

Title:

Endoscopic ultrasound (EUS) guided fine needle biopsy (FNB) with the Procore™ needle provides inadequate material for the histological diagnosis of early chronic pancreatitis

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OR 5164

Endoscopic ultrasound (EUS) guided fine needle biopsy (FNB) with the Procore™ needle provides inadequate material for the histological diagnosis of early chronic pancreatitis

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CONFLICT OF INTEREST

Julio Iglesias-García has acted as a speaker for Pentax Medical. J. Enrique Domínguez-Muñoz has acted as an advisor for Pentax Medical. The needles for EUS-guided FNB used in this study were provided by Cook-Medical at no cost. The other authors have no conflicts of interest to disclose.

ABSTRACT

Background: diagnosis of early chronic pancreatitis (CP) is hampered due to the low accuracy of current imaging techniques and the absence of methods for histological confirmation. We aimed to evaluate the efficacy of endoscopic ultrasound-guided fine needle biopsy (EUS-FNB) for the histological diagnosis of early CP.

Methods: a prospective, cross-sectional, single-center study was designed. Consecutive patients referred for EUS with a clinical suspicion of CP were evaluated for inclusion into the study. Inclusion criteria were age > 18 years and indeterminate EUS findings for the diagnosis of CP according to the Rosemont classification. EUS-FNB of the body of the pancreas was performed with Procore™ needles. Tissue samples were immersed into a methanol-based buffered preservative solution for cytohistological evaluation. The quality of the samples obtained and the histological findings were evaluated. Procedure-related complications were recorded.

Results: the study was stopped after eleven patients were included due to safety concerns and poor diagnostic yield. The mean age of the patients was 50.3 years (range 33-70 years) and six were male. Samples were of poor quality in five cases, but were sufficient for cell-block evaluation. An inflammatory infiltration with mild fibrosis was identified in two cases and neither inflammatory infiltration nor fibrosis was identified in three cases. With regard to the other six cases, isolated inflammatory cells were observed in one case, although the cellularity was poor and unsuitable for cytological evaluation in five cases. There was one major complication (9.1%) of acute pancreatitis that required hospitalization for 48 hours.

Conclusion: EUS-FNB is technically feasible in patients with EUS findings categorized as indeterminate for a CP diagnosis. However, the diagnostic yield is poor and there is a non-negligible risk of complications.

Key words: Tissue acquisition. Chronic pancreatitis. Endoscopic ultrasound.

INTRODUCTION

Chronic pancreatitis (CP) is defined as the presence of chronic inflammation of the pancreas, fibrosis and loss of function of the parenchymal cells (1,2). Diagnosis of CP is usually easy during late stages of the disease. Severe morphological changes such as pancreatic calcifications, atrophy and a disturbed morphology of the pancreatic duct can be demonstrated by routine imaging techniques. These include computer tomography (CT), magnetic resonance imaging (MRI), transabdominal ultrasound and endoscopic ultrasound (EUS) (3,4). However, these methods are not sensitive enough

for the detection of early changes that occur in CP. In addition, mild morphological changes that may be detected are not specific to CP, thus making early diagnosis a challenge (4,5).

A safe and simple method for the acquisition and evaluation of pancreatic tissue in patients with suspected CP could be useful for an early diagnosis of CP, as well as for differentiating this condition from other pancreatopathies (6,7). A histological grading of pancreatic inflammation and fibrosis has the additional potential benefit of being used as an endpoint in future therapeutic trials. The implementation of tissue acquisition via EUS in solid pancreatic lesions has had a major impact on the diagnosis and subsequent management of pancreatic cancer (8-12). Moreover, EUS-guided fine needle aspiration (FNA) or biopsy (FNB) is safe and reliable (13-15). Data on the role of EUS-FNB in early CP are scarce (16).

Different types of needles for EUS-FNB have been developed over the last few years. The Procore™ needle (Cook Endoscopy, Winston-Salem, NC) provides adequate samples for the histological evaluation of different lesions, including solid pancreatic tumors (17-19).

The aim of the present study was to evaluate the feasibility, safety and sample quality of a pancreatic EUS-FNB performed with standard Procore™ needles in patients with a clinical suspicion of CP and minimal changes of the disease on EUS.

MATERIAL AND METHODS

Study design

This was a pilot, prospective, cross-sectional, single-center study.

