Differentiation of Malignant and Benign Intraductal Papillary Mucinous Neoplasm by Repeated Pancreatic Juice Cytology Combined with Carcinoembryonic Antigen Level in Pancreatic Juice

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ABSTRACT

Objective The sensitivity of pancreatic juice cytology for intraductal papillary mucinous neoplasm is insufficient. We evaluated the usefulness of repeated pancreatic juice cytology via an endoscopic naso-pancreatic drainage tube combined with carcinoembryonic antigen level of pancreatic juice for the diagnosis of malignant intraductal papillary mucinous neoplasm. **Methods** Between April 2004 and February 2015, conventional pancreatic juice cytology and repeated pancreatic juice cytology were performed in 30 and 45 patients with intraductal papillary mucinous neoplasm, respectively. The carcinoembryonic antigen cutoff level of pancreatic juice for differentiation of malignant intraductal papillary mucinous neoplasm was determined from examination of 46 patients. The relationship between carcinoembryonic antigen level in pancreatic juice and carcinoembryonic antigen immunohistochemical expression of 32 resected tissues was also evaluated. **Results** The sensitivity, specificity, and accuracy of repeated pancreatic juice cytology were 52%, 83%, and 60%, respectively; the sensitivity was significantly higher than that of the conventional method (p=0.01). When repeated pancreatic juice cytology values and carcinoembryonic antigen level in pancreatic juice >72 ng/mL were combined, the sensitivity, specificity, and accuracy were 67%, 88%, and 72%, respectively and were 83%, 75%, and 80%, respectively, in patients with "worrisome features" and main pancreatic duct diameters from 5 to 9 mm. Post-ERCP pancreatitis was detected frequently by repeated pancreatic juice cytology in branch duct type intraductal papillary mucinous neoplasm. The carcinoembryonic antigen level of pancreatic juice in immunohistochemical analysis was correlated with carcinoembryonic antigen expression in resected specimens. **Conclusions** The repeated pancreatic juice cytology method was feasible for intraductal papillary mucinous neoplasm patients with main pancreatic duct diameters ≥5 mm. This method combined with the carcinoembryonic antigen level of pancreatic juice may be useful for patients with "worrisome features" and main pancreatic duct diameters of 5–9 mm.

INTRODUCTION

IPMN of the pancreas is characterized by papillary proliferation of columnar mucin-producing epithelial cells [1]. IPMNs show a wide histological spectrum and are suspected of progressing to invasive carcinomas in the adenoma–carcinoma sequence [2]. Surgical resection provides the best chance for cure in patients with malignant IPMN. According to the 2012 revised consensus guidelines (Fukuoka guidelines) [3], patients with Main duct (MD)-IPMN and Branch duct (BD)-IPMN with "high-risk stigmata," including obstructive jaundice, enhanced solid components, or dilation of the main pancreatic duct (MPD) to a diameter >10 mm, are strongly recommended for surgical resection. On the other hand, the surgical indication for patients with "worrisome features" is still unclear.

If image modalities, including computed tomography (CT), magnetic resonance cholangiopancreatography (MRCP), and endoscopic ultrasonography (EUS), do not provide sufficient information to decide the surgical indication, further approaches, such as EUS-fine-needle aspiration (EUS-FNA) or endoscopic retrograde pancreatography (ERP) may be considered. Although routine EUS-FNA or ERP for the evaluation of malignant IPMN is not recommended in the international consensus guideline, cytological results with fluid collection or pancreatic juice sometimes are critical for the management of IPMN, and molecular analysis, including the use of tumor markers, with these samples has been reported to be useful for evaluating malignant IPMN [4, 5, 6].

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Abbreviations BD branch duct; ENPD endoscopic naso-pancreatic drainage; IPMN intraductal papillary mucinous neoplasm; MD main duct; MPD main pancreatic duct; RPJC repeated pancreatic juice cytology
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Cytological examination of pancreatic juice obtained during endoscopic retrograde cholangiopancreatography (ERCP) is well established. In the last decade, EUS-FNA-based cytology has been increasingly used worldwide; however, the cytological diagnostic ability has not been satisfactory because of its low sensitivity [7, 8, 9, 10, 11]. A recent meta-analysis of EUS-FNA-based cytology revealed that it had good specificity but poor sensitivity in differentiating malignant from benign IPMN [12]. In addition, EUS-FNA-based cytology for mucinous cystic lesions is not generally performed in Japan because of the potential for seeding of tumor cells after EUS-FNA [13]. Recently, the usefulness of the repeated pancreatic juice cytology (RPJC) method via endoscopic naso-pancreatic drainage (ENPD) tube has been reported for diagnosis of pancreatic neoplasm, especially of early pancreatic cancer [14, 15, 16]; however, it is unclear whether this method is useful and feasible for diagnosis of IPMN [17].

