Pancreatic cancer is one of the most lethal neoplasms, with five-year survival rates of less than 10%. It also is the fourth-leading cause of cancer deaths in Canada and is seen mainly in urbanized nations of the world. The peak incidence of pancreatic cancer is in the seventh to eighth decades of life and the disease rarely occurs prior to the age of 50. Males have a slightly higher risk than females — this difference being highest in the younger age groups.1

**Genetics Of Pancreatic Cancer: What Are The Risks?**

Although the risk of pancreatic cancer and ultimate prognosis in asymptomatic mutation carriers is yet to be defined, genetic screening may be justified in the future.

**By Geeta Lal, MD, MSc, FRCSC; and Steven Gallinger, MD, MSc, FRCSC**

Pancreatic cancer is one of the most lethal neoplasms, with five-year survival rates of less than 10%. It also is the fourth-leading cause of cancer deaths in Canada and is seen mainly in urbanized nations of the world. The peak incidence of pancreatic cancer is in the seventh to eighth decades of life and the disease rarely occurs prior to the age of 50. Males have a slightly higher risk than females — this difference being highest in the younger age groups.1

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Primary pancreatic malignancies may be of epithelial or mesenchymal origin. Ductal adenocarcinoma, an infiltrating epithelial malignancy that forms glandular ductal structures, is the most common primary pancreatic malignancy, representing 75% of all primary nonendocrine pancreatic tumors. This tumor type will form the focus of the rest of this article.

Patients with ductal pancreatic adenocarcinoma usually present with weight loss, jaundice and pain. Surgery remains the mainstay of treatment for resectable disease and is the only potentially curative treatment option available to date. Radiation therapy in combination with chemotherapy may be beneficial for locally advanced, unresectable disease.

In spite of improvements in all these modalities, five-year survival rates from this malignancy remain low. The most important reason for this poor prognosis is that greater than 80% of patients present with unresectable tumors at the time of diagnosis. This has prompted a better understanding of etiologic factors, which may help identify the disease at an earlier stage and improve survival.

Etiology

Environmental Factors — Diet, Lifestyle and Occupation

Several studies have implicated high-fat diets and meat products, coupled with a decreased intake of fruits and vegetables, as risk factors for pancreatic cancer. An association also has been suggested between alcohol and coffee consumption and increased pancreatic cancer risk. Exposure to industrial carcinogens, particularly b-naphthylamine and benzidine, as well as ionizing radiation, has been reported to play a significant role in the development of pancreatic cancer.
causative role in this malignancy. The overall contribution of these exposures to pancreatic cancer risk, however, is not significant. Cigarette smoking is the most consistent lifestyle risk factor for developing pancreatic cancer, with a dose-response relationship between the number of pack-years (one pack per day times one year) smoked and pancreatic cancer risk.4

**Acquired Host Factors — Past Medical History**
Pancreatic cancer risk may be elevated in patients who have undergone gastric surgery for peptic ulcer disease. Although the mechanism of this increased risk has not been elucidated, some investigators have suggested that it may be related to hypo-acidity. Hypo-acidity results in bacterial overgrowth and increased production of circulating carcinogens, particularly N-nitroso compounds.5 A history of chronic pancreatitis also has been associated with an elevated risk for pancreatic cancer.6 Diabetes mellitus has long been associated with pancreatic cancer, but the controversy over cause versus effect has been resolved by recent studies that suggest no etiologic relationship between the two conditions.7,8

**Inherited Host Factors — Genetic Susceptibility**
Recently, two lines of evidence have emerged to suggest that genetic factors are important determinants of pancreatic cancer risk. The first comes from case series and epidemiologic studies which report that a family history of pancreatic cancer is an important risk factor for the disease.9-11 The second comes from the observation that this malignancy occurs in excess of expected frequencies in the presence of several familial cancer syndromes. These syndromes are associated with germline mutations in various cancer-predisposing genes and are discussed in the following sections.

Cigarette smoking is the most consistent lifestyle risk factor for developing pancreatic cancer.

