Table 1 Characteristics of patients and comparison of results of percutaneous biopsy with those of endoscopic ultrasound-guided fine-needle aspiration

	Percutaneous biopsy Group A	EUS-FNA Group B	*P* value
Patients	46	75	
Site of puncture			
Pancreas	46	74	> 0.9999
Head/body/tail	12/32/2	34/31/9	0.0114
Sex (male/female)	25/21	39/36	> 0.8525
Age, yr			> 0.8466
≥ 65	28	48	
< 65	18	27	
Tumor diameter, mm (range)	44.8 (18-111)	25.5 (7-70)	
≥ 40	30	25	0.0007
< 40	16	50	
Passes (range)	2.26 (1-4)	2.85 (2-5)	< 0.0001
Adequate specimens obtained[1] *n* (%)			
Cytology	42 (91.3)	75 (100)	0.0192
Histology	41 (89.1)	65 (86.7)	0.7812
Positivity for cancer *n* (%)			
Cytology	33 (78.6)	72 (94.6)	0.0079
Histology	33 (80.5)	51 (78.4)	> 0.9999
Total *n* (%)	43 (93.5)	73 (97.3)	0.3672
Complications *n* (%)	2 (4.3) Fever[1] Bleeding[1]	1 (1.3) Peritonitis[1]	> 0.5567
Time from puncture to definitive diagnosis			
Cytology, d (range)	4.05 (0-8)	1.65 (0-5)	< 0.0001
Histology, d (range)	3.95 (2-7)	3.18 (2-10)	0.7066

[1]An on-site pathologist was available for endoscopic ultrasound-guided fine-needle aspiration (EUS-FNA) but not for ultrasonography-guided-FNA.

RESULTS

US-FNA was performed in 48 patients from February 2006 until October 2009. Two cases (renal cell carcinoma and malignant lymphoma) were excluded from the analysis of US-FNA because the patients did not have primary PC. EUS-FNA was attempted in 125 cases and was successfully performed in 123 cases from November 2009 until September 2011. Among these, 48 patients did not meet the selection criteria (lymph node metastasis, 34 cases; other pancreatic tumor, 10 cases; other abdominal tumor, three cases, and mediastinum tumor, one case). EUS-FNA could not be performed in two patients because of difficulty of visualization due to total gastrectomy in one case, and impossibility of ensuring the puncture route in the other. Thus, 46 patients who underwent US-FNA (Group A) and 75 who underwent EUS-FNA (Group B) were eligible for analysis.

Table 1 shows the characteristics of the study subjects. The distribution of the target tumor in the pancreas differed significantly between the two groups, with the tumor location more frequent in the pancreatic head/tail than in the pancreatic body in Group B. The maximum diameter of the target tumor ranged from 18 to 111 mm (median, 44.8 mm) in Group A and from 7 to 70 mm (median, 25.5 mm) in Group B. A significantly larger number of target tumors were < 40 mm in Group B than in Group A ($P = 0.0007$).

Table 1 shows a comparison of the results of percutaneous biopsy with those of EUS-FNA. Adequate cytological and histological specimens were obtained in 42 (91.3%) and 41 (89.1%) Group A patients ($n = 46$), respectively, and in 75 (100%) and 65 (86.7%) Group B patients ($n = 75$).

Results of cytology indicated the presence of cancer cells in 33 Group A patients (78.6%) and in 72 Group B patients (94.6%). Histological studies showed cancer tissue in 33 (80.5%) and 51 (78.4%) patients in Group A and Group B, respectively. In total, a cancer diagnosis was made in 43 Group A (93.5%) and 73 Group B (97.3%) patients by cytology and/or histology. These 116 patients were diagnosed with pancreatic adenocarcinoma by cytology/histology as well as by imaging and their subsequent clinical course. The final diagnosis of PC in the remaining five patients for whom there was no cytological or histological proof was confirmed by the clinical course until April 2012. The positive cytology/histology rate did not differ between the two groups.

Total puncture procedures per patient varied from one to five, with a median of 3. The frequency of multiple punctures, that is, > 2, was significantly higher in Group B than in Group A. Time from the day of puncture until the day of the final cytological diagnosis varied from 0 to 8 d (median, 4.1 d) in Group A and from 0 to 5 d (median, 1.7 d) in Group B. The period was significantly shorter in Group B than in Group A. The time from the day of puncture until the day of the final histological diagnosis varied from 2 to 7 d (median, 4.0 d) in Group A and 2 to 10 d (median, 3.2 d) in Group B, with no significant difference between the two groups.

Severe adverse events occurred in two Group A patients (4.3%) and in one Group B patient (1.3%). In Group A, one patient developed a high fever, which required hospitalization but resolved with only symptomatic treatment. The other Group A patient experienced upper gastrointestinal bleeding, which was confirmed by endoscopy to be related to the needle biopsy. This patient was treated by blood transfusion and antiulcer medication and was hospitalized for 1 wk without surgical intervention. The adverse event in Group B was an abdominal abscess that required surgical drainage. The patient experienced continuous abdominal pain one night after EUS-FNA, and dynamic CT demonstrated an abscess in front of the pancreatic body tumor, which was clearly related to the EUS-FNA puncture. Fortunately, she recovered after surgery and antibiotic therapy and could receive chemotherapy thereafter. There was no cancer seeding event up to 6 mo from the time of puncture in any patient in either group.

DISCUSSION

The aim of the current study was to investigate the results of two different approaches to obtain pancreatic biopsy specimens, which are a percutaneous approach and EUS-FNA, because this issue has seldom been ad-

dressed[12]. Our results confirmed the usefulness of EUS-FNA, especially with regard to cytology. The National Comprehensive Cancer Network Guidelines (2012) require that cytological or histological confirmation is needed for the diagnosis of unresectable pancreatic carcinoma[13]. In patients with stage Ⅳ PC, a biopsy of the metastatic lesion is preferred for proof of cancer. However, in those with stage Ⅲ PC and some patients with stage Ⅳ PC in whom it is difficult to access metastatic sites for biopsy procedures, the primary tumor of the pancreas must be targeted to obtain proof of cancer. Pancreatic juice cytology was developed in the early 1980s and is still being performed; however, cancer cells cannot easily be observed by collection of pancreatic juice[1,2,14]. Percutaneous needle biopsy was developed with the expectation of a more definitive method to obtain proof of cancer from the primary pancreatic tumor[3,15,16]. Our institute then used percutaneous needle biopsy under extracorporeal US guidance as the standard for histological confirmation of the pancreatic primary tumor. Recently, EUS-FNA was introduced and was used mainly in high-volume cancer centers in Japan[17-22]. As a result of the risk of cancer seeding as well as other risks with percutaneous biopsy, we adopted EUS-FNA beginning in November 2009 in place of percutaneous biopsy. We expected that EUS-FNA would have advantages over a percutaneous procedure with regard to efficacy in confirmation of cancer and avoiding adverse reactions before administering chemotherapy to patients with PC.

Our results demonstrated that EUS-FNA is effective and feasible for obtaining proof of cancer in candidates for PC chemotherapy. In fact, EUS-FNA might have merits with regard to obtaining specimens from small tumors or tumors in the pancreatic tail, for which performance of percutaneous biopsy is difficult[2,23-27]. In this study, the location of the target tumor was most frequent at the body of the pancreas in Group A. In addition, the target tumors were larger in Group A than in Group B. These findings suggest that patients might have been excluded from Group A in which difficulty could be expected in making a puncture because the tumor was either small or difficult to delineate. In these cases, endoscopic retrograde cholangiopancreatography or liver biopsy might have been performed to obtain confirmation of malignancy, if possible.

Horwhat *et al*[12] have performed a randomized controlled trial of EUS-FNA and percutaneous biopsy of the pancreas (US- and CT-guided) in 2006. Although there was no statistically significant difference in accuracy between the two methods, the results showed that EUS-FNA had the advantage in the diagnosis of pancreatic malignancy. In our study, the diameters of the target tumors in the EUS-FNA group (Group B) were smaller than those in the US-FNA group (Group A) and the deviation of distribution around the puncture site was smaller in the EUS-FNA than the US-FNA group. Our results indicated high performance through the use of EUS-FNA and are not inconsistent with those of Horwhat *et al*[12]. In the present study, there was no analysis of accuracy in the two groups, because our institution is an oncology hospital and we rarely perform biopsies of benign cases.

