Endoscopic ultrasound-guided fine needle aspiration in diagnosis of cystic pancreatic lesions

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ABSTRACT

Background and study aims: pancreatic cysts are commonly found lesions and proper diagnosis is very important for planning further management. The study aims to evaluate the role of cyst fluid amylase and tumour markers as cancer antigen (CA 19-9) and carcinoembryonic antigen (CEA) in addition to mucin stain in diagnosing pancreatic cysts and differentiating malignant from benign lesions.

Patients and methods: This prospective study was conducted on 184 patients diagnosed to have pancreatic cystic lesions from January 2013 to January 2018. Fluid analysis for CA 19-9, CEA, amylase, mucin stain and cytopathology were done. We compared these data with the final diagnosis based on histopathology after surgical resection, positive cytopathology and long period of follow up of the patients for at least 18 months.

Results: The highest AUC was that of cystic CEA with cut-off value of 160 ng/ml; it had a sensitivity of 60.4% and a specificity of 85%. The best cut-off value for cystic CA 19-9 was 1318 U/ml with a sensitivity of 64.1% and a specificity of 68.1%. The cut-off value of cyst amylase level was 5500 U/L, with 84.2% sensitivity and 37.1% specificity. The sensitivity of mucin stain in detecting mucinous cystic neoplasm was 85.45%, specificity was 86.05% with accuracy 85.87%.

Conclusion: Cyst fluid analysis by investigating amylase, mucin, CA 19-9, CEA and EUS examination improves the diagnosis of different pancreatic cysts.

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Introduction

Pancreatic cystic lesions are common and vary from simple cyst to potentially neoplastic lesion [1–3]. Mucinous cysts (intraductal papillary mucinous neoplasms (IPMN) and mucinous cystic neoplasms (MCN)) have unpredictable potential for malignant transformation [4]. Given the poor outcomes associated with pancreatic malignancy, differentiation and staging of these cystic lesions represent important requirements for reaching an exact diagnosis and eventually appropriate management [5]. It has been reported that combined sonographic features of cystic lesions and FNA yield have low negative predictive value [6] and the negative cytopathological finding dose not totally exclude neoplasia [7].

Existing studies have suggested that measuring cystic fluid tumour markers namely carbohydrate antigen (CA19-9) and carcinoembryonic antigen (CEA) in addition to amylase level and mucin stain provide an important contributing role in diagnosis of pancreatic cysts [8–10]. Therefore, combining both EUS-FNA findings with these biomarkers might enhance the diagnostic accuracy of pancreatic cystic lesions and help to construct a novel diagnostic model.

This work aims to evaluate the role of cyst fluid amylase and tumour markers (CA-19 and CEA) in addition to mucin staining in diagnosing pancreatic cysts and to differentiate between malignant and benign lesions.

Patients and methods

Study design and population

A prospective study was conducted on 184 patients with pancreatic cystic lesions on imaging modalities, CT, EUS, abdominal ultrasound or MRI. Study subjects were recruited from Endoscopic unit of Internal Medicine Department at Cairo University Hospital during the study period from Jan 2013 to Jan 2018.
Patients older than 18 years old who had large pancreatic cyst complicated by obstructive jaundice, those with suspicious papillary mucinous neoplasm (IPMN), and patients with pancreatic duct dilatation and common bile duct strictures proved by ERCP or MRCP were included in the study, while patients with small cyst (less than 1 cm), calcular cholecystitis, those with high risk for sedation and bleeding diathesis, or risk of deep sedation were rolled out. In addition, patients who refused to sign the consent to be enrolled in the study and those who missed the follow up were ruled out from the study.

The study protocol was revised and approved by the Medical Research Committee in the Internal Medicine Department, Cairo University. Each participant provided signed informed consent after proper orientation about the study procedures. The study was treated with confidentiality according to Helsinki declarations.

Methodology

After full history taking and clinical examination, the patients were subjected to:

1) Endoscopic ultrasound examination with a Pentax echoendoscope, EG3830UT (HOYA Corporation, PENTAX Life care Division, Showanomori Technology Center, Tokyo, Japan) attached to Hitachi machine EUB-7000. EUS guided FNA was done using 22 or 19G Echotip needless (Cook, Endoscopy, Winston-Salem, NC). Deep sedation with IV Propofol was used in all patients. Prophylactic ceftriaxone (1gm) was administrated prior to the procedure.

2) Analysis of cystic fluid

The cystic fluid was near completely evacuated with a single needle pass. Aspirated material inside the needle was spread over dry slides and also part of the fluid sample (at least 2 ml) was sent for cytopatological examination including mucin staining using alcian blue stain. At least 3 ml of cyst fluid were analyzed for CEA and CA-19-9 using two-site immunoassays (Beckman Coulter), amylase was measured by enzymatic colorimetric assay on a modular system (Roche).

