

Pancreatic Cancer: From Genes to Patient Care

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There has been a revolution in our understanding of the molecular biology of pancreatic cancer. In the last 5 years, pancreatic cancer has gone from one of the most poorly understood cancers to one of the best. It is now clear that pancreatic cancer is a genetic disease – a disease of inherited and acquired mutations in cancer-causing genes. These alterations include the activation of an oncogene, *K-ras*, in ~90% of the cancers; the inactivation of several tumor-suppressor genes including *p16* (~95% of the cancers), *p53* (50-70%), *DPC4* (55%), and *BRCA2* (10%, almost all inherited); and microsatellite instability in 4%.

TABLE 1: Molecular Alterations in Invasive Pancreatic Adenocarcinoma

Gene (Chromosomal Region)	# of tumors and/or cell lines studied	% of Tumors with <u>Genetic Alteration</u>	Reference
K-ras(12p)	82	>90%	(1)
p16(9p)	37	>95%	(2)
p53(17p)	27	50%-70%	(3)
DPC4(18q)	84	55%	(4)
ATK2 (19q)	18	10%-20%	(5)
MYB (6q)	35	10%	(6)
AIB1 (20q)	9	10%	(7)
BRCA2(13q)	42	7%-10%	(8)
LKB1/STK11(19p)	100	<5%	(9)
MKK4(17p)	137**	<5%	(10)
TGFβ-R1 (9q) or TGFβ-R2 (3p)	97	<5%	(11)
RB1(13q)	62	<5%	(12)

**45 of these were prescreened for LOH

An understanding of the genetic alterations associated with the development of pancreatic cancer is important, not only because these genetic alterations may one day form the basis of screening tests, but also because we can use molecular genetics to understand the familial aggregation of pancreatic cancer and histopathologically distinct neoplasms that arise in the pancreas.

Familial Pancreatic Cancer

It has been estimated that familial aggregation and genetic susceptibility play a role in as many as 10% of pancreatic cancers (13). The genetic basis for this aggregation is only now coming to light. To date, at least five genetic syndromes associated with the familial aggregation of pancreatic cancer have been identified. These include

1. Familial breast cancer with germline mutations in the *BRCA2* gene (8).
2. Familial Atypical Multiple Mole Melanoma (FAMMM) Syndrome with germline mutations in the *p16* gene (14).
3. The Peutz-Jeghers syndrome with germline mutations in the *STK11/LKB1* gene (9).
4. Hereditary Nonpolyposis Colorectal Cancer (HNPCC) with germline mutations in one of the DNA mismatch repair genes (15) and
5. Hereditary pancreatitis with germline mutations in the cationic trypsinogen gene (16; 17).

TABLE 2: Genetic Disorders and Germ-line Genetic Alterations Associated with Familial Pancreatic Cancer

Disorder	Gene (location)	Increased Risk of Pancreatic Cancer	References
Hereditary Pancreatitis	<i>PRSS1</i> (7q35)	50x	(16)
Hereditary Nonpolyposis Colorectal Cancer Lynch II variant	<i>hMSH2, hMLH1</i>	?	(15)
Hereditary Breast and Ovarian Cancer	<i>BRCA2</i> (13q12-q13)	10x	(18);(8)
Familial Atypical Multiple Mole Melanoma Syndrome (FAMMM)	<i>p16</i> (9p21)	20x	(14)
Peutz-Jeghers Syndrome	<i>STK11/LKB1</i> (19p13)	132x	(19)

Family member of kindreds with one of these syndromes can now be screened to see if they inherited one of these mutations. Those who are found to carry a germline mutation can benefit from careful cancer screening programs or even prophylactic surgery, while those found not to carry a mutation will be relieved of their anxiety.

These syndromes, however, do not explain the vast majority of cases in which there is a familial aggregation of pancreatic cancer. We have established the National Familial Pancreas Tumor Registry (NFPTR) with the hope of identifying the causes of the aggregation of pancreatic cancer in these kindred. Physicians wishing to refer patients to this research registry may contact the NFPTR at:

National Familial Pancreas Tumor Registry
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720 Rutland Avenue
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We have followed kindred with an aggregation of pancreatic cancer and found that apparently healthy members of these kindred have an elevated risk for prospectively developing pancreatic cancer (20). These kindred may benefit from novel screening tests.

In addition to a better understanding of the familial aggregation of pancreatic cancer, a better understanding of the genetics of pancreatic cancer has improved our understanding of the variants of pancreatic cancer and led to the development of new markers that can aid pathologists evaluating biopsies of the pancreas. Let us look at a few examples.

New Markers of pancreatic cancer:

Two types of new immunohistochemical markers have recently been developed for invasive ductal adenocarcinomas of the pancreas. The first are markers that are lost in the immune cancers, while the second are markers that are overexpressed by the cancers.

DPC4 is genetically deleted in ~55% of pancreatic carcinomas and this deletion is relatively specific for pancreatic cancer (it is only rarely seen in other tumors). Wilentz et al. have recently demonstrated that immunohistochemical labeling for the DPC4 gene product (Anti-DPC4 Antibody B8, DAKO) mirrors DPC4 gene status (21). Neoplasms with genetic inactivation of DPC4 show complete loss of labeling with this antibody. The loss of immunolabeling for DPC4 therefore can be used to support the diagnosis of carcinoma in difficult pancreatic biopsies and the loss of immunolabeling for DPC4 in a metastatic neoplasm of unknown primary can also be used to suggest a pancreatic primary is a metastatic carcinoma of unknown primary.