**Cancer Genetics: Basic Concepts**
Unlike other “traditional” genetic diseases (e.g., cystic fibrosis) that result from inherited mutations, cancers result from both inherited mutations and somatically acquired changes. Each of these alterations provide an individual cell and its progeny with a growth advantage over surrounding cells and, ultimately, lead to a transformed phenotype.12 Genes predisposing to cancer development usually play key roles in cellu-
Figure 1. Cancer predisposing genes.
lar growth regulation.

**Oncogenes, Tumor Suppressor Genes And Mismatch Repair Genes**

Oncogenes were initially recognized as the transforming genetic material of deoxyribonucleic acid (DNA) and ribonucleic acid (RNA) viruses. Subsequently, similar sequences called proto-oncogenes were discovered in the genome of normal host cells.13 Proto-oncogenes include growth factors and their receptors, intracellular transducer proteins, transcription factors and cell-cycle-control proteins. Oncogenes arise from the activation of proto-oncogenes by various mechanisms, such as somatic point mutations, amplification and gene rearrangements. These changes cause the protein products of the oncogenes to be constitutively active, or lead to their increased transcription and expression; thus, leading to a malignant phenotype.14 Mutated oncogenes, therefore, lead to a gain of function in a dominant fashion.

In contrast, under normal circumstances, tumor suppressor genes act as barriers to cell proliferation and growth. Knudson was the first to propose a mechanism for the role of tumor suppressor genes in tumorigenesis through his statistical work on retinoblastoma and Wilm’s tumor.15 His “two-hit” hypothesis suggested the development of these hereditary tumors was the result of an inherited mutated allele and an acquired mutation of the other wild-type allele. Subsequently, it was demonstrated that both alleles also were mutated in the sporadic form of retinoblastoma. Thus, tumor suppressor genes are functionally recessive since their tumorigenic activity requires loss of both (maternal and paternal) alleles. An initial mutation event inactivates one copy of the gene and the second event inactivates the remaining allele via various mechanisms, which include mutations, deletions, chromosomal rearrangements and mitotic recombination.

The third broad group of cancer predisposing genes consists of mismatch repair genes, which are critical in maintaining the fidelity of DNA replication by repairing errors that occur during this process. Inactivation of mismatch repair genes leads to an increase in the spontaneous mutation rate of various genes, including oncogenes and tumor suppressors. This leads to uncontrolled cell proliferation.16 Like tumor suppressors, mismatch repair genes are functionally recessive (i.e., loss of both alleles is necessary for tumorigenesis) (Figure 1).

**Familial Cancer Syndromes Predisposing To Pancreatic Cancer**

Pancreatic adenocarcinoma forms an integral part of a number of inherited cancer syndromes that are caused by germline mutations in various cancer-predisposing genes (Tables 1 and 2).

**The Familial Atypical Multiple Mole Melanoma (FAMMM) Syndrome And The p16 Gene**

The FAMMM syndrome is characterized by the development of multiple nevi and malignant melanomas.17 Germline p16 mutations have been shown to co-segregate with melanoma in FAMMM kindreds, showing linkage to chromosome 9p21.18 These mutations impair p16 binding to its ligands CDK4 and/or CDK6.19 In addition to their role in FAMMM kindreds, germline
p16 mutations also predispose to disease in some patients with multiple primary melanomas.\(^{20}\)

Melanoma kindreds have long been noted to have an excess of pancreatic cancers, and this tumor is the second most commonly reported malignancy in melanoma families.\(^{21}\) The role of p16 germline mutations in pancreatic cancer was first demonstrated when Whelan \textit{et al.} reported a kindred in which p16 germline mutations cosegregated with melanoma, pancreatic cancer and squamous cell carcinoma of the oropharynx.\(^{22}\) In fact, melanoma kindreds with p16 germline mutations that impair protein function have a thirteen-fold increased risk of pancreatic adenocarcinoma.\(^{23}\) A shared susceptibility to the development of pancreatic adenocarcinoma and melanoma was further suggested by later studies, which demonstrated a nearly twofold increased risk of subsequent pancreatic adenocarcinoma in melanoma patients diagnosed prior to 50 years of age.\(^{24}\) These studies also demonstrated an increased mortality due to pancreatic adenocarcinoma observed in some melanoma kindreds.\(^{25}\) Despite this apparent association

