Efficacy of Endoscopic Ultrasound-Guided Fine Needle Aspiration with and without Stylet and Suction for Cytopathological Diagnosis of Different Solid Lesions

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ABSTRACT

Background: The EUS-guided fine needle aspiration (EUS-FNA) procedure has a low rate of side effects, excellent sensitivity, and specificity, and allows for cytological confirmation of imaging findings. It is thought that the quality and diagnostic sample yield are improved during EUS-guided FNA (EUS-FNA) when a suction syringe and stylet are used. **Objectives:** To compare the diagnostic yield and cytological properties of samples collected by EUS-FNA with and without suction and stylet.

Patients and Methods: 62 individuals who had been recommended for EUS-FNA due to solid upper gastrointestinal lesions participated in this comparative study. With and without a suction syringe and stylet, each lesion was sampled twice. A predetermined set of cytological criteria was used to evaluate the samples' quality.

Results: In this comparative study, 62 patients were prospectively enrolled, and 44 underwent EUS-FNA at the pancreas, 11 at the common bile duct, 5 at the stomach, and 2 at the lymph nodes. With stylet and suction, 29/62 [46.8%] vs. without stylet and suction, 29/62 [46.8%], P = 0.901), interim analysis showed that no difference in the overall diagnostic outcome of malignancy between the specimens collected using the two procedures. Regarding cellularity (P = 0.494), contamination (P = 0.511), and specimen sufficiency (P = 0.471), there was no difference. The no suction, no stylet approach considerably reduced blood contamination (P < 0.0001).

Conclusion: Both techniques offered comparable diagnostic outcomes. However, no suction, no stylet technique showed less blood contamination resulting in a sample of higher quality.

Key Words: Endoscopic ultrasound, fine needle aspiration, stylet, suction.

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INTRODUCTION

Endoscopic ultrasonography (EUS) was first utilized in clinical practice in 1980. It has shown to perform better than computed tomography (CT), endoscopic retrograde cholangiopancreatography (ERCP), and transabdominal ultrasonography in the diagnosis of tiny pancreatic neoplasms.^[1]

For the diagnosis and staging of malignant and benign lesions of the gut wall and associated structures of the mediastinum, abdomen, and pelvis, EUS has become a crucial technique. Additionally, it is employed as a diagnostic tool for identifying pancreatic endocrine tumors, assessing vascular disease, and evaluating submucosal masses of the upper gastrointestinal tract and the rectosigmoid. Interventional uses, such as tissue/fluid sample collection using EUS-guided fine-needle aspiration (EUS-FNA), this imaging technique clinical efficacy and financial viability will probably be improved by using it for pseudocyst draining as well as for administering local therapy. The diagnosis and staging of gastric, esophageal, rectal, and pancreaticobiliary carcinomas is where EUS is most widely used. Clinical management strategy of a considerable number of patients have modified by EUS to a less expensive and hazardous one.^[2]

The usage of EUS has changed from being a tool for diagnosis to a therapeutic treatment for a number of conditions affecting the pancreas, kidneys, retroperitoneum, adrenal glands, and lymph nodes surrounding the gastrointestinal tract. It can be useful in separating benign from malignant lesions and can frequently take the place of surgery.^[3]

Depending on the intervention used, it is conducted as an outpatient operation that can take up to 60 minutes and can be done with sedation or general anesthesia. Depending on the indication, further instruments in the scope may include a core biopsy, fine-aspiration, a neurolysis and a celiac plexus blockage needle, and a metal or plastic stent. EUS is performed using a flexible broad endoscope with a small ultrasonic probe and camera at the tip.^[4]

A plane-transecting ultrasonic image that extends beyond the scope's axis is produced by performing the procedure using either a radial (360°) or a transverse (180°). The first to be created, this type of instrument still serves as the standard for diagnostic imaging. It has a linear view configuration that generates an ultrasonic image in a plane parallel to the scope's axis.^[5]

Compared to other imaging modalities, EUS provides a number of benefits. There is no radiation risk like in CT or positron emission tomography, and magnetic resonance imaging (MRI) contraindications like metal implants or claustrophobia are not present. When paired with Doppler, EUS can give high-resolution real-time imaging for assessing the vasculature.^[6]

PATIENTS AND METHODS:

Study design:

This was a prospective comparative study. It was conducted at Kobry Elkobba Military Medical Complex in the period from October 2021 to June 2022 on 62 patients who undergone EUS-FNA for pancreatic or upper gastrointestinal masses. All patients included in the study were fulfilling the inclusive criteria, ethical approval was obtained from hospital ethical committee and Armed Forces College of Medicine and patient medical consent.