The benefits of EUS-FNA might be maximized to make a pathological diagnosis in patients with an abdominal tumor of an uncertain type. The definite merit of our EUS-FNA procedure was thought to be rapid cytological results, but perhaps success in this regard was mainly due to the contribution of an on-site cytotechnologist and not to the EUS-FNA procedure itself. Iglesias-Garcia *et al*[28] have claimed that on-site cytological evaluation improves the diagnostic yield of EUS-guided FNA for the cytological diagnosis of solid pancreatic masses. Savoy *et al*[29] have pointed out that even trained endosonographers have variable and, in some cases, inferior abilities in interpreting on-site cytology in comparison with cytotechnologists. In the present study, we had adequate specimens for all cases in the EUS-FNA group. This is natural because we continued the examination until we obtained a sufficient quantity of specimens that were checked by the on-site cytotechnologist. On the contrary, there was no difference in the rate of adequate specimens obtained for histological examination between the EUS-FNA and US-FNA groups, because the collected tissue was checked by the examiner's naked eye in both groups. The presence of an on-site cytotechnologist to accompany EUS-FNA is considered to be necessary, at least, in high-volume centers.

In the present study, the positivity rate for malignancy was higher for EUS-FNA cytology than for histology. Supporting the current results, another study has shown that the positivity rate for malignancy in EUS-FNA cytology of the pancreas was higher than that in histology[30].

As previously reported, EUS-needle core biopsy is useful for histological and cytological diagnosis in terms of sample volume[31]. In addition, the combined results of EUS-FNA cytology and EUS-needle core biopsy have been reported to improve diagnosis[32-34]. However, to confirm the malignancy, EUS-FNA cytology is more useful than EUS-needle core biopsy[35]. This result is similar to the results of our study, indicating that cytology might be more useful than histology for the diagnosis of malignancy.

In the current study, there was no cancer seeding in any patient in either group. As previously reported, there were rare cases of seeding among patients who underwent US-guided FNA[36]. With regard to the puncture route, we suggest that there is less possibility of seeding in patients who undergo EUS-FNA than in patients who undergo US-FNA, although some recent studies have shown the possibility of seeding in patients who undergo EUS-FNA[37-39]. We did inform patients who were scheduled to undergo EUS-FNA about the possibility of this complication.

The limitations of our study included its retrospective nature. Furthermore, there were no cases of benign pancreatic conditions to enable an evaluation of US and EUS-FNA for accurate differentiation between malignant

and benign diseases.

In conclusion, EUS-FNA, as well as percutaneous needle aspiration, is an effective modality to obtain cytopathological confirmation in patients with advanced PC. EUS-FNA cytology was able to detect malignancy at a high rate. We believe that EUS-FNA has advantages for smaller tumors located deeply and for tumors in which the diagnosis is uncertain by various other imaging modalities.

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COMMENTS

Background

Ultrasonography-guided fine-needle aspiration (US-FNA) biopsy or computed tomography (CT)-guided FNA biopsy was used for histological/cytological diagnosis of pancreatic cancer (PC). US-FNA is limited to masses in the pancreatic tail. CT-guided FNA is time-consuming and limited by a substantial false-negative rate. There have been concerns about percutaneous cancer seeding and difficulty in puncturing for small tumors. Endoscopic ultrasound (EUS)-guided FNA has been developed as a more feasible method of obtaining definitive specimens for the diagnosis of PC. Studies on the results of the two different approaches to obtain pancreatic biopsy specimens, which are the percutaneous approach and EUS-FNA, have rarely been conducted.

Research frontiers

The benefits of EUS-FNA might be maximized to be able to make a pathological diagnosis in patients with an abdominal tumor of an uncertain type.

Innovations and breakthroughs

EUS-FNA is effective and feasible for obtaining proof of cancer in PC chemotherapy candidates. In fact, EUS-FNA might have advantages with regard to obtaining specimens from small tumors or tumors in the pancreatic tail, for which performance of percutaneous biopsy is difficult.

Applications

The results suggest that EUS-FNA is the best method of obtaining cytological samples for diagnosis of unresectable PC. This method can be used for other types of cancer.

Terminology

On-site cytotechnologist: An on-site cytotechnologist should attend the puncture examination to confirm quickly the existence of atypical cells. The information of the cytotechnologist is more appropriate than that of the endoscopist.

Peer review

This is a good descriptive study in which EUS-FNA is a feasible and safe technique to acquire pancreatic specimens. The results are interesting in that the advantages of EUS-FNA over the percutaneous procedure are time between examination and diagnosis, the possibility of puncture of small tumors, and tumors in the tail of the pancreas.

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RESEARCH ARTICLE Open Access

Association between variations in the fat mass and obesity-associated gene and pancreatic cancer risk: a case–control study in Japan

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Abstract

Background: It is clear that genetic variations in the fat mass and obesity-associated (FTO) gene affect body mass index and the risk of obesity. Given the mounting evidence showing a positive association between obesity and pancreatic cancer, this study aimed to investigate the relation between variants in the FTO gene, obesity and pancreatic cancer risk.

Methods: We conducted a hospital-based case–control study in Japan to investigate whether genetic variations in the FTO gene were associated with pancreatic cancer risk. We genotyped rs9939609 in the FTO gene of 360 cases and 400 control subjects. An unconditional logistic model was used to estimate the odds ratio (OR) and 95% confidence interval (CI) for the association between rs9939609 and pancreatic cancer risk.

Results: The minor allele frequency of rs9939609 was 0.18 among control subjects. BMI was not associated with pancreatic cancer risk. Compared with individuals with the common homozygous TT genotype, those with the heterozygous TA genotype and the minor homozygous AA genotype had a 48% (OR=1.48; 95%CI: 1.07–2.04), and 66% increased risk (OR=1.66; 95%CI: 0.70–3.90), respectively, of pancreatic cancer after adjustment for sex, age, body mass index, cigarette smoking and history of diabetes. The per-allele OR was 1.41 (95%CI: 1.07–1.85). There were no significant interactions between TA/AA genotypes and body mass index.

Conclusions: Our findings indicate that rs9939609 in the FTO gene is associated with pancreatic cancer risk in Japanese subjects, possibly through a mechanism that is independent of obesity. Further investigation and replication of our results is required in other independent samples.

Keywords: The fat mass and obesity-associated gene, Pancreatic cancer, rs9939609, Case–control study

Background

In 2010, approximately 28,000 Japanese subjects died from pancreatic cancer, making it the fifth leading cause of cancer deaths in Japan [1]. Despite extensive research efforts, the etiology of pancreatic cancer remains poorly understood. Cigarette smoking and long-standing type II diabetes are two well-established risk factors, based on consistent findings from epidemiologic studies [2,3]. In addition, being overweight and obese have been implicated in the development of pancreatic cancer [4], with statistically significant, positive associations observed in large cohort studies conducted in Western countries [5-7], and corroborated in at least four meta-analyses [8-11] and three pooled analyses [12-14]. The positive association between body mass index (BMI) and pancreatic cancer, however, has not been clearly observed in Asian populations. To date, four cohort studies have examined the association between BMI and pancreatic cancer in Asians, but the results have been inconsistent and inconclusive [15-18].

Recently, genome-wide association (GWA) studies have identified at least 30 loci that affect BMI and the

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risk of obesity [19]. Among these loci, the fat mass and obesity-associated (FTO) gene, which was first identified in a GWA study of diabetes in 2007 [20], has the strongest influence on BMI and obesity. Rs9939609, located in the first intron of the FTO gene, was found to be associated with both BMI and type II diabetes in subsequent GWA studies in diverse populations [21-23]. The association of rs9939609 with various traits, including hip circumference, energy intake and total mortality has also been studied [24-26]. In addition, rs9939609 genotypes have been linked with the risk of prostate, breast and endometrial cancers [27-29]. The association between genetic variations in the FTO gene and the risk of pancreatic cancer, however, is not clear. Of the three studies that examined this association, only one case–control study, conducted at the MD Anderson Cancer Center in the United States, reported that the minor A allele of FTO, rs9939609, was associated with an increased risk of pancreatic cancer among overweight subjects [30]. Another two studies examined rs8050136 of the FTO gene, with one study reporting a positive association [31], and the other no association [32].