### Table 1

<table>
<thead>
<tr>
<th>Syndrome</th>
<th>Associated Cancers/Lesions</th>
<th>Germline Mutation</th>
</tr>
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<tbody>
<tr>
<td>FAMMM</td>
<td>Melanoma</td>
<td>p16</td>
</tr>
<tr>
<td>Breast-ovarian cancer</td>
<td>Breast and ovarian cancer</td>
<td>BRCA1</td>
</tr>
<tr>
<td>Male breast cancer, breast cancer</td>
<td></td>
<td>BRCA2</td>
</tr>
<tr>
<td>Hereditary non-polyposis colorectal cancer*</td>
<td>Colorectal cancer, ovarian cancer, endometrial cancer, transitional cell cancer</td>
<td>hMSH2, hMLH1 and other mismatch repair genes</td>
</tr>
<tr>
<td>Peutz-Jeghers syndrome</td>
<td>GI hamartomas, mucocutaneous melanin deposits</td>
<td>STK11/LKB1</td>
</tr>
<tr>
<td>Li-Fraumeni syndrome</td>
<td>Sarcomas, breast cancer, brain tumors, leukemias, adrenocortical cancers</td>
<td>p53</td>
</tr>
<tr>
<td>FAP</td>
<td>Colorectal cancer, gastric and duodenal polyps, desmoid tumors, osteomas, congenital hypertrophy of retinal pigmented epithelium</td>
<td>APC</td>
</tr>
<tr>
<td>Hereditary pancreatitis</td>
<td>Severe chronic pancreatitis</td>
<td>Cationic trypsinogen gene</td>
</tr>
<tr>
<td>Williams syndrome</td>
<td>Infantile hypercalcemia</td>
<td></td>
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</tbody>
</table>

*Refer to text for Amsterdam Criteria

FAMMM = familial atypical multiple mole melanoma; GI = gastrointestinal; FAP = familial adenomatous polyposis
between p16 and pancreatic adenocarcinoma, there has been no report of a family in which a p16 germline mutation segregates with pancreatic cancer in the absence of a family history of melanoma.

The Hereditary Breast-Ovarian Cancer Syndrome, And The BRCA1 And BRCA2 Genes

Approximately 5% to 10% of breast cancers are thought to arise due to an inherited predisposition to the disease. At least 45% of families with site-specific breast cancer, and more than 80% of families with breast and ovarian cancer, were linked to BRCA1. In BRCA1 mutation carriers, the lifetime risk of breast and ovarian cancer was estimated to be approximately 90% and 65%, respectively. Germline mutations in BRCA2 were thought to account for about 70% of inherited breast cancers not linked to BRCA1, and also for the majority of familial male breast cancers. More recent studies, however, have shown that these mutation frequencies and risk assignments were likely overestimated, arising mainly from linkage studies of families with multiple cases of breast and ovarian cancer. Newer studies estimate that BRCA1 and BRCA2 together account for probably only 40% to 50% of hereditary breast cancers.

Most mutations reported in BRCA1 and BRCA2 families are either nonsense or frameshift mutations that lead to a prematurely terminated protein product, and are distributed throughout these large genes. The cloning of BRCA2 was aided by a novel technique called representational difference analysis. Using this method, Schutte et al identified a homozygous deletion that mapped to chromosome 13q in a pancreatic cancer. This provided the first evidence of BRCA2 inactivation in some pancreatic cancers and a probable association between BRCA2 and pancreatic cancer. Other studies showed that an excess of pancreatic cancer cases occur in families with BRCA1 and BRCA2 germline mutations.

Furthermore, individuals with pancreatic adenocarcinoma in these families were found to have inherited the at-risk haplotype and developed pancreatic cancer at a young age, indicating the pancreatic tumors were likely due to mutations in these predisposing genes. The breast cancer-pancreatic cancer association also was strengthened by the observation that a family history of pancreatic cancer was predictive of the presence of one of the above mutations in breast cancer families of Jewish descent (odds ratio = 3.74, p < 0.01). In particular, a family history of pancreatic cancer strongly predicted...
the presence of a BRCA2 mutation (odds ratio = 6.1, p = 0.01).35,36 A recent cohort study of BRCA2 families showed a significant increase in the risk of pancreatic cancer in these families (relative risk = 3.51, 95% confidence interval = 1.87-6.58).37