Study population

This study was conducted on 62 patients who were referred for EUS-FNA of upper gastrointestinal solid lesions, which were detected by different imaging techniques. All patients involved in the study were fulfilling the inclusion criteria, ethical approval was obtained from hospital ethical committee and Armed Forces College of Medicine and patient medical consent.

Inclusion criteria:

• The minimum age to participate in the study was 18 years old.

• Both Sexes were eligible for the study.

• All patients were identified as having either pancreatic or upper gastrointestinal lesions and had undergone a

Triphasic CT, dynamic MRI or MRCP before being referred for EUS.

Exclusion criteria:

- Age <18 years.
- Severe coagulopathy (INR >1.5).
- Low platelet count (<50,000),

• Failure to acquire samples from the lesion because of the intervening blood vessels.

• Patient inability to tolerate the procedure.

• Patient management plan would not be altered by EUS-FNA findings.

Procedure technique:

Using linear array echoendoscopes (Pentax EG38UG attached to a Hitachi Avius US machine), an experienced endosonographer performed EUS. The patients were sedated moderately to deeply with intravenous propofol while the endoscopy was performed with the patients lying on their left side. On a case report form, the lesions' location, size (long axis), echogenicity, and lymph node characteristics were noted.

To prevent interfering vessels, EUS-guided sample was carried out using a 22G needle (Boston Scientific[®], USA) under EUS and Color-Doppler guidance. A 20-ml suction syringe attached to the needle's proximal end was used for standard suction after the first sample from each lesion was taken using the no-suction, no-stylet technique. The second sample was taken using the stylet slow pull technique to establish low pressure at initially. Every lesion received two to three passes with a single needle, each including 8 to 15 consistent to and fro motions.

Preparation of cytologic specimen:

The samples were discharged on various, numbered glass slides. All of the slides underwent air drying and Papanicolaou staining. The materials were analyzed by a cytopathologist who was unaware of the technique utilized during staining.

Cytological criteria

The main comparison criteria was based on the data obtained from the laboratory database and cytology reports of the biopsies obtained using both techniques. Then the quality of samples was assessed using the following cytological criteria: **1- Sample Cellularity** (percentage of area of slide that contains cells of the representative lesion):

• No representative cells present.

 \bullet Representative cells present in less than 25% of the slide.

• Representative cells present in 25-50% of the slide.

• Representative cells present in more than 50% area of the slides.

2- Adequacy of specimen:

• Inadequate for histological interpretation (total material is less than $10 \times$ power field in length).

• Adequate histological interpretation (total material is more than a $10 \times$ power field in length).

3- Contamination (Percentage of area of slide that represents GI contamination):

- No contamination seen.
- Contamination present in less than 25% of the slide.
- Contamination present in 25%-50% of the slide.
- Contamination present in more than 50% of the slide.

4- Degree of sample contamination by blood: Was categorized into 3 grades: Minimal, Moderate and Significant.

After the cytological examination, a final conclusion was established and the outcomes were divided into one of the following groups: malignant, suspicious, benign or inadequate for reporting.

Data entry and Statistical analysis:

Data were entered on Microsoft Office Excel Program for Windows and analysis of data was done by IBM computer using SPSS (statistical program for social science version 23) as follows:

• **Description** of quantitative variables as Mean, SD, median and IQR.

• Shapiro test of normality used to check the data distribution.

• **Description** of qualitative variables as number and percentage.

• Chi-square test was used to compare qualitative variables between groups.

• Fisher exact test was used when one expected cell or more are less than 5.

• **Kappa measure of agreement** was used to assess the agreement between both techniques according to the previously mentioned cytological criteria.

P-value: level of significance

P value >0.05 insignificant.

P<0.05 significant.

RESULTS:

In total, the study included 70 patients. 62 of whom met the inclusion criteria. No solid mass lesion was identified, FNA was not recommended in 2 patients, a cystic lesion in 3 patients, and early operation termination due to medical instability in 3 patients are the reasons for exclusion. As Kobry Elkobba Military Medical Complex, the facility where the procedures were done, only provided services to males, all of our patients were males.

Comparison between no stylet, no suction EUS guided FNA technique with the other technique using stylet and suction syringe in sampling of solid lesion in pancreas, stomach, common bile duct or lymph nodes regarding the diagnostic outcome of malignancy, cellularity, adequacy and degree of blood and GI contamination of specimens was done in the period from October 2021 to June 2022.