Cyst fluid CEA level has been reported to be a useful marker for differentiation of mucinous from non-mucinous cysts [7, 18], but it is of limited value for differentiation of benign from malignant pancreatic cystic lesions [19]. When only patients with IPMN were analyzed, the diagnostic ability of cyst fluid CEA level to distinguish malignant from benign IPMN has given inconsistent results [4, 20, 21, 22]. A more recent report indicated that CEA level in pancreatic juice was a useful predictor of malignant IPMN [23, 24]. So, far, studies analyzing CEA level of pancreatic juice have not been reported from other institutions.

CEA immunohistochemical expression has been reported to correlate with histological grade of IPMN in resected tissues [25]. There has been only one study examining the relationship between cyst fluid CEA levels and degree of dysplasia. This study reported that CEA levels increased as the histologic grade of dysplasia progressed from low to high; however, CEA levels declined once invasive cancer developed [22]. To the best of our knowledge, there has been no report on the relationship of CEA level of cyst fluid or pancreatic juice and CEA expression in resected tissues of IPMN.

In this study, we compared the usefulness and feasibility of the RPJC method with those of the conventional method in patients with IPMN. We also evaluated use of the CEA level of pancreatic juice combined with the RPJC method for differentiation of malignant IPMN. In addition, we accessed the relationship between CEA level of pancreatic juice and immunohistochemical CEA expression in resected species of IPMN.

Methods

Patients

The subjects were 75 consecutive patients (56 men and 19 women; mean age, 68.3 years) with IPMN who underwent ERCP and aspiration cytology of pure pancreatic juice between March 2004 and February 2015. The patient characteristics diagnosed by EUS, CT, or MRCP were consistent with the 2006 criteria, which included branch type of IPMN with cyst size >30 mm and presence of nodules or MPD dilatation or the main duct type of IPMN. The histology of IPMN was based on analysis obtained during surgery (n=72). All resected specimens were examined pathologically and classified into four groups: IPMN with low-grade dysplasia (LGD), IPMN with intermediate-grade dysplasia (IGD), IPMN with high-grade dysplasia (HGD), and IPMN with an associated invasive carcinoma, according to the World Health Organization classification [1]. Malignant IPMN was defined as HGD and invasive carcinoma. In the absence of surgery, the reference standard for the diagnosis of IPMC was based on cytopathological detection of cancer cells from pancreatic juice or liver tissues obtained by biopsy from liver metastasis coupled with clinical and/or radiological evidence of progressive disease (n=3). This study protocol was approved by the ethics committee of our institution.

Cytological Examination

ERCP was performed by using a duodenoscope (JF 240 and JF 260V; Olympus Optical Co. Ltd., Tokyo, Japan). Between March 2004 and January 2008, conventional sampling of pure pancreatic juice for cytology and CEA examination was performed during ERCP in 25 patients with IPMN. These patients were examined by the conventional method and classified as the "conventional group." Between January 2008 and January 2015, we attempted to collect cytological samples of pancreatic juice repeatedly by using an ENPD tube in 50 patients with IPMN and could place the tube in 45 patients. These patients were classified as the "RPJC group". In this period, we failed to place the ENPD tube in the MPD in five patients in whom pancreatic juice could be obtained. These patients were included as the "conventional cytology group". The RPJC method was performed as previously reported [15]. If possible, the tip of the tube was placed close to the cyst or nodule in the MPD.

Pancreatic juice samples were centrifuged, and then smears of cell pellets were made on slide glasses, fixed in 95% ethyl alcohol, and stained by using the Papanicolaou technique. The cytological diagnoses were categorized into the following five groups: benign/reactive process (class 1, 2), atypical cells (class 3), severe atypical cells (class 3b), malignancy strongly suspected (class 4), and cytology conclusive for malignancy (class 5). Cytological results in which malignancy was suspected (classes 3b and 4) or conclusive (class 5) were regarded as cancer-positive, and atypical results were regarded as cancer-negative.

