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ORIGINAL ARTICLE

Performance of EUS-FNB in solid pancreatic masses: a lesson from 463 consecutive procedures and a practical nomogram

Nico Pagano[1] · Claudio Ricci[2,6] · Carlo Ingaldi[2,6] · Sinan Sadalla[1,6] · Andrea Fabbri[1,6] · Laura Alberici[2,6] · Giovanna Impellizeri[1,6] · Socrate Pallio[3] · Rocco Maurizio Zagari[1,6] · Antonio De Leo[4,7] · Matteo Cescon[5,6] · Riccardo Casadei[2,6]

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Abstract

The study's main goal was the diagnostic adequacy of pancreatic endoscopic ultrasonographic (EUS) fine-needle biopsy (FNB) and associated predictive factors. The secondary objective was to define the diagnostic accuracy of EUS-FNB in the diagnosis of pancreatic masses and pancreatic malignancies. None of the studies reported the diagnostic adequacy and accuracy of EUS. We retrospectively identified patients with solid pancreatic lesions that underwent EUS-FNB between 2013, and 2018. We calculated diagnostic adequacy and related factors. Using definitive histology on the surgically resected specimen as the gold standard, we calculated diagnostic accuracy, sensitivity, specificity, positive predictive value, and negative predictive value of EUS-FNB. We identified a total of 463 procedures. Diagnostic specimens were adequate in 436 procedures (94.1%), while 27 biopsies provided insufficient samples (5.9%). The multivariate analysis showed that lesion size and needle caliper were the only factors influencing diagnostic adequacy. The use of a biopsy needle (OR 0.69, 95% CI 0.30–0.1.63, *P* 0.400) did not improve sample adequacy. We calculated sensitivity (100%), specificity (93.2%), diagnostic accuracy (93.2%), positive predictive value (97.1%), and negative predictive value (100%) using resected specimen as the gold standard. We found no significant complications. EUS-FNB is a reliable technique for the histological characterization of solid pancreatic masses.

Keywords Endoscopic Ultrasound · Fine Needle Biopsy · Pancreatic cancer

Introduction

Pancreatic solid lesions comprise many different diseases, malignant as pancreatic ductal adenocarcinoma (PDAC), neuroendocrine tumors (NET), lymphomas, metastasis, or benign, such as chronic pancreatitis (CP) and autoimmune pancreatitis (AIP) [1–3]. EUS-guided sampling represents the technique of choice for tissue acquisition in most gastrointestinal lesions, including pancreatic lesions, liver nodules, lymph nodes, and subepithelial lesions [4]. At the beginning of the EUS era, the sampling was mostly cytological. In the last years, thanks to advances in technology, we can acquire real tissue cores by EUS [5]. Tissue samples, with preserved histological architecture, allow a better classification of pancreatic malignancies that is fundamental in the choice of personalized treatments [6]. Our study aim was to report the diagnostic adequacy of EUS-FNB in a tertiary center.

✉ Claudio Ricci
claudio.ricci6@unibo.it

1 Division of Gastroenterology, IRCCS Azienda Ospedaliero, Universitaria Di Bologna, via Albertoni 15, Bologna, Italy

2 Division of Pancreatic Surgery, IRCCS Azienda Ospedaliero, Universitaria Di Bologna, via Albertoni 15, Bologna, Italy

3 Dipartimento di Medicina Clinica e Sperimentale, University of Messina, Messina, Italy

4 Pathology Unit, IRCCS Azienda Ospedaliero, Universitaria Di Bologna, via Albertoni 15, Bologna, Italy

5 Transplantation and Hepatobiliary Unit, IRCCS Azienda Ospedaliero, Universitaria Di Bologna, via Albertoni 15, Bologna, Italy

6 Department of Internal Medicine and Surgery (DIMEC), Alma Mater Studiorum, University of Bologna, Bologna, Italy

7 Department of Specialistic, Diagnostic and Experimental Medicine (DIMES), Alma Mater Studiorum, University of Bologna, Bologna, Italy

