SURVEILLANCE OF INDIVIDUALS AT HIGH RISK FOR DEVELOPING PANCREATIC CANCER

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op het krijgen van pancreascarcinoom

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CHAPTER 1
GENERAL INTRODUCTION & AIMS OF THE THESIS

The clinical burden of pancreatic cancer

We still face great difficulties to treat and cure patients with pancreatic ductal adenocarcinoma (henceforth referred to as pancreatic cancer). The survival is dismal even in those who undergo intended curative surgery in case of a localized tumor. Despite the relatively low incidence of 9-12 per 100.000 per year in Western populations (approximate lifetime-risk 1.0%), pancreatic cancer is ranked among the top five causes of cancer-related death in Western populations (1, 2). Unfortunately, as clearly demonstrated by Figure 1, major efforts in the fields of surgery and (neo)adjuvant treatment have not yielded a significant improvement in prognosis. With a mean survival of less than 6 months and an overall 5-year survival of less than 6% (1, 2), patients with pancreatic cancer still face one of the worst prognosis of all human cancers.

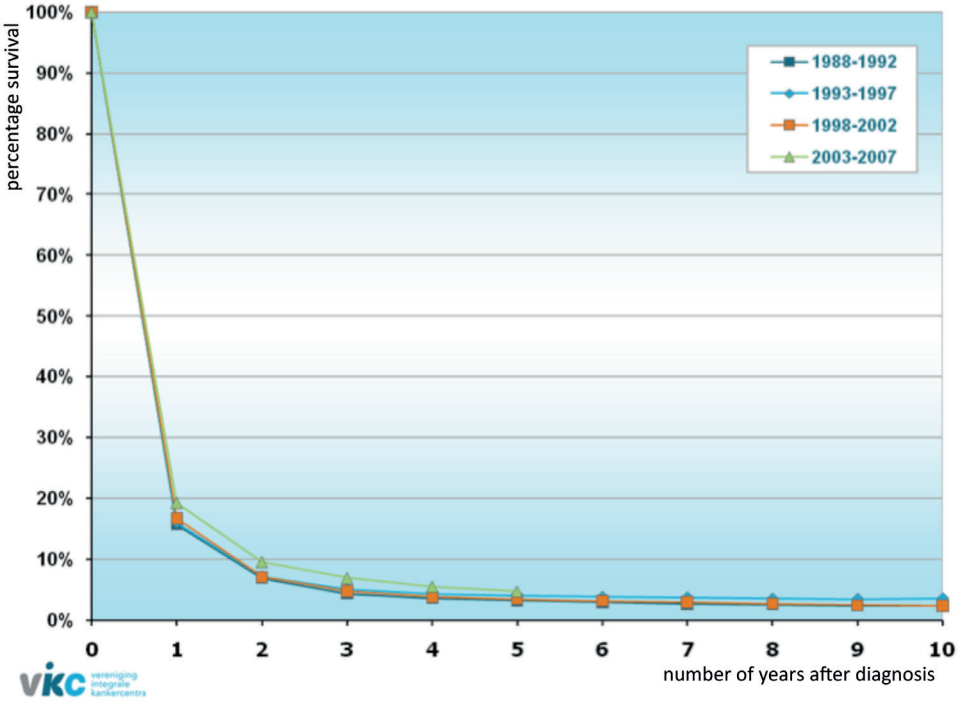


Figure 1. Relative survival of pancreatic cancer per period of diagnosis (Source IKCnet)

One of the main reasons for this problem and urge for early detection

One of the main reasons of the high mortality of pancreatic cancer and the current inability to cure this disease is the late occurrence of pancreatic cancer-related symptoms. Consequently, less than 20% of all patients present with localized disease and is therefore eligible for surgical resection, which is currently the only treatment with a curative potential. Unfortunately, even such intended curative resection proves only effective for the minority of patients as the overall 5-year survival after surgical resection is less than 10% (1-3). The best prognosis is obtained in patients with early stage disease; the 5-year

survival of patients with stage I pancreatic cancer (T1-2N0M0) is 28% (4). However, due to the late occurrence of symptoms, only 7% of all operated patients are diagnosed and treated with stage I disease (2). Given these facts, screening for pancreatic cancer and its precursor lesions is an important and promising tool to improve the prognosis and outcome of this disease since screening aims to detect a disease prior to the point of clinical presentation. Consequently, screening aims to decrease mortality and morbidity of the disease (5). Mainly due to the current lack of a non-invasive and affordable screening test, screening of the general population is neither useful nor feasible. However, it may be worthwhile when offered to individuals at high risk for developing pancreatic cancer.

Individuals at high risk for pancreatic cancer

It is currently estimated that about 10% of all pancreatic cancer cases occur in the background of familial clustering, representing a population of individuals at high risk for developing pancreatic cancer. On the basis of clinical and genetic criteria, these high-risk individuals can be divided into two groups. In the first group, pancreatic cancer develops within the framework of a known hereditary cancer syndrome or hereditary disease. The second group, referred to as familial pancreatic cancer (FPC), consists of families with clustering of pancreatic cancer and not meeting diagnostic criteria of specific hereditary cancer syndromes.

Known hereditary cancer syndromes and hereditary diseases with an increased pancreatic cancer risk include (1) Peutz-Jeghers syndrome (germline mutation *LKB1*) (6)), (2) familial cutenous malignant melanoma (germline mutation *CDKN2A*) (7, 8), (3) Hereditary Breast and Ovarian Cancer syndrome (germline mutation *BRCA1* and *BRCA2*) (9-11), (4) Lynch syndrome (germline mutation *MLH1*, *MSH2*, *MSH6* and *PMS2*) (12), (5) hereditary pancreatitis (germline mutation *PRSS1*) (13), and (6) Li-Fraumeni syndrome (germline mutation *p53*) (14). In the majority of families (80%) with a strong family history of pancreatic cancer, the disease is apparently unrelated to any currently recognized hereditary syndrome/disease and these families are therefore referred to as FPC family. A strong family history is defined as pancreatic cancer in either ≥2 first-degree relatives (FDR), ≥3 relatives or 2 relatives of whom one being <50 years at time of diagnosis (15). Prospective studies of families with a family history of pancreatic cancer demonstrate an increased risk of developing pancreatic cancer in FDR that increases depending on the number of affected relatives (16). The 'classical' phenotype of FPC-families (with pancreatic cancer in subsequent generations and affecting both male and female family members) suggests an autosomal dominant inheritance of the disease with variable penetrance. However, at present, the major gene(s) involved in the development of pancreatic cancer in FPC kindreds is/are unknown. Consequently, it is impossible to identify family members at true risk within FPC-families. This has important implications with all family members offered screening while only 50% of family members carry the (unknown) autosomal dominant gene mutation.

In the past decades, our knowledge about the level of pancreatic cancer risk for the different high-risk populations has increased substantially, but for the majority of syndromes the associated pancreatic cancer risk has not yet been firmly established. Table 1 lists the estimated level of pancreatic cancer-risk for the different high-risk populations based on the currently available data.

Syndromic pancreatic cancer	**Relative risk**
Peutz-Jeghers syndrome (6, 26)	76-132
Familial cuteneous malignant melanoma (7, 8, 27)	14.8-52
Hereditairy pancreatitis (13, 28)	57-87
Hereditairy breast and ovarium cancer (*BRCA2*) (9, 10)	3.51-5.9
Hereditairy breast and ovarium cancer (BRCA1) (11)	2.26
Li Fraumeni (14)	7.5
Lynch syndrome (12)	8.0
Familial Pancreatic Cancer	
≥3 first degree relatives with pancreatic cancer (16)	32.0
2 first degree relatives (16)	6.4

Table 1. Relative pancreatic cancer risk within different hereditary cancer syndromes/diseases and familial pancreatic cancer kindreds.

Early stage pancreatic cancer and premalignant lesions of pancreatic cancer

The evidence is strong that long-term survival can be achieved following surgical resection of small non-metastatic pancreatic cancer (17-19). This is particularly true if negative margins (R0 resection) can be achieved (20, 21). Resection of high-grade premalignant lesions of pancreatic cancer leads to an even better survival since at this stage of disease there is yet no hazard of local recurrence and/or distant metastases.

Well-defined premalignant lesions of pancreatic cancer are Pancreatic Intraepithelial Neoplasia (PanIN) and Intraductal Papillary Mucinous Neoplasm (IPMN), both are intraductal lesions. The rate at which PanINs and IPMNs progress to invasive carcinoma is currently insufficiently understood. Based on results of studies on sporadic pancreatic cancer, it is estimated that it takes at least 15 years for metastatic pancreatic cancer to develop (22). Patients with non-invasive IPMNs are, on average, 3-5 years younger than patients with an IPMN with an associated invasive carcinoma, suggesting it takes 3-5 years for a clinically detectable non-invasive lesion to progress to an invasive one (23). In addition, patients with a known small branch type IPMN have been carefully followed and over 5-years only 2.4% to 6.9% of these lesions progress to invasive ductal adenocarcinoma (24, 25). It should be noted that these estimates are derived from sporadic pancreatic cancer. No data is available to show whether the same chain of events and speed of progression applies to individuals with a strong family history of pancreatic cancer.

Effective screening program

It is internationally accepted that effective (and ethical) screening programs should comply with several criteria (5). Screening of individuals at high risk of developing pancreatic cancer will ultimately be effective if the benefits of screening, defined as a reduction in mortality due to pancreatic cancer and life years gained compared to individuals who do not undergo screening, outweigh the potentially negative side-effects of screening including overtreatment, false positive and negative case findings and costs. Despite encouraging preliminary data and sound theoretical reasoning, we currently lack data driven evidence to show that the benefits of surveillance outweigh its negative side effects, even in high-risk individuals. In order to prevent inefficient, ineffective and even potentially harmful screening practices, it is therefore key to judiciously evaluate such pancreatic cancer surveillance programs in high-risk individuals.

AIMS OF THIS THESIS

The aims of this thesis are to:

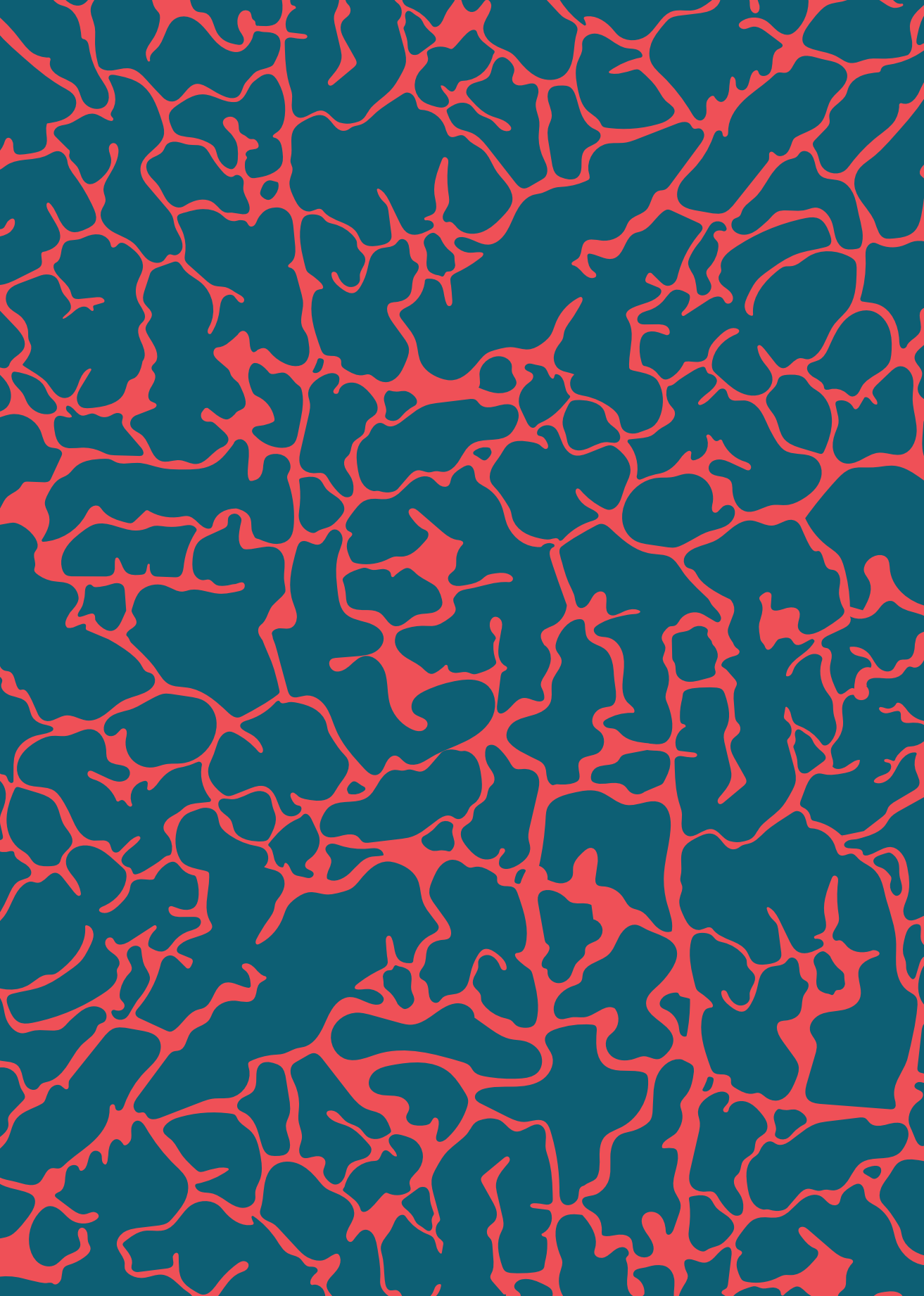
1. Investigate the feasibility, outcome and effectiveness of a pancreatic cancer surveillance program
2. Expand our knowledge on pathophysiology and risk profiles of the various high-risk groups
3. Examine the psychological impact of participating in a pancreatic cancer surveillance program

REFERENCES

1. Jemal A, Bray F, Center MM, Ferlay J, Ward E, Forman D. Global cancer statistics. CA Cancer J Clin. 2011;61(2):69-90. Epub 2011/02/08.
2. Canto MI, Harinck F, Hruban RH, Offerhaus GJ, Poley JW, Kamel I, et al. International Cancer of the Pancreas Screening (CAPS) Consortium summit on the management of patients with increased risk for familial pancreatic cancer. Gut. 2013;62(3):339-47. Epub 2012/11/09.
3. Shaib Y, Davila J, Naumann C, El-Serag H. The impact of curative intent surgery on the survival of pancreatic cancer patients: a U.S. Population-based study. The American journal of gastroenterology. 2007;102(7):1377-82. Epub 2007/04/04.
4. Bilimoria KY, Bentrem DJ, Ko CY, Ritchey J, Stewart AK, Winchester DP, et al. Validation of the 6th edition AJCC Pancreatic Cancer Staging System: report from the National Cancer Database. Cancer. 2007;110(4):738-44. Epub 2007/06/21.
5. Wilson JM, Jungner G. Principles and Practice of Screening for Disease. Geneva:World Health Organisation. 1968.
6. Giardiello FM, Brensinger JD, Tersmette AC, Goodman SN, Petersen GM, Booker SV, et al. Very high risk of cancer in familial Peutz-Jeghers syndrome. Gastroenterology. 2000;119(6):1447-53. Epub 2000/12/13.
7. de Snoo FA, Bishop DT, Bergman W, van Leeuwen I, van der Drift C, van Nieuwpoort FA, et al. Increased risk of cancer other than melanoma in CDKN2A founder mutation (p16-Leiden)-positive melanoma families. Clin Cancer Res. 2008;14(21):7151-7.
8. Goldstein AM, Struewing JP, Fraser MC, Smith MW, Tucker MA. Prospective risk of cancer in CDKN2A germline mutation carriers. J Med Genet. 2004;41(6):421-4.
9. Cancer risks in BRCA2 mutation carriers. The Breast Cancer Linkage Consortium. J Natl Cancer Inst. 1999;91(15):1310-6. Epub 1999/08/05.
10. van Asperen CJ, Brohet RM, Meijers-Heijboer EJ, Hoogerbrugge N, Verhoef S, Vasen HF, et al. Cancer risks in BRCA2 families: estimates for sites other than breast and ovary. J Med Genet. 2005;42(9):711-9. Epub 2005/09/06.
11. Thompson D, Easton DF. Cancer Incidence in BRCA1 mutation carriers. J Natl Cancer Inst. 2002;94(18):1358-65. Epub 2002/09/19.
12. Kastrinos F, Mukherjee B, Tayob N, Wang F, Sparr J, Raymond VM, et al. Risk of pancreatic cancer in families with Lynch syndrome. JAMA. 2009;302(16):1790-5. Epub 2009/10/29.
13. Howes N, Lerch MM, Greenhalf W, Stocken DD, Ellis I, Simon P, et al. Clinical and genetic characteristics of hereditary pancreatitis in Europe. Clin Gastroenterol Hepatol. 2004;2(3):252-61. Epub 2004/03/16.
14. Ruijs MW, Verhoef S, Rookus MA, Pruntel R, van der Hout AH, Hogervorst FB, et al. TP53 germline mutation testing in 180 families suspected of Li-Fraumeni syndrome: mutation detection rate and relative frequency of cancers in different familial phenotypes. J Med Genet. 2010;47(6):421-8. Epub 2010/06/05.

15. Brand RE, Lerch MM, Rubinstein WS, Neoptolemos JP, Whitcomb DC, Hruban RH, et al. Advances in counselling and surveillance of patients at risk for pancreatic cancer. Gut. 2007;56(10):1460-9.
16. Klein AP, Brune KA, Petersen GM, Goggins M, Tersmette AC, Offerhaus GJ, et al. Prospective risk of pancreatic cancer in familial pancreatic cancer kindreds. Cancer Res. 2004;64(7):2634-8. Epub 2004/04/03.
17. Ferrone CR, Finkelstein DM, Thayer SP, Muzikansky A, Fernandez-delCastillo C, Warshaw AL. Perioperative CA19-9 levels can predict stage and survival in patients with resectable pancreatic adenocarcinoma. J Clin Oncol. 2006;24(18):2897-902. Epub 2006/06/20.
18. Schnelldorfer T, Ware AL, Sarr MG, Smyrk TC, Zhang L, Qin R, et al. Long-term survival after pancreatoduodenectomy for pancreatic adenocarcinoma: is cure possible? Ann Surg. 2008;247(3):456-62. Epub 2008/04/01.
19. Winter JM, Cameron JL, Campbell KA, Arnold MA, Chang DC, Coleman J, et al. 1423 pancreaticoduodenectomies for pancreatic cancer: A single-institution experience. J Gastrointest Surg. 2006;10(9):1199-210; discussion 210-1. Epub 2006/11/23.
20. Fatima J, Schnelldorfer T, Barton J, Wood CM, Wiste HJ, Smyrk TC, et al. Pancreatoduodenectomy for ductal adenocarcinoma: implications of positive margin on survival. Arch Surg. 2010;145(2):167-72. Epub 2010/02/17.
21. Matthaei H, Hong SM, Mayo SC, dal Molin M, Olino K, Venkat R, et al. Presence of pancreatic intraepithelial neoplasia in the pancreatic transection margin does not influence outcome in patients with R0 resected pancreatic cancer. Ann Surg Oncol. 2011;18(12):3493-9. Epub 2011/05/04.
22. Yachida S, Jones S, Bozic I, Antal T, Leary R, Fu B, et al. Distant metastasis occurs late during the genetic evolution of pancreatic cancer. Nature. 2010;467(7319):1114-7. Epub 2010/10/29.
23. Sohn TA, Yeo CJ, Cameron JL, Hruban RH, Fukushima N, Campbell KA, et al. Intraductal papillary mucinous neoplasms of the pancreas: an updated experience. Ann Surg. 2004;239(6):788-97; discussion 97-9. Epub 2004/05/29.
24. Sawai Y, Yamao K, Bhatia V, Chiba T, Mizuno N, Sawaki A, et al. Development of pancreatic cancers during long-term follow-up of side-branch intraductal papillary mucinous neoplasms. Endoscopy. 2010;42(12):1077-84. Epub 2010/12/02.
25. Uehara H, Nakaizumi A, Ishikawa O, Iishi H, Tatsumi K, Takakura R, et al. Development of ductal carcinoma of the pancreas during follow-up of branch duct intraductal papillary mucinous neoplasm of the pancreas. Gut. 2008;57(11):1561-5. Epub 2008/05/15.
26. Korsse SE, Harinck F, van Lier MG, Biermann K, Offerhaus GJ, Krak N, et al. Pancreatic cancer risk in Peutz-Jeghers syndrome patients: a large cohort study and implications for surveillance. Journal of medical genetics. 2013;50(1):59-64. Epub 2012/12/15.

27. Goldstein AM, Chan M, Harland M, Gillanders EM, Hayward NK, Avril MF, et al. High-risk melanoma susceptibility genes and pancreatic cancer, neural system tumors, and uveal melanoma across GenoMEL. Cancer Res. 2006;66(20):9818-28.
28. Rebours V, Boutron-Ruault MC, Schnee M, Ferec C, Maire F, Hammel P, et al. Risk of pancreatic adenocarcinoma in patients with hereditary pancreatitis: a national exhaustive series. The American journal of gastroenterology. 2008;103(1):111-9. Epub 2008/01/11.



PART I

SURVEILLANCE OF INDIVIDUALS AT HIGH RISK FOR DEVELOPING PANCREATIC CANCER

F. Harinck, J-W. Poley, I. Kluijt, P. Fockens, M.J. Bruno, on behalf of the Dutch research group of pancreatic cancer surveillance in high-risk individuals

Digestive Diseases 2010; 28: 670-678

CHAPTER 2
IS EARLY DIAGNOSIS OF PANCREATIC CANCER FICTION? SURVEILLANCE OF INDIVIDUALS AT HIGH RISK FOR PANCREATIC CANCER

ABSTRACT

Pancreatic cancer represents one of the most deadly human malignancies with an overall 5-years survival less than 5 percent. Despite improvements in imaging techniques and surgical techniques, survival statistics have hardly improved over the past decades. To improve the dismal outlook it would be highly desirable to develop a program to detect precursor lesions or small asymptomatic early cancers at the time when the disease is still at a curable stage. Screening the general population for disease presence is not feasible at present because of the relatively low disease incidence and the lack of a non-invasive, reliable and cheap screening tool. Targeted surveillance programs however, in individuals at high risk for developing pancreatic cancer, like mutation carriers of pancreatic cancer prone hereditary (tumor) syndromes or individuals with a strong family history of pancreatic cancer without a known underlying genetic defect, might be feasible. Careful consideration of the criteria put forward by Wilson and Jungner as published by the World Health Organization on the principles and practice of screening for disease, indicate that surveillance in this high risk population by means of endosonography (EUS) and/or magnetic resonance imaging (MRI) represents a promising development, though experimental. It nicely points out which open questions need to be addressed. Among others, these include how to acquire a better understanding of the natural behavior and progression of precursor lesions towards invasive cancer, how to firmly establish the performance characteristics of EUS and MRI for the detection of (early) lesions in individuals at high risk for pancreatic cancer, and how to determine which lesions can be safely observed with continued surveillance and which lesions justify resection.

INTRODUCTION

Is early detection of pancreatic cancer fiction? There is no definite answer to this burning question at this time, but based on theoretical reasoning and preliminary (pre)clinical data early detection of pancreatic cancer offers a promising outlook to fight the high death toll of this devastating disease. Pancreatic cancer still remains one of the most deadly cancers in which incidence nearly equals the mortality rate. One explanation for the high mortality is that the majority of patients develop symptoms late in the course of the disease, at the time when they already have locoregional spread and/or distant metastases. But even in the vast majority of patients with localized disease, surgical treatment with curative intention eventually proves not to be effective. The poor survival statistics, with an overall median survival of less than 6 months and an overall 5-year survival rate of less than 5%, have hardly changed over the past decades despite advancements in the fields of radiology, surgery, oncology and radiotherapy (1, 2).

When contemplating that treatment of pancreatic cancer by surgical resection and (neo) adjuvant therapy has not brought about a significant change in survival statistics, other strategies to lower cancer mortality like primary and secondary prevention come into focus. Primary prevention by modifying established risk factors like cessation of smoking has a considerable potential to reduce the number of pancreatic cancer deaths, but it is well known that such individual and societal behavioral change is very difficult to accomplish (3). Secondary prevention by screening the general population for disease presence does not seem feasible because of the relatively low incidence and the lack of a noninvasive, reliable and cheap screening tool. However, screening might be feasible in a selected group of individuals at high risk for developing pancreatic cancer. Detection of nonsymptomatic cancer or its precursor lesions and subsequent early treatment will hopefully have a favorable effect on disease outcome and improve survival rates. In this paper, we will apply the principles of screening and practice for disease as proposed by Wilson and Jungner (4) to appraise the validity of surveillance using endosonography (EUS) and/or magnetic resonance imaging (MRI) in individuals at high risk for developing pancreatic cancer (Box 1).

1. The condition sought should be an important health problem
2. There should be an accepted treatment for patients with recognized disease
3. Facilities for diagnosis and treatment should be available
4. There should be a recognized latent or early symptomatic stage
5. There should be a suitable test or examination
6. The test should be acceptable to the population
7. The natural history of the condition, including development from latent to declared disease, should be adequately understood
8. There should be an agreed policy on whom to treat as patients
9. The cost of case-finding (including diagnosis and treatment of patients diagnosed) should be economically balanced in relation to possible expenditure on medical care as a whole
10. Case-finding should be a continuing process and not a “once and for all” project

Box 1. Principles of screening by Wilson and Jungner

Principle 1: The condition sought should be an important health problem

'To be considered an important health problem, a disease need not necessary have a high degree of prevalence… but also conditions with serious consequences to the individual and his or her family may warrant relatively uneconomic screening measures' (4).

It is not the incidence of pancreatic cancer that puts this disease in the spotlight as an important health problem, as pancreatic cancer is a fairly rare disease with a yearly incidence of 8.5 per 100,000 in Europe (2, 5). What does make pancreatic cancer a serious health problem is its high death toll, which approaches almost 100%, ranking it the 5th leading cause of all cancer deaths in Europe (5, 6). Despite the evolution of surgical techniques and the utilization of (neo)adjuvant chemo(radiation) therapies, the prospects for surviving this dismal disease have hardly increased over the past decades.

Interestingly, it is estimated that about 10% of all pancreatic cancer cases are caused by inherited (genetic) factors (7). On the basis of clinical and genetic criteria, these high-risk individuals can be divided into 2 groups. The first group consists of mutation carriers of pancreatic cancer prone hereditary (tumor) syndromes (syndromic pancreatic cancer). The 2nd, and largest, group consists of individuals with a strong family history of pancreatic cancer, but without a known underlying genetic defect (familial pancreatic cancer (FPC)). A family with at least 1 pair of 1st-degree relatives with pancreatic cancer or families with at least 3 affected relatives is referred to as FPC-kindred (8). In these selected cases the lifetime risk for developing pancreatic cancer is strongly increased and depending on the gene involved this risk can exceed up to 17% in carriers of a p16/CDKN2A mutation (9), 36% in Peutz-Jeghers syndrome patients (10) and 55% in patients with hereditary pancreatitis (11). The lifetime risk of developing pancreatic cancer for members of FPC-kindreds increases to 40% when this member has 3 affected 1st-degree relatives (12). An overview of high-risk conditionsfor pancreatic cancer is listed in Table 1.

Although inherited pancreatic cancer encompasses a relatively smaller part of the total incidence of pancreatic cancer, the social, psychological and clinical implications for family members are immense and beyond comprehension for non-affected individuals. This

	Lifetime risk	Potential candidates for surveillance
Syndromic pancreatic cancer		
Familial Atypical Multiple Mole Melanoma syndrome (FAMMM) (9)	17%	Yes
Peutz Jegers syndrome (10, 13)	11-36%	Yes
Hereditairy pancreatitis (11)	55%	Yes
Hereditairy breast and ovarium cancer (*BRCA2*) (14)	5%	if ≥ 2* affected
Hereditairy breast and ovarium cancer (*BRCA1*) (15)	?	if ≥ 2* affected
Li Fraumeni	?	if ≥ 2* affected
Lynch syndrome (16)	?	if ≥ 2* affected
Familial Pancreatic Cancer		
≥3 first degree relatives with pancreatic cancer (12)	40%	Yes
2 first degree relatives (12)	8-12%	Yes

Table 1. Hereditary conditions with increased risk for pancreatic cancer

**confirmed pancreatic cancer in at least two proven mutation carriers*

together with minimal odds for cure once pancreatic cancer has become symptomatic demonstrates the potential implication, impact and relevance of surveillance for precursor lesions or early pancreatic cancer in individuals from high-risk families.

Principle 2: There should be an accepted treatment for patients with a recognized disease

'Of all the criteria a screening test should fulfill, the ability to treat the condition adequately, when discovered, is perhaps the most important … A better prognosis should be given by treating the conditions found at an earlier stage than was previously the practice' (4).

Surgery offers the only chance for cure for patients with pancreatic cancer. In the old days, pancreatic head resections (classic Whipple's resection or pylorus-preserving pancreatoduodenectomy) were associated with a high morbidity and mortality rate, the latter reaching up to 20% even at major academic institutions. Nowadays, owing to refinements in surgical techniques and improved preoperative and postoperative care, mortality rates are well below 4% in high-volume centers (17).

The prognosis of pancreatic cancer is strongly dependent on stage. Nearly all long-term survivors had early stage disease at the time of resection (18, 19). After curative surgery, the 5-year survival for patients with a tumor smaller than 2 cm, negative margins and no nodal involvement (T1N0M0 AJCC stage IA) is 31.4%. This survival drops dramatically in more advanced stages with a 5-year survival rate of only 7.7% in patients with a tumor that extends beyond the pancreas but without involvement of the celiac axis or the superior mesenteric artery (AJCC stage IIB) (20).

Pancreatic cancer in asymptomatic individuals is likely to be smaller and, hence, should offer a better prognosis. In a surveillance setting, it might even be possible to detect and resect noninvasive precursor lesions, avoiding the risk of metastases and postsurgical tumor recurrence and thereby improving the prognosis. To date, screening programs have detected 4 asymptomatic cancers. In one of these, there was a favorable outcome; 5 years after surgery and adjuvant chemoradiation therapy for a 28-mm T2N1M0 this patient is still alive and disease-free (21). The other 3 cases eventually died either because resection was proved not radical (n = 2) or because of tumor recurrence despite a R0 resection (22). One could argue that for these patients the start of this screening program came too late. If screening would have been commenced a year earlier, smaller cancers or even precursor lesions might have been detected with a better disease outcome. However, this is highly speculative and remains to be proven prospectively.

Principle 3: Facilities for diagnosis and treatment should be available

The availability of advanced imaging modalities, including multidetector computed tomography (CT), MRI and EUS has increased exponentially over the past decades and these techniques are currently widely used. Nevertheless, despite their widespread availability, we strongly believe that screening and surveillance efforts should be centered in specialized facilities with a dedicated and experienced multidisciplinary pancreatic

team. There are multiple reasons to support this. For one, the reliability of EUS is strongly dependent on the experience and skills of the operator. This may even be more true when screening relatively normal pancreases; one should not overinterpret variations of normal as being abnormal, but also not negate features that matter. This is hampered by the fact that, as yet, there are no firmly established guidelines that aid in deciding when to continue surveillance or when to intervene with surgical resection in case of an abnormality. Therefore, it is pivotal that patients are managed within a well-defined research protocol clearly defining which step is to be taken at what time. Outcomes should be carefully documented and discussed at multidisciplinary conferences. Another important consideration for centralization of surveillance efforts is that complication rates of pancreatic surgery have proven to be the lowest when performed in high-volume centers by experienced pancreatic surgeons (17).

Principle 4: There should be a recognized latent or early symptomatic stage

Recognized and well-defined precursor lesions of invasive pancreatic cancer are pancreatic intraductal neoplasia (PanIN), intraductal papillary mucinous neoplasms (IPMN) and mucinous cystic neoplasms (23). PanIN and IPMN have been reported to occur in individuals individuals at high risk of developing pancreatic cancer (21, 22, 24–28). Both are intraductal lesions predominately composed of columnar, mucin-producing cells that may grow in a flat configuration or may produce papillae.

PanINs are microscopic lesions that typically arise in the smaller ducts (<5 mm) whereas IPMNs usually involve the larger ducts (23). Based on the degree of architectural and nuclear atypia, PanINs can be further classified into 3 different grades; PanIN-1, PanIN-2 and PanIN-3. PanIN-1 lesions are flat (PanIN-1a) or papillary (PanIN-1b), whereas PanIN-2 lesions are architecturally more complex and exhibit more nuclear changes. PanIN-3 lesions show the highest degree of dysplasia (29).

IPMNs arise within the main pancreatic duct (mainduct IPMN) or one of its main branches (branch-duct IPMN) causing a varying degree of duct dilatation that is identifiable on imaging examinations. Based on architectural and cytological changes, IPMNs can be further classified into IPMN with low-grade dysplasia (IPMNadenoma), IPMN with moderate dysplasia (IPMN-borderline) and IPMN with high-grade dysplasia (in situ carcinoma) (30). Radiological and clinical features that are predictive of malignant degeneration include mural nodules or a solid mass component, a markedly dilated main pancreatic duct, clinical symptoms (e.g. abdominal pain, newly onset diabetes, weight loss) and a cyst size >30 mm in case of a branch-duct IPMN (31).

Principle 5: There should be a suitable test or examination

Although the optimal approach for screening and surveillance is still unknown, EUS and MRI currently are regarded as the most promising tests for the detection of early cancer and its precursors. CT does not seem to be a suitable surveillance technique

since preliminary results indicated that CT failed to visualize clinical significant lesions (32). Furthermore, the repeated radiation exposure with each follow-up CT examination does not make CT a suitable surveillance modality. At present EUS holds the best cards for detection of small mass lesions (early cancer) as it is considered the most sensitive imaging modality to detect (asymptomatic) cancerous lesions, in particular for lesions less than 2 cm which are often missed by MR or CT (33, 34).

Detection of PanIN lesions is not straightforward since we currently lack a technique that can reliably detect this type of lesions in vivo, at least when they appear isolated. However, limited data support that EUS might be able to detect the secondary parenchymal changes caused by PanIN lesions. It has been suggested that multifocal PanIN lesions (even low-grade) are associated with lobular atrophy of the surrounding parenchyma. These histological changes have been shown to correlate with the chronic pancreatitis-like changes detected by EUS (24). It is still too early to determine whether this association is invariably present and can provide a reliable morphological approach to screen for PanIN lesions in vivo. For this, further research is needed.

Both EUS and MRI/MRCP have proven to be valuable techniques for the detection of IPMN-like lesions and risk stratification by assessing potentially malignant features including a size greater than 3 cm or the presence of intracystic nodules (35, 36).

More work needs to be done in order to distinguish high-grade lesions and early cancers from low-grade lesions and non-neoplastic lesions to reduce the chance of overtreatment. For example, judgment calls based on EUS reports has led to the resection of benign (serous cystadenomas) (21, 26) or low-grade dysplastic lesions (21, 25, 26).

Principle 6: The test should be acceptable for the population

To date there is hardly any report addressing the acceptability and experiences of surveillance techniques among high-risk individuals for pancreatic cancer. One could argue that the invasiveness of EUS might result in a lower acceptability than, for instance, the acceptability of MRI. However, in most centers, EUS is performed under conscious sedation or propofol sedation, which largely prevents patients from experiencing excessive burden, also because some sedatives cause retrograde amnesia (37). Based on our own (preliminary) experience, participants of our surveillance study do not report a significant difference in the acceptability and experiences between EUS and MRI (38). The incidence of cases in which claustrophobia prevented the MRI scan from being performed or to be terminated prematurely is low, only 2.0 and 1.2%, respectively. For these selected cases, sedation and scanning in a prone position might help to overcome this problem (39).

Principle 7: The natural history of the condition, including development from latent to declared disease, should be adequately understood

While our understanding of precursor lesions of pancreatic cancer has significantly improved over the past decades, the natural history of both PanINs (29) and IPMNs (23) is still

incompletely elucidated. Several important questions with respect to their natural history remain unanswered. A widely accepted hypothesis is that the development and progression of the majority of invasive pancreatic cancers is analogue to the adenomacarcinoma sequence seen in, for instance, colorectal cancer (29, 40). Accumulation of genetic and chromosomal abnormalities (which have largely been identified) may cause normal tissue to derange into a PanIN-1 lesion which may then progress into a PanIN-2 or PanIN-3 lesion and finally into invasive cancer. It is currently unclear what the probability is that a single PanIN lesion will progress to invasive cancer and how fast this evolution occurs. We also do not know whether PanIN lesions that arise in high-risk individuals have the same biological behavior as PanIN lesions in sporadic cases (29).

As for IPMN lesions, genetic analysis also revealed genetic and chromosomal abnormalities accumulating from low-grade to histological high-grade IPMN (23). Similar to PanINs, it is currently not established at what speed and frequency this progression takes place. Based on series in which sporadic IPMNs were studied, it is well known that both main-duct as well as branch-duct IPMNs carry a risk of harboring malignancy. This risk is particularly high for IPMNs arising from the main branch in which the frequency of malignancy (in situ and invasive) in main-duct IPMNs equals 70% (range 60–92%). In branch-duct IPMNs the frequency of malignancy is 25% (range 6–46%) and the frequency of invasive cancer is 15% (range 0–31%) with the lower range prevalence in asymptomatic patients (31). When detected in its early stage (without invasive cancer and positive margins) the risk of recurrence following resection with curative intent is less than 10%. However, the risk of recurrence is high (60–70%) when invasive cancer is present (41). As for PanIN, it is currently unknown if IPMNs arising in high-risk individuals behave biologically differently than their sporadic counterparts. In their screening series in individuals at high-risk for pancreatic cancer, Canto *et al.* (25) reported on 3 cystic lesions with a more rapid progression in size compared to what is usually observed when following sporadic branch-duct IPMNs. After resection, all proved to be IPMN-adenoma. In another case from this same series, it is reported that a small 6-mm possible branch-type IPMN progressed into a 2.5-cm adenocarcinoma within 3 months time (25). The question remains whether this is truly part of the spectrum of biologic development of IPMN, which would be quite worrisome as such millimeter-sized IPMN lesions are encountered quite frequently, or that it represents a false negative observation from the preceding investigation. In our pilot series, the prevalence of IPMN-'like' lesions was 16% with none of the lesions larger then 15 mm (22). During limited follow-up of maximally 3 years, no significant changes occurred in these lesions (unpubl. data).

Principle 8: There should be an agreed upon policy whom to treat as a patient

Like in any process of clinical decision making, the benefits of treatment should outweigh its potential risks. The ultimate consequence of finding suspicious lesions while examining an individual at high risk for developing pancreatic cancer is to resect it. Although risks are

considered acceptable when pancreatic surgery is performed in a high-volume center, it is certainly not without complications and this cannot be neglected in the decision-making process when choosing between surgery or continued surveillance. The key issue is to rightfully recognize and identify (precursor) lesions that have a high risk to progress into an invasive malignancy. Individuals with truly benign lesions should not be exposed to unwarranted surgery, while patients with truly suspicious (pre)malignant lesions should not be withheld a rightful resection, so as not to miss the opportunity to cure cancer. Probably the least difficult cases to deal with are those with main-branch or branch-duct IPMN with morphological features suspicious of malignancy. From studies dealing with sporadic IPMNs, it is well known that dilatation of the main duct >10 mm, a cyst size >30 mm, or the presence of intramural nodules are risk factors for malignancy and established indications for surgical resection. When these features are absent, it is considered safe to follow a wait-and-follow up policy (31).

