Pancreatic Cyst Fluid Vascular Endothelial Growth Factor A and Carcinoembryonic Antigen: A Highly Accurate Test for the Diagnosis of Serous Cystic Neoplasm

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BACKGROUND: Accurate differentiation of pancreatic cystic lesions is important for early detection and prevention of pancreatic cancer, as well as avoidance of unnecessary surgical intervention. Serous cystic neoplasms (SCNs) have no malignant potential, but can mimic the following premalignant mucinous cystic lesions: mucinous cystic neoplasm and intraductal papillary mucinous neoplasm (IPMN). We recently identified vascular endothelial growth factor (VEGF)-A as a novel pancreatic fluid biomarker for SCN. We hypothesize that combining cyst fluid CEA with VEGF-A will improve the diagnostic accuracy of VEGF-A.

METHODS: Pancreatic cyst/duct fluid was collected from consenting patients undergoing surgical cyst resection with corresponding pathologic diagnoses. Pancreatic fluid VEGF-A and CEA levels were detected by ELISA.

RESULTS: One hundred and forty-nine patients with pancreatic cystic lesions met inclusion criteria. Pathologic diagnoses included pseudocyst (n = 14), SCN (n = 26), mucinous cystic neoplasm (n = 40), low-/moderate-grade IPMN (n = 34), high-grade IPMN (n = 20), invasive IPMN (n = 10), and solid pseudopapillary neoplasm (n = 5). Vascular endothelial growth factor A was significantly elevated in SCN cyst fluid compared with all other diagnoses ($p < 0.001$). With a threshold of >5,000 pg/mL, VEGF-A alone has 100% sensitivity and 83.7% specificity to distinguish SCNs from other cystic lesions. With a threshold of ≤10 ng/mL, CEA alone identifies SCN with 95.5% sensitivity and 81.5% specificity. Sensitivity and specificity of the VEGF-A/CEA combination are 95.5% and 100%, respectively. The c-statistic increased from 0.98 to 0.99 in the receiver operating characteristic analysis when CEA was added to VEGF-A alone.

CONCLUSIONS: Although VEGF-A alone is a highly accurate test for SCN, the combination of VEGF-A with CEA approaches the gold standard for pathologic diagnosis, importantly avoiding false positives. Patients with a positive test indicating benign SCN can be spared a high-risk surgical pancreatic resection. (J Am Coll Surg 2017;225:93−100. © 2017 Published by Elsevier Inc. on behalf of the American College of Surgeons.)

Disclosure Information: Nothing to disclose.

Support: This study was supported by the Indiana Clinical and Translational Sciences Institute, funded in part by Grant Number UL1TR001108 from the National Institutes of Health, National Center for Advancing Translational Sciences, Clinical and Translational Sciences Award, and by a grant from the Lustgarten Foundation and the Indiana Genomics Initiative of Indiana University, supported in part by Lilly Endowment Inc.

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Presented at the Western Surgical Association 124th Scientific Session, Coronado, CA, November 2016.

Received January 4, 2017; Revised April 3, 2017; Accepted May 5, 2017.

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http://dx.doi.org/10.1016/j.jamcollsurg.2017.05.003
ISSN 1072-7515/17

Abbreviations and Acronyms

EUS = endoscopic ultrasonography
FNA = fine-needle aspiration
IPMN = intraductal papillary mucinous neoplasm
MCN = mucinous cystic neoplasm
PNET = pancreatic neuroendocrine tumor
ROC = receiver operating characteristic
SCN = serous cystic neoplasm
SPN = solid pseudopapillary neoplasm
VEGF-A = vascular endothelial growth factor A
VHL = von Hippel-Lindau

Pancreatic cysts are being increasingly diagnosed due to advancements in radiographic imaging allowing for higher diagnostic sensitivity and higher test volume; 2.6% of patients undergoing abdominal imaging will have incidentally diagnosed cystic lesions of the pancreas.[1] Pancreatic cysts can be differentiated based on malignant potential. Although mucinous cysts, such as intraductal papillary mucinous neoplasm (IPMN) and mucinous cystic neoplasm (MCN), harbor malignant potential, serous cystic neoplasm (SCN) and pseudocysts virtually never progress to invasive carcinoma.[2] Consequently, management ranges from observation to aggressive pancreatic resection. Pancreatic surgery is associated with considerable mortality and morbidity, according to the contemporary literature.[3] Diagnostic accuracy is therefore of paramount importance.