The sensitivities, specificities, positive predictive values, negative predictive values, and overall accuracies of the RPJC and conventional method for malignant IPMN were compared by $\chi 2$ test. In addition, the sensitivities of RPJC for malignant IPMN were examined according to clinical features, including type of IPMN, MPD diameter, size of mural nodule, numbers of cytological samplings, tumor location, and existence of invasion. A p value < 0.05 was considered to indicate statistical significance.

CEA Levels in Pancreatic Juice and CEA Immunohistochemistry

The CEA levels in the supernatant were measured by means of a CEA immunometric chemiluminescent assay kit (Fujirebio, Tokyo, Japan). The cutoff levels for pancreatic juice CEA level were determined to maximize the difference between benign and malignant IPMNs by receiver operating characteristic (ROC) curve analysis.

Sections of formalin-fixed paraffin-embedded tissue were subjected to immunohistochemistry using the avidin–biotin complex method to characterize the tumor cells. We used antibodies against carcinoembryonic antigen (NCL-CEA-2; Novocastra, Laboratories, UK; 1:100 dilution). The extent of immunohistochemical staining was graded by using a three-tiered scale. Positivity with apical or cytoplasmic staining in <10% of the tumor cells was defined as low, cases with 10–50% were defined as intermediate, and cases with >50% were defined as high expression. The relationship between CEA level of Pancreatic juice and CEA immunohistochemical expression was evaluated by using Spearman correlation coefficients.

Complications

After the procedure, the patients were carefully observed for any complications. Procedure-induced pancreatitis was defined as persistent abdominal pain continuing for ≥24 hours in association with the serum concentration of pancreatic enzyme (amylase) ≥3× the upper limit of normal. Pancreatitis was graded according to a modification of the 1991 consensus guidelines: mild, requiring fasting and treatment for ≤3 days; moderate, requiring fasting and treatment for 4–10 days; and severe, requiring fasting and treatment for >10 days, intensive care, or surgical intervention.

Frequencies of pancreatitis in the conventional and RPJC groups were compared by using Fisher's exact test. A p value <0.05 was considered to indicate statistical significance. IBM SPSS Statistics software version 20.0 (IBM Corporation, Chicago, IL) was used to perform all statistical analyses.

Ehics

In this retrospective study, written informed consent was not provided by the participants, but the documents that explain how the data included in this study would be used were displayed on bulletin board in Chiba university hospital. The study protocol conforms to the ethical guidelines of the "World Medical Association Declaration of Helsinki - Ethical Principles for Medical Research Involving Human Subjects" adopted by the 18th WMA General Assembly, Helsinki, Finland, June 1964 and amended by the 59th WMA General Assembly, Seoul, South Korea, October 2008, as reflected in a priori approval by our institutional review committee.

RESULTS

In the baseline characteristics, the conventional cytology group tended to include more patients with the branch duct type of IPMN; therefore, the group tended to have more patients with benign IPMN than did the RPJC group **(Table 1)**. All 30 patients in the conventional cytology group and 42 patients (93%) in the RPJC group underwent surgery. Surgery could not be performed on three patients because two had metastatic cancer and one rejected surgery because of old age. In the RPJC group, the mean number of samples of collected pancreatic juice was 4.9. The overall sensitivity, specificity, positive predictive value, negative predictive value, and accuracy of the conventional method for malignant IPMN were 12.5%, 100%, 100%, 52%, and 54%, respectively, and those of the RPJC method were 52%, 83%, 88%, 43%, and 60%, respectively, which showed significantly higher sensitivity for the RPJC method than for the conventional method **(Table 2)**. In the RPJC group, there were two false-positive cases: class 5 was detected in one patient with branch duct type, and class 4 was detected in one patient with the other main duct type. The sensitivities of the RPJC method according to clinical features, including the type of IPMN, MPD diameter, size of mural nodule, numbers of cytological samplings, tumor location, and existence of invasion, are shown in **Table 3**. In these groups, 22 (88%) of 25 patients who had high-risk stigmata, including the main duct type, and 11 (55%) of 20 patients who had worrisome features were malignant. The sensitivities of the RPJC method were 59% in the patients with high-risk stigmata and 39% in the patients with worrisome features. The sensitivities of the RPJC method tended to be higher in the main duct and combined type of IPMN than in the branch duct type (59% vs. 17%, p=0.07) and were not significantly different with respect to the size of mural nodules, numbers of cytological samplings, tumor location, and invasive or non-invasive IPMC.

