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Genetic Markers of Malignant Transformation in Intraductal Papillary Mucinous Neoplasm of the Pancreas:

A Meta-Analysis

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Abstract

Objectives—The objective of this study was to determine the relationship between specific genetic alterations and malignant transformation in intraductal papillary mucinous neoplasm (IPMN) of the pancreas.

Methods—Quantitative meta-analysis was conducted of studies through October 2010 that adhered to the 1996 World Health Organization guidelines for distinguishing adenoma and borderline IPMN versus carcinoma in surgically resected specimens using a random-effects model. We developed a 6-point scoring system to assess study quality.

Results—Thirty-nine studies (1235 IPMN samples) satisfied the inclusion criteria, and we conducted pooled analysis of 8 genetic markers: *MUC1*, *MUC2*, *MUC5AC*, *kRas*, *p53*, *hTERT* (human telomerase reverse transcriptase), *cyclooxygenase 2*, and *Shh* (Sonic hedgehog). Markers having the strongest association with malignant IPMN were *hTERT* (odds ratio [OR], 11.4; 95% confidence interval [CI], 3.5–36.7) and *Shh* (OR, 6.9; 95% CI, 2.4–20.2), whereas *MUC5AC* (OR, 1.0; 95% CI, 0.1–13.9) and *kRas* (OR, 2.0; 95% CI, 1.0–4.3) showed weak association with IPMN histologic progression.

Conclusions—Expression of *hTERT* is strongly associated with malignant transformation in IPMN, consistent with up-regulation of *hTERT* as a key step in progression of IPMN to cancer. Expression of *kRas* and *MUC5AC* is common but not strongly associated with IPMN histologic progression. The quality criteria used here may guide future reporting of genetic markers related to malignant transformation of IPMN.

Keywords

telomerase; mucin; pancreatic cancer; intraductal papillary mucinous neoplasm; IPMN; Sonic hedgehog; cyclooxygenase 2; p53; *hTERT*; *Cox2*; *kRas*; marker

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Intraductal papillary mucinous neoplasms (IPMNs) describe papillary proliferations of the exocrine pancreas epithelium that secrete copious amounts of thick mucin causing cystic dilatation of the ducts.¹ Since the pathologic entity was first described by Ohashi et al in 1982 and designated *intraductal papillary mucinous neoplasm* by Sessa et al² in 1994, it has been characterized radiologically, histologically, and molecularly and is thought to represent a lesion that is distinct from and less clinically aggressive than ductal adenocarcinoma.³⁻⁵

Histologic analysis of IPMN samples reveals a spectrum of progressive cytoarchitectural atypia. The spectrum of this histologic progression is reflected in the 1996 World Health Organization (WHO) classification of IPMN into 3 categories based on increasing nuclear atypia and mitotic rate: adenoma (IPM-A), borderline (IPM-B), and carcinoma (IPM-C, both in situ and invasive).⁶ The histologic progression of IPMN from a benign (IPM-A or IPM-B) into a malignant (IPM-C) lesion has a significant impact on patient survival. In patients who underwent pancreatic resection between 1990 and 2007, the 5-year survival rate of patients with benign IPMN ranged from 89% to 95% versus 63% to 65% for patients with malignant IPMN.⁷⁻⁹ Moreover, malignant transformation is not rare: the frequency of malignancy in IPMNs in the main pancreatic duct ranges from 60% to 92% in various reports.¹⁰⁻¹⁵

A longstanding question has been whether the histologic progression of IPMN reflects an accumulation of genetic mutations leading to increasing atypia. Many studies have therefore sought to identify specific genetic mutations associated with malignant transformation in IPMN. The objective of the present study was to enhance our understanding of malignant progression in IPMN based on literature published to date. Specifically, our aim was to perform a quantitative meta-analysis of studies from the past 14 years to determine the relationship between individual genetic alterations and malignant transformation in IPMN.

METHODS

Search Methods and Study Selection Criteria

A computerized literature search was performed independently in the PubMed (National Library of Medicine, Bethesda, Md), Cochrane Library, and EMBASE databases by 2 of the authors (S.N., G.E.I.). To identify studies investigating gene expression in IPMN, we used the following search terms: “intraductal papillary mucinous neoplasm,” “IPMN,” “intraductal papillary mucinous tumor,” “IPMT,” “IPMA,” “IPMB,” “IPMC,” “gene expression,” “molecular marker,” “*Ras*,” “*p53*,” “*MUC*,” “telomerase,” and “mutations.” Bibliographies from relevant publications were further reviewed to identify additional published articles not indexed by the major databases.

Studies published from January 1996 to October 2010 were potentially eligible for inclusion based on publication of WHO guidelines for classification of IPMN in 1996.¹ For the present meta-analysis, we included studies that satisfied the following criteria: (1) human subjects, (2) histologic confirmation of IPMN in surgically resected tissue, (3) classification of IPMN in accordance to WHO guidelines, (4) inclusion of data regarding genotype frequency and/or risk estimates, and (5) use of validated molecular methods for genotyping.

Individual case reports, editorials, review articles, and duplicate publications were excluded. We further excluded studies that incorporated patients with chronic pancreatitis, bile duct IPMN, and high-risk familial cohorts.

Data Extraction

Data were extracted independently by each reviewer using a standardized data abstraction form. Any disagreements between the 2 reviewers were examined by all 3 investigators and resolved by consensus.

Data Synthesis

The primary outcome was defined as IPMN with presence of malignant transformation (IPM-C). During the initial phase of data extraction, IPMN grade was extracted as presented in the original publication. For purposes of the present meta-analysis, IPMN grade was further dichotomized to either benign (IPM-A, IPM-B) or malignant (IPM-C) lesions.

Assessment of Study Quality

There is currently no validated method to rate the quality of observational studies on IPMN. To provide a means to appraise the methodologic quality of studies included in the present meta-analysis, we formulated a scoring system based on recommendations from the STROBE¹⁶ and PRAISE guidelines.¹⁷ This checklist consisted of 6 factors summarized in Figure 1A: review of histologic classification of IPMN, in the case of dysplasia or carcinoma review of histology by at least 2 independent pathologists, an explicit definition of genetic assay positivity, blinded genetic assay, description of statistical analysis, and mention of potential sources of bias. One point was assigned for the presence of each of the individual factors.

Statistical Analysis

Effect size was expressed as an odds ratio (OR) with the corresponding 95% confidence interval (CI). Quantitative meta-analysis was performed when 3 or more studies evaluated the same gene candidate. In the meta-analysis, pooled ORs were generated based on the individual studies using a random-effects model (DerSimonian and Laird) chosen based on the ability to incorporate between-study variance potentially related to differences among the populations included among studies. The I^2 test was used to evaluate study heterogeneity. For genetic markers with 5 or more studies, we performed further subgroup analyses based on methodologic quality (high quality, 3 points). Analyses were performed using Quantitative Meta-analysis version 2.2 (Biostat, Englewood, NJ).