Given the mounting evidence showing a positive association between obesity and pancreatic cancer, we hypothesized that variants in the FTO gene may be associated with pancreatic cancer risk through effects on obesity or other mechanisms. In a search of the literature for obesity-related genetic variants, we found that FTO rs9939609 was the most widely studied single nucleotide polymorphism (SNP), and has been found to exert strong effects on BMI, as well as diabetes. Furthermore, it showed strong linkage disequilibrium with other SNPs in the FTO gene, such as rs8050135 and rs17817449 [22]. We therefore investigated the association between FTO rs9939609 and pancreatic cancer risk in a case–control study in Japan.

Methods

Study subjects

Our study is an ongoing hospital-based case–control study focusing on the role of genetic polymorphisms and gene-environment interaction in pancreatic cancer. For the present analysis, eligible cases were patients aged older than 20 years, who were newly diagnosed with pancreatic cancer in five hospitals located in central, north and Tokyo metropolitan areas from April 1, 2010 through May 15, 2012. The diagnosis of pancreatic cancer was based on imaging modalities or pathologic reports. The response rate among cases was 85% (441/516) as of July 1, 2012. Almost all of the cases were approached within a week after the diagnosis of pancreatic cancer, and very few cases died before they were invited to participate in our study. During the same period, we recruited control subjects with no diagnosis of cancer from inpatients and outpatients from the participating hospitals where the cases were enrolled, as well as relatives of inpatients, and individuals undergoing a medical checkup in one of the participating hospitals. Control subjects were eligible if they were more than 20 years old and had no prior cancer diagnoses. Recruitment of controls was accomplished by approaching eligible participants in the hospitals who satisfied the study requirements, and the response rate was 98% (525/534). Control subjects had a variety of diseases, such as anemia, gastric ulcer, and irritable bowel syndrome. Control subjects were matched with case patients according to sex and age (within 10-year categories). As a result, data from 360 case patients and 400 control subjects were included in the present analysis.

All subjects provided written, informed consent. This study was approved by the ethical board of Aichi Medical University (Nagakute, Japan), the Institutional Review Board (IRB) of Cancer Institute Hospital (Tokyo, Japan), the IRB of Kanagawa Cancer Center Hospital (Kanagawa, Japan), the IRB of Tokyo Metropolitan Komagome Hospital (Tokyo, Japan), and the IRB of Sapporo Medical University (Sapporo, Japan).

Data collection

Study subjects were asked to fill out a self-administered questionnaire including information on demographic characteristics, medical history, and lifestyle factors, such as cigarette smoking, alcohol consumption and dietary intake. For body weight, data on usual weight over the year prior to study entry as well as weight at age 20 were reported by the study participants. For current or former smokers, we collected detailed data on smoking exposure, including smoking status (never, former, or current smokers), average number of cigarettes smoked per day, age at starting and quitting, and duration of smoking. For subjects with type II diabetes, we recorded the age at diagnosis. In addition to the questionnaire survey, all consenting participants provided a 7-mL venous blood sample. Genomic DNA was extracted from peripheral lymphocytes at SRL Hachioji Laboratory and then stored at -30°C at the Department of Public Health, Aichi Medical University.

Genotyping assays

Genotyping was performed using the Taqman SNP Genotyping Assay (Applied Biosystems, Foster City, CA, USA) at the laboratory of Aichi Cancer Center Research Institute, Nagoya, Japan. Laboratory staff were blinded to case or control status. Four quality control samples were included in each assay, and the successful genotyping rate was 100%.

Statistical analysis

Case–control differences in selected demographic characteristics and risk factors were evaluated using t tests (for continuous variables) and Chi-square tests (for categorical variables). A chi-square test was used to test genotype frequencies in control subjects for Hardy-Weinberg equilibrium (HWE) by comparing observed genotype frequencies with those expected under HWE. A co-dominant genomic model was assumed for SNP effects. Unconditional logistic regression methods were used to estimate odds ratios (ORs) and 95% confidence intervals (CIs) for the association between rs9939609 genotypes and pancreatic cancer risk. Homozygous carriers of the common FTO rs9939609 T allele served as the reference group. All analyses were adjusted for age (continuous), sex (male or female), BMI (<20, 20–22.4, 22.5-24.9, ≥25.0), history of diabetes (yes or no), and cigarette smoking (current, former, never smokers). ORs were also estimated for the variant allele on the basis of a log-additive model. The interaction of genotype-BMI and genotype-history of diabetes with respect to pancreatic cancer risk was assessed using the likelihood ratio test. Because recent-onset diabetes may result from pancreatic cancer, we performed an analysis excluding cases who had onset of diabetes within 2 years prior to the diagnosis of pancreatic cancer.

All P-values were two-sided, with $P<0.05$ indicating statistical significance. All statistical analyses were performed using SAS 9.2 (SAS Institute, Inc., Cary, NC, USA).

Results

The distribution of genotypes among control subjects did not deviate from the Hardy-Weinberg equilibrium (P=0.94). The minor allele frequency (MAF) was 0.18 among control subjects. Table 1 summarizes the characteristics of cases and controls. Both groups had a similar distribution of sex and 10-year age groups. The mean age was 65.1±8.1 years for cases, and 58.5±9.1 years for controls. Cases were more likely to be current smokers and have a history of diabetes compared with controls. Current smokers had an approximately 2.9-fold increased risk of pancreatic cancer compared with nonsmokers, after adjustment for age, sex, BMI, and history of diabetes (OR=2.86; 95%CI: 1.79-4.57). Individuals who had a BMI of 30 or more had a 1.21-fold increased risk, but the association was not statistically significant. Similar results were obtained in an additional analysis in which BMI at age 20 was used (data not shown). Risk of pancreatic cancer was significantly increased among subjects reporting a history of diabetes (OR=2.94; 95%CI: 1.90-4.57). The significant, positive association remained after excluding pancreatic cancer cases with recent-onset diabetes (OR=1.92; 95%CI: 1.20–3.08). Among control subjects, the mean BMI was 22.7±3.1 for the TT genotype, 23.2±3.3 for the TA genotype, and 21.1±2.9 for the AA genotype.

Table 1 Association between variations in the fat mass and obesity-associated gene and pancreatic cancer risk: a case–control study in Japan

Characteristics	Case patients (N=360)	Control subjects (N=400)	OR (95% CI)
Age group			Matching factor
<50	12 (3.3)	19 (4.8)	
50-59	44 (12.2)	79 (19.8)	
60-69	141 (39.2)	170 (42.5)	
70-79	138 (38.3)	115 (28.8)	
≥80	25 (6.9)	17 (4.3)	
Sex			Matching factor
Female	215 (59.7)	226 (56.5)	
Male	145 (40.3)	174 (43.5)	
Body mass index (kg/m^2)			
<25	278 (77.2)	312 (78.0)	1.00
25.0-29.9	64 (17.8)	75 (18.7)	0.96 (0.65-1.43)
≥30	16 (4.4)	12 (3.0)	1.21 (0.53-2.77)
Unknown	2 (0.6)	1(0.3)	-
Smoking status			
Non-smokers	145 (40.2)	202 (50.5)	1.00
Former smokers	119 (33.1)	140 (35.0)	1.23 (0.82-1.85)
Current Smokers	96 (26.7)	58 (14.5)	2.86 (1.79-4.57)
History of diabetes			
No	269 (74.7)	362 (90.5)	1.00
Yes	87 (24.2)	35 (8.7)	2.94 (1.90-4.57)
Unknown	4 (1.1)	3 (0.8)	-

OR: odds ratio; CI: confidence interval.
OR was adjusted for sex, age, smoking status and history of diabetes.