CEA Levels in Pancreatic Juice and CEA Immunohistochemistry

We examined CEA levels in the pancreatic juice of 46 IPMN patients (15 patients from the conventional cytology group and 31 patients from the repeated cytology group) obtained during ERP. The mean CEA level of pancreatic juice was 1336 ng/mL in malignant IPMN and was 35.5 ng/mL in benign IPMN, (p=0.02). According to the ROC curves for CEA levels of pure pancreatic juice, the diagnostic cutoff levels for differentiation from malignant IPMN was 72 ng/mL and the area under the curve was 0.74 **(Figure 1)**. The sensitivity, specificity, and accuracy of the CEA cutoff level were 48%, 85%, and 61%, respectively. If either the CEA level in pancreatic juice was >72 ng/mL or cytological malignancy was interpreted as positive, the sensitivity, specificity, and accuracy were 67%, 88%, 72% in 32 IPMN patients, were 67%, 83%, and 73% in the patients with worrisome features (n=15), and were 83%, 75%, and 80% in the patients with worrisome features and MPD ≥5 mm (n=10) **(Table 4)**.

CEA immunohistochemistry was evaluated in 32 IPMN patients, including 22 with malignant and 10 with benign IPMN whose CEA levels of pancreatic juice were examined. The relationship between CEA level of pancreatic juice

Table 1. Baseline characteristics of RPJC and conventional cytology group.

	RPJC group (N=45)	Conventional cytology group (N=30)	P
Mean age (range)	66.9 (43-80)	70.4 (55-85)	0.13
Sex male: female	33:12:00	23:07	0.79
Location head: body+tail	26:16:00	20:10	0.8
Surgery	42	30	0.27
Main+Mixed: Branch	35:10:00	17:13	0.07
Cyst size (mm)	34.4	31.7	0.56
MPD diameter	8.1	7.6	0.13
Nodule size	9 (n=30)	10.4 (n=24)	0.9
Malingnant: Benign	33:12:00	16:14	0.09
Mean number of cytological samplings	4.9	1	

Table 2. Diagnostic yields of RPJC and Conventional method for malignant IPMN.

	RPJC method	Conventional method	P
Sensitivity	17/33 (52%)	2/16 (12.5%)	0.01
Specificity	10/12 (83%)	14/14 (100%)	0.2
Positive predictive value	15/17 (88%)	2/2(100%)	1
Negative predictive value	12/28 (43%)	14/28 (50%)	0.59
Accuracy	27/45 (60%)	16/30 (53%)	0.63

Table 3. Sensitivity of RPJC method for malignant IPMN according to clinical factors.

		Sensitivity	P
2012 Guideline	High risk stigmata (n=25)*	13/22 (59%)	0.28
	Worrisome features (n=20)**	4/11 (36%)	
Type	Main duct /Combined	8/10 (80%) /8/17(47%)	0.07
	Branch duct	1/6 (17%)	
MPD diameter	10 mm<	7/12 (58%)	0.07
	/5-10 mm	/9/15 (60%)	
	<5 mm	1/6 (17%)	
Size of nodule	7.5 mm<	9/18 (50%)	1
	7.5 mm≧	6/12 (50%)	
Number of cytological samplings	≧4 times	13/23 (57%)	0.46
	≦3 times	4/10 (40%)	
Location	Head	10/19 (53%)	1
	Body and tail	5/11 (45%)	
Invasive ca or non-invasive ca	Invasive	9/16 (56%)	1
	Non-invasive	8/17 (47%)	

* Including MD-IPMN with high risk stigmata (n=9); ** Including MD-IPMN with worrisome features (n=3)

and CEA immunoreactivity of the tissue sample is shown in **Figure 2**. Malignancy was detected in 6 of 13 patients with low or intermediate expression and in 16 of 19 with high expression of CEA immunoreactivity. Malignancy was significantly more often detected in patients with high expression than in those with low and intermediate expression (p=0.049). The mean CEA levels of the low, intermediate, and high expression categories of CEA immunoreactivity were 2.8 ng/mL, 32.9 ng/mL, and 1616.7 ng/mL, respectively. The CEA levels of pancreatic juice were significantly different between the patients with low and intermediate expression (p=0.04) and between those with intermediate and high expression of CEA immunoreactivity (p=0.01). The CEA level of pancreatic juice positively correlated with CEA immunohistochemical expression (r=0.67, p<0.01).

