

Prospective Study

Can the wet suction technique change the efficacy of endoscopic ultrasound-guided fine-needle aspiration for diagnosing autoimmune pancreatitis type 1? A prospective single-arm study

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Abstract

BACKGROUND

Other than surgery, endoscopic ultrasound-guided fine-needle aspiration (EUS-FNA) is the only procedure for histologically diagnosing autoimmune pancreatitis (AIP). However, adequate specimens are difficult to obtain. Recently, more adequate specimens were reported to be obtained with EUS-FNA with a wet suction technique (WEST) than with conventional EUS-FNA.

AIM

To histologically diagnose AIP by EUS-FNA with a WEST.

METHODS

Eleven patients with possible type 1 AIP between February 2016 and August 2018 underwent EUS-FNA with a WEST (WEST group), with four punctures by 19 or 22 G needles. As a historical control, 23 type 1 AIP patients who underwent no fewer than four punctures with 19 or 22 G needles were enrolled (DRY group). Patient characteristics and histological findings were compared between the two groups.

RESULTS

Three histopathological factors according to the International Consensus

approved by the Institutional Review Board of Fukushima Medical University.

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Informed consent statement: All study participants, or their legal guardian, provided written consent prior to study enrollment.

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Diagnostic Criteria were significantly greater in the WEST group than the DRY group [lymphoplasmacytic infiltrate without granulocytic infiltration: 9 (81.8%) vs 6 (26.1%), $P = 0.003$, storiform fibrosis: 5 (45.5%) vs 1 (4.3%), $P = 0.008$, abundant (> 10 cells/HPF) IgG4-positive cells: 7 (63.6%) vs 5 (21.7%), $P = 0.026$]. Level 1 or level 2 histopathological findings were observed more often in the WEST group than in the DRY group [8 (72.7%) vs 3 (13.0%), $P = 0.001$].

CONCLUSION

EUS-FNA with a WEST was more successful than standard EUS-FNA in histologically diagnosing AIP.

Key words: Autoimmune pancreatitis; Endoscopic ultrasound-guided fine needle aspiration; Wet suction technique

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Core tip: Recently, more adequate specimens were reported to be obtained with endoscopic ultrasound-guided fine-needle aspiration (EUS-FNA) with a wet suction technique (WEST) than with the conventional method (DRY) of EUS-FNA. This study aimed to histologically diagnose autoimmune pancreatitis (AIP) by EUS-FNA with a WEST. Patient characteristics and histological findings were compared between the WEST group and the DRY group. Level 1 or level 2 histopathological findings were observed more often in the WEST group than in the DRY group. EUS-FNA with a WEST was more useful than standard EUS-FNA for histologically diagnosing AIP.

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INTRODUCTION

Autoimmune pancreatitis (AIP) was determined by Yoshida *et al*^[1]. Some autoimmune pathogenesis caused pancreatitis with pancreatic enlargement, pancreatic duct narrowing, or invasion of lymphocytes with fibrosis. Hamano *et al*^[2] described elevated serum IgG4 in AIP patients. The 2010 International Consensus Diagnostic Criteria (ICDC) for AIP defined pancreatitis with other organ involvement and elevated serum IgG4 as "type 1"; lymphoplasmacytic sclerosing pancreatitis (LPSP) was the most prominent histological characteristic^[3]. Four items were mentioned as being significant to histologically diagnose LPSP. These items are periductal lymphoplasmacytic infiltrate without granulocytic infiltration, obliterative phlebitis, storiform fibrosis, and abundant (> 10 cells/HPF) IgG4-positive cells. If three of those four items are observed, that is defined as level 1 histological findings. If two items are observed, it is defined as level 2 histological findings.

