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MicroRNAs as Biomarkers of High-risk Pancreatic Cysts: A Possible Holy Grail for the Early Detection of Pancreatic Cancer

Jennifer Permuth-Wey^{1,2}, Christina Georgeades³, Mokenge Malafa²

¹Department of Cancer Epidemiology, H. Lee Moffitt Cancer Center and Research Institute, Tampa, FL, USA

²Department of Gastrointestinal Oncology, H. Lee Moffitt Cancer Center and Research Institute, Tampa, FL, USA

³Morsani College of Medicine, University of South Florida, Tampa, FL, USA

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The clinical challenge

Each year in the United States, more than 150,000 people are diagnosed with an asymptomatic pancreatic cyst when undergoing a computed tomography scan or magnetic resonance imaging for an unrelated medical problem [1]. This scenario involving the detection of an “incidental pancreatic cyst” has become increasingly more common, with radiologists detecting a cyst in up to 2.6% of computed tomography scans and 19.9% of magnetic resonance imaging studies [2,3]. However, their detection represents a double-edged sword, as these cysts can be benign, premalignant, or malignant, and imaging modalities and biomarkers cannot accurately differentiate between these categories. For patients, clinical work-up typically involves an invasive procedure such as endoscopic ultrasound-fine needle aspiration, in which nearly half of these incidentally detected cysts are ultimately diagnosed as intraductal papillary mucinous neoplasms (IPMNs) [4].

IPMNs are one of the only radiographically identifiable lesions that represent premalignant precursors to pancreatic cancer, the deadliest of the top five causes of cancer death in the United States, with a 5-year relative survival rate of only 6% [5]. Thus, the detection of an IPMN is a prime opportunity for intervention before progression to invasive, fatal malignancy. Imagine that a patient’s care can be personalized by offering him or her a blood test to determine the severity of the detected IPMN: the blood test can accurately distinguish “low-risk/benign” IPMNs, which can be monitored, from “high-risk/malignant” IPMNs, which warrant a risky surgical procedure to remove all or part of their pancreas. If this idea of personalized risk stratification based on a blood test sounds like science fiction, it is.

However, researchers are beginning to make important steps toward making this a reality by studying biomarkers known as microRNAs (miRNAs).

IPMNs were initially described in 1982 as mucin-producing tumors of the pancreas. In 1996, they were formally defined by the World Health Organization as “an intraductal papillary mucin-producing neoplasm arising in the main pancreatic duct or its major branches” [6]. Their incidental detection has increased 14-fold between 1985 and 2005 [7]. With the increased frequency of detecting IPMNs comes the need to accurately determine their degree of malignancy, which spans from low- and moderate-grade to high-grade and invasive [2]. IPMNs can also be characterized by ductal involvement and histomorphological type [2]. However, despite advances in imaging, cytology, and molecular studies, there is no accurate way to preoperatively differentiate between low-risk and high-risk IPMNs [8]. Currently, the only way to treat IPMNs and accurately identify the grade of dysplasia is through surgical resection and pathological evaluation, but the rates of morbidity (40% to 50%) and mortality (2% to 4%) associated with an extensive surgery could outweigh the benefits, especially if the disease is low grade [9]. Alternatively, a purely observational approach could result in a missed opportunity to detect invasive disease requiring surgery [10].

miRNAs as biomarkers for the early detection of pancreatic cancer

A hopeful solution exists in the form of biomarker assays. miRNAs are a class of small non-coding RNA molecules that regulate (and serve as) tumor suppressor genes and oncogenes by binding to the 3' untranslated region of the messenger RNA [11]. An important feature of miRNAs is their stability in tissues and biofluids, making them amenable to reliable measurement and excellent candidate biomarkers of early progression to pancreatic malignancy [10].

To date, a growing number of studies have evaluated miRNAs as biomarkers of IPMNs using various biological samples, including tumor tissue, pancreatic cyst fluid, and blood. Three recent studies [8,10,11] investigated genome-wide miRNA expression using archived IPMN tissue pathologically confirmed to be low-risk (low- or moderate-grade) or high-risk (high-grade or invasive). Matthaei *et al.* [10] examined archived tissue (and cyst fluid) from patients with IPMNs and other types of pancreatic cysts and discovered 9 miRNAs (miR-24, miR-18a, miR-30a-3p, miR-92a, miR-106b, miR-342-3p, miR-99b, miR-142-3p, miR-532-3p) that predicted malignant potential with a sensitivity and specificity of 89% and 100%, respectively. Meanwhile, Lubezky *et al.* [11] highlighted 4 different miRNAs (miR-217, miR-21, miR-708, miR-155). A study conducted by Permuth-Wey *et al.* [8] identified 6 miRNAs (miR-100, miR-99b, miR-99a, miR-342-3p, miR-126, and miR-130a) as differentially expressed between groups, with expression levels observed to be down-regulated in high-risk versus low-risk IPMNs. The authors [8] provided evidence to support a tumor suppressor role of the identified miRNAs. Additionally, associations were reported between the miRNAs of interest and relevant clinical and pathological factors [8]. For example, the presence of main pancreatic duct involvement and low levels of preoperative serum albumin levels were associated with decreased expression of the candidate miRNAs in the high-risk compared to the low-risk group, suggesting these factors could potentially be useful in preoperatively determining the grade of IPMN dysplasia [8]. Therefore, the