Table 2 shows the association between variants in the FTO gene (rs9939609) and pancreatic cancer risk. Compared with individuals with the TT genotype, the multivariate adjusted OR for developing pancreatic cancer was 1.48 (95%CI: 1.07–2.04) among those with the TA

Table 2 Association between the FTO rs9939609 and pancreatic cancer risk

FTO rs9939609	Cases	Control subjects	Age- and sex-adjusted OR	Multivariable-adjusted OR
TT	213	271	1.00	1.00
TA	133	116	1.49 (1.09-2.03)	1.48 (1.07-2.04)
AA	14	13	1.49 (0.67-3.29)	1.66 (0.70-3.90)

OR: odds ratio ; CI: confidence interval.
Multivariable adjusted OR: adjusted for age, sex, body mass index, cigarette smoking and history of diabetes.

genotype, and 1.66 (95%CI: 0.70–3.90) among those with the AA genotype. Under the dominant model, the OR was 1.49 (95%CI: 1.09–2.05) among carriers of the TA/AA genotype. Under the log-additive model, each additional copy of minor allele A was associated with a 1.4-fold increased risk of pancreatic cancer (OR=1.41, 95% CI: 1.07–1.85).

We found no significant interaction between FTO rs9939609 and BMI (Table 3). Individuals with both a TA/AA genotype and a history of diabetes had a 3.7-fold increased risk of pancreatic cancer compared with those with a TT genotype and no history of diabetes (Table 4), but a test for the interaction was not statistically significant.

Table 4 Joint associations of the FTO rs 9939609 and history of diabetes with respect to pancreatic cancer risk

Genotype	History of diabetes	Cases/ control subjects	Age- and sex-adjusted OR	Multivariable-adjusted OR
TT	No	163/243	1.00	1.00
TA/AA	No	106/119	1.38 (0.99-1.93)	1.41 (1.00-1.98)
TT	Yes	34/26	1.76 (1.01-3.07)	1.70 (0.96-3.00)
TA/AA	Yes	24/8	4.03 (1.75-9.24)	3.70 (1.59-8.63)
				P for interaction=0.28

Cases were excluded if the onset of diabetes was within 2 years prior to the diagnosis of pancreatic cancer.
Multivariable OR: adjusted for age, sex, body mass index, and cigarette smoking.

Discussion

This was a hospital-based case–control study in Japan to investigate whether genetic variations in the FTO gene were associated with pancreatic cancer risk. The main findings of our study were: 1) individuals with the FTO rs9939609 TA genotype had a significant 1.5-fold increased risk of pancreatic cancer compared with those with the TT genotype; and 2) a combination of the FTO rs9939609 TA/AA genotype and a history of diabetes significantly increased the pancreatic cancer risk, with an OR of 3.70 (95%CI: 1.59–8.63).

We found that obesity, defined as a BMI of 30 or more, was associated with 1.2-fold increased risk of pancreatic cancer, but this association was not statistically significant. In contrast to evidence of a positive association between obesity and pancreatic cancer in Western countries, available data on the role of obesity in pancreatic cancer in Japanese are inconclusive. There have been no prospective studies that have observed a clear, dose–response relation between baseline BMI and pancreatic cancer risk in the Japanese population [15,16]. Given that less than 5% of the subjects were obese in this study, it might be difficult to observe significant associations. The small percentage of obese people may be the main reason for the inconclusive results on BMI and pancreatic cancer in Asians, including Japanese [15-18]. In addition, differences in body fat distribution, in genetic predisposition to obesity and in lifestyle factors between Caucasians and Asians may contribute to the inconsistent results on BMI and pancreatic cancer risk in Asian populations [33,34].

Table 3 Joint associations of the FTO rs9939609 and BMI with respect to pancreatic cancer risk

Genotype	BMI	Cases/control subjects	Age- and sex-adjusted OR	Multivariable-adjusted OR
TT	<25	166/220	1.00	1.00
TA/AA	<25	112/92	1.69 (1.20-2.40)	1.68 (1.18-2.41)
TT	≥25	45/51	1.29 (0.81-2.04)	1.20 (0.75-1.94)
TA/AA	≥25	35/36	1.35 (0.81-2.25)	1.21 (0.71-2.07)
				P for interaction=0.29

Multivariable OR: adjusted for age, sex, cigarette smoking and history of diabetes.

Because of the positive association between obesity and pancreatic cancer in Caucasians and the plausible mechanisms, several research groups have hypothesized that variants in obesity-related genes might be associated with pancreatic cancer risk. The association between rs9939609 in the FTO gene was reported in one previous hospital–based case–control study conducted at the MD Anderson Cancer Center, Texas, USA [30]. Of the 15 obesity- and diabetes-associated genotypes in the FTO gene, rs9939609 was found to be positively associated with pancreatic cancer risk in persons who were overweight, whereas no increased risk was observed in persons who had a BMI of less than 25 kg/m^2 [30]. In contrast, our study showed a significant, positive association between rs9939609 TA/AA genotype and pancreatic cancer risk in individuals with a BMI of less than 25 kg/m^2. We consider that the difference in minor allele frequency (MAF) may be the main reason, given the fact that the MAF was 18% in our study, much lower than the 38% in the MD Anderson Cancer Center case–control study. The possible differences in selection of cases and controls, patterns of linkage disequilibrium and effects of gene-gene interactions may also account for the inconsistent findings. In addition to rs9939609, rs8050136 in the FTO gene was found to be associated with pancreatic cancer risk in individuals of European ancestry [31]; however, no association was noted in another case–control study [32].

In our study, FTO rs9939609 genotypes were associated with pancreatic cancer risk. However, the mean BMI did not differ among rs9939609 genotypes for control subjects, and no significant interaction was observed between rs9939609 TA/AA genotypes and BMI with

respect to pancreatic cancer risk. It is possible that the positive association observed between rs9939609 genotypes and pancreatic cancer risk may be driven by a mechanism other than adiposity. Diabetes, a well-established risk factor for pancreatic cancer, is a possible candidate. There is evidence suggesting that Asian people are more susceptible to insulin resistance at a lesser degree of obesity than Caucasians [33,34]. Besides its close association with adiposity, FTO has been shown to be associated with susceptibility to type II diabetes [21,22]. We found that individuals with a TA/AA genotype and a history of diabetes were at a 3.7-fold increased risk of pancreatic cancer. However, a test for the interaction was not statistically significant. Another possibility is that FTO is just a proxy of as yet unidentified causal variants, and it is those variants that exert their effects on rs9939609 and influence pancreatic cancer risk. Given that the function of the FTO gene is largely unknown, further studies are needed to comprehensively evaluate multiple SNPs in the FTO gene and elucidate the mechanisms by which FTO rs9939609 influences pancreatic cancer risk.

Our study has several limitations. First, it is well-known that two significant issues, namely selection bias and recall bias, plague case–control studies. Our results might have been biased if hospital controls did not represent the same population from which the cases were derived. However, the allele frequencies observed among control subjects in our study were similar to those reported in the studies of Asian populations [22]. In particular, the MAF of FTO rs9939609 was 18% in our control subjects, which is very close to that reported from a sample of 100 Japanese included in the HapMap project. Moreover, the risk estimates for current smokers and individuals with a history of diabetes were comparable to those estimated from cohort or population-based case–control studies [2,3], providing indirect evidence that selection bias might not be a serious concern in our study. Second, as for recall bias, while the analysis of the association between pancreatic cancer and BMI based on self-reported weight and height might be affected by recall bias, the association with the obesity-related genotype was not. Third, although our study included a relatively large sample size compared with previous studies conducted in Japan, the sample size may not have been large enough to detect significant gene-environment interactions in subgroups. Finally, it is possible that the results could represent a chance association and therefore replication in other independent samples is required. Despite these limitations, there are several advantages of the hospital-based design adopted in our study, including rapid case ascertainment, a high response rate from both cases and controls, and high quality genotyping.

Conclusion

Our findings indicate that rs9939609 in the FTO gene is associated with pancreatic cancer risk in Japanese subjects, possibly through a mechanism that is independent of obesity. Because of the limited statistical power, our results need replication in other independent samples. The fast-increasing prevalence of overweight/obesity and type II diabetes in Asians provides a good opportunity to further address this association and its underlying mechanisms.

Competing interests

The authors declare no conflict of interest.