Evidently, from a biological and pathophysiological point of view, one would also like to resect individuals with PanIN-3 lesions. In clinical practice, however, this is not as strait forward as it may seem. At present we lack objective morphological criteria to reliably indentify such lesions on EUS or MR, although development of early features of chronic pancreatitis may provide a clue (24, 25, 27). A somewhat disturbing feature in this regard is the observation that individuals at high risk for pancreatic cancer may develop hypoechoic lesions of several millimeters in size that have been shown to disappear and could represent areas of transient focal acute pancreatitis (42).

The true clinical significance of these lesions is as yet unknown, but clearly one wants to avoid surgery in patients with a transient lesion unless it is unequivocally proved that this is a feature associated with a high risk of (future development of) malignancy. The fact that at present no ‘evident based’ (or even agreed upon) policy exists on whom to treat as a patient, the medical community should not routinely offer surveillance to individuals at high risk to develop pancreatic cancer. Instead, it should only be offered in the context of a scientific study with established criteria on whom to include and when to operate or continue surveillance in case of abnormal findings. Preferably, this data should be made publically available to a web-based world-wide registry database for which initiatives have already been undertaken by the Erasmus Medical Center in Rotterdam (the Netherlands) and the John Hopkins University in Baltimore (USA).

Principle 9: The cost of case-finding (including diagnosis and treatment of patients diagnosed) should be economically balanced in relation to possible expenditure on medical care as a whole

Cost-effectiveness analysis is a comparison tool to help evaluate choices. It may not always indicate a clear choice, but it will evaluate options quantitatively based on a defined model. For pancreatic cancer surveillance, this is currently not clearly established. Obviously, the costs of repeated MRI and/or EUS investigations can be calculated easily. However,

the impact of surveillance on generating costs on the one hand (e.g. more surgical interventions) and saving costs (e.g. avoiding expensive palliative treatments including chemotherapy and improved labor productivity) on the other is largely unknown.
The question also remains if one of both imaging modalities suffices and at which interval investigations should be repeated. Most studies use an interval of 1 year. Should this interval be the same for all individuals screened or is it possible, for example, to extend this interval in case 2 consecutive investigations are negative? Most importantly, the performance characteristics of tests for this particular surveillance purpose needs to be firmly established and the verdict is still out if EUS or MRI will detect potential malignant lesions in due time at a point when cure is still possible. On theoretical grounds and based on preliminary results from ongoing surveillance studies, expectations are favorable, but this needs to be unequivocally proved by prospective data.

Principle 10: Case-finding should be a continuing process and not a 'once and for all' project

'Single-occasion examination is clearly only of limited value, since the screening picks up those persons in the population who happen at that particular time to have the condition sought; it cannot touch the future incidence at all. … Regular offers of examination are likely gradually to cover more and more of the population at risk, including by re-examination, those patients presenting with new disease' (4).

There are multiple reasons why a surveillance program is more suited than a single screening offer for this particular situation. For one, the target population are individuals at high risk for developing cancer, not at average risk. Contrary to colon cancer screening in which precursor lesions (adenoma) can be easily removed with endoscopic polypectomy providing a considerable risk reduction for developing colon cancer (43), precursor lesions in the pancreas cannot be removed except by surgical resection. For low-risk lesions the risks of surgery do not outweigh the benefits of resection. Hence the only means to deal with such situations is to offer continued surveillance. Moreover, a normal single test outcome in these high-risk individuals will be no guarantee that pancreatic cancer will not develop in subsequent years.
Previous reports have shown that the increased risk becomes apparent as early as 45 years of age (12) and that the mean age at cancer diagnosis among FPC-probands is significantly younger than the mean age at cancer diagnosis for sporadic cases (65.3 8 11.6 years vs. 70 8 12.1 years) (44). Indeed, most screening and surveillance protocols start at the age of 45 years or 10 years earlier than the youngest age of onset of pancreatic cancer in the family. In most series, a yearly follow-up regime is adopted and preliminary data indicate that no interval cancers have developed to date.

SUMMARY AND CONCLUSION

Based on theoretical reasoning and preliminary (pre) clinical data surveillance, of individuals at high risk for pancreatic cancer by EUS and/or MRI, the outlook to fight the high death toll of this devastating disease seems promising. Careful consideration of the criteria put forward by Wilson and Jungner (4) as published by the World Health Organization on the principles and practice of screening for disease indicate that surveillance in this high-risk population should be regarded as a promising development, though experimental. It nicely points out which outstanding questions need to be addressed. Among others, these include acquiring a better understanding of the natural behavior and progression of precursor lesions towards invasive cancer, to firmly establish the performance characteristics of EUS and MRI for the detection of (early) lesions in individuals at high risk for pancreatic cancer, and to determine which lesions can be safely observed with continued surveillance and which lesions justify resection.

REFERENCES

1. Starling N, Cunningham D: Survival from cancer of the pancreas in England and Wales up to 2001. British J Cancer 2008; 99(suppl 1):S24–S25.
2. Soreide K, Aagnes B, Moller B, Westgaard A, Bray F: Epidemiology of pancreatic cancer in Norway: trends in incidence, basis of diagnosis and survival 1965–2007. Scand J Gastroenterol 2010;45: 82–92.
3. Gollust SE, Schroeder SA, Warner KE: Helping smokers quit: understanding the barriers to utilization of smoking cessation services. Milbank Q 2008; 86: 601–627.
4. Wilson JMG, Jungner G: Principles and Practice of Screening for Disease. Public Health Papers, No. 34. Geneva, WHO, 1968.
5. Intergrale Kankercentra, Nederland. www. ikcnet.nl.
6. Ferlay J, Autier P, Boniol M, Heanue M, Colombet M, Boyle P: Estimates of the cancer incidence and mortality in Europe in 2006. Ann Oncol 2007; 18: 581–592.
7. Lynch HT, Smyrk T, Kern SE, Hruban RH, Lightdale CJ, Lemon SJ, Lynch JF, Fusaro LR, Fusaro RM, Ghadirian P: Familial pancreatic cancer: a review. Sem Oncol 1996; 23: 251–275.
8. Brand RE, Lerch MM, Rubinstein WS, Neoptolemos JP, Whitcomb DC, Hruban RH, Brentnall TA, Lynch HT, Canto MI: Advances in counselling and surveillance of patients at risk for pancreatic cancer. Gut 2007; 56: 1460–1469.
9. Vasen HF, Gruis NA, Frants RR, van der Velden PA, Hille ET, Bergman W: Risk of developing pancreatic cancer in families with familial atypical multiple mole melanoma associated with a specific 19 deletion of p16 (p16-Leiden). Int J Cancer 2000; 87: 809–811.
10. Giardiello FM, Brensinger JD, Tersmette AC, Goodman SN, Petersen GM, Booker SV, Cruz-Correa M, Offerhaus JA: Very high risk of cancer in familial Peutz-Jeghers syndrome. Gastroenterology 2000; 119: 1447–1453.
11. Rebours V, Boutron-Ruault MC, Schnee M, Ferec C, Maire F, Hammel P, Ruszniewski P, Levy P: Risk of pancreatic adenocarcinoma in patients with hereditary pancreatitis: a national exhaustive series. Am J Gastroenterol 2008; 103: 111–119.
12. Klein AP, Brune KA, Petersen GM, Goggins M, Tersmette AC, Offerhaus GJ, Griffin C, Cameron JL, Yeo CJ, Kern S, Hruban RH: Prospective risk of pancreatic cancer in familial pancreatic cancer kindreds. Cancer research 2004; 64: 2634–2638.
13. Hearle N, Schumacher V, Menko FH, Olschwang S, Boardman LA, Gille JJ, Keller JJ, Westerman AM, Scott RJ, Lim W, Trimbath JD, Giardiello FM, Gruber SB, Offerhaus GJ, de Rooij FW, Wilson JH, Hansmann A, Moslein G, Royer-Pokora B, Vogel T, Phillips RK, Spigelman AD, Houlston RS: Frequency and spectrum of cancers in the Peutz-Jeghers syndrome. Clin Cancer Res 2006; 12:3209–3215.

14. van Asperen CJ, Brohet RM, Meijers-Heijboer EJ, Hoogerbrugge N, Verhoef S, Vasen HF, Ausems MG, Menko FH, Gomez Garcia EB, Klijn JG, Hogervorst FB, van Houwelingen JC, van't Veer LJ, Rookus MA, van Leeuwen FE: Cancer risks in BRCA2 families: estimates for sites other than breast and ovary. J Med Genet 2005; 42: 711–719.
15. Thompson D, Easton DF: Cancer Incidence in BRCA1 mutation carriers. J Natl Cancer Inst 2002; 94: 1358–1365.
16. Kastrinos F, Mukherjee B, Tayob N, Wang F, Sparr J, Raymond VM, Bandipalliam P, Stoffel EM, Gruber SB, Syngal S: Risk of pancreatic cancer in families with Lynch syndrome. JAMA 2009; 302: 1790–1795.
17. Birkmeyer JD, Finlayson SR, Tosteson AN, Sharp SM, Warshaw AL, Fisher ES: Effect of hospital volume on in-hospital mortality with pancreaticoduodenectomy. Surgery 1999; 125: 250–256.
18. Cleary SP, Gryfe R, Guindi M, Greig P, Smith L, Mackenzie R, Strasberg S, Hanna S, Taylor B, Langer B, Gallinger S: Prognostic factors in resected pancreatic adenocarcinoma: analysis of actual 5-year survivors. J Am Coll Surg 2004; 198: 722–731.
19. Kuhlmann KF, de Castro SM, Wesseling JG, ten Kate FJ, Offerhaus GJ, Busch OR, van Gulik TM, Obertop H, Gouma DJ: Surgical treatment of pancreatic adenocarcinoma; actual survival and prognostic factors in 343 patients. Eur J Cancer 2004; 40: 549–558.
20. Bilimoria KY, Bentrem DJ, Ko CY, Ritchey J, Stewart AK, Winchester DP, Talamonti MS: Validation of the 6th edition AJCC Pancreatic Cancer Staging System: report from the National Cancer Database. Cancer 2007; 110: 738–744.
21. Canto MI, Goggins M, Yeo CJ, Griffin C, Axilbund JE, Brune K, Ali SZ, Jagannath S, Petersen GM, Fishman EK, Piantadosi S, Giardiello FM, Hruban RH: Screening for pancreatic neoplasia in high-risk individuals: an EUS-based approach. Clin Gastroenterol Hepatol 2004; 2: 606–621.
22. Poley JW, Kluijt I, Gouma DJ, Harinck F, Wagner A, Aalfs C, van Eijck CH, Cats A, Kuipers EJ, Nio Y, Fockens P, Bruno MJ: The yield of first-time endoscopic ultrasonography in screening individuals at a high risk of developing pancreatic cancer. Am J Gastroenterol 2009; 104: 2175–2181.
23. Maitra A, Fukushima N, Takaori K, Hruban RH: Precursors to invasive pancreatic cancer. Adv Anat Pathol 2005; 12: 81–91.
24. Brune K, Abe T, Canto M, O'Malley L, Klein AP, Maitra A, Volkan Adsay N, Fishman EK, Cameron JL, Yeo CJ, Kern SE, Goggins M, Hruban RH: Multifocal neoplastic precursor lesions associated with lobular atrophy of the pancreas in patients having a strong family history of pancreatic cancer. Am J Surg Pathol 2006; 30: 1067–1076.

25. Canto MI, Goggins M, Hruban RH, Petersen GM, Giardiello FM, Yeo C, Fishman EK, Brune K, Axilbund J, Griffin C, Ali S, Richman J, Jagannath S, Kantsevoy SV, Kalloo AN: Screening for early pancreatic neoplasia in high-risk individuals: a prospective controlled study. Clin Gastroenterol Hepatol 2006; 4: 766–781, quiz 665.
26. Langer P, Kann PH, Fendrich V, Habbe N, Schneider M, Sina M, Slater EP, Heverhagen JT, Gress TM, Rothmund M, Bartsch DK: Five years of prospective screening of highrisk individuals from families with familial pancreatic cancer. Gut 2009; 58: 1410–1418.
27. Brentnall TA, Bronner MP, Byrd DR, Haggitt RC, Kimmey MB: Early diagnosis and treatment of pancreatic dysplasia in patients with a family history of pancreatic cancer. Ann Intern Med 1999; 131: 247–255.
28. Tersmette AC, Petersen GM, Offerhaus GJ, Falatko FC, Brune KA, Goggins M, Rozenblum E, Wilentz RE, Yeo CJ, Cameron JL, Kern SE, Hruban RH: Increased risk of incident pancreatic cancer among first-degree relatives of patients with familial pancreatic cancer. Clin Cancer Res 2001; 7: 738–744.
29. Hruban RH, Maitra A, Goggins M: Update on pancreatic intraepithelial neoplasia. Int J Clin Exp Pathol 2008; 1: 306–316.
30. Hruban RH, Takaori K, Klimstra DS, Adsay NV, Albores-Saavedra J, Biankin AV, Biankin SA, Compton C, Fukushima N, Furukawa T, Goggins M, Kato Y, Kloppel G, Longnecker DS, Luttges J, Maitra A, Offerhaus GJ, Shimizu M, Yonezawa S: An illustrated consensus on the classification of pancreatic intraepithelial neoplasia and intraductal papillary mucinous neoplasms. Am J Surg Pathol 2004; 28: 977–987.
31. Tanaka M: Intraductal papillary mucinous neoplasm of the pancreas: diagnosis and treatment. Pancreas 2004; 28: 282–8.
32. Canto MI, Schulick RD, Kamel IR, Fishman EK, Topazian MD, Takahashi N, Lee JH, Tamm EP, Vikram R, Syngal S, Saltzman JR, Mortele KJ, Farrell JJ, Margolis D, Zhang Z, Petersen GM, Hruban RH, Goggins MG: 415g: screening for familial pancreatic neoplasia: a prospective, multicenter blinded study of EUS, CT, and secretin-MRCP (The NCI-Spore Lustgarten Foundation Cancer of the Pancreas CAPS 3 Study). Gastrointest Endosc 2010;71:AB119.
33. Hunt GC, Faigel DO: Assessment of EUS for diagnosing, staging, and determining resectability of pancreatic cancer: a review. Gastrointest Endosc 2002; 55: 232–237.
34. Muller MF, Meyenberger C, Bertschinger P, Schaer R, Marincek B: Pancreatic tumors: evaluation with endoscopic US, CT, and MR imaging. Radiology 1994; 190: 745–751.
35. Farrell JJ, Brugge WR: Intraductal papillary mucinous tumor of the pancreas. Gastrointest Endosc 2002; 55: 701–714.
36. Pilleul F, Rochette A, Partensky C, Scoazec JY, Bernard P, Valette PJ: Preoperative evaluation of intraductal papillary mucinous tumors performed by pancreatic magnetic resonance imaging and correlated with surgical and histopathologic findings. J Magn Reson Imaging 2005; 21: 237–244.

37. Westerterp M, van Westreenen HL, Deutekom M, Stoker J, Fockens P, Comans EF, Plukker JT, Bossuyt PM, van Lanschot JJ, Sloof GW: Patients' perception of diagnostic tests in the preoperative assessment of esophageal cancer. Patient preference and adherence 2008; 2: 157–162.
38. Harinck F, Nagtegaal T, Kluijt I, Aalfs CM, Smets E, Poley J-W, Wagner A, Fockens P, Hooft van JE, Bruno MJ, Bleiker E: W1155 burden of undergoing EUS-MRI based pancreatic cancer screening in high-risk individuals; first experiences. Gastroenterology 2010;138:S663.
39. Eshed I, Althoff CE, Hamm B, Hermann KG: Claustrophobia and premature termination of magnetic resonance imaging examinations. J Magn Reson Imaging 2007; 26: 401–404.
40. Fearon ER, Vogelstein B: A genetic model for colorectal tumorigenesis. Cell 1990; 61: 759–767.
41. Chari ST, Yadav D, Smyrk TC, DiMagno EP, Miller LJ, Raimondo M, Clain JE, Norton IA, Pearson RK, Petersen BT, Wiersema MJ, Farnell MB, Sarr MG: Study of recurrence after surgical resection of intraductal papillary mucinous neoplasm of the pancreas. Gastroenterology 2002; 123: 1500–1507.
42. Harinck F, Poley J-W, Hooft Van JE, Gouma DJ, Van Eijck CH, Nio CY, Hermans J, Cats A, Wagner A, Aalfs CM, Kluijt I, Fockens P, Bruno MJ: T1463: spontaneous resolvement of focal pancreatic lesions in EUS screening of individuals at high risk of pancreatic cancer. Gastrointest Endosc 2010;71:AB283–AB284.
43. Winawer SJ, Zauber AG, Ho MN, et al: Prevention of colorectal cancer by colonoscopic polypectomy. The National Polyp Study Workgroup. N Engl J Med 1993; 329: 1977–1981.
44. Petersen GM, de Andrade M, Goggins M, Hruban RH, Bondy M, Korczak JF, Gallinger S, Lynch HT, Syngal S, Rabe KG, Seminara D, Klein AP: Pancreatic cancer genetic epidemiology consortium. Cancer Epidemiol Biomarkers Prev 2006; 15: 704–710.

F. Harinck, I.C.A.W. Konings, I. Kluijt, J-W. Poley, J.E. van Hooft, H.M. van Dullemen, C.Y. Nio, N.C. Krak, J.J. Hermans, C.M. Aalfs, A. Wagner, R. Sijmons, K. Biermann, C.H.J. van Eijck, D.J. Gouma, M.G.W. Dijkgraaf, P. Fockens, M.J. Bruno, on behalf of the Dutch research group on pancreatic cancer surveillance in high-risk individuals.

Submitted

CHAPTER 3
A MULTICENTER COMPARATIVE PROSPECTIVE BLINDED ANALYSIS OF EUS AND MRI FOR SCREENING OF PANCREATIC CANCER IN HIGH-RISK INDIVIDUALS

ABSTRACT

Objective | Endoscopic ultrasonography (EUS) and magnetic resonance imaging (MRI) are promising tests to detect precursors and early stage pancreatic ductal adenocarcinoma (PDAC) in high-risk individuals (HRI). It is unclear which screening technique is to be preferred. We aimed to compare the efficacy of EUS and MRI in their ability to detect clinically relevant lesions in HRI.

Design | Multicenter prospective study. The results of 139 asymptomatic HRI (>10-fold increased risk) undergoing first time screening by EUS and MRI are described. Clinically relevant lesions were defined as solid lesions, main duct IPMNs and cysts ≥10mm. Results were compared in a blinded, independent fashion.

Results | Two solid lesions (mean size 9mm) and nine cysts ≥10mm (mean size 17mm) were detected in nine HRI (6%). Both solid lesions were detected by EUS only and proved to be a stage I PDAC and a multifocal PanIN-2. Of the nine cysts ≥10 mm, six were detected by both imaging techniques and three were detected by MRI only. The agreement between EUS and MRI for the detection of clinically relevant lesions was 55%. Of these clinically relevant lesions detected by both techniques, there was a good agreement for location and size.

Conclusion | EUS and/or MRI detected clinically relevant pancreatic lesions in 6% of HRI. Both imaging techniques were complementary rather than interchangeable: contrary to EUS, MRI was found to be very sensitive for the detection of cystic lesions of any size, MRI however might have some important limitations with regard to the timely detection of solid lesions.

INTRODUCTION

Despite all efforts in past decades, the prognosis of pancreatic ductal adenocarcinoma (PDAC) is still dismal. With a mean survival of less than 6 months and a 5-year survival of less than 5%, PDAC ranks among the top five causes of cancer related deaths in the Western world despite its relatively low incidence (1). Survival rates are strongly dependent on the stage at which PDAC is detected. Once symptoms develop, the disease is usually at an advanced stage and consequently beyond cure. Therefore, there is great interest in pancreatic screening to detect PDAC at an earlier and potentially curable stage or, even more preferable, to detect high-grade precursor lesions.

Screening of the general population is not feasible as we currently lack a simple, reliable and inexpensive screening tool. However, evidence is starting to accumulate that screening might be worthwhile when offered to individuals at high risk of developing PDAC (2). High-risk individuals include mutation carriers of PDAC-prone gene mutations (e.g. *CDKN2A*, *BRCA1, BRCA2*, *STK11/LKB1*) and relatives of patients with familial PDAC. The risk of developing PDAC within these well-defined populations of high-risk individuals is estimated to be at least 10-fold increased compared to the general population and exceeds 76-fold in selected cases (2, 3). Previous studies have shown that screening these high-risk individuals leads to the detection of early stage PDAC and premalignant lesions (4-13).

At present, endoscopic ultrasound (EUS) and magnetic resonance imaging (MRI) are considered the most accurate techniques for pancreatic imaging within a screening setting (2, 8). Only one study (8) has prospectively compared the diagnostic yields of EUS and MRI in a blinded fashion. In this study (8), good concordance for lesion size, number and location between EUS and MRI was seen.

We conducted a prospective head-to-head blinded comparison between EUS and MRI for the detection of clinically relevant pancreatic lesions at first time screening in individuals at high risk for developing PDAC.

METHODS

Study design and sites

We conducted a multicenter prospective blinded cohort study. Participating centers were Erasmus MC-University Medical Center Rotterdam, Academic Medical Center Amsterdam, University Medical Center Groningen and the Netherlands Cancer Institute-Antoni van Leeuwenhoek Hospital.

Objective

A prospective head-to-head blinded comparison between EUS and MRI for the detection of pancreatic lesions at first time screening in individuals at high risk for developing PDAC.

Participants

Data were collected within the framework of our ongoing Familial Pancreatic Cancer Surveillance Study. Eligible for inclusion are asymptomatic individuals with an estimated ≥10-fold increased familial or inherited PDAC-risk compared to the general population (see inclusion criteria below). The minimal age for inclusion is 45 years or 10 years younger than the age of the youngest relative with PDAC, whichever occurred first. For patients with Peutz Jeghers syndrome the minimal age for inclusion is 30 years or 10 years younger than the age of the youngest relative with PDAC, whichever occurred first. Potential candidates are evaluated and recruited by a clinical geneticist to check whether inclusion criteria are fulfilled. This evaluation includes (1) obtaining a detailed personal and family medical history, (2) verification of clinical diagnoses reported by patients and family members by review of medical and pathologic records and revision of histological slides whenever available, and (3) based on the medical information including genetic testing for the suspected gene mutation(s).

Inclusion criteria

(1) Carriers of *CDKN2A* gene mutations, regardless of the family history of PDAC
(2) Peutz-Jeghers Syndrome patients (diagnosis based on a proven *LKB1/STK11* gene mutation and/or clinical diagnosis), regardless of the family history of PDAC
(3) Carriers of gene mutations in *BRCA1*, *BRCA2*, *p53*, or Mismatch Repair Gene with a family history of PDAC in at least 2 family members
(4) First degree relatives (FDR) of patients with familial pancreatic cancer. Familial pancreatic cancer-patients were defined as having at least (1) one FDR with PDAC, (2) two second degree relatives (SDR) with PDAC, or (3) one SDR relative with PDAC aged <50 years at time of diagnosis. This means that a screened individual has at least one FDR affected by PDAC and at least one SDR (scenario 1), two third degree relatives (TDR) (scenario 2) or one TDR aged <50 years at time of diagnosis (scenario 3).

Exclusion criteria

(1) Personal history of PDAC
(2) Age younger than 18 years
(3) Individuals unable to provide informed consent due to mental retardation or language barrier
(4) Upper gastrointestinal tract obstruction or stricture that does not allow passage of the endoscope
(5) Severe medical illness; ASA score ≥3

The study protocol was approved by the Ethical Committee of all participating centers and the study was conducted in accordance with the Declaration of Helsinki. All patients gave written informed consent prior to the performance of EUS and MRI.

Experimental methods

Screening techniques

EUS

All EUS procedures were carried out by five experienced endosonographers (J.W.P., P.F., M.B., H.v.D., J.v.H). Both electronic radial (Olympus UC-160 AE, Olympus Europe, Hamburg, Germany with Aloka α 5 ultrasoundprocessor, Zug, Switzerland or Pentax EG-3670 URK, Pentax Medical Europe Headquarters, Hamburg, Germany with Hitachi ultrasoundprocessor, Hitachi Medical Systems Europe, Zug, Switzerland) and curvilinear (Olympus UCT / UCP 160, Olympus Europe, Hamburg Germany with Philips HDI 5000 ultrasoundprocessor, Philips Healthcare Medical Systems, Best, The Netherlands or Aloka α 10 ultrasoundprocessor, Zug, Switzerland) instruments were used according to the personal preference of the endosonographer. Procedures were performed under conscious sedation with midazolam/fentanyl or propofol. Imaging of the pancreas was carried out from the duodenum and stomach and was digitally recorded with lossy compression (Endobase, Olympus, Hamburg). In case a relevant clinical lesion or a lesion of unknown significance was detected, both a case description and video recordings were distributed amongst all participating endosonographists for independent review. The outcome of this independent review was then presented to the local multidisciplinary Hepato-Pancreato-Biliary team consisting of gastroenterologists, surgeons and radiologists for final decision making regarding further management.

MRI

MRI was performed at a 1.5 or 3.0 Tesla machine (Signa HDxt, Discovery 450 or 750, GE Healthcare, Milwaukee, Wisconsin, United States of America; Siemens Avanto or Philips). Parameters were kept constant between different scanners where possible. The following sequences were obtained: coronal balanced steady state free precession (bSSFP) imaging with 6 mm slices, coronal and axial T2-weighted single-shot fast spin echo (SSFSE) series with 6 mm slices, axial respiratory triggered (RT) fat suppressed T2-weighted FSE series with 6 mm slices, 3D heavily T2-weighted coronal MRCP with 1,4 mm slices (with subsequent axial reconstructions) and breath-hold axial diffusion weighted imaging (DWI) series including ADC-mapping with 6 mm slices, using 3 different b-values (b = 50, 400, and 800 sec/mm2). The dynamic sequence involved fat suppressed 3D T1-weighted spoiled gradient-echo (SPGRE) series using 2 or 3 mm slices before and after intravenous administration of gadobutrol (Gadovist 1.0 mmol/mL, Bayer Schering Pharma, Berlin, Germany) at a dose of 0.1 mmol/kg body weight using automated infusion with a power injector at a flow rate of 2 mL/sec. Series were timed in the arterial, pancreatic and portal phase using bolus tracking. MRIs were scored by three highly experienced radiologists (C.N., N.K. and J.H.).

Image Interpretation and Reporting

Participating gastroenterologists and radiologists were blinded to the baseline results of either EUS or MRI imaging. Reporting of imaging findings was standardized across EUS and MRI using a Case Record Form. Items that were scored for each EUS were: type of scope (curvilinear or radial); duration of procedure; quality of video-taping; degree of visualization of pancreatic head, body and tail; presence of lesion, if yes: location, diameter, signal intensity, type, calcification, border, vessel involvement, liver metastasis, lymphadenopathy, aspect (benign/malignant/unsure), consensus reading recommended; and features of chronic pancreatitis (hyperechoic foci, hyperechoic stranding, parenchymal calcifications, parenchymal atrophy, lobularity, ductal calculi, ductectasia (defined as visible dilated side branches), irregular pancreatic duct contour, hyperechoic pancreatic duct margin, main duct dilatation (normal values 3 mm in tail, 2 mm in body and 1 mm in head), cysts. Items that were scored for each MRI were: presence of lesion; if yes: type, diameter, signal intensity, shape, border, connection with pancreatic duct (PD); stricture of PD; PD diameter; CBD dilatation; vessel involvement; lymphadenopathy; liver metastasis; parenchymal atrophy; and quality of MRI images. We specifically looked for clinically relevant abnormalities defined as solid lesions of any size and cystic lesions larger than 10 mm, see also below (14). The imaging diagnosis used for the present analysis was based on the initial description/ diagnosis provided by either the attending radiologist or gastroenterologist. Whenever there was a discrepancy between the findings of EUS and MRI with respect to clinically relevant lesions, the EUS video and MR-images were reviewed to determine whether the lesion(s) was (were) indeed not detectable by the other technique.

Clinically relevant lesions

In this manuscript we mainly focus on the detection of clinically relevant lesions. These include all solid lesions suspicious for a malignancy as well as all lesions that fulfill the revised Sendai criteria for surgery or close follow-up (14): cysts ≥3 cm, cysts with thickened/enhancing cyst walls and/or mural nodules and/or a solid component, main branch intraductal papillary mucinous neoplasms (IPMNs) with main pancreatic duct ≥10 mm in size, and side branch IPMNs with side duct dilations/cysts >10mm.

Surgical outcomes considered 'a success'

Detection and surgical treatment of (1) invasive cancer ≥T1N0M0 with negative margins, (2) multifocal PanIN 3 lesions and (3) high-grade IPMNs were defined as a successful outcome of surveillance (2).

Follow-up policy

The follow-up policy was based on the agreement of an expert panel consisting of experienced endosonographists, surgeons, radiologists and pathologists and was as follows:

(1) Annually, when EUS and/or MRI detected no pancreatic abnormalities or cystic lesions <10 mm
(2) Three months in case EUS and/or MRI detected a lesion for which a morphological diagnosis could not be readily made, hereinafter referred to as lesions with unknown clinical significance
(3) Six months in case of a detected cysts or side branch IPMN with a diameter >10mm and <30mm without malignant features (see below)
(4) Surgical resection, in case of the detection of a solid lesion morphologically suspicious for a malignancy, cystic lesion >30mm, cystic lesions with malignant features (thickened/enhancing cyst walls and/or mural nodules) or, main branch IPMN with main pancreatic duct ≥10 mm (14).

Statistical methods

Descriptive statistics were generated to describe patient and lesion characteristics. To compare both imaging test results, a percentage agreement was calculated for the detection of lesions and for location of lesions, and a Spearman's rho correlation coefficient was calculated for the size of lesions. We considered an agreement of 0.00 as poor agreement, 0.01-0.20 as slight agreement, 0.21-0.40 as fair agreement, 0.41-0.60 as moderate agreement, 0.61-0.80 as substantial agreement and 0.81-1.00 as almost perfect agreement (15). The Spearman's rho value is a single value between -1 and +1, with a value of 0 signifying no relationship between the variables and the closer the value to 1 or -1, the more positive or negative correlation exists. All analyses were conducted using the Statistical Package for the Social Sciences (version 21, SPSS Institute, Chicago, IL).

RESULTS

Patient characteristics

At September 1st 2013 a total of 166 high risk individuals were prospectively included in this study. Twenty-two individuals underwent some form of pancreatic screening prior to inclusion and were therefore excluded from this blinded baseline analysis. Furthermore, five high-risk individuals were excluded from this analysis because they either had underwent only EUS or only MRI (Figure 1). Therefore, a total of 139 individuals from 81 unique families were included in this blinded analysis of whom the baseline characteristics are summarized in Table 1. The mean age at inclusion was 51 years (SD 9.7, range 20-73 years). Sixty-three individuals (45%) were male. Sixteen individuals (12%) were current smokers at time of inclusion. Forty individuals (29%) had a medical history affected by cancer; in 24 of these individuals (60%) the cancer type was melanoma. Seventy-one individuals (51%) carried a pancreatic cancer prone gene mutation, whereas the remaining individuals stemmed from FPC-families. No FNA was performed and no procedure related adverse events occurred.

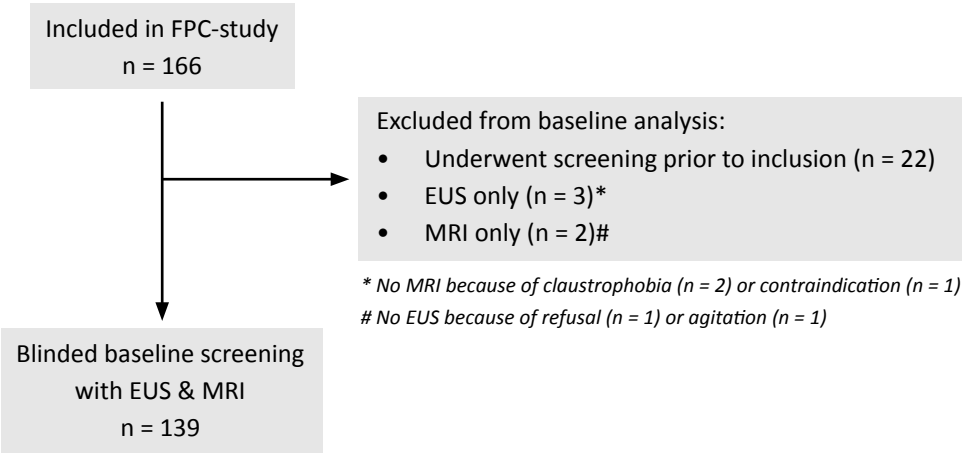


Figure 1. Flowchart

	Number included, n (%)	Mean age at inclusion, yrs (range)	Male gender, n (%)	Mean number of family members with PDAC (range)	Mean age of youngest family member with PDAC, yrs
Familial pancreatic cancer	68 (49)	53 (32-74)	32 (47)	2.7 (2-5)	53
Familial CMM *(CDKN2A)*	38 (27)	48 (20-66)	16 (42)	2.5 (0-7)	51
HBOC *(BRCA1)*	3 (2)	48 (43-57)	1 (33)	2.7 (2-3)	39
HBOC *(BRCA2)*	20 (14)	52 (39-71)	8 (40)	2.4 (2-3)	52
Peutz-Jeghers syndrome *(LKB1)*	7 (5)	52 (35-65)	5 (71)	0.2 (0-1)	54
Li-Fraumeni syndrome *(p53)*	3 (2)	43 (34-54)	1 (33%)	2 (2)	44

Table 1. Characteristics of asymptomatic high-risk individuals who underwent baseline screening with EUS and MRI (n=139). EUS, endoscopic ultrasonography; familial CMM, familial cutaneous malignant melanoma; HBOC, hereditary breast and ovarian cancer; PDAC, pancreatic ductal adenocarcinoma

Diagnostic yield

Clinically relevant lesions, as defined previously, were detected by either EUS and/or MRI in 9 out of 139 high risk individuals (6%). Two of these 9 individuals (22%) had two clinically relevant lesions. Therefore, a total of 11 clinically relevant lesions were identified in nine individuals: 2 solid lesions and 9 cysts larger than 10 mm. Main branch IPMNs and cysts with malignant features were not detected. Further characteristics are summarized in Table 2. Additionally, 8 hypo-echoic areas with unknown clinical relevance were detected by EUS in 8 individuals and 2 lesions with reduced signal intensity on TI-weighted series were detected by MRI in 2 individuals. Together with the remaining 58 cysts that were smaller than 10 mm (in 34 individuals) and 9 ductectasias (in 6 individuals), a total of 88 lesions were identified in 46 out of 139 high risk individuals (33%). Characteristics of these lesions are summarized in Table 3. No difference in findings was seen between individuals that carried a PDAC-prone gene mutation and individuals that stemmed from a FPC family.

Lesion no.	Case no.	Genetic background (no. of affected family members with PDAC)	Lesion type	Detection test(s)	Size EUS / size MRI (mm)	Location on EUS / location on MRI	Remark	Outcome
#1	1	CDKN2A (7)	Solid	EUS	11 / NA	Body / NA	Also in retrospect, the lesion was not visible on MRI	Distal pancreatectomy with splenectomy, pathology: 12mm T1N0M0 PDAC
#2	2	FPC (2)	Solid	EUS	7 / NA	Head / NA	Due to respiratory motion, the T2 sequence of MRI was of poor quality	Pancreaticoduodenectomy, pathology: multifocal PanIN2
#3	3	FPC (3)	Cystic	EUS & MRI	14 / 17	Head / Head		Unchanged at FU 12 months
#4	4	LKB1 (0)	Cystic	EUS & MRI	6 / 18	Head / Head	Endosonographist mentioned that lesion would probably be >6 mm because of elongated appearance	Again detected by both EUS and MRI at FU 12 months, EUS then described it as 20 mm
#5	5	FPC (3)	Cystic	EUS & MRI	27 / 36	Head / Head	Diagnosed as pseudocyst (history of acute pancreatitis)	Regression of the pseudocyst
#6	6	CDKN2A (2)	Cystic	EUS & MRI	12 / 12	Body / Body		Unchanged at FU 12 months
#7	6	CDKN2A (2)	Cystic	EUS & MRI	12 / 13	Tail / Tail	Same patient as #6	Unchanged at FU 12 months
#8	7	FPC (2)	Cystic	EUS & MRI	5 / 10	Tail / Tail		At FU 12 months measured at 3 mm by EUS and 5 mm by MRI
#9	8	FPC (2)	Cystic	MRI	NA / 24	NA / Head		At FU 12 months unchanged MRI findings, then also detected by EUS (8 mm)
#10	8	FPC (2)	Cystic	MRI	NA / 12	NA / Body	Same patients as #9	At FU 12 months unchanged MRI findings, then also detected by EUS (8 mm)
#11	9	FPC (3)	Cystic	MRI	NA / 10	NA / Head	MRI described lesion in uncinate process, this area could not be visualized by EUS	At FU 12 months unchanged MRI findings, again not detected by EUS

Table 2. Characteristics of all morphologically clinically relevant lesions detected at baseline screening with EUS and MRI (n=11)
No, number; PDAC, pancreatic ductal adenocarcinoma; EUS, endoscopic ultrasonography; MRI, magnetic resonance imaging; FPC, familial pancreatic cancer; FU, follow-up

	Total number detected n (%)	Number detected by EUS&MRI n (%)	Number detected by EUS only n (%)	Number detected by MRI only n (%)	Mean size, mm (range)
Solid lesions	2 (2)	-	2 (100)	-	9.0 (7-11)
Cystic lesions					
≥ 10 mm	9 (10)	6 (67)	-	3 (33)	16.9 (10-36)
< 10 mm	58 (66)	13 (22)	7 (12)	38 (66)	4.8 (2-9)
any size (total)	67 (76)	19 (28)	7 (10)	41 (61)	5.4 (2-36)
Hypo-echoic areas with unknown relevance	8 (9)	-	8 (100)	-	5.1 (2-11)
Hypo-intense areas with unknown relevance	2 (2)	-	-	2 (100)	7.0 (5-9)
Ductectasias	9 (10)	4 (44)	1 (11)	4 (44)	2.2 (2-3)

Table 3. Characteristics of all detected lesions at baseline screening with EUS and MRI (n=88)
EUS, endoscopic ultrasonography; MRI, magnetic resonance imaging

Of all 11 clinically relevant lesions, six (55%) were detected by both modalities. EUS detected a total of 8 (73%) and MRI detected a total of 9 (82%) clinically relevant lesions. When analyzing all lesions (clinically relevant lesions, hypo-echoic areas of unknown clinical relevance, hypo-intense areas of unknown clinical relevance and cysts <10mm), MRI was very sensitive for the detection of cystic lesions (of all 67 cystic lesions, 60 (90%) were detected by MRI and 26 (39%) by EUS and of all 58 cystic lesions <10 mm, 51 (88%) were detected by MRI (smallest cyst detected by MRI was 2 mm) and 20 (35%) by EUS). Conversely, EUS detected two solid lesions that were not detected by MRI, also not after re-evaluation of the MRI: (1) a 11 mm solid lesion in the body of the pancreas (Table 2, lesion 1 and Figure 2A) and (2) a 7 mm solid lesion in the head of the pancreas (Table 2, lesion 2C). For both lesions, resection was performed. The former lesion proved to be a 12 mm T1N0M0 moderately differentiated adenocarcinoma (Figure 2B). Although post-surgical staging suggested a favorable outcome (R0 resection of a small tumor of 12 mm) the patient developed local disease recurrence with liver and peritoneal metastases a few months later and died within 36 months after initial diagnosis. The 7 mm solid lesion in the head of the pancreas proved to be two separate 3 mm lesions very close to each other and was therefore classified as multifocal pancreatic intraepithelial neoplasm 2 (PanIN2) (Figure 2D). Characteristics of all detected lesions by EUS and MRI are summarized in Table 4.
Both EUS and MRI detected areas of (yet) unknown clinical relevance; these were lesions that were not cystic in nature and without the distinct morphology according to the consensus panel to be classified as a solid lesion or hypoechoic lobule. For that reason interval screening was recommended to follow these lesions. Table 5 provides a detailed description of these lesions of unknown clinical relevance. None of these cases had a history of (acute) pancreatitis or chronic ethanol overuse; only one was a heavy smoker (>15 cigarettes per day for over 40 years, case no. 5, Table 5). In all cases, except one (case no. 8, in

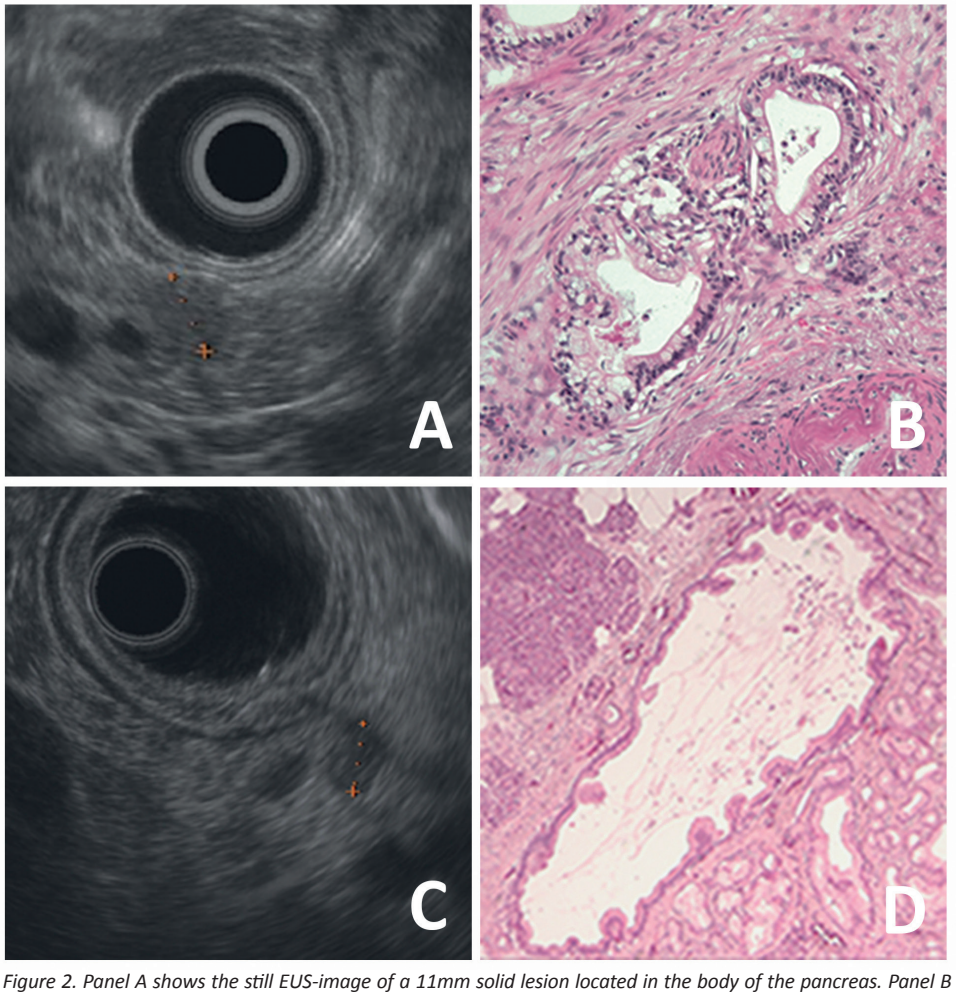


Figure 2. Panel A shows the still EUS-image of a 11mm solid lesion located in the body of the pancreas. Panel B shows the histologic image after resection of the lesion shown in panel A., which proved to be a 12mm T1N0M0 moderately differentiated pancreatic ductal adenocarcinoma. Panel C shows the still EUS-image of a 7mm solid lesion in the head of the pancreas. Panel D the histologic image after resection of the lesion shown in Panel C, which proved to be two seperate 3mm lesions, within 2 mm distance of each other, classified as multifocal PanIN2.