RESULTS

The computerized literature search yielded 253 studies. Abstracts of these studies were reviewed, and 119 studies were excluded for the reasons delineated in Figure 1B. A full manuscript review was performed on the remaining 134 studies, and 95 additional studies were excluded. The most common reasons for exclusion were studies totaling less than 3 for a particular gene marker and studies that did not stratify IPMN samples by histologic grade according to the 1996 WHO guidelines.

A total of 39 studies between January 1996 and October 2010 (representing a total of 1235 IPMN samples) met our predetermined inclusion criteria (Table 1). A pooled analysis was performed to determine the risk of malignant transformation for each genetic marker. Results are summarized according to individual gene candidates.

MUC Expression

Mucins are large, heavily glycosylated proteins that are differentially expressed in epithelial cells of glandular tissues and various tumor types.^{19,21,57} Of the 19 mucin genes identified, *MUC1*, *MUC2*, and *MUC5AC* genes have been most frequently characterized in the pancreas. Eleven studies with 417 IPMN samples reported *MUC* expression.^{18–21,23–26,58} These studies examined *MUC* expression at the mRNA level by in situ hybridization²⁰ or at the protein level by immunohistochemistry (IHC).^{18,19,21,23–26,58} Because the expression of different mucin genes appears to vary with grade of IPMN, we pooled studies according to analysis of the most frequently reported mucin genes: *MUC1*, *MUC2*, and *MUC5AC*. Figure 2 summarizes the individual ORs.

Eight studies representing a total of 322 IPMN samples examined the expression of *MUC1* by IHC.^{18–25} *MUC1* was expressed in 8.6% (15/174) of IPM-A/B samples and 35.8% (53/148) of IPM-C samples. The pooled OR for *MUC1* and malignant transformation was 5.9 (95% CI, 1.8–19.8). Grouped according to study quality, high-quality studies' pooled OR was 3.6 (95% CI, 0.4–32.1), whereas low-quality studies' pooled OR was 7.4 (95% CI, 1.7–31.2).

Eight studies representing a total of 322 IPMN sample examined the expression of *MUC2* by IHC.^{18–25} *MUC2* was expressed in 51.7% (90/174) of IPM-A/B samples and 68.9% (102/148) of IPM-C samples. The overall pooled OR for *MUC2* and malignant transformation was 4.2 (95% CI, 1.7–10.1). Grouped according to study quality, high-quality studies' pooled OR was 14.5 (95% CI, 4.1–51.8), whereas low-quality studies' pooled OR was 1.3 (95% CI, 0.4–4.4).

Three studies representing a total of 231 IPMN samples examined the expression of *MUC5AC* by IHC.^{20,24,26} Five studies were excluded from pooled analysis because all IPMN samples were positive for *MUC5AC*.^{18,21,25,58,59} *MUC5AC* was expressed in 84.7% (149/176) of IPM-A/B samples and 92.4% (97/105) of IPM-C samples. The pooled OR for *MUC5AC* and malignant transformation was 1.0 (95% CI, 0.1–13.9).

Oncogenic *kRas* Expression

Activating point mutations in the GTP-binding protein *kRas* have been found in virtually all advanced pancreatic ductal adenocarcinoma and are thought to be an important step in pancreatic oncogenesis.⁶⁰ Thirteen studies representing a total of 285 samples examined the presence of the *kRas* oncogenic mutation by polymerase chain reaction (PCR) and sequencing^{24,27–36,39,40} or IHC²⁹ on resected tissue. Figure 3 summarizes the individual ORs.

In pooled analysis, the oncogenic mutation in *kRas* was found in 48.3% (71/147) of IPM-A/B samples and 55.1% (76/138) of IPM-C samples. The pooled OR for *kRas* mutation and

malignant transformation was 2.0 (95% CI, 1.0–4.3). Grouped according to study quality, high-quality studies' pooled OR was 1.3 (95% CI, 0.4–4.2), whereas low-quality studies' pooled OR was 2.8 (95% CI, 1.1–7.1).

p53 Nuclear Expression

Most studies investigating p53 mutations in IPMN examined nuclear immunostaining of p53. This positivity criterion is based on the observation that wild-type p53 protein is present in the nucleus only at low levels, but mutant forms common to many types of cancer accumulate in the nucleus and can be visualized by IHC. Nuclear localization of p53 has been investigated as a prognostic factor in several kinds of gastrointestinal tumors including pancreatic cancer,⁶¹ hepatocellular cancer,⁶² colorectal cancer,⁶³ and gastric cancer.⁶⁴

Six studies^{24,41,42,44–46} measured p53 mutations according to recommendations by Kawai et al,⁶⁵ in which samples considered positive had greater than 10% of cells with positive nuclear immunostaining of p53. These 6 studies representing a total of 293 IPMNs identified positive p53 staining in 16.9% (31/183) of IPM-A/B samples and 40.9% (45/110) of IPM-C samples.^{24,41,42,44–46} Three studies used a threshold of 5% positive cells to define positive p53 staining.^{21,39,43} Together, these 3 studies examined 111 IPMN samples and identified positive staining in 17.1% (13/76) of IPM-A/B samples and 27.5% (11/40) of IPM-C samples.^{21,39,43} Uemura et al³² examined 15 IPMN samples and, using a criterion of focal aggregates of more than 30 cells with positive nuclear staining, found positive p53 staining in 0% (0/7) of IPM-A/B samples and 25% (2/8) of IPM-C samples. Ueda et al²³ examined 24 IPMN samples and found that no IPM-A/B samples had any p53-positive nuclear staining, but 27% (3/11) of IPM-C samples had scattered or diffuse positive cells. Mueller et al³¹ performed PCR and single-strand conformation polymorphism analysis to find sequence changes in 13 IPMN samples and in this manner identified mutations in 10% (1/10) of IPM-A/B samples and 33.3% (1/3) of IPM-C samples. Wada et al³⁴ used fluorescence-labeled microsatellite markers to detect loss of heterozygosity in the 17p13 chromosomal locus of *p53* and found loss of heterozygosity in no IPM-A/B samples but in 66.7% (6/9) of IPM-C samples.

Pooling all the above studies examining p53 mutations by IPMN histologic grade, a total of 478 IPMN samples are examined, of which p53 mutations were identified in 14.9% (45/302) of IPM-A/B samples and 38.6% (68/176) of IPM-C samples. The pooled OR for p53 mutations and malignant transformation was 6.3 (95% CI, 2.9–13.8). Grouped according to study quality, high-quality studies' pooled OR was 3.5 (95% CI, 1.3–9.6), whereas low-quality studies' pooled OR was 15.6 (95% CI, 4.4–55.3). Figure 4 summarizes the individual ORs.

