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Role of serial EUS-guided FNA on pancreatic cystic neoplasms: a retrospective analysis of repeat carcinoembryonic antigen measurements

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Abstract

Background and Aims—Pancreatic cystic neoplasms (PCNs) often need interval surveillance, including repeat EUS, but the role of repeat FNA with fluid analysis is poorly defined. The aim of this analysis is to evaluate the potential clinical significance of serial carcinoembryonic antigen (CEA) measurements by EUS-guided FNA (EUS-FNA) in the surveillance for PCNs.

Patients—Patients who underwent EUS-FNA for PCNs were studied retrospectively. EUS-FNA findings were compared between index and prior procedures among patients who underwent repeat EUS-FNA.

Results—A total of 400 patients with PCNs underwent EUS-FNA. Eighty-seven of those patients had prior EUS-FNA with cyst fluid analysis. Patients with repeat FNA were significantly more likely to have multiple cysts (57% vs 41%; $P = .008$), multilocular cysts (75% vs 62%; $P = .042$), connection to pancreatic duct (33% vs 18%; $P = .005$), and higher initial CEA levels (94.8 vs 25.6 ng/mL; $P = .003$) compared with patients who had only a single FNA. A comparison of prior and index FNAs did not show significant differences in EUS or cyst fluid analysis findings. After log transformation, the association between CEA level at prior and index FNA was moderate ($R^2 = 0.626$; $P < .001$), but cystic fluid CEA classification with a cutoff value of 192 ng/mL changed in 17 patients (20%), without significant changes in EUS findings.

Conclusions—Repeat surveillance EUS-FNA resulted in stable CEA levels in the majority of patients, with spurious fluctuations of CEA in approximately 20% of patients. These data call into question any clinical significance attributed to an isolated interval rise in CEA level, especially in light of a stable EUS examination.

We increasingly encounter asymptomatic pancreatic cystic neoplasms (PCNs) in clinical practice because of the increase in opportunities of abdominal imaging such as US or CT as well as the improvement in imaging modalities.¹ Given the wide range of diagnoses from benign to malignant, the accurate diagnosis of PCN is essential, albeit challenging, for treatment decisions.^{2,3}

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Cytology and cyst-fluid analysis are performed routinely in EUS-guided FNA (EUS-FNA) for PCN.^{4,5} Specificity of cytology in the diagnosis of malignant PCNs is high, but its sensitivity is low. Meanwhile, carcinoembryonic antigen (CEA) levels in the cyst fluid are useful to differentiate mucinous versus nonmucinous cysts, but they are not useful for the diagnosis of malignant PCNs.^{6,7} Although various molecular markers have been reported,⁸ they are not yet readily available for clinical use, and cyst fluid CEA measurement remains the mainstay for evaluation of PCNs. Given the malignant potential of PCNs such as side-branch intraductal papillary mucinous neoplasms (IPMNs), many patients with PCN need continued surveillance. The revised Sendai consensus guideline⁹ recommends EUS and magnetic resonance imaging (MRI) as the procedures for surveillance; however, little is known about the role of repeat EUS-FNA during surveillance of PCNs. We, therefore, conducted this retrospective analysis of EUS-FNA for PCNs to evaluate the potential clinical significance of serial CEA measurements by EUS-FNA in patients under surveillance for PCNs.

PATIENTS AND METHODS

Patients

Consecutive patients with clinically suspected PCNs who underwent EUS-FNA with cyst fluid analysis (index FNA) between June 2009 and May 2012 at the H.H. Chao Comprehensive Digestive Disease Center, University of California, Irvine Medical Center, were included in the analysis. Medical records including age, sex, EUS-FNA findings, cyst fluid analysis, and cytology results were retrospectively reviewed. Cyst characteristics (cyst number, location, diameter, macrocystic versus microcystic, multilocular versus unilocular, the presence of connection to the pancreatic duct, mural nodule) on EUS, and FNA procedure details (needle size, the amount of obtained cyst fluid) were retrieved from EUS reports. Data from previous EUS-FNA (prior FNA), if any, also were retrieved. This study was approved by the local Institutional Review Board.

EUS-FNA procedure

After careful EUS examination, the cyst was punctured by using a 19-, 22-, or 25-gauge FNA needle, and cyst fluid was aspirated with suction. A 22-gauge needle was used primarily for EUS-FNA for PCNs, but if the fluid was too viscous to come through the smaller needle, a 19-gauge needle was used. A 25-gauge needle was used only in cases with a risk of bleeding because of cysts located adjacent to a vessel. The cyst fluid was sent for fluid analysis (CEA and amylase) and cytology. Cyst fluid CEA measurement was performed in a 1-step sandwich immunoassay on Immulite 2000 XPI (Siemens Medical Solutions USA Inc, Malvern, Pa). Cyst fluid CEA was categorized by using a cut-off value of 192 ng/mL. When the amount of the aspirated cystic fluid was small, the fluid was preferentially sent for fluid analysis with dilution by normal saline solution, if necessary.¹⁰ A prophylactic oral quinolone was routinely administered for 5 days after the procedure.