Evidence has emerged to suggest a role for germline BRCA2 mutations in patients with apparently sporadic pancreatic cancers. This was subsequently confirmed by the authors’ group, which demonstrated BRCA2 germline mutations in 4.9% of unselected pancreatic cancer cases. The same study also identified the recurrent BRCA2 6174delT mutation in four out of 39 (10%) unrelated Jewish individuals with pancreatic cancer. The lifetime risk of pancreatic cancer in 6174delT mutation carriers was estimated to be 7% (95% confidence interval 1.9% to 19%), as compared to a risk of 0.85% in the general population.38 The specificity of the high frequency of this mutation is further highlighted by the fact that the other common founder mutation, BRCA1 185delAG, was not identified in the same series of Jewish patients (unpublished results). There is little evidence to date, therefore, to suggest a role for germline BRCA1 mutations in sporadic pancreatic cancers.

The authors’ group has studied 199 breast cancer families with known BRCA2 mutations — 31 of whom had at least one member affected with pancreatic cancer. There was no difference in the frequencies of particular BRCA2 mutations in families with or without pancreatic cancer (unpublished results). Hence, there does not appear to be a genotype-phenotype correlation between the location of BRCA2 mutations and pancreatic cancer risk.

HNPCC is characterized by early onset of chiefly predominantly right-sided colon cancers, which often are mucinous and poorly differentiated.

HNPCC is an autosomal dominant disorder, accounting for 1% to 2% of all colorectal cancer cases. It is characterized by early onset of predominantly right-sided colon cancers, which often are mucinous and poorly differentiated. Synchronous and metachronous lesions are commonly observed in this syndrome.39 Due to difficulties inherent in classifying HNPCC, the International Collaborative Group on HNPCC established the Amsterdam Criteria to identify individuals with the disorder. The criteria are entirely clinical and include:

1. Three or more relatives with histologically verified colorectal cancer, one of whom is a first-degree relative of the other two;
Figure 2: Sample pedigrees of mutation carriers.

Probands are indicated with an arrow. Affected individuals are designated by filled circles or squares (diagnosis verified by review of pathology or medical records) or + symbols (unable to verify diagnosis). Age at diagnosis, if known, is indicated by numerals next to the cancer sites, which are designated as follows: B = breast, C = colon, K = kidney, Li = liver, M = melanoma, O = ovary and P = pancreas. In order to protect confidentiality, the birth order of siblings has been modified and most unaffected family members are not shown.
2. Colorectal cancer involving at least two generations; and
3. One or more cases of colorectal cases diagnosed before the age of 50 in the family.\(^{40}\)

The genetic basis of HNPCC has recently been elucidated in 1993. This syndrome is caused by inherited mutations in the mismatch repair genes hMSH2, hMLH1, hPMS1, hPMS2 and hMSH6/GTBP.\(^{41}\) Patients with HNPCC inherit one mutated copy of a mismatch repair gene and the second allele is inactivated somatically in the colorectal epithelium. This presumably leads to a loss of mismatch repair protein function, accumulation of further mutations and malignant transformation.

HNPCC kindreds have an increased frequency of various extra-colonic tumors, a feature that led Lynch \(et\ al\) to sub-classify this disorder into Lynch I and Lynch II syndromes.\(^{39}\) The latter was characterized not only by familial colorectal cancers but also by tumors of the endometrium, ovaries, stomach, small bowel, urinary tract and the hepatobiliary system, including the pancreas. Thus, HNPCC predisposes affected individuals to pancreatic cancer. Since HNPCC is caused by inherited mutations in mismatch repair genes, this suggests a possible association between mismatch repair genes and pancreatic cancer.

Microsatellites are 1-5 base pair tandemly repeated units of DNA present throughout the intronic regions of the human genome. Mismatch repair gene inactivation also leads to mutations in these microsatellites. This mutator phenotype is designated microsatellite instability and serves as a marker for mismatch repair gene deficiency.\(^{42}\) Although mismatch repair gene mutations have not been reported in the germline of pancreatic cancer patients, the association between the two entities is supported by the finding of microsatellite instability, a feature of HNPCC tumors, in a proportion of pancreatic adenocarcinomas.\(^{43}\) The incidence of mismatch repair gene mutations in a population of unselected pancreatic cancer patients is not known.