A single Endosonographer performed EUS and the pathologist was blinded to EUS results. Follow up of patients after the procedures revealed no post-procedures serious complications.

Final diagnosis was based on the presence of one of the following:

1. Malignant cytology in the FNA
2. Presence of distant metastasis
3. Follow up of benign lesions for at least 18 months with no change in the size or development of distant metastasis.
4. Surgical resection and postoperative histopathological examination.

Surgical excision was done for 48 surgically fit patients. 42 out of 72 mucinous cysts and 6 malignant masses with duct adenocarcinoma with cystic breakdown (2 patients), solid pseudopapillary tumours (2 patients) and serous cystadenocarcinoma (2 patients). Postoperative histopathological examination proved the operative diagnosis.

Data collection and statistic analysis

Analysis of data was done using SPSS (statistical program for social science version12. Data were expressed in median and IQR. T-test was used; significance was taken at the level of 5%. The sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), and accuracy were all calculated manually based on published methodology.

Results

The study included 184 patients who underwent EUS guided-biopsy. Patients were 103 males and 81 females. Their age ranged between 12 and 78 years, with a mean (SD) of 50.47 (13.97) years. The commonest finding detected by abdominal ultrasound was body cyst (35.89%) followed by head cyst (29.89%). The demographic, clinical and laboratory data of the studied patients are summarized in Table 1.

Out of 184 enrolled patients, 106 (57.6%) were diagnosed to have benign lesions, while 78 (42.4%) had malignant lesions. The detailed final diagnoses are shown in Table 2. Non-mucinous lesions were encountered in 129 patients.

The incidence of positive mucin stain was significantly higher in patients with mucinous cystic lesions 47 (85.5%) compared to those with non-mucinous cysts 18 (14%) with p < 0.0001.

Cyst fluid CA19-9 and CEA levels were significantly higher in malignant patients with median (IQR) values of 1900 (19874) and 418 (1379) respectively, compared to those with benign
lesions \(355 (11,995)\) and \(3 (86)\) respectively with \(p = 0.033\) and \(< 0.0001\), respectively; however median value of cystic amylase was 224 (14,484) in benign lesions and did not differ statistically from malignant lesions 145 (1,726), \(p = 0.163\) as demonstrated in Table 3 and Fig. 2a–2c.

Median of serum amylase was 2,378 with IQR 1,402 while the median of serum CA19-9 was 395,498 with IQR 43,418.

It is worth mentioning that CEA level in patients with infected pseudocyst (8 subjects) was found to be high \(392 \pm 511.192\) with no significant difference compared to malignant lesions.

Using the Roc curve analyses of the three markers as tools of diagnosis of malignancy, the highest AUC was that of cystic CEA. With cut-off value of 160 ng/ml, it had 60.4% sensitivity and 85% specificity. The best cut-off value for cystic CA 19-9 was 1,318 IU/L with 64.1% sensitivity and 68.1% specificity. The criterion of cystic amylase level was 5,500 IU/L with 84.2% sensitivity and 37.1% specificity. The sensitivity of mucin stain in detecting mucinous cystic neoplasm was 85.45%, specificity 86.05% with accuracy of 85.87% (shown in Table 4 and Fig. 3a–3c).

<table>
<thead>
<tr>
<th>Variables</th>
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<th>Max</th>
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<th>IQR</th>
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<td>Malignant</td>
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<td>59,500</td>
<td>59,499</td>
<td>418.00</td>
</tr>
</tbody>
</table>

Fig. 1a. Inflammatory pseudocysts with multiple septations.

Fig. 1b. Microcystic serous cystadenoma with honey-comp appearance.

Fig. 1c. Solid pseudopapillary neoplasm with minute areas of haemorrhage.

Fig. 1d. Mucinous cystic neoplasm with mural nodules and mucous plugs.

Fig. 2a. Cystic amylase level of benign and malignant pancreatic lesions.

Fig. 2b. Cystic CA-19 level of benign and malignant pancreatic lesions.

Fig. 2c. Cystic CEA level of benign and malignant pancreatic lesions.
Discussion

The current study suggests that combined cystic fluid analysis for amylase, mucin, CA 19-9, CEA and EUS examination could improve the differentiation between different pancreatic cysts. Some criteria are considered to be indicative of malignancy in pancreatic cysts, namely mural nodules, dilated pancreatic duct, thickened wall and septa, lymph node enlargement and elevated tumour markers. In this study, we aimed to find the role of EUS and EUS-FNA in differentiating between different pancreatic cysts and to highlight the value of EUS and EUS-FNA before surgery.