Telomerase Expression

The human telomerase reverse transcriptase (*hTERT*) gene encodes the catalytic component of telomerase required to overcome telomere shortening and cellular senescence. Therefore, activation of *hTERT* is considered a hallmark of cancer.^{66,67} Three studies representing a total of 82 samples examined expression of *hTERT* by IHC^{48,49} or reverse transcriptase–PCR⁴⁷ on resected tissue specimens (Fig. 5). *hTERT* expression was found in 23.7% (9/38)

of IPM-A/B samples and 88.6% (39/44) of IPM-C samples. The pooled OR for hTERT mutation and malignant transformation was 11.4 (95% CI, 3.5–36.7).

Cyclooxygenase 2 Expression

The biosynthesis of prostaglandins by the cyclooxygenase 2 (*Cox2*) enzyme is thought to mediate many properties of carcinogenesis. Expression of *Cox2* is upregulated in pancreatic cancer^{68,69} and is thought to stimulate invasion⁷⁰ and angiogenesis.⁷¹ Five studies representing a total of 158 IPMN samples examined the expression of *Cox2* by IHC (Fig. 5).^{24,50–53} *Cox2* was expressed in 53.3% (48/90) of IPM-A/B samples and 75.4% (52/69) of IPM-C samples. The pooled OR for *Cox2* and malignant transformation was 2.9 (95% CI, 1.1–7.7). Grouped according to study quality, high-quality studies' pooled OR was 2.6 (95% CI, 0.6–11.9), whereas low-quality studies' pooled OR was 3.1 (95% CI, 0.9–11.1).

Shh Expression

The secreted factor Sonic hedgehog (*Shh*) has an important role in regulating normal pancreas development and has been implicated in tumorigenesis in pancreatic ductal adenocarcinoma.⁷² Three studies representing a total of 136 IPMN samples examined the expression of *Shh* by IHC (Fig. 5).^{54–56} *Shh* was expressed in 68.7% (57/83) of IPM-A/B samples and 90.6% (48/53) of IPM-C samples. The pooled OR for *Shh* expression and malignant transformation was 6.9 (95% CI, 2.4–20.2).

Heterogeneity and Impact of Quality

The estimated I^2 was low (<25%) for studies evaluating *telomerase*, *Cox2*, and *Shh*. Estimated I^2 for studies on *kRas* and *p53* was moderate, 32% and 36%, respectively. Estimated I^2 exceeded 50% for each of the MUC genes analyzed (*MUC1*, 54%; *MUC2*, 68%; and *MUC5AC*, 79%). Heterogeneity was significantly lower among high-quality studies of *MUC2* (I^2 , 0%) and *MUC5AC* (I^2 , 16%). However, this was not the case for *MUC1* (I^2 among high-quality studies, 63%).

DISCUSSION

We have performed a quantitative meta-analysis of genetic markers associated with histologic progression of IPMN. Through a computerized literature search of online databases using predetermined inclusion criteria, we identified 39 studies between January 1996 and October 2010 (representing a total of 1235 IPMN samples) that examined the expression of 8 different genetic markers in benign versus malignant IPMN: *MUC1*, *MUC2*, *MUC5AC*, *kRas*, *p53*, *hTERT*, *Cox2*, and *Shh*. Pooled analysis of these studies revealed expression of *hTERT*, *Shh*, and *MUC1* to have the strongest association with malignant progression of IPMN and expression of *MUC5AC* to have the weakest association with malignant progression of IPMN.

Intraductal papillary mucinous neoplasm of the pancreas has presented challenges in terms of pathophysiology and clinical management. Although the pathologic entity has been recognized for decades, the mechanisms of malignant transformation remain poorly understood. A key hypothesis has been that IPMN is fundamentally a genetic lesion and that

an accumulation of somatic mutations drives the histologic progression, ultimately leading to malignant transformation. This hypothesis is a basis for pursuit of genetic markers that can be utilized to improve diagnosis, guide optimal management, and potentially design new therapeutic targets. In this meta-analysis, we investigated the hypothesis that specific genetic alterations are associated with the histologic progression of IPMN.

The risk of cancer associated with various gene mutations ranged from 1.0 to 11.41. The marker found to have the strongest association with malignant IPMN was *hTERT* (OR, 11.4; 95% CI, 3.5–36.7). One interpretation of this finding is that the genetic alterations leading to abnormal expression of *hTERT* occur later in the histologic progression of IPMN toward cancer. This is consistent with the hypothesis of “telomere crisis” in carcinogenesis. This model postulates that telomeres are progressively shortened with cell proliferation until cells reach “crisis” at which point most cells will die; the ability of rare cells to overcome crisis by upregulating *hTERT* is a critical step in carcinogenesis.⁶⁶ Shortening of telomeres has been reported in some IPM-A but has been noted to progressively worsen with histologic progression.⁴⁹ Therefore, the dramatic upregulation of *hTERT* observed in malignant compared with benign IPMN supports the notion of a crisis point in the development of malignancy.⁴⁹

In the pooled analysis, we also identified markers that had little or no association with malignant progression of IPMN. Although fairly common, *MUC5AC* and *kRas* were not strongly associated with malignancy among IPMN lesions. A weaker association with malignant progression may suggest that alterations leading to expression of these markers occur early in the histologic progression of IPMN toward cancer. Activating mutations of *kRas* are recognized to be among the earliest mutations leading to pancreatic cancer, detected in more than 40% of early pancreatic intraepithelial neoplasia lesions.^{60,73,74} Alternatively, markers with no association with malignant IPMN may have no role in the pathobiology of IPMN progression. This could be the case with *MUC5AC*, which had no association (OR, 1.0; 95% CI, 0.1–13.9) as opposed to *MUC1*, which had a strong association (OR, 5.9; 95% CI, 1.8–19.8) with malignant transformation. *MUC5AC* is a secreted mucin speculated to form a protective gel around tumors,⁷⁵ a property that may be advantageous for all neoplasms, benign or malignant. In contrast, *MUC1* is a membrane-associated mucin that has been demonstrated to bind and signal through β -catenin and mitogen-activated protein kinase pathways critical in cell proliferation, and thus upregulation of *MUC1* may be necessary for the progression of IPMN from benign to malignant lesions.^{76–78} Interestingly, a knockout of *Muc1* in mice significantly slowed the growth rate of oncogene-induced breast tumors and decreased the rates of tumor metastasis.⁷⁹

A proposed model for genetic alterations associated with malignant progression of IPMN based on the present meta-analysis is presented in Figure 6. How well these ORs in fact correspond to the pathobiology of IPMN progression remains to be validated in future functional studies. A key question is whether the genetic markers are so-called “passenger” or “driver” alterations: do these genetic alterations cause IPMN lesions to acquire malignant behavior, or are they simply a consequence of other genetic alterations that in fact drive IPMN progression?

The strengths of the approach taken in this meta-analysis include a comprehensive and unbiased search of IPMN literature, the use of standardized systematic review and meta-analysis techniques using predefined inclusion and exclusion criteria, and elaboration of a scoring system to assess quality of included studies.