Statistical analyses

EUS and EUS-FNA results of index and prior EUS-FNAs were compared among patients who underwent repeated EUS-FNAs for PCNs. The Pearson correlation coefficient was used

to assess the correlation between cyst fluid CEA levels of prior and index EUS-FNAs. Cyst fluid CEA category (≥ 192 vs <192 ng/mL) was compared between index and prior EUS-FNAs by using the McNemar test.

Continuous variables are presented as the mean and standard deviation (SD) or the median and range. Statistical comparisons were performed with a chi-square test or the Fisher exact test for categorical variables and the *t* test or the Wilcoxon rank-sum test for continuous variables. A *P* value $< .05$ in a 2-tailed test was considered a statistically significant difference. JMP software version 10.0 (SAS Institute, Inc, Cary, NC) was used for all statistical analyses.

RESULTS

Patients

A total of 400 patients with PCNs underwent EUS-FNA between June 2009 and May 2012, and among these 400 patients, 87 patients had prior EUS-FNA with cyst fluid analysis with a median interval of 8.7 months (range 0.8-91.7 months). These patients underwent repeat EUS-FNA for various reasons as clinically dictated, including CEA values inconsistent with gross fluid characteristics, unusual EUS features, and patient or physician preferences.

Patient characteristics and FNA results in patients with repeat FNAs ($n = 87$) compared with those with 1-time FNA ($n = 313$) are shown in Table 1. Patients with repeat FNA were more likely to have multiple cysts (57% vs 41%; $P = .008$), multilocular cysts (75% vs 62%; $P = .042$), connection to a pancreatic duct (33% vs 18%; $P = .005$), and higher cyst fluid CEA levels (94.8 vs 25.6 ng/mL; $P = .003$).

EUS-FNA results in 87 patients with repeat FNAs

A comparison of index and prior EUS-FNA results in 87 patients with repeat FNA is shown in Table 2. There were no significant differences in EUS and EUS-FNA findings between index and prior EUS procedures. The median cyst fluid CEA was 94.8 ng/mL at index FNA and 118 ng/mL at prior FNA. Cyst fluid CEA levels decreased 18.1% at median (range 1241% decrease to 100% increase) from prior FNA. Although cyst fluid CEA levels at index and prior EUS-FNAs were well correlated with each other after log transformation ($R^2 = 0.626$; $P < .001$), the CEA category with a cutoff value of 192 ng/mL changed in 17 patients (20%). Cyst fluid CEA changed from low to high in 6 cases (7%) and high to low in 11 cases (6%). Details of these 17 discordant cases are shown in Table 3.

After repeat EUS-FNA, only 3 patients underwent surgical resection, although EUS findings with CEA category did not change between 2 EUS-FNA procedures. Surgery was performed at the discretion of patients, and the pathologic diagnoses of these patients were 1 IPMN with concomitant pancreatic intraepithelial neoplasia (PanIN) and 2 PanIN.

Serial cyst fluid CEA change in patients undergoing 3 or more EUS-FNAs

Among 87 patients with repeat EUS-FNAs, 14 patients underwent 3 or more EUS-FNAs for PCNs; 3 EUS-FNAs in 11 patients, 4 EUS-FNAs in 1, and 5 EUS-FNAs in 2. Serial CEA change is shown in Figure 1. Cyst fluid CEA levels showed considerable fluctuations over

time. CEA category was not consistent in 7 patients (50%), although EUS findings did not change considerably.

DISCUSSION

In our retrospective analysis of EUS-FNA in the surveillance of PCNs, cyst fluid CEA levels did not change significantly in the majority of patients during surveillance, but fluctuations of CEA without changes in EUS imaging were observed in approximately 20% of patients.

