Clinical Cancer Research

Cyst Fluid Analysis in Pancreatic Intraductal Papillary Mucinous Neoplasms

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Pancreatic intraductal papillary mucinous neoplasms (IPMN) are surgically treatable cancer precursors, if high-risk features can be identified. Analysis of proteins and genes in cyst fluid helps diagnostically, but prior biomarkers had limited ability to detect high-risk cases. Cyst fluid telomerase activity may be a useful biomarker for high-risk IPMNs. *Clin Cancer Res; 22(20); 4966–7. ©2016 AACR.*

See related article by Hata et al., p. 5141

In this issue of *Clinical Cancer Research*, Hata and colleagues (1) propose the use of telomerase activity as a novel cyst fluid biomarker for high-risk intraductal papillary mucinous neoplasms (IPMNs) of the pancreas. Pancreatic cancer is almost uniformly fatal because of the late stage at diagnosis and the ineffectiveness of surgical resection and current systemic therapies for invasive carcinoma; even in the <15% of patients diagnosed prior to the development of identifiable metastases and treated with resection and adjuvant systemic therapy, the probability of survival beyond 5 years is less than 10%. Identification of high-risk precursor lesions, such as IPMNs with high-grade dysplasia, provides an opportunity to intervene surgically before invasive carcinoma develops.

The epithelial lining of IPMNs is believed to evolve from low-grade dysplasia to high-grade dysplasia to invasive carcinoma (2). Diagnostic technologies to identify these mucinous precursor lesions have improved, to the extent that distinguishing them from non-neoplastic cysts and purely benign cystic neoplasms is becoming less problematic. A variety of cyst fluid proteins (CEA, CA19-9, CA15-3, MUC-1, and amylase) have been evaluated as potential diagnostic markers of cyst subtype. Although all of these can be elevated in mucinous cysts, a prospective study by Brugge and colleagues of 112 patients with cystic lesions showed that an elevated cyst fluid CEA level (>192 ng/mL) was the best predictor of a mucinous lesion (IPMN or mucinous cystic neoplasm) and accurately identified them in 79% of cases (3). Elevated CEA levels and the presence of extracellular mucin have been shown to have a positive predictive value for mucinous lesions as high as 85% (4). However, the timing and frequency of malignant progression in IPMNs are unknown, and the management of patients with these cystic precursor lesions remains controversial (5, 6), primarily because laboratory, endoscopic, cytologic, and imaging technologies are unable to reliably distinguish between low-risk (low-grade dysplasia) and high-risk (high-grade dysplasia) IPMNs. The degree to which the cyst fluid CEA level is elevated has not been found to predict the grade of dysplasia in IPMNs (7), a finding confirmed in the study by Hata and colleagues. Even microinvasive carcinoma is difficult to recognize without an operation. The recognition that certain somatic mutations (such as *KRAS*) are specific for ductal neoplasia and that others such as *GNAS* and *RNF43* are essentially restricted to IPMNs and MCNs raised the possibility that detection of these mutations in cyst fluid could provide important diagnostic information (8). *KRAS* mutations are present in 78% of IPMNs, either alone or in association with a *GNAS* mutation. Unlike *KRAS* mutations that may be found in other pancreatic cancer precursor lesions, activating *GNAS* mutations are highly specific for IPMNs, found in 58% of cases when considering all histologies compared with 0% of mucinous cystic neoplasms, serous cystadenomas, and solid pseudopapillary neoplasms, which may mimic an IPMN clinically and radiographically. Inactivating mutations in *RNF43* are present in 38% of IPMNs, and when present together with a *GNAS* mutation and other key molecular features provide a 76% sensitivity and 97% specificity in correctly identifying the subtype of pancreatic cyst. Again, however, the presence of these mutations does not indicate a high-risk IPMN and is therefore not an ideal means to determine which IPMNs require surgical resection.

Presently, the most accurate test for identifying high-risk IPMNs is the radiographic finding of a dilated main pancreatic duct (main-duct IPMN). Patients who undergo resection for a main-duct IPMN have a 50%–60% chance of having high-grade dysplasia (9), whereas high-grade dysplasia is present in only 10%–15% of patients who undergo resection in the absence of a dilated main duct (branch duct IPMN; ref. 6). Consensus guidelines now recommend resection for all patients who have dilation of the main pancreatic duct, with observation for branch duct IPMNs that are less than 3 cm in diameter (10). But as Hara and colleagues point out, these criteria are inadequate to identify all IPMNs with high-grade dysplasia or carcinoma, and some high-risk cases based on radiographic criteria prove to have only low-grade dysplasia upon resection. Improving our ability to define the grade of dysplasia in patients with IPMNs would therefore significantly improve

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clinical care. Patients with high-risk lesions could undergo resection prior to the development of invasive disease, and patients with low-risk lesions could be radiographically monitored and avoid a life-threatening operation until high-risk features develop, a strategy already being employed but using suboptimal means to assess the risk of high-grade dysplasia (11).