The study revealed that cystic level of CA19-9 and CEA were higher in malignant lesions than benign lesions which is consistent with a previously reported significant difference between mucinous (potentially malignant and malignant) and benign non-mucinous cysts [11].

In this clinical setting, cyst fluid analysis for CEA, CA19-9, amylase, mucin, and cytopathological examination could help to identify the nature of the pancreatic cyst whether malignant or benign and the consequent management.

It has been proved that EUS-FNA is more accurate for cytological diagnosis of pancreatic neoplasm than ERCP cytology [12] however both procedures are complementary in the evaluation of patients with pancreatic neoplasms [13].

In our study, median cystic CEA level was 418 ng/ml and that of CA 19.9 was 1900 U/ml in malignant pancreatic lesions which is significantly higher than the levels in benign cystic lesions. An earlier study concluded that elevated CEA in cystic fluid was noted in mucinous cysts, with low levels in serous cystadenomas (SCA) and pseudocysts and also it demonstrated that CA 19-9 and 125 levels were variable and heterogeneous.

These findings are along with what were suggested in many previous multicenter studies where both CEA and CA 72-4 concentrations were high in the cyst fluid of mucinous cystic neoplasms at different values with altered accuracy [12–14].

Nevertheless, Brugge and colleagues suggested that combination of tumour markers (CEA, CA 72-4, CA 125, CA 19-9 and CA 15-3) and cytology of cystic fluid was of no value in diagnosing pancreatic cysts [15].

Concerning amylase level in cystic fluid, it was found to be increased in benign lesions than in malignant lesions. This is along with a study identified a significantly higher mean level of amylase in benign compared with malignant pancreatic cysts [16,17]. Cyst amylase levels is elevated in pseudocysts as well and mucinous cysts especially IPMN, however, cyst amylase level less than 250 U/L virtually excludes the diagnosis of inflammatory pseudocysts [18,19].

The diagnostic performance of cyst tumour markers reported various cut-off values [8,11,20]. In our study, Roc curves analysis
revealed that cut-off value of 160 ng/ml of cyst CEA level had 60.4% sensitivity and 85% specificity. The cut-off value for cyst fluid CA-19-9 of 1318 U/ml had 64.1% sensitivity and 68.1% specificity. A review done by Van der Waaij et al noted a level of CEA > 800 ng/ml significantly suggests underlying mucinous pathology at higher specificity of 98% and poor sensitivity of 48%. Similarly, CA 19-9 at criterion of less 37 U/ml was indicative of SCA with a specificity of 98% and sensitivity 19% [21].

Leung et al calculated CEA cut-off value for differentiating a mucinous from non-mucinous cyst at 109.9 ng/ml, with 81% sensitivity and 98% specificity [11].

In the current study, the cyst amylase level was of 5500 U/L and it had a sensitivity of 84.2% and a specificity of 37.1%. This is not in accordance with one study that revealed a cut-off value for cyst amylase level above 479 U/L yielded a sensitivity of 73% and a specificity of 90% for distinguishing cystic neoplasms from non-neoplastic lesions [22].

On contrary to our findings, cyst amylase sensitivity was lower, with nearly similar specificity (62.5 and 69.4%, respectively) compared to CEA (91.8 and 63.9% respectively) and for CA 19-9 (81.3 and 69.4%, respectively) [16] for diagnosis of malignant or potentially malignant lesions, inconsistent with previous studies [23-25].

Regarding the role of mucin staining in distinguishing mucinous from non mucinous cysts, we noticed in pancreatic cysts it had a PPV of 72.4% which is lower than reported in another study in which mucin staining had better performance and accuracy [26,27].

Our study should be viewed with some points of limitations due to the lack of DNA analysis of cyst fluid for more accurate reliable results. Further studies addressing new markers are awaited which provide a panel of clinical, imaging and laboratory data to better recognize the malignant and potentially malignant lesions.

To conclude, cystic fluid analysis by investigating amylase, mucin, CA 19-9, CEA and EUS examination improves the differentiation between different pancreatic cysts. This helps in some special situations as in pancreatic cysts with no clear typical morphology particularly before invasive surgical intervention.

Declaration of Competing Interest
None

Acknowledgment
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Disclosures
The authors certify that they have no conflict of interest with any organization or entity with any financial or non-financial interest in the subject matter or materials discussed in this manuscript.

Appendix A. Supplementary data
Supplementary data to this article can be found online at https://doi.org/10.1016/j.ajg.2019.05.008.

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