EUS plays a central role in the management of PCNs¹¹ because it provides accurate imaging with its high resolution and because of the anatomic proximity of the pancreas and upper GI tract. However, EUS diagnosis is confounded by the apparent subjectivity of its interpretation, and imaging alone is not sufficient for the accurate diagnosis of PCNs.¹² EUS-FNA for PCNs has been reported to increase the diagnostic yield of EUS.⁶ The incremental increase in the diagnostic yield of EUS and fluid analysis over CT and MRI for prediction of neoplastic cysts is reported to be 36% and 54%, respectively.¹³ Cyst fluid CEA has been widely used for the differentiation of mucinous and nonmucinous cysts, but recent reports cast doubt on its usefulness.^{14,15} Although mucinous cysts are potentially malignant, surgical resection is not always the first choice because of its invasiveness. It was estimated that in elderly patients with comorbidities, noninvasive surveillance, or even doing nothing, can be the management of choice in terms of quality-adjusted life-years.¹⁶ In the revised Sendai guideline,⁹ close surveillance by using EUS and MRI is recommended in patients with mucinous cysts >2 cm. Although EUS-FNA is widely used in the diagnosis of PCN, the role of repeat EUS-FNA is not fully discussed in the Sendai guideline. The recently published American Gastroenterological Association guideline¹⁷ emphasized the role of MRI for surveillance of asymptomatic PCNs. EUS-FNA is recommended only in PCNs with at least 2 high-risk features or in PCNs newly developing those high-risk features during follow-up. Our results, in part, support the American Gastroenterological Association guideline in that repeat EUS-FNA (for the sole purpose of repeating the CEA level) is unlikely to change the management of PCNs in the absence of morphologic changes on EUS. However, as described in the guideline, those recommendations were based on very low-quality evidence, and Canto and Hruban¹⁸ clearly pointed out the problems with a non-EUS/FNA strategy. Given the higher detection rate of mural nodules using EUS than using MRI, EUS should be incorporated into the surveillance of PCNs, perhaps alternating with MRI, and repeat FNA may be considered for cytologic evaluation in the presence of a new mural nodule. If cytology on repeat EUS-FNA suggested the presence of malignancy, surgery should be recommended. Meanwhile, given our results, there is no clear evidence that routine EUS-FNA for the sole purpose of remeasuring the CEA level should be performed in the absence of change in EUS image. Certainly, a decision to recommend surgery for a pancreatic resection based on an isolated rise in CEA level is not supported by our data. The reason for these spurious fluctuating CEA levels is not entirely clear. However, we noticed variability of simultaneous CEA levels taken from different cysts within the same pancreas, possibly because the mucin concentration may vary (clumping) within the same cyst as seen routinely during EUS-guided cystoscopy by using through-the-needle fiber optic imaging¹⁹ (described as “mucin clouds”). It is quite possible that the more highly concentrated mucin clumps may yield higher CEA levels. Finally, a recent study analyzing

the CEA level among various IPMN subtypes showed that the highest CEA levels were among the most benign gastric subtype.²⁰ Therefore, we should consider these possible explanations for rising CEA levels before sending a patient to surgery.

Repeat EUS-FNA should be considered if it provides additional information beyond CEA for the purpose of risk stratification. For example, recently described EUS-guided through-the-needle imaging needle-based confocal laser endomicroscopy (nCLE)²¹ enables in vivo endomicroscopy during EUS procedures.²² We reported on the feasibility of dual through-the-needle imaging (cystoscope and nCLE).¹⁹ In addition, nCLE has potential for in vivo immunohistologic staining by antibody-tagged contrast material,²³ which might enable future risk stratification of PCNs, that is, subtypes of IPMNs.²⁴

There are some limitations in this study. Selection bias could not be avoided because of the retrospective design. The lack of pathologic diagnosis by surgical specimen is another limitation, but this was inevitable given that our study population was under surveillance for PCNs. In addition to the pathologic diagnosis, data on long-term outcomes were not available. Therefore, we could not evaluate the association of changes in repeat EUS-FNA with long-term outcomes of PCNs.

In conclusion, this study showed that repeat surveillance EUS-FNA resulted in stable CEA levels in the majority of patients, with unexplained changes of CEA category seen in approximately 20% of patients. These data call into question any clinical significance attributed to an isolated interval change in CEA category, especially in light of a stable EUS examination. However, the long-term impact of repeat EUS-FNA on the management of PCNs should be further investigated, especially in cases with high-risk stigmata.

Abbreviations

CEA	carcinoembryonic antigen
EUS-FNA	EUS-guided FNA
IPMN	intraductal papillary mucinous neoplasm
MRI	magnetic resonance imaging
nCLE	needle-based confocal laser endomicroscopy
PCN	pancreatic cystic neoplasm

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Figure 1. Serial carcinoembryonic antigen changes in 14 patients who underwent 3 or more EUS-guided FNAs for pancreatic cystic neoplasms.

Table 1

Patient characteristics of patients with multiple FNAs and with 1-time FNA

	Multiple FNA n = 87	1-time FNA n = 313	P value
Age, mean ($\pm$ SD), y	69.4 (11.6)	67.6 (11.4)	.339
Sex, male/female, %	40/60	38/62	.710
Multiple cysts, %	57	41	.008
Head/body or tail, %	31/69	37/63	.315
Cyst diameter, mean ($\pm$ SD), mm	24.7 (16.2)	25.7 (20.3)	.635
Macro-/microcystic/mixed, %	94/0/6	92/2/6	.776
Multilocular/unilocular, %	75/25	62/38	.042
Connection to PD on EUS, %	33	18	.005
Mural nodule, %	9	8	.665
Needle size, 19/22/25 gauge, %	20/80/0	11/88/1	.054
Fluid amount, median (range), mL	1 (0.2-57)	1 (0.1-960)	.576
Dilution with saline solution, %	25	25	1.000
Viscous cyst fluid, %	34	30	.431
CEA, median (range) ng/mL	94.8 (0.3-6147)	25.6 (0.2-104473)	.003
CEA >192 ng/mL, %	33	21	.024
Amylase, median (range), IU/L	800 (<5-800)	800 (<5-800)	.240
Cytology test performed, %	76	81	.368
Malignancy on cytology*, %	2	4	.539
Mucin on cytology, %	11	14	.721

