patients with intraductal papillary mucinous tumor developed acute mild pancreatitis after stent placement. This type of tumor secretes mucin; therefore, copious mucin fills the main and/or branch pancreatic ducts. Mucin can be associated with the stagnation of the pancreatic juice in a pancreatic stent. This stagnation of the pancreatic juice may result in the development of acute pancreatitis. To this end, preoperative stenting seems unsuitable for patients with this type of tumor, but intraoperative stenting (by intraoperative duodenoscopy or via duodenostomy) following removal of the tumor can be applicable to them.[21] We have previously experienced the case of a patient in whom intraoperative pancreatic stenting via duodenostomy prevented pancreatic fistula development after local pancreatic resection.[21]

Although no pancreatic stent occlusion was found in this series, a stent occlusion may directly induce postoperative pancreatic fistula development. If technically possible, large and short (7 Fr., 3 cm) stent placement, as in the present cases, seems desirable. Long-term stent placement may induce pancreatic ductal changes that resemble chronic pancreatitis.[32] Pancreatic fistula usually occurs 2–7 days after surgery.[2,16] Therefore, a stent should be retrieved within 2 weeks of surgery unless a pancreatic fistula has developed.[21]

In conclusion, preoperative endoscopic pancreatic stenting may be an effective prophylactic measure against pancreatic fistula after DP in selected patients.

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