Descriptive data of the studied patients

Table 1: Mean age of the patients

Age (Mean \pm SD)	59±11	59 (52.7)
	Number	%
Smoking	17	27.40
Diabetic	22	35.50
Hypertensive	15	24.20
Presenting symptom	Ν	%
Abdominal pain	20	32.30
Jaundice	34	54.80
Weight loss	4	6.50
Fever	4	6.50

The most common comorbidities in the patients under study were diabetes mellitus and hypertension, which were present in 22 patients (35.5%) and 15 patients (24.2%), respectively. Jaundice and stomach discomfort were observed to be the presenting symptoms in 20 (32.3%) and 34 (54.8%) of patients, respectively, as shown in the (Table 1).

Outcome data

Table 2: Site of the lesions detected by CT, MRI and MRCP

Lesions detected by CT, MRI and MRCP	Ν	%
Pancreatic mass	52	83.90
Intraabdominal lymph node	2	3.20
Gastric mass	5	8.10
Extrahepatic biliary stricture	3	4.80

Regarding CT data, (100%) of patients had solid lesion detected by CT, MRI and MRCP, (83.9%) of these lesions were pancreatic masses, (8.1%) were gastric masses, (4.8%) were extrahepatic biliary masses and (3.2%) were intraabdominal lymphadenopathies (Table 2-4), (Figure 1-3).



Fig. 1: Site of the lesions detected by CT, MRI and MRCP

Table 3: Site of the lesion detected by EUS

Site of the lesion detected by EUS	N	%
Pancreatic head	30	48.40
Distal CBD	8	12.90
Pancreatic body or tail	14	22.60
Intraabdominal lymph nodes	2	3.20
Proximal CBD	3	4.80
Gastric mass	5	8.10

Of the 62 lesions that undergone EUS FNA (48%) were in the pancreatic head, (22.6%) were in the pancreatic body or tail, (8.1%) were submucosal gastric lesions, (12.9%) were in the distal CBD, (4.8%) were in the proximal CBD and (3.2%) were intraabdominal lymph nodes.

Table 3: Echogenicity of the lesions detected by EUS

Lesions Echogenicity	N	%
Hypoechoic	56	90.30
Isoechoic	6	9.70
Table 4: Size of the lesions		
Size of the lesion (cm) Mean± SD	3.45 ± 1.08	1.6:6.2



Fig. 2: Site of the lesion detected by EUS



Fig. 3: Echogenicity of the lesions detected by EUS

Table 8:	Cytological	characteristics	of	samples	obtained	with
both techi	niques					

Cytological criteria	No suction No stylet	suction and stylet	p value
Cellularity	N(%)	N(%)	0.494
No representative cells	17(27.4)	12(19.4)	
Representative cells < 25%	9(14.5)	13(21)	
Representative cells 25–50%	25(40.3)	22(35.5)	
Representative cells >50	11(17.7)	15(24.2)	
Percentage of area of slide that represents GI contamination	N(%)	N(%)	0.511
No contaminations seen	50(80.6)	44(71)	
Contamination present in < 25%	10(16.1)	12(19.4)	
Contamination present in 25- 50%	1(1.6)	2(3.2)	
Contamination present in >50	1(1.6)	4(6.5)	
Amount of blood	N(%)	N(%)	<0.001

Mild	17(27.4)	4(6.5)	
Moderate	36(58.1)	24(38.7)	
Significant	9(14.5)	34(54.8)	
Adequacy of	N(%)	N(%)	0.471
the specimen			
Adequate	41(66.1)	44(70.9)	
Inadequate	21(33.9)	18(29.1)	
Final diagnosis	N(%)	N(%)	0.901
Malignant	29(46.8)	29(46.8)	
Benign	7(11.3)	9(14.5)	
Suspicious for	8(12.9)	9(14.5)	
malignancy			
Inadequate for	18(29)	15(24.2)	
reporting			

For all lesions collected without the usage of suction or a stylet, the final cytological diagnosis was malignancy in 29 (46.8%), suspected for malignancy in 8 (12.9%), benign in 7 (11.3%), and inadequate for reporting in 18 (29%) lesions.

In specimens collected with a suction and stylet, the final diagnosis was malignant in 29 (46.8%), suspected for malignancy in 9 (14.5%), benign in 9 (14.5%), and inadequate for reporting in 15 (24.2%) lesions.

Diagnostic outcome of malignancy

The total diagnostic outcome of malignancy did not differ between the specimens acquired using the two procedures (with stylet and suction, 29/62 [46.8%] vs. without stylet and suction, 29/62 [46.8%], P = 0.901). In the same way, there was no significant difference in the two groups' final diagnoses. For the specimens that were found to be suggestive for malignancy (with stylet and suction, 9/62 [14.5%] vs. without a stylet and suction, 8/62 [12.9%], P = 0.901) (Figure 4, 5).