Material and methods

We conducted a retrospective study based on a prospectively maintained database, which included all EUS-guided pancreatic tissue acquisition performed in the endoscopy center of the Gastroenterology Unit at IRCCS (Scientific Institute for Research, Hospitalization and Health Care) S.Orsola-Malpighi Hospital, Bologna between January 1 2013 and October, 31 2018. The informed consent was obtained from each patient included in the study. The study protocol conforms to the ethical guidelines of the 1975 Declaration of Helsinki. The local ethics committee approved data acquisition and analysis (code 401/2019/Oss/AOUBo). Enrollment criteria included aged ≥ 18 at the time of the procedure, solid pancreatic masses, availability of endoscopy and histologic reports, and informed consent. The flow-chart of the selection process is reported in Fig. 1. Briefly, we report the endoscopic ultrasound sampling procedures. We performed EUS-FNB in outpatients and inpatients, with a fastening period of at least 8 h and managing anticoagulant and antiaggregant therapy according to current guidelines [7, 8]. During the procedures, the patients were in left lateral decubitus and received oxygen support. Conscious or deep sedation was provided by the endoscopist or the anesthesiologist when present, with continuous monitoring of vital signs. We employed a conventional linear EUS scope for all procedures (GF-UCT 180 Olympus Medical System Europe). We used both a trans-gastric and a trans-duodenal approach for biopsy, depending on the lesion's site. We chose the type of needle according to lesion's size and site (available needles at the time of the study: Expect™ Slimline (SL) 19G/22G/25G, Acquire™ 22G, EchoTip ProCore™ HD 19G/20G/22G/25G). According to the macroscopic visual examination of the collected samples (MOSE), we decided the number of needle passes on a case-by-case basis. We used a single administration of antibiotics (ceftriaxone 2 g or levofloxacin 500 mg) only when considered necessary according to the patient's clinical situation, as current guidelines do not recommend routine antibiotic prophylaxis [9, 10]. At the end of the procedure, patients were stationed under observation in the Endoscopy Unit for 1 h, after which they were dismissed if no symptoms suggestive of a complication occurred. The outpatients were contacted by phone at home the day after the procedure according a pre-established protocol. An experienced endoscopist (NP) performed all the