Because of the limitations in the radiographic and endoscopic determination of high-risk IPMNs, many investigators have looked at other biomarkers that may better predict high-grade dysplasia or carcinoma (12). Array comparative genomic hybridization studies showed that while low-risk IPMNs had no detectable chromosomal abnormalities, high-risk IPMNs had several copy number alterations that included chromosomes 5q, 6q, and 11q (13). However, because these alterations were detected in tumor tissue, it may be difficult to detect them in cyst fluids that contain contaminating normal DNA, limiting utility as a preoperative diagnostic test. Recently reported evidence supports the hypothesis that a proinflammatory environment exists in patients who have high-risk IPMNs. Initial studies in 40 patients with single inflammatory markers revealed that all the patients with low-risk IPMNs had undetectable IL1β levels, whereas 89.5% of patients with high-risk lesions had elevated levels in their cyst fluid (14). IL5 and IL8 were also overexpressed in high-risk IPMNs. Other studies utilizing a bead array with 87 different protein markers on the cyst fluid from patients with resected IPMNs found an AUC in the validation set of 0.80 for a model that incorporated MMP9 and CA72-4.

The study by Hara and colleagues proposes that a telomerase activity level greater than 730 copies/μL of cyst fluid not only allows the detection of mucinous cysts but is highly sensitive and specific for the presence of high-grade dysplasia or carcinoma. Furthermore, the results retain high positive and negative predictive values (73.7% and 90.6%, respectively) for high-risk IPMNs even within the group of 72 cysts with "worrisome features", which represents the key cohort for which surgical management remains a dilemma. Telomerase activity was independent of other conventional clinical and radiographic risk factors as a predictor of high-risk IPMNs. When only the 42 cases of IPMN with worrisome features were considered, the negative predictive value dropped to 78.3%, still a reasonable number but an indication that telomerase measurements will likely show the greatest utility when used in a selected patient population. The patients studied by Hara and colleagues all underwent resection, which indicates a much greater rate of indication for surgery than would be encountered in the general population of pancreatic cyst patients, in whom the pretest probability of high-grade dysplasia or carcinoma would be much lower. These data indeed argue that this relatively simple assay can be used to more effectively stratify the management of patients with ambiguous indications for surgery. Validation in a prospective cohort is clearly indicated to better define how this and other cyst fluid assays can be incorporated into the management algorithm for IPMNs, but the promise of telomerase activity providing significant added value appears great.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

Authors' Contributions

Conception and design: P.J. Allen, D.S. Klimstra
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References

1. Hata T, Dal Molin M, Suenaga M, Yu J, Pittman M, Weiss M, et al. Cyst fluid telomerase activity predicts the histologic grade of cystic neoplasms of the pancreas. Clin Cancer Res 2016;22:5141–51.
2. Maitra A, Fukushima N, Takaori K, Hruban RH. Precursors to invasive pancreatic cancer. Adv Anat Pathol 2005;12:81–91.
3. Brugge WR, Lewandrowski K, Lee-Lewandrowski E, Centeno BA, Szydlo T, Regan S, et al. Diagnosis of pancreatic cystic neoplasms: a report of the cooperative pancreatic cyst study. Gastroenterology 2004;126:1330–6.
4. Walsh RM, Henderson JM, Vogt DP, Baker ME, O'Malley CM Jr, Herts B, et al. Prospective preoperative determination of mucinous pancreatic cystic neoplasms. Surgery 2002;132:628–33.
5. Tanaka M, Fernandez-del Castillo C, Adsay V, Chari S, Falconi M, Jang JY, et al. International consensus guidelines 2012 for the management of IPMN and MCN of the pancreas. Pancreatology 2012;12:183–97.
6. Allen PJ. The management of intraductal papillary mucinous neoplasms of the pancreas. Surg Oncol Clin N Am 2010;19:297–310.
7. Nagula S, Kennedy T, et al. Evaluation of cyst fluid CEA analysis in the diagnosis of mucinous cysts of the pancreas. J Gastrointest Surg 2010;14:1997–2003.
8. Springer S, Wang Y, Dal Molin M, Masica DL, Jiao Y, Kinde I, et al. A combination of molecular markers and clinical features improve the classification of pancreatic cysts. Gastroenterology 2015;149:1501–10.
9. Salvia R, Fernandez-del Castillo C, Bassi C, Thayer SP, Falconi M, Mantovani W, et al. Main-duct intraductal papillary mucinous neoplasms of the pancreas: clinical predictors of malignancy and long-term survival following resection. Ann Surg 2004;239:678–85; discussion 85–7.
10. Tanaka M, Chari S, Adsay V, Fernandez-del Castillo C, Falconi M, Shimizu M, et al. International consensus guidelines for management of intraductal papillary mucinous neoplasms and mucinous cystic neoplasms of the pancreas. Pancreatology 2006;6:17–32.
11. Rocha FG, Lee H, Katabi N, DeMatteo RP, Fong Y, D'Angelica MI, et al. Intraductal papillary neoplasm of the bile duct: a biliary equivalent to intraductal papillary mucinous neoplasm of the pancreas? Hepatology 2012;56:1352–60.
12. Brugge WR, Lauwers GY, Sahani D, Fernandez-del Castillo C, Warshaw AL. Cystic neoplasms of the pancreas. N Engl J Med 2004;351:1218–26.
13. Fritz S, Fernandez-del Castillo C, Mino-Kenudson M, Crippa S, Deshpande V, Lauwers GY, et al. Global genomic analysis of intraductal papillary mucinous neoplasms of the pancreas reveals significant molecular differences compared to ductal adenocarcinoma. Ann Surg 2009;249:440–7.
14. Maker AV, Katabi N, Qin LX, Klimstra DS, Schattner M, Brennan MF, et al. Cyst fluid interleukin-1beta (IL1beta) levels predict the risk of carcinoma in intraductal papillary mucinous neoplasms of the pancreas. Clin Cancer Res 2011;17:1502–8.