Complications

Post-ERCP pancreatitis was identified in 4 patients (8.9%) in the RPJC group and in two patients (7.1%) in the conventional cytology group, but the difference was not significant. In the RPJC group, two patients (5.7%) developed mild pancreatitis in the main duct and combined type of IPMN; on the other hand, in the branch duct type of IPMN, two patients (20%) developed pancreatitis, including one with mild and one with moderate severity. All of them were cured by conservative treatment.

DISCUSSION

In this study, we first separately examined the diagnostic abilities of the RPJC method and CEA level of pancreatic juice and found that their sensitivities were insufficient

for differentiation of malignant IPMN. However, if the RPJC method results were combined with a CEA level in pancreatic juice >72 ng/mL, the sensitivity and accuracy improved to >80%, especially in patients with "worrisome features" whose MPD size was 5–9 mm.

Cytological examination of pancreatic juice obtained during ERCP has been performed since ERCP was first introduced, and its sensitivity for malignant IPMN has been reported to range from 30–40% [10, 11]. Recently, excellent results of PJC using balloon catheter or lavage cytology with cell block have been reported [26, 27]. Regarding cytological interpretation, severe atypical cells (class 3b) were interpreted as cancer-positive in our study because high-grade epithelial atypia has been reported as being a more sensitive predictor of malignancy than positive cytology [28, 29]. However, the 12.5% sensitivity for the conventional method was relatively lower than the levels reported in other studies. This result may be because of the unavailability of secretin, which was discontinued in Japan in 2004. The sensitivity of the repeated cytology method was 57%, which was significantly higher than that of the conventional method (p=0.01). The sensitivities of the repeated cytology method tended to be higher for the main duct and combined types of IPMN than for the branch duct type (59% vs. 17%, p=0.07). Positive results of pancreatic juice cytology were more likely to be easily obtained when malignant IPMN was present in the MPD. However, the sensitivity of this method was insufficient for differentiation of malignant from benign IPMN, especially in the 39% of patients with "worrisome features" **(Figure 3)**.

According to the 2012 revised consensus guidelines, patients with MD-IPMN and BD-IPMN with "high-risk stigmata" are recommended for surgical resection. In contrast, patients with BD-IPMN with "worrisome features" are recommended for evaluation without immediate resection. In fact, our data showed that 22 (88%) of 25 patients who had high-risk stigmata, including MD-IPMN, and 11 (55%) of 20 patients who had worrisome features, including MD-IPMN with MPD<10 mm, had malignancies. With regard to MD-IPMN, 8 (89%) of 9 patients with high-risk stigmata and 2 (67%) of 3 with worrisome features had malignancies. Therefore, further improvements are required to differentiate benign from malignant IPMN, especially in patients with worrisome features, including MD-IPMN with MPD<10 mm.

Marie F *et al*. reported that a cyst fluid CEA concentration >200 ng/mL had sensitivity and specificity of 90% and 71%, respectively, for diagnosis of malignant IPMN [4]; however, these data have not been reproduced in subsequent studies, so its ability to distinguish malignant IPMN is controversial. Recent meta-analysis showed that CEA cutoff levels for determining a malignant cyst have ranged from 109.9–6000 ng/mL, and pooled estimates of CEA in malignant cysts have led to poor prediction. On the other hand, Hirono S *et al*. reported that a CEA concentration >110 ng/mL in pancreatic juice was the only independent predictive factor for malignant IPMN [23]. More recently, they also reported that a CEA concentration >30 ng/mL had sensitivity, specificity, and accuracy of 94%, 85%, and 80%, respectively, for diagnosis of malignant BD-IPMN [24]. These results were difficult to compare because the specimens (cyst fluid or pancreatic juice), sampling method (EUS-FNA or ERCP), and variety of pancreatic cyst were different. We speculated that CEA levels of pancreatic juice may reflect secretion of CEA from tumor cells of the MPD as well as the branch duct. In this study, the CEA cutoff concentration was 72 ng/mL for differentiation of malignant from benign IPMN, and a CEA concentration >72 ng/mL had sensitivity and specificity of 48% and 85%, respectively. From our results, it appeared that the CEA level of pancreatic juice was of limited value in differentiating malignant from benign IPMN. However, if the RPJC method results and a CEA level >72 ng/mL in pancreatic juice were combined, the sensitivity and accuracy improved.