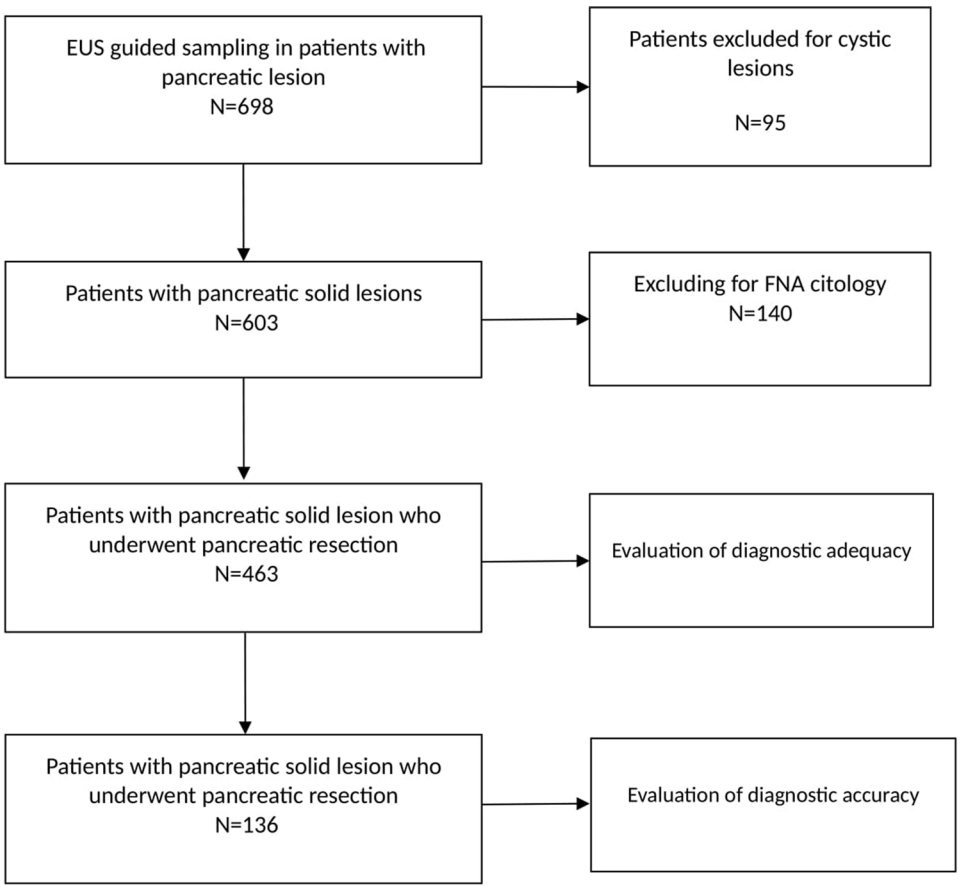

Fig. 1 Flow chart of patient selection

procedures. The study's main goal was to define the diagnostic adequacy of EUS-FNB and the associated clinical and technical factors. The secondary endpoints were: (1) to evaluate the diagnostic accuracy using surgical specimen as the gold-standard reference, (2) to evaluate procedure-related adverse events such as bleeding, pancreatitis, infection, and perforation. The demographic characteristics of patients are descriptive. We presented quantitative variables as proportion and mean ± SD, while categorical variables as relative and absolute frequencies. We used a backward logistic regression model to determine predictive variables of diagnostic adequacy (defined as the percentage of patients in whom EUS-FNB obtained a histologically interpretable specimen). A P value > 0.10 was used to remove the variables in backward multivariate, and a $P < 0.05$ was considered statistically significant. The multivariate analysis was reported as an odds ratio (OR) with a 95% confidence interval (CI 95%). Basing on the β coefficient of logistic regression and using a dedicated algorithm, we generated a nomogram predicting the diagnostic adequacy. Starting from the nomogram, we obtained a score that was calibrated using logistic regression and margin estimation. We also reported sensitivity (Se), specificity (Sp), negative (NPV), positive predictive values (PPV), and accuracy based on the final pathological diagnosis in predicting PDAC. The PPV was defined as the number of true positive out of all positive. The NPV was defined as the number of true negative out of all negative. PPV measures the odds that a patient with a positive biopsy is diseased. On the contrary, NPV calculates the odds that a patient with a negative biopsy is diseased. We used Stata 15 software (Stata Corp LP, TX) for statistical analysis.

Results

The flow-chart of patients' selection is reported in Fig. 1: starting from 698 cases, we excluded 95 patients because they had a cystic lesion and 140 because only cytology was available. In the final analysis, we included only 463 patients whose baseline characteristics are reported in Table 1. The overall diagnostic adequacy was 94.1%. The histologic diagnosis of the 436 adequate sample were: 255 (58.5%) pancreatic ductal adenocarcinoma (PDAC), 83 (19%) neuroendocrine Tumors (NET), 40 (9.2%) chronic pancreatitis/autoimmune pancreatitis (CP/AIP), 21 (4.8%) metastasis/lymphomas, 14 (3.2%) intraductal papillary mucinous neoplasia (IPMN), 3 (0.7%) serous neoplasia and 2 (0.5%) atypia. In 18 cases (4.1%) the specimen resulted in normal pancreatic parenchyma. The histological diagnosis is reported in Supplementary Table 1. The multivariate analysis showed that the only factors influencing sample adequacy were: the size of the lesion with an OR of 1.05 (1.01–1.10; P 0.019) for each mm and the needle caliper in Gauge with an OR of 0.45 (0.57–0.99; P 0.049). Sex, age, size, and needle type were not significantly related to diagnostic accuracy. The lesion site did not reach a statistical relevance but showed a trend: the diagnostic adequacy seems to drop comparing head-isthmus vs. body-tail location (OR 0.44; 0.17–1.16; P 0.088) (Table 2). The nomogram derived from the multivariate model is plotted in Fig. 2. Three parameters contributed to the final score: (i) needle caliper (from 0 points of 25 Gauge to 2.4 points of 19 Gauge); (ii) lesion location (from 0 points of lymph nodes to 1.9 points of head); (iii) lesion size (from 0.8 points of 10 mm to 7.8 points of 100 mm). The final score ranged from 0 to 21 points. The calibration of the score was graphically reported in Fig. 3 and exhaustively described in Supplementary Table 2. Starting from 0 value, for each incremental point, we observed a statistically significant increase in diagnostic adequacy. For a score greater than 9 points, the diagnostic adequacy was constantly higher than 90%. From 9 points on, for every further increase, the gain was progressively smaller. A definitive diagnosis, based on the analysis of surgically resected specimens, was available in 136 patients. The results showed 84 (61.8%) cases of pancreatic ductal adenocarcinoma, 30 (22.1%) cases of neuroendocrine