Table 5), follow-up showed these lesions to remain stable or being not detectable anymore. In case no. 8, EUS detected two 5 mm hypo-echoic lesions, one located in the body and one in the tail of the pancreas (lesion #8 and #9 in Table 5). Interval screening at three months was performed at which both lesions had not changed. This case was rescheduled for screening at six months during which again no morphological changes were seen. However, at follow-up at 12 months, both lesions had a more solid appearance and one of these lesions discretely increased in size (from 5 to 7 mm). Based on these morphological changes, it was decided to resect both lesions. A partial spleen preserving body/tail resection was performed and pathological examination showed multifocal PanIN2 lesions.

	n (%)	Mean size of lesions, mm	Location of lesions (n, %)		
			head	body	tail
Detected by EUS	**41**	**6.1**	**14 (34)**	**18 (44)**	**9 (22)**
- Solid	2 (5)	9.0	1 (50)	1 (50)	-
- Cystic					
≥ 10 mm	6 (15)	12.7	3 (50)	1 (17)	2 (33)
< 10 mm	20 (49)	5.2	6 (30)	10 (50)	4 (20)
any size (total)	26 (63)	6.9	9 (35)	11 (42)	6 (23)
- Unclear	8 (20)	5.1	2 (25)	4 (50)	2 (25)
- Ductectasia	5 (12)	2.0	2 (40)	2 (40)	1 (20)
Detected by MRI	**70**	**6.1**	**26 (37)**	**24 (34)**	**20 (29)**
- Solid	-	-	-	-	-
- Cystic					
≥ 10 mm	9 (13)	16.9	6 (67)	1 (11)	2 (22)
< 10 mm	51 (73)	4.8	17 (33)	19 (37)	15 (29)
any size (total)	60 (86)	6.6	23 (38)	20 (33)	17 (28)
- Unclear	2 (3)	7.0	2 (100)	-	-
- Ductectasia	8 (11)	2.3	1 (13)	4 (50)	3 (38)

Table 4. Characteristics of lesions detected by EUS and by MRI respectively at baseline screening
EUS, endoscopic ultrasonography; MRI, magnetic resonance imaging

A total of 41 out of 139 high-risk individuals (30%) had at least one feature of chronic pancreatitis: lobularity was the most frequently detected feature (19%), as well as hyperechoic pancreatic duct margins (17%) and hyperechoic stranding (15%). Twenty individuals (14%) had 3 or more features of chronic pancreatitis. No differences in features of chronic pancreatitis were seen between individuals that carried a PDAC-prone gene mutation and individuals that stemmed from a FPC family. Also, no correlation with the presence of cysts, alcohol use or tobacco use was found.

Agreement between EUS and MRI at baseline screening (blinded analysis)

The agreement between EUS and MRI for the detection of clinically relevant lesions (n=11) was moderate with a 55% agreement, see Table 6. Not surprisingly, the agreement was only fair for detection of all lesions regardless of size (n=88, agreement 26%). However, there was a perfect agreement between EUS and MRI for location of both clinically relevant lesions (n=6) and all lesions (n=26) (agreement 100%). Also, there was a substantial to almost perfect agreement between EUS and MRI on the size of clinically relevant lesions (Spearman's rho correlation coefficient of 0.638) and the size of all detected lesions (Spearman's rho correlation coefficient of 0.859).

Follow-up

A total of 135 out of 139 high-risk individuals underwent repeated surveillance after 12 months; one patient developed metastatic disease (case no. 1 in Table 2) and 3

Lesion no.	Case no.	Genetic background (no. of affected family members with PDAC)	Lesion	Detection test	Management	Pathology report	Outcome
#1	1	FPC (2)	Unclear lesion 11 mm in body of pancreas	EUS	Interval 3 months	NA	No abnormality detected at FU 3 and 12 months
#2	2	CDKN2A (3)	Unclear lesion 5 mm in head of pancreas	EUS	Interval 6 months	NA	At FU 6 and 12 months characterized as ductectasia
#3	3	CDKN2A (0)	Unclear lesion 9 mm in head of pancreas	MRI	Standard FU at 12 months	NA	No abnormality detected at FU 12 months
#4	4	CDKN2A (5)	Unclear lesion 5 mm in tail of pancreas	EUS	Interval 3 months	NA	Lesion unchanged at FU 3 and 12 months, characterized as pronounced lobule
#5	5	FPC (3)	Unclear lesion 5 mm in body of pancreas	EUS	Interval 3 months	NA	Lesion unchanged at FU 3 and 12 months
#6	6	CDKN2A (2)	Unclear lesion 2 mm in head of pancreas	EUS	Standard FU at 12 months	NA	Lesion unchanged at FU 12 months
#7	7	BRCA2 (2)	Unclear lesion 5 mm in head of pancreas	MRI	Standard FU at 12 months	NA	No abnormality detected at FU 12 months, lesion characterized as blood vessel
#8	8	FPC (4)	Unclear lesions 5 mm in body of pancreas	EUS	Interval 3 months	Multifocal PanIN2	Lesion unchanged at FU 3 months, at FU 12 months solid component → resection
#9	8	FPC (4)	Unclear lesions 5 mm in tail of pancreas	EUS	Interval 3 months	Multifocal PanIN2	Lesion unchanged at FU 3 months, at FU 12 months solid component → resection
#10	9	FPC (3)	Unclear lesion 3 mm in body of pancreas	EUS	Interval 3 months	NA	Lesion unchanged at FU 3 months, characterized as cyst at FU 12 months

Table 5. Characteristics of lesions/areas of (yet) unknown clinical relevance that were detected at baseline screening (n=10)

No, number; EUS, endoscopic ultrasonography; MRI, magnetic resonance imaging; FPC, familial pancreatic cancer; FU, follow-up

	Clinically relevant lesions	Clinically relevant lesions + lesions with unknown relevance	All lesions	Agreement
Detection				
Baseline				
Agreement per lesion	55% (n=11)	29% (n=21)	26% (n=88)	Fair to moderate
Agreement per participant	56% (n=9)	28% (n=18)	35% (n=46)	
Follow-up 12 months				
Agreement per lesion	67% (n=12)	50% (n=16)	24% (n=106)	Fair to substantial
Agreement per participant	50% (n=8)	67% (n=9)	35% (n=49)	
Location				
Baseline				
Agreement per lesion	100% (n=6)	100% (n=6)	100% (n=26)	Perfect
Agreement per participant	100% (n=9)	100% (n=18)	100% (n=46)	
Follow-up 12 months				
Agreement per lesion	100% (n=8)	100% (n=8)	100% (n=24)	Perfect
Agreement per participant	100% (n=8)	100% (n=9)	100% (n=48)	
Size				
Baseline				Substantial to
Spearman's rho per lesion	0.638 (n=6)	0.638 (n=6)	0.859 (n=26)	almost perfect
Follow-up 12 months				
Spearman's rho per lesion	0.270 (n=8)	0.518 (n=8)	0.619 (n=24)	Fair to substantial

Table 6. Agreement between endoscopic ultrasonography and magnetic resonance imaging for different variables and subsets of pancreatic lesions

individuals withdrew from the surveillance program (one individual had emigraded and two individuals provided no reason for withdrawal). At 12 months follow-up, 12 clinically relevant lesions were detected in 8 individuals (6%). Seven of these 12 lesions were unchanged compared to baseline screening (lesion #3, 4, 6, 7, 9, 10 and 11, Table 2). Two lesions increased in size: in case no. 6 (Table 2) a cyst in the pancreatic head grew from 5 to 10 mm, and in another case, a 9 mm large cyst in the tail of the pancreas grew to 13 mm, both without secondary signs of malignancy. Three newly developed clinically relevant pancreatic lesions were identified: (1) case no. 6 developed a cyst of 13 mm in the body of the pancreas which was detected by both imaging modalities; (2) case no. 2, who had underwent a pancreaticoduodenectomy, developed a new 10 mm large cyst in the pancreatic tail detected by MRI; and (3) in another case, one new 10 mm large cyst in the body of the pancreas was detected by MRI, all without secondary signs of malignancy.

Agreement between EUS and MRI at follow-up 12 months (unblinded analysis)

The agreement between EUS and MRI for the detection of clinically relevant lesions increased from 55% at baseline screening (blinded results) to 67% agreement at follow-up 12 months (unblinded results).

DISCUSSION

To determine the effectiveness of EUS and MRI in their ability to detect pancreatic lesions in high-risk individuals, we conducted a multicenter prospective study in which we compared baseline results in a blinded fashion. This nationwide, blinded prospective study shows that for detection of pancreatic lesions, in this series both tests were complementary rather than interchangeable. EUS and/or MRI showed a total of 11 morphologically clinically relevant lesions at baseline screening in 6% of participating high-risk individuals. To date, results of 12 screening studies for pancreatic cancer have been published (4-13, 18, 19). Based on these results, EUS and MRI are currently regarded as the most promising screening techniques as they are relatively widely accessible, have low morbidity rates, and, in particular, are superior to any other imaging modality with regard to the detection of small pancreatic lesions. However, data on which of these two imaging techniques is to be preferred for screening purposes are largely lacking since only one of these series was conducted in a blinded fashion (8). In this study (8), good concordance for lesion size, number and location between EUS and MRI was seen.

In our cohort however, we found a moderate to fair agreement between EUS and MRI on the detection of both clinically relevant lesions and all pancreatic lesions, but a good to perfect agreement on size and location of detected lesions. The moderate agreement between EUS and MRI on the detection of pancreatic lesions is a reflection of the fact that only 55% of the clinically relevant lesions (6 of 11) were detected by both EUS and MRI. For baseline imaging, both radiologist and endosonographists were blinded to the results of the competing imaging modality. Since both modalities were performed on the same day as much as possible, the order being dependent on availability and logistics, it was not possible to unblind investigators after the initial investigation. For follow-up investigations after 12 months however, radiologists and endosonographers were aware of the baseline results. The agreement per lesion between both techniques increased from 55% at baseline screening to 67% at follow-up surveillance. The disagreement between EUS and MRI lies mostly in the detection of cysts by EUS, and the detection of solid lesions by MRI. As a result, in this series both techniques were complementary rather than interchangeable.

EUS proved to be particularly sensitive for the detection of small solid lesions. Two solid lesions detected by EUS, including a stage I PDAC, were not detected by MRI. When MRI investigations in both cases were re-evaluated these lesions were indeed not detectable. Our results are in line with the results of previous studies which were conducted in a clinical setting (sporadic cases) that showed EUS has the highest sensitivity for the detection of <20mm pancreatic cancers when compared to other imaging modalities including MRI (18,19). Unfortunately, the long term outcome of the case with early stage PDAC was disappointing; although post-surgical staging suggested a favorable outcome (R0 resection of a small tumor of 12 mm) this patient developed local disease recurrence with liver and peritoneal metastases a few months later and died 36 months after initial diagnosis.

MRI was particularly sensitive for the detection of (small) cystic lesions. All nine cystic lesions sized ≥10mm were detected by MRI, whereas EUS detected six (66%). There are multiple possible explanations why these lesions were missed by EUS. The 24 mm cyst in the head of the pancreas (Table 2, lesion #9) was composed of multiple microcysts (Figure 3). This composition influences the penetration of the ultrasound waves with the walls of the microcysts reflecting the ultrasound waves causing the lesion not to appear as a cystic lesion on EUS. However, one still would expect the lesion to be discordant compared to the surrounding pancreatic parenchyma and thus identified as a potential 'lesion'. Indeed, at follow-up 12 months, a different endosonographer detected both lesion #9 and #10. The location of cyst #11 in the uncinate process (Table 2), could be the reason why this particular lesion was missed. This part of the pancreas is sometimes more challenging to visualize by EUS. Lastly, in both cases a radial scope was used. Although in this multicenter study the choice of the device was left to the discretion of the attending investigator, most endosonographists prefer a linear device to scan the pancreas.

Furthermore, MRI was more sensitive for the detection of subcentimeter cystic lesions. At present, the clinical relevance of detecting these subcentimeter cysts seems to be limited in particular in individuals undergoing yearly screening (14). However, longer prospective follow-up is required to understand more about the natural course of these small cystic lesions in these high-risk individuals.

Strengths of our nationwide, multicenter, prospective study are that at baseline screening participating gastroenterologists and radiologists were blinded to the results of either EUS or MRI imaging. Moreover, as a result of the extensive genetic evaluation prior to inclusion in this study and rigid inclusion criteria, our cohort consists of individuals truly at high risk for developing PDAC.

This study is limited by the fact that we lack a definitive diagnosis of the vast majority of cases in whom an abnormality was detected, in particular if detected by one imaging

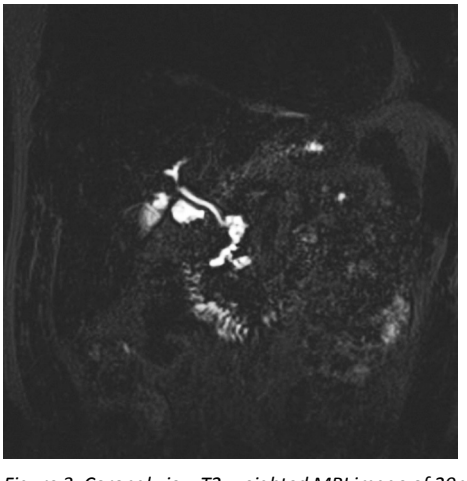


Figure 3. Coronal view T2-weighted MRI image of 20mm multicystic lesion located in the head of the pancreas.

modality only. As a resultant of this baseline screening, only two of all cases (1.4%) were operated. Consequently, it is yet impossible to make a final judgement with regard to the clinical relevance of the different types and sizes of pancreatic lesions detected. For instance, the importance of the hypo-echoic areas of unknown significance that were detected by EUS but not by MRI remains to be determined. Only longer follow-up will learn whether such findings bare clinical relevance. We are currently conducting a prospective follow-up study to assess the clinical relevance of various lesions detected by EUS and MRI and whether screening high-risk individuals is truly effective in reducing PDAC-related morbidity and mortality.

The true challenge in pancreatic cancer surveillance is to adequately identify the stage of pre-neoplastic lesions to avoid resections of early stage lesions (e.g. PanIN 1 and 2 lesions), but timely resect advanced lesions before cancer has developed. Based on the present study, it is not possible to draw definite conclusions about the (potential) merits of surveillance to prevent pancreatic cancer death. To answer this pivotal question, long-term follow-up studies are required in a large number of individuals. In this regard, it should be recognized that it has taken many years to prove that colon cancer screening saves lives. Also, despite many patients undergoing surveillance investigations, definite proof about the ability of Barrett's surveillance to prevent esophageal cancer death is still lacking to date.

In conclusion, for individuals at high risk for developing pancreatic cancer that undergo screening, EUS and MRI are rather complementary than interchangeable imaging modalities. For future screening therefore, we will continue to use both imaging modalities in the follow-up of our cohort of high-risk individuals. Given the lack of data-driven evidence of the effectiveness of PDAC screening in high-risk individuals, we believe this should be conducted within the framework of a research protocol. In contrast to EUS, MRI is very sensitive for the detection of even the smallest cysts (as small as 2 mm) of which the clinical relevance is unknown in particular in these individuals who are being screened yearly. EUS seems to be most sensitive for the early detection of (small) solid lesions, which from a clinical perspective is an important property of this imaging modality.

REFERENCES

1. Jemal A, Bray F, Center MM, Ferlay J, Ward E, Forman D. Global cancer statistics. CA Cancer J Clin. 2011;61(2):69-90. Epub 2011/02/08.
2. Canto MI, Harinck F, Hruban RH, Offerhaus GJ, Poley JW, Kamel I, et al. International Cancer of the Pancreas Screening (CAPS) Consortium summit on the management of patients with increased risk for familial pancreatic cancer. Gut. 2013;62(3):339-47. Epub 2012/11/09.
3. Korsse SE, Harinck F, van Lier MG, Biermann K, Offerhaus GJ, Krak N, et al. Pancreatic cancer risk in Peutz-Jeghers syndrome patients: a large cohort study and implications for surveillance. J Med Genet. 2013;50(1):59-64. Epub 2012/12/15.
4. Al-Sukhni W, Borgida A, Rothenmund H, Holter S, Semotiuk K, Grant R, et al. Screening for Pancreatic Cancer in a High-Risk Cohort: An Eight-Year Experience. J Gastrointest Surg. 2011. Epub 2011/12/01.
5. Brentnall TA, Bronner MP, Byrd DR, Haggitt RC, Kimmey MB. Early diagnosis and treatment of pancreatic dysplasia in patients with a family history of pancreatic cancer. Ann Intern Med. 1999;131(4):247-55. Epub 1999/08/24.
6. Canto MI, Goggins M, Hruban RH, Petersen GM, Giardiello FM, Yeo C, et al. Screening for Early Pancreatic Neoplasia in High-Risk Individuals: A Prospective Controlled Study. Clin Gastroenterol Hepatol. 2006;4(6):766-81.
7. Canto MI, Goggins M, Yeo CJ, Griffin C, Axilbund JE, Brune K, et al. Screening for pancreatic neoplasia in high-risk individuals: An EUS-based approach. Clin Gastroenterol Hepatol. 2004;2(7):606-21.
8. Canto MI, Hruban RH, Fishman EK, Kamel IR, Schulick R, Zhang Z, et al. Frequent detection of pancreatic lesions in asymptomatic high-risk individuals. Gastroenterology. 2012;142(4):796-804; quiz e14-5. Epub 2012/01/17.
9. Langer P, Kann PH, Fendrich V, Habbe N, Schneider M, Sina M, et al. Five years of prospective screening of high-risk individuals from families with familial pancreatic cancer. Gut. 2009;58(10):1410-8. Epub 2009/05/28.
10. Ludwig E, Olson SH, Bayuga S, Simon J, Schattner MA, Gerdes H, et al. Feasibility and yield of screening in relatives from familial pancreatic cancer families. The American journal of gastroenterology. 2011;106(5):946-54. Epub 2011/04/07.
11. Poley JW, Kluijt I, Gouma DJ, Harinck F, Wagner A, Aalfs C, et al. The yield of first-time endoscopic ultrasonography in screening individuals at a high risk of developing pancreatic cancer. Am J Gastroenterol. 2009;104(9):2175-81. Epub 2009/06/06.
12. Verna EC, Hwang C, Stevens PD, Rotterdam H, Stavropoulos SN, Sy CD, et al. Pancreatic cancer screening in a prospective cohort of high-risk patients: a comprehensive strategy of imaging and genetics. Clinical cancer research : an official journal of the American Association for Cancer Research. 2010;16(20):5028-37. Epub 2010/09/30.

13. Vasen HF, Wasser M, van Mil A, Tollenaar RA, Konstantinovski M, Gruis NA, et al. Magnetic resonance imaging surveillance detects early-stage pancreatic cancer in carriers of a p16-Leiden mutation. Gastroenterology. 2011;140(3):850-6. Epub 2010/12/07.
14. Tanaka M, Fernandez-del Castillo C, Adsay V, Chari S, Falconi M, Jang JY, et al. International consensus guidelines 2012 for the management of IPMN and MCN of the pancreas. Pancreatology. 2012;12(3):183-97. Epub 2012/06/13.
15. Landis JR, Koch GG. The measurement of observer agreement for categorical data. Biometrics. 1977;33(1):159-74. Epub 1977/03/01.
16. Kimmey MB, Bronner MP, Byrd DR, Brentnall TA. Screening and surveillance for hereditary pancreatic cancer. Gastrointestinal endoscopy. 2002;56(4 Suppl):S82-6. Epub 2002/09/26.
17. Schneider R, Slater EP, Sina M, Habbe N, Fendrich V, Matthai E, et al. German national case collection for familial pancreatic cancer (FaPaCa): ten years experience. Fam Cancer. 2011;10(2):323-30. Epub 2011/01/06.
18. Harinck F, Bruno MJ. Endosonography in the management of biliopancreatic disorders. Best practice & research Clinical gastroenterology. 2009;23(5):703-10. Epub 2009/09/12.
19. Kamata K, Kitano M, Kudo M, Sakamoto H, Kadosaka K, Miyata T, et al. Value of EUS in early detection of pancreatic ductal adenocarcinomas in patients with intraductal papillary mucinous neoplasms. Endoscopy. 2014;46(1):22-9. Epub 2013/11/13.

M.I. Canto, F. Harinck, R.H. Hruban, G.J. Offerhaus, J-W Poley, I. Kamel, C.Y. Nio, R.S. Schulick, C. Bassi, I. Kluijt, M.J. Levy, A. Chak, P. Fockens, M. Goggins[1], and M.J. Bruno, on behalf of the International Cancer of the Pancreas Screening (CAPS) Consortium

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CHAPTER 4
INTERNATIONAL CANCER OF THE PANCREAS SCREENING (CAPS) CONSORTIUM SUMMIT ON THE MANAGEMENT OF PATIENTS WITH INCREASED RISK FOR FAMILIAL PANCREATIC CANCER

ABSTRACT

Background | Screening individuals at increased risk for pancreatic cancer (PC) detects early, potentially curable, pancreatic neoplasia.

Objectives | To develop consortium statements on screening, surveillance and management of high-risk individuals with an inherited predisposition to PC.

Methods | A 49-expert multidisciplinary international consortium met to discuss pancreatic screening and vote on statements. Consensus was considered reached if ≥75% agreed or disagreed.

Results | There was excellent agreement that, to be successful, a screening program should detect and treat T1N0M0 margin-negative PC and high-grade dysplastic precursor lesions (pancreatic intraepithelial neoplasia and intraductal papillary mucinous neoplasm). It was agreed that the following were candidates for screening: first-degree relatives (FDR) of patients with PC from a familial PC kindred with at least two affected FDR; patients with Peutz-Jeghers syndrome; and p16, BRCA2 and hereditary non-polyposis colorectal cancer (HNPCC) mutation carriers with ≥ 1 affected FDR. Consensus was not reached for the age to initiate screening or stop surveillance. It was agreed that initial screening should include endoscopic ultrasonography (EUS) and/or MRI/magnetic resonance cholangiopancreatography not CT or endoscopic retrograde cholangiopancreatography. There was no consensus on the need for EUS fine-needle aspiration to evaluate cysts. There was disagreement on optimal screening modalities and intervals for follow-up imaging. When surgery is recommended it should be performed at a high-volume center. There was great disagreement as to which screening abnormalities were of sufficient concern to for surgery to be recommended.

Conclusions | Screening is recommended for high-risk individuals, but more evidence is needed, particularly for how to manage patients with detected lesions. Screening and subsequent management should take place at high-volume centers with multidisciplinary teams, preferably within research protocols.

INTRODUCTION

Pancreatic ductal adenocarcinoma (PC) is a deadly disease. It remains the fourth most common cause of death from cancer in the USA (1) and one of the deadliest cancers in the world. Although treatments have improved, average PC 5-year survival is <5%. Because pancreatic neoplasia detected early is potentially curable, there is interest in pancreatic screening. Because of the low incidence of PC in the general population, population-based screening has not been recommended.

Selective screening of individuals at increased risk for PC (high-risk individuals (HRIs)) based on their family history or identifiable genetic predisposition is considered worthwhile. Over the past decade, centres in the USA and Europe initiated pancreatic screening programmes. Single (2–10) and multicenter (8, 11) cohort studies have evaluated the diagnostic yield of screening (detection of asymptomatic precursor lesions and PCs at baseline and follow-up) using different imaging modalities and study populations (Table 1).

Study	High-risk Group	Imaging tests	Diagnostic yield* n (%)
Brentnall 1999(46) n=14	FPC	EUS + ERCP + CT	7/14(50) †
Kimmey 2002(47) n=46‡	FPC	EUS; ERCP§	12/46 (26) †
Canto 2004(5) n=38	FPC, PJS	EUS; ERCP§, EUS-FNA§, CT§	2/38(5.3)†
Canto 2006(4) n=78	FPC, PJS	EUS; CT§,EUS-FNA§, ERCP§	8/78(10.3) ¶, †
Poley 2009(8) n=44	FPC, *BRCA*, PJS, *p16*, *p53*, HP	EUS;CT§,MRI§	10/44(23)
Langer 2009(6) n=76	FPC, *BRCA*	EUS + MRCP; EUS-FNA§	1/76(1.3) ¶, †
Verna 2010(10) n=51	FPC, *BRCA*, *p16*	EUS and/or MRCP	6/51(12)†
Ludwig 2011 (7) n=109	FPC, *BRCA*	MRCP; EUS§, EUS-FNA§	9/109(8.3) ¶
Vasen 2011(9) n=79	*p16*	MRI/MRCP	14/79† (18)
Al-Sukhni 2011(2) n=262	FPC, *BRCA*, PJS, *p16*, HP	MRI; CT§, EUS§, ERCP§	19/262¶ (7.3)
Schneider 2011(34)** n=72	FPC,*BRCA*,*PALB2*	EUS+MRCP	11/72 (15)¶
Canto 2012(11) n=216	FPC, *BRCA*, PJS	CT, MRI/MRCP, EUS; ERCP§	5/216(2.3) † -92/216(43)

Table 1. Summary of diagnostic yield of familial pancreatic cancer screening and surveillance programmes

**Yield is defined as the detection of any pathologically proven (pre)malignant lesion (≥PanIN-2/IPMN and pancreatic adenocarcinoma) and lesions that are morphologically suspicious for branch-duct IPMNs.*

†Includes only pathologically proven pancreatic neoplasms (histology or cytology)

‡Continuation of Brentnall 1999, included 14 high-risk individuals from Brentnall 1999.

§Test performed only as an additional test for detected abnormalities.

¶Includes baseline and follow-up.

***Continuation of Langer 2009, includes high-risk individuals from this series.*

ERCP, endoscopic retrograde cholangiopancreatography; EUS, endoscopic ultrasonography; FNA, fine-needle aspiration; FPC, familial pancreatic cancer; IPMN, intraductal papillary mucinous neoplasm; MRCP, magnetic resonance cholangiopancreatography; PJS, Peutz–Jeghers syndrome; PanIN, pancreatic intraepithelial neoplasia.

AIMS AND METHODS

Scope and purpose

The International Cancer of the Pancreas Screening (CAPS) Consortium was formed in 2010 to help organise global pancreatic screening. After prior formulation of key topics for discussion, the CAPS Consortium held a multidisciplinary consensus conference. The statements developed at this conference provide recommendations related to the following questions: (1) Who should be screened? (2) How should HRIs be screened and followed up? (3) When should surgery be performed? (4) What are the goals of screening and what outcome should be considered a success?

Consortium participants

The conference chairs (Professors Canto and Bruno) selected an international multidisciplinary group of 50 experts from 10 countries in four continents representing the fields of epidemiology, genetics, gastroenterology, radiology, oncology, surgery and pathology. Participant selection was based upon expertise, publications and participation in ongoing PC screening and surveillance programmes. The group included physicians, scientists, nurses and genetic counsellors from community-based practices, academic institutions and cancer centres.

Conference proceedings

The international CAPS Consortium held a 2-day conference in February 2011, in Baltimore, Maryland, USA. A comprehensive literature search was performed and relevant publications were reviewed by conference chairs and representative experts. At the meeting, experts outlined the current state of the field. Thereafter, workgroups comprising geneticists/epidemiologists, gastroenterologists, radiologists, surgeons and pathologists met to discuss topics relevant to each specialty. Multidisciplinary groups met in breakout sessions to formulate concise statements for voting (see table 1, online Appendix). Gaps in knowledge and areas of disagreement and agreement were also identified and specifically discussed. The statements were presented after plenary and workgroup discussions, and anonymous voting was performed using touchpad technology at the end of the meeting. The resulting 100 consensus statements were reviewed and refined after the meeting and then voted on by 49 of 50 participants by anonymous electronic survey. Participants voted on multiple choice questions on a five-point scale (eg, a=definitely agree, b=moderately agree, c=neutral, d=moderately disagree, e=definitely disagree) or five-item selection list. A statement was accepted if ≥75% of the participants voted ‘agree’ or ‘disagree’. Statements that did not reach consensus are listed separately (Appendix 1).

The Appraisal of Guidelines Research and Evaluation process for assessment of quality of evidence and strength of recommendations (12, 13) was used to determine if the available literature was sufficient to make and grade recommendations. Evidence was graded

based upon study design (randomised controlled trial=high, observational study=low, any other evidence=very low), study quality, consistency and directness of evidence (12). The grade of evidence was modified if there was strong evidence of association (relative risk (RR)>2) (eg, consistent evidence from multiple observational studies, or evidence of a dose–response gradient) (12). The strength of the group's recommendation statement was based upon Grading of Recommendations Assessment, Development and Evaluation (GRADE) definitions for quality improvement and guidelines development (12, 14): 1 (strong)= 'definitely do it', 2 (weak) ='probably do it', 3 (no recommendation), 4 (weak)= 'probably don't do it' and 5 (strong)= 'definitely don't do it'.

RECOMMENDATION STATEMENTS

Each statement includes its grade of evidence, the voting results (Table 2) and a brief discussion.

Who should be screened?

Since the incidence of PC in the general population is low (lifetime risk 1.3%), screening is not recommended for the general population, but instead for individuals considered to be at high risk of developing the disease (ie, >5% lifetime risk, or fivefold increased RR). The main tool used to quantify PC risk is still the family history; risk stratification is determined from the number of affected family members and the relationships among at-risk individuals (15). Gene testing can identify a family's underlying genetic susceptibility, but it has a limited role because the genetic basis of much of the inherited susceptibility to PC remains unexplained. Additional PC susceptibility genes may be discovered in the near future that should improve our ability to identify individuals who would benefit most from pancreatic screening.

Patients with a family history of PC

Individuals with three or more blood relatives with PC, with at least one affected first-degree relative (FDR,) should be considered for screening (agree 91.9%, grade moderate, 'probably do it'). Those with at least two affected FDRs should be considered for screening (agree 91.9%, grade moderate, 'probably do it'). Individuals with two affected blood relatives with PC, with at least one FDR, should be considered for screening (agree 77.5%, grade low, 'probably do it').

These recommendations for screening are primarily based on evidence of increased risk, rather than a proven efficacy of screening. Prospective studies demonstrate an increased risk of developing PC in unaffected FDRs that depends on the number of relatives with PC (16). This risk has been estimated to be 6.4-fold greater in individuals with two FDRs with PC (lifetime risk 8–12% (17)) and 32-fold greater in individuals with three or more FDRs

Who should be screened?

	Statements
A1	Individuals with three or more affected blood relatives, with at least one affected FDR, should be considered for screening
A2	Individuals with at least two affected FDRs with PC, with at least one affected FDR, should be considered for screening once they reach a certain age.
A3	Individuals with two or more affected blood relatives with PC, with at least one affected FDR, should be considered for screening.
A4	All patients with Peutz–Jeghers syndrome should be screened, regardless of family history of PC.
A5	*p16* carriers with one affected FDR should be considered for screening.
A6	*BRCA2* mutation carriers with one affected FDR should be considered for screening.
A7	*BRCA2* mutation carriers with two affected family members (no FDR) with PC should be considered for screening.
A8	*PALB2* mutation carriers with one affected FDR should be considered for screening.
A9	Mismatch repair gene mutation carriers (Lynch syndrome) with one affected FDR should be considered for screening..

How should high risk individuals be screened?

	Statement
B1	Initial screening should include (multiple answers allowed): EUS 83.7%, MRI/MRCP 73.5%, CT 26.5%, abdominal ultrasound 14.3%, ERCP 2.0%.
B2	When previous screening did not detect an abnormality that met criteria for shortening of the interval or surgical resection, follow-up screening should include (multiple answers allowed): EUS 79.6% MRI/MRCP 69.4%, CT 22.4%, abdominal ultrasound 4.1%, ERCP 2.0%.
B3	Standardised nomenclature should be used to define chronic pancreatitis-like abnormalities.
B4	Whenever a cystic lesion is detected, an additional ERCP should not be performed.
B5	Patients with a cystic lesion without worrisome features for malignancy should have an imaging test after 6–12 months.
B6	When a solid lesion is detected, CT should also be performed.
B7	When a solid lesion is detected, ERCP should not be performed.
B8	When a solid lesion is detected at baseline with an indeterminate diagnosis and the patient is not referred for immediate surgery, imaging should be repeated after 3 months.
B9	When a new solid lesion is detected at follow-up with an indeterminate diagnosis and the patient is not referred for immediate surgery, imaging should be repeated after 3 months.
B10	If an indeterminate main pancreatic duct stricture without a mass is detected, repeat imaging should be performed within 3 months.

When should surgery be performed?

	Statement
C1	Screening should only be offered to individuals who are candidates for surgery.
C2	Pancreatic resections should be performed at specialty centres (taking into account volume, morbidity and mortality rates and expertise available).
C3	*Intraoperatively*, further pancreatectomy (up to a possible total) should be performed in patients with otherwise reasonable life expectancy to achieve R0 resection of cancer.
C4	*Intraoperatively*, further pancreatectomy (up to a possible total) should not be performed in a patient with otherwise reasonable life expectancy and no cancer but with unifocal PanIN-2 in the resected specimen but not at the margin.
C5	*Postoperatively*, further pancreatectomy (up to a possible total) should be not performed in patients with otherwise reasonable life expectancy in a patient without cancer in the resected specimen but with PanIN-2 at margin.
C6	*Postoperatively*, further pancreatectomy (up to possible total) should be not be performed in patients with otherwise reasonable life expectancy in a patient who did not have cancer but had unifocal PanIN-2 in the resected specimens but not at the margin.
C7	*Postoperatively*, further pancreatectomy (up to a possible total) should be not performed in patients with otherwise reasonable life expectancy in a patient without cancer but who has multifocal PanIN-2 in the resected specimens but not at the margin.

What are the goals of screening? What outcome(s) would be considered a "success"?

	Statement
D1	Resectable carcinoma is a potential target for early detection and treatment.
D2	PanINs are a potential target for early detection and treatment.
D3	IPMNs are a potential target for early detection and treatment.
D4	Detection and treatment of multifocal PanIN-3 should be considered a success of a screening/surveillance programme.
D5	Detection and treatment of IPMNs with high-grade dysplasia should be considered a success of a screening/surveillance programme.
D6	Detection and treatment of invasive cancer-T1N0M0 detected at baseline should be considered a success of a screening programme.
D7	Treatment of invasive cancer-T1N0M0 detected at follow-up should be considered a success of a screening programme.
D8	Detection and treatment of invasive cancer >T1N0M0 resectable with margins negative at baseline, should be considered a success of a screening programme.

Table 2. Summary of consensus statements for the management of high-risk individuals

ERCP, endoscopic retrograde cholangiopancreatography; EUS, endoscopic ultrasonography; FDR, first-degree relative; IPMN, intraductal papillary mucinous neoplasm; MRCP, magnetic resonance cholangiopancreatography; PC, pancreatic cancer; PanIN, pancreatic intraepithelial neoplasia.

with PC (lifetime risk 40% (17)). Among kindreds with familial PC, risk is higher in kindreds with a young-onset PC (age <50 years, RR=9.3) compared with kindreds without (18). No consensus was reached on whether to screen individuals without an affected FDR, including individuals with a young-onset PC relative, or patients with new-onset diabetes (Appendix Table 1).

Mutation carriers

Germline mutations in the *BRCA2*, *PALB2*, *p16*, *STK11*, ATM, *PRSS1* genes and the hereditary colon cancer (Lynch syndrome) genes, are associated with significantly increased risk of PC (19). Mutations in these genes explain only ~10% of the familial susceptibility to PC. Individuals with PC susceptibility gene mutations may not have many affected family members.