Patients

Patients referred to the Endoscopy Unit of the Gastroenterology Department, Hospital Universitario de Santiago de Compostela in Spain, due to epigastric pain and the clinical suspicion of CP were eligible for inclusion into the study. Additional inclusion criteria were age > 18 years and EUS findings classified as “indeterminate for CP” according to the Rosemont classification (20). Exclusion criteria included the following: prior pancreatic or upper-GI surgery, thrombocytopenia (platelet levels < 100,000/mm³

), anemia (Hb < 10 g/dl), advanced heart disease, chronic obstructive pulmonary disease, chronic liver disease with portal hypertension, known systemic malignancy, coagulopathy (international normalized ratio ≥ 1.3), use of anticoagulants, inability to discontinue platelet aggregation inhibitors, acute pancreatitis during the previous twelve weeks, active infection or fever during the previous seven days, pregnancy, breast-feeding or the inability to provide informed consent. Complications such as pain, bleeding and acute pancreatitis, were monitored in-hospital for 24 hours after the procedure and seven days thereafter via a phone call.

Endoscopic ultrasound (EUS) and tissue acquisition

EUS was performed using the slim linear echoendoscope (EG-3270-UK; Pentax Europe GmbH, Hamburg, Germany) and the HITACHI-Preirus platform (Hitachi Medical Systems Europe, Zug, Switzerland). All procedures were performed under anesthesiologist-guided propofol sedation by two experienced endosonographers (JIG and JLN), with more than 2,500 and 1,200 EUS-guided FNA/FNB procedures performed respectively at the time of the study. The Rosemont criteria for the diagnosis of CP were evaluated and the presence of vessels that may impede EUS-guided puncture was investigated using the doppler function. EUS-guided tissue acquisition from the body of the pancreas was performed from the stomach with 19, 22 and 25 gauge Procore™ histological needles (Wilson Cook Procore Echotip; Cook Endoscopy, Winston-Salem, NC) with the stylet in place. The selection of the needle size was left to the discretion of the endosonographer. Special attention was given to avoid puncture of the main pancreatic duct (Fig. 1). When the needle was in place within the pancreas, the stylet was pushed in order to expel any mucosal tissue and was then removed. A 10-ml syringe was attached to the hub of the needle to apply a negative suction. Five to ten to-and-fro movements were performed within the pancreas. The needle was withdrawn into the sheath, followed by the withdrawal of the entire system from the biopsy channel. The specimen was expelled into a methanol-based buffered preservative solution (ThinPrep® CytoLyt solution, Hologic, Marlborough, MA) by flushing the needle with 2-3 ml of the buffer solution. Tissue samples were sent immediately for pathological evaluation.

After each biopsy, the pancreatic parenchyma was observed by real-time EUS for at least one minute for any evidence of hemorrhage. In the case that hemorrhaging was observed, the period of observation continued until its resolution. Patients were hospitalized for 24 hours after completion of the biopsy and discharged thereafter provided that there were complications. Patients were called seven days later in order to control any clinically relevant late complications.

Cytohistological evaluation

Samples were processed at the Department of Pathology for histological evaluation. Tissue was embedded in paraffin and stained with hematoxylin-eosin. Samples were processed as a cell block for cytological evaluation when a tissue core was not obtained. All samples were analyzed by one expert pathologist with more than 1,500 pancreatic samples from EUS-guided tissue acquisition evaluated. The quality of the samples was classified as: a) non-diagnostic, no pancreatic tissue or pancreatic cells; b) scant pancreatic tissue, only suitable for cytological evaluation; and c) a pancreatic core sample suitable for histological evaluation. Pathological diagnostic criteria for CP were scored based on a modification of pathological classification proposed by Amman et al. (21). The degree of fibrosis (1, mild; 2, moderate; and 3, severe) and the degree of lymphocyte and plasma cell infiltration (1, mild; 2, moderate; and 3, severe) were evaluated. Figure 2 shows an example of the sample obtained.

Statistical analysis

A descriptive analysis of the data was performed. Data are shown as mean or median and percentage, as appropriate. Based on a prevalence of chronic pancreatitis of 50 cases per 100,000 inhabitants, one third will have indeterminate changes of the disease on EUS. The expected proportion of adequate tissue samples was 85%, therefore a sample size of 33 patients was calculated for a confidence level of 0.95 and a precision of 10%.