Serial Analysis of Gene Expression (SAGE) and gene expression arrays have recently been used to identify genes overexpressed in pancreatic cancer. For example, prostate stem cell antigen (PSCA) and mesothelin were both found to be overexpressed in SAGE libraries of pancreatic cancer (<http://www.ncbi.nlm.nih.gov/SAGE/>) and immunohistochemical labeling has confirmed that they are both specifically overexpressed in pancreatic ductal carcinomas as compared to normal (22).

Medullary carcinoma of the pancreas

The clinical and pathologic features of carcinomas of the pancreas with DNA replication errors (RER+) have recently been characterized. Goggins et al. screened 82 carcinomas of the pancreas for DNA replication errors using polymerase chain reaction amplification of microsatellite markers (15). Three (3.7%) of the carcinomas were RER+ and all 3 had medullary histology. That is, they were poorly differentiated, and they all had a syncytial growth pattern, and pushing borders. All 3 carcinomas were also wild-type for *K-ras*. Furthermore, patients with this

distinctive type of neoplasm seem to have a better prognosis than patients with the usual infiltrating ductal carcinoma of the pancreas, and they often have a family history of cancer. Based on these findings, Goggins et al. suggested that this newly recognized type of pancreatic cancer should be classified separately.

Furthermore, neoplasms with DNA replication errors may be caused by inherited (germline) mutations in one of the DNA mismatch repair genes (*hMSH2*, *hMLH1*, etc.). Germline mutations in DNA mismatch repair genes are associated with the development of a variety of neoplasms including colorectal cancer, and therefore patients with RER+ pancreatic carcinomas may benefit from genetic screening. Of interest, we recently have seen a patient with synchronous colonic and pancreatic carcinomas, both of which had medullary histology. Both neoplasms showed a high degree of microsatellite instability (RER+), and immunohistochemical stains for the *hMLH1* gene product were negative, suggesting inactivation of this mismatch repair gene. It is likely that this patient had a germline mutation in the *hMLH1* mismatch repair gene and that he developed two synchronous carcinomas, one in the colon and one in the pancreas. The following course of action appears appropriate once you recognize a medullary carcinoma of the pancreas.

Medullary histology → get immunostains for hMLH1 and hMSH2 → If loss of labeling is seen → test for MSI → If microsatellite unstable, check family history, consider genetic testing.

Undifferentiated carcinoma with osteoclast-like giant cells

These rare neoplasms are typically well circumscribed, yellow-pink, and fleshy. By light microscopy, they are composed of multinucleated, benign-appearing, osteoclast-like giant cells dispersed among infiltrating atypical mononuclear cells. The multinucleated giant cells in these tumors more closely resemble the osteoclast of resorbing bone than the pleomorphic giant cells of the anaplastic giant cell carcinoma. Furthermore, these giant cells are consistently reactive for monocyte/macrophage markers such as KP-1. Some osteoclast-like giant tumors of the pancreas are associated with mucinous cystic neoplasms or an adenocarcinoma, suggesting an epithelial origin to these neoplasms. Indeed, at the molecular level, these distinctive neoplasms frequently harbor activating point mutations in codon 12 of *K-ras*. Furthermore, the same *k-ras* mutations have been demonstrated in both the epithelial (that is, the mucinous cystic neoplasm or adenocarcinoma) and the infiltrating mononuclear cell components of these neoplasms, helping to establish that undifferentiated carcinomas with osteoclast-like giant cells arise from the epithelial components. These neoplasms are therefore best considered carcinomas which elicit a non-neoplastic giant cell response and not mesenchymal neoplasms. The term “undifferentiated carcinoma with osteoclast-like giant cells” is therefore preferred over “osteoclast-like giant cell tumor (23).”

Pancreatoblastomas and other variants

S. Abraham and colleagues have recently characterized the genetic alterations in a number of neoplastic variants in the pancreas, including pancreatoblastoma, solid and pseudopapillary tumors, and acinar cell carcinomas (24). Remarkably, these morphologically distinct neoplasms have genetic profiles that are distinct from usual ductal adenocarcinomas of the pancreas (Table 3). For example, loss of heterozygosity at 11p is common in pancreaticoblastomas, while alterations in the APC/ β -catenin pathway are seen in virtually all solid and pseudopapillary tumors.

TABLE 3: Molecular Alterations in Pancreatic Neoplasms

Gene	Ductal Adenocarcinoma	Pancreatoblastoma	Solid-Pseudopapillary Tumor	Acinar Cell Carcinoma
K-ras	90%	0%	0%	0%
DPC4	55%	25%	0%	0%
p53	50%-70%	0%	16%	0%
LOH 11p	40%	86%	-	50%
APC/ β -catenin	0%	67% (5/8 sporadic β -catenin) (1/1 FAP with APC)	90%	24%

From, S. Abraham et al

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