Table 1 Baseline characteristics of the 463 patients included

EUS-FNB parameters	Patients (*n* 463)
Sex, *n* (%)	
Male	231 (49.9)
Female	232 (50.1)
Age (years), mean (SD)	66.0 (13.5)
Lesion dimensions (mm), mean (SD)	25.6 (13.6)
Lesion site, *n* (%)	
Head	267 (57.7)
Isthmus	35 (7.5)
Body-tail	161 (34.8)
Needle type, *n* (%)	
Traditional needle	283 (61.1)
Biopsy needle	180 (38.9)
Needle caliper (Gauge), *n* (%)	
19	207 (44.7)
20	18 (3.9)
22	226 (48.8)
25	12 (2.6)
Number of passages, mean (SD)	2.1 (0.9)
Diagnostic adequacy, *n* (%)	
Adequate sample	436 (94.1)
Inadequate sample	27 (5.9)

EUS-FNB endocscopic ultrasound fine-needle biopsy, *SD* standard deviation

Table 2 Multivariate analysis for predictive factors of diagnostic adequacy

Parameters	OR (95% CI)	*P* value	Step exclusion
Sex			4th
Female	1		
Male	2.05 (0.78–5.38)	0.156	
Age (years)	1.01 (0.98–1.10)	0.780	1st
Size (mm)	1.05 (1.01–1.10)	0.019	Final
Localization			Final
Head	1		
Isthmus	0.70 (0.18–2.73)	0.605	
Body-tail	0.44 (0.17–1.16)	0.088	
Lymph-node	0.30 (0.05–1.83)	0.194	
Needle type			2nd
Traditional needle	1		
Biopsy needle	0.69 (0.29–1.63)	0.408	
Needle caliper (Gauge)	0.45 (0.57–0.99)	0.049	Final
Number of passages	1.39 (0.85–2.27)	0.221	3rd

OR Odds ratio, *CI* confidence Interval

tumors, 15 (11%) cases of metastasis/lymphoma, 3 (2.2%) cases of IPMNs, 3 (2.2%) cases of CP/AIP and 1 (0.7%) case of serous lesion (Supplementary Table 3). Based on the above findings, EUS-FNB for PDAC showed 100% sensitivity, 93.2% specificity, 93.2% diagnostic accuracy, 97.1% positive predictive value, and 100% negative predictive value (Table 3).

Discussion

Our study shows that EUS-FNB achieves an adequate sample for histological diagnosis in more than 94% of the cases. Endoscopic ultrasound-guided sampling has a fundamental role in the diagnosis and management of gastrointestinal lesions, and it is considered the first choice to biopsy pancreatic masses [4, 11]. When EUS affirmed its position in the clinical context, the first technique used for tissue sampling was EUS-FNA with cytology assessment [12]. A recent meta-analysis found no significant difference in diagnostic adequacy between EUS-FNA and EUS-FNB when rapid onsite evaluation (ROSE) was available during FNA. Without ROSE, FNB showed better diagnostic adequacy in the characterization of solid