Patients with apparently sporadic pancreatic cancer can have mutations in *BRCA2*, as can those without a family history of breast, ovarian cancer (20). Incomplete or low penetrance is a common feature of familial PC susceptibility gene mutations.

Patients with Peutz–Jeghers syndrome, regardless of family history, should be considered for screening (agree 96%, grade moderate, 'do it').

Patients with Peutz–Jeghers syndrome (who generally carry germline *STK11* gene mutations) have a very high (132-fold (21)) risk of PC. Lifetime cumulative risk to age 65–70 for PC in patients with Peutz–Jeghers syndrome is 11–36% (22).

BRCA2 mutation carriers with one or more affected FDR with PC (agree 85.7%, grade low, 'probably do it') and those with two or more affected family members (even without a FDR) (agree 89.8%, grade low, 'probably do it') should be considered for screening.

Germline *BRCA2* gene mutations account for the highest percentage of known causes of inherited PC. These have been identified in 5–17% of familial PC kindreds (23–25). The RR of PC in *BRCA2* gene mutation carriers is 3.5 (95% CI 1.87 to 6.58) (26, 27). Individuals with Jewish ancestry and a family history of PC should be considered for genetic counselling and testing for the founder *BRCA2* gene mutation, 6174delT, present in 1% of Ashkenazi Jewish individuals (28) and 4% of patients with PC (29). It has not been established that the risk of PC in *BRCA1* gene mutation carriers is increased. One cohort study found a modest increased risk of pancreatic cancer (RR=2.3) (30). No agreement was reached on the question of screening *BRCA2* mutation carriers with no family history of PC (agree 51.1%), or for *BRCA1* mutation carriers with one affected FDR or two affective relatives but no FDR (agree 69.4%).

PALB2 mutation carriers with one or more affected FDR with PC should be screened (agree 77.5%, grade very low, 'probably do it').

PALB2 (partner and localiser of *BRCA2*) was recently identified as a PC susceptibility gene (31). Germline mutations have been detected in up to 3% of patients with familial PC (31–35). The magnitude of PC risk in *PALB2* mutation carriers has not been established. However, given the function of the *PALB2* gene, the risk of PC among *PALB2* gene mutation carriers is estimated to be similar to that found for *BRCA2* gene mutation carriers.

p16 mutation carriers with one or more affected FDR with PC should be considered for screening. (agree 87.8%, grade low, 'probably do it').

Germline *p16* gene mutations are found in families with familial atypical multiple mole melanoma syndrome (FAMMM syndrome), an autosomal dominant disease with variable penetrance. PC risk among *p16* gene mutation carriers is estimated to be increased 13- to 22-fold, compared with the general population (36–38).

Patients with Lynch syndrome and one affected FDR with PC should be considered for screening. (agree 87.5%, grade low, 'probably do it').

Patients with mismatch repair gene (*MLH1*, *MSH2*, *MSH6*, *PMS2*) gene mutations (Lynch syndrome) have an estimated lifetime risk of 3.7% of developing PC (8.6-fold higher risk) (39 40). Patients with PC having histology characteristic of mismatch repair-deficient cancers ('medullary' histology) (41) should have their pedigree evaluated for possible hereditary non-polyposis colorectal cancer.

The estimated lifetime risk of PC in individuals with hereditary pancreatitis is high (about 40%) (42). Many of these individuals have germline *PRSS1* gene mutations. This PC risk is directly related to the duration of recurrent pancreatitis and chronic inflammation (43). Screening of *PRSS1* mutation carriers with longstanding chronic pancreatitis is being performed within established programmes (44) but it is controversial whether healthy siblings with a *PRSS1* mutation should also be screened.

At what age should screening begin and end?

There was disagreement about the age to initiate screening in HRIs (Appendix Table 1). For individuals with familial PC, the average age at diagnosis is 68 (18). Fifty-one per cent voted to recommend starting screening at age 50. In contrast, screening typically begins at age 40 in *PRSS1* mutation carriers with hereditary pancreatitis owing to younger age of onset of PC (45). Smokers with a family history of PC have a greater risk of developing PC than non-smokers (18), but there was no consensus as to whether to recommend initiating screening at an earlier age for current smokers (Appendix Table 1). There was also no consensus recommendation about the age to end screening for HRIs without pancreatic lesions (Appendix Table 1).

How should high-risk individuals be screened?

Published screening studies have employed different screening tests. Direct interpretation of screening modalities is limited by differences in study populations, and reported diagnostic yields have ranged between 1.3% and 50%, depending on whether resected neoplasms or pancreatic lesions were tabulated (Table 1) (2, 4, 6–9, 34, 46, 47). Of 1040 HRIs screened, to date only 70 (6.7%) had a pancreatic lesion or suspected neoplasm resected (2, 4, 6–9, 34, 46, 47). Small cysts (branch-duct intraductal papillary mucinous neoplasms or BD-IPMNs) are the most common abnormality detected (in 34% and 53% of HRIs aged 50–59 and 60–69, respectively (11)). Solid masses are rarely detected (20 of 70 resected were pancreatic ductal adenocarcinomas) (2, 4, 6–9, 34, 46, 47). The number of incident PCs detected in published studies is likely to be unreported (only four were cohort studies, all with limited follow-up), but eight of 20 (40%) of the PCs diagnosed in screened HRIs were not detected at baseline screening.

Initial screening should include (multiple answers allowed): endoscopic ultrasonography (EUS) (agree 83.7%, grade moderate, 'do it'), MRI/magnetic resonance cholangiopancreatography (MRCP) (agree 73.5%, grade moderate, 'do it'), CT (26.5%), abdominal ultrasound (14.3%), endoscopic retrograde cholangiopancreatography (ERCP) (2%).

EUS and MRI are considered the most accurate tools for pancreatic imaging and do not involve ionising radiation. Few studies have compared the diagnostic yield of imaging tests for HRIs in screening, and most comparisons have not been performed in a blinded, randomised fashion. The prospective CAPS3 study (published after the CAPS summit) performed blinded comparisons of standardised pancreatic-protocol CT, secretin-enhanced MRI/MRCP and EUS for one-time screening (11). It showed that EUS and MRI are better than CT for the detection of small, predominantly cystic, pancreatic lesions, with good to excellent concordance of lesion number, size and location between EUS and MRI/MRCP. EUS, MRI/MRCP and CT identified pancreatic lesions in 42.6%, 33.3% and 11% of screened HRIs, respectively (11). MRCP provided the best visualization of cyst communication with the main pancreatic duct.

Incorrect diagnosis of lesions identified by EUS and/or MRI is a significant concern, particularly in the screening process. Some cysts are found at resection to be benign serous cystadenomas, while other resected pancreata have only low-grade pancreatic intraepithelial neoplasia (PanIN) associated with lobulocentric parenchymal atrophy (4–7, 34). These results highlight the risk of overtreatment using available screening tests. The risk of overtreatment for pancreatic screening is magnified by the risks of morbidity and mortality (~1–2%) of pancreatic surgery. The risk of incorrect diagnosis is particularly true for EUS, an operator-dependent test with only modest interobserver agreement (48).

There was excellent agreement that radiation exposure and the suboptimal detection

rate preclude CT from being a routine pancreatic screening test (2, 11, 46). Abdominal ultrasound and ERCP were also not recommended for screening, owing to their low diagnostic sensitivity and the risk of pancreatitis, respectively.

Additional tests

ERCP should be performed as an additional test if a solid lesion (disagree 77.5%, grade high, 'don't do it') or cystic lesion (disagree 77.5%, grade moderate, 'don't do it') is detected. When a solid lesion is detected, CT should be performed (agree 75.5%, grade, low, 'do it').

When ERCP was performed routinely for abnormal EUS results, it did not improve diagnostic yield and was associated with a 7% pancreatitis rate (4).

No consensus was reached on the role of EUS-guided fineneedle aspiration (FNA) to evaluate solid or cystic lesions in asymptomatic HRIs (Appendix). The role of EUS-FNA in the clinical management of most pancreatic cysts is limited, given the low accuracy of cytology in cystic lesions (49, 50) and the low volume of cyst fluid aspirated from small cysts. False-positive cytology from subcentimetre solid indeterminate lesions may also lead to unnecessary surgery (5 9).

Multidetector pancreatic-protocol CT was recommended for evaluation of solid lesions identified by EUS or MRI. The level of evidence that supports this agreement is low.

Surveillance

For routine follow-up, the best imaging test is (multiple answers allowed): EUS (79.6%), MRI/MRCP (69.4%), CT (22.4%), abdominal ultrasound (4.1%), ERCP (2%).

How should surveillance be performed after baseline screening? Published studies have generally used the same imaging tests for follow-up as for baseline imaging. There was no consensus reached on the ideal screening interval in the absence of pancreatic abnormalities at baseline, but 73.5% of participants suggested a 12-month interval. There is only indirect and limited evidence to support this recommendation. The vast majority of individuals in whom a clinically relevant lesion developed during follow-up had pancreatic abnormalities at baseline (4, 9). Furthermore, HRIs who presented with an advanced pancreatic malignancy after prior normal or indeterminate imaging were diagnosed ≥12 months later (2, 9).

Patients with a non-suspicious cyst should have an imaging test after 6–12 months (agree 83.7%, grade moderate, 'do it'). Patients with a newly detected indeterminate solid lesion should have follow-up screening at 3 months, if surgery is not imminent (agree 85.7%, grade low, 'do it'). If an indeterminate main pancreatic duct stricture is detected, repeat imaging should be performed within 3 months (agree 95.9%, grade low, 'do it').

Cystic branch-duct lesions (presumed BD-IPMNs) without concerning features indicating malignancy (51) should be re-evaluated at intervals depending on size, similar to accepted international consensus guidelines for sporadic BD-IPMNs (51). The majority of such BD-IPMNs remained stable during follow-up (2, 6, 7, 9, 34).

Small (<1 cm diameter) lesions identified as solid by EUS are difficult to manage, particularly when not detected by MRI or CT. These lesions can be aspirated but the yield for these lesions is low. Some indeterminate solid lesions identified only by EUS are cancers, but they can be benign lesions, such as non-metastatic pancreatic neuroendocrine tumours (2, 4, 11) or lowgrade PanIN with focal associated lobulocentric parenchymal atrophy (5). There was no consensus reached on the need for additional tests such as CT, ERCP, FNA, or timing of repeat imaging (although 73.5% suggested 3 months) to evaluate these lesions (Appendix Table 1).

Long-term follow-up of PC screening cohorts is lacking, (maximum follow-up period; 10 years (9, 34), mean follow-up time of 4 (9) to 4.2 years (2)). Importantly, the group acknowledged that until there are additional studies we will not know if screening HRIs saves lives.

When should surgery be performed? What type of surgery should be performed?

Screening should only be offered to individuals who are candidates for surgery (agree 75.5%, grade moderate, 'do it'). Pancreatic resections should be performed at high-volume specialty centres (agree 100%, grade moderate, 'do it').

Determining when surgery is required for pancreatic lesions is difficult and is best individualised after multidisciplinary assessment, preferably within research studies.

There is little consensus about which lesions detected by screening require surgery. The few published reports are based on limited numbers of patients (52, 53). Because of the risks of pancreatectomy, prophylactic surgery is not recommended for asymptomatic HRIs without an identifiable lesion. When indicated, pancreatic surgery is best performed at a high-volume specialty centre. Multiple studies have shown volume directly correlates with outcomes (54, 55).

Unambiguous solid lesions (≥1 cm, or seen by multiple imaging modalities) are ominous and the threshold for removing them is much lower. There was no consensus as to whether any indeterminate solid lesions detected by EUS should be resected (Appendix Table 1).

The majority of cystic lesions detected by screening appear to be low-risk branch-duct IPMNs (Table 1). The Sendai international consensus guidelines have been developed and updated (56) to help stratify patients with an IPMN as low risk versus high risk for either developing or currently harbouring a malignancy (51). In subjects with suspected BD-IPMNs, resection is considered if the patient has symptoms attributable to the cyst(s), if the cysts are >3 cm in size, or if the cysts contain mural nodules. Logic would dictate that if these are the recommendations for subjects without a strong family history of PC, then

these thresholds for resection should be either the same or lower in subjects with a strong family history. There was no consensus on the size criterion for resection of suspected BD-IPMNs or other cysts in HRIs but the majority agreed that surgery should be considered for suspected BD-IPMNs which were ≥2 cm (Appendix Table 1). Pathologically confirmed PanIN-3 lesions have been found in the pancreata of individuals who had resections of IPMNs smaller than 1 cm (5, 11). High-grade dysplasia and main-duct involvement have been identified at resection of some individuals who had surgery for one or more small (<3 cm) BD-IPMNs (4, 5, 11, 34). However, there is insufficient evidence to lower the threshold criteria for surgery for patients with lesions identified during pancreatic screening.
Management of patients with resected lesions was discussed, particularly how the preliminary and final pathology results, including margin status, should influence operative treatment.

Intraoperatively, further pancreatectomy (including total pancreatectomy) should generally be performed to achieve R0 resection of cancer (agree 75.5%, grade low, 'do it'). Intraoperatively, further pancreatectomy (including total pancreatectomy) should not be performed on patients with only unifocal PanIN-2 in the resected specimen, (agree 77.6%, grade low, 'don't do it').

The presence of PanIN-3 at the margin should be dealt with in consideration of the overall medical condition and life expectancy of the patient. The presence of PanIN-2, low-grade IPMN or intermediate-grade IPMN (on either frozen or permanent sections) at the margin or in the resection specimen should not drive further resection.
In patients undergoing surgery for invasive PC, complete resection of the cancer is recommended. If only PanIN is at the margin, it is considered unlikely that resection of additional parenchyma would be beneficial, even if the PanIN is high grade (57). Importantly, it is difficult to grade PanIN in intraoperative frozen sections.

Postoperatively, further resection of the pancreas to remove PanIN-2 at the margin should be performed in high-risk patients without PC (disagree 79.5%, grade low, 'don't do it'). Postoperatively, further resection of the pancreas should be performed because unifocal PanIN-2 (disagree 81.6%, grade low, 'don't do it') or multifocal PanIN-2 (disagree 77.5%, grade low, 'don't do it') was found anywhere in the resected specimen.

Multiple scenarios for consideration of further pancreatectomy after R0 resection of cancer did not reach consensus agreement, including management of PanIN-3 at the margin (Appendix Table 1). PanIN-3 at the resection margin in non-familial patients treated for PC does not significantly affect the postoperative course (57). However, follow-up imaging was recommended less than 6 months after surgery if there was any PanIN-3 in the resected pancreas of individuals without PC.

What are the goals of screening? What outcome(s) would be considered a 'success'?
Screening HRIs can be considered successful if it can be shown that the benefits outweigh the costs of screening. The goal of screening is the reduction of pancreatic cancer-related mortality. Evidence for success is best provided by large randomised controlled trials in which the outcomes of subjects who undergo surveillance are compared with appropriate controls, as has been demonstrated for colonoscopy screening (58). However, given the relatively low incidence of familial PC, such trials are difficult to undertake. Surrogate end points that define the success of pancreatic screening are therefore needed. Pathological staging is critical and a standard protocol for handling pancreatic resection specimens is recommended (Appendix). The following questions (What are the goals of screening? What outcome(s) would be considered a success?) were designed to define surrogate end points of screening, such as resection of potentially curable lesions (high-grade precursor neoplasms and early invasive carcinomas), as these lesions, if left untreated, can progress to incurable and lethal disease.

One target for early detection and treatment is resectable carcinoma (agree 83.7%, grade high, 'do it'). Detection and treatment of early invasive cancer (T1N0M0) (agree 89.8%, grade high, 'do it') at baseline or follow-up (agree 77.5%, grade moderate, 'do it') should be considered a success. Detection and treatment of any invasive resectable PC at baseline screening should also be considered a success of the screening programme (agree 89.8%, grade high, 'do it').

Long-term survival can be achieved by resecting small nonmetastatic PCs (59, 60), particularly if margins are negative for invasive PC (R0 resection) (57, 61). However, most patients who undergo an R0 resection of their pancreatic cancer will die from their disease. Survival is most likely for patients with the smallest cancers (T1N0M0). A critical statistic which screening and surveillance programmes should track is the number of high-risk patients that need to be screened and treated (62), which considers the likelihood that treatment will prevent the target event of PC at the expense of adverse events. In one prospective screening study of high-risk Leiden *p16* mutation carriers using MRI/MRCP, nine invasive pancreatic PCs were detected and treated in 79 patients followed up for a median of 4 years. The number of patients needed to be screened to detect and treat one PC was 11.9
Well-differentiated neuroendocrine tumours (PanNETs) have been detected within familial PC screening programmes (2, 4, 11). PanNETs <0.5 cm (microadenomas) are essentially benign lesions. Resection of PanNETs between 0.5 and 1.0 cm is generally curative (63). There was no consensus as to whether detecting and treating PanNETs should be considered a success of screening (Appendix Table 1).

One potential target for early detection is PanIN (agree 81.7%, grade high, 'do it'). Detecting and treating multifocal PanIN-3 should be considered a success (agree 83.7%, grade moderate, 'do it'). Whether to detect and treat unifocal PanIN-3 did not reach consensus (agree 73.5%)

The strength of the evidence linking sporadic PanIN-3 lesions to invasive carcinoma is based on clinical associations and genetic analyses (64–66). Similarly, strong evidence supports the hypothesis that some of the invasive PCs that arise in patients with a family history of pancreatic cancer arise from PanIN lesions (67, 68). Although PanINs are a well-accepted precursor of sporadic and familial PC, the frequency and rate at which PanINs progress to invasive carcinoma is not known. In a nonfamilial population, it is estimated that the average adult pancreas has five PanINs and that 0.86% of these progress to invasive cancer (69). It may take a decade or more for an early precursor cell (a low-grade PanIN) to progress to PC, and initial estimates suggest that the first invasive cancer cell may take several years to extend beyond the pancreas or metastasize. Importantly, the 'window' for clinical detection of an invasive PC is shorter since these lesions are only detected once they reach a certain size (70). These estimates may not apply to patients with a strong family history of PC, especially those with specific genetic mutations that may increase the rate at which precursor lesions progress.

Another target for early detection and treatment is IPMN (agree 87.7%, evidence high, 'do it'). Detection and treatment of IPMN with high-grade dysplasia should be considered a success of a screening/surveillance programme (agree 95.9%, evidence high,'do it').

IPMNs, particularly IPMNs with high-grade dysplasia, are associated with a significantly increased risk of invasive PC (67, 68, 71, 72). The IPMN phenotype has been described in familial PC relatives and gene mutation carriers (73). The frequency and rate at which IPMNs in HRIs progress to invasive PC are not well known. In patients with apparently sporadic noninvasive IPMNs it may take 3–5 years for a clinically detectable non-invasive lesion to progress to an invasive PC (74). Furthermore, in patients with small BD-IPMN(s) followed up over 5 years, only 2.4–6.9% of these lesions progressed to invasive PC (75, 76).

Important areas where there was lack of consensus: areas for future research

Topics that did not reach consensus voting are listed in the Appendix Table 1. Additional evidence is required to more accurately answer important questions, such as who to screen, when to begin screening and the frequency of screening. Some of the important gaps in knowledge pertain to the optimal age at which to begin screening, the role of as yet unidentified PC susceptibility genes as a guide to optimising screening and how to incorporate environmental risk factors such as smoking, diabetes, obesity and other exposures into risk stratification.

The CAPS summit recommended prioritising research into areas where there was lack of consensus: diagnostic evaluation and management of cystic and solid lesions detected by screening and postoperative management. Importantly, the group recommends collaborative multicentre institutional review board-approved studies to collect data on demographics, family history, risk factors, and to bank tissue, juice, aspirated fluid and blood to improve biomarker prediction of the risk of progression to PC in HRIs. Ultimately, research into how to improve screening methods, the outcomes of screening and surveillance for PC and the cost-effectiveness of alternative approaches, is of the highest priority.

SUMMARY

Screening studies have identified pancreatic neoplasms in asymptomatic patients with strong family histories of PC. However, available evidence supporting screening and surveillance is limited to observational studies. The diagnostic yield from pancreatic screening depends on many factors, including the extent of an individual's family history, the age at which screening begins and the screening modality used. Screening may also lead to the discovery of incidental or indeterminate lesions, resulting in diagnostic confusion and uncertain management. There is a clear need to improve approaches to screening of HRIs. The management of asymptomatic pancreatic lesions detected by imaging tests remains the most challenging aspect of screening and surveillance programmes. Individualised decision-making within multidisciplinary programmes and prospective research studies is essential. The findings of this workgroup should standardise current efforts and serve as a platform for the development of future multidisciplinary research protocols.

Who should be screened?

%Agree	%Neutral	%Disagree	Statement
25.0	12.5	62.5	Individuals with 2 relatives on the same side of the family (no FDR) with PC should be screened once they reach a certain age.
57.2	22.4	20.4	All p16 carriers should be screened, regardless of family history.
53.0	26.5	20.4	HNPCC carriers with 2 affected relatives, no FDR, should be screened.
43.9	34.7	22.4	HNPCC carriers with 1 affected relative, no FDR, should be screened.
37.5	12.5	37.5	Individuals with 2 blood relatives (no FDR) with PC one <50 years at diagnosis should be screened once they reach a certain age.
			In general, for the defined high-risk groups, screening should begin at age ___, or 10 years younger than earliest PC in the family (except PJ**S**). 40 yr 18.4% 45 yr 28.6% 50 yr 51%
			Peutz-Jegher syndrome patients should have screening beginning at age ___. 30 yr 36.7% 35 yr 14.3% 40 yr 36.7%
71.5	12.2	16.3	New-onset diabetes in a high risk individual should lead to initiation of screening, regardless of age.
55.1	28.6	16.4	Current smokers should start screening at 5 years earlier than nonsmokers.
			Screening should stop at age ___ for an individual in a surveillance program with no evidence of a lesion. Never 16.3% 85 18.4% 80 36.7% 75 26.5% 70 2.0% ***Stop at age 75: 81.6%***

Appendix Table 1. Statements for the managment of high-risk individuals without consensus agreement or disagreement.

How should high risk individuals be screened?

%Agree	%Neutral	%Disagree	Statement
26.5	40.8	32.6	When performing MRI, one should always use secretin.
34.7	32.7	32.7	In the presence of severe chronic pancreatitis, EUS-screening should be discontinued
30.6	8.2	61.3	In case of detection of a cystic lesion, EUS-FNA should always be performed.
			In case of detection of a cystic lesion, EUS-FNA should be performed only when the size is larger than: 5 mm 8.2% 10mm 30.6% 20mm 32.7% 30mm 6.1% EUS-FNA should never be performed. 22.4% ***Do EUS-FNA for cyst ≥ 10 mm: 69.4%***
32.6	18.4	49.0	Whenever a cystic lesion is detected, CT should be performed.
			After a cystic lesion is detected at baseline screening and the morphological characteristics do not meet criteria for surgical resection (56), an imaging test should be repeated after ___ months. 3 months 16.3%, 6 months 53.1%, 12 months 30.6%, 24 months 0, 36 months 0 ***Repeat imaging at 6-12 months: 83.7%***
			After a cystic lesion is newly detected at follow-up screening and the morphological characteristics do not meet criteria for surgical resection (56), an imaging test should be repeated after ___ months. 3 months 30.6%, 6 months 53.1%, 12 months 16.3%, 24 months 0, 36 months 0 ***Follow-up screening at 3-6 months: 83.7%***
67.4	12.2	20.4	In case of the detection of a solid lesion, EUS-FNA should always be performed.
			In case of the detection of a solid lesion, EUS-FNA should only be performed when the size is larger than: 5 mm 53.1%, 10 mm 24.5% 15 mm 0, 20 mm 0 EUS-FNA should never be performed 22.4% ***Do EUS-FNA for solid lesions (regardless of size): 77.6%***
51.0	14.3	34.7	In case of a MPD-stricture, EUS-FNA should always be performed.
38.8	18.4	42.8	In case of an indeterminate MPD-stricture, without a mass by EUS, EUS-FNA should be performed.
67.3	14.3	18.4	In case of a MPD-stricture, CT should also be performed
44.9	34.7	20.4	In case of a MPD-stricture, ERCP should also be performed

Continuation of Appendix Table 1. Statements for the managment of high-risk individuals without consensus agreement or disagreement.

When should surgery be performed?

%Agree	%Neutral	%Disagree	Statement
59.2	20.4	20.5	Enucleation of pancreatic lesions is not indicated.
61.2	6.1	32.7	Prophylactic resection is performed for a patient with no pancreatic lesion but strong family history or genetic syndrome.
69.4		30.6	Any detectable solid lesion by EUS (and not biopsy proven or highly suspicious to be neuroendocrine, autoimmune, and other known benign conditions) should be resected.
			In making a decision to resect a solid lesion, size should be considered. The size should be at least: Any size 34.7%, 5 mm 34.7%, 7 mm 8.2%, 10 mm 22.4% 15 mm 0% ***Resect any solid lesion ≥ 5 mm: 65.3%***
67.4	16.3	16.4	Each of the following criteria are indications for resection of IPMN[1] when detected in a high-risk individual: cyst > 2 cm (different from sporadic); mural nodule in cyst (= to sporadic); symptoms including pancreatitis, jaundice, pain (= to sporadic); main duct diameter > 5 mm (= to sporadic)
			Each of the following are criteria for resection of IPMN[1] when detected in a high-risk individual: cyst size > ___ cm. 1 cm 10.2%, 2 cm 36.7%, 3 cm 38.8%, 4 cm 2%,disregard size 12.2% ***IPMN size ≥ 2 cm:77.5%***
Intra-operatively, further pancreatectomy (up to a possible total) should be performed in patients with otherwise reasonable life expectancy in the following situations:			
49.0	16.3	34.7	patient with R0 resection of an invasive N0 cancer BUT with the presence of PanIN-3 at margin
24.5	20.4	55.1	patient with R0 resection of an invasive N0 cancer BUT with the presence of PanIN-2 at margin
49.0	20.4	30.6	patient without cancer BUT with PanIN-3 at the margin
12.2	22.4	65.3	patient without cancer BUT with PanIN-2 at the margin
32.7	22.4	44.9	patient with R0 resection of cancer and multifocal high grade dysplasia in the resected specimen but NOT at the margin
28.6	18.4	53.0	patient with R0 resection of cancer and unifocal high grade dysplasia in the resected specimen but NOT at the margin
32.6	22.4	44.9	patient with no cancer BUT with multifocal PanIN-3 in the resected specimen but NOT at the margin
8.1	24.5	67.4	patient without cancer BUT with multifocal PanIN-2 in the resected specimen but NOT at the margin
18.4	26.5	55.1	patient without cancer BUT with the presence of unifocal PanIN-3 in the resected specimens, NOT at the margin
Postoperatively, further pancreatectomy (up to a possible total) should be performed in patients with otherwise reasonable life expectancy in the following situations:			
61.2	12.2	26.5	To achieve R0 resection of cancer.
25.0	16.3	38.8	patient who already had R0 resection of an invasive N0 cancer but has PanIN-3 at the margin
4.0	22.4	73.4	patient who already R0 resection of an invasive N0 cancer but has PanIN-2 at the margin

Continuation of Appendix Table 1. Statements for the managment of high-risk individuals without consensus agreement or disagreement.

42.9	26.5	30.6	patient without cancer in the resected specimen BUT has PanIN-3 at the margin
22.4	18.4	59.2	patient who had R0 resection of cancer but had multifocal high grade dysplasia in the resected specimen but NOT at the margin
10.2	18.4	71.4	patient who underwent R0 resection of cancer but had unifocal high grade dysplasia in the resected specimen but NOT at the margin
22.4	26.5	51.0	patient who did not have cancer but had multifocal PanIN-3 in the resected specimen but NOT at the margin
10.2	20.4	69.4	patient who did not have cancer but had unifocal PanIN-3 in the resected specimen but NOT at the margin
0	18.4	81.6	patient who did not have cancer but had unifocal PanIN-2 in the resected specimens but NOT at the margin
			Follow-up imaging should be performed __ months after surgery with any PanIN 3 in the resected pancreas. 3 months 32.7%, 6 months 46.9%, 12 months 20.4%, 24 months, 36 months 0

What are the goals of screening? What outcome(s) would be considered a "success"?

%Agree	%Neutral	%Disagree	Statement
38.8	28.6	32.7	One of the pathologic lesions that is a potential target for early detection and treatment is extra-pancreatic neoplasm
73.5	12.2	14.3	Detection and treatment of unifocal PanIN-3 should be considered a success of a screening program.
59.1	14.3	26.6	Detection and treatment of IPMN with low or intermediate grade dysplasia should be considered a success of a screening program.
61.2	4.1	34.7	Detection and treatment of invasive cancer >T1N0M0 resectable with margins negative on follow-up, should be considered a success of a screening program.
			Detection and treatment of pancreatic neuroendocrine tumor (PancNet) should be considered a success of a screening program. Irrespective of size 34.7%, > 5mm 22.4%, > 10mm 36.7%, > 15mm 0, > 20 mm 6.1% ***Detection and treatment of any pancreatic neuroendocrine tumor should be considered a success: 65.2%***
16.3	32.7	51	There is evidence-based medicine that supports the contention that precursor lesions in high risk groups PROGRESS FASTER to invasive cancer than do precursor lesions in the general population.
30.6	26.5	42.9	There is evidence-based medicine that supports the contention that precursor lesions in high-risk individuals ARE MORE LIKELY TO PROGRESS to invasive cancer than precursor lesions in the general population.
16.3	32.7	51.0	I suspect that precursor lesions in high risk groups PROGRESS FASTER to invasive cancer than do precursor lesions in the general population.
30.6	26.5	42.9	I suspect that precursor lesions in high risk individuals ARE MORE LIKELY TO PROGRESS to invasive cancer than precursor lesions in the general population.

Continuation of Appendix Table 1. Statements for the managment of high-risk individuals without consensus agreement or disagreement.

APPENDIX

Standard pathology protocols for the handling of pancreatic resections and their reporting

The goals of the pathologic examination of pancreata removed as part of screening studies are to establish the diagnoses and to prepare well-oriented biosamples for future studies. Since the lesions removed are often small, and the pancreas is prone to autodigestion, the preparation of these biosamples in a timely manner is critical. Resected specimens should be examined fresh. Surgical margins and any other clinical frozen sections for intraoperative consultations should obviously take priority. The specimen should be carefully oriented.If invasive cancer is not seen grossly, the pathologist should start at one end and serially bread-loaf the pancreas in 1-2mm slices. Each slice should be examined grossly and any lesions can be photographed.For research purposes, consideration should be given to harvesting tissue for laser capture microdissection. To harvest this tissue, every ~4th slice and any gross lesions should then be placed in Optimal Controlled Temperature (OCT) media and sectioned for frozen section.A 5-micron hematoxylin and eosin (H & E) section should be prepared on a regular slide, and then 20 unstained frozen sections (cut at 10 microns) should be prepared on slides, to allow for laser capture microdissection, and immediately placed in deep freezer.These sections then represent well-oriented, well-preserved representative sections of the pancreas and all grossly visible lesions on appropriate slides for laser capture microdissection. If an invasive PC is grossly identified, then after the processing described above, the specimen should be prepared for research studies if appropriate institutional review board (IRB) approval is in place, such as storing the cancer fresh-frozen in a tumor bank and xenografting. One also needs to bank fresh-frozen normal tissue, including spleen, normal pancreas, and/or normal duodenum for research.

REFERENCES

1. Jemal A, Bray F, Center MM, et al. Global cancer statistics. CA Cancer J Clin 2011;61:69–90.
2. Al-Sukhni W, Borgida A, Rothenmund H, et al. Screening for Pancreatic Cancer in a High-Risk Cohort: An Eight-Year Experience. J Gastrointest Surg 2011;16:771–83.
3. Brentnall TA. Pancreatic cancer surveillance: learning as we go. Am J Gastroenterol 2011;106:955–6.
4. Canto MI, Goggins M, Hruban RH, et al. Screening for early pancreatic neoplasia in high-risk individuals: a prospective controlled study. Clin Gastroenterol Hepatol 2006;4:766–81; quiz 665.
5. Canto M, Goggins M, Yeo C, et al. Screening for pancreatic neoplasia in high-risk individuals. Clin Gastro Hepatol 2004;2:606–21.
6. Langer P, Kann PH, Fendrich V, et al. Five years of prospective screening of high-risk individuals from families with familial pancreatic cancer. Gut 2009;58:1410–18.
7. Ludwig E, Olson SH, Bayuga S, et al. Feasibility and yield of screening in relatives from familial pancreatic cancer families. Am J Gastroenterol 2011;106:946–54.
8. Poley JW, Kluijt I, Gouma DJ, et al. The yield of first-time endoscopic ultrasonography in screening individuals at a high risk of developing pancreatic cancer. Am J Gastroenterol 2009;104:2175–81.
9. Vasen HF, Wasser M, van Mil A, et al. Magnetic Resonance Imaging Surveillance Detects Early-Stage Pancreatic Cancer in Carriers of a p16-Leiden Mutation. Gastroenterology 2011;140:850–6.
10. Verna EC, Hwang C, Stevens PD, et al. Pancreatic Cancer Screening in a Prospective Cohort of High-Risk Patients: A Comprehensive Strategy of Imaging and Genetics. Clin Cancer Res 2010;16:5028–37.
11. Canto MI, Hruban RH, Fishman EK, et al. Frequent Detection of Pancreatic Lesions in Asymptomatic High-Risk Individuals. Gastroenterology 2012.
12. Atkins D, Best D, Briss PA, et al. Grading quality of evidence and strength of recommendations. BMJ 2004;328:1490–4.
13. Guyatt GH, Oxman AD, Vist GE, et al. GRADE: an emerging consensus on rating quality of evidence and strength of recommendations. BMJ 2008;336:924–6.
14. Jaeschke R, Guyatt GH, Dellinger P, et al. Use of GRADE grid to reach decisions on clinical practice guidelines when consensus is elusive. BMJ 2008;337:a744.
15. Wang W, Chen S, Brune KA, et al. PancPRO: risk assessment for individuals with a family history of pancreatic cancer. J Clin Oncol 2007;25:1417–22.
16. Klein AP, Brune KA, Petersen GM, et al. Prospective risk of pancreatic cancer in familial pancreatic cancer kindreds. Cancer Res 2004;64:2634–8.
17. Grover S, Syngal S. Hereditary Pancreatic Cancer. Gastroenterology 2010;139:1076–80.

18. Brune KA, Lau B, Palmisano E, et al. Importance of age of onset in pancreatic cancer kindreds. J Natl Cancer Inst 2010;102:119–26.
19. Hruban RH, Canto MI, Goggins M, et al. Update on familial pancreatic cancer. Adv Surg 2010;44:293–311.
20. Goggins M, Schutte M, Lu J, et al. Germline BRCA2 gene mutations in patients with apparently sporadic pancreatic carcinomas. Cancer Res 1996;56:5360–4.
21. Giardiello FM, Brensinger JD, Tersmette AC, et al. Very high risk of cancer in familial Peutz-Jeghers syndrome. Gastroenterology 2000;119:1447–53.
22. van Lier MG, Wagner A, Mathus-Vliegen EM, et al. High cancer risk in Peutz-Jeghers syndrome: a systematic review and surveillance recommendations. Am J Gastroenterol 2010;105:1258–64; author reply 65.
23. Couch FJ, Johnson MR, Rabe KG, et al. The prevalence of BRCA2 mutations in familial pancreatic cancer. Cancer Epidemiol Biomarkers Prev 2007;16:342–6.
24. Hahn SA, Greenhalf B, Ellis I, et al. BRCA2 germline mutations in familial pancreatic carcinoma. J Natl Cancer Inst 2003;95:214–21.
25. Murphy KM, Brune KA, Griffin C, et al. Evaluation of candidate genes MAP2K4, MADH4, ACVR1B and BRCA2 in familial pancreatic cancer: deleterious BRCA2 mutations in 17%. Cancer Res 2002;62:3789–93.
26. Yilmaz EN, van Heek NT, van der Spoel JI, et al. Myocardial contusion as a result of isolated sternal fractures: a fact or a myth? Eur J Emerg Med 1999;6:293–5.
27. van Asperen CJ, Brohet RM, Meijers-Heijboer EJ, et al. Cancer risks in BRCA2 families: estimates for sites other than breast and ovary. J Med Genet 2005;42:711–19.
28. Struewing JP, Abeliovich D, Peretz T, et al. The carrier frequency of the BRCA1 185delAG mutation is approximately 1 percent in Ashkenazi Jewish individuals. Nat Genet 1995;11:198–200.
29. Ferrone CR, Levine DA, Tang LH, et al. BRCA germline mutations in Jewish patients with pancreatic adenocarcinoma. J Clin Oncol 2009;27:433–8.
30. Thompson D, Easton DF. Cancer Incidence in BRCA1 mutation carriers. J Natl Cancer Inst 2002;94:1358–65.
31. Jones S, Hruban RH, Kamiyama M, et al. Exomic sequencing identifies PALB2 as a pancreatic cancer susceptibility gene. Science 2009;324:217.
32. Slater EP, Langer P, Niemczyk E, et al. PALB2 mutations in European familial pancreatic cancer families. Clin Genet 2010;78:490–4.
33. Tischkowitz MD, Sabbaghian N, Hamel N, et al. Analysis of the gene coding for the BRCA2-interacting protein PALB2 in familial and sporadic pancreatic cancer. Gastroenterology 2009;137:1183–6.
34. Schneider R, Slater EP, Sina M, et al. German national case collection for familial pancreatic cancer (FaPaCa): ten years experience. Fam Cancer 2011;10:323–30.

35. Harinck F, Kluijt I, van Mil SE, et al. Routine testing for PALB2 mutations in familial pancreatic cancer families and breast cancer families with pancreatic cancer is not indicated. Eur J Hum Genet 2012;20:577–9.
36. Goldstein AM, Chan M, Harland M, et al. Features associated with germline CDKN2A mutations: a GenoMEL study of melanoma-prone families from three continents. J Med Genet 2007;44:99–106.
37. Lynch HT, Fusaro RM, Lynch JF, et al. Pancreatic cancer and the FAMMM syndrome. Fam Cancer 2008;7:103–12.
38. de Snoo FA, Bishop DT, Bergman W, et al. Increased risk of cancer other than melanoma in CDKN2A founder mutation (p16-Leiden)-positive melanoma families. Clin Cancer Res 2008;14:7151–7.
39. Win AK, Young JP, Lindor NM, et al. Colorectal and other cancer risks for carriers and noncarriers from families with a DNA mismatch repair gene mutation: a prospective cohort study. J Clin Oncol 2012;30:958–64.
40. Kastrinos F, Mukherjee B, Tayob N, et al. Risk of pancreatic cancer in families with Lynch syndrome. JAMA 2009;302:1790–5.
41. Goggins M, Offerhaus GJ, Hilgers W, et al. Pancreatic adenocarcinomas with DNA replication errors (RER+) are associated with wild-type K-ras and characteristic histopathology. Poor differentiation, a syncytial growth pattern and pushing borders suggest RER+. Am J Pathol 1998;152:1501–7.
42. Lowenfels AB, Maisonneuve P, DiMagno EP, et al. Hereditary pancreatitis and the risk of pancreatic cancer. International Hereditary Pancreatitis Study Group. J Natl Cancer Inst 1997;89:442–6.
43. Whitcomb DC. Inflammation and Cancer V. Chronic pancreatitis and pancreatic cancer. Am J Physiol Gastrointest Liver Physiol 2004;287:G315–19.
44. Vitone LJ, Greenhalf W, Howes NR, et al. Hereditary pancreatitis and secondary screening for early pancreatic cancer. Rocz Akad Med Bialymst 2005;50:73–84.
45. Ulrich CD. Pancreatic cancer in hereditary pancreatitis: consensus guidelines for prevention, screening and treatment. Pancreatology 2001;1:416–22.
46. Brentnall TA, Bronner MP, Byrd DR, et al. Early diagnosis and treatment of pancreatic dysplasia in patients with a family history of pancreatic cancer. Ann Intern Med 1999;131:247–55.
47. Kimmey MB, Bronner MP, Byrd DR, et al. Screening and surveillance for hereditary pancreatic cancer. Gastrointest Endosc 2002;56:S82–6.
48. Topazian M, Enders F, Kimmey M, et al. Interobserver agreement for EUS findings in familial pancreatic-cancer kindreds. Gastrointest Endosc 2007;66:62–7.
49. Brugge WR, Lewandrowski K, Lee-Lewandrowski E, et al. Diagnosis of pancreatic cystic neoplasms: a report of the cooperative pancreatic cyst study. Gastroenterology 2004;126:1330–6.
50. Cizginer S, Turner B, Bilge AR, et al. Cyst fluid carcinoembryonic antigen is an accurate diagnostic marker of pancreatic mucinous cysts. Pancreas 2011;40:1024–8.