Authors' contribution

SK supervised the study, SK, YL, KY designed the study, YL drafted the manuscript and conducted the statistical analysis. JU and KM performed genotyping and SNP data analysis. HI, MU, NE, HN, MM participated in data collection. All authors read and approved the final manuscript.

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ORIGINAL ARTICLE

Randomized phase II study of gemcitabine and S-1 combination versus gemcitabine alone in the treatment of unresectable advanced pancreatic cancer (Japan Clinical Cancer Research Organization PC-01 study)

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Abstract
Purpose To evaluate the efficacy and safety of the combination of gemcitabine (GEM) and S-1 (GS) in comparison to GEM alone (G) for unresectable pancreatic cancer.
Methods In this multicenter randomized phase II study, we randomly assigned unresectable pancreatic cancer patients to either the GS group or the G group. The GS group regimen consists of intravenous 1,000 mg/m^2 GEM during 30 min on days 1 and 8, combined with 80 mg/m^2 oral S-1 twice daily on days 1–14, repeated every 3 weeks. On the other hand, the G group regimen consists of intravenous 1,000 mg/m^2 GEM on days 1, 8, and 15, repeated every 4 weeks. The primary endpoint was objective response rate (ORR). Secondary end points included treatment toxicity, clinical response benefit, progression-free survival (PFS), and overall survival.

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Results We registered 117 patients from 16 institutions between June 2007 and August, 2010. The ORR of the GS group was 28.3%, whereas that of the G group was 6.8%. This difference was statistically significant ($P = 0.005$). The disease control rate was 64.2% in the GS group and 44.1% in the G group. Median PFS was 6.15 months in the GS group and 3.78 month in the G group. This was also statistically significant ($P = 0.0007$). Moreover, the median overall survival (OS) of the GS group was significantly longer than that of the G group (13.7 months vs. 8.0 months; $P = 0.035$). The major grade 3–4 adverse events were neutropenia (54.7% in the GS group and 22.0% in the G group), thrombocytopenia (15.1% in the GS group and 5.1% in the G group), and skin rash (9.4% in the GS group).
Conclusions The GS group showed stronger anticancer activity than the G group, suggesting the need for a large randomized phase III study to confirm GS advantages in a specific subset.

Keywords Unresectable pancreatic cancer · Chemotherapy · Gemcitabine · S-1 · Gemcitabine+S-1

Introduction

Pancreatic cancer (PC) currently is the fifth leading cause of cancer-related mortality in Japan, with an estimated 25,960 deaths attributable to the disease in 2010 [1]. Although surgical complete removal of the tumor is the only chance of cure, almost all PC patients are diagnosed at an advanced unresectable stage, despite recent improvements in diagnostic techniques. Moreover, since PC recurs in about 20% of patients even after surgical resection, development of effective chemotherapy is essential to improve the prognosis of this disease.

Gemcitabine (Gem) is widely used as a standard systemic chemotherapeutic agent for advanced PC [2]. Although some combination therapies including Gem have shown survival benefit, these are not considered as standard regimens [3, 4]. S-1 is a fourth generation oral fluoropyrimidine, which contains tegafur/gimeracil/oteracil potassium at a molar ratio of 1.0:0.4:1.0. The efficacy of S-1 has already been shown in a variety of solid tumors, particularly gastric cancer [5, 6]. A phase II trial of S-1 alone for PC metastatic to other organ has shown a response rate of 37.5% and a median survival of 9.2 months [7, 8]. Moreover, non-randomized phase II trials of a combination of Gem and S-1 (GS) therapy have demonstrated excellent results as to ORR of 44–48% and median survival of 10–12 months [9–13].

The current study (PC-01) was a randomized phase II trial to clarify the effectiveness of GS, prior to an anticipated phase III trial comparing GS with Gem alone, because there are many chemotherapy regimens that did not prove survival benefit despite the fact that one-arm phase II studies showed extremely promising results. Consequently, we, investigators of the Japan Clinical Cancer Research Organization (JACCRO), considered the current study (PC-01) could accurately elucidate the true activity of GS, because selection bias frequently seen in one-arm trials may be minimized by prospective randomization studies.

Patients and methods

Patients

The eligibility criteria for enrollment into this study (March 2007–August 2010) were patients with histologically or cytologically proven pancreatic adenocarcinoma, patients with International Union Against Cancer clinical stage III (locally advanced disease: T4N0-1 and M0) or IV (metastatic disease: T1-4N0-1 and M1), patients with measurable lesions as defined in the Response Evaluation Criteria in Solid Tumors (RECIST) version 1.0 guidelines, age $\geq$20 and $\leq$80, no prior anticancer treatment for any malignancies, an Eastern Cooperative Oncology Group performance status (PS) $\leq$2, adequate bone marrow (leukocyte count $\geq$4,000/mm^3, neutrophil $\geq$2,000/mm^3, platelet count $\geq$100,000/mm^3, and hemoglobin $\geq$8.0 g/dl), adequate renal function (serum creatinine concentration $\leq$1.5 mg/dl and creatinine clearance $\geq$60 ml/min), adequate hepatic function (serum bilirubin level $\leq$2.0 mg/dl, serum alanine and aspartate transaminase levels $\leq$2.5 times the upper limit of the institutional normal; if biliary drainage was performed for jaundice before registration, the former $\leq$5 times the upper limit of the institutional normal and the

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latter ≤2.5 times the upper limit of the institutional normal), oxygen saturation ≥93%, adequate nourishment, no serious complications, life expectancy of at least 8 weeks, and provision of written informed consent from the patient.

Before randomization, a complete history was obtained and physical examination, routine hematology and biochemistry, ECG, chest X-ray, and abdominal computed tomography (CT) scan were performed.

Study design

PC-01 was an open-label, screening design, randomized phase II study. The primary end point was ORR. Secondary end points included treatment toxicity, clinical response benefit, PFS, and OS.

Patients were randomly assigned to the G group or the GS group in a 1:1 ratio. Random assignment was performed centrally by a web-based assistant system (flexible license assisted data server, JACCRO, Tokyo), using a computer-driven minimization procedure. Stratification factors were stage (III vs. IV), PS (0 or 1 vs. 2), and pain due to cancer (present vs. absent).

This study protocol was approved by the Protocol Review Committee of the JACCRO and Institutional Review Board of each institution, ClinicalTrials.gov identifier number was NCT00514163.

Protocol treatment

Eligible patients were randomly assigned to either the G group or the GS group. The G group patients received 1,000 mg/m^2 Gem intravenously during 30 min on days 1, 8, and 15, as 1 course repeated every 4 weeks. Patients with grade 4 hematological toxicities or grade 3 non-hematological toxicities underwent dose reduction to 800 mg/m^2 in the next course. The GS group patients received 1,000 mg/m^2 Gem intravenously during 30 min on days 1 and 8, and 40 mg/m^2 S-1 taken orally twice daily on days 1–14, every 3 weeks. When patients developed grade 4 hematological toxicities or grade 3 non-hematological toxicities by day 8, treatment was delayed by 1 week, and the S-1 dose was reduced to 60 mg/m^2 in the next course. In neither arms, prophylactic granulocyte-colony stimulating factor support allowed. Treatment was continued until progression, unacceptable toxicity, or patient refusal to continue the protocol treatment. The discontinuation of the protocol treatment for the reasons mentioned above was defined as protocol cessation.

Response and toxicity assessment

Toxicities were evaluated at each patient visit, according to the Common Terminology Criteria for Adverse Events version 3.0. CT or magnetic resonance imaging scans were performed at the baseline and after every 4 weeks to assess radiological response according to the RECIST version 1.0. Radiological tumor shrinkage of the primary tumor of the pancreas was assessed for all patients in the current study. ORR and DCR were set at the frequency of complete response plus partial response, in addition to stable disease among patients in each arm, respectively.

Clinical response benefit was assessed using daily analgesic consumption (measured in oral morphine-equivalent milligrams). Among patients who required opioid before the protocol treatment, patients whose opioid administration decreased to better than half of the baseline by day 1 of course 3 (8 weeks later in the G group and 6 weeks later in the GS group) were defined to be responders.