There were several limitations to the present study. In this meta-analysis, IPMN lesions were stratified according to the WHO IPM-A/B versus IPM-C grades, but insufficient data were available to stratify according to histologic cell type (eg, gastric, intestinal, pancreatobiliary, oncocytic). There are also potential biases derived from limiting our search to published studies. Unpublished data may have an increased proportion of null results in which no association is detected between a genetic marker and IPMN progression. The genetic markers eligible for pooled analysis were limited by the number of studies that met the inclusion criteria. A number of markers such as *p16*^{23,43} and *Smad4*^{80,81} have been frequently investigated but have not been used as consistent criteria for measuring genetic alterations. Additional candidate genetic markers that have been examined in IPMN tissue were unable to be included in the quantitative meta-analysis because of availability of fewer than 3 published reports in the medical literature. Finally, as it is derived from published reports, this meta-analysis is limited to previously characterized markers. Unbiased approaches such as RNASeq will be critical to identify novel genetic alterations that play a role in malignant progression of IPMN.

We developed a 6-point scoring system based on the STROBE¹⁶ and PRAISE reporting guidelines¹⁷ for observational studies. We found considerable variability in quality among the studies included in the present meta-analysis. In particular, many studies failed to confirm the histologic diagnosis of malignancy by 2 independent pathologists. In our analysis, we found that the risk of malignant transformation associated with a number of gene mutations varied according to study quality. Therefore, we advocate establishment of standardized quality reporting criteria to reduce heterogeneity and enhance accuracy of future studies evaluating genetic risk factors for malignant transformation of IPMN.

Ultimately, the motivation for this and future studies is to elucidate genetic markers that can improve diagnosis, guide optimal management, and offer new therapeutic targets. In addition, elucidating the genetic alterations underlying malignant transformation of IPMN may have relevance to pancreatic cancer. A recent meta-analysis identified *Cox2* as a marker associated with poor survival outcome in pancreatic cancer.⁸² Here, we similarly find that *Cox2* expression is associated with malignant IPMN (OR, 2.9), highlighting potential parallels in the pathogenesis of malignant IPMN and pancreatic cancer.

In summary, numerous studies have evaluated candidate gene mutations associated with malignant transformation in IPMN. Using quantitative meta-analysis, we identified a strong association of *hTERT* expression with malignant transformation of IPMN, consistent with up-regulation of *hTERT* as a key step in progression of IPMN to cancer. Although *kRas* was a commonly detected mutation, its presence was not strongly associated with histologic progression of IPMN. In the context of this meta-analysis, we have also proposed a set of quality criteria for reporting of genetic studies related to malignant transformation of IPMN.

We believe the present findings can be used as a framework to help guide further research aimed at elucidating the genetic basis of malignant progression in IPMN of the pancreas.

Abbreviations

CI	confidence interval
Cox2	cyclooxygenase 2
OR	odds ratio
IPMN	intraductal mucinous neoplasm
<i>hTERT</i>	human telomerase reverse transcriptase
<i>Shh</i>	Sonic hedgehog
IHC	immunohistochemistry

References

1. Hruban RH, Takaori K, Klimstra DS, et al. An illustrated consensus on the classification of pancreatic intraepithelial neoplasia and intraductal papillary mucinous neoplasms. *Am J Surg Pathol.* 2004; 28(8):977–987. [PubMed: 15252303]
2. Sessa F, Solcia E, Capella C, et al. Intraductal papillary-mucinous tumours represent a distinct group of pancreatic neoplasms: an investigation of tumour cell differentiation and K-ras, p53 and c-erbB-2 abnormalities in 26 patients. *Virchows Arch.* 1994; 425(4):357–367. [PubMed: 7820300]
3. Shimada K, Sakamoto Y, Sano T, et al. Invasive carcinoma originating in an intraductal papillary mucinous neoplasm of the pancreas: a clinicopathologic comparison with a common type of invasive ductal carcinoma. *Pancreas.* 2006; 32:281–287. [PubMed: 16628084]
4. Maire F, Hammel P, Terris B, et al. Prognosis of malignant intraductal papillary mucinous tumours of the pancreas after surgical resection. Comparison with pancreatic ductal adenocarcinoma. *Gut.* 2002; 51(5):717–722. [PubMed: 12377813]
5. Sohn TA, Yeo CJ, Cameron JL, et al. Intraductal papillary mucinous neoplasms of the pancreas: an increasingly recognized clinicopathologic entity. *Ann Surg.* 2001; 234(3):313–321. discussion 312–321. [PubMed: 11524584]
6. Kloppel, G.; Solcia, E.; Longnecker, D., et al. Histological Typing of Tumours of the Exocrine Pancreas. World Health Organization International Histological Classification of Tumours. 2. Berlin: Springer-Verlag; 1996.
7. Jang JY, Kim SW, Ahn YJ, et al. Multicenter analysis of clinicopathologic features of intraductal papillary mucinous tumor of the pancreas: is it possible to predict the malignancy before surgery? *Ann Surg Oncol.* 2005; 12(2):124–132. [PubMed: 15827792]
8. Murakami Y, Uemura K, Hayashidani Y, et al. Predictive factors of malignant or invasive intraductal papillary-mucinous neoplasms of the pancreas. *J Gastrointest Surg.* 2007; 11(3):338–344. [PubMed: 17458608]
9. Shin SH, Han DJ, Park KT, et al. Validating a simple scoring system to predict malignancy and invasiveness of intraductal papillary mucinous neoplasms of the pancreas. *World J Surg.* 2010; 34(4):776–783. [PubMed: 20127242]
10. Kobari M, Egawa S, Shibuya K, et al. Intraductal papillary mucinous tumors of the pancreas comprise 2 clinical subtypes: differences in clinical characteristics and surgical management. *Arch Surg.* 1999; 134(10):1131–1136. [PubMed: 10522860]
11. Doi R, Fujimoto K, Wada M, et al. Surgical management of intraductal papillary mucinous tumor of the pancreas. *Surgery.* 2002; 132(1):80–85. [PubMed: 12110799]