51. Tanaka M, Chari S, Adsay V, et al. International consensus guidelines for management of intraductal papillary mucinous neoplasms and mucinous cystic neoplasms of the pancreas. Pancreatology 2006;6:17–32.
52. Del Chiaro M, Zerbi A, Capurso G, et al. Familial pancreatic cancer in Italy. Risk assessment, screening programs and clinical approach: a position paper from the Italian Registry. Dig Liver Dis 2010;42:597–605.
53. Jacobs EJ, Chanock SJ, Fuchs CS, et al. Family history of cancer and risk of pancreatic cancer: a pooled analysis from the Pancreatic Cancer Cohort Consortium (PanScan). Int J Cancer 2010;127:1421–8.
54. Finks JF, Osborne NH, Birkmeyer JD. Trends in hospital volume and operative mortality for high-risk surgery. N Engl J Med 2011;364:2128–37.
55. Lemmens VE, Bosscha K, van der Schelling G, et al. Improving outcome for patients with pancreatic cancer through centralization. Br J Surg 2011;98:1455–62.
56. Tanaka M, Fernandez-Del Castillo C, Adsay V, et al. International consensus guidelines 2012 for the management of IPMN and MCN of the pancreas. Pancreatology 2012;12:183–97.
57. Matthaei H, Hong SM, Mayo SC, et al. Presence of pancreatic intraepithelial neoplasia in the pancreatic transection margin does not influence outcome in patients with R0 resected pancreatic cancer. Ann Surg Oncol 2011;18:3493–9.
58. Zauber AG, Winawer SJ, O'Brien MJ, et al. Colonoscopic polypectomy and long-term prevention of colorectal-cancer deaths. N Engl J Med 2012;366:687–96.
59. Winter JM, Cameron JL, Campbell KA, et al. 1423 pancreaticoduodenectomies for pancreatic cancer: A single-institution experience. J Gastrointest Surg 2006;10:1199–210; discussion 210–1.
60. Schnelldorfer T, Ware AL, Sarr MG, et al. Long-term survival after pancreatoduodenectomy for pancreatic adenocarcinoma: is cure possible? Ann Surg 2008;247:456–62.
61. Fatima J, Schnelldorfer T, Barton J, et al. Pancreatoduodenectomy for ductal adenocarcinoma: implications of positive margin on survival. Arch Surg 2010;145:167–72.
62. Laupacis A, Sackett DL, Roberts RS. An assessment of clinically useful measures of the consequences of treatment. N Engl J Med 1988;318:1728–33.
63. Sellner F, Thalhammer S, Stattner S, et al. TNM stage and grade in predicting the prognosis of operated, non-functioning neuroendocrine carcinoma of the pancreas–a single-institution experience. J Surg Oncol 2011;104:17–21.
64. Brat DJ, Lillemoe KD, Yeo CJ, et al. Progression of pancreatic intraductal neoplasias to infiltrating adenocarcinoma of the pancreas. Am J Surg Pathol 1998;22:163–9.
65. Hruban RH, Adsay NV, Albores-Saavedra J, et al. Pancreatic intraepithelial neoplasia: a new nomenclature and classification system for pancreatic duct lesions. Am J Surg Pathol 2001;25:579–86.

66. 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;25:942–8.
67. Meckler KA, Brentnall TA, Haggitt RC, et al. Familial fibrocystic pancreatic atrophy with endocrine cell hyperplasia and pancreatic carcinoma. Am J Surg Pathol 2001;25:1047–53.
68. Shi C, Klein AP, Goggins M, et al. Increased Prevalence of Precursor Lesions in Familial Pancreatic Cancer Patients. Clin Cancer Res 2009;15:7737–43.
69. Terhune PG, Phifer DM, Tosteson TD, et al. K-ras mutation in focal proliferative lesions of human pancreas. Cancer Epidemiol Biomarkers Prev 1998;7: 515–21.
70. Yachida S, Jones S, Bozic I, et al. Distant metastasis occurs late during the genetic evolution of pancreatic cancer. Nature 2010;467:1114–17.
71. Wu J, Matthaei H, Maitra A, et al. Recurrent GNAS mutations define an unexpected pathway for pancreatic cyst development. Sci Transl Med 2011;3:92ra66.
72. Matthaei H, Schulick RD, Hruban RH, et al. Cystic precursors to invasive pancreatic cancer. Nat Rev Gastroenterol Hepatol 2011;8:141–50.
73. Sato N, Rosty C, Jansen M, et al. STK11/LKB1 Peutz-Jeghers gene inactivation in intraductal papillary-mucinous neoplasms of the pancreas. Am J Pathol 2001;159:2017–22.
74. Sohn TA, Yeo CJ, Cameron JL, et al. Intraductal papillary mucinous neoplasms of the pancreas: an updated experience. Ann Surg 2004;239:788–97; discussion 97–9.
75. Sawai Y, Yamao K, Bhatia V, et al. Development of pancreatic cancers during long-term follow-up of side-branch intraductal papillary mucinous neoplasms. Endoscopy 2010;42:1077–84.
76. Uehara H, Nakaizumi A, Ishikawa O, et al. Development of ductal carcinoma of the pancreas during follow-up of branch duct intraductal papillary mucinous neoplasm of the pancreas. Gut 2008;57:1561–5.

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Submitted

CHAPTER 5
EXPLORING THE EFFECTS OF FACTORS ASSOCIATED WITH THE OUTCOME OF PANCREATIC CANCER SCREENING IN HIGH-RISK INDIVIDUALS

ABSTRACT

Background & Aims | We currently lack scientific evidence to recommend screening for pancreatic ductal adenocarcinoma (PDAC) in high-risk individuals. We explored the effects of such screening under a range of plausible assumptions and analyzed which factors have the highest impact.

Methods | Effects of PDAC screening were estimated using the MISCAN-model. We modelled two different disease pathways: Model A with only progressive disease and model B with progressive and slow developing disease. We varied screening test-characteristics,,follow-up strategies after a positive screening, procedure related mortality and level of risk for developing PDAC.

Results | In case of screening every 5 year, in Model B the mortality reduction (MR) was 17% per 100,000 Life years (LYs) and in Model A 35%. Annual screening resulted in a MR of 41% in Model B and 58% in Model A. For 5 yearly screening, the number needed to screen (NNS) to prevent one cancer death was 166 in Model A and 326 in model B. The number needed to treat (NNT) were 3.3 and 6.0 in Models A and B, respectively. NNT was lowest in case all screen positives with pre-invasive stage 3 or cancer are treated (4.8 in model B, MR 18%). If only persons are treated who are already in an invasive stage of disease, the NNT was 8.4 (MR 6%). Results were also sensitive for PDAC risk, but less sensitive for test characteristics.

Conclusions | Modeling shows that there is potential for pancreatic screening to be effective in high-risk individuals. Follow-up strategy of screen positives and duration of the preclinical stage have the highest impact on the outcome of PDAC screening, as is inclusion of patient populations that are exposed to a certain risk to develop PDAC.

INTRODUCTION

Pancreatic ductal adenocarcinoma (PDAC) has one of the poorest survivals of all human cancers (1). Approximately 5-10% of all PDAC cases represent patients with an increased familial and/or inherited risk for this fatal disease. Within this population of high-risk individuals, the lifetime risk of developing PDAC may exceed 40% (2). High-risk individuals include carriers of PDAC prone gene mutations (e.a. *BRCA2*, *CDKN2A* and *LKB1*) and first-degree relatives of familial PDAC-patients. Familial PDAC patients are patients with PDAC and a family history affected by at least (1) one first degree relative with PDAC, (2) one second degree relative <50 years at time of diagnosis or (3) two relatives with PDAC.

Cancer screening aims to detect precancerous lesions or cancer prior to the point of clinical presentation in order to decrease morbidity and mortality of cancer (3). There are several disease-related aspects of PDAC that indicate that screening for this cancer type could be worthwhile (2, 4), especially when screening is selectively applied to individuals with an increased risk of developing PDAC. We, however, currently lack undisputed evidence that screening for PDAC in high-risk populations is effective as scientific data with respect to some crucial parameters (eg. natural history and dwell time, screening test characteristics) are scare. Based on evidence of increased risk, rather than proven efficacy of screening, screening for PDAC is recommended for high-risk individuals (ideally within a research setting) (4). To ensure that the benefits of screening practices outweigh the harms, more evidence on the effect of PDAC screening is needed. Exploration of effects of screening under plausible assumptions would give input on what to expect when more evidence would be sought.

It has been shown that decision analytic models are extremely useful for this exploration since they provide a structure for ordering and synthesizing information from a wide range of sources (5). Furthermore, by building and fine-tuning these decision analytic models it becomes readily apparent which knowledge gaps exist. Sensitivity analyses can be used to quantify the impact of the lack of knowledge of individual factors (6). These analyses help us to identify which factors have the most impact on the overall effect of PDAC screening. By using the microsimulation model MISCAN ('Microsimulation Screening Analysis') we aimed to explore the uncertainties concerning early detection of PDAC in high-risk individuals, to analyze their impact on the effect of screening and consequently to highlight the areas to which further research should be directed.

METHODS

We explored the effects of PDAC screening by using the MISCAN-model (7), which has been made and applied for cancer of the cervix, breast, colon, and prostate (8-14). The MISCAN-model generates a large study population with fictitious individual life histories, in which persons can, at a certain rate, develop a pre-invasive lesion and/or PDAC, and

some will die from this disease. This simulation results in an age-specific and time-specific output of disease incidence and mortality. This fictitious population then undergoes PDAC screening, which will change some of the life histories. These changes constitute the effects of PDAC screening and are represented by the numbers of events and stages induced or prevented by screening. The stochastic model underlying the simulation is specified by the input parameters. These parameters relate to demographic characteristics (e.g., the life table), epidemiology and natural history of the disease (e.g., duration of preclinical cancer), and the characteristics of screening (e.g., the sensitivity of the screening test(s)).

Model Specifications and Assumptions

The majority of assumptions are based on the recommendations as stated in the consensus paper of the international Cancer of the Pancreas Screening (CAPS)-consortium (4)
The model that has a decision analytic structure containing source data based on the best available data from the literature is referred to as the base case model. Through sensitivity analyses of differential factors the effects of variations of base case assumptions were studied.

Base case assumptions

Demography, Epidemiology, and Natural History. The models we present are cohort models. We simulated a Dutch population based on demographic data; mortality from other causes was estimated using the observed age-specific mortality in the Netherlands in 2005 [Source: Statistics Netherlands]. Because the population at high risk for pancreatic cancer is also at higher risk for other (lethal) diseases (4), we assumed that the simulated population has a decreased life expectancy compared to the general population (70 years instead of 80 years). Disease was subdivided into eight sequential stages: three pre-invasive stages (i.e. pancreatic intraductal neoplasia (PanIN)I/ intraductal papillary mucinous neoplasm (IPMN) low-grade, PanIN2/IPMN moderate grade, and PanIN3/IPMN high-grade), and five invasive stages (TNM stages IA, IB, IIA, IIB and III/IV). Although future studies might show a difference in natural behavior of both PanIN lesions and IPMNs, for this explorative study we have modelled the duration of each stage (dwelling time) and probability to transit to a subsequent stage (transition) as being one entity. The parameter values are than a weighted average for both. We have done so since we currently lack evidence that proves that and how much they differ. Pre-invasive stages can only be diagnosed by screening and not clinically because they are asymptomatic, whereas invasive stages can be diagnosed by screening as well as clinically (Appendix Figure 1).
Because much is still unknown about the natural history of PDAC, more in particular the relation of the pancreatic neoplastic lesions detected by screening to clinical PDAC, we modelled two contrasting disease progression patterns from pre-invasive disease onset to clinical cancer. In model A, we assumed all durations were governed by an exponential probability distribution and that durations in each disease stage were assumed to be

100% associated with each other. The average total duration of pre-clinical disease is 14.3 years. The latter was based on the recent results of Yachima et al (15) showing that it takes at least a decade between the occurrence of the initiating mutation and the birth of the parental, non-metastatic founder cell and at least five more years for the acquisition of metastatic ability. In model B, we assumed much more variance in duration: disease can be either progressive as in model A (exponential distributed) but with an average duration of only 5 years, or indolent (progression is that slow that the disease will never develop in lethal or even clinical cancer, no matter how old the person gets; henceforth referred to as slow developing disease). In case of slow developing disease, pre-invasive stages will at most progress to preclinical cancer stage 1 (Appendix Figure 1).
For both models (A and B), we used the observed age specific mortality rate of PDAC and age distribution [Source: CBS, death registry] in the Netherlands in 2008 as calibration target for the incidence and age distribution of pre-invasive neoplasia that will eventually become cancer. We used mortality instead of incidence, since at older ages the cancer registry data is not complete. Moreover, mortality rates almost equal incidence rates, since most incidence cases are fatal within short time. To simulate a high-risk population, we multiplied the incidence by a factor 10 (4) Consequently, the lifetime risk for developing PDAC is 7.5% in the population simulated. For model B, we moreover calibrated the incidence of slow developing disease to reproduce the same pre-invasive detection rates, pre-invasive stage-distribution and total preclinical invasive detection rates similar to model A (Figure 1 shows for model A and B the prevalence of each stage of disease by age). For both models, the stage-specific survival used in the model for clinical cases (i.e., cases diagnosed based on symptoms as opposed to screen detection) was based on observed survival in the Netherlands. Detailed assumptions about the natural history of pancreatic cancer and its precursors are given in Table 1.

Base Case Assumptions Regarding Screening and Early Treatment. High-risk individuals were screened annually from age 50 to 75. Analyses were performed for high-risk individuals that adhere to all screening rounds. As endoscopic ultrasonography (EUS) and magnetic resonance imaging (MRI) are currently considered the most accurate tests for pancreatic imaging within a screening setting (4), the assumptions with respect to the test characteristics were based on the results of these two tests. The sensitivity of the test for different disease stages was assumed to be 60% for pre-invasive stage 1 and 2, 75% for pre-invasive stage 3, 90% for preclinical invasive cancer stage I, 93% for preclinical invasive cancer stage II, and 99% for preclinical invasive cancer stage III/IV (16-19). Would the pre-invasive stage only include IPMNs (cystic lesions), one would expect a higher sensitivity. However, since in this model pre-invasive lesions include both PanINs as well as IPMN the estimated (average) sensitivity is lower based on the fact that it is more difficult to correctly identify and stage PanIN with the current available techniques. Specificity of the test was assumed to be 90%. In case of a positive test result, persons were referred to

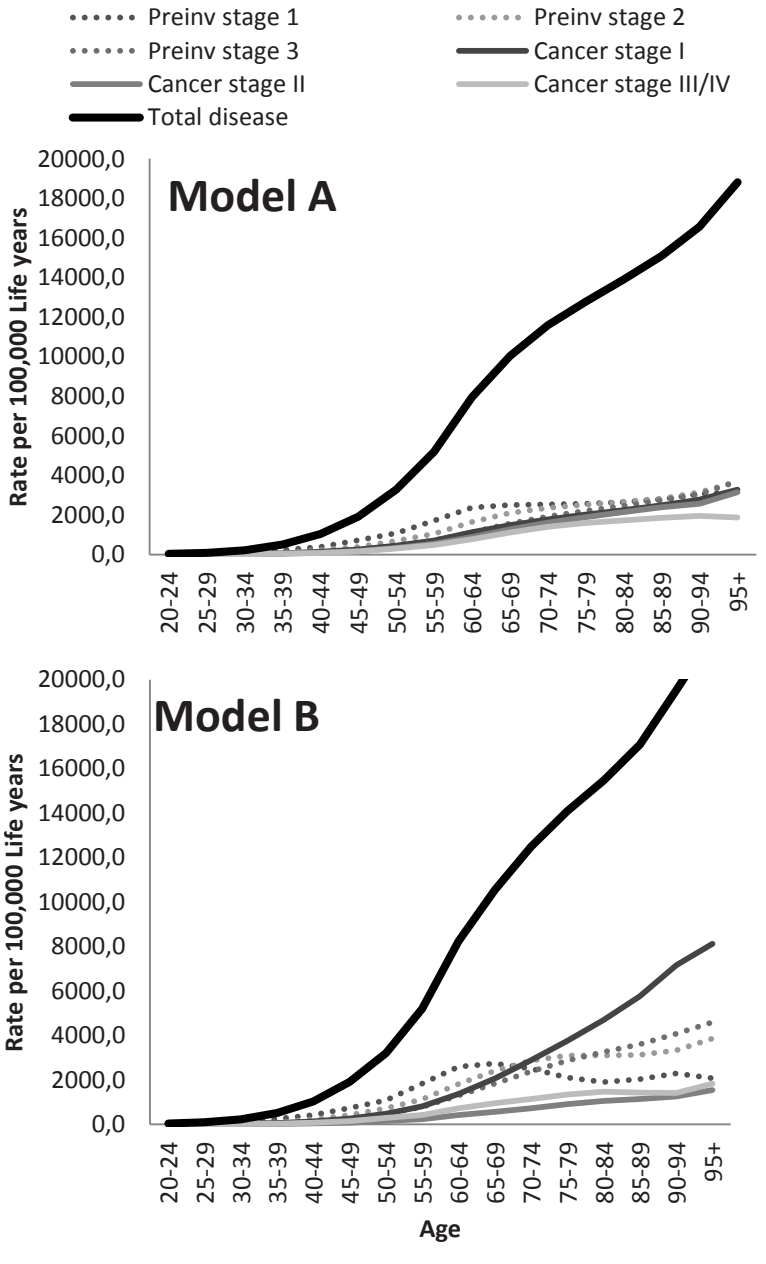


Figure 1. Prevalence by age of the different stages of disease (pre-invasive stages and cancer (preclinical and clinical cases)) in the two models for high-risk individuals in the base case MISCAN-pancreas model, without screening, for men and women together.

Stage	Value	
	Model A	**Model B**
	Mean duration of progressive stages	
Pre-invasive stage 1	3.33 year	1.11 year
Pre-invasive stage 2	3.33 year	1.11 year
Pre-invasive stage 3	3.33 year	1.11 year
Preclinical 1a	1 year	0.33 year
Preclinical 1b	1 year	0.33 year
Preclinical 2a	1 year	0.33 year
Preclinical 2b	1 year	0.33 year
Preclinical 3/4	1 year	0.33 year
Estimated mean total pre-clinical	14.3 year	4.8 years
	Mean duration of slow developing disease	
Pre-invasive stage 1	n.a.	7.08 year*
Pre-invasive stage 2	n.a.	11.78 year*
Pre-invasive stage 3	n.a.	24.15 year*
Preclinical 1a	n.a.	untill death other causes*
	Probability of being clinically diagnosed, before moving to the next stage[28]	
Cancer stage 1a	4.4%	
Cancer stage 1b	5.6%	
Cancer stage 2a	11.2%	
Cancer stage 2b	14.7%	
Cancer stage 3/4	100%	
	*5-year relative survival***	
Clinical 1a	31.4%	
Clinical 1b	27.2%	
Clinical 2a	15.7%	
Clinical 2b	7.7%	
Clinical 3/4	0%	

Table 1. Model assumptions about the natural history of PDAC and its precursors for the two base case models.
** Optimized parameters*
*** A linear distribution of the PDAC mortality was assumed over 5 years. If someone has survived the first 5 years after diagnosis, we assumed lifelong relative PDAC survival. Detection (and the associated management of pre-invasive lesions) was assumed to lead to a 100% cure rate. For resection, we assumed a 3% mortality risk. For screen-detected invasive cancers, stage-specific survival in the model was based on observed survival of clinically detected cancer in the Netherlands.*

surveillance or cases were directly referred to surgery for resection. Assumptions (based on expert opinions) with respect to the probability that a patient undergoes resection (rightfully or wrongfully as a result of misclassification) after a positive test were as following: 5% in case the true state was pre-invasive stages 1, 25% in case the true state was pre-invasive stages 2, and 40% in case the true state was pre-invasive stage 3. In case of preclinical cancer stages 1, 2 and 3/4 these probabilities were 90%, 93% and 1%, respectively. These probabilities, together with the sensitivity determine the proportion of patients that undergoes resection (Figure 2). For example, 81% of the patients with a preclinical stage 1 cancer will undergo resection, because 90% of the cases will have a positive screening test and 90% of these positive cases will be treated (90%*90%=81%). Some persons directly returned to the regular screening, based on the interpretation of the positive test result (Figure 2). The sensitivity of the surveillance test was assumed to be equal to the screening test, the specificity was assumed to be 100%. We assumed that a proportion (depended on true stage, see Figure 2) of the persons with a positive surveillance test will undergo resection and that surveillance tests would be performed until resection or end of life. Detection and the associated management of pre-invasive lesions (i.e. partial pancreatic resection) was assumed to lead to a 100% cure rate (i.e. no cancer development). Patients are, however, still at risk to develop new lesions. For resection, we assumed a 3% mortality risk associated with treatment (20, 21) For screen-detected invasive cancers, stage-specific survival in the model was based on observed survival of clinically detected cancer in the Netherlands, so we did not assume any of the potential benefit from within stage shift at screen-detection.

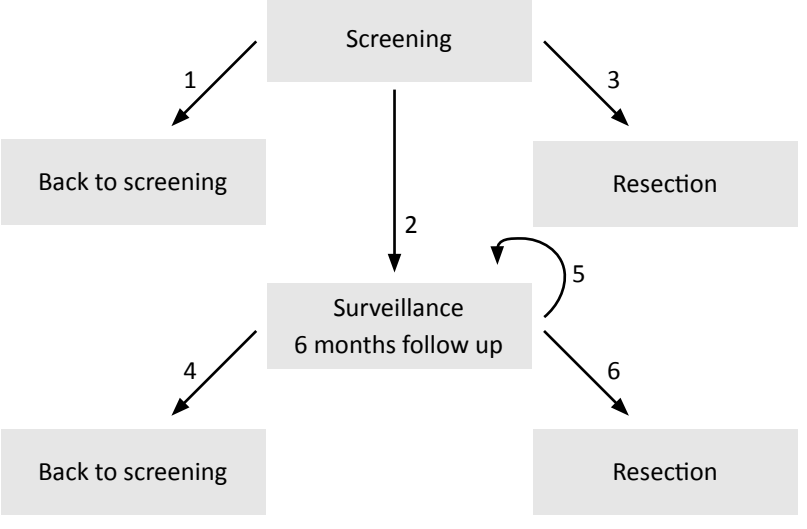


Figure 2. Screening scenario evaluated in the MISCAN-pancreas model. Transition 1 ('back to screening') is the effect of (false-) negative test result. Values in table are for the base case situation.

	Assumptions				Transitions in Figure 2					
True state (i.e. pathologic diagnosis)	A Probability positive screening or surveillance test	B Probability resection after positive screening test	C Probability surveillance after positive screening test	D Probability resection after positive surveillance test	1. Back to screening* (100%-2.- 3.)	2. Surveillance, 6 months follow up (A*C)	3. Resection (A*B)	4. Back to screening*	5. Surveillance, 6 months follow up (100%-6.- 4.)	6. Resection (A*D)
Normal	10%	0%	100%	0%	90%	10%	0%	100%	0%	0%
Preinvasive stage 1	60%	5%	20%	33%	85%	12%	3%	0%	80%	20%
Preinvasive stage 2	60%	25%	45%	60%	58%	27%	15%	0%	64%	36%
Preinvasive stage 3	75%	40%	58%	55%	26%	44%	30%	0%	59%	41%
Preclinical 1	90%	90%	10%	100%	10%	9%	81%	0%	10%	90%
Preclinical 2	93%	93%	7%	100%	7%	7%	86%	0%	7%	93%
Preclinical 3/4	99%	100%**	0%	n.a.	1%	0%	99%**	n.a	n.a	n.a

Figure 2. Screening scenario evaluated in the MISCAN-pancreas model. Transition 1 ('back to screening') is the effect of (false-) negative test result. Values in table are for the base case situation.

** 0.5, 1, 2 or 5 year interval*

*** In case of cancer stage 3/4, patients only receive palliative care (no resection)*

Base case and Sensitivity Analyses

We simulated 1,000,000 individuals for each screening strategy. The simulated results account for the lifelong effects. The effects are presented as numbers of screening tests, numbers of surveillance tests, numbers of resections, incidence, mortality, numbers of deaths prevented, interval cancer rate (in the first 5 years and in the total period after screening), and life years gained (LYsG). For each screening scenario, we calculated the number needed to screen (NNS), number needed to surveil (NNSurv) and number needed to treat (NNT) to prevent one cancer death. The NNS is defined as the number of screening tests and the NNSurv is the number of surveillance tests (after a positive screening test) that need to be performed to prevent one death.

Through sensitivity analyses, the following variations in assumptions were simulated to study the impact on uncertainties: (1) sensitivity of the screening test, (2) specificity of the screening test, (3) level of risk for developing PDAC. In addition, we varied follow up strategy (i.e. cut off for resection).

Sensitivity To examine the impact of the sensitivity of the screening test, for each stage, we decreased the probability that the test will be positive with 5% and 10%. Conversely, we also increased this probability with 5% and 10%. The probability that a patient undergoes resection given a positive test was equal to the base case situation (i.e 5%, 25%, and 40% in case of pre-invasive stages and 90%, 93% and 1% in case of cancers). As a result, the probabilities from Figure 2 change, as showed in Table 1 of the Appendix.

Specificity We varied the probability that a person without disease will have a positive screening test. We increased the probability from 10% (base case) to 15% (specificity 85%), and decreased it to 5% and 0% (specificity 95% and 100%). After a false positive screen result (i.e. a positive test result in a person without (preinvasive) PDAC), persons undergo one surveillance test after 6 months. If this test is negative (normal), persons return to the regular screening program (Figure 2).

Risk The risk of developing PDAC was halved and doubled (base case lifetime risk for developing PDAC 7.5%).

Follow up strategy In the base case situation we assumed that a proportion of patients undergoes resection after a positive screening test (Figure 2). To examine the consequences of referring an increased or decreased number of people for surgery we evaluated the situation that (1) all persons with a positive screening test undergo resection, (2) that all persons with a positive screening test that are in true disease stage 'pre-invasive stage 3' or higher undergo resection, or (3) that that all persons with a positive screening test that are in true disease stage cancer undergo resection. In case of a positive screen result and no resection, persons will undergo one surveillance test after 6 months. If this test is negative (in case of analysis 2: <preinvasive stage 3, or in case of analysis 3: <cancer), persons are send back to the regular screening program. As a result, the probabilities from Figure 2 change, as shown in Table 1 of the Appendix.

Treatment mortality In the base case situation we assumed that the treatment mortality risk was equal to 3%, we increased this risk to 5%.

RESULTS

Table 2 shows the effects of screening under the base case assumptions, for screening annually or every 5 year from age 50 to age 75 in a high-risk population, compared to no screening, for model A and model B.

In case of screening every 5 year, we found that with slow developing and progressive disease (Model B) the mortality rate decreases from 108 to 89 per 100,000 Life years (LYs) (-18%). Per 10,000 persons simulated this corresponds with 31 cancer cases prevented and 129 cancer death cases prevented. Without slow developing disease (Model A) the mortality rate decreases from 108 to 71 (-34%) per 100,000 LYs which corresponds with 198 cancer cases and 256 cancer deaths prevented.

In case of annual screening the mortality rate decreases from 108 to 64 per 100,000 Life years (LYs) (-41%) in Model B (259 cancer cases and 309 cancer death prevented) and from 108 to 46 per 100,000 Life years (LYs) (-58%) in Model A (455 cancer cases and 432 cancer death prevented).

In case of annual screening, relatively more resections are done in patients with a less invasive stage of disease, compared to screening with an interval of 5 years. Significantly more interval cancers are found in case of rapid growth of progressive lesions (Model B) compared to Model A. In the base case annual screening situation, to prevent one PDAC death 431 persons need to be screened (NNS) without indolent disease, 2.9 patients need to be treated (NNT) (i.e. resection) and 47 surveillance tests (NNSurv) need to be performed. In case of a 5 year interval, less persons need to be screened (n=166) and less surveillance tests need to be performed (n=19), but more patients need to be treated (n=3.3) to prevent one PDAC death. In case of slow developing and progressive disease, results are less positive: with annual screening NNS is 600, NNSurv is 65 and NNT is 4.1. With 5 yearly screening NNS is 326, NNSurv is 38 and NNT is 6.0.

Sensitivity analyses

The influence of differences in epidemiologic and screening characteristics were investigated in sensitivity analyses (Figure 3, Appendix Tables 2 and 3).

Sensitivity In both models the effect of the sensitivity (within the +10% range investigated) on the screening result is minimal. In case of only progressive disease screening every 5 year, we found that a 10% increased probability to detect (pre-invasive) PDAC decreased the incidence and mortality rate by <10%, and lowered the number of tests to prevent one PDAC death by 6.6%. In case of a decreased sensitivity, incidence and mortality rates increased slightly by <10%, and the number of tests to prevent one PDAC death increased. In case of annual screening the effect is smaller.

Specificity The specificity has a significant effect on the number of surveillance tests needed to be performed to prevent one PDAC death. If there is only progressive disease, in case of a screening interval of 5 year and 85% specificity 27 surveillance tests are needed,

	BASE CASE MODEL A			BASE CASE MODEL B		
	NO SCREENING	1 year interval	5 year interval	NO SCREENING	1 year interval	5 year interval
Number screening tests	0	186,504	42,415	0	185,594	42,175
Number of resections	0	1,232	845	0	1,279	778
Number of surveillance tests	0	20,182	4,945	0	20,025	4,941
Number of Preinv stage 1 resections	0	292 (23.7%)	82 (9.7%)	0	286 (22.4%)	88 (11.3%)
Number of Preinv stage 2 resections	0	414 (33.6%)	201 (23.8%)	0	391 (30.6%)	205 (26.3%)
Number of Preinv stage 3 resections	0	233 (18.9%)	207 (24.5%)	0	254 (19.9%)	204 (26.2%)
Number of SD cancer stage 1 resections	0	252 (20.4%)	256 (30.3%)	0	276 (21.6%)	203 (26.1%)
Number of SD cancer stage 2 resections	0	40 (3.2%)	97 (11.5%)	0	66 (5.2%)	74 (9.5%)
Number of SD cancer stage 3/4 of resections	0	2 (0.2%)	3 (0.4%)	0	5 (0.4%)	4 (0.5%)
Cancer cases	921	466	723	918	659	887
Cancer deaths	751	319	495	753	444	624
Incidence (per 100,000 LYs)	132	67	104	132	94	127
Mortality (per 100,000 LYs)	108	46	71	108	64	89
LYs gained	0	4,015	2,291	0	3,052	1,233
Interval cancers, <5 yr (per 100,000 LYs)	0	25	123	0	79	237
Interval cancers, total (per 100,000 LYs)	0	40	136	0	106	257
NNS	n.a.	431,4	165.7	n.a.	600.4	325.8
NNSurv	n.a.	46.7	19.3	n.a.	64.8	38.2
NNT	n.a.	2.9	3.3	n.a.	4.1	6.0

Table 2. Base case results for screening at ages 50 to 75, with a 1 and 5 year interval, per 10,000 simulated persons (i.e. 9,250 and 9,251 screened individuals) for models A and B. The interval cancer rate is presented as the number of cancer cases (per 100,000 LYs) in the first 5 years after a negative screening test and in the total period after a negative screening test (including after age 75) (SD cancer cases after a negative screening test are not included).

Preinv = preinvasive, SD = screen detected, LYs = Life years, NNS = Number of screening tests need to perform to prevent one cancer death, NNT = Number needed to treat to prevent one cancer death, NNSurv = Number of surveillance test need to performed to prevent one cancer death.

compared to 4 in case of 100% specificity. With annual screening the effect is even larger.
Risk The risk of developing PDAC in the screened population is one of the most important parameters that determine the number of tests needed to prevent one PDAC death. In case of a screening interval of 5 years and a halved PDAC risk, the NNS is 323 compared to 166 in the base case situation in model A. If the risk is doubled, the NNS is only 87. For model B these figures are 639 compared to 326, and 167 if the risk is doubled.
Follow up strategy The management strategy after a positive screening test, that is the cut off for resection, is the most important parameter that determines the effect of the screening. As expected, the mortality reduction is largest if all persons with a positive screening test are resected. However, the NNT to prevent one PDAC death is lowest in case the screen positives with preinvasive stage 3 or cancer are resected. For instance, in case of annual screening with only progressive disease (Model A) the mortality reduction is 66% when all persons with a positive screening test are resected and 64% when persons with a positive screening test that are in true disease stage 'pre-invasive stage 3' or higher undergo resection. However, the NNT is 2.9 in case all persons with a positive test are resected and drops to 2.2 when persons with a positive screening test that are in true disease stage 'pre-invasive stage 3' or higher undergo resection. If only persons are resected who are already in an invasive stage of disease, the effect of screening is significantly lower.
Treatment mortality Increasing the treatment mortality from 3% to 5% resulted in a 4%-8% increase in cancer deaths per 10 000 persons in both models. In case of screening every 5 years, for Model A the number of cancer deaths prevented dropped from 256 to 247 (-3.5%) and for Model B from 130 to 120 (-7.7%). In case of annual screening, for Model A the number of cancer deaths prevented dropped from 433 to 414 (-4.4%) and for Model B from 309 to 291 (-5.8%).

DISCUSSION

Based on the results of our explorative study, we conclude that the natural history (differently modeled in the two models) is one of the key factors that determine the potential success of pancreatic screening in high-risk individuals. Other factors that have major influence on the effect of screening are the follow up strategy of screen positives and the level of risk for developing PDAC. The sensitivity (+/-10%) of the screening test has a much smaller effect on mortality reduction. The specificity of the test is particularly important for the number of surveillance tests (associated with burden) needed to prevent one PDAC death.
Our results clearly show that screening is most efficient if patients are treated before the disease becomes invasive. In this regard however it must be acknowledged that for this explorative analysis, the only negative aspect of treatment included is a 3% or 5% surgical mortality risk. Morbidity and loss in quality of life as a result of the resection were not

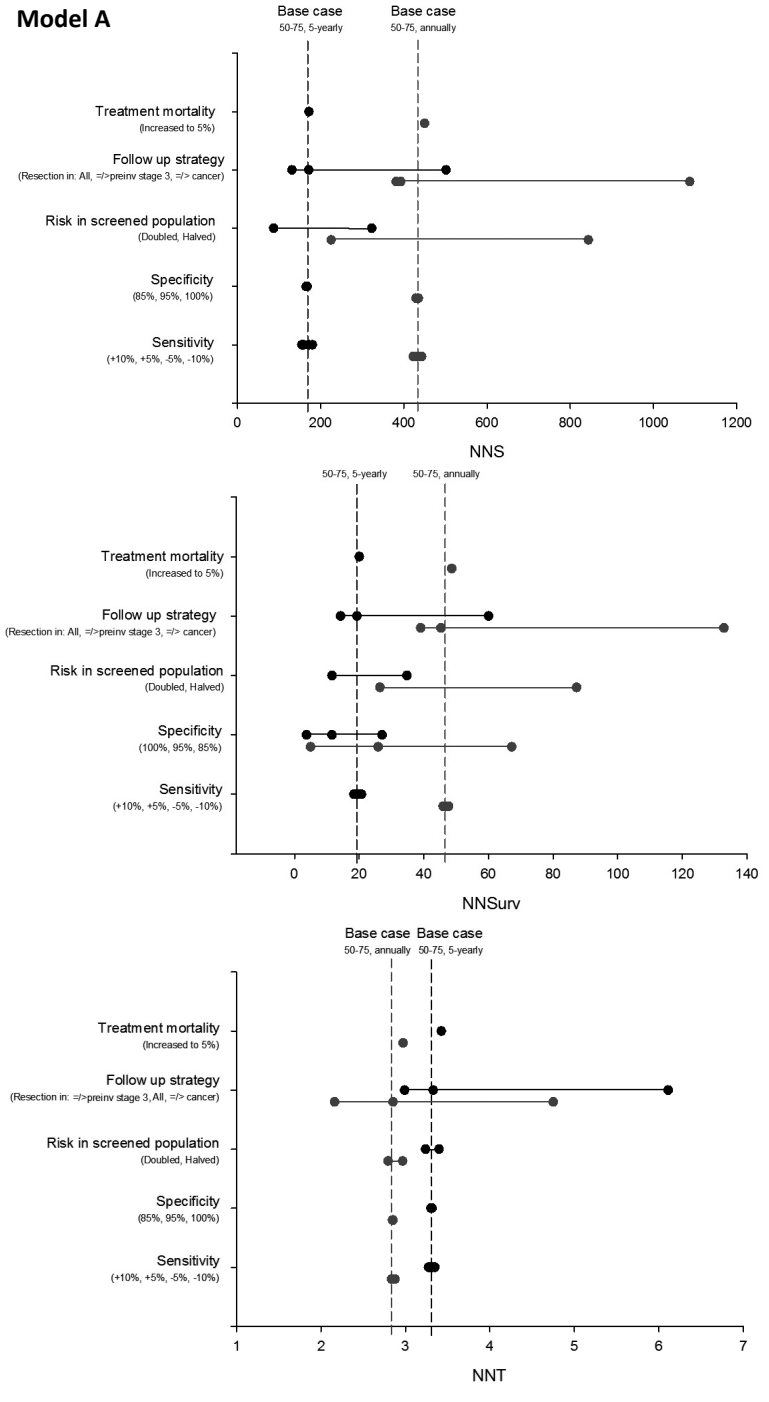


Figure 3. Results of the sensitivity analyses for screening at ages 50 to 75, with an 1 and 5 year interval for model A and model B. The order (from left to right) of the variations in the factors (between brackets) corresponds to the order (from left to right) in the figure. The dashed lines represent the results of the base case analyses. NNS = Number of screening tests need to perform to prevent one cancer death, NNT = Number needed to treat to prevent one cancer death, NNSurv = Number of surveillance test need to performed to prevent one cancer death.