AIP can be diagnosed by imaging characteristics and elevated serum IgG4 or by other criteria. Nevertheless, level 1 pancreatic histological findings of the ICDC are necessary to histologically diagnose AIP type 1. Other than surgical biopsy, endoscopic ultrasound-guided fine needle aspiration (EUS-FNA) is the only procedure that can histologically diagnose AIP. However, it is very hard to acquire a sufficient specimen via EUS-FNA^[4-6]. The ICDC recommended using EUS-FNA for ruling out malignancy before a diagnostic steroid administration^[3]. In 2017, Sugimoto *et al*^[7] reported that the clinical characteristics of AIP differed from those of pancreatic cancer and that malignancy was ruled out by EUS-FNA in AIP patients.

However, recently, with a wet suction technique (WEST) of EUS-FNA, more adequate specimens were reported to be obtained than by the conventional method of EUS-FNA^[8]. Therefore, we hypothesized that more AIP patients could be histopathologically diagnosed by WEST EUS-FNA. The aim of our study is to clarify the efficacy of EUS-FNA with a WEST for diagnosing AIP type 1.

MATERIALS AND METHODS

Study design and ethics

This study was a single arm prospective study intended to clarify the efficacy of WEST EUS-FNA for diagnosing type 1 AIP. This study was performed at Fukushima Medical University. The Institutional Review Board of Fukushima Medical University approved this study. All patients agreed to participate in this study. This trial was registered in UMIN (ID: 000019768).

Patients

We recruited 11 patients who were suspected to have type 1 AIP at Fukushima Medical University between February 2016 and August 2018. They showed diffuse or focal pancreatic swelling on abdominal enhanced computed tomography with elevated serum IgG4 levels (> 135 mg/dL). To prove the efficacy of WEST EUS-FNA for diagnosing AIP, historical controls were used. The historical controls included 42 AIP patients who were diagnosed at Fukushima Medical University between July 2003 and July 2018 before the initiation of this study. Among these 42 patients, 35 had elevated IgG4 levels (≥ 135 mg/dL). In this study, WEST EUS-FNA was performed with four punctures using 19 or 22 G needles. The gauge of needle used during EUS-FNA was randomly chosen by each endoscopist. Therefore, 23 type 1 AIP patients who underwent procedures with no fewer than four punctures with 19 or 22 G needles were selected as the control group (Figure 1). The targets of this study were termed the WEST group. On the other hand, historical controls were termed the DRY group. The targets and historical controls were all diagnosed with type 1 AIP according to the ICDC.

Method of WEST EUS-FNA

All procedures were performed under the guidance of a professional endoscopist who was well versed in EUS-FNA (TT) and who had performed no less than 1500 EUS-FNA procedures. After a patient was well sedated with intravenous midazolam, an echoendoscope was inserted. The swollen pancreas was viewed on the monitor, and the vessels along the puncture line were confirmed by color doppler echo imaging. Then, the puncture needle was prepared for WEST. The EUS-FNA with a WEST was performed according to the methods described in a previous report by Attam *et al*^[8]. First, the stylet was removed from the needle, and saline solution was injected until the needle was filled (Figure 2A). After a suction syringe was loaded to 20 mL of suction in a locked position, the syringe was set at the edge of the needle without an extension tube (Figure 2B). The needle was inserted into the target lesion, and the lock of the suction syringe was opened. Saline solution flowed into the suction syringe because of the negative pressure (Figures 2C and D), and the needle was moved back and forth 20 times per puncture. The punctures were performed 4 times according to the instructions in the past report by Suzuki *et al*^[9]. In patients with focal pancreatic swelling, samples were obtained from the swollen tissue. In patients with diffuse pancreatic swelling, samples were obtained at the farthest point from the puncture. If the target was the pancreatic head, a transduodenal puncture was performed. On the other hand, if the target was a pancreatic body or tail, the transgastric approach was performed.

In the WEST group, the echoendoscope used was GF-UCT260 or GF-UC240AL-5 (Olympus Medical Systems, Tokyo, Japan). The ultrasonography apparatus was EU-ME2 (Olympus Medical Systems). The biopsy needles were Expect 22 or 19 G (Boston Scientific, MA, United States). The biopsy needles were randomly selected by the endoscopists.