Fig. 4: Final cytological diagnosis of the lesion acquired without suction or stylet



Fig. 5: Final diagnosis of the lesion acquired using suction syringe and stylet

Cytopathological characteristic

The difference between the number of insufficient specimens acquired using the suction and stylet approach and those obtained without them was not statistically significant (P = 0.471).

As compared to specimens obtained without the use of a suction syringe and stylet, those obtained with them had a higher level of bloodiness, including mild amounts [4 (6.5%) vs. 17 (27.4%], moderate amounts [24 (38.7%) vs. 36 (58.1%), and significant amounts [34 (54.8%) vs. 9 (14.5%), (P = 0.001).

With no representative cells present in [17 (27.4%) vs. 12 (19.4%), P = 0.494, representative cells present in [9 (14.5%) vs. 13 (21%), P = 0.001], representative cells present in 25-50% of the slide[25 (40.3%) vs. 22 (35.5%), P = 0.494], and representative cells present in >50% of the slide, there was no significant statistical difference in cellularity between the specimens obtained with both techniques.

In terms of GI contamination, there was also no significant statistical difference between samples obtained without using a suction syringe and stylet and those obtained using a suction syringe and stylet, with no contaminations observed in [50 (80.6%) vs. 44 (71%), P = 0.511], contamination present in 25% of the slide [10 (16.1%) vs. 12 (19.4%), P = 0.511], contamination present in 25-50% [1 (1.6%) vs. 2 (3.2%), P = 0.511] and contamination in 50% of the slide [1 (1.6%) vs. 4 (6.5%), P = 0.511]. (Figure 6-8)



Fig. 6: Comparison between both techniques regarding the amount of blood



Fig. 7: Cellularity of the specimens acquired using both techniques



Fig. 8: Adequacy of the specimens acquired using both techniques

 Table 9: Measure of agreement between both techniques in cytological criteria and final diagnosis

	Kappa measure of agreement	95 th CI
Cellularity	0.713	0.575 to 0.851
Percentage of area of slide that represents GI contamination	0.551	0.358 to 0.743
Amount of blood	0.167	0.014 to 0.321
Adequacy of the specimen	0.813	0.658 to 0.969
Final diagnosis	0.833	0.720 to 0.946

Regarding agreement between both techniques in cytological criteria assessed by kappa measure of agreement, it was found that there was substantial agreement in cellularity (0.713), moderate agreement in GI contamination (0.551), slight agreement in bloodiness (0.167), perfect agreement in adequacy of the specimens (0.813) and final diagnosis (0.833).

DISCUSSION

FNA is a straightforward, low-cost, and safe procedure for collecting cytological samples for the diagnosis of various tumors. It is employed frequently to sample lymph nodes, thyroid, breast, and other readily accessible tumors. The ability of the procedure to collect cytologic specimens from practically every tumor, regardless of location, has improved with the emergence of new imaging techniques like EUS.^[7]

Today, EUS-FNA is a crucial investigative method in the detection and staging of several types of cancer. Many endosonographers routinely perform EUS-FNA while using a stylet and suction syringe. The unfounded idea that the utilization of a stylet enhances the quality of samples by preventing needle clumping by GI cells, hence reducing contamination and enhancing the diagnostic outcome during collection, lends support to this. Additionally, it is thought that applying high negative pressure while performing EUS-FNA by attaching a 20 ml suction syringe to the needle's proximal end enhances the cellularity of the specimen that is obtained.^[8]

The employing of a stylet lengthens the procedure and raises the price of the EUS-FNA needles. When a mass or lesion has been penetrated, it might be challenging to push or remove the stylet, making EUS FNA with a stylet more challenging in some situations. This can make the process more difficult for the assisting technician and raises the hazard of needle stick injuries. It happens when the needle is kinked or there is a loop in the echoendoscope. When dealing with fibrotic lesions, a stylet may be required to stiffen the needle and aid in puncturing the tough lesions. The stylet's ability to better express the material on the slides than blowing it with air is another benefit.^[9]

According to recent statistics, utilizing a stylet does not improve the quality of the specimens or the yield of malignancy diagnostics. In a randomized, controlled trial, 101 patients with 118 solid lesions were used to compare the two methods. The yield of malignancy did not differ significantly between the two groups (23% with a stylet vs. [28%] without a stylet, P =0.29). The cytological parameters of GI contamination (P = 0.92), adequacy (P = 0.26), the percentage of representative cells (P = 0.98), and bloodiness (P = 0.61) did not differ from each other.^[10] The diagnostic outcome of malignancy of samples acquired with EUS-FNA with and without a stylet was compared in another study. Additionally, comparison of cytopathological traits like cellularity, bloodiness, and sufficiency were made. Between the two groups, there were no appreciable differences in the cytological traits or the diagnostic outcome of malignancy.^[11]