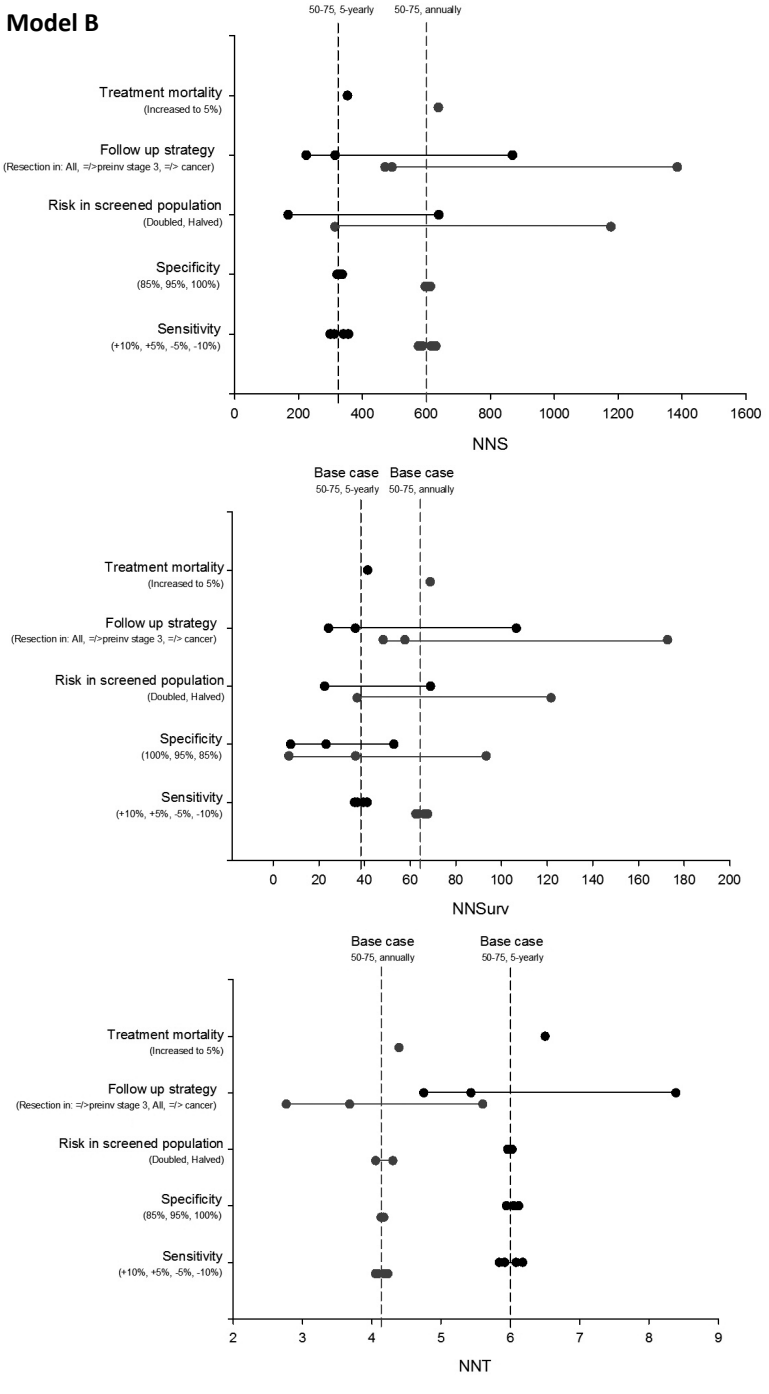


Figure 3. Results of the sensitivity analyses for screening at ages 50 to 75, with an 1 and 5 year interval for model A and model B. The order (from left to right) of the variations in the factors (between brackets) corresponds to the order (from left to right) in the figure. The dashed lines represent the results of the base case analyses. NNS = Number of screening tests need to perform to prevent one cancer death, NNT = Number needed to treat to prevent one cancer death, NNSurv = Number of surveillance test need to performed to prevent one cancer death.

considered. Pancreatic surgery carries a considerably probability of morbidity (40-60%) (20, 21) the most frequently seen complications being delayed gastric emptying, wound infections and postoperative pancreatic fistulae. Development of diabetes mellitus and/or exocrine insufficiency as a result of pancreatic resection may also influence quality of life. Integrating these harms in the model will likely result in a less favorable effect. Our results are driven by the assumption that treatment in a pre-invasive stage of disease leads to 100% cure, in other words, patients resected in a pre-invasive stage will not die of that lesion. They are, however, still at risk to develop new lesions in their remnant pancreas after partial resection. The survival of cancer after treatment is much lower (Table 1).

A remarkable finding of our study was that the influence of the sensitivity of the screening test on the NNS, NNT and NNSurv seems negligible. This is partly caused by the fact that the probability that someone is referred to resection after a positive test was not varied (Table 1 appendix). Consequently, the increase (or decrease) in resections (and life years gained) is lower than the increase (or decrease) in screen positives. Still, in case of a screening interval of 5 year and a 10% lower sensitivity, 16% more screening tests need to be performed to prevent one PDAC death compared to the 10% higher sensitivity scenario (Model A). For the same comparison, the mortality rate was 9% lower. So although there is an effect of the sensitivity, it is much less pronounced compared to the effect of the PDAC risk, the duration of the preclinical stage and the follow up strategy.

Our modeling shows that there certainly is potential for pancreatic screening in high-risk individuals to be worthwhile. This will be highly dependent on the risk level of the population at risk, the probability that a lesion will progress to cancer and its growth rate. The NNS estimated for breast-, cervical-, colorectal, or prostate cancer screening range from approximately 1000 to 2000 (22, 23). Our estimation that approximately 500 (high-risk) persons need to be screened to prevent one PDAC death is significantly lower than the NNS for the already established screening programmes. Although many of the factors that influence the NNS in our model are based on assumptions, none of the sensitivity analyses resulted in a NNS of >1500. However, the screening instruments (i.e. EUS, MRI) to detect (pre-invasive) PDAC are more invasive tests compared to the tests used in the known screening programmes. It is also more expensive (approximately €700 for EUS (27)) compared to <€60 for other screening programmes (9, 11, 13, 24). Consequently, in an analysis of costs and quality of life, screening for PDAC might probably not be more efficient than screening for breast-, cervical-, colorectal- or prostate cancer.

Two PDAC screening studies estimating the cost-effectiveness of PDAC screening have been published (25, 26). Since these studies differed from our study with regard to some major starting points, they are difficult to compare. Rulyak *et al.* (26) found that endoscopic screening in high-risk individuals increases patient life expectancy and was cost-effective. This study assumed only one-time screening of 50 year old members of familial PDAC kindreds. After a positive EUS, endoscopic retrograde cholangiopancreatography (ERCP) was performed. If ERCP was positive, a total pancreatectomy was done. Compared to

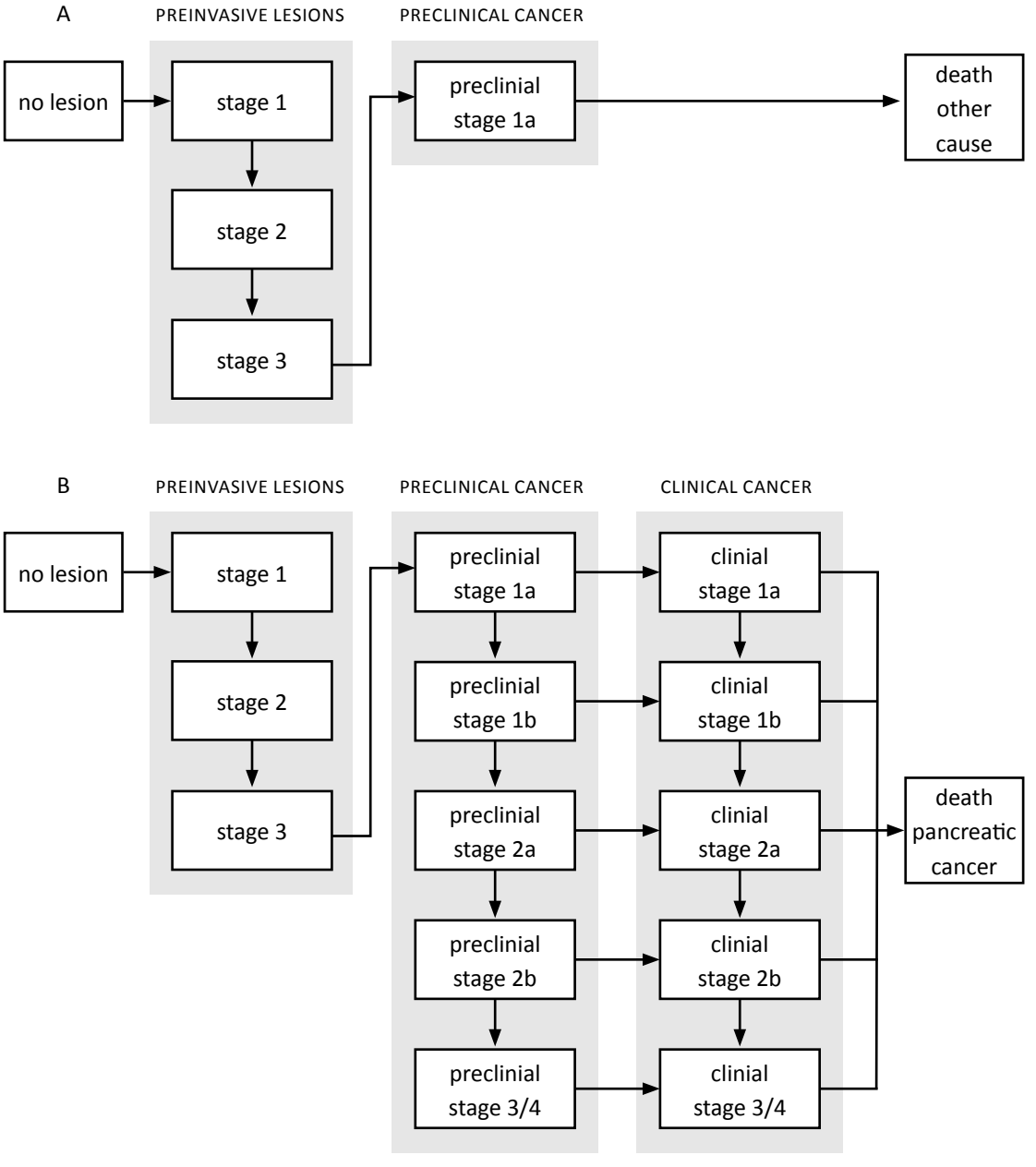
our simulation not only the screen population differed, but also the screening tests, the screen policy and the management strategy. Rubenstein *et al.* (25) concluded that for men with features of chronic pancreatitis, which are at high risk for developing PDAC, the most effective strategy to manage is no intervention. This study compared 4 different management strategies; no intervention, total pancreatectomy, EUS surveillance and EUS - Fine Needle Aspiration surveillance. Apart from the fact that their screening population entailed patients with a diseased pancreas with features of chronic inflammation (scars and calcifications), much different from our population in which most pancreatic glands have a normal architecture, this study took into account high morbidity rates and loss of quality of life in persons with total pancreatectomy.

Microsimulation is driven by assumptions. Ideally, these assumptions are based on high quality evidence. In the case of surveillance for pancreatic cancer in high-risk individuals scientific data are scarce which is a limitation of the current study. This paucity of high quality evidence can be partly explained by the relatively short period of time in which the effects of PDAC screening are being studied (the first report dates from 1999) (27) Among others, these data are needed because they contain information about test characteristics and the natural behavior of detected lesions. Furthermore, as previously mentioned, we lack high quality data with respect to the level of PDAC risk and natural behavior and development of PDAC. For example, we could not use data with information on the prevalence of preinvasive lesions. As a result, more or less indolent but detectable lesions may be missing in the model, underestimating overdiagnosis and - treatment of screening and surveillance. For the current study we have based our assumptions on the recommendations of the International Cancer of the Pancreas (CAPS) consortium (4) and thorough literature review. Our model therefore is designed and inputted by assumptions and data that represent current state in the field. Another strength of our study is that we have used a formal microsimulation model (in which the assumptions are well defined, can be reproduced and of which outputs can be compared with observations), and performed comprehensive sensitivity analyses, through which we have tested the robustness of our results.

We emphasize that this paper does not give the final answer to the question whether we need to screen for PDAC in high-risk individuals or not. By developing this comprehensive microsimulation model for PDAC, we were in the first place able to show that screening for PDAC in high-risk individuals looks, under plausible assumptions, promising and that therefore further research is warranted. Secondly, we could highlight which specific research questions are of major importance to further expand and refine the model with more reliable estimates of key input factors in order to improve upon the prediction value and applicability of the model. We found that the follow up strategy of screen positives is one of the key factors that determine the potential success of pancreatic cancer screening. Regarding the natural history, we showed that the probability of progression, the growth rate and the duration of the preclinical stage is important. We also showed

that it is important to include populations with a high enough risk to develop PDAC. To identify these risk groups, more epidemiological research needs to be done. Finally we showed how different interval cancers rates are expected with different natural history and test characteristic assumptions, which, conditional on a well-established risk level of the study group, can be compared with observed rates (the model in this way can also be used for powering such studies). Indeed, the final effect of screening can only be derived from clinical trials.

APPENDIX



Appendix Figure 1. Schematic overview of the natural history in the MISCAN-pancreas model. Slow developing (A) versus progressive (B) disease pathway.

	True stage	1. Back to screening	2. Surveillance, 6 months follow up	3. Resection
Base case	Normal	90%	10%	0%
	Preinvasive stage 1	85%	12%	3%
	Preinvasive stage 2	68%	17%	15%
	Preinvasive stage 3	26%	44%	30%
	Preclinical 1	10%	9%	81%
	Preclinical 2	7%	7%	86%
	Preclinical 3/4	0%	0%	100%*
Sensitivity -10%	Normal	90%	10%	0%
	Preinvasive stage 1	86%	11%	3%
	Preinvasive stage 2	62%	24%	14%
	Preinvasive stage 3	44%	33%	23%
	Preclinical 1	19%	8%	73%
	Preclinical 2	16%	6%	78%
	Preclinical 3/4	0%	0%	100%*
Sensitivity -5%	Normal	90%	10%	0%
	Preinvasive stage 1	86%	11%	3%
	Preinvasive stage 2	60%	26%	14%
	Preinvasive stage 3	30%	41%	29%
	Preclinical 1	14%	9%	77%
	Preclinical 2	12%	6%	82%
	Preclinical 3/4	0%	0%	100%*
Sensitivity +5%	Normal	90%	10%	0%
	Preinvasive stage 1	84%	13%	3%
	Preinvasive stage 2	66%	22%	12%
	Preinvasive stage 3	33%	40%	27%
	Preclinical 1	5%	10%	86%
	Preclinical 2	2%	7%	91%
	Preclinical 3/4	0%	0%	100%*
Sensitivity +10%	Normal	90%	10%	0%
	Preinvasive stage 1	83%	14%	3%
	Preinvasive stage 2	54%	29%	17%
	Preinvasive stage 3	9%	54%	37%
	Preclinical 1	1%	10%	89%
	Preclinical 2	0%	7%	93%
	Preclinical 3/4	0%	0%	100%*

All screen positives to resection	Normal	90%	10%	0%
	Preinvasive stage 1	40%	0%	60%
	Preinvasive stage 2	40%	0%	60%
	Preinvasive stage 3	25%	0%	75%
	Preclinical 1	10%	0%	90%
	Preclinical 2	7%	0%	93%
	Preclinical 3/4	0%	0%	100%*
≥preinvasive stage 3 to resection	Normal	90%	10%	0%
	Preinvasive stage 1	40%	60%	0%
	Preinvasive stage 2	40%	60%	0%
	Preinvasive stage 3	25%	0%	75%
	Preclinical 1	10%	0%	90%
	Preclinical 2	7%	0%	93%
	Preclinical 3/4	0%	0%	100%*
≥cancer to resection	Normal	90%	10%	0%
	Preinvasive stage 1	40%	60%	0%
	Preinvasive stage 2	40%	60%	0%
	Preinvasive stage 3	25%	75%	0%
	Preclinical 1	10%	0%	90%
	Preclinical 2	7%	0%	93%
	Preclinical 3/4	0%	0%	100%*

Appendix Table 1. Assumptions for the sensitivity analyses on the sensitivity of the screening test and the follow-up strategy (i.e. management). See Figure 2. Transition 1 ('back to screening') is the effect of (false-) negative test result.

**In case of cancer stage 3/4, 99% of the patients only receives palliative care (no resection)*

Model A	Cancer cases prevented (per 10,000 persons)	Cancer deaths prevented (per 10,000 persons)	Incidence (per 100,000 LYs)	Mortality (per 100,000 LYs)	Interval cancers <5 years (per 100,000 LYs)	All Interval cancers (per 100,000 LYs)	NNS	NNT	NNSurv
NO SCREENING			132	108	n.a.	n.a.	n.a.	n.a.	n.a.
Sensitivity analyses for screening annually from age 50 to 75									
BASE CASE	454	433	67	46	25	40	431	2.9	47
Sensitivity +5%	461	437	66	45	23	38	427	2.8	46
Sensitivity +10%	470	442	64	44	21	36	422	2.8	46
Sensitivity -5%	445	427	68	46	27	43	437	2.9	47
Sensitivity -10%	436	421	69	47	30	45	443	2.9	48
Specificity 85%	459	435	66	45	25	40	429	2.8	67
Specificity 95%	451	431	67	46	24	39	433	2.8	26
Specificity 100%	447	429	68	46	24	39	435	2.9	5
Risk Halved	240	225	34	23	12	20	844	2.8	87
Risk Doubled	822	800	130	89	49	79	225	3.0	26
All screen positives resection	588	494	47	37	13	26	381	2.9	39
≥Preinv stage 3 screen positives resection	538	480	55	39	20	42	392	2.2	45
≥Cancer screen positives resection	-133	168	151	84	53	77	1087	4.8	133
Treatment mortality 5%	455	414	67	48	25	40	450	3.0	49
Sensitivity analyses for screening every 5 year, from age 50 to 75									
BASE CASE	198	256	104	71	123	136	166	3.3	19
Sensitivity +5%	206	266	102	70	114	127	160	3.3	19
Sensitivity +10%	216	274	101	68	106	120	155	3.3	18
Sensitivity -5%	187	246	105	72	133	146	172	3.3	20
Sensitivity -10%	178	235	106	74	143	156	180	3.4	21
Specificity 85%	199	257	103	71	123	136	165	3.3	27
Specificity 95%	195	255	104	71	123	136	167	3.3	12
Specificity 100%	192	253	104	71	123	137	168	3.3	4
Risk Halved	105	134	53	36	62	69	323	3.2	35
Risk Doubled	345	470	200	137	244	269	87	3.4	12
All screen positives resection	329	325	85	61	89	101	131	3.3	14
≥Preinv stage 3 screen positives resection	185	248	105	72	123	139	172	3.0	19
≥Cancer screen positives resection	-93	84	146	96	190	207	502	6.1	60
Treatment mortality 5%	198	247	103	72	123	136	172	3.4	20

Appendix Table 2. Results of the sensitivity analyses for screening at ages 50 to 75, with an 1 and 5 year interval in model A (progressive disease). The interval cancer rate is presented as the number of cancer cases (per 100,000 LYs) in the first 5 years after a negative screening test and in the total period after a negative screening test (including at age 75). NNS = Number of screening tests need to perform to prevent one cancer death, NNT = Number needed to treat to prevent one cancer death, NNSurv = Number of surveillance test need to performed to prevent one cancer death.

** Cancer cases and deaths per 10,000 persons*

Model B	Cancer cases prevented (per 10,000 persons)	Cancer deaths prevented (per 10,000 persons)	Incidence (per 100,000 LYs)	Mortality (per 100,000 LYs)	Interval cancers <5 years (per 100,000 LYs)	All Interval cancers (per 100,000 LYs)	NNS	NNT	NNSurv
NO SCREENING			132	108	n.a.	n.a.	n.a.	n.a.	n.a.
Sensitivity analyses for screening annually from age 50 to 75									
BASE CASE	259	309	94	64	79	106	600	4.1	65
Sensitivity +5%	268	316	93	63	74	101	587	4.1	64
Sensitivity +10%	277	323	92	62	69	97	575	4.1	62
Sensitivity -5%	249	302	96	65	85	112	615	4.2	66
Sensitivity -10%	241	295	97	66	91	118	630	4.2	68
Specificity 85%	264	311	94	63	80	107	596	4.1	93
Specificity 95%	256	307	95	64	79	105	605	4.1	36
Specificity 100%	249	303	96	64	78	105	613	4.2	7
Risk Halved	137	161	48	33	41	55	1178	4.1	122
Risk Doubled	460	569	182	123	157	206	314	4.3	37
All screen positives resection	419	399	71	51	51	77	471	3.7	48
≥Preinv stage 3 screen positives resection	374	381	78	53	67	97	492	2.8	58
≥Cancer screen positives resection	-201	132	161	89	106	133	1385	5.6	173
Treatment mortality 5%	260	291	94	66	79	106	638	4.4	69
Sensitivity analyses for screening every 5 year, from age 50 to 75									
BASE CASE	31	130	127	89	237	257	326	6.0	38
Sensitivity +5%	37	136	126	89	230	250	311	5.9	37
Sensitivity +10%	44	141	125	88	224	244	299	5.8	36
Sensitivity -5%	24	124	128	90	244	264	340	6.1	40
Sensitivity -10%	18	119	129	91	251	270	356	6.2	41
Specificity 85%	34	132	127	89	237	256	321	5.9	53
Specificity 95%	29	128	128	90	237	257	331	6.1	23
Specificity 100%	918	753	128	90	238	257	336	6.1	8
Risk Halved	16	67	65	46	120	130	639	6.0	69
Risk Doubled	53	242	242	171	467	501	167	6.0	22
All screen positives resection	137	189	112	81	194	214	224	5.4	24
≥Preinv stage 3 screen positives resection	44	134	125	89	235	255	314	4.8	36
≥Cancer screen positives resection	-171	48	156	101	281	299	870	8.4	107
Treatment mortality 5%	32	120	127	91	237	257	353	6.5	41

Appendix Table 3. Results of the sensitivity analyses for screening at ages 50 to 75, with an 1 and 5 year interval in model B (including indolent disease). The interval cancer rate is presented as the number of cancer cases (per 100,000 LYs) in the first 5 years after a negative screening test and in the total period after a negative screening test (including at age 75). NNS = Number of screening tests need to perform to prevent one cancer death, NNT = Number needed to treat to prevent one cancer death, NNSurv = Number of surveillance test need to performed to prevent one cancer death.

** Cancer cases and deaths per 10,000 persons*

† An increased preclinical duration has negligible impact mortality reduction when screening annually. The increased NNSurv compared to the base case NNSurv is a result coincidence.

REFERENCES

1. Jemal A, Bray F, Center MM, Ferlay J, Ward E, Forman D. Global cancer statistics. CA Cancer J Clin 2011;61:69-90.
2. Harinck F, Poley JW, Kluijt I, Fockens P, Bruno MJ, Surv DRGPC. Is Early Diagnosis of Pancreatic Cancer Fiction? Surveillance of Individuals at High Risk for Pancreatic Cancer. Digestive Diseases 2010;28:670-678.
3. Wilson JMG, Jungner G. Principles and practice of screening for disease. Geneva: WHO, 1968.
4. Canto MI, Harinck F, Hruban RH, Offerhaus GJ, Poley JW, Kamel I, Nio Y, Schulick RS, Bassi C, Kluijt I, Levy MJ, Chak A, Fockens P, Goggins M, Bruno M. International Cancer of the Pancreas Screening (CAPS) Consortium summit on the management of patients with increased risk for familial pancreatic cancer. Gut 2013;62:339-47.
5. van Oortmarssen GJ, Boer R, Habbema JD. Modelling issues in cancer screening. Stat Methods Med Res 1995;4:33-54.
6. Karnon J, Goyder E, Tappenden P, McPhie S, Towers I, Brazier J, Madan J. A review and critique of modelling in prioritising and designing screening programmes. Health Technol Assess 2007;11:iii-iv, ix-xi, 1-145.
7. Habbema JDF, Vanoortmarssen GJ, Lubbe JTN, Vandermaas PJ. The Miscan Simulation Program for the Evaluation of Screening for Disease. Computer Methods and Programs in Biomedicine 1985;20:79-93.
8. de Kok IMCM, van Ballegooijen M, Habbema JDF. Cost-Effectiveness Analysis of Human Papillomavirus Vaccination in the Netherlands. Journal of the National Cancer Institute 2009;101:1083-1092.
9. de Kok IMCM, van Rosmalen J, Dillner J, Arbyn M, Sasieni P, Iftner T, van Ballegooijen M. Primary screening for human papillomavirus compared with cytology screening for cervical cancer in European settings: cost effectiveness analysis based on a Dutch microsimulation model. British Medical Journal 2012;344.
10. Draisma G, Boer R, Otto SJ, van der Cruijsen IW, Damhuis RAM, Schroder FH, de Koning HJ. Lead times and overdetection due to prostate-specific antigen screening: Estimates from the European randomized study of screening for prostate cancer. Journal of the National Cancer Institute 2003;95:868-878.
11. Heijnsdijk EA, Wever EM, Auvinen A, Hugosson J, Ciatto S, Nelen V, Kwiatkowski M, Villers A, Paez A, Moss SM, Zappa M, Tammela TL, Makinen T, Carlsson S, Korfage IJ, Essink-Bot ML, Otto SJ, Draisma G, Bangma CH, Roobol MJ, Schroder FH, de Koning HJ. Quality-of-life effects of prostate-specific antigen screening. N Engl J Med 2012;367:595-605.
12. Lansdorp-Vogelaar I, van Ballegooijen M, Zauber AG, Habbema JD, Kuipers EJ. Effect of rising chemotherapy costs on the cost savings of colorectal cancer screening. J Natl Cancer Inst 2009;101:1412-22.

13. Saadatmand S, Tilanus-Linthorst MM, Rutgers EJ, Hoogerbrugge N, Oosterwijk JC, Tollenaar RA, Hooning M, Loo CE, Obdeijn IM, Heijnsdijk EA, de Koning HJ. Cost-effectiveness of screening women with familial risk for breast cancer with magnetic resonance imaging. J Natl Cancer Inst 2013;105:1314-21.
14. Wilschut JA, Habbema JD, van Leerdam ME, Hol L, Lansdorp-Vogelaar I, Kuipers EJ, van Ballegooijen M. Fecal occult blood testing when colonoscopy capacity is limited. J Natl Cancer Inst 2011;103:1741-51.
15. Yachida S, Jones S, Bozic I, Antal T, Leary R, Fu B, Kamiyama M, Hruban RH, Eshleman JR, Nowak MA, Velculescu VE, Kinzler KW, Vogelstein B, Iacobuzio-Donahue CA. Distant metastasis occurs late during the genetic evolution of pancreatic cancer. Nature 2010;467:1114-7.
16. Canto MI, Hruban RH, Fishman EK, Kamel IR, Schulick R, Zhang Z, Topazian M, Takahashi N, Fletcher J, Petersen G, Klein AP, Axilbund J, Griffin C, Syngal S, Saltzman JR, Mortele KJ, Lee J, Tamm E, Vikram R, Bhosale P, Margolis D, Farrell J, Goggins M, CAPS ACPS. Frequent Detection of Pancreatic Lesions in Asymptomatic High-Risk Individuals. Gastroenterology 2012;142:796-804.
17. Hunt GC, Faigel DO. Assessment of EUS for diagnosing, staging, and determining resectability of pancreatic cancer: a review. Gastrointestinal Endoscopy 2002;55:232-7.
18. Kim YC, Choi JY, Chung YE, Bang S, Kim MJ, Park MS, Kim KW. Comparison of MRI and endoscopic ultrasound in the characterization of pancreatic cystic lesions. AJR Am J Roentgenol 2010;195:947-52.
19. Othman MO, Wallace MB. The role of endoscopic ultrasonography in the diagnosis and management of pancreatic cancer. Gastroenterol Clin North Am 2012;41:179-88.
20. Sohn TA, Yeo CJ, Cameron JL, Koniaris L, Kaushal S, Abrams RA, Sauter PK, Coleman J, Hruban RH, Lillemoe KD. Resected adenocarcinoma of the pancreas - 616 patients: Results, outcomes, and prognostic indicators. Journal of Gastrointestinal Surgery 2000;4:567-579.
21. Winter JM, Cameron JL, Campbell KA, Arnold MA, Chang DC, Coleman J, Hodgin MB, Sauter PK, Hruban RH, Riall TS, Schulick RD, Choti MA, Lillemoe KD, Yeo CJ. 1423 pancreaticoduodenectomies for pancreatic cancer: A single-institution experience. Journal of Gastrointestinal Surgery 2006;10:1199-1210.
22. Fatima J, Schnelldorfer T, Barton J, Wood CM, Wiste HJ, Smyrk TC, Zhang L, Sarr MG, Nagorney DM, Farnell MB. Pancreatoduodenectomy for ductal adenocarcinoma: implications of positive margin on survival. Arch Surg 2010;145:167-72.
23. Hewitson P, Glasziou P, Watson E, Towler B, Irwig L. Cochrane systematic review of colorectal cancer screening using the fecal occult blood test (hemoccult): an update. American Journal of Gastroenterology 2008;103:1541-9.
24. Schnelldorfer T, Ware AL, Sarr MG, Smyrk TC, Zhang L, Qin R, Gullerud RE, Donohue JH, Nagorney DM, Farnell MB. Long-term survival after pancreatoduodenectomy for pancreatic adenocarcinoma: is cure possible? Ann Surg 2008;247:456-62.

25. Rubenstein JH, Scheiman JM, Anderson MA. A clinical and economic evaluation of endoscopic ultrasound for patients at risk for familial pancreatic adenocarcinoma. Pancreatology 2007;7:514-25.
26. Rulyak SJ, Kimmey MB, Veenstra DL, Brentnall TA. Cost-effectiveness of pancreatic cancer screening in familial pancreatic cancer kindreds. Gastrointestinal Endoscopy 2003;57:23-9.
27. Brentnall TA, Bronner MP, Byrd DR, Haggitt RC, Kimmey MB. Early diagnosis and treatment of pancreatic dysplasia in patients with a family history of pancreatic cancer. Annals of Internal Medicine 1999;131:247-55.
28. Bilimoria KY, Bentrem DJ, Ko CY, Ritchey J, Stewart AK, Winchester DP, Talamonti MS. Validation of the 6th edition AJCC Pancreatic Cancer Staging System: report from the National Cancer Database. Cancer 2007;110:738-44.

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Submitted

CHAPTER 6
CLINICOPATHOLOGICAL CHARACTERISTICS OF PATIENTS WITH INHERITED OR FAMILIAL PANCREATIC DUCTAL ADENOCARCINOMA

ABSTRACT

Since the extreme poor prognosis of pancreatic cancer is mainly due to the late occurrence of symptoms, there is a growing interest towards the early detection of pancreatic cancer in individuals with an increased inherited or familial risk for this disease. When designing screening programs aiming to identify high-risk lesions for early resection, knowledge of the pathology of the disease is essential. In this current study we focus on the clinicopathological characteristics of patients with inherited or familial pancreatic cancer in comparison to sporadic cases. Our results showed that high grade precursor lesions were more frequently found in inherited or familial pancreatic cancer cases. Since these high-grade precursor lesions are key targets for early detection, our findings have important implications within the context of screening/surveillance of individuals at high risk for developing pancreatic cancer.

INTRODUCTION

Pancreatic ductal adenocarcinoma (PDAC) is still one of the deadliest cancer types worldwide (1, 2). Since its extreme poor prognosis is mainly caused by the late occurrence of symptoms, there is a growing interest towards early detection of PDAC in individuals with an increased inherited or familial risk of PDAC (3). It is currently estimated that about 10% of all PDAC-cases are caused by inherited and/or familial factors (4).

Evidence is slowly beginning to accumulate that screening and surveillance of high-risk individuals with EUS and/or MRI leads to the detection of non-invasive precursor lesions and asymptomatic early stage PDAC (3) at a time when the disease is still curable. Though these results are promising, proof that screening is effective and lowers mortality is currently lacking (5).

Several questions concerning PDAC remain unanswered (3). Thorough knowledge of the natural history and pathology of a disease is essential in order to conclude whether individuals with an increased risk for developing such condition may benefit from screening/surveillance and by which means (5). Pancreatic carcinogenesis in both inherited/familial and sporadic PDAC involves stepwise progression of distinctive pathologic precursor lesions including pancreatic intraepithelial neoplasia (PanIN) and intraductal papillary mucinous neoplasm (IPMN) (6-8). The occurrence, distribution, and speed of progression of these precursor lesions might differ between inherited or familial PDAC and sporadic PDAC patients. Such observation could partly explain the increased frequency of PDAC in high-risk individuals and the observation that, within specific inherited tumor syndromes, PDAC occurs earlier compared to sporadic cases (3). It may also highlight which type of lesion is most important to look for and it may have implications for choosing the most appropriate screening technique. Thus far only one study has investigated the prevalence of precursor lesions in patients with familial pancreatic cancer (9). The aim of the current study therefore was to study the prevalence and grade of precursor lesions in patients with inherited and familial PDAC compared to sporadic cases.

METHODS

Selection of cases

Familial and inherited cases

Data were gathered from the PC-family registry of our working group; the Dutch Research Group of Pancreatic Cancer Surveillance in high-risk individuals. This working group is a multicenter collaboration between three academic centres (University Medical Center Rotterdam - Erasmus MC (Rotterdam), University Medical Center Amsterdam – Academic Medical Center Amsterdam (Amsterdam) and University of Groningen - University Medical Center Groningen (Groningen)) and one specialized oncological center (The Netherlands Cancer Institute - Antoni van Leeuwenhoek Hospital (Amsterdam)). The registry consists

of Dutch families affected by a (1) familial or (2) inherited form of PDAC. From this registry we selected family members affected by PDAC and in whom (intended) curative pancreatectomy was performed. We contacted the hospitals were these patients were operated to collect formalin fixed and paraffin-embedded pathological material.

Sporadic cases
Data were gathered from a registry consisting of all patients who received pancreatectomy at the Erasmus MC. Patients who received (intended) curative pancreatectomy because of PDAC in the period between 1/1/2006 and 25/11/2011 were selected. From this selection we randomly included patients for this analysis.

At the time of surgery, all patients had given written informed consent to use their pathological material for research purposes.

Data collection
For all selected cases, we recorded the demographic parameters (age, gender, genetic status (if applicable)) and reviewed the pathology reports for information concerning the size of the tumor as measured during the pathological examination, the location of the tumor (head, body, or tail), the TNM-tumor stage based on results of the pathological examination and information about the resection margin.

Microscopic examination
For each of the selected cases, all available histological slides were reviewed and scored by an experienced pancreatic pathologist (KB). The microscopic examination focused on areas with non-invasive carcinoma tissue. This tissue was scored for the presence, number, and grade of foci of PanIN lesions and incipient IPMNs according to the consensus definitions described by Hruban (10). One field corresponded with 0.2375 cm^2. Furthermore, we scored for the presence of acinar-ductal metaplasia, atrophy and chronic inflammation.

Statistical analysis
Data were analyzed using the SPSS 20.0 statistical software for Windows (IBM, Somers, New York, USA). Depending on the level of measurement, Chi-square/Fisher's exact tests or Student's t-test were used to assess the differences between the two groups. Furthermore, sub-analyses were performed within the group of inherited/familial PDAC to test for differences between patients with a proven gene mutation and patients with FPC. A two-sided p value <0.05 was considered to be statistically significant.

RESULTS

Study population

We evaluated pancreatectomy specimens from 16 patients with inherited/familial PDAC and 19 patients with sporadic PDAC. The 16 cases with inherited/familial PDAC originated from 13 distinct families. All sporadic cases originated from independent families. Twelve cases (75%) from the inherited/familial group had familial PDAC, 3 (19%) were carriers of *CDKN2A* mutations and one (6.3%) carried a *BRCA2* mutation. Table 1 shows the characteristics of the studied population. An average of 49 fields were available for evaluation (SD 38). There was no significant difference between both groups with respect to the mean age at time of diagnosis, tumor stage, tumor size and resection margins, but the number of fields available for evaluation per case was significantly higher in the inherited/familial group compared to the sporadic group (70 vs. 31, p=0.005).

	Cases n=16	Controls n=19	p-value
Mean age, yrs (SD)	63 (8.9)*	66 (8.9)**	0.352
Gender, n (%)			
Female	8 (50)	7 (37)	
Male	8 (50)	12 (63)	0.448
Stage (based on pathology), n (%)			
T1N0	2 (13)	0 (0)	
T1N1	1 (6)	0 (0)	
T2N0	0 (0)	3 (16)	
T2N1	1 (6)	1 (5)	
T3NO	1 (6)	6 (32)	
T3N1	10 (63)	9 (47)	
Unknown	1 (6)	0 (0)	0.107
Mean size tumor, mm (SD)	31 (15)*	34 (12)	0.515
Tumor location, n (%)			
Head	12 (75)	15 (79)	
Body	0 (0)	0 (0)	
Tail	3 (19)	4 (21)	
Unknown	1 (6)	0 (0)	0.541
Resection margin, n (%)			
Positive	10 (63)	16 (84)	
Negative	5 (31)	3 (16)	
Unknown	1 (6)	0	0.266
Mean number of fields studied, n (SD)***	70 (45)	31 (19)	0.005

Table 1. Baseline characteristics

results of 2 cases missing; **results of 1 case missing; * 1 field = 0.2375 cm^2*

Microscopic examination

Table 2 shows the results of the microscopic examination. PanIN lesions were the most often found precursor lesions for both groups. A significant difference was observed between the mean number of PanIN lesions (9.2 vs. 2.7, p=0.04) between the two groups. The number of patients in whom at least two high-grade precursors were detected was significantly higher in the inherited/familial group. More patients within the inherited/familial group had PanIN-3 lesions. Furthermore, in significantly more patients within the inherited/familial group multiple PanIN lesions were detected. Incipient IPMNs were rarely detected; an incipient IPMN grade I lesion was observed in two cases (13%) and one control patient (5%). The number of patients in whom parenchymal changes of acinar-ductal metaplasia, atrophy and chronic inflammation were detected did not differ between the two groups.

The sub-analyses within the inherited/familial PDAC group did not result in significant differences between patients with a proven gene mutation and those with familial PDAC.

	Cases n=16	Controls n=19	p-value
# Pts with presence of:			
Any type of precursor lesion[*]*, n*	11	12	0.728
High-grade precursor lesion[**]*, n*	6	1	0.032
Both PanINs and incIPMNs present, n	2	1	0.582
Both high-grade PanINs and incIPMNs present[**]*, n*	0	0	n.a.
# Pts with presence of:			
PanIN-I, n	10	7	0.130
PanIN-II, n	9	7	0.251
PanIN-III, n	6	1	0.032
Multiple PanINs, n	8	3	0.030
Multiple PanIN-IIIs, n	3	1	0.312
Mean number of:			
Total PanINs, n (SD)	9.2 (12.5)	2.7 (4.0)	0.04
PanIN-I, %	26	41	0.177
PanIN-II, %	62	56	0.061
PanIN-III, %	12	4.1	0.05
Number of patients with CAM[±], n (%)	13	11	0.138
Number of patients with CI[¥], n (%)	12	10	0.172

Table 2. Pathological findings

**PanIN and/or incipient IPMN, any degree; **PanIN-III and/or incipient IPMN grade 3; ± Acinar-ductal metaplasia*

¥ Chronic inflammation; n.a.: Not applicable

DISCUSSION

Our data show that the number of patients with presence of high-grade precursor lesions was significantly higher in the inherited/familial cases compared to the sporadic cases. Interestingly, all of these high-grade precursor lesions were PanIN-lesions as high-grade (incipient)IPMNs were not detected. Furthermore, a higher number of precursor lesions were detected in the inherited/familial cases.

Our findings are partly in line with the results of Shi *et al.* (9) which to date is the only other study investigating the prevalence of precursor lesions in familial pancreatic cancer patients. In that retrospective study it was also shown that PanIN-3 lesions were the predominantly found precursor lesions. Shi *et al.* also found that the density of the precursor lesions was significantly higher in the familial PDAC population. Unfortunately, we were not able to calculate the density in our series as there was a significantly difference in the number of slides available for pathological examination between both groups.

This significant difference in available slides was the main limitation of this current study. Unfortunately, this is the consequence of the retrospective design of our study in which there was only limited availability of pathology slides. By no means the whole resected pancreatic specimen was investigated. Also a limited number of cases were included which consequently restricts the ability to detect a possible difference.