In the DRY group, the echoendoscope used was GF-UCT260, GF-UCT240AL-5, or GF-UC240AL-5 (Olympus Medical Systems, Tokyo, Japan). SSD5000 (ALOKA, Tokyo, Japan), EU-ME2, or EU-ME1 (Olympus Medical Systems) was used as the ultrasonography apparatus. The biopsy needles were Expect 22 or 19 G (Boston Scientific, MA, United States); EZ Shot 22 G or NA11J-KB (Olympus Medical System); or EchoTip 19, 22, EchoTip ProCore 19 G, or Quick-Core 19 G (Cook Medical Inc., NC, United States).

Examination items

The primary endpoint was the level 1 or level 2 histopathological finding according to the ICDC^[3]. Biopsy of the pancreatic duct by EUS-FNA is difficult; therefore, an item on the ICDC such as "periductal lymphoplasmacytic infiltrate without granulocytic infiltration" was replaced with "Lymphoplasmacytic infiltrate without granulocytic infiltration" according to the Clinical Diagnostic Criteria for AIP 2011 of Japan^[10]. The secondary outcomes were each of the four items involved in the ICDC histopathological findings, namely, lymphoplasmacytic infiltrate without granulocytic

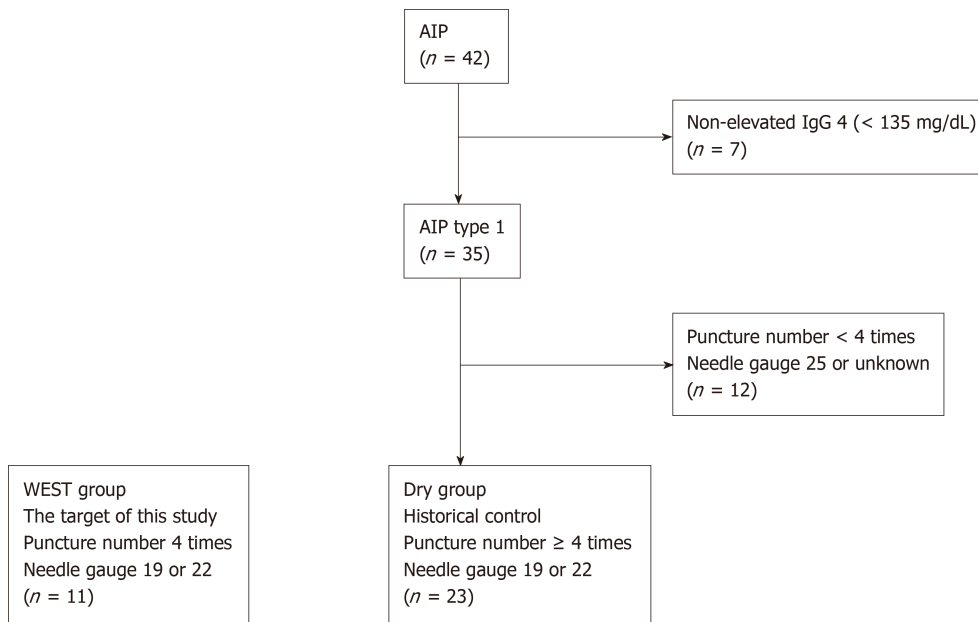


Figure 1 Target patients in this study and historical controls. The historical controls were 23 autoimmune pancreatitis patients who underwent endoscopic ultrasonography-guided fine-needle aspiration with no fewer than 4 punctures by 19 or 22 G needles. The target subjects in this study underwent endoscopic ultrasonography-guided fine-needle aspiration with a wet suction technique. The historical controls were considered the DRY group. AIP: Autoimmune pancreatitis; WEST: Wet suction technique.

infiltration, obliterative phlebitis, storiform fibrosis, and abundant (> 10 cells/HPF) IgG4-positive cells). All pathological diagnoses were performed by more than two pathologists.

Patient characteristics (age, sex, type of pancreatic swelling, serum level of IgG4), items related to EUS-FNA (19 or 22 G needle, number of needle passes, histology, or adverse events), primary outcome and secondary outcomes were compared between the WEST group and the DRY group.