SD, Standard deviation; *PD*, pancreatic duct; *CEA*, carcinoembryonic antigen.

* Malignancy on cytology is defined as cancer, atypical cells, or neuroendocrine tumor diagnosed on a cytologic specimen.

Table 2

A comparison of prior and index EUS-guided FNA results in patients with repeat EUS-guided FNAs

	Prior FNA	Index FNA	P value
Age, mean ($\pm$ SD), y	68.2 (11.6)	69.4 (11.6)	–
Sex, male/female, %	40/60	40/60	–
Multiple cysts, %	55	57	.617
Head/body or tail, %	31/69	31/69	–
Diameter, mean ($\pm$ SD), mm	25.6 (23.3)	24.7 (16.2)	.646
Macro-/microcystic/mixed, %	97/0/3	94/0/6	.480
Multilocular/unilocular	77/23	75/25	.617
Connection to PD on EUS, %	24/76	33/67	.013
Mural nodule, %	6	9	.450
Needle size, 19/22 gauge, %	9/91	20/80	.027
Amount of fluid	1 (0.2-760)	1 (0.2-57)	.548
Viscous cyst fluid, %	36	33	1.000
CEA, ng/mL, median (range)	118 (0.5-27540)	94.8 (0.3-6147)	.286
CEA >192 ng/mL, %	39	33	.332
Amylase, median (range), IU/L	800 (8-800)	800 (<5-800)	.316
Cytology test performed, %	75	76	1.000
Malignancy on cytology, %	2	2	1.000
Mucin on cytology	10	11	1.000

SD, Standard deviation; –, not applicable; *PD*, pancreatic duct; *CEA*, carcinoembryonic antigen.

Table 3

Cases with changes in CEA classifications

	Age	Sex	Multiple cysts	H/BT	Diameter, prior; index, mm	Macroscopic	Multilocular	Connection to PD	Mural nodule prior; index	CEA, prior; index, ng/mL	Interval, d
Increased CEA level											
Case 1	83	M	No	BT	20.3, 16.1	Yes	Yes	No	No, no	130, 308	98
Case 2	75	F	Yes	BT	18.5, 23.1	Yes	Yes	Yes	No, no	158, 335	280
Case 3	68	F	Yes	BT	10.2, 9.0	Yes	No	No	No, no	135.6, 446	437
Case 4	79	M	No	H	24.1, 23.6	Yes	Yes	No	No, yes (5.7 mm)	176.5, 378	99
Case 5	76	F	No	H	20.4, 23.5	Yes	Yes	Yes	No, no	83.6, 643	364
Case 6	82	F	Yes	H	25.0, 18.0	Yes	No	No	No, no	37.3, 294	429
Decreased CEA level											
Case 1	81	M	No	H	20, 20.4	Yes	Yes	Yes	No, no	837, 28	107
Case 2	65	M	No	BT	16.6, 19.0	Yes	Yes	No	No, no	366, 105	91
Case 3	73	M	Yes	BT	27.5, 31.0	Yes	Yes	Yes	6.4 mm, 6.4 mm	2140, 134	280
Case 4	78	F	No	H	17.0, 21.0	Yes	Yes	Yes	No, no	222, 137.2	716
Case 5	70	F	Yes	H	15.1, 15.0	Yes	Yes	No	Yes (3.9 mm), no	4675, 20	93
Case 6	50	F	No	BT	10.3, 10.9	Yes	Yes	No	No, no	382, 138	105
Case 7	64	F	Yes	BT	14.3, 13.3	Yes	Yes	Yes	No, no	272, 167.2	77
Case 8	60	M	No	BT	16.0, 16.3	Yes	Yes	No	No, no	514, 107	220
Case 9	67	M	No	BT	10.0, 12.0	Yes	No	Yes	7.0 mm, 10.0 mm	247, 88.1	116
Case 10	75	M	No	BT	10.8, 16.8	Yes	Yes	No	No, no	202, 94.8	273
Case 11	82	F	Yes	BT	37.5, 34.6	Yes	Yes	Yes	No, no	27540, 44.4	213

H, Head; BT, body + tail; PD, pancreatic duct; CEA, carcinoembryonic antigen; M, male; F, female.