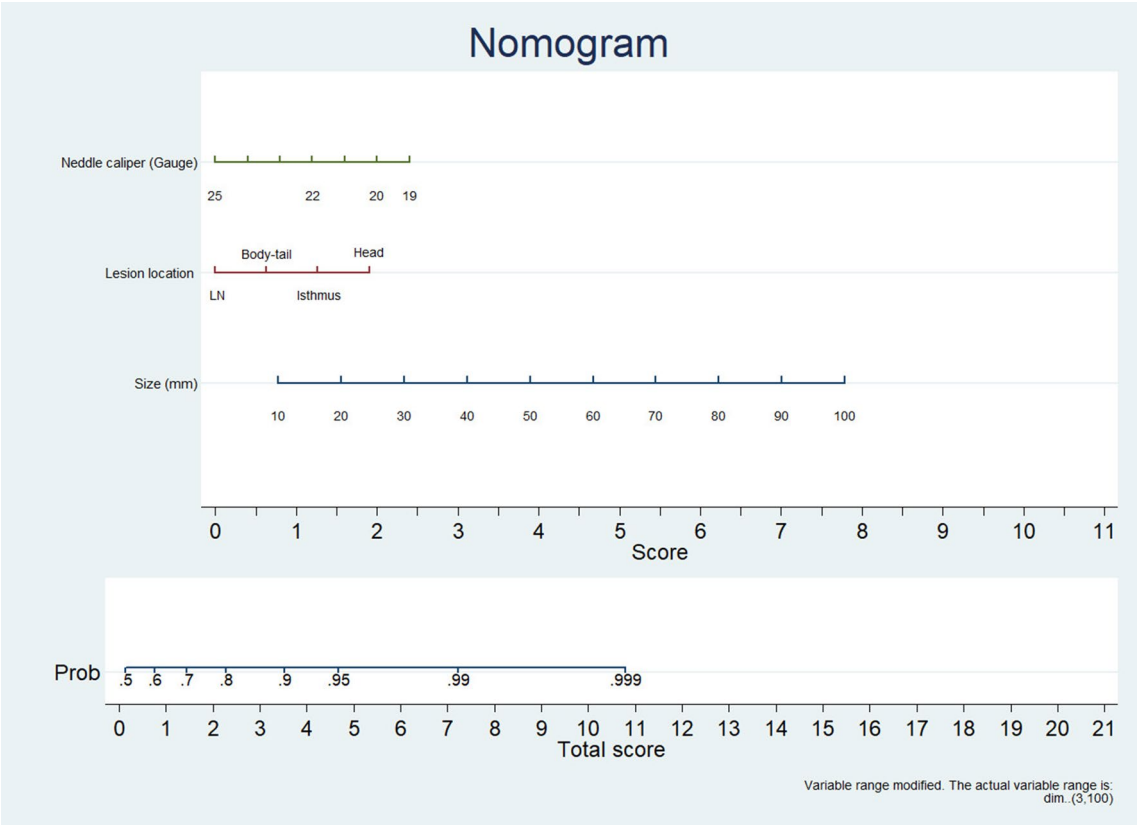

Fig. 2 Nomogram

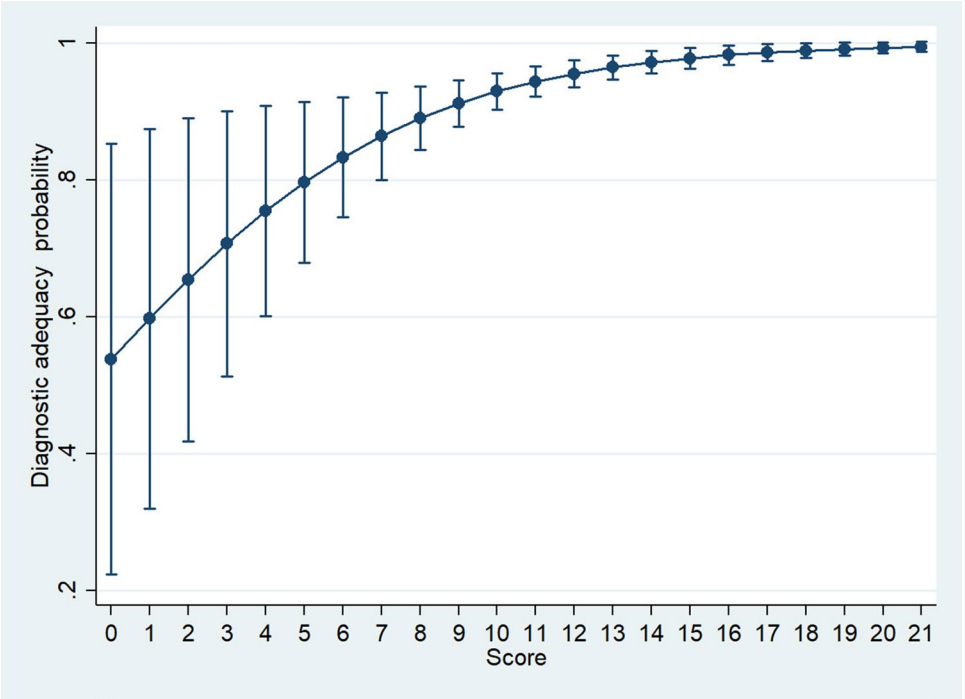

Fig. 3 Nomogram score calibration

Table 3 Comparison between EUS-FNB and resected specimen

Comparison between EUS-FNB and resected specimen		Biopsy							
		Carcinoma (%)	IPMN (%)	NET (%)	Metastasis (%)	PSC (%)	CP (%)	SPT (%)	Total (%)
Surgical specimen	Carcinoma	81 (95.2)	0	0	0	0	0	0	81 (59.6)
	IPMN	2 (2.4)	2 (100)	0	0	0	0	0	4 (2.9)
	NET	0	0	28 (100)	0	0	0	0	28 (20.6)
	Metastasis	0	0	0	15 (100)	0	0	0	15 (11.1)
	PSC	0	0	0	0	1 (100)	0	0	1 (0.7)
	CP	2 (2.4)	0	0	0	0	3 (100)	0	5 (3.7)
	SPT	0	0	0	0	0	0	2 (100)	2 (1.4)
	Total	85 (100)	2 (100)	28 (100)	15 (100)	1 (100)	3 (100)	2 (100)	136 (100)

Observed agreement	Expected random agreement	Kappa ± SD	*P* value
97.06%	42.83%	0.949 ± 0.056	< 0.001

EUS-FNB endoscopic ultrasound fine-needle biopsy, *IPMN* Intraductal papillary mucinous neoplasm, *NET* neuroendocrine tumors, *PSC* pancreatic serous cystadenoma, *CP* chronic pancreatitis, *SPT* solid pseudopapillary tumor, *SD* standard deviation

pancreatic lesions [13]. FNB also seems to require fewer needle passes than FNA to establish malignancy diagnosis [14–17]. Endoscopic ultrasound-guided fine-needle biopsy (FNB) primary goal is to overcome FNA limitations, and besides adequacy issues, the major pitfall of EUS-FNA is the inability to preserve tissue architecture [18, 19]. Immunohistochemical and molecular characterization is possible only on tissue cores, so tissue sampling allows the choice of optimal treatment for each patient with a personalized approach[4,5]. In our series, we had diagnostic adequacy on the lower end of the range reported in the literature [5]. A plausible explanation is that our series included a significant rate of non-malignant pancreatic masses, reducing the pre-test probability of pancreatic malignancy, and a high number of chronic pancreatitis, in which the diagnostic accuracy for pancreatic malignancies is reduced [20]. The analysis of the factors influencing adequacy showed that lesion size and needle caliper are the only factors reaching statistical significance. We observed an increase in the diagnostic adequacy of 5% for each mm of lesion's size. This result confirms previous studies showing better adequacy of the specimen in larger lesions, which was