Sample size

The results of conventional EUS-FNA for AIP type 1 patients in our hospital indicated that level 1 or level 2 histopathological findings were observed in 13% (3/23) of patients. The results of WEST EUS-FNA were expected to be 60%, which was better than a previous multicenter study^[11]. Eleven patients were needed in this study for an α error of 0.05 and statistical power of 0.8.

Statistical analysis

Age was compared by Student's *t* test. Serum IgG4 level and the number of needle passes were compared using the Mann-Whitney *U* test. Nominal variables were compared by Fisher's exact test. A *P* value < 0.05 was defined as statistically significant. All statistical analyses were performed using EZR (Saitama Medical Centre, Jichi Medical University, Saitama, Japan), which is a graphical user interface for R (The R Foundation for Statistical Computing, Vienna, Austria)^[12].

RESULTS

No significant differences were observed in patient characteristics between the WEST and DRY groups except for sex (Table 1). Females were significantly more common in the WEST group than in the DRY group (male/female, WEST group 6/5, DRY group 21/2, $P = 0.024$).

Regarding the comparison of procedures and results related to EUS-FNA, several items were significantly different between the two groups (Table 2). The number of needle passes was significantly lower in the WEST group than in the DRY group [median (range), 4 (4-4) vs 5 (4-9), $P < 0.001$]. In terms of the histopathological findings according to the ICDC, three items were observed to be significantly greater in the WEST group than in the DRY group [lymphoplasmacytic infiltrate without granulocytic infiltration: 9 (81.8%) vs 6 (26.1%), $P = 0.003$, storiform fibrosis: 5 (45.5%) vs 1 (4.3%), $P = 0.008$, abundant (> 10 cells/HPF) IgG4-positive cells: 7 (63.6%) vs 5 (21.7%), $P = 0.026$]. Level 1 histopathological findings were observed more in the