evaluation of biofluids is warranted in those with newly diagnosed IPMNs and with increased genetic risk for these cysts [8].

miRNAs circulating in pancreatic cyst fluid

Pancreatic cyst fluid represents a rich reservoir of biomarkers of IPMN severity [10]. Historically, markers such as carcinoembryonic antigen, CA19–9, and amylase levels have all been used diagnostically to differentiate between mucinous and nonmucinous cysts, but these markers cannot accurately predict the grade of IPMN dysplasia [12]. Using cyst fluid samples, Matthaei *et al.* [10] identified an miRNA panel that successfully differentiated between different grades of IPMN. The development of next-generation sequencing has also led to increased power and accuracy in testing for differential miRNA expression, with a recent study showing the differential expression between low-risk and high-risk IPMNs for 12 miRNAs [13]. Collectively, findings from these tissue and cyst fluid-based studies show promise for using miRNAs to more accurately characterize IPMNs preoperatively for personalized medical management, but they rely on invasive procedures for specimen collection.

miRNAs circulating in blood

The development of a blood-based test for miRNA detection would provide a minimally invasive, cost-effective, and convenient method to preoperatively characterize IPMN grade. miRNAs are ideal biomarkers for a blood-based test because they are released from tissues into circulation in a stable form and are therefore protected from endogenous RNase activity [14]. Although previous investigations [15–17] have evaluated miRNA expression in blood of pancreatic cancer patients and healthy controls, the implications for early detection were limited because most patients had locally advanced or metastatic disease. Of two studies [18,19] that focused on evaluating blood-based genome-wide miRNA expression in early-stage PDAC patients, one [19] included preoperative serum from 20 patients with IPMNs (4 with low grade and 16 with moderate/high grade). Li *et al.* [19] observed that serum miR-1290 levels were higher among the 20 IPMN patients than levels shown in the control group (area underneath the curve=0.76 [95% CI: 0.61–0.91]). Using novel nCounter technology™ (Nanostring, Inc) to evaluate 800 miRNAs, Permuth-Wey *et al.* [20] showed that a 30-miRNA signature distinguished 42 IPMN cases from 24 healthy individuals without a disease involving their pancreas (AUC = 74.4 [95% CI, 62.3–86.5]; $P=0.002$). The signature included novel miRNAs and miRNAs previously implicated in pancreatic carcinogenesis that had 2- to 4-fold higher expression in patients than in healthy individuals. The group [20] also generated a 5-miRNA signature that discriminated between 21 malignant (high-grade dysplasia and invasive carcinoma) and 21 benign (low- and moderate-grade dysplasia) IPMNs (AUC = 73.2 [95% CI, 57.6–73.2]; $P=0.005$) and showed that paired plasma and tissue samples from patients with IPMNs can have distinct miRNA expression profiles. These blood-based studies [19,20] suggest feasibility of using new cost-effective technology to develop a miRNA-based blood test to aid in preoperative identification of high-risk IPMNs that warrant resection while sparing individuals with low-risk IPMNs and the morbidity associated with overtreatment.

Next steps on the horizon for miRNA diagnostics

Taken together, the future looks bright for development of an effective blood-based miRNA assay to classify newly diagnosed IPMNs preoperatively based on their malignant potential. Large-scale, multicenter studies that prospectively collect biofluids (such as blood and cyst fluid) at several time points from individuals newly diagnosed with IPMNs and other types of pancreatic cysts are needed to advance these efforts. Additionally, to increase diagnostic accuracy and improve outcomes, it will be prudent to integrate novel classes of molecular markers, imaging features, and clinical and epidemiologic variables. Moreover, further assessment of relevant miRNAs and their target genes may be helpful in understanding the progression to pancreatic malignancy so that prevention and early detection methods can be developed [8]. Importantly, a multidisciplinary approach with a team of providers, including radiologists, gastroenterologists, pancreatic surgeons, oncologists, and epidemiologists, will be essential to effectively risk-stratify patients and provide personalized care [18].