Ethical aspects

The study was approved by the local ethical committee and conducted in accordance with the Declaration of Helsinki and its amendments and the Good Clinical Practice guidelines. All patients provided written informed consent for the study. Register code of the ethical committee of Galicia: 2011/281.

RESULTS

The investigators decided to interrupt the study after the inclusion of eleven patients due to the poor yield of EUS-FNB and the development of a major complication in one patient (complication rate of 9.1%). The mean age of patients was 50.3 years (range 33-70 years), six were male. Four and three Rosemont features of CP were observed via EUS in nine and two patients respectively. Eight patients were mild drinkers and smokers, whereas the remaining three patients had no risk factor for CP (4,22).

The EUS-guided tissue acquisition procedure was considered to be feasible in all cases. A 19 gauge needle was used in four cases, 22 gauge in three and 25 gauge in four cases. The general impression of the endosonographers was the fact that the softness of the pancreatic parenchyma made it difficult to puncture and move the needle inside the parenchyma. A tissue core was not obtained in any of the cases. Microscopic evaluation demonstrated a generally poor sample quality. Scant pancreatic cells that were sufficient for cell-block evaluation was obtained in five cases; three cases punctured with a 19-gauge needle and two cases with a 25-gauge needle. Inflammatory infiltration with mild fibrosis was found in two cases; one punctured with the 19-gauge needle and one, with the 25-gauge needle. Thus, supporting the diagnosis of CP (Fig. 2). In the remaining three cases, neither inflammatory infiltration nor fibrosis was detected and thus a CP diagnosis could not be confirmed. Among the remaining six patients, inflammatory cells but no pancreatic cells were observed in one case punctured with the 19-gauge needle. However, the cellularity of samples was poor and did not allow for a cytological evaluation in the other five cases; three punctured with a 22-gauge needle and two, with a 25-gauge needle. The results are summarized in table 1.

As previously mentioned, one patient with three Rosemont features for a CP diagnosis developed mild acute pancreatitis after the procedure that required hospitalization for

48 hours. There were no other complications related to the EUS-FNB.

DISCUSSION

This study shows that EUS-guided tissue acquisition with Procore™ needles in patients with indeterminate EUS findings for a CP diagnosis is technically feasible, but provides inadequate material for cytohistological analysis. It was not possible to obtain a core biopsy for histological evaluation in any of the patients. Even though both endosonographers and the pathologist involved in this study are experts in obtaining and processing EUS-guided pancreatic tissue samples, adequate samples for cytological evaluation were only obtained in five of the patients (45.4%). Furthermore, the development of one major complication of acute pancreatitis in one case questions the safety of this procedure in patients with suspected early CP. Solid pancreatic lesions usually have dense fibrosis and the safety of EUS-guided tissue acquisition in this scenario is well documented (23-25). However, the cases in the current study represent a different risk category. Until the safety and efficacy of EUS-guided FNB in mildly altered pancreas is well documented, ideally in animal studies, EUS-guided biopsy should be avoided in these patients, even for research purposes.

The diagnosis of CP is a clinical challenge, mainly during early stages of the disease (1,4,5). Chronic pancreatitis is characterized by the presence of a chronic infiltration of inflammatory cells and the progressive loss of acinar cells which are replaced by fibrosis. However, due to the absence of histology, the diagnosis of chronic pancreatitis relies on the demonstration of morphological abnormalities suggestive of pancreatic fibrosis (20,26,27). This makes the diagnosis at an early stage of the disease challenging, as well as the differential diagnosis with regard to similar conditions like pancreatopathy (7). In addition, the lack of histological analysis renders the appropriate evaluation of any potential new therapy of the disease difficult.

EUS-guided FNA and FNB are safe and accurate techniques for the cytohistological evaluation of solid pancreatic lesions (13-15,24,25,28). The use of reverse-beveled Procore™ needles provides a high diagnostic yield in this setting (17-19,29,30). However, data with regard to the cytohistological diagnosis of chronic pancreatitis are scarce. We previously reported on the accuracy of EUS-guided FNA for the

cytohistological evaluation of moderate to severe chronic pancreatitis (31). DeWitt et al. obtained an appropriate tissue sample for a histological diagnosis of chronic pancreatitis in only one of nine patients with mild chronic pancreatitis (16). In the present study, we also failed to obtain adequate pancreatic tissue by EUS-guided FNB for the evaluation of indeterminate EUS findings for a diagnosis of chronic pancreatitis. The needles used in the DeWitt' study and our study were different (QuickCore® and Procore™ needles, respectively), even though the diagnostic yield was similar. Difficulties to obtain a core biopsy was probably related to the softness of the pancreatic parenchyma in early chronic pancreatitis.