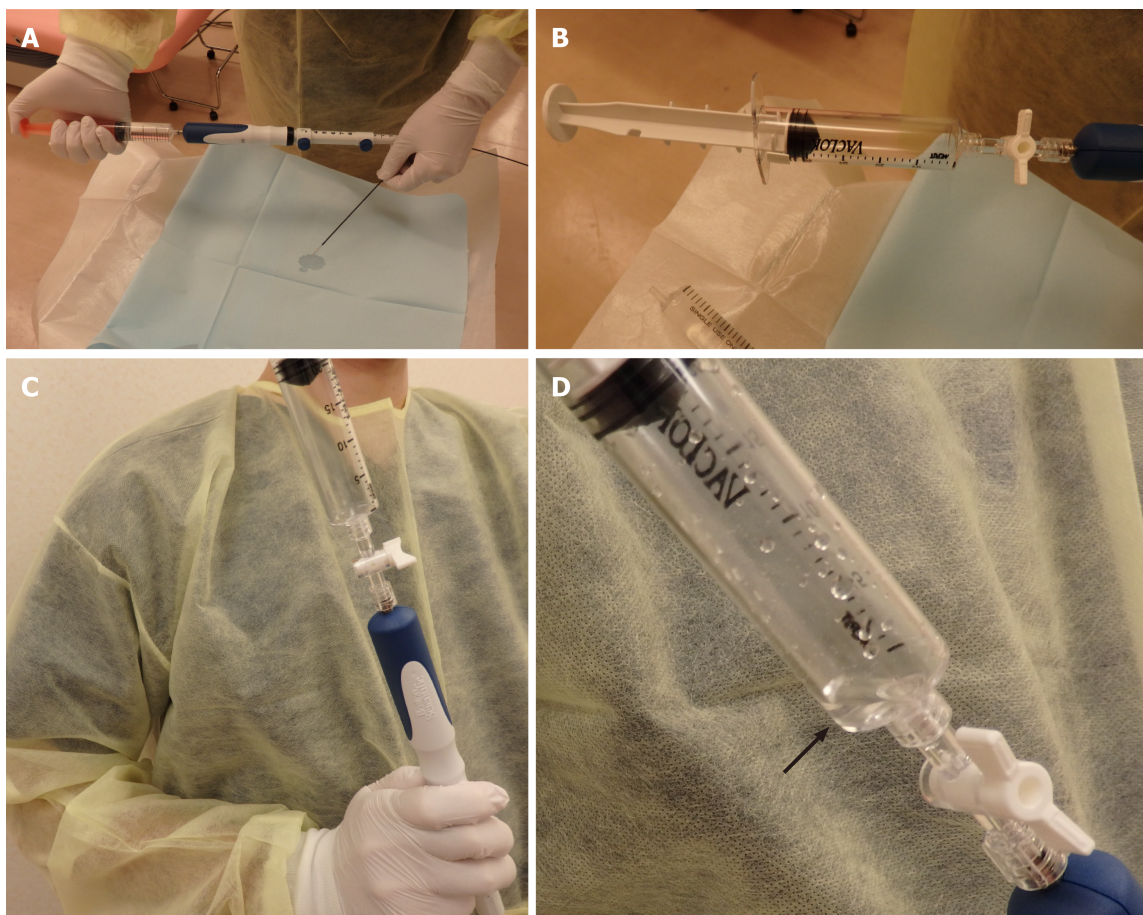


Figure 2 The wet suction technique method of endoscopic ultrasonography-guided fine-needle aspiration. A: After the stylet of the needle was withdrawn, saline solution was injected into the needle; B: A locked suction syringe with 20 mL of negative pressure was set at the edge of the needle; C: The needle was used to puncture the target lesion; D: then, the lock on the syringe was opened. Saline solution flowed into the suction syringe due to the negative pressure.

WEST group than in the DRY group [4 (36.4%) *vs* 1 (4.3%), $P = 0.029$]. Level 1 or level 2 histopathological findings were observed more often in the WEST group than in the DRY group [8 (72.7%) *vs* 3 (13.0%), $P = 0.001$]. Adverse events were not observed in either group.

In **Figure 3**, a representative case of a patient diagnosed with AIP by WEST EUS-FNA is shown.

DISCUSSION

In this study, we investigated the efficacy of WEST EUS-FNA for diagnosing AIP. Three histopathological items according to the ICDC were observed more frequently in the WEST group than in the DRY group. Level 1 histopathological findings and level 2 histopathological findings were observed more in the WEST group than in the DRY group.

Regarding EUS-FNA for AIP, the results of procedures performed with standard needles and more complex needles were reported. Regarding previous reports using standard needles, Iwashita *et al*^[13] reported that 17 of 44 AIP patients could be diagnosed by EUS-FNA with a 19 G needle. The reports using a 22 G needle are described below. Ishikawa *et al*^[4] diagnosed LPSP in 9 of 47 AIP patients. Imai *et al*^[5] could not histopathologically diagnose AIP. Interestingly, a multicenter study found that the diagnosability of AIP by EUS-FNA was poor^[6], and another multicenter study reported that 57.7% of patients were diagnosed with level 2 or higher based on the histopathological findings^[14].

Second, some previous reports involved more complex needles. Mizuno *et al*^[15] reported that EUS Tru-Cut biopsy (EUS-TCB) resulted in a diagnosis of LPSP in 45.5% of cases. In a report by Kanno *et al*^[11], EUS-FNA using 22 G automated spring-loaded PowerShot needles resulted in a diagnoses of ICDC level 1 and 2 histological findings in 56% and 24% of AIP cases. The histopathological diagnosability of EUS-FNA for

Table 1 Comparisons of patient characteristics between the wet suction technique group and the DRY group

	WEST group (n = 11)	DRY group (n = 23)	P value
Age (yr, means \pm SD)	62.9 \pm 12.4	61.0 \pm 9.6	0.626
Sex, male/female	6/5	21/2	0.024
Pancreatic swelling type, diffuse/focal	6/5	11/12	1.0
Serum IgG4 level [mg/dL, median (range)]	568 (177-2100)	447 (149-1480)	0.663

WEST: Wet suction technique.