12. Kitagawa Y, Unger TA, Taylor S, et al. Mucus is a predictor of better prognosis and survival in patients with intraductal papillary mucinous tumor of the pancreas. *J Gastrointest Surg.* 2003; 7:12–18. discussion 18–19. [PubMed: 12559180]
13. Sugiyama M, Izumisato Y, Abe N, et al. Predictive factors for malignancy in intraductal papillary-mucinous tumours of the pancreas. *Br J Surg.* 2003; 90(10):1244–1249. [PubMed: 14515294]
14. Salvia R, Fernández-del Castillo C, Bassi C, 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(5):678–685. discussion 677–685. [PubMed: 15082972]
15. Takuma K, Kamisawa T, Anjiki H, et al. Predictors of malignancy and natural history of main-duct intraductal papillary mucinous neoplasms of the pancreas. *Pancreas.* 2011; 40(3):371–375. [PubMed: 21206326]
16. Sanderson S, Tatt ID, Higgins JP. Tools for assessing quality and susceptibility to bias in observational studies in epidemiology: a systematic review and annotated bibliography. *Int J Epidemiol.* 2007; 36(3):666–676. [PubMed: 17470488]
17. Moher D, Liberati A, Tetzlaff J, et al. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. *Ann Intern Med.* 2009; 151(4):264–269. W264. [PubMed: 19622511]
18. Luttges J, Zamboni G, Longnecker D, et al. The immunohistochemical mucin expression pattern distinguishes different types of intraductal papillary mucinous neoplasms of the pancreas and determines their relationship to mucinous noncystic carcinoma and ductal adenocarcinoma. *Am J Surg Pathol.* 2001; 25(7):942–948. [PubMed: 11420467]
19. Adsay NV, Merati K, Andea A, et al. The dichotomy in the preinvasive neoplasia to invasive carcinoma sequence in the pancreas: differential expression of *MUC1* and *MUC2* supports the existence of two separate pathways of carcinogenesis. *Mod Pathol.* 2002; 15(10):1087–1095. [PubMed: 12379756]
20. Terris B, Dubois S, Buisine MP, et al. Mucin gene expression in intraductal papillary-mucinous pancreatic tumours and related lesions. *J Pathol.* 2002; 197(5):632–637. [PubMed: 12210083]
21. Ito H, Endo T, Oka T, et al. Mucin expression profile is related to biological and clinical characteristics of intraductal papillary-mucinous tumors of the pancreas. *Pancreas.* 2005; 30:e96–e102. [PubMed: 15841035]
22. Moriya T, Kimura W, Semba S, et al. Biological similarities and differences between pancreatic intraepithelial neoplasias and intraductal papillary mucinous neoplasms. *Int J Gastrointest Cancer.* 2005; 35(2):111–119. [PubMed: 15879625]
23. Ueda M, Miura Y, Kunihiro O, et al. *MUC1* overexpression is the most reliable marker of invasive carcinoma in intraductal papillary-mucinous tumor (IPMT). *Hepatogastroenterology.* 2005; 52(62):398–403. [PubMed: 15816444]
24. Jang JY, Park YC, Song YS, et al. Increased K-ras mutation and expression of S100A4 and *MUC2* protein in the malignant intraductal papillary mucinous tumor of the pancreas. *J Hepatobiliary Pancreat Surg.* 2009; 16(5):668–674. [PubMed: 19412570]
25. Cao W, Adley BP, Liao J, et al. Mucinous nonneoplastic cyst of the pancreas: apomucin phenotype distinguishes this entity from intraductal papillary mucinous neoplasm. *Hum Pathol.* 2010; 41:513–521. [PubMed: 19954814]
26. Kanno A, Satoh K, Kimura K, et al. The expression of *MUC4* and *MUC5AC* is related to the biologic malignancy of intraductal papillary mucinous neoplasms of the pancreas. *Pancreas.* 2006; 33:391–396. [PubMed: 17079945]
27. Z'graggen K, Rivera JA, Compton CC, et al. Prevalence of activating K-ras mutations in the evolutionary stages of neoplasia in intraductal papillary mucinous tumors of the pancreas. *Ann Surg.* 1997; 226(4):491–498. discussion 498–500. [PubMed: 9351717]
28. Mulligan NJ, Yang S, Andry C, et al. The role of p21ras in pancreatic neoplasia and chronic pancreatitis. *Hum Pathol.* 1999; 30(6):602–610. [PubMed: 10374765]
29. Paal E, Thompson LD, Przygodzki RM, et al. A clinicopathologic and immunohistochemical study of 22 intraductal papillary mucinous neoplasms of the pancreas, with a review of the literature. *Mod Pathol.* 1999; 12(5):518–528. [PubMed: 10349991]

30. Yoshizawa K, Nagai H, Sakurai S, et al. Clonality and K-ras mutation analyses of epithelia in intraductal papillary mucinous tumor and mucinous cystic tumor of the pancreas. *Virchows Arch.* 2002; 441(5):437–443. [PubMed: 12447672]
31. Mueller J, Gansauge S, Mattfeldt T. P53 mutation but not p16/MTS1 mutation occurs in intraductal papillary mucinous tumors of the pancreas. *Hepatogastroenterology.* 2003; 50(50):541–544. [PubMed: 12749268]
32. Uemura K, Hiyama E, Murakami Y, et al. Comparative analysis of K-ras point mutation, telomerase activity, and p53 overexpression in pancreatic tumours. *Oncol Rep.* 2003; 10(2):277–283. [PubMed: 12579258]
33. Kitago M, Ueda M, Aiura K, et al. Comparison of K-ras point mutation distributions in intraductal papillary-mucinous tumors and ductal adenocarcinoma of the pancreas. *Int J Cancer.* 2004; 110(2): 177–182. [PubMed: 15069678]
34. Wada K, Takada T, Yasuda H, et al. Does “clonal progression” relate to the development of intraductal papillary mucinous tumors of the pancreas? *J Gastrointest Surg.* 2004; 8:289–296. [PubMed: 15019925]
35. Kobayashi N, Inamori M, Fujita K, et al. Characterization of K-ras gene mutations in association with mucinous hypersecretion in intraductal papillary-mucinous neoplasms. *J Hepatobiliary Pancreat Surg.* 2008; 15(2):169–177. [PubMed: 18392710]
36. Schonleben F, Allendorf JD, Qiu W, et al. Mutational analyses of multiple oncogenic pathways in intraductal papillary mucinous neoplasms of the pancreas. *Pancreas.* 2008; 36:168–172. [PubMed: 18376308]
37. Schonleben F, Qiu W, Bruckman KC, et al. BRAF and KRAS gene mutations in intraductal papillary mucinous neoplasm/carcinoma (IPMN/IPMC) of the pancreas. *Cancer Lett.* 2007; 249:242–248. [PubMed: 17097223]
38. Schonleben F, Qiu W, Remotti HE, et al. PIK3CA, KRAS, and BRAF mutations in intraductal papillary mucinous neoplasm/carcinoma (IPMN/C) of the pancreas. *Langenbecks Arch Surg.* 2008; 393(3):289–296. [PubMed: 18343945]
39. Chadwick B, Willmore-Payne C, Tripp S, et al. Histologic, immunohistochemical, and molecular classification of 52 IPMNs of the pancreas. *Appl Immunohistochem Mol Morphol.* 2009; 17(1): 31–39. [PubMed: 18813127]
40. Fritz S, Fernandez-del Castillo C, Mino-Kenudson M, 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(3):440–447. [PubMed: 19247032]
41. Biankin AV, Biankin SA, Kench JG, et al. Aberrant p16(INK4A) and DPC4/Smad4 expression in intraductal papillary mucinous tumours of the pancreas is associated with invasive ductal adenocarcinoma. *Gut.* 2002; 50(6):861–868. [PubMed: 12010891]
42. Jinfeng M, Kimura W, Sakurai F, et al. Histopathological study of intraductal papillary mucinous tumor of the pancreas: special reference to the roles of Survivin and p53 in tumorigenesis of IPMT. *Int J Gastrointest Cancer.* 2002; 32:73–81. [PubMed: 12794243]
43. Sasaki S, Yamamoto H, Kaneto H, et al. Differential roles of alterations of p53, p16, and SMAD4 expression in the progression of intraductal papillary-mucinous tumors of the pancreas. *Oncol Rep.* 2003; 10(1):21–25. [PubMed: 12469138]
44. Nishikawa N, Kimura Y, Okita K, et al. Intraductal papillary mucinous neoplasms of the pancreas: an analysis of protein expression and clinical features. *J Hepatobiliary Pancreat Surg.* 2006; 13(4): 327–335. [PubMed: 16858545]
45. Abe K, Suda K, Arakawa A, et al. Different patterns of p16INK4A and p53 protein expressions in intraductal papillary-mucinous neoplasms and pancreatic intraepithelial neoplasia. *Pancreas.* 2007; 34:85–91. [PubMed: 17198188]
46. Miyasaka Y, Nagai E, Yamaguchi H, et al. The role of the DNA damage checkpoint pathway in intraductal papillary mucinous neoplasms of the pancreas. *Clin Cancer Res.* 2007; 13:4371–4377. [PubMed: 17671118]
47. Nodari F, Baiocchi GL, Salvi A, et al. Telomerase gene expression in intraductal papillary-mucinous tumors (IPMT): preliminary findings. *Acta Biomed.* 2003; 74(suppl 2):59–64. [PubMed: 15055037]