CEA immunohistochemical analysis revealed that high CEA expression was significantly more often detected in malignant IPMN than in benign IPMN and that the CEA

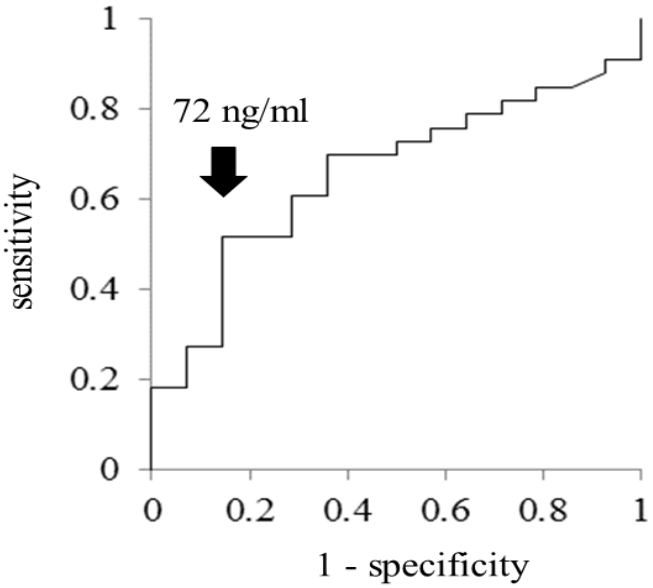

Figure 1. According to the ROC curves for CEA levels of pure pancreatic juice, the diagnostic cutoff levels for differentiation from malignant IPMN was 72 ng/mL and the area under the curve was 0.74.

Table 4. Diagnostic yields of RPJC method combined with CEA level in PJ for malignant IPMN.

	Sensitivity	Specificity	Accuracy
RPJC method (n=45)	17/33 (52%)	10/12 (83%)	27/45 (60%)
CEA≧72 (n=46)	17/33 (48%)	11/13 (85%)	28/46 (61%)
RPJC method or CEA≧72 (All patients n=32)	16/24 (67%)	7/8 (88%)	23/32 (72%)
RPJC method or CEA≧72 (patients with worrisome features n=15)	6/9 (67%)	5/6 (83%)	11/15 (73%)
RPJC method or CEA≧72 (patients with worrisome features+MPD≧5 mm n=10)	5/6 (83%)	3/4 (75%)	8/10 (80%)