Suction during EUS-FNA is still up for discussion. Numerous studies have suggested that capillary sampling, or EUS-FNA without suction, is a highly sensitive and accurate method for sampling and diagnosing a variety of gastrointestinal and nongastrointestinal tumours.^[13]

Suction generally increases the amount of representative cells per slide while decreasing the collected specimens' quality by increasing the amount of blood contamination.^[1]

Despite the fact that recent guidelines advise utilizing suction during EUS-FNA of solid lesions, there are many different ways to do so.^[14] According to the type of lesion, suction is often provided with a 10-ml suction syringe either constantly or intermittently. It is preferable to employ no or little suction when dealing with bloody aspirates. In fibrotic masses and sparse aspirates, suction is preferred.^[15]

Various studies have revealed the superior quality of samples obtained from solid lesions using the capillary suction technique, which relies on the low negative pressure produced by the slow stylet withdrawal. The capture of a larger specimen with less blood contamination is the justification for better specimen quality.^[16]

Another study that involved 97 patients and either the capillary suction technique (CST) or the syringe suction technique (SST) discovered that the latter was associated with potentially higher diagnostic yield (sensitivity 90.0% vs 67.9%) and less blood contamination with the 25G needle.^[17]

The diagnostic outcome of malignancy has been compared in this prospective, single-blind, randomized, controlled trial between specimens obtained with and without a 20 ml suction syringe and stylet during EUS-FNA. A cytopathologist who was blind to the suction and stylet arm conducted another comparison on the cytological parameters, such as cellularity, amount of blood, contamination, and sufficiency, based on specified criteria.

Between the two groups, there was no difference in the final diagnosis (P = 0.901). This study reveals that there were no appreciable variations in cellularity (P = 0.494), contamination (P = 0.511), or specimen adequacy (P = 0.471) between the EUS-FNA technique with and without suction and stylet. The sample quality was improved by the EUS-FNA without suction and stylet group, which had a significantly lower blood contamination percentage (P = 0.001).

The prospective design of this study is one of its advantages. An experienced endosonographer who performs more than 300 EUS-FNAs annually performed the procedure in a consistent manner.

Consideration should be given to a few study limitations. The group to which the passes belonged could not be concealed from the endosonographer. Apart from the use of a suction and stylet, there were deliberate measures made to guarantee that all passes in the lesion were performed with exactly the same technique.

This study's drawback is the cytopathologist subjective evaluation of the samples. However, for this study, extensive, predetermined criteria were employed to evaluate the cytopathological traits of the specimens with and without a suction and stylet. Additionally, one cytopathologist who was blinded to the group to which the slides belonged assessed each slide from a single patient.

The accuracy rates of the two techniques for cancer detection were not the focus of this investigation because patients were not tracked longitudinally. This study only employed 22-gauge EUS-FNA needles, thus it's possible that the results don't apply to other needle sizes.

The outcomes of this randomized, controlled trial show that the collected specimen's quality or the overall diagnostic yield of malignancy are unaffected by the use of a suction syringe and stylet during EUS-FNA. It is okay to perform EUS-FNA without using a suction syringe or a stylet, and this technique may even increase the procedure's overall effectiveness and diagnostic output.

CONCLUSION

The use of a suction syringe and stylet during EUS-FNA to improve the collected samples quality is still up for debate.

The findings of our study cast doubt on the notion that using a suction syringe, stylet, or both during EUS-FNA enhances the quality and diagnostic outcome of the acquired samples.

Regarding the sufficiency, cellularity, and diagnostic yield, there was no significant statistical difference between the samples obtained by EUS-FNA with and without

suction and stylet. The lack of suction and stylet approach demonstrated a decreased level of blood contamination, improving sample quality.

The practice of not using a suction syringe or stylet during EUS-FNA would be adopted if more multicenter randomized controlled trials supported these findings. As a result, EUS-FNA would be simpler, less labor-intensive, and more time- and money-efficient.

RECOMMENDATIONS

• It's more convenient to use no suction, no stylet technique during EUS-FNA of upper gastrointestinal solid lesion.

• There should be more multicentric prospective randomized studies conducted with blinded cytological analysis, but with more participants.

• Further large scale studies are needed with different groups of specific target lesions: esophageal, mediastinal, rectal masses, and so on.

CONFLICT OF INTEREST

There are no conflicts of interest.

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