**Familial Site-Specific Pancreatic Cancer**
As indicated above, a number of families containing multiple individuals affected with pancreatic cancer have been reported over the years. Although these case reports suggest an inherited predisposition to pancreatic cancer, such familial aggregation of cases can result from chance association or common environmental exposures. To overcome this problem of small numbers, a number of familial pancreatic cancer registries have been established.\(^{44,45}\) Studies of these families show there is an autosomal dominant mode of inheritance for the disease. However, the age at diagnosis, histologic subtypes and survival of patients from these families is no different compared to unselected patients with pancreatic cancer.

Recently, Brentnall \(et\ al\) described a large family of 75 members spanning five generations, with nine family members who had died of pancreatic adenocarcinoma.\(^{46}\) Of interest, no other types of cancer were found in this kindred. This family was tested for mutations of p16, mismatch repair genes and the hereditary pancreatic-
The authors also recently completed a family history case-control study of 174 pancreatic cancer cases and 136 controls to further study cancer family history as an independent risk factor. Pancreatic cancer was the only cancer site statistically in excess in the case relatives, as compared to the control relatives (relative risk = 5.0, p = 0.01). Moreover, the lifetime risk of pancreatic cancer was high (7.2%) for relatives of those patients diagnosed before age 60, as was the risk for relatives of patients with multiple primary cancers (12.3%).

Other Familial Cancer Syndromes
Pancreatic cancer forms part of the tumor spectrum of other familial cancer syndromes, albeit less frequently. The syndromes, along with associated germline genetic defects include the Peutz-Jeghers syndrome (STK11), Li-Fraumeni syndrome (p53), familial adenomatous polyposis (FAP) (APC), Williams syndrome and hereditary pancreatitis (cationic trypsinogen gene).45

Role Of Various Cancer-Predisposing Genes
The authors’ group recently conducted a genetic analysis of 102 patients with pancreatic adenocarcinoma. The patients were unselected for age, sex, family history or ethnic origin. They completed a family history questionnaire and provided blood for mutation analysis. Pedigrees were constructed and classified as high-risk/familial, intermediate-risk/familial, intermediate-risk/nonfamilial or low-risk, according to defined criteria. Thirty-eight (37%) cases were characterized as high- or intermediate-risk, and 13% of these were found to harbor germline mutations — one in p16, one in BRCA1 and three in BRCA2 (Figure 2). Four of the mutation carriers had strong family histories of the syndromes associated with the mutated genes. No mutations were identified in patients in whom the sole risk factor was a family history of pancreatic cancer.47 Furthermore, in a separate retrospectively identified series, germline p16 mutations were found to predispose to disease in a proportion of population-based patients with both pancreatic adenocarcinoma and malignant melanoma.48

Summary
Despite the above findings, the lack of identifiable germline mutations in a large fraction of patients thought to be at increased risk, including those with histories indicative of familial pancreatic cancer, suggests there are likely as yet unidentified gene loci predisposing to pancreatic cancer. Further investigation is needed in this area. The ability to identify a population of individuals at increased risk, based on genetic tests, offers a unique opportunity to intervene with a variety of primary and secondary prevention strategies.

There are currently no cost-effective and clinically proven clinical screening protocols for pancreatic adenocarcinoma. However, since it is possible to identify individuals at increased risk for pancreatic adenocarcinoma by genetic
screening for germline mutations known to predispose to the disease, this group of individuals offers the opportunity to develop and validate various screening procedures. Although the risk of pancreatic cancer and ultimate prognosis in asymptomatic mutation carriers is yet to be defined, genetic screening can be justified on several grounds. First, as discussed earlier, pancreatic cancer has a significant disease burden. Second, early diagnosis will probably lead to an improved outcome. Third, genetic screening leads to the identification of at-risk carriers, and a possible reduction in anxiety for relatives who are found to be noncarriers. Fourth, germline mutation carriers can undergo clinical screening for melanoma, breast cancer and colon cancer (diseases that occur in association with pancreatic cancer) according to established guidelines.

Of course, it must be emphasized that given the current stage of knowledge, such genetic and clinical screening for susceptibility to pancreatic adenocarcinoma should only be conducted in research settings with appropriate pre- and post-test counseling.

References

Suggested Reading