Statistical considerations

The primary endpoint was ORR. A sample size of 49 was required for a one-sided alpha value of 0.05 and a beta value of 0.20 with an expected response rate of 30% in the GS group and a threshold response rate of 10% in the G group. The protocol was activated in June 2007, and a total of 110 patients were planned for recruitment accounting for some drop-off

Table 1 Patient characteristics

Characteristics	G group ($n = 59$)	GS group ($n = 53$)	P value
	n	n	
Gender			
Male	35	32	1.00
Female	24	21	
Age			
<65	31	28	1.00
≥65	28	25	
ECOG PS			
0	45	44	0.66
1 or 2	14	9	
Locally advanced	18	13	0.53
Metastatic	41	40	
Metastatic sites			
Liver	30	28	0.85
Lymph node	10	6	0.43
Peritoneum	7	12	0.14
Lung	3	8	0.11
Ascites and/or pleural effusion			
Present	4	7	0.34
Absent	55	46	
Pain			
Present	20	17	1.00
Absent	39	36	

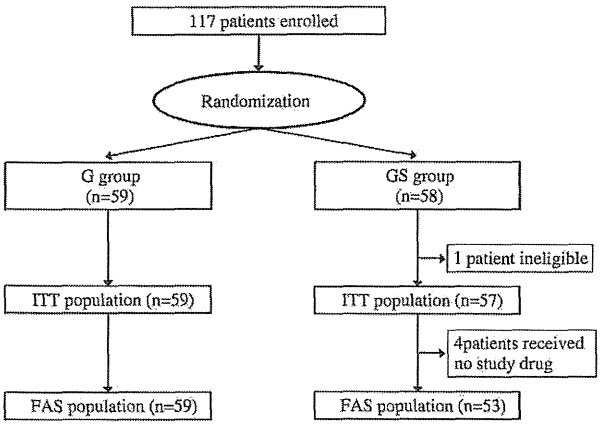

Fig. 1 Trial profile

cases within 1 year. If the null hypothesis (response rate) was not attained, the subsequent phase III trial would be designed to confirm the superiority of GS therapy to Gem alone.

The frequencies of each characteristic in Table 1 and each ORR and DCR in Table 3 were analyzed by the chi-square test.

OS was determined as the time from the date of registration to the date of death due to any cause and was censored at the date of the last follow-up for surviving patients. PFS was measured from the date of registration to the date of the first evidence of radiological or clinical progression, or death due to any cause and was censored at the date of the last follow-up CT for surviving patients with no clinical progression. OS and PFS were estimated by the Kaplan–Meier method, and the confidence interval (CI) was calculated with the Greenwood formula. Comparison of survival probability was conducted by the log-rank test. P values of less than 0.05 were considered to indicate statistically significant differences in the current study. The analysis was carried out with the SAS 9.2 statistical software (SAS Institute, Cary, NC, USA).

Results

Because of the poor recruitment rate, the protocol was amended twice, in January 2008 and February 2009, and a total of 117 patients were enrolled by August 2010 from 16 hospitals (see "Appendix"). One patient was judged to be ineligible after registration, because the final pathological diagnosis was not cancer. Accordingly, a total of 116 were allocated into either the G group ($N = 59$) or the GS group ($N = 57$) from among the intent-to-treat (ITT) population. Of the 116 patients, 4 in the GS group received supportive care instead of protocol treatment because of early deterioration or patient refusal. The full analysis set (FAS) consisted of 112, i.e., 59 and 53 patients in the G group and the GS group, respectively (Fig. 1).

Patient data registration was closed in June 2011, 10 months after the last patient registration. At the time of analysis, protocol treatment had been continued in 1 of 53 patients in the GS group. All analyses in comparison between the G group and the GS group were done in the FAS population, except OS.

Patient characteristics

Patient characteristics are shown in Table 1. The median age in the G group was 64 (41–79) years old, and that in the GS group was also 64 (45–77) years old. Although the protocol allowed enrollment of patients with PS 2, almost all patients were in good general condition (PS 0:1:2 was 79%:18%:3%, respectively). Metastatic disease was found in 72% of the patients. Analgesics (including opioids) were used in 33% (19%) of the patients at the baseline.

Toxicity

The major grade 3–4 adverse events are shown in Table 2. Although the frequency of grade 3–4 adverse events in the GS group was higher than that in the G group regarding both hematological and non-hematological toxicities, the toxicities were predictable and manageable. Discontinuation of the protocol treatment due to toxicity was seen in 13 (22%) of 59 protocol-cessation patients in the G group, and 14 (27%) of 52 protocol-cessation patients in the GS group. Treatment-related death was reported in 1 patient in each arm.

Clinical response benefit

At baseline, 12 and 10 patients required opioids in the G group and the GS group, respectively. There were 0 responders to opioids of 12 in the G group, and 2 of 10 in the GS group.

Objective response

Radiological responses are shown in Table 3. There was no complete response. The ORR in the GS group (28.3%) was significantly higher than that in the G group (6.8%), and the null hypothesis was rejected (two-sided $P = 0.005$). Also the DCR in the GS group was significantly higher.

In 31 patients with locally advanced disease, partial response was demonstrated in 1 (5.6%) of 18 patients in the G group, and 3 (23%) of 13 patients in the GS group. In the remaining 81 patients with metastatic disease, partial response was seen in 3 (7.3%) of 41 patients in the G group, and 12 (30%) of 40 patients in the GS group.

Table 2 Summary of maximum toxicity grades

Event	G group ($n = 59$)			GS group ($n = 53$)		
	Grade 3 (%)	Grade 4 (%)	Grade 3/4 (%)	Grade 3 (%)	Grade 4 (%)	Grade 3/4 (%)
Hematological						
WBC	5.1	0	5.1	20.8	5.7	26.4
Hemoglobin	5.1	0	5.1	7.5	0	7.5
Neutrophil	20.3	1.7	22.0	41.5	13.2	54.7
Platelet	3.4	1.7	5.1	7.5	7.5	15.1
Non-hematological						
Fatigue	5.1	1.7	6.8	3.8	0	3.8
Anorexia	5.1	0	5.1	3.8	0	3.8
Nausea	1.7	0	1.7	3.8	0	3.8
Diarrhea	0	0	0	3.8	0	3.8
Stomatitis	0	0	0	3.8	0	3.8
Skin rash	0	0	0	7.5	1.9	9.4
AST	3.4	0	3.4	1.9	0	1.9
ALT	6.8	0	6.8	3.8	0	3.8
ALP	6.8	0	6.8	3.8	0	3.8
Bilirubin	6.8	0	6.8	1.9	0	1.9
Albumin	0	0	0	1.9	0	1.9
C-reactive protein	0	0	0	1.9	0	1.9
Treatment-related death	1.7			1.9		

Progression-free survival

PFS curves are shown in Fig. 2. Discontinuation of the protocol treatment due to progression was seen in 34 (58%) of 59 protocol-cessation patients in the G group, and 20 (38%) of 52 protocol-cessation patients in the GS group. The median progression survival time in the GS group (6.15 months) was significantly longer than that in the G group (3.78 months, $P = 0.0007$).

Post-study treatment

After discontinuation of the protocol treatment, 37 (67%) of 55 patients in the G group and 23 (44%) of 52 patients in the GS group received various second-line treatments, most of which consisted of Gem or S-1 or both.

Overall survival in the ITT population

OS curves in the G group ($N = 59$) and the GS group ($N = 57$) are shown in Fig. 3. The GS group included 4 patients who deteriorated early or refused before protocol treatment, and subsequently received best supportive care without any anti-cancer treatment. The median survival time and 1-year survival probability in the G group and the GS group were 8.0 months and 29.0%, and 13.7 months and 55.9%, respectively. OS was

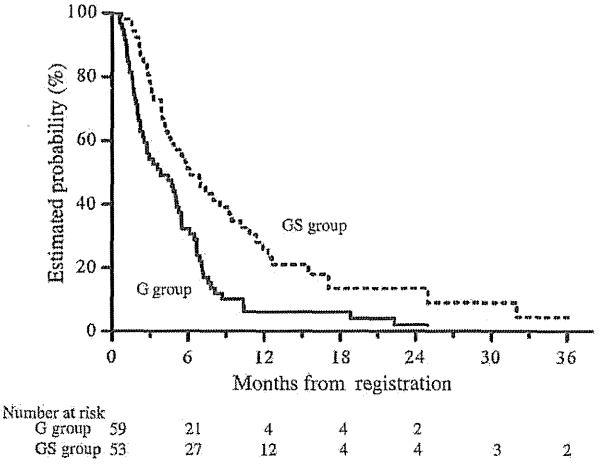

Fig. 2 Kaplan–Meier estimates of progression-free survival ($n = 112$)

significantly better in the GS group ($P = 0.035$), and its hazard ratio was 0.63 (95%, 0.41–0.97).