48. Hashimoto Y, Murakami Y, Uemura K, et al. Detection of human telomerase reverse transcriptase (hTERT) expression in tissue and pancreatic juice from pancreatic cancer. *Surgery*. 2008; 143:113–125. [PubMed: 18154939]
49. Hashimoto Y, Murakami Y, Uemura K, et al. Telomere shortening and telomerase expression during multistage carcinogenesis of intraductal papillary mucinous neoplasms of the pancreas. *J Gastrointest Surg*. 2008; 12(1):17–28. discussion 19–28. [PubMed: 17960465]
50. Koshiba T, Hosotani R, Miyamoto Y, et al. Immunohistochemical analysis of cyclooxygenase-2 expression in pancreatic tumors. *Int J Pancreatol*. 1999; 26(2):69–76. [PubMed: 10597402]
51. Kokawa A, Kondo H, Gotoda T, et al. Increased expression of cyclooxygenase-2 in human pancreatic neoplasms and potential for chemoprevention by cyclooxygenase inhibitors. *Cancer*. 2001; 91:333–338. [PubMed: 11180079]
52. Aoki T, Nagakawa Y, Tsuchida A, et al. Expression of cyclooxygenase-2 and vascular endothelial growth factor in pancreatic tumors. *Oncol Rep*. 2002; 9(4):761–765. [PubMed: 12066205]
53. Nijima M, Yamaguchi T, Ishihara T, et al. Immunohistochemical analysis and in situ hybridization of cyclooxygenase-2 expression in intraductal papillary-mucinous tumors of the pancreas. *Cancer*. 2002; 94:1565–1573. [PubMed: 11920515]
54. Jang KT, Lee KT, Lee JG, et al. Immunohistochemical expression of Sonic hedgehog in intraductal papillary mucinous tumor of the pancreas. *Appl Immunohistochem Mol Morphol*. 2007; 15:294–298. [PubMed: 17721274]
55. Liu MS, Yang PY, Yeh TS. Sonic hedgehog signaling pathway in pancreatic cystic neoplasms and ductal adenocarcinoma. *Pancreas*. 2007; 34:340–346. [PubMed: 17414057]
56. Satoh K, Kanno A, Hamada S, et al. Expression of Sonic hedgehog signaling pathway correlates with the tumorigenesis of intraductal papillary mucinous neoplasm of the pancreas. *Oncol Rep*. 2008; 19(5):1185–1190. [PubMed: 18425375]
57. Kim YS, Gum JR Jr. Diversity of mucin genes, structure, function, and expression. *Gastroenterology*. 1995; 109(3):999–1001. [PubMed: 7657131]
58. Ji Y, Lou WH, Jin DY, et al. A series of 64 cases of pancreatic cystic neoplasia from an institutional study of China. *World J Gastroenterol*. 2006; 12(45):7380–7387. [PubMed: 17143961]
59. Handra-Luca A, Flejou JF, Rufat P, et al. Human pancreatic mucinous cystadenoma is characterized by distinct mucin, cytokeratin and CD10 expression compared with intraductal papillary-mucinous adenoma. *Histopathology*. 2006; 48:813–821. [PubMed: 16722930]
60. Hezel AF, Kimmelman AC, Stanger BZ, et al. Genetics and biology of pancreatic ductal adenocarcinoma. *Genes Dev*. 2006; 20(10):1218–1249. [PubMed: 16702400]
61. Harada N, Gansauge S, Gansauge F, et al. Nuclear accumulation of p53 correlates significantly with clinical features and inversely with the expression of the cyclin-dependent kinase inhibitor p21(WAF1/CIP1) in pancreatic cancer. *Br J Cancer*. 1997; 76(3):299–305. [PubMed: 9252195]
62. Bressac B, Galvin KM, Liang TJ, et al. Abnormal structure and expression of p53 gene in human hepatocellular carcinoma. *Proc Natl Acad Sci U S A*. 1990; 87(5):1973–1977. [PubMed: 2155427]
63. Rodrigues NR, Rowan A, Smith ME, et al. p53 mutations in colorectal cancer. *Proc Natl Acad Sci U S A*. 1990; 87(19):7555–7559. [PubMed: 1699228]
64. Uchino S, Noguchi M, Hirota T, et al. High incidence of nuclear accumulation of p53 protein in gastric cancer. *Jpn J Clin Oncol*. 1992; 22(4):225–231. [PubMed: 1279244]
65. Kawai A, Noguchi M, Beppu Y, et al. Nuclear immunoreaction of p53 protein in soft tissue sarcomas. A possible prognostic factor. *Cancer*. 1994; 73(10):2499–2505. [PubMed: 8174045]
66. Maser RS, DePinho RA. Connecting chromosomes, crisis, and cancer. *Science*. 2002; 297(5581):565–569. [PubMed: 12142527]
67. O'Hagan RC, Chang S, Maser RS, et al. Telomere dysfunction provokes regional amplification and deletion in cancer genomes. *Cancer Cell*. 2002; 2(2):149–155. [PubMed: 12204535]
68. Okami J, Yamamoto H, Fujiwara Y, et al. Overexpression of cyclooxygenase-2 in carcinoma of the pancreas. *Clin Cancer Res*. 1999; 5(8):2018–2024. [PubMed: 10473081]
69. Yip-Schneider MT, Barnard DS, Billings SD, et al. Cyclooxygenase-2 expression in human pancreatic adenocarcinomas. *Carcinogenesis*. 2000; 21(2):139–146. [PubMed: 10657949]