Serous cystic neoplasm is a benign lesion that represents 16% of resected pancreatic cysts and <1% of all pancreatic lesions.[4,5] Only 3 serous cystadenocarcinomas were found within the largest series of SCN patients to date (n = 2,622), demonstrating the extremely low rate of malignant progression.[6] In addition, SCN-specific mortality approaches nil, at 0.1%.[6] For these reasons, in addition to the low rate of symptoms, the majority of SCNs can be managed nonoperatively.[6-8] Despite this, the retrospective multinational series by Jais and colleagues[6] found only 39% of SCN patients avoid surgical resection. Major indications for surgery included presence of symptoms, cyst size or growth rate, and lack of preoperative diagnosis (60%).[6] Although exact thresholds for cyst size or growth rate are controversial, most agree that diagnostic uncertainty is unacceptable.[2,7,9,10]

Existing diagnostic modalities (CT, MRI, endoscopic ultrasonography [EUS], and fine-needle aspiration [FNA] with cyst fluid analysis) are imperfect and leave many cysts undiagnosed or incorrectly diagnosed.[8,11] Biomarker research is being pursued in hopes of improving the diagnostic accuracy for pancreatic cysts. However, the majority of emerging biomarker investigations focus on mucinous cysts with malignant potential. It is equally important to develop diagnostic tools for benign lesions to avoid unnecessary morbidity and mortality of surgery. We previously identified vascular endothelial growth factor (VEGF)-A as a valuable pancreatic fluid biomarker for differentiating benign SCN from all other pancreatic cysts.[12] In the current study, we aim to validate VEGF-A in a larger cohort and establish an improved VEGF-A/CEA diagnostic test for SCN. This test will facilitate avoidance of unnecessary pancreatic resection for SCN.

METHODS

Patient samples

Samples were obtained from the Indiana University Pancreatic Tissue-Fluid Bank after approval by Indiana University IRB or were kindly provided by Johns Hopkins University (n = 12, Dr Anne Marie Lennon). Patients signed informed consent for collection of pancreatic fluid at the time of routine endoscopy (EUS or ERCP) or operation. We have previously confirmed that the method of fluid procurement does not affect VEGF-A measurement.[12] Fluid specimens were placed on ice immediately after procurement and aliquoted for storage at −80°C. In total, samples from 149 patients collected between 2003 and 2015, including serous cystic neoplasm (n = 26), pseudocyst (n = 14), mucinous cystic neoplasm (MCN; n = 40), low-/moderate-grade IPMN (n = 34), high-grade IPMN (n = 20) or invasive IPMN (n = 10), and solid pseudopapillary neoplasm (SPN; n = 5), were pathologically confirmed after surgical resection. Intraductal papillary mucinous neoplasm dysplasia was determined according to WHO criteria. Vascular endothelial growth factor A levels were reported previously for 87 of these patients.

Vascular endothelial growth factor A and carcinoembryonic antigen measurement

Pancreatic fluid samples (1 to 50 μL) were analyzed for VEGF-A by Quantikine ELISA (R&D Systems) according to manufacturer's protocol. Carcinoembryonic antigen was determined by Beckman Coulter DxI 800 analyzer or by ELISA in cases of low fluid volume (Sigma-Aldrich). Carcinoembryonic antigen values obtained by ELISA were converted to the Beckman automated analyzer scale using linear regression.

Molecular genetic analysis of von Hippel-Lindau

DNA was extracted from 3 SCN samples and their matched adjacent normal tissue using the QIAamp DNA Mini Kit (Qiagen). A custom multiplex polymerase