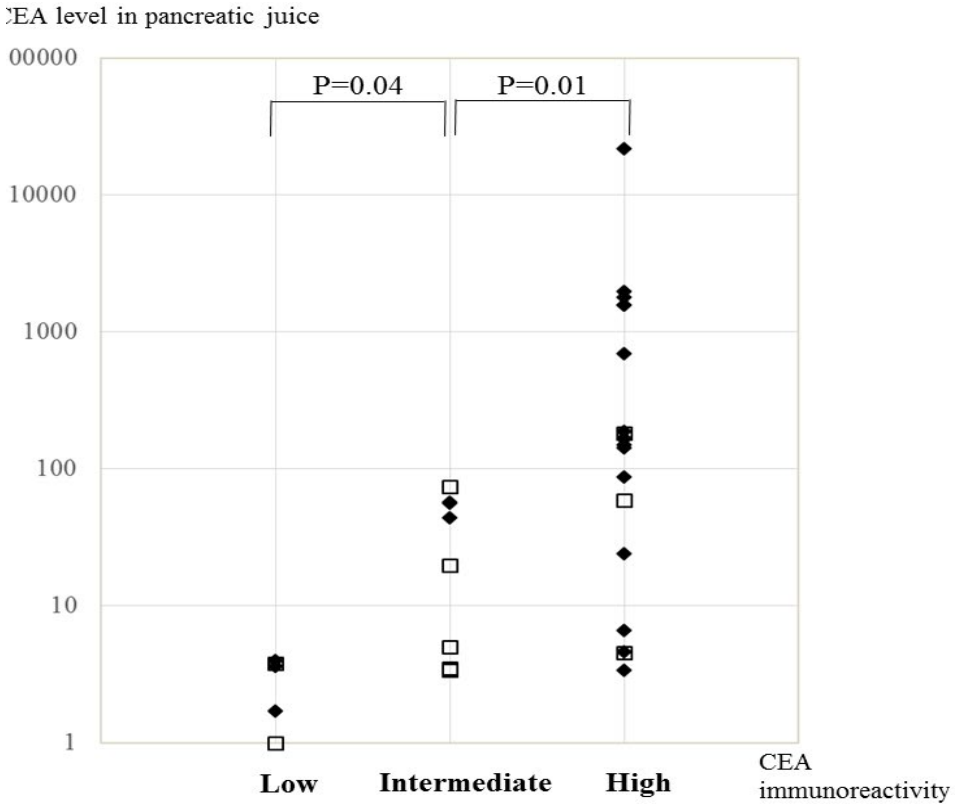

Figure 2. The relationship between CEA level of pancreatic juice and CEA immunoreactivity of tissue samples is shown. High CEA expression was detected in 16 of 22 patients with malignant IPMN and in 3 of 10 patients with benign IPMN. High CEA expression was significantly more often detected in malignant IPMN than in benign (p = 0.049). The mean CEA levels of low, intermediate, and high expression of CEA immunoreactivity are 2.8 ng/mL, 32.9 ng/mL, and 1616.7 ng/mL, respectively. The CEA levels of pancreatic juice are significantly different between the patients with low and intermediate expressions (p = 0.04) and between those with intermediate and high expressions of CEA immunoreactivity (p = 0.01). The CEA level of pancreatic juice positively correlates with CEA immunohistochemical expression (r = 0.67, p < 0.01).

□ indicates benign IPMN, and ◆ indicates malignant IPMN.