70. Ito H, Duxbury M, Benoit E, et al. Prostaglandin E2 enhances pancreatic cancer invasiveness through an Ets-1-dependent induction of matrix metalloproteinase-2. *Cancer Res.* 2004; 64(20): 7439–7446. [PubMed: 15492268]
71. Chu J, Lloyd FL, Trifan OC, et al. Potential involvement of the cyclooxygenase-2 pathway in the regulation of tumor-associated angiogenesis and growth in pancreatic cancer. *Mol Cancer Ther.* 2003; 2(1):1–7. [PubMed: 12533667]
72. Morris, JPt; Wang, SC.; Hebrok, M. KRAS, Hedgehog, Wnt and the twisted developmental biology of pancreatic ductal adenocarcinoma. *Nat Rev Cancer.* 2010; 10(10):683–695. [PubMed: 20814421]
73. Klimstra DS, Longnecker DS. K-ras mutations in pancreatic ductal proliferative lesions. *Am J Pathol.* 1994; 145(6):1547–1550. [PubMed: 7992857]
74. Hruban RH, van Mansfeld AD, Offerhaus GJ, et al. K-ras oncogene activation in adenocarcinoma of the human pancreas. A study of 82 carcinomas using a combination of mutant-enriched polymerase chain reaction analysis and allele-specific oligonucleotide hybridization. *Am J Pathol.* 1993; 143(2):545–554. [PubMed: 8342602]
75. Hollingsworth MA, Swanson BJ. Mucins in cancer: protection and control of the cell surface. *Nat Rev Cancer.* 2004; 4(1):45–60. [PubMed: 14681689]
76. Li Y, Bharti A, Chen D, et al. Interaction of glycogen synthase kinase 3beta with the *DF3/MUC1* carcinoma-associated antigen and beta-catenin. *Mol Cell Biol.* 1998; 18(12):7216–7224. [PubMed: 9819408]
77. Schroeder JA, Thompson MC, Gardner MM, et al. Transgenic *MUC1* interacts with epidermal growth factor receptor and correlates with mitogen-activated protein kinase activation in the mouse mammary gland. *J Biol Chem.* 2001; 276(16):13057–13064. [PubMed: 11278868]
78. Yamamoto M, Bharti A, Li Y, et al. Interaction of the *DF3/MUC1* breast carcinoma-associated antigen and beta-catenin in cell adhesion. *J Biol Chem.* 1997; 272(19):12492–12494. [PubMed: 9139698]
79. Spicer AP, Rowse GJ, Lidner TK, et al. Delayed mammary tumor progression in Muc-1 null mice. *J Biol Chem.* 1995; 270(50):30093–30101. [PubMed: 8530414]
80. Sunamura M, Lefter LP, Duda DG, et al. The role of chromosome 18 abnormalities in the progression of pancreatic adenocarcinoma. *Pancreas.* 2004; 28:311–316. [PubMed: 15084978]
81. Inoue H, Furukawa T, Sunamura M, et al. Exclusion of SMAD4 mutation as an early genetic change in human pancreatic ductal tumorigenesis. *Genes Chromosomes Cancer.* 2001; 31(3):295–299. [PubMed: 11391801]
82. Jamieson NB, Carter CR, McKay CJ, et al. Tissue biomarkers for prognosis in pancreatic ductal adenocarcinoma: a systematic review and meta-analysis. *Clin Cancer Res.* 2011; 17(10):3316–3331. [PubMed: 21444679]

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FIGURE 1. Methodology used in this meta-analysis. A, We developed a 6-point scoring system to assess quality of studies based on recommendations from the STROBE¹⁶ and PRAISE¹⁷ guidelines. B, Flow diagram of the literature search and study selection.

FIGURE 2.

Forest plot of studies examining association of *MUC1* (A), *MUC2* (B), or *MUC5AC* (C) expression and malignant transformation of IPMN. Methodologic quality of each study was assigned a score from 1 to 6 based on criteria summarized in Table 1. For each study, the quality score, OR; 95% CI, and relative weight are shown. The size of the data markers (squares) represents the statistical weight that each study contributed to the random-effects summary estimates; horizontal lines represent the 95% CI. The diamonds indicate the summary OR. I^2 test P values evaluating study heterogeneity are shown.

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FIGURE 3.

Forest plot of studies examining association of *kRas* expression and malignant transformation of IPMN.

FIGURE 4.

Forest plot of studies examining association of altered *p53* expression and malignant transformation of IPMN.

FIGURE 5.

Forest plot of studies examining association of *hTERT* (A), *Cox2* (B), or *Shh* (C) expression and malignant transformation of IPMN.

FIGURE 6.

A model for genetic alterations associated with malignant progression of IPMN based on the findings of this meta-analysis.