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References

1. Sachs T, Pratt WB, Callery MP, Vollmer CM Jr. The incidental asymptomatic pancreatic lesion: nuisance or threat? *J. Gastrointest. Surg* 13(3), 405–415 (2009). [PubMed: 19130153]
2. Matthaei H, Schulick RD, Hruban RH, Maitra A. Cystic precursors to invasive pancreatic cancer. *Nat. Rev. Gastroenterol. Hepatol* 8(3), 141–150 (2011). [PubMed: 21383670]
3. Megibow AJ, Baker ME, Gore RM, Taylor A. The incidental pancreatic cyst. *Radiol. Clin. North Am* 49(2), 349–359 (2011). [PubMed: 21333781]
4. Hruban RH, Maitra A, Kern SE, Goggins M. Precursors to pancreatic cancer. *Gastroenterol. Clin. North Am* 36(4), 831–849, vi (2007). [PubMed: 17996793]
5. American Cancer Society. Cancer Facts and Figures 2015 (American Cancer Society, Atlanta, GA, 2015).
6. Kawamoto S, Horton KM, Lawler LP, Hruban RH, Fishman EK. Intraductal papillary mucinous neoplasm of the pancreas: can benign lesions be differentiated from malignant lesions with multidetector CT? *Radiographics*. 25(6), 1451–1468 (2005). [PubMed: 16284127]
7. Klibansky DA, Reid-Lombardo KM, Gordon SR, Gardner TB. The clinical relevance of the increasing incidence of intraductal papillary mucinous neoplasm. *Clin. Gastroenterol. Hepatol* 10(5), 555–558 (2012). [PubMed: 22210438]
8. Permuth-Wey J, Chen YA, Fisher K et al. A genome-wide investigation of microRNA expression identifies biologically-meaningful microRNAs that distinguish between high-risk and low-risk intraductal papillary mucinous neoplasms of the pancreas. *PLoS One*. 10(1), e0116869 (2015). [PubMed: 25607660]
9. Hines OJ, Reber HA. Pancreatic surgery. *Curr. Opin. Gastroenterol* 24(5), 603–611 (2008). [PubMed: 19122502]
10. Matthaei H, Wylie D, Lloyd MB et al. miRNA biomarkers in cyst fluid augment the diagnosis and management of pancreatic cysts. *Clin. Cancer Res* 18(17), 4713–4724 (2012). [PubMed: 22723372]

11. Lubezky N, Loewenstein S, Ben-Haim M et al. MicroRNA expression signatures in intraductal papillary mucinous neoplasm of the pancreas. *Surgery*. 153(5), 663–672 (2013). [PubMed: 23305591]
12. Rockacy M, Khalid A. Update on pancreatic cyst fluid analysis. *Ann. Gastroenterol* 26(2), 122–127 (2013). [PubMed: 24714589]
13. Wang J, Paris PL, Chen J et al. Next generation sequencing of pancreatic cyst fluid microRNAs from low grade-benign and high grade-invasive lesions. *Cancer Lett*. 356(2 Pt B), 404–409 (2015). [PubMed: 25304377]
14. Mitchell PS, Parkin RK, Kroh EM et al. Circulating microRNAs as stable blood-based markers for cancer detection. *Proc. Natl. Acad. Sci. U S A* 105(30), 10513–10518 (2008). [PubMed: 18663219]
15. Liu R, Chen X, Du Y et al. Serum microRNA expression profile as a biomarker in the diagnosis and prognosis of pancreatic cancer. *Clin. Chem* 58(3), 610–618 (2012). [PubMed: 22194634]
16. Schultz NA, Dehlendorff C, Jensen BV et al. MicroRNA biomarkers in whole blood for detection of pancreatic cancer. *JAMA*. 311(4), 392–404 (2014). [PubMed: 24449318]
17. Wang J, Chen J, Chang P et al. MicroRNAs in plasma of pancreatic ductal adenocarcinoma patients as novel blood-based biomarkers of disease. *Cancer Prev. Res. (Phila)* 2(9), 807–813 (2009). [PubMed: 19723895]
18. Law JK, Hruban RH, Lennon AM. Management of pancreatic cysts: a multidisciplinary approach. *Curr. Opin. Gastroenterol* 29(5), 509–516 (2013). [PubMed: 23872487]
19. Li A, Yu J, Kim H et al. MicroRNA array analysis finds elevated serum miR-1290 accurately distinguishes patients with low-stage pancreatic cancer from healthy and disease controls. *Clin. Cancer Res* 19(13), 3600–3610 (2013). [PubMed: 23697990]
20. Permuth-Wey J, Chen DT, Fulp WJ. Plasma microRNAs as novel biomarkers of disease for patients with intraductal papillary mucinous neoplasms of the pancreas. *Cancer Prev. Res. (Phila)* In Press (2015).