Though speculative, our data might suggest that the prevalence of high-grade precursors within inherited/familial cases is by all means not lower compared to sporadic cases. This is based on the fact that with just a twofold increased number of pathological slides available, the number of patients with presence of high-grade precursors was six fold higher within the inherited/familial cases. Had we found no difference between both groups, this would have suggested an lower prevalence.

What do these findings signify within the context of PDAC screening? When PanIN-3 lesions are detected and surgically resected this is considered a successful outcome of the screening/surveillance program according to a recently published consensus report (3). In the present study, we found an increased prevalence of PanIN-3 lesions in the pancreatic parenchyma of the areas adjacent to PDAC in patients with inherited/familial PDAC.). This suggests an accelerated progression of PanIN lesions to clinically relevant lesions in these high-risk individuals. Unfortunately, we currently lack a diagnostic test that reliably detects high-grade PanIN-lesions while differentiating them from lower-grade PanIN lesions. Although higher grades of PanIN lesions may be associated with (early) features of fibrosis that can be visualized by EUS, it has not yet been proven that this feature can be used in clinical practice to reliably identify patients with PanIN 3 lesions (11, 12). When not correctly diagnosed, such a lack in diagnostic and discriminative power could result in either a false negative test outcome when PanIN-3 lesions are missed, or a false positive test outcome when lower grade PanIN lesions resemble and are mistaken for PanIN-3 lesions. In the latter scenario, individuals undergo a pancreatic resection without a

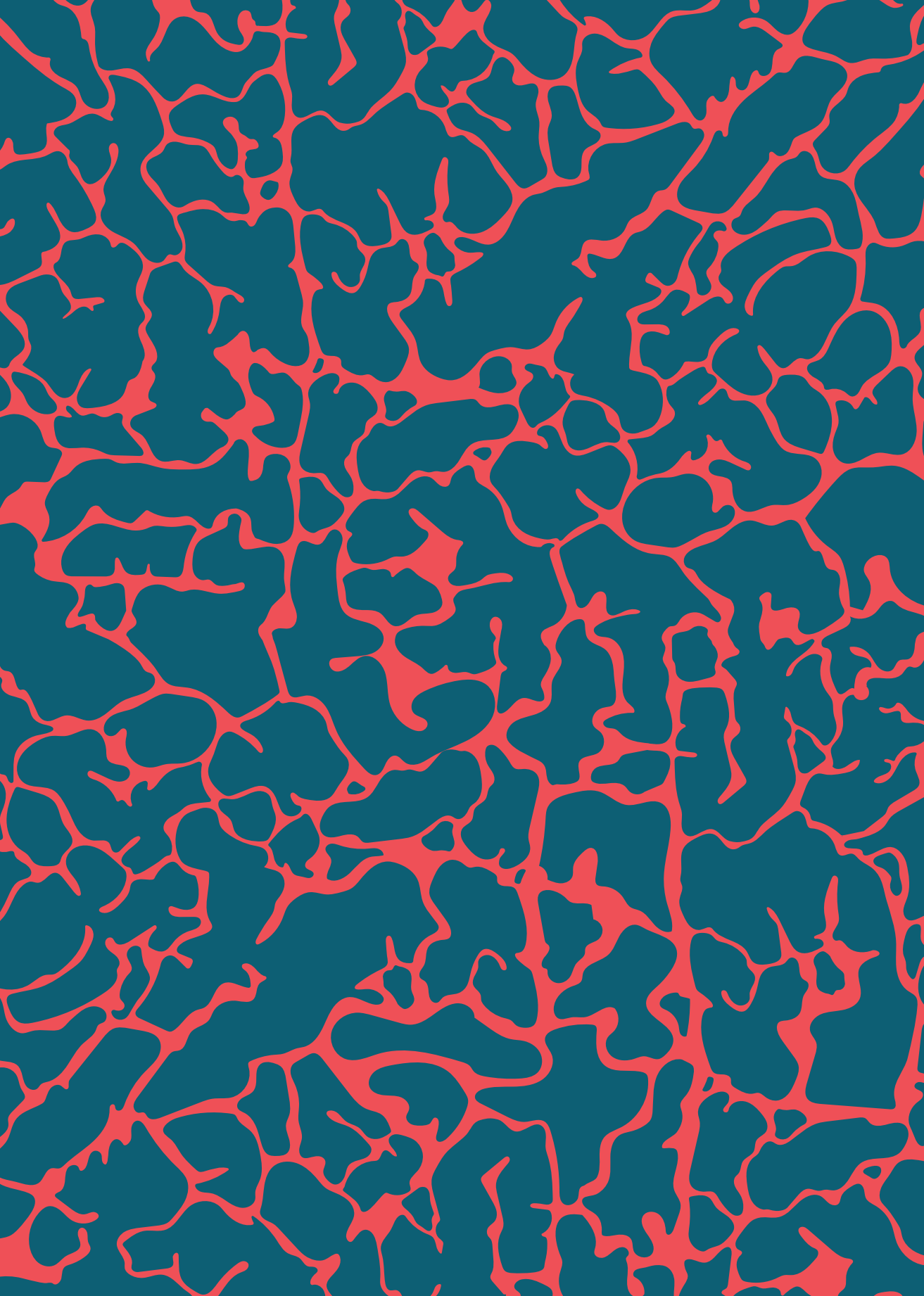
clinically relevant lesion being identified at final pathology review. With the current rapid developments in the fields of molecular markers and micro-RNA (13), it might be possible to test for these lesions using molecular analysis of duodenal collections of pancreatic juice, blood, or feces. A recently published study showed that duodenal collections of secretin-stimulated pancreatic juice are an excellent source of mutant DNA from the pancreas (14).

In conclusion, our findings show that both the number of patients with presence of high-grade precursor lesions and the number of high-grade PanIN-lesions in patients with inherited or familial PDAC is higher than in patients with sporadic PDAC. These high-grade precursor lesions are an important target for screening and surveillance of high-risk individuals for which the most suitable test has yet to be identified.

REFERENCES

1. van Asperen CJ, Brohet RM, Meijers-Heijboer EJ, Hoogerbrugge N, Verhoef S, Vasen HF, et al. Cancer risks in BRCA2 families: estimates for sites other than breast and ovary. J Med Genet. 2005;42(9):711-9. Epub 2005/09/06.
2. Brand RE, Lerch MM, Rubinstein WS, Neoptolemos JP, Whitcomb DC, Hruban RH, et al. Advances in counselling and surveillance of patients at risk for pancreatic cancer. Gut. 2007;56(10):1460-9.
3. Canto MI, Harinck F, Hruban RH, Offerhaus GJ, Poley JW, Kamel I, et al. International Cancer of the Pancreas Screening (CAPS) Consortium summit on the management of patients with increased risk for familial pancreatic cancer. Gut. 2013;62(3):339-47. Epub 2012/11/09.
4. Lynch HT, Smyrk T, Kern SE, Hruban RH, Lightdale CJ, Lemon SJ, et al. Familial pancreatic cancer: a review. Seminars in oncology. 1996;23(2):251-75. Epub 1996/04/01.
5. Wilson JM, Jungner G. Principles and Practice of Screening for Disease. Geneva:World Health Organisation. 1968.
6. Brat DJ, Lillemoe KD, Yeo CJ, Warfield PB, Hruban RH. Progression of pancreatic intraductal neoplasias to infiltrating adenocarcinoma of the pancreas. Am J Surg Pathol. 1998;22(2):163-9. Epub 1998/03/21.
7. Hruban RH, Wilentz RE, Maitra A. Identification and analysis of precursors to invasive pancreatic cancer. Methods Mol Med. 2005;103:1-13. Epub 2004/11/16.
8. Wu J, Matthaei H, Maitra A, Dal Molin M, Wood LD, Eshleman JR, et al. Recurrent GNAS mutations define an unexpected pathway for pancreatic cyst development. Sci Transl Med. 2011;3(92):92ra66. Epub 2011/07/22.
9. Shi C, Klein AP, Goggins M, Maitra A, Canto M, Ali S, et al. Increased Prevalence of Precursor Lesions in Familial Pancreatic Cancer Patients. Clin Cancer Res. 2009;15(24):7737-43. Epub 2009/12/10.
10. Hruban RH, Takaori K, Klimstra DS, Adsay NV, Albores-Saavedra J, Biankin AV, et al. An illustrated consensus on the classification of pancreatic intraepithelial neoplasia and intraductal papillary mucinous neoplasms. Am J Surg Pathol. 2004;28(8):977-87. Epub 2004/07/15.
11. Brune K, Abe T, Canto M, O'Malley L, Klein AP, Maitra A, et al. Multifocal neoplastic precursor lesions associated with lobular atrophy of the pancreas in patients having a strong family history of pancreatic cancer. Am J Surg Pathol. 2006;30(9):1067-76. Epub 2006/08/26.
12. Takaori K, Matsusue S, Fujikawa T, Kobashi Y, Ito T, Matsuo Y, et al. Carcinoma in situ of the pancreas associated with localized fibrosis: a clue to early detection of neoplastic lesions arising from pancreatic ducts. Pancreas. 1998;17(1):102-5. Epub 1998/07/17.

13. Matthaei H, Wylie D, Lloyd MB, Dal Molin M, Kemppainen J, Mayo SC, et al. miRNA biomarkers in cyst fluid augment the diagnosis and management of pancreatic cysts. Clinical cancer research : an official journal of the American Association for Cancer Research. 2012;18(17):4713-24. Epub 2012/06/23.
14. Kanda M, Knight S, Topazian M, Syngal S, Farrell J, Lee J, et al. Mutant GNAS detected in duodenal collections of secretin-stimulated pancreatic juice indicates the presence or emergence of pancreatic cysts. Gut. 2013;62(7):1024-33. Epub 2012/08/04.



PART II

WHO ARE AT RISK?

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CHAPTER 7
HEREDITARY AND FAMILIAL FORMS OF PANCREATIC CANCER

ABSTRACT

With a median survival of four to six months and a five-year survival of less than 5%, the prognosis of pancreatic cancer is poor. The cause lies mainly in the late occurrence of symptoms and the aggressiveness of this tumour type, whereby fewer than 20% of symptomatic patients have resectable disease at time of diagnosis and even in these patients, radical resection produces a five-year survival of less than 20%. screening for precursor lesions or malignancies at an early asymptomatic stage could potentially offer a way to improve the prognosis, especially when offered to individuals with an already high baseline risk of developing pancreatic cancer. This article addresses these high-risk individuals.

INTRODUCTION

Despite the low incidence of pancreatic cancer (PC) (8.5 per 100,000 per year in Europe), PC is the fourth leading cancer-related cause of death in Europe (1) with a median survival of less than six months and a five-year survival of less than 5%.(2) The poor prognosis is mainly due to the late onset of symptoms and the aggressiveness of this tumour, such that the majority of patients present with incurable disease. Screening for precursor lesions or malignancies at an early asymptomatic stage could potentially offer a way of improving the prognosis. Since the incidence of PC is low and we currently lack a non-invasive, reliable and cheap surveillance tool it is neither useful nor feasible to offer PC surveillance to the general population. However, surveillance may be worthwhile when offered to individuals at high risk of developing PC.

Currently, several groups of individuals at high risk of developing PC have been identified. (3) On the basis of clinical and genetic criteria, these high-risk individuals can be divided into two groups. In the first group, PC develops within the framework of a known hereditary cancer syndrome or hereditary disease. In these cases, the underlying causative genetic factor is known. The second group, referred to as familial PC (FPC), consists of families with clustering of PC and not meeting diagnostic criteria of specific hereditary cancer syndromes.

In this article, we provide an overview of PC-prone hereditary cancer syndromes and diseases, discuss the level of PC risk for the different genetic syndromes and FPC families and list diagnostic criteria that may help clinicians to determine whether there is an indication to refer to a clinical genetics centre because of a suspicion of an inherited or familial form of PC.

Hereditary cancer syndromes and diseases with an increaded PC-risk

Just a small fraction (~20%) of all inherited PC cases develop within a framework of a hereditary disease or cancer syndrome. In most of these syndromes, the risk of developing other types of cancer is higher than the risk of developing PC and therefore clustering of PC can be absent or less obvious. Known genetic diseases and hereditary cancer syndromes with an increased PC risk include: hereditary pancreatitis; Peutz–Jeghers syndrome (PJS); familial atypical multiple mole melanoma syndrome (FAMMM); hereditary breast and ovarian cancer syndrome; Lynch syndrome; and, to a lesser extent, Li–fraumeni syndrome.

Hereditary pancreatitis

Among all high-risk individuals, patients with hereditary pancreatitis (HP) are probably at the highest risk of developing PC. Several genes can cause a predisposition to HP (OMIM #167800) of which *PRSS1* causes the highest risk, leading to an autosomal dominant pattern of inheritance. This disease is characterised by recurrent attacks of acute pancreatitis which start in childhood or early adolescence and usually progress

to chronic pancreatitis with endocrine and exocrine failure. Patients with HP are at an increased risk of developing PC that has been attributed to the long duration of exposure to inflammation.

The majority of available data about PC risk in HP patients derives from three study groups: Rebours *et al*. (French cohort) (4,5), European Registry of Hereditary Pancreatitis and Pancreatic Cancer (EUROPAC) (6) and International Hereditary Pancreatitis Study Group (worldwide) (7). The relative risk (RR) found varied from 57 to 87 with a cumulative risk at age 70 that varied from 25.3 (6) to 53.5%.(4). The median age at PC diagnosis was around 55 years (5, 7) smoking doubles the risk of PC development in HP patients and lowers the age of PC onset which was found to be 20 years younger than seen in the general population (8).

Interesting findings with respect to mortality risk were published by Rebours *et al.* (9). Their results showed that HP patients do not have excess mortality risk compared with the general population. The median overall survival in a cohort of 189 HP patients was 74 years. The only factor of influence on survival was having PC. These findings emphasise the need for early detection of PC in HP patients. Unfortunately, proper examination of the pancreas in this group of patients is difficult since parenchymal changes due to chronic inflammation including calcifications make it hard to judge the presence or absence of (pre)malignant lesions.

Peutz–Jeghers Syndrome

PJS (OMIM #175200) is a rare autosomal dominant inherited syndrome, characterised by gastrointestinal hamartomas and mucocutaneous pigmentations. The incidence has been estimated between 1 in 8,300 to 1 in 280,000 individuals (10, 11) A clinical diagnosis of PJS can be made in individuals with at least two of the following criteria: ≥2 PJS polyps of the small bowel; characteristic mucocutaneous pigmentation on buccal mucosa, lips or digits; or a family history of PJs (12) Germline mutations in *STK11* cause PJS and with the currently available techniques a pathogenic *STK11* mutation can be detected in 80–94% of families with the PJS phenotype (13, 14).

PJS patients are at increased risk of developing cancer particularly for gastrointestinal cancers at a mean young age (15). PC forms part of the PJS, however risk estimates for PC differ largely between studies. In the largest study, a meta-analysis by Giardiello *et al.* (16) the RR of developing PC was 132 and the cumulative risk by age 65 was 36%. The mean age of onset of PC in this cohort was just 40.8 years which is significantly lower in comparison to the onset of sporadic PC (mean age 65 years) (17). A more recent collaboration study (18) among 419 PJS patients reported a life-time risk (LTR) of 11% by the age of 70. Worth mentioning is that in another collaboration study (19) no PCs were found in a cohort of 149 *STK11*-positive PJS patients. These contradictory findings emphasise the need for further research to better study the actual PC risk. This should ideally be done in a homogenous cohort of PJS families.

Familial Atypical Multiple Mole Melanoma Syndrome

Clinical diagnostic criteria of FAMMM syndrome (OMIM #606719) are a family history of melanomas in either ≥2 first-degree relatives (FDRs) or ≥3 relatives (20). The disease is inherited as an autosomal dominant trait, with germline mutations in *CDKN2A* having been reported in at east a quarter of all FAMMM families (21). In Europe, carriers of mutations in this gene run a LTR of 58% to develop melanomas (22). In addition, *CDKN2A* mutation carriers are at risk of other types of cancer, particularly PC (20, 23-25). Between the different *CDKN2A* mutations, the level of PC risk seems to differ. For instance, the RR for carriers of the Dutch founder mutation p16-Leiden (c.225_243del, p.Ala76fs) is 46 (24), whereas a RR of 14.8 is found in carriers of the Ligurian *CDKN2A* founder mutation (c.301g>T, gly101Trp). Goldstein et al (26) studied 15 *CDKN2A* mutation positive families with a total of eight different mutations and found a 52-times elevated risk of developing PC compared to the general population.

Hereditary Breast and Ovarian Cancer Syndrome

The hereditary breast and ovarian cancer syndrome (HBOC) is an autosomal dominant cancer syndrome, caused by germline mutations in either *BRCA1* (OMIM #604370) or *BRCA2* (OMIM #612555), with strongly increased risks of developing breast cancer (BC; LTR 60–80%), and of ovarian cancer (OC; LTRs 30–60% for *BRCA1* and 5–20% for *BRCA2*) (27).

PC is one of the other malignancies besides BC and OC associated with germline mutations in *BRCA1* and *BRCA2* (28-32). Data indicating that *BRCA1* and *BRCA2* play a causative role in the development of PC were published by Al-Sukhni *et al.* (33) and Skoulidis *et al.* (34). They studied a population of *BRCA1* and *BRCA2* mutation positive PC cases and found loss of heterozygosity in at least some of the cases.

The level of PC risk in carriers of *BRCA1/2* was studied by the Breast Cancer Linkage Consortium (35, 36). They reported RRs of 3.51 for PC in 173 *BRCA2* mutation families and RRs of 2.26 in 699 *BRCA1* mutation families. In a Dutch nationwide study, van Asperen (37) calculated a RR of 5.9 for PC (95% CI 3.2–10) in 139 *BRCA2* families, with higher risk for males, age >65 and mutations outside the ovarian cancer cluster region (OCCR). Risch *et al.* however showed in a Canadian cohort of OC patients a total RR of 3.4 (95% CI 1.4–8.5) for pancreatic, gastric and prostate cancer in family members of 21 *BRCA2* mutation carriers, but only for mutations within the OCCR (38). Kim *et al*. demonstrated a significant lower mean age at diagnosis of PC in mutation carriers from *BRCA1/2* families in comparison to the population mean, calculated from the national cancer institute (SEER) database. The mean age at diagnosis was 62.9 years for *BRCA1* (SD 12.0; p=0.0014) and 62.9 years for *BRCA2* mutation carriers (SD 11.7; p=0.011) compared to 70.0 years (SD 12.1) in SEER (39).

Lynch Syndrome

The Lynch syndrome (Hereditary Nonpolyposis Colorectal Carcinoma [HNPCC]) is an autosomal dominant tumour syndrome, with a strongly increased risk of developing

colorectal cancer and in females, endometrial cancer. The LTRs for both cancers is 25–70%, depending on the involved mismatch repair gene (*MLH1*, OMIM #609310; *MSH2*, OMIM #120435; *MSH6*, OMIM #600678; or *PMS2*, OMIM #600259). In addition, carriers of these mutations run an increased risk of developing other tumour types, among which is PC. Lynch reported in 1985, before the discovery of the mismatch repair genes, a HNPCC family with three PC cases, all before age 60, in subsequent generations.
Kastrinos *et al.* (40) demonstrated in familial cancer registries from two US cancer centres one or more PC cases in 31 of 147 families with a germline mutation in one of the mismatch repair genes, with in total 47 PC cases (13 from *MLH1* families, 31 from *MSH2* families and three from *MSH6* families). The mean ages of onset was 51.5 in males and 56.5 in females. The cumulative risk was 1.31% by age 50 and 3.68% by age 70 years, counting for a an eight-fold risk (95% CI 4.7–15.7) compared to the general population.
In studies by Wilentz *et al.* (41), Banville *et al.* (42) and Goggins *et al.* (43), it was suggested that PCs in Lynch syndrome have a distinctive medullary appearance, similar to medullary carcinomas of the colon. This could signify an indication for microsatellite instability (MSI) testing in the rarely occurring medullary variant of PC.

Li–Fraumeni Syndrome
Li–fraumeni syndrome (LFS; OMIM #151623) is another highly penetrant autosomal dominant cancer syndrome. Clinical criteria of LFS are a proband with sarcoma diagnosed under the age of 45 years, a first-degree relative with any cancer under 45 years and another first- or second-degree relative with either cancer under 45 years or a sarcoma at any age (44). Patients with this syndrome are at increased risk of multiple primary tumours. Breast cancer, sarcomas, brain tumours, leukaemia and adrenal cortical cancer are the most frequent associated malignancies, but a much broader tumour spectrum is reported (45). In most cases, LFS is caused by a germline mutation in *TP53*.
Birch *et al.* (46) showed six PCs in 28 *TP53* positive families and regarded PC, after exclusion of the strongly LFS-associated cancers as BC and sarcomas from the analyses, as a moderately associated cancer (p=0.007). Ruijs *et al.* (47) found in four of 24 Dutch *TP53* positive LFS-families four PCs at ages 41, 45, 49 and 52 years, indicating a RR of 7.5.

Familial pancreatic cancer
In the majority of families (80%) with a strong family history of PC, the disease is apparently unrelated to any currently recognized hereditary syndrome and these families are therefore referred to as FPC family. A strong family history is defined as PC in either ≥2 first-degree relatives (FDR), ≥3 relatives or two relatives of whom one being <50 years at time of diagnosis (3). The 'classical' phenotype of FPC-families (with PC in subsequent generations and affecting both male and female family members) suggests an autosomal dominant inheritance of the disease with variable penetrance. However, at present, the major gene(s) involved in the development of PC in FPC kindreds is/are unknown. Germline

BRCA2 mutations seem to cause a fraction of the familial clustering of PC, the prevalence of mutations in this gene was between 2.8 and 17% in non-syndromic FPC-families (48-50). A recently discovered gene, *PALB2*, seems to be responsible for the development of PC in a small fraction of the FPC kindreds (51). Furthermore, *CDKN2A*-mutations have been reported in families with clustering of PC and not meeting the diagnostic criteria of FAMMM (52). Previous studies have shown that the risk of PC is significantly increased in FPC kindreds and that this risk rises with increasing numbers of affected FDRs; individuals with one affected FDR have a 4.6-fold increased risk, with two affected FDRs a 6.4-fold increased risk and with three affected FDRs a 32-fold increased risk compared to the general population (53).

Why is it important that a clinician is aware of the existence of familial/hereditary pancreatic cancer?

First, it bears clinical relevance for family members of PC patients. They may have an increased risk of the development of PC but also for other tumour types in case PC developed within the framework of an inherited cancer syndrome. Referral to genetic services for clinical genetic counselling and DNA testing will be helpful in analysing their personalised cancer risks and advise them about screening or preventive options; e.g. dermatological surveillance in *CDKN2A* mutation carriers or BC screening in *BRCA1/2* mutation carriers. Furthermore, these individuals must be made aware of the fact that the cessation of smoking is critical since it is well known that cigarette smoking doubles the risk of PC development (54). Because of their high baseline risk of developing PC, the cessation of smoking in these individuals is even more important than in the general population. Second, future developments may show that PC patients with *BRCA1/2* mutations benefit from targeted therapy (e.g. PARP-inhibitors) and/or high dose chemotherapy, since results of first trials with these therapeutical options are promising in patients with *BRCA1/2* associated BC and other cancers (55-57).

Which characteristics raise suspision of familial or hereditary pancreatic cancer?

Family and patient characteristics that should raise suspicion of familial/hereditary PC are: PC in two or more family members; PC at a young age; and PC among other primary malignancies in one individual. In addition, PC in combination with certain other tumour types in a family should raise suspicion of a hereditary tumour syndrome, especially combinations of PC with melanoma, with BC and with colorectal cancer, diagnosed in one patient or in members from the same family, may point towards a shared genetic factor and is reason for referral to a clinical genetics centre.

Pancreatic cancer surveillance among high-risk individuals

Surveillance of individuals at high risk of PC is emerging. Currently, several studies are being conducted to evaluate the effectiveness of PC surveillance. Although the first results

are hopeful, the effectiveness still needs to be proven. Therefore, PC surveillance should only be performed in a research setting and offered to those who carry a substantial (e.g. 10-fold) increased risk of developing PC (3). It is currently estimated that HP patients, patients with PJS, carriers of *CDKN2A* mutations and FDR of FPC patients carry such risk. However, as mentioned before, examination of the pancreas is difficult in HP patients because of parenchymal changes due to chronic inflammation. For families with germline mutations in *BRCA1/2*, mismatch repair genes or in *TP53*, we presume a LTR>10% for PC only in those families with two or more carriers affected with PC. It is worth mentioning that the estimates of PC risk are based on results of either large collaborative studies evaluating heterogeneous groups of patients or small cohort studies leading to a wide variety in the reported cancer risk. Because of these limitations it is difficult to get a reliable risk estimate for PC development for these inherited syndromes. In addition, PC risks are derived from families that have been preselected by the presence of tumours that are typical for that particular syndrome, e.g. BC and OC in HBOC syndrome, melanoma in FAMMM syndrome and colorectal cancer in Lynch syndrome. It is not known to what extent the involved genes play a role in what is now referred to as sporadic or familial PC and in combinations of PC with other tumour types.

CONCLUSION

Pancreatic cancer is one of the most fatal human malignancies, with an incidence rate that nearly equals mortality rate. Recent advances in the field of radiology, surgery and (neo)adjuvant therapy have not led to a significant improvement in survival. Surveillance in order to detect precursor lesions or early cancers emerges as a potentially realistic opportunity to fight this devastating disease and hopefully lower mortality rates, especially when offered to a selected group of individuals that carry a significantly increased risk. In the past decades, our knowledge about the level of PC risk for the different syndromes has increased substantially, but for the majority of syndromes the associated PC risk has not yet been firmly established. The key focus of research in the coming years, therefore, should be directed towards a more precise PC risk assessment of the various syndromes in larger patient series as well as identifying the gene(s) involved in the development of FPC. Such knowledge would be most helpful in better identifying target populations that may benefit most from PC surveillance.

REFERENCES

1. Ferlay J, Parkin DM, Steliarova-Foucher E, Estimates of cancer incidence and mortality in Europe in 2008, Eur J Cancer, 2010;46(4):765–81.
2. Raimondi S, Maisonneuve P, Lowenfels AB, Epidemiology of pancreatic cancer: an overview, Nat Rev Gastroenterol Hepatol, 2009;6(12):699–708.
3. Brand RE, Lerch MM, Rubinstein WS, et al., Advances in counselling and surveillance of patients at risk for pancreatic cancer, Gut, 2007;56(10):1460–9.
4. Rebours V, Boutron-Ruault MC, Schnee M, et al., The natural history of hereditary pancreatitis: a national series, Gut, 2009;58(1):97–103.
5. Rebours V, Boutron-Ruault Mc, Schnee M, et al., Risk of pancreatic adenocarcinoma in patients with hereditary pancreatitis: a national exhaustive series, Am J Gastroenterol, 2008;103(1):111–9.
6. Howes N, Lerch MM, Greenhalf W, et al., Clinical and genetic characteristics of hereditary pancreatitis in europe, Clin Gastroenterol Hepatol, 2004;2(3):252–61.
7. Lowenfels AB, Maisonneuve P, DiMagno EP, et al., Hereditary pancreatitis and the risk of pancreatic cancer. International Hereditary Pancreatitis study group, J Natl Cancer Inst, 1997;89(6):442–6.
8. Lowenfels AB, Maisonneuve P, Whitcomb DC, et al., Cigarette smoking as a risk factor for pancreatic cancer in patients with hereditary pancreatitis, JAMA, 2001;286(2): 169–70.
9. Rebours V, Boutron-Ruault MC, Jooste V, et al., Mortality rate and risk factors in patients with hereditary pancreatitis: uni- and multi-dimensional analyses, Am J Gastroenterol, 2009;104(9):2312–7.
10. Finan MC, Ray MK, Gastrointestinal polyposis syndromes, Dermatol Clin, 1989;7(3):419–34.
11. Hemminki A, The molecular basis and clinical aspects of Peutz-Jeghers syndrome, Cell Mol Life Sci, 1999;55(5):735–50.
12. Giardiello FM, Welsh SB, Hamilton SR, et al., Increased risk of cancer in the Peutz-Jeghers syndrome, N Engl J Med, 1987;316(24):1511–4.
13. Aretz S, Stienen D, Uhlhaas S, et al., High proportion of large genomic sTK11 deletions in Peutz-Jeghers syndrome, Hum Mutat, 2005;26(6):513–9.
14. Volikos E, Robinson J, Aittomaki K, et al., LKB1 exonic and whole gene deletions are a common cause of Peutz-Jeghers syndrome, J Med Gen, 2006;43(5):e18.
15. van Lier MG, Westerman AM, Wagner A, et al., High cancer risk and increased mortality in patients with Peutz-Jeghers syndrome, Gut, 2011;60(2):141–7.
16. Giardiello FM, Brensinger JD, Tersmette AC, et al., Very high risk of cancer in familial Peutz-Jeghers syndrome, Gastroenterology, 2000;119(6):1447–53.
17. Vitone LJ, Greenhalf W, Mcfaul CD, et al., The inherited genetics of pancreatic cancer and prospects for secondary screening, Best Pract Res Clin Gastroenterol, 2006;20(2):253–83.

18. Hearle N, Schumacher V, Menko FH, et al., Frequency and spectrum of cancers in the Peutz-Jeghers syndrome, Clin Cancer Res, 2006;12(10):3209–15.
19. Mehenni H, Resta N, Park JG, et al., Cancer risks in LKB1 germline mutation carriers, Gut, 2006;55(7):984–90.
20. Leachman SA, Carucci J, Kohlmann W, et al., Selection criteria for genetic assessment of patients with familial melanoma, J Am Acad Dermatol, 2009;61(4):677 e1–14.
21. Gruis NA, Sandkuijl LA, van der velden PA, et al., CDKN2 explains part of the clinical phenotype in Dutch familial atypical multiple-mole melanoma (FAMMM) syndrome families, Melanoma Res, 1995;5(3):169–77.
22. Bishop DT, Demenais F, Goldstein AM, et al., Geographical variation in the penetrance of CDKN2A mutations for melanoma, J Natl Cancer Inst, 2002;94(12):894–903.
23. Borg A, Sandberg T, Nilsson K, et al., High frequency of multiple melanomas and breast and pancreas carcinomas in CDKN2A mutation-positive melanoma families, J Natl Cancer Inst, 2000;92(15):1260–6.
24. de Snoo FA, Bishop DT, Bergman W, et al., Increased risk of cancer other than melanoma in CDKN2A founder mutation (p16-Leiden)-positive melanoma families, Clin Cancer Res, 2008;14(21):7151–7.
25. Goldstein AM, Chan M, Harland M, et al., High-risk melanoma susceptibility genes and pancreatic cancer, neural system tumors, and uveal melanoma across GenoMEL, Cancer Res, 2006;66(20):9818–28.
26. Goldstein AM, Struewing JP, Fraser MC, et al., Prospective risk of cancer in CDKN2A germline mutation carriers, J Med Gen, 2004;41(6):421–4.
27. Antoniou A, Pharoah PD, Narod S, et al., Average risks of breast and ovarian cancer associated with BRCA1 or BRCA2 mutations detected in case series unselected for family history: a combined analysis of 22 studies, Am J Hum Genet, 2003;72(5): 1117–30.
28. Friedenson B, BRCA1 and BRCA2 pathways and the risk of cancers other than breast or ovarian, MedGenMed, 2005;7(2):60.
29. Greer JB, Whitcomb DC, Role of BRCA1 and BRCA2 mutations in pancreatic cancer, Gut, 2007;56(5):601–5.
30. Jakubowska A, Scott R, Menkiszak J, et al., A high frequency of BRCA2 gene mutations in Polish families with ovarian and stomach cancer, Eur J Hum Genet, 2003;11(12): 955–8.
31. Lubinski J, Phelan CM, Ghadirian P, et al., Cancer variation associated with the position of the mutation in the BRCA2 gene, Fam Cancer, 2004;3(1):1–10.
32. Mohamad HB, Apffelstaedt JP, Counseling for male BRCA mutation carriers: a review, Breast, 2008;17(5):441–50.
33. Al-sukhni W, Rothenmund H, Borgida AE, et al., Germline BRCA1 mutations predispose to pancreatic adenocarcinoma, Hum Genet, 2008;124(3):271–8.

34. Skoulidis F, Cassidy LD, Pisupati V, et al., Germline BRCA2 heterozygosity promotes Kras(g12D)-driven carcinogenesis in a murine model of familial pancreatic cancer, Cancer Cell, 2010;18(5):499–509.
35. Cancer risks in BRCA2 mutation carriers. The Breast cancer Linkage consortium, J Natl Cancer Inst, 1999;91(15):1310–6.
36. Thompson D, Easton DF, Cancer incidence in BRCA1 mutation carriers, J Natl Cancer Inst, 2002;94(18):1358–65.
37. van Asperen CJ, Brohet RM, Meijers-Heijboer EJ, et al., Netherlands collaborative group on hereditary Breast cancer (HEBON), cancer risks in BRCA2 families: estimates for sites other than breast and ovary, J Med Genet, 2005;42(9):711–9.
38. Risch HA, McLaughlin JR, Cole DE, et al., Prevalence and penetrance of germline BRCA1 and BRCA2 mutations in a population series of 649 women with ovarian cancer, Am J Hum Genet, 2001;68(3):700–10.
39. Kim DH, Crawford B, Ziegler J, Beattie MS, Prevalence and characteristics of pancreatic cancer in families with BRCA1 and BRCA2 mutations, Fam Cancer, 2009;8(2):153–8.
40. Kastrinos F, Mukherjee B, Tayob N, et al., Risk of pancreatic cancer in families with Lynch syndrome, JAMA, 2009;302(16):1790–5.
41. Wilentz RE, Goggins M, Redston M, et al., Genetic, immunohistochemical, and clinical features of medullary carcinoma of the pancreas: a newly described and characterized entity, Am J Pathol, 2000;156(5):1641–51.
42. Banville N, Geraghty R, Fox E, et al., Medullary carcinoma of the pancreas in a man with hereditary nonpolyposis colorectal cancer due to a mutation of the MSH2 mismatch repair gene, Hum Pathol, 2006;37(11):1498–502.
43. Goggins M, Offerhaus GJ, Hilgers W, et al., Pancreatic adenocarcinomas with DNA replication errors (rer+) are associated with wild-type K-ras and characteristic histopathology. Poor differentiation, a syncytial growth pattern, and pushing borders suggest rer+, Am J Pathol, 1998;152(6):1501–7.
44. Li FP, Fraumeni JF, Jr., Mulvihill JJ, et al., A cancer family syndrome in twenty-four kindreds, Cancer Res, 1988;48(18):5358–62.
45. Petitjean A, Mathe E, Kato S, et al., Impact of mutant p53 functional properties on TP53 mutation patterns and tumor phenotype: lessons from recent developments in the IARC TP53 database, Hum Mutat, 2007;28(6):622–9.
46. Birch JM, Alston RD, Mcnally RJ, et al., Relative frequencyand morphology of cancers in carriers of germline TP53 mutations, Oncogene, 2001;20(34):4621–8.
47. Ruijs MW, Verhoef S, Rookus MA, et al., TP53 germline mutation testing in 180 families suspected of Li-fraumeni syndrome: mutation detection rate and relative frequency of cancers in different familial phenotypes, J Med Genet, 2010;47(6):421–8.
48. Couch FJ, Johnson MR, Rabe KG, et al., The prevalence of BRCA2 mutations in familial pancreatic cancer, Cancer Epidemiol Biomarkers Prev, 2007;16(2):342–6.

49. Hahn SA, Greenhalf B, Ellis I, et al., BRCA2 germline mutations in familial pancreatic carcinoma, J Natl Cancer Inst, 2003;95(3):214–21.
50. Murphy KM, Brune KA, Griffin C, et al., Evaluation of candidate genes MAP2K4, MADH4, ACVR1B, and BRCA2 in familial pancreatic cancer: deleterious BRCA2 mutations in 17%, Cancer Res, 2002;62(13):3789–93.
51. Jones S, Hruban RH, Kamiyama M, et al., Exomic sequencing identifies PALB2 as a pancreatic cancer susceptibility gene, Science, 2009;324(5924):217.
52. Bartsch DK, Langer P, Habbe N, et al., Clinical and genetic analysis of 18 pancreatic carcinoma/melanoma-prone families, Clin Genet, 2010;77(4):333–41.
53. Klein AP, Brune KA, Petersen GM, et al., Prospective risk of pancreatic cancer in familial pancreatic cancer kindreds, Cancer Res, 2004;64(7):2634–8.
54. Gold EB, Goldin SB, Epidemiology of and risk factors for pancreatic cancer, Surg Oncol Clin N Am, 1998;7(1):67–91.
55. Fong PC, Boss DS, Yap TA, et al., Inhibition of poly(ADP-ribose) polymerase in tumors from BRCA mutation carriers, N Engl J Med, 2009;361(2):123–34.
56. Tutt A, Robson M, Garber JE, et al., Oral poly(ADP-ribose) polymerase inhibitor olaparib in patients with BRCA1 or BRCA2 mutations and advanced breast cancer: a proof-of-concept trial, Lancet, 2010;376(9737):235–44.
57. Vollebergh MA, Lips EH, Nederlof PM, et al., An aCGH classifier derived from BRCA1-mutated breast cancer and benefit of high-dose platinum-based chemotherapy in HER2-negative breast cancer patients, Ann Oncol, 2011;22(7);1561–70.

F. Harinck, I. Kluijt, N. van der Stoep, R. Oldenburg, A. Wagner, C.M. Aalfs, R.H. Sijmons, J-W. Poley, E.J. Kuipers, P. Fockens, T.A.M. van Os, M.J. Bruno, on behalf of the Dutch research group of pancreatic cancer surveillance in high-risk individuals

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CHAPTER 8
INDICATION FOR *CDKN2A*-MUTATION ANALYSIS IN FAMILIAL PANCREATIC CANCER FAMILIES WITHOUT MELANOMAS

ABSTRACT

Background | *CDKN2A*-mutation carriers run a high risk of developing melanomas and have an increased risk of developing pancreatic cancer (PC). Familial PC (FPC) patients with a personal history or family history of melanomas are therefore offered *CDKN2A*-mutation analysis. In contrast, *CDKN2A* testing in FPC families without a history of melanomas is not generally recommended. The aim of this study was to evaluate the frequency of *CDKN2A*-mutations in FPC families without melanomas.

Methods | Data were gathered from PC family registers. FPC families were defined as families with clustering of PC without meeting diagnostic criteria of familial cutaneous malignant melanoma (familial CMM) or other inherited cancer syndromes. Blood samples were obtained for DNA isolation from PC patients or first degree relatives and analysed for *CDKN2A*-mutations.

Results | Among 40 FPC families, DNA analyses were carried out in 28 families (70%), leading to identification of *CDKN2A*-mutations in six families (21%). None of the *CDKN2A*-mutation-positive families fulfilled the diagnostic criteria for familial CMM and in three *CDKN2A* families no melanomas were observed. Two *CDKN2A*-mutations were found; the Dutch founder mutation p16-Leiden (c.225_243del, p.Ala76fs) and the c.19_23dup, p.Ser8fs-mutation. After disclosure of the *CDKN2A*-mutation in one of the families, a curable melanoma was diagnosed at dermatological surveillance in a 17-year-old family member.

Conclusion | *CDKN2A*-mutation can be found in a considerable proportion of families with FPC. *CDKN2A*-mutation analysis should therefore be included in genetic testing in FPC families, even in the absence of reported melanomas. This strategy will enhance the recognition of individuals at risk for PC and facilitate the early detection of melanomas.