TABLE 1

Studies Meeting Inclusion Criteria for Meta-Analysis Organized by Genetic Marker

First Author	Year	Total No. IPMNs	Study Area	Gene	Assay, Definition of Positivity	OR	95% CI	Study Quality (0-6)
Luttges et al ¹⁸	2001	51	Germany, Italy, United States	<i>MUC1</i>	IHC	22.077	1.210–402.833	1
Adsay et al ¹⁹	2002	74	United States	<i>MUC1</i>	>10% Cells positive by IHC	3.158	0.636–15.671	2
Terris et al ²⁰	2002	57	France	<i>MUC1</i>	IHC	21.375	3.861–118.345	1
Ito et al ²¹	2005	21	Japan	<i>MUC1</i>	>5% Cells positive by IHC	55.8	2.246–1386.315	5
Moriya et al ²²	2005	37	Japan	<i>MUC1</i>	>10% Cells positive by IHC	3.125	0.377–25.918	3
Ueda et al ²³	2005	24	Japan	<i>MUC1</i>	IHC	31.909	1.523–668.749	1
Jang et al ²⁴	2009	41	South Korea	<i>MUC1</i>	>10% Cells positive by IHC	0.733	0.123–4.367	3
Cao et al ²⁵	2010	17	United States	<i>MUC1</i>	IHC	0.333	0.014–7.875	1
Luttges et al ¹⁸	2001	51	Germany, Italy, United States	<i>MUC2</i>	IHC	0.316	0.016–6.288	1
Adsay et al ¹⁹	2002	74	United States	<i>MUC2</i>	>10% Cells positive by IHC	2.659	0.970–7.286	2
Terris et al ²⁰	2002	57	France	<i>MUC2</i>	IHC	0.296	0.090–0.975	1
Ito et al ²¹	2005	21	Japan	<i>MUC2</i>	>5% Cells positive by IHC	6.810	0.321–144.629	5
Moriya et al ²²	2005	37	Japan	<i>MUC2</i>	>10% Cells positive by IHC	11.250	1.245–101.635	3
Ueda et al ²³	2005	24	Japan	<i>MUC2</i>	IHC	2.700	0.507–14.372	1
Jang et al ²⁴	2009	41	South Korea	<i>MUC2</i>	>10% Cells positive by IHC	22.500	3.689–137.235	3
Cao et al ²⁵	2010	17	United States	<i>MUC2</i>	IHC	7.000	0.306–160.322	1
Terris et al ²⁰	2002	57	France	<i>MUC5AC</i>	IHC	0.096	0.017–0.547	1
Kanno et al ²⁶	2006	51	Japan	<i>MUC5AC</i>	IHC	11.471	0.617–213.276	4
Jang et al ²⁴	2009	41	South Korea	<i>MUC5AC</i>	>10% Cells positive by IHC	1.714	0.298–9.869	3
Z'graggen et al ²⁷	1997	16	United States	<i>kRas</i>	Sequence mutation	6.000	0.257–140.045	4
Mulligan et al ²⁸	1999	7	United States	<i>kRas</i>	Sequence mutation	1.500	0.055–40.633	3
Paal et al ²⁹	1999	22	United States	<i>kRas</i>	Immunohistochemistry	41.000	1.115–1507.260	2
Yoshizawa et al ³⁰	2002	7	Japan	<i>kRas</i>	Sequence mutation	18.000	0.812–399.154	2
Mueller et al ³¹	2003	13	Germany	<i>kRas</i>	Sequence mutation	1.167	0.074–18.346	2
Uemura et al ³²	2003	15	Japan	<i>kRas</i>	Sequence mutation	4.000	0.447–35.788	2
Kitago et al ³³	2004	20	Japan	<i>kRas</i>	Sequence mutation	1.364	0.112–16.577	3

First Author	Year	Total No. IPMNs	Study Area	Gene	Assay, Definition of Positivity	OR	95% CI	Study Quality (0-6)
Wada et al ³⁴	2004	23	Japan	<i>kRas</i>	Sequence mutation	3.063	0.472–19.879	2
Kobayashi et al ³⁵	2008	4	Japan	<i>kRas</i>	Sequence mutation	0.200	0.005–8.825	2
Schonleben et al ³⁶	2008	38	United States	<i>kRas</i>	Sequence mutation	1.429	0.270–7.549	2
Schonleben et al ³⁷	2007	same data as above	United States	<i>kRas</i>	Sequence mutation			2
Schonleben et al ³⁸	2008	same data as above	United States	<i>kRas</i>	Sequence mutation			1
Chadwick et al ³⁹	2009	52	United States	<i>kRas</i>	Sequence mutation	0.406	0.119–1.381	3
Fritz et al ⁴⁰	2009	20	United States	<i>kRas</i>	Sequence mutation	0.333	0.047–2.366	3
Jang et al ²⁴	2009	41	South Korea	<i>kRas</i>	Sequence mutation	5.867	1.395–24.673	4
Blankin et al ⁴¹	2002	18	Australia	<i>p53</i>	>10% Cells positive by IHC	3.889	0.543–27.866	4
Jinfeng et al ⁴²	2002	22	Japan	<i>p53</i>	>10% Cells positive by IHC	25.000	1.168–535.241	2
Mueller et al ³¹	2003	13	Germany	<i>p53</i>	PCR amplification and single-strand conformation polymorphism analysis of exons 5, 6, 7, 8	4.500	0.190–106.823	2
Sasaki et al ⁴³	2003	38	Japan	<i>p53</i>	>5% Cells positive by IHC	33.000	1.647–661.051	2
Uemura et al ³²	2003	15	Japan	<i>p53</i>	focal aggregates of >30 positive cells with nucleus stained brown	5.769	0.232–143.371	2
Wada et al ³⁴	2004	23	Japan	<i>p53</i>	Fluorescence-labeled microsatellite markers to detect loss of heterozygosity of 17p13	50.143	2.242–1121.352	1
Ito et al ²¹	2005	21	Japan	<i>p53</i>	>5% Cells positive by IHC	3.250	0.340–31.074	5
Ueda et al ²³	2005	24	Japan	<i>p53</i>	A few positive cells, scattered staining, diffuse staining	11.118	0.508–243.105	1
Nishikawa et al ⁴⁴	2006	37	Japan	<i>p53</i>	>10% Cells positive by IHC	3.300	0.475–22.942	4
Abe et al ⁴⁵	2007	47	Japan	<i>p53</i>	>10% Cells positive by IHC	4.688	1.016–21.619	4
Miyasaka et al ⁴⁶	2007	128	Japan	<i>p53</i>	>10% Cells positive by IHC	25.000	3.153–198.205	4
Chadwick et al ³⁹	2009	52	United States	<i>p53</i>	>5% Cells positive by IHC	0.535	0.146–1.962	3
Jang et al ²⁴	2009	41	South Korea	<i>p53</i>	>10% Cells positive by IHC	8.767	0.458–167.937	3
Nodari et al ⁴⁷	2003	2	Italy	<i>Telomerase</i>	Reverse transcriptase-PCR	9.000	0.097–831.780	0
Hashimoto et al ⁴⁸	2008	12	Japan	<i>Telomerase</i>	IHC	39.667	1.279–1229.867	4
Hashimoto et al ⁴⁹	2008	68	Japan	<i>Telomerase</i>	IHC	9.750	2.676–35.528	5
Koshiba et al ⁵⁰	1999	26	Japan	<i>Cox2</i>	IHC	23.000	1.070–494.573	2
Kokawa et al ⁵¹	2001	29	Japan	<i>Cox2</i>	>5% Cells positive by IHC	1.697	0.332–8.666	2
Aoki et al ⁵²	2002	30	Japan	<i>Cox2</i>	IHC	5.833	1.119–30.403	3
Nijijima et al ⁵³	2002	32	Japan	<i>Cox2</i>	>10% Cells positive by IHC	1.244	0.262–5.915	3

First Author	Year	Total No. IPMNs	Study Area	Gene	Assay, Definition of Positivity	OR	95% CI	Study Quality (0–6)
Jang et al ²⁴	2009	41	South Korea	<i>Cox2</i>	IHC	2.955	0.310–28.140	2
Jang et al ⁵⁴	2007	55	South Korea	<i>Shh</i>	IHC	6.691	1.946–23.000	3
Liu et al ⁵⁵	2007	18	Taiwan	<i>Shh</i>	IHC	10.455	0.425–256.957	0
Satoh et al ⁵⁶	2008	63	Japan	<i>Shh</i>	>20% Cells positive by IHC	5.940	0.322–109.490	4