AIP was improved by needle choice; however, it was difficult to statistically compare the sensitivity and accuracy between the standard needles and more complex needles.

On the other hand, the histopathological accuracy by WEST EUS-FNA was statistically superior to that by the standard EUS-FNA method or EUS-TCB in this study (seven historical control cases underwent EUS-TCB). Moreover, the positive result of this study was achieved with standard needles. Though the reasons underlying the increased size of the specimens collected by WEST are not clear, it is thought that the saline solution coating the lining of the needle leads to better transmission of the applied suction or that the saline solution acts as a stylet, reducing the contamination from GI tissue^[8].

In recent reports, EUS-FNA using a 22 G SharkCore needle was effective for histologically diagnosing AIP^[16,17]. By the development of puncture methods such as WEST or special puncture needles, the diagnosability of AIP by EUS-FNA will be improved.

Some limitations exist in this study. First, a small number of patients at a single institution were involved in this study. Second, historical controls were used as a control group. In the future, a multicenter randomized controlled trial (RCT) with more cases is needed. However, AIP patients are rare, rendering an RCT difficult to conduct. Third, we used EUS-TCB needles such as Quick-Core 19 G or ProCore 19 G (Cook Medical Inc., NC, United States) in the historical control patients. In past reports, these needles were useful for obtaining more specimens^[18,19]. However, these needles were not used in the WEST group. Therefore, they were not a factor in the superiority of the WEST technique.

In conclusion, WEST EUS-FNA was more useful for histologically diagnosing AIP than was standard EUS-FNA.

Table 2 Comparisons of the procedures and results between wet suction technique and DRY techniques for endoscopic ultrasonography-guided fine-needle aspiration, n (%)

	WEST group (n = 11)	DRY group (n = 23)	P value
EUS-FNA needle (19 G/22 G)	1/10	8/15	0.214
Number of needle passes [median (range)]	4 (4-4)	5 (4-9)	< 0.001
Histopathological findings			
Lymphoplasmacytic infiltrate without granulocytic infiltration	9 (81.8)	6 (26.1)	0.003
Obliterative phlebitis	2 (18.2)	0 (0)	0.098
Storiform fibrosis	5 (45.5)	1 (4.3)	0.008
Abundant (> 10 cells/HPF) IgG4 positive cells	7 (63.6)	5 (21.7)	0.026
Level 1 histopathological findings	4 (36.4)	1 (4.3)	0.029
Level 1 or level 2 histopathological findings	8 (72.7)	3 (13.0)	0.001
Adverse events	0 (0)	0 (0)	

WEST: Wet suction technique; EUS-FNA: Endoscopic ultrasonography-guided fine-needle aspiration.