also reported in EUS-FNA studies [21]. The more interesting result is the correlation between needle caliper and diagnostic adequacy. We observed a significant increase in adequacy between the use of minor caliper needles and larger ones. This is a remarkable result, and previous studies showed this trend [22]. Technological advancement allowed the development of biopsy needles, improving, in theory, the ability to obtain a tissue core [23]. We found no impact of the dedicated needles in increasing adequacy rate compared to the standard needle, and the number of needle passes did not correlate with an improvement in diagnostic yield, as previously reported in other studies [20]. Our data also showed a trend towards incremental diagnostic yield from the body-tail to isthmus-head, though not statistically significant. We used the dataset to generate a nomogram able to predict the probability of diagnosis. After creating and calibrating the nomogram, we created a score for the adequacy probability. Observing the nomogram, a message arises: given the non-modifiable variables such as location and size, the choice of the needle caliper is crucial. In particular, the smaller the lesion the larger must be the needle to acquire enough tissue to obtain a diagnosis.

It seems fair to affirm that this study explores the diagnostic performance of EUS-FNB in a real-world practice of non-selected patients so that these results can apply to everyday practice.

Regarding FNB performance, diagnostic accuracy is lower than in previous studies [24–26] but still good, over 90%. This is related to the application of stringent criteria to declare correct a diagnosis: even a little discrepancy in histology between EUS biopsy and the surgical specimen was considered a diagnostic failure. We did not test the mere capacity of EUS-FNB to obtain a diagnosis but to make a correct classification of the lesion with definitive histology as the gold standard. Regarding the most common and important lesion (PDAC), the false positives were near 2.9%. In our series, the false-positive cases were due: (1) to chronic pancreatitis in which the structural changes were similar to fibrotic peritumor modifications due to obstructive PDAC; (2) to IPMN in which the mild-moderate dysplasia could be erroneously interpreted as in situ-invasive carcinoma.

As we can see from Table 3 we had a correct classification of the lesion most of the time, even with rare diagnoses such as a solid pseudopapillary tumor.

This means that EUS-FNB is a reliable method to characterize a lesion, obtaining a tissue core that allows histological diagnosis and ancillary methods, like the immunohistochemical analysis.

In all adequate specimens, immunohistochemical analysis was feasible, granting a complete diagnostic definition, thus confirming that EUS-FNB samples had a preserved tissue architecture. This is fundamental in FNB and differentiates this approach from FNA, in which samples usually consist of macro cell-aggregates, often not suitable for further characterization [27].

Regarding safety and feasibility, EUS-FNB was performed both in outpatients and in hospitalized patients, no significant adverse events occurred during or after the biopsy, showing that this is a safe procedure, especially if we consider the high number of cases and the absence of technical failures.

We found no significant difference in diagnostic yield between head and body/tail lesion, so there is no difference in performance between trans-duodenal and trans-gastric approach, confirming a recent study [28].

Our study has some limitations. First, it is a retrospective study and could be affected by selection bias, but it is worth noticing that our database was maintained prospectively, so this kind of bias should be limited. In the second place, long-time follow-up was not available for all patients, but the use of histology as a gold standard overcame this limitation, though narrowing the evaluation of diagnostic accuracy only to patients undergoing surgery. In conclusion, our data confirm current evidence that EUS-FNB is a feasible procedure defined by a high safety profile and a high technical success rate. The diagnostic yield was 94%, and in most cases, the material allowed histological and immunohistochemical analysis. We must consider that pancreatic pathologists, especially in Europe, are more confident with histologic samples than with cytology so that the biopsy approach can be considered more applicable in clinical practice. Avoiding the need for ROSE, EUS-FNB leans the endoscopic suite workflow; moreover, reducing the number of needles passes, the technique is less time consuming and, virtually, safer [9, 17, 29]. In the attempt to obtain an adequate tissue sample, the use of needles with a large caliper is related, in our experience, to a higher success rate with no difference between dedicated and standard needles. In particular, our nomogram shows that the smaller the lesion, the larger has to be the needle to compensate for the lower adequacy probability. Our experience suggests that EUS-FNB is a reliable technique for obtaining tissue samples that can be processed as histology with all the related implications.

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Declarations

Conflict of interest The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Informed consent The research involved human partecipants. The informed consent was obtained from each patient included in the study.

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