OS curves in the relation to extent of original disease are shown in Figs. 4 and 5. The median survival time in locally advanced and metastatic disease in the G group and the GS group were 8.7 and 7.7 months, and 14.6 and 12.9 months, respectively. OS in metastatic disease was significantly better in the GS group ($P = 0.029$).

Table 3 Objective response

Total (n = 112)	G group (n = 59)	GS group (n = 53)	P value
	n (%)	n (%)	
Complete response	0	0	–
Partial response	4 (6.8)	15 (28.3)	
Stable disease	22 (37.3)	19 (35.9)	
Progressive disease	23 (39.0)	7 (13.2)	
Not evaluable	10 (17.0)	12 (22.6)	
Objective response rate (%)	6.8	28.3	0.005
(95% CI)	(2.7–16.2)	(18.0–41.6)	
Disease control rate (%)	44.1	64.2	0.039
(95% CI)	(32.2–56.7)	(50.7–75.7)	
Locally advanced (n = 31)	G group (n = 18)	GS group (n = 13)	P value
	n (%)	n (%)	
Complete response	0	0	–
Partial response	1 (5.6)	3 (23.1)	
Stable disease	7 (38.9)	5 (38.5)	
Progressive disease	5 (27.8)	0	
Not evaluable	5 (27.8)	5 (38.5)	
Objective response rate (%)	5.6	23.1	0.284
(95% CI)	(1.0–25.8)	(8.2–50.3)	
Disease control rate (%)	44.4	61.5	0.473
(95% CI)	(24.6–66.3)	(35.5–82.3)	
Metastatic (n = 81)	G group (n = 41)	GS group (n = 40)	P value
	n (%)	n (%)	
Complete response	0	0	–
Partial response	3 (7.3)	12 (30.0)	
Stable disease	15 (36.6)	14 (35.0)	
Progressive disease	18 (43.9)	7 (17.5)	
Not evaluable	5 (12.2)	7 (17.5)	
Objective response rate (%)	7.3	30	0.011
(95% CI)	(2.5–19.4)	(18.1–45.4)	
Disease control rate (%)	43.9	65	0.075
(95% CI)	(29.9–59.0)	(49.5–77.9)	

Discussion

We set out to determine whether a combination of S-1 plus GS would obtain better results than GEM alone in a phase II study of unresectable pancreatic cancer.

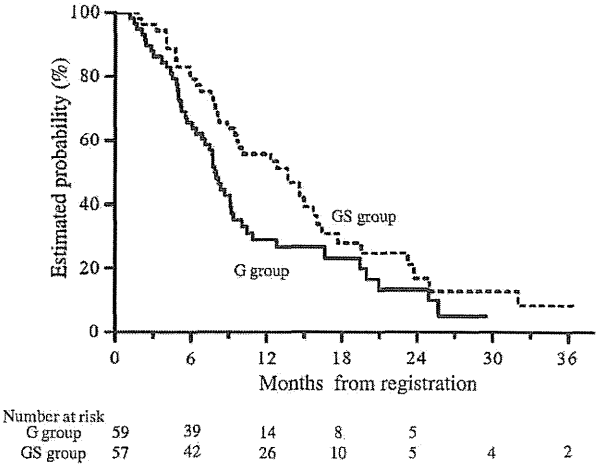

Fig. 3 Kaplan–Meier estimates of overall survival (n = 116)

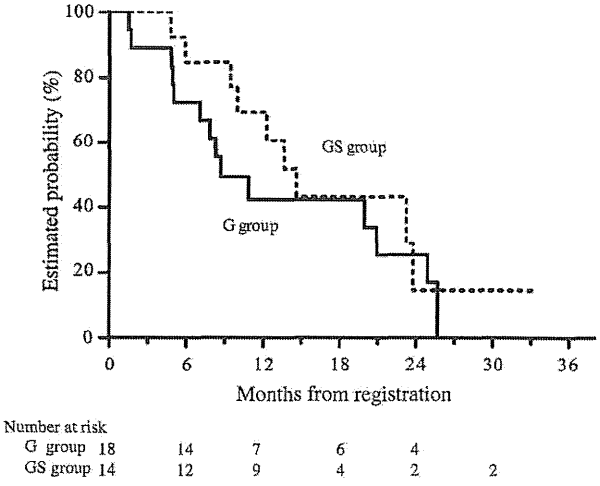

Fig. 4 Kaplan–Meier estimates of overall survival in locally advanced (n = 32)

The current PC-01 study, which was intended to screen GS as a promising investigation for a phase III trial comparing to standard Gem alone, successfully met this primary endpoint. Although the response rate obtained in the current study was lower than that in the previous one-arm phase II trials, the anticancer activity of GS was confirmed to be stronger than Gem alone [9–13]. Favorable results of GS as to PFS and OS data also encouraged us to plan a large phase III study comparing GS to standard Gem alone. However, results of large randomized phase III study of GS and Gem alone, known as the GEST trial, which was started by another Japanese cooperative group after our PC-01, were reported at the latest annual meeting of American Society of Clinical Oncology 2011 [14]. This large-scale (N = 600) GEST did not show OS superiority of GS compared to Gem alone. In terms of the survival benefit, this study seems to contradict the present PC-01 study.

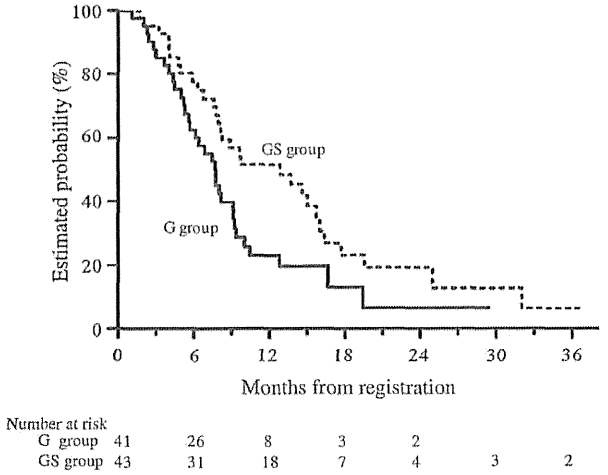

Fig. 5 Kaplan–Meier estimates of overall survival in Metastatic ($n = 84$)

Fluoropyrimidine and its derivatives have been intensively examined in combination with Gem for PC [15, 16]. All of those combinations have failed to show OS superiority compared to Gem alone in phase III settings, whereas relatively favorable results were generally reported in terms of response rate and survival. Accordingly, it may be important to explore a specific population in whom benefit would be maximized by GS therapy, though it may be difficult to develop Gem and fluoropyrimidine combination as a conventional frontline regimen for standard risk cases with advanced PC.

The main limitation of the PC-01 study derived from its inclusion of a relatively large number of patients who were found to be non-evaluable, mainly due to either the deterioration of the disease or patient refusal, which might well have affected the outcome of local response. On the other hand, randomized comparison of GS and Gem alone was one of the strengths of the current study. The ORR of GS in a previous non-randomized phase II study was extremely high, around 40%, perhaps due to selection bias [9–13]. However, in actual practice, since the response rate is usually below 30%, the PC-01 demonstrated a response rate acceptable to medical oncologists. Although PC-01 was not a phase III trial designed to confirm survival benefit, the OS and PFS data in the ITT population were impressive. The GS group showed a significant survival advantage against Gem group, even though the GS group included 3 cases of early deterioration. In the subset analysis, there was some discrepancy for the favorable population for GS between the current PC-01 and the GEST study. For example, GS was favorable in metastatic disease in PC-01; on the other hand, it was favorable in locally advanced disease in the GEST. GEMSAP, another Japanese study group, also carried out a randomized phase II trial of GEM and GS comparison and reported GS superiority to GEM in PFS in ASCO2011 [17].