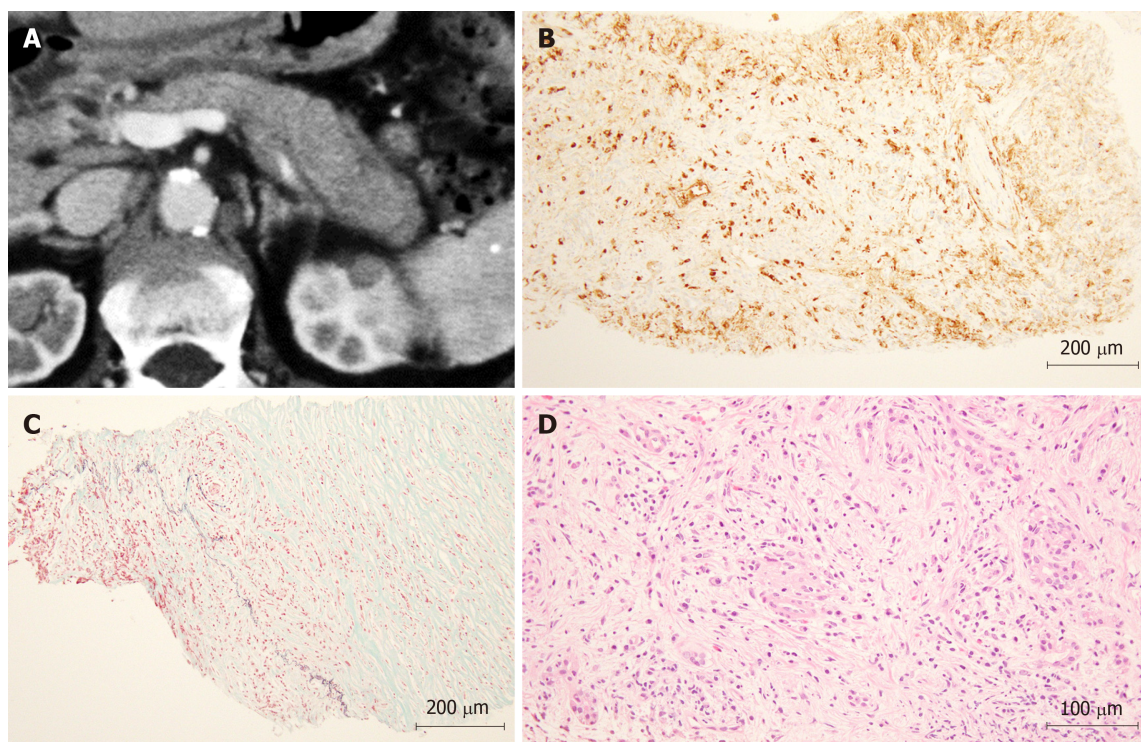


Figure 3 The features of a patient with autoimmune pancreatitis diagnosed by endoscopic ultrasonography-guided fine-needle aspiration with a wet suction technique. A: The pancreatic tail is swollen with a capsule-like rim sign as observed on abdominal computed tomography; B: Specimen acquired by wet suction technique (WEST) endoscopic ultrasonography-guided fine-needle aspiration (EUS-FNA) (HE × 200): IgG4-positive plasma cells were observed; C: Specimen acquired by WEST EUS-FNA (EM × 200): obliterative phlebitis was observed; D: Specimen acquired by WEST EUS-FNA (HE × 400): fibrosis with a storiform pattern of plasma cells was observed.

ARTICLE HIGHLIGHTS

Research background

Apart from surgical biopsy, endoscopic ultrasound-guided fine needle aspiration (EUS-FNA) is the only method used to histologically diagnose autoimmune pancreatitis (AIP) type 1. Nevertheless, it is very difficult to obtain an adequate specimen by EUS-FNA for a histological diagnosis of AIP type 1. However, recently, with EUS-FNA with a wet suction technique (WEST), adequate specimens were reported to be obtained more often than those obtained by conventional EUS-FNA.

Research motivation

EUS-FNA with a WEST could overcome the difficulty of histologically diagnosing AIP type 1.

Research objectives

To investigate the efficacy of EUS-FNA with a WEST for diagnosing AIP type 1.

Research methods

Eleven AIP type 1 patients were recruited for this study and underwent EUS-FNA with a WEST for histological diagnosis. The histological diagnosability was compared between the WEST group and a historical control group that included AIP type 1 patients who underwent conventional EUS-FNA before this study (DRY group).

Research results

Three histopathological features [lymphoplasmacytic infiltrate without granulocytic infiltration, storiform fibrosis, and abundant (>10 cells/HPF) IgG4-positive cells] according to the International Consensus Diagnostic Criteria were significantly greater in the WEST group than in the DRY group. Level 1 or level 2 histopathological findings were observed more often in the WEST group than in the DRY group.

Research conclusions

EUS-FNA with a WEST was more useful than standard EUS-FNA in the histological diagnosis of AIP.

Research perspectives

The histological diagnosability of EUS-FNA for AIP will be developed by the improvement of puncture needles and puncture methods, such as WEST.

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