Furthermore, the accuracy of a histological diagnosis of chronic pancreatitis based on single biopsies may not be high due to the patchy distribution of the disease (32). However, since the FNB is guided by EUS to target the most abnormal looking area, this cannot be confirmed as appropriate needles to obtain core biopsies from the pancreatic parenchyma are not available. Finally, there are still safety concerns related with EUS-FNB. In the present study, one case developed acute pancreatitis after the procedure. This, together with the limited success of the technique led to the study being interrupted prematurely.

The strengths of the current study include the prospective design with a consecutive inclusion of patients. During the study period, no patient that fulfilled inclusion and exclusion criteria was excluded. The skills and experience of endosonographers and the pathologist involved in the study should also be highlighted. The open study design may be considered as a limitation.

In conclusion, EUS-guided FNB for the histological diagnosis of chronic pancreatitis in patients with indeterminate EUS findings of the disease is technically feasible. However, the yield is poor. In addition, the safety of this technique is also a concern. Based on the present data, EUS-FNB for mild chronic pancreatitis should be avoided in humans until the efficacy and safety of this technique has been demonstrated in animal studies.

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Accepted Article

Table 1. Results of EUS-FNB in patients with indeterminate EUS findings for a diagnosis of chronic pancreatitis

<i>Case number</i>	<i>Number of EUS criteria</i>	<i>Needle size</i>	<i>EUS-FNB result</i>	<i>Complications</i>
1	4	19G	Scant pancreatic tissue. No inflammation. No fibrosis	No
2	4	19G	Scant pancreatic tissue. No inflammation. No fibrosis	No
3	4	19G	Scant pancreatic tissue. Inflammatory infiltration. Mild fibrosis	No
4	4	19G	Scant inflammatory cells	No
5	4	22G	Poor cellularity	No
6	4	22G	Poor cellularity	No
7	4	22G	Poor cellularity	No
8	4	25G	Scant pancreatic tissue. No inflammation. No fibrosis	No
9	3	25G	Poor cellularity	No
10	3	25G	Poor cellularity	Acute pancreatitis
11	4	25G	Scant pancreatic tissue. Inflammatory infiltration. Mild fibrosis	No



Fig. 1. EUS image of the pancreatic parenchyma accessed for EUS guided tissue acquisition.

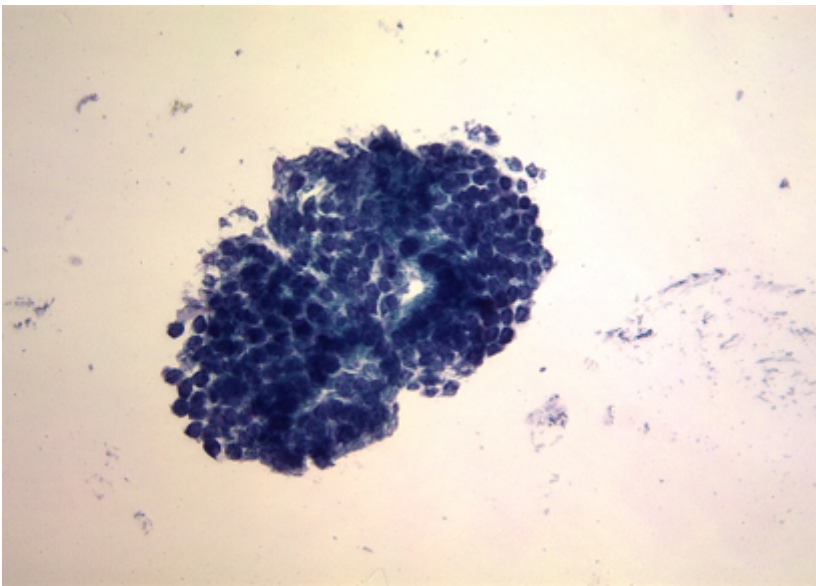


Fig. 2. Cytohistological image obtained by EUS guided tissue acquisition with a 19 gauge Procore™ needle.