Further accumulation of GEM and GS data might warrant an integrated meta-analysis to identify the population most likely to benefit from GS. Subsequently, a large randomized phase III trial to confirm GS advantages in a specific patients subset may be justified.

In conclusion, PC-01 demonstrated that GS had strong anticancer activity, and we believe that GS in some situations would be beneficial to give advanced PC patients.

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Conflict of interest No authors have any conflict of interest.

Appendix

The following investigators registered patients for this study:

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Yuki Hirose (Japan Red Cross Fukui Hospital, Fukui, Japan); Takaaki Ikari (Tobu Chiiki Hospital Tokyo Metropolitan Health and Medical Treatment Corporation, Tokyo, Japan).

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Original Article

Gastroduodenal stenting with Niti-S stent: Long-term benefits and additional stent intervention

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Background and Aim: Self-expandable metallic stents have mainly been used for the palliation of malignant gastric outlet obstruction (GOO). However, their use in long-term survivors and the feasibility, safety and benefit of additional intervention for stent dysfunction remain controversial. The present study examined the long-term benefits of endoscopic gastroduodenal stenting.

Methods: We reviewed 61 patients treated with Niti-S stents at several hospitals and estimated the efficacy of stent intervention, stent patency, eating period and factors related to poor effectiveness.

Results: All 61 first stent interventions and 14 additional stent interventions (11 second interventions and 3 third interventions) were successfully carried out. Clinical success rates were 83.6% and 85.7%, and median stent patency was 214 days and 146 days ($P = 0.47$), respectively. Fifty patients could be treated with a first stent only, and 11 patients received additional stents. At the time of study termination or death, 70.0% of the former group and 63.6% of the latter group maintained oral intake ($P = 0.71$), and each 86% and 100% among the group could maintain oral intake for a period exceeding half of their remaining lives after first stent intervention. Karnofsky performance status ≤50 ($P = 0.03$), ascites ($P = 0.009$), and peritoneal dissemination ($P = 0.001$) appeared to be factors related to poor effectiveness.

Conclusions: Despite the presence of factors related to poor effectiveness, endoscopic gastroduodenal stenting would be the first treatment of choice for GOO and provide long-term benefits. If stent dysfunction occurs, additional stent intervention enables continued oral intake safely.

Key words: additional stent intervention, factors related to poor effectiveness, gastric outlet obstruction (GOO), long-term benefit, Niti-S gastroduodenal stenting

INTRODUCTION

FOR PALLIATION OF symptomatic malignant gastric outlet obstruction (GOO), endoscopic gastroduodenal stent intervention is an effective and minimally invasive procedure, and has become an alternative to surgical gastroenterostomy.[1–3] With the availability of through-the-scope (TTS)-type stents, stent placement has become technically easier and more widespread.[4,5]

The Niti-S Pyloric Duodenal D-type stent (Niti-S stent; Taewoong Medical, Seoul, Korea) is an uncovered nitinol stent, and has unique features of low axial force, little foreshortening, and high expansible force. We use the Niti-S stent to palliate most of our patients with malignant GOO. However, only a few published reports on Niti-S gastroduodenal stenting exist.[6,7] Some studies have reported that the long-term outcome of endoscopic stent intervention is less favorable compared with surgical gastroenterostomy.[8,9] In our experience, many patients experience long-term survival after endoscopic gastroduodenal stenting, with continued palliation with additional stent interventions among those with stent dysfunction. However, the use of this approach in patients with long expected survival remains controversial.[10]

Estimating first and additional gastroduodenal stent interventions, the present study aimed to determine the feasibility, efficacy and benefit of long-term palliation of malignant GOO using Niti-S stents.

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METHODS

Patients

WE RETROSPECTIVELY STUDIED a total of 61 patients with symptomatic malignant GOO who received initial treatment with a Niti-S Stent from March 2011 to April 2013. Patients were enrolled at two tertiary medical centers in Japan: Aichi Cancer Center Hospital and Yokohama City University Hospital. All patients were assessed clinically, and all malignancies were staged with standard cross-sectional imaging, including computed tomography (CT) before stenting. We excluded patients who had been treated with stents other than Niti-S, those with prior surgical resection of the stomach or duodenum, and patients with multiple levels of obstruction in the intestinal tract.

Equipment and procedure

We used uncovered Niti-S stents of 6 cm, 8 cm, 10 cm, and 12 cm lengths, with fully expanded diameters of 20 mm or 22 mm. The 10-Fr delivery system enabled through-the-scope stent placement using the TJF-260V, JF-260V, CF-H260AI, and CF-Q240 (Olympus, Tokyo, Japan) endoscopes with a working channel diameter of ≥3.7 mm.

Patients were sedated with i.v. pethidine hydrochloride and midazolam. After identifying the site and length of obstruction with a water-soluble radiographic contrast medium using endoscopic retrograde cholangiography (ERCP) catheter (Tandem XL; Boston Scientific Japan, Tokyo, Japan), the stenosis was negotiated with a biliary guidewire (Jagwire; Boston Scientific Japan or RevoWave-J; Piolax Medical Devices, Inc., Kanagawa, Japan). Without pre-dilation, the stent delivery system was inserted over the guidewire through the working channel and deployed across the stricture with fluoroscopic guidance.

After stent intervention and follow up

One day after stent intervention, stent position and expansion were assessed with abdominal X-rays in all patients. If there was no abdominal pain, oral intake was allowed starting with liquids, followed by semisolids and then solids. If the patients' nutritional status improved sufficiently, they were considered for chemotherapy and/or radiotherapy, as appropriate. The primary physician followed up patients for symptom resolution until study termination (July 2013), or death.

The Gastric Outlet Obstruction Scoring System (GOOSS) was used before and after stent intervention for assessing symptomatic status of patients.[11] This score divides oral intake ability into the following categories: 0, no oral intake; 1, liquids only; 2, soft solids; 3, low-residue diet; and 4, normal diet. In the present paper, score 4 is our original score.

Outcomes

Primary outcome was long-term improvement in oral intake as estimated by eating period. Secondary outcomes were feasibility and safety of additional stent interventions, and factors related to poor effectiveness.

We compared the following factors between patients who were palliated with a single stent, and those who required additional stenting: technical success, procedure time, time to oral fluids and solids, length of hospitalization, oral intake ability, clinical effect, stent patency, stent dysfunction, and complications. The clinical effect was assessed 1 week after any interventions and classified into three levels: good, improvement of both oral intake and symptoms; moderate, improvement of either oral intake or symptoms; poor, improvement of neither oral intake nor symptoms. Early and late complications were defined as those occurring within 1 week, and later than 1 week after the procedure, respectively. The following factors were studied for prediction of poor stent efficacy: age, sex, site of obstruction, tumor stage, pre-intervention GOOSS, Karnofsky performance status (KPS), previous gastroduodenal or biliary stenting, ascites and peritoneal dissemination. We differentiated ascites from peritoneal dissemination by the presence of peritoneal nodules in the latter condition, with or without ascites, as detected by imaging modalities.

To estimate long-term outcome, patients with first-stent intervention only were compared to patients with additional stent intervention according to the following: final oral intake ability, eating period, survival time after first stent intervention, and percent eating period. Eating period and survival time were defined as the time from the date of the first stent intervention to the final follow up or death, respectively. We also reviewed post-procedure therapy and biliary drainage.

The present study was approved by the ethical committees of Aichi Cancer Center Hospital (3–145) and Yokohama City University Hospital (B140109014) and registered as a clinical trial (UMIN000012784).

Statistical analysis

Summary statistics are presented as means ± standard deviation for parametric data and as medians and interquartile range for non-parametric data. Standard statistical comparisons were made as appropriate. Multivariate analysis of factors related to poor effectiveness was done with the logistic regression method. Patency of duodenal stents, cumulative eating period, and cumulative survival of the two groups