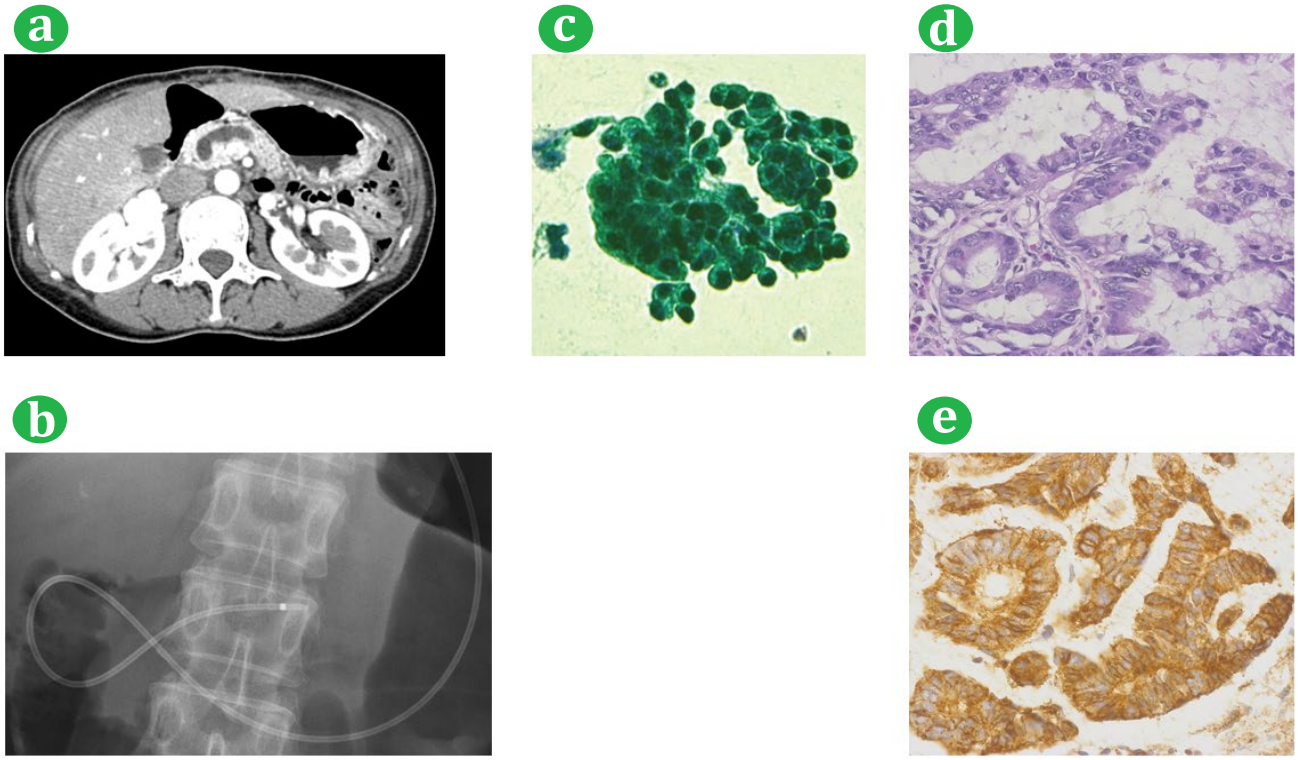

Figure 3. (a). In a 58-year-old woman, multidetector-row CT showed main pancreatic duct dilatation with a diameter of 8 mm without an apparent nodule that is categorized as MD-IPMN with "worrisome features." **(b).** ERP was performed, the ENPD tube was left in place for 3 days, and cytological samples were collected six times; the samples are class 3, 3, 3, 5, 3, and 4, respectively. **(c).** Cytological diagnosis is positive at the fourth and sixth samplings. **(d).** After examination by pancreatoscopy, a total pancreatectomy was performed. Pathological examination of the resected specimen showing micro invasive IPMC. **(e).** The CEA levels of pancreatic juice are 1580 ng/mL, and CEA immunohistochemical analysis indicates high expression.

level of pancreatic juice correlated with CEA expression. However, 4 of 17 malignant patients with IPMN and high CEA expression had low CEA concentrations of pancreatic juice, which caused the sensitivity of CEA level of pancreatic juice for differentiation of malignant IPMN to be low. Interestingly, these four patients all had invasive IPMC. Kucera S *et al*. reported that cyst fluid CEA level increased with increasing histological grade but declined with development of invasive cancer. They speculated that fewer cells with tight junctions were present and therefore, there was less CEA at the luminal surface available for release into the cystic fluid.

The major complication associated with ERCP is post-ERCP pancreatitis, the incidence of which varies widely from 1–8%. In the present study, post-ERCP pancreatitis occurred in two patients (6.7%) in the conventional group and in four patients (8.9%) of the repeated cytology group, all of which resolved with conservative treatment. The incidence of pancreatitis was slightly higher in the conventional group than in the repeated cytology group, possibly because the viscosity of the pancreatic juice may have slowed flow through the ENPD tube. In addition, pancreatitis tended to occur more in BD-IPMN patients (20%) than in MD and combined IPMN patients (5.7%). This finding might have been related to the presence of chronic obstructive pancreatitis by mucin, which could have developed in more cases of MD and combined IPMN than in cases of BD-IPMN and resulted in a lower incidence of post-ERCP pancreatitis. These findings were similar to those reported previously in which pancreatic stent placement increased the post-ERCP pancreatitis in male patients with IPMN, possibly because of obstruction by mucin, but no patients with a dilatation of the MPD >6 mm had post-ERCP pancreatitis [30]. Considering this frequency of pancreatitis and its low sensitivity, the RPJC method does not appear to be suitable for patients with BD-IPMN.

There were some limitations in this study. This was a retrospective study with a small number of patients conducted at a single tertiary center. In addition, there may have been a selection bias because we could not perform ERCP with pancreatic juice cytology and CEA analysis of pancreatic juice consequently.

CONCLUSION

In conclusion, the repeated cytological method was found to be feasible for IPMN patients with MPD diameters ≥5 mm. This method combined with a CEA level of pancreatic juice may be useful for patients with "worrisome features" and MPD diameters from 5–9 mm. Further studies with larger numbers of patients will be needed to confirm the reliability of this method.

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Conflict of Interest

The authors have no conflicts to disclose. All authors disclosed no financial relationships relevant to this publication.

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