INTRODUCTION

Approximately 10% of all pancreatic cancer (PC) cases occur in a background of familial clustering (1). In about 20% of these cases the underlying gene mutation is recognized (1). One such inherited cancer syndrome with a known increased risk for PC is familial cutaneous malignant melanoma (familial CMM), referred to in the past as the familial atypical multiple mole melanoma syndrome (OMIM 155600) (2-5).
This syndrome is characterised by the familial occurrence of melanomas (5) and inherits as an autosomal dominant trait. Germ-line mutations in *CDKN2A* have been found in at least a quarter of all melanoma prone families (6-7).
In addition to an increased risk of developing melanomas, *CDKN2A*-mutation carriers are also at risk of other types of cancer, particularly PC (2-4). Previous studies have shown that the risk of developing PC among *CDKN2A* carriers may be 50 times greater than in the general population (3).
Therefore, families with any combination of PC and melanomas should be offered *CDKN2A* analysis (8-14). However, *CDKN2A* analysis is not recommended in families with a clustering of PC but without melanomas (10).
The aim of this study was to evaluate the frequency of *CDKN2A*-mutations in familial PC (FPC) families without melanomas.

MATERIALS AND METHODS

Patients and families

We conducted a retrospective cohort study. Data were gathered from PC family registries from four Dutch clinical genetic centres (Academic Medical Center Amsterdam, Erasmus MC-University Medical Center Rotterdam, University Medical Center Groningen and the Netherland Cancer Institute-Antoni van Leeuwenhoek Hospital). The total number of PC families was 70. Based on their phenotype, PC families were divided into FPC families (n=40) and syndromic PC families (n=30).
FPC families were defined as families with clustering of PC (≥2 first degree relatives (FDR), ≥3 relatives (FDR or second degree relatives (SDR)) or two SDR relatives, one <50 years at diagnosis) and not meeting diagnostic criteria of known inherited cancer syndromes (listed below) (10). Syndromic PC families were defined as families with a known inherited cancer syndrome predisposing them to PC (including familial CMM, Peutz-Jeghers, Lynch, Li-Fraumeni and hereditary breast and ovarian cancer syndromes). The diagnosis of familial CMM was made based on the Dutch clinical criteria of familial atypical multiple mole melanomas in either ≥2 affected FDR or ≥3 affected relatives (FDR and/or SDR) (15). The total number of familial CMM-families within the cohort of syndromic PC families was 16 (53%).
In this study, we included only the FPC families and analysed whether DNA-mutation analysis for *CDKN2A/CDK4* was performed, and if so what the outcome of this mutation

analysis was. DNA, either from blood samples or available paraffin embedded tumour samples, originated from individuals affected with PC or, in families without available DNA from affected individuals, from healthy FDRs. For each family, a complete three-generation pedigree was made. Clinical diagnoses reported by patients and family members were verified by a review of medical and pathological records, and by revision of histological slides whenever available. At the time of genetic counselling, patients or their family members had given written informed consent to use the DNA results for future research projects.
All individuals from *CDKN2A*-mutation-positive families were advised to undergo dermatological examination to detect dysplastic nevi and/or melanoma.

***CDKN2A/CDK4*-mutation analysis**
Direct sequencing of all *CDKN2A* exons and *CDK4* exon 2 was performed on samples from the index cases, and subsequently on relatives of mutation-positive cases. In brief, all exons with flanking intronic regions were amplified by PCR using the following primers: CDKN2A-EX1BF 5-GTGCGTGGGTCCCA GTCT-3', CDKN2A-EX1BR 5'-TAGCCTGGGCTAGAGACG AA-3' (Ta=57◦C), CDKN2A-EX1AF 5'-TTCGCTAAGTGCTC GGAGTT-3', CDKN2A-EX1AR 5'-GAGAATCGAAGCGCTACCT- 3' (Ta=57◦C), CDKN2A-EX2F 5'-GGAAATTGGAAACTGGAAGC-3', CDKN2A-EX2R 5'-GCTGAACTTTCTGTGCTGGAAAAATG-3'(Ta=55◦C), CDKN2A-EX3F 5'-GCAGTGGACTAGCTGCTGGA- 3', CDKN2A-EX3R 5'-TTTACGGTAGTGGGGGAAGG-39 (Ta=57◦C), and CDK4-EX2F 5'-TTGTTGCTGCAGGCTCATAC-3', CDK4-EX2R 5'-TCAGGGTCCCCACTTCTCTA 3'(Ta=57◦C). All primers were flanked with respectively m13forward or reverse tags to allow direct sequencing. PCR reactions were carried out using GoTaq® DNA Polymerase (Promega, Benelux b.v.) based on the standard protocol at annealing temperature (Ta) as indicated at the primers. Subsequently, the sequence PCR products were analysed on an ABI3730 sequencer using BigDye® Terminator v3.1 Cycle Sequencing Kit (Applied Biosystems, Foster City, California, USA) and genotypes were assigned using SeqScape software (Applied Biosystems). The reference sequences for *CDKN2A* and for *CDK4* respectively with GenBank accession numbers NT_008413 v17 and NC_000012 v10 were used to analyse the sequence results and all detected variants were described according to the HGVS nomenclature recommendations.
CNV analysis of *CDKN2A* was performed for all samples by Multiplex Ligation-dependent Probe Amplification using the MRC-Holland probe-mixME024-A1 (MRC-Holland, Amsterdam, Netherlands) as indicated by the manufacturer. CNV data for the *CDKN2A* region specific probes were analysed using GeneMarker software package (SoftGenetics, Pennsylvania, USA).

Data analyses
Continuous variables are presented as mean (SD) or median (IQR), where appropriate. Continuous variables were compared using the t test or the ManneWhitney test.

Categorical variables were compared using the c2 or Fisher's exact test. All analyses were conducted using the Statistical Package for the Social Sciences (V.17.0; SPSS Institute). A two-sided p value <0.05 was considered to be statistically significant.

RESULTS

In our series of 40 FPC families, DNA analyses were carried out in 28 families (70%). Of the remaining 12 families, DNA was not available. Twenty-seven of the 28 analysed families (96%) were of Caucasian descent and one family (3.6%) was of Indonesian descent (Maluku Islands). These 28 FPC families had a total of 74 affected patients with PC. In 14 families (50%) two family members were diagnosed with PC, in 12 families (43%) three family members were diagnosed with PC, in one family (3.6%) four family members were diagnosed with PC and in one family (3.6%) seven family members were diagnosed with PC. Of the 74 PC cases, 41 (55%) were male subjects. The mean age of diagnosis of PC was 59.0 years (range 30-84 years, SD 12.3). Nine patients (12%) were younger than 45 at the time of diagnosis. In addition to the PC cases, 24 families (86%) were affected by other types of cancer. Four families (14%) were affected by melanomas, and nine families (32%) were affected by breast cancer.

In 21 FPC families (75%), DNA was available from affected PC cases, isolated from blood samples in 19 families and from PC tumour tissue in two families. In the remaining families, DNA analyses were carried out in DNA of healthy FDR (six families; mean number of FDR tested 1.7, range 1-3) or suspected carriers (one family) because of their position in the pedigree.

DNA analyses of mutations in *CDKN2A/CDK4* led to the identification of a causal genetic factor in six (21.4%) FPC families. In three (50%) of these *CDKN2A*-mutation-positive FPC families, no melanomas and/or dysplastic nevi had been reported at the time of DNA analyses. In the other three *CDKN2A*-mutation-positive FPC families, two had one family member diagnosed with melanoma and in the third family two second degree members were diagnosed with melanoma.

Two different *CDKN2A*-mutations were found, the Dutch founder mutation p16-Leiden (c.225_243del, p.Ala76fs) and the c.19_23dup.p.Ser8fs-mutation. The p16-Leiden-mutation was found in all three melanoma-positive (100%) families and in two of the melanoma-negative (67%) families.

Table 1A shows the characteristics of the *CDKN2A*-mutationpositive FPC families without melanomas; supplementary figures A to C display the pedigrees. These three families included a total of eight PC cases of whom five were male subjects (63%) and the mean age at the time of diagnosis was 51.5 years (SD 9.2). Two patients (25%) were younger than 45 years at the time of diagnosis. The Dutch founder mutation was found in two Caucasian families. The c.19_23dup.p.Ser8fsmutation was found in the family of Indonesian descent. The *CDKN2A*-mutation-positive FPC families without melanoma (Table 1A) and with

melanomas (Table 1B) included a total of 19 PC cases of whom 10 were male subjects (53%) and the mean age at the time of diagnosis was 55.7 (SD 10.2). These characteristics were not statistically significantly different from the PC cases of the other FPC families: mean age at the time of diagnosis was 60.1 (SD 12.8), 56% were male persons.

A	No. family members with PC/ Gender & Age at diagnosis (years)	No. family members with melanoma	Other tumor types*	Type of mutation
1.	3/♂48, ♂55, ♂67 (Figure 1A)	0	Lung cancer n=1	c.19_23dup.p.Ser8fs
2.	3/♂39, ♀41, ♀52 (Figure 1B)	0	Cancer unkown origin n=2	c.225_243del, p.Ala76fs
3.	2/♀51, ♂59 (Figure 1C)	0¶	Basal cell carcinoma n=1	c.225_243del, p.Ala76fs
B				
4.	2/♂50, ♀67	1	Pharyngeal cancer n=1± Gastric cancer n=1±	c.225_243del, p.Ala76fs
5.	2/♂62, ♂73	1	No	c.225_243del, p.Ala76fs
6.	7/♀50, ♂50, ♂51, ♀59, ♀65, ♀74, ♀76	2	Prostate cancer n=1 Thyroid cancer n=1	c.225_243del, p.Ala76fs

Table 1. Characteristics of CDKN2A-positive FPC families without melanomas (A) and CDKN2A-positive FPC-families with melanomas not fulfilling diagnostic criteria of familial-CMM (B).

**Other associated tumor types in family*

¶After disclosure of the CDKN2A-mutation in this family, a melanoma was diagnosed at dermatological surveillance in a 17-year old female family member

±Same patient

DISCUSSION

In six out of 28 FPC families tested, we identified a *CDKN2A*-mutation (21%), in three of whom (50%) no melanomas and/or dysplastic nevi had been reported at the time of DNA analysis. If the current recommendation would have been followed to test only for *CDKN2A*-mutations in FPC families with at least one melanoma case, these three families would have gone unnoticed (10). This recommendation is based on three previous studies on the role of *CDKN2A*-mutations in FPC families, which failed to identify *CDKN2A*-mutations in non-melanoma FPC families (9, 12, 13) (Table 2). Recently, the prevalence of *CDKN2A*-mutations in a large series of unselected PC cases was studied and this turned out to be low (0.6%) (16). A subanalysis of this series in which only FPC cases were included showed *CDKN2A*-mutations in 3.3% of cases. The discrepancy between these and our series is not readily explained, although the method of patient selection between our series (gathered from PC family registries) and the latter large series (unselected PC cases) may account for part of the discrepancy. It does not seem likely that the differences in results can be explained by the type of *CDKN2A*-mutations that we found. The Dutch founder mutation p16-Leiden, which two of our *CDKN2A*-mutaion-positive non-melanoma FPC families carried, is a well-researched mutation; the phenotype of its carriers is not restricted to clustering of PC only (3). Although, a part of the differences might be explained by the founder role of this mutation. The third *CDKN2A*-mutation-positive non-melanoma

Study	Study population	*CDKN2A* mutation found	Type(s) of mutation	Phenotype
Slater *et al.* (2010)[13]	56 FPC families	No	-	-
Bartsch *et al.* (2002)[9]	18 FPC families	No	-	-
Moskaluk *et al.* (1998)[12]	21 FPC families	Yes	c.457 G>T	Mel-PC family¶
Current study	28 FPC-families	Yes	c.19_23dup.p.Ser8fs	FPC family
			c.225_243del,p.Ala76fs	FPC family
			c.225_243del,p.Ala76fs	FPC- family¥
			c.225_243del,p.Ala76fs	Mel-PC family¶
			c.225_243del,p.Ala76fs	Mel-PC family¶
			c.225_243del,p.Ala76fs	Mel-PC family¶

Table 2. Overview of literature on role CDKN2A-mutations in FPC

FPC family carried a c.19_23dup. p.Ser8fs-mutation. To our knowledge, this is a new mutation that has not been previously reported. The frame shift that is caused by this mutation causes a stop-codon. Based on the results of this one family, it is too early to claim that this specific mutation causes a phenotype without melanomas. The family in whom the c.19_23dup.p.Ser8fs-mutation was detected is of Indonesian descent and their darker-skinned complexion might have offered them added protection from developing melanomas. It is well known that the risk of developing melanoma is higher in fair-skinned people, especially those with blond or red hair and who sunburn and freckle easily than in people with darker complexions (17). The two families with the Dutch founder mutation were Caucasian.

Differences in lifestyle may also explain some of the difference between the results of earlier studies and our series. For example, it is well known that cigarette smoking is associated with an increased risk of PC (18). Unfortunately, we lack detailed information about smoking status. We therefore cannot exclude the possibility that in our series the PC affected family members of non-melanoma *CDKN2A*-mutation-positive families smoked more than affected individuals in the previously published studies which also did not report on smoking status.

Another limitation of this current series is that in some families affected relatives were unavailable for DNA testing and, instead, unaffected FDRs were tested. A negative genetic test result in such cases does not exclude the presence of a pathogenetic mutation unless a specific genetic mutation is been found in another relative. This may have caused an underestimation of the prevalence of *CDKN2A/CDK4* in this current study.

We were able to collect detailed information on the family history of all FPC families in whom DNA analyses were carried out. In addition, patients not known with any skin lesions before they were genetically tested were seen by a dermatologist, minimizing the chance that we missed a diagnosis of melanoma or dysplastic nevi negligible. Since in both family B and family C some family members died of cancer of unknown origin, we cannot state with 100% certainty that none of these individuals died of melanoma.

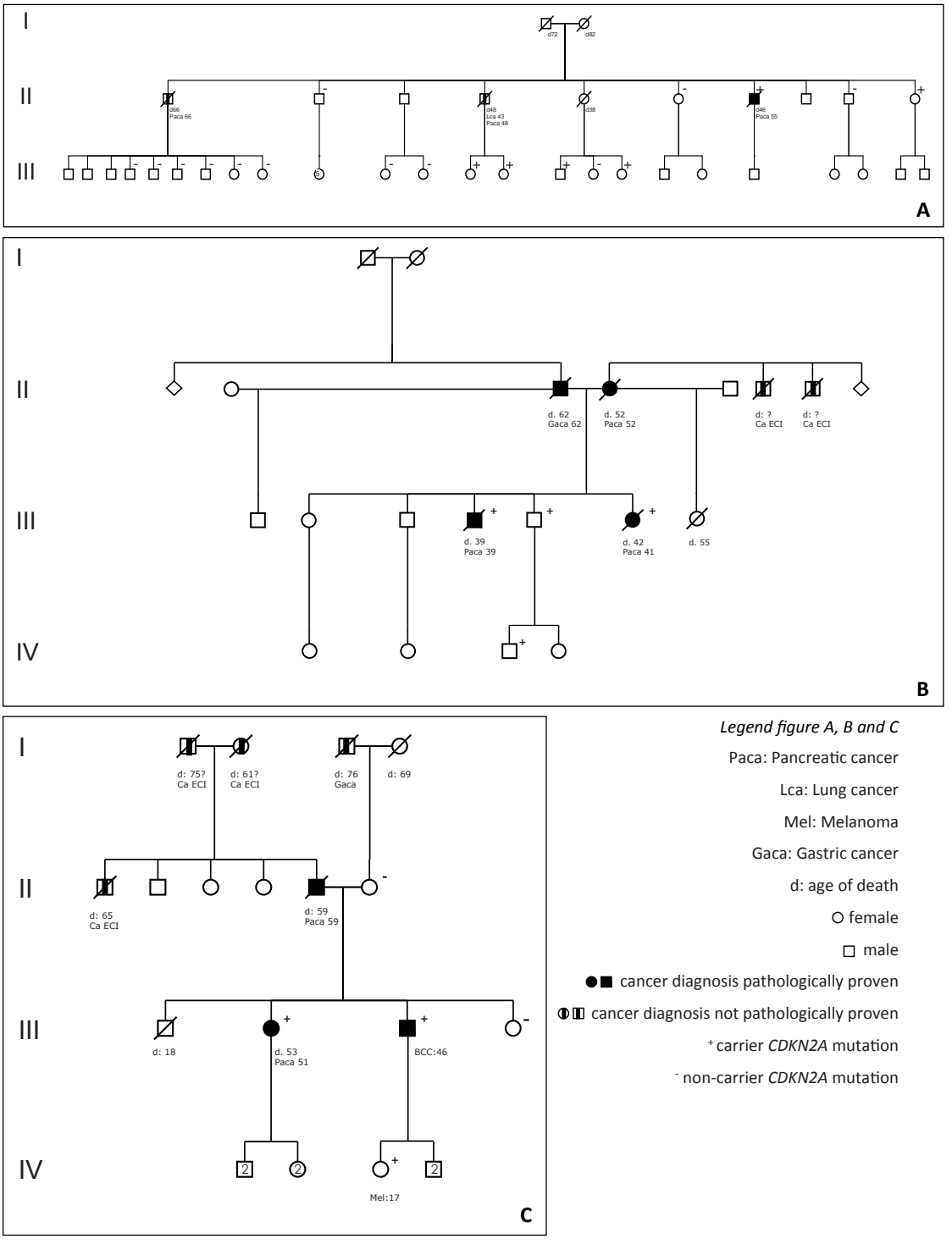
The three non-melanoma FPC families in which a *CDKN2A*-mutation was found were of moderate to large size. It is therefore less likely that melanoma or dysplastic nevi were not observed because of a low a priori change based on numbers and RR. Interestingly,

during dermatological follow-up of one of our *CDKN2A*-positive FPC families we detected an early stage melanoma in a 17-year-old female family member. A year after resection of a 5-mm superficial spreading melanoma (Breslow 0.8 mm), this patient is disease-free. Among the 28 families in which *CDKN2A*-mutation was performed, four families (14%) were affected by ≥1 melanoma (s). It is of interest that in three (75%) of these melanomapositive families, a mutation in *CDKN2A* was found. These findings are in line with previous reports showing that *CDKN2A*-mutations are frequently found in families affected by both PC and melanomas (8, 9). The prevalence of *CDKN2A*-mutations in the remaining melanoma-negative FPC families was 12%

A number of previous reports have shown an indication for *BRCA2*-mutation analysis in FPC families that did not meet the criteria of familial breast and ovarian cancer (19-21). In a similar way, our findings emphasise the need to include *CDKN2A*-mutation analysis in genetic testing for FPC families, even in the absence of reported melanomas. It will help to better identity those at risk of developing PC and/or melanoma.

Surveillance of individuals at a high risk of malignant melanomas has proved to lead to early detection of melanomas and will consequently have a favourable effect in prognosis (22-24). Surveillance of individuals at high risk of PC is emerging and may lead to an improvement of prognosis and a decline in PC incidence (25-30).

In conclusion, the results of this series show that *CDKN2A*-mutation can be found in a considerable proportion of families with FPC. *CDKN2A*-mutation analysis should therefore be included in genetic testing in FPC families, even in the absence of reported melanomas. This strategy will enhance the recognition of individuals at risk for PC and facilitate the early detection of melanomas.



Legend figure A, B and C

Paca: Pancreatic cancer

Lca: Lung cancer

Mel: Melanoma

Gaca: Gastric cancer

d: age of death

○ female

□ male

●■ cancer diagnosis pathologically proven

◑◧ cancer diagnosis not pathologically proven

⁺ carrier *CDKN2A* mutation

⁻ non-carrier *CDKN2A* mutation

Figure 1. Pedigrees of CDKN2A positive families without melanomas.

REFERENCES

1. Lynch HT, Smyrk T, Kern SE, Hruban RH, Lightdale CJ, Lemon SJ, Lynch JF, Fusaro LR, Fusaro RM, Ghadirian P. Familial pancreatic cancer: a review. Semin Oncol 1996;23:251e75.
2. Goldstein AM, Chan M, Harland M, Gillanders EM, Hayward NK, Avril MF, Azizi E, Bianchi-Scarra G, Bishop DT, Bressac-de Paillerets B, Bruno W, Calista D, Cannon Albright LA, Demenais F, Elder DE, Ghiorzo P, Gruis NA, Hansson J, Hogg D, Holland EA, Kanetsky PA, Kefford RF, Landi MT, Lang J, Leachman SA, Mackie RM, Magnusson V, Mann GJ, Niendorf K, Newton Bishop J, Palmer JM, Puig S, Puig-Butille JA, de Snoo FA, Stark M, Tsao H, Tucker MA, Whitaker L, Yakobson E. Highrisk melanoma susceptibility genes and pancreatic cancer, neural system tumors, and uveal melanoma across GenoMEL. Cancer Res 2006;66:9818e28.
3. de Snoo FA, Bishop DT, Bergman W, van Leeuwen I, van der Drift C, van Nieuwpoort FA, Out-Luiting CJ, Vasen HF, ter Huurne JA, Frants RR, Willemze R, Breuning MH, Gruis NA. Increased risk of cancer other than melanoma in CDKN2A founder mutation (p16-Leiden)-positive melanoma families. Clin Cancer Res 2008;14:7151e7.
4. Borg A, Sandberg T, Nilsson K, Johannsson O, Klinker M, Masback A, Westerdahl J, Olsson H, Ingvar C. High frequency of multiple melanomas and breast and pancreas carcinomas in CDKN2A mutation-positive melanoma families. J Natl Cancer Inst 2000;92:1260e6.
5. Online Mendelian Inheritance in Man. Baltimore, MD: Johns Hopkins University. MIM Number: 155600. http://omim.org/ (accessed 7 Jul 2011).
6. Gruis NA, Sandkuijl LA, van der Velden PA, Bergman W, Frants RR. CDKN2 explains part of the clinical phenotype in Dutch familial atypical multiple-mole melanoma (FAMMM) syndrome families. Melanoma Res 1995;5:169e77.
7. Goldstein AM, Chan M, Harland M, Hayward NK, Demenais F, Bishop DT, Azizi E, Bergman W, Bianchi-Scarra G, Bruno W, Calista D, Albright LA, Chaudru V, Chompret A, Cuellar F, Elder DE, Ghiorzo P, Gillanders EM, Gruis NA, Hansson J, Hogg D, Holland EA, Kanetsky PA, Kefford RF, Landi MT, Lang J, Leachman SA, MacKie RM, Magnusson V, Mann GJ, Bishop JN, Palmer JM, Puig S, Puig-Butille JA, Stark M, Tsao H, Tucker MA, Whitaker L, Yakobson E; Lund Melanoma Study Group; Melanoma Genetics Consortium (GenoMEL). Features associated with germline CDKN2A mutations: a GenoMEL study of melanoma-prone families from three continents. J Med Genet 2007;44:99e106.
8. Bartsch DK, Langer P, Habbe N, Matthai E, Chaloupka B, Sina M, Hahn SA, Slater EP. Clinical and genetic analysis of 18 pancreatic carcinoma/melanoma-prone families. Clin Genet 2010;77:333e41.
9. Bartsch DK, Sina-Frey M, Lang S, Wild A, Gerdes B, Barth P, Kress R, Grutzmann R, Colombo-Benkmann M, Ziegler A, Hahn SA, Rothmund M, Rieder H. CDKN2A germline mutations in familial pancreatic cancer. Ann Surg 2002;236:730e7.

10. Brand RE, Lerch MM, Rubinstein WS, Neoptolemos JP, Whitcomb DC, Hruban RH, Brentnall TA, Lynch HT, Canto MI. Advances in counselling and surveillance of patients at risk for pancreatic cancer. Gut 2007;56:1460e9.
11. Lynch HT, Brand RE, Lynch JF, Fusaro RM, Smyrk TC, Goggins M, Kern SE. Genetic counseling and testing for germline p16 mutations in two pancreatic cancer-prone families. Gastroenterology 2000;119:1756e60.
12. Moskaluk CA, Hruban H, Lietman A, Smyrk T, Fusaro L, Fusaro R, Lynch J, Yeo CJ, Jackson CE, Lynch HT, Kern SE. Novel germline p16(INK4) allele (Asp145Cys) in a family with multiple pancreatic carcinomas. Mutations in brief no. 148. Online. Hum Mutat 1998;12:70.
13. Slater EP, Langer P, Fendrich V, Habbe N, Chaloupka B, Matthai E, Sina M, Hahn SA, Bartsch DK. Prevalence of BRCA2 and CDKN2a mutations in German familial pancreatic cancer families. Fam Cancer 2010;9:335e43.
14. Whelan AJ, Bartsch D, Goodfellow PJ. Brief report: a familial syndrome of pancreatic cancer and melanoma with a mutation in the CDKN2 tumor-suppressor gene. N Engl J Med 1995;333:975e7.
15. Oncoline Cancer Clinical Practice Guidelines. Vereniging van Integrale Kankercentra the Netherlands. http://oncoline.nl/fammm (accessed 1 Jul 2011).
16. McWilliams RR, Wieben ED, Rabe KG, Pedersen KS, Wu Y, Sicotte H, Petersen GM. Prevalence of CDKN2A mutations in pancreatic cancer patients: implications for genetic counseling. Eur J Hum Genet 2011;19:472e8.
17. Marrett LD, King WD, Walter SD, From L. Use of host factors to identify people at high risk for cutaneous malignant melanoma. CMAJ 1992;147:445e53.
18. Fuchs CS, Colditz GA, Stampfer MJ, Giovannucci EL, Hunter DJ, Rimm EB, Willett WC, Speizer FE. A prospective study of cigarette smoking and the risk of pancreatic cancer. Arch Intern Med 1996;156:2255e60.
19. Murphy KM, Brune KA, Griffin C, Sollenberger JE, Petersen GM, Bansal R, Hruban RH, Kern SE. Evaluation of candidate genes MAP2K4, MADH4, ACVR1B, and BRCA2 in familial pancreatic cancer: deleterious BRCA2 mutations in 17%. Cancer Res 2002;62:3789e93.
20. Hahn SA, Greenhalf B, Ellis I, Sina-Frey M, Rieder H, Korte B, Gerdes B, Kress R, Ziegler A, Raeburn JA, Campra D, Grutzmann R, Rehder H, Rothmund M, Schmiegel W, Neoptolemos JP, Bartsch DK. BRCA2 germline mutations in familial pancreatic carcinoma. J Natl Cancer Inst 2003;95:214e21.
21. Couch FJ, Johnson MR, Rabe KG, Brune K, de Andrade M, Goggins M, Rothenmund H, Gallinger S, Klein A, Petersen GM, Hruban RH. The prevalence of BRCA2 mutations in familial pancreatic cancer. Cancer Epidemiol Biomarkers Prev 2007;16:342e6.
22. Vasen HF, Bergman W, van Haeringen A, Scheffer E, van Slooten EA. The familial dysplastic nevus syndrome. Natural history and the impact of screening on prognosis. A study of nine families in the Netherlands. Eur J Cancer Clin Oncol 1989;25:337e41.

23. Rhodes AR, Weinstock MA, Fitzpatrick TB, Mihm MC Jr, Sober AJ. Risk factors for cutaneous melanoma. A practical method of recognizing predisposed individuals. JAMA 1987;258:3146e54.
24. Masri GD, Clark WH Jr, Guerry D 4th, Halpern A, Thompson CJ, Elder DE. Screening and surveillance of patients at high risk for malignant melanoma result in detection of earlier disease. J Am Acad Dermatol 1990;22:1042e8.
25. Canto MI, Goggins M, Hruban RH, Petersen GM, Giardiello FM, Yeo C, Fishman EK, Brune K, Axilbund J, Griffin C, Ali S, Richman J, Jagannath S, Kantsevoy SV, Kalloo AN. Screening for early pancreatic neoplasia in high-risk individuals: a prospective controlled study. Clin Gastroenterol Hepatol 2006;4:766e81; quiz 665.
26. Kimmey MB, Bronner MP, Byrd DR, Brentnall TA. Screening and surveillance for hereditary pancreatic cancer. Gastrointest Endosc 2002;56:S82e6.
27. Langer P, Kann PH, Fendrich V, Habbe N, Schneider M, Sina M, Slater EP, Heverhagen JT, Gress TM, Rothmund M, Bartsch DK. Five years of prospective screening of high-risk individuals from families with familial pancreatic cancer. Gut 2009;58:1410e18.
28. Poley JW, Kluijt I, Gouma DJ, Harinck F, Wagner A, Aalfs C, van Eijck CH, Cats A, Kuipers EJ, Nio Y, Fockens P, Bruno MJ. The yield of first-time endoscopic ultrasonography in screening individuals at a high risk of developing pancreatic cancer. Am J Gastroenterol 2009;104:2175e81.
29. Vasen HF, Wasser M, van Mil A, Tollenaar RA, Konstantinovski M, Gruis NA, Bergman W, Hes FJ, Hommes DW, Offerhaus GJ, Morreau H, Bonsing BA, de Vos tot Nederveen Cappel WH. Magnetic resonance imaging surveillance detects early-stage pancreatic cancer in carriers of a p16-Leiden mutation. Gastroenterology 2011;140:850e6.
30. Verna EC, Hwang C, Stevens PD, Rotterdam H, Stavropoulos SN, Sy CD, Prince MA, Chung WK, Fine RL, Chabot JA, Frucht H. Pancreatic cancer screening in a prospective cohort of high-risk patients: a comprehensive strategy of imaging and genetics. Clin Cancer Res 2010;16:5028e37.

F. Harinck, I. Kluijt, S.E. van Mil, Q. Waisfisz, T.A.M. van Os, C.M. Aalfs, A. Wagner, M. Olderode-Berends, R.H. Sijmons, E.J. Kuipers, J-W Poley, P. Fockens, M.J. Bruno, on behalf of the Dutch research group of pancreatic cancer surveillance in high-risk individuals

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CHAPTER 9
ROUTINE TESTING FOR *PALB2* MUTATIONS IN FAMILIAL PANCREATIC CANCER FAMILIES AND BREAST CANCER FAMILIES WITH PANCREATIC CANCER IS NOT INDICATED

ABSTRACT

PALB2-mutation carriers not only have an increased risk for breast cancer (BC) but also for pancreatic cancer (PC). Thus far, *PALB2* mutations have been mainly found in PC patients from families affected by both PC and BC. As it is well known that the prevalence of gene mutations varies between different populations, we studied the prevalence of *PALB2* mutations in a Dutch cohort of non-*BRCA1/2* familial PC (FPC) families and in non-*BRCA1/2* familial BC (FBC) families with at least one PC case. Mutation analysis included direct sequencing and multiplex ligation-dependent probe amplification (MLPA) and was performed in a total of 64 patients from 56 distinct families (28 FPC families, 28 FBC families). In total, 31 patients (48%) originated from FPC families; 24 were FPC patients (77%), 6 had a personal history of BC (19%) and 1 was a suspected carrier (3.2%). The remaining 33 patients (52%) were all female BC patients of whom 31 (94%) had a family history of PC and 2 (6.1%) had a personal history of PC. In none of these 64 patients a *PALB2* mutation was found. Therefore, *PALB2* does not have a major causal role in familial clustering of PC and BC in non-*BRCA1/2* families in the Dutch population.

INTRODUCTION

Recently, it has become clear that the Fanconi gene *FANCN/PALB2* (partner and localizer of *BRCA2*) should not only be considered as a susceptibility gene for breast cancer (BC) (1) but also as a susceptibility gene for pancreatic cancer (PC) (2). Mutations in this gene may be associated with familial clustering of PC and BC (1-10).
Previous studies have shown that *PALB2*-mutation-positive familial BC (FBC) patients were significantly more likely to have a relative with PC (5), and that nearly all *PALB2*-mutation positive familial PC (FPC) families were affected by at least one BC case (4).
Given these findings and the fact that the prevalence of gene mutations varies between different populations, we aimed to determine the prevalence of *PALB2* mutations in Dutch cohorts of non-*BRCA1/2* FPC patients and of non-*BRCA1/2* FBC patients with a personal or family history of PC.

MATERIALS AND METHODS

The prevalence of germline mutations in *PALB2* was investigated in Dutch non-*BRCA1/2* FPC patients and non-*BRCA1/2* FBC patients with a personal or family history of PC.
FPC families were defined as families with PC in either ≥2 first-degree relatives (FDRs), ≥3 relatives (FDR and second-degree relative (SDR)) or 2 SDRs of whom one was <50 years at diagnosis and did not meet diagnostic criteria of specific other cancer syndromes (11). These families were identified in the registries of the Clinical Genetic Centres of Amsterdam (Academic Medical Centre-University Medical Centre Amsterdam and Netherlands Cancer Institute), Rotterdam (Erasmus MC-University Medical Centre Rotterdam), and Groningen (University Medical Centre Groningen), consisting of a total of 40 FPC families. In 28 of these families, DNA was available for *PALB2* mutation analysis. In families in which DNA was available of multiple family members affected by PC, *PALB2* mutation analysis was performed in DNA of all cases. In families without available DNA from PC patients, mutation analysis was performed in family members affected by BC. In one family, mutation analysis was performed in a suspected carrier; this suspicion was based on the position of this individual in the pedigree; this specific case had a sibling with PC and a child with PC.
The FBC patients were taken from the registry from the Netherlands Cancer Institute and consisted of non-*BRCA1/2* BC patients that fulfilled the Dutch clinical criteria for *BRCA1* and *BRCA2* mutation testing, which include (1) BC diagnosis at age <35 years, (2) bilateral BC of which one diagnosis at age <50 years, (3) at least two FDR with BC at an age <50 years, (4) at least three FDR or SDR with BC, and (5) one <50 years at diagnosis. From this registry, patients with a personal history of both BC and PC, and BC patients with a FDR or SDR with PC were selected. In families in which DNA was available of multiple affected family members, *PALB2* mutation analysis was performed in all cases.

At the time of genetic counselling, patients had given written informed consent to use their DNA for the search for new cancer susceptibility genes.

Sequencing and multiplex ligation-dependent probe amplification (MLPA)
The presence of germline mutations in *PALB2* was evaluated by direct sequencing of the entire coding region and by sequencing intron–exon boundaries on genomic DNA isolated from whole blood. Primer pairs that were used have been previously described (12).
The presence of large genomic deletions in *PALB2* was analysed by MLPA using the MLPA P057 kit of MRC-Holland (Am sterdam, The Netherlands) as previously described (13). As a positive control, genomic DNA from the previously described *PALB2* FA patient EUFA1341 was included in the analysis (12).

RESULTS

PALB2 mutation analysis was performed in a total of 64 patients from 56 distinct families (28 FPC families, 28 FBC families; Table 1). In total, 31 patients (48%) originated from FPC families; 24 were FPC patients (77%), 6 had a personal history of BC (19%) and 1 was a suspected carrier (3.2%). The remaining 33 patients (52%) were all female BC patients of whom 31 (94%) had a family history of PC and 2 (6.1%) had a personal history of PC.
The 28 FPC families had a total of 70 affected patients with PC of which 57 (81%) were confirmed by medical (n=11) or pathology reports (n=46) (Table 2). The mean age at time of PC diagnosis was 61 years (SD±11.5). In total, 38 of the PC patients (54%) were male. Ten FPC families (36%) had at least three family members diagnosed with PC. Fourteen FPC families (50%) were affected by BC. Among the 33 tested patients from 28 FBC families, the mean age at BC diagnosis was 42 years (SD±9.5). The total number of PC cases was 29, of which 24 (83%) were confirmed by medical (n¼13) or pathology report (n=11). The mean age at PC diagnosis was 70 years (SD±10.9) and 41% (n=12) of all PC cases were male. In none of these cases was a *PALB2* mutation found by direct sequencing and MLPA.

DISCUSSION

Our data provide further evidence that there is a limited causal role for *PALB2* mutations in both FPC and FBC, as we did not identify any *PALB2* mutations in our Dutch cohort of 28 FPC families and 28 FBC families affected with at least one case of PC.
Since the recent recognition of *PALB2* as BC- and PC-susceptibility gene (1,2), a number of studies have been carried out to investigate the role of *PALB2* in different patient populations. Our results are in line with the results of these previous reports in which no *PALB2* mutations (9,14) or low prevalence of *PALB2* mutations (2,4,7–8,10) were found. It should be mentioned that the relatively small sample size could be a possible explanation for that no mutation carrier was identified in the current study. Even if a larger sample size might have detected sporadic cases, its results do show that the role of *PALB2* in this

FPC-families n=28	**n**	**%**
Total number of cases tested	31	
Type of case in whom *PALB2* mutation analysis was performed		
- Personal history of FPC	24	77
- Personal history of BC	6	19
- Suspected carrier	1	3.2
BC-PC family n=28		
Total number of cases tested	33	
Type of case in whom *PALB2* mutation analysis was performed		
- Personal history of BC with FDR or SDR with PC	31	94
- Personal history of both BC and PC	2	6.1
Type of Dutch clinical criteria for *BRCA1/2* mutation testing per family		
- BC diagnosis at age <35 years	6	21
- Bilateral BC, one diagnosis at age <50 years	2	7.1
- ≥2 FDR with BC at age <50 years	12	43
- ≥3 FDR or SDR with BC, one <50 years at diagnosis	8	29

Table 1. Characteristics of tested families (n=56) and cases (n=64)

FPC families n=28		
Total number of PC-cases (n)	70	
Confirmed by medical or pathology report (n , %)	*57*	*81*
Mean age at PC-diagnosis (years, SD)	61	11.5
Mean age at PC-diagnosis in tested case n=24 (years, SD)	60	11.4
Male gender of PC-case (n, %)	38	54
Number of families ≥3 family members diagnosed with PC (n, %)	10	36
Number of families affected by BC (n, %)	14	50
BC-PC families n=28 and cases n=33		
Mean age at BC-diagnosis in tested case (years, SD)	42	9.5
Total number of PC-cases (n)	29	
Confirmed by medical or pathology report (n, %)	*24*	*83*
Mean age at PC-diagnosis (years, SD)	70	11.9
Male gender of PC-case (n, %)	12	41

Table 2. Detailed description of FPC-families (n=28) and PC-affected FBC families

particular setting is insignificant. When combining our data with the previously published data, *PALB2* is involved in only 2.3% (7/306, range 0–3.7%, 95% CI 0.6–40%) of all FPC families and in 1.6% (5/306, range 0–4.8%, 95% CI 0.21–31%) of all FBC families with PC cases.

Although *PALB2* is involved in the clustering of both PC and BC, it explains only a small fraction of the clustering, and it is therefore crucial that future research is directed towards identifying the gene(s) that are involved in the development of both FPC and FBC. Knowledge of additional PC- and BC-susceptibility genes will be helpful in the counselling of family members from FPC and FBC families, as this will improve our ability to identify individuals at increased risk of developing PC and BC. Furthermore, it will have implications on the effectiveness of screening which will be highest when only directed towards individuals at risk.

Role of *PALB2*-mutations in FPC						
Study	*Country*	*Population (families)*			*PALB2 + families*	*PALB2 + families with BC*
		Total	*FPC*	*FPC with BC*		
		n	*n*	*n*	*n (%, 95% C.I.)*	
Jones ('09)(2)	USA	96	96	n.s.	3 (3.1%, n.s.)	2/3 (75%)
Tischkowitsh ('09)(4)	Canada	101	80	21	1 (1.0%, n.s.)	1/1 (100%)
Slater ('10)(10)	Europe	81	67	15	3 (3.7%, 0.8-10.4%)	3/3 (100%)
Current study	Netherlands	28	14	14	0 (0%, n.a.)	n.a.
Role of *PALB2*-mutations in BC-families affected by PC						
Study	*Country*	*Population (families)*			*PALB2 + families*	
		Total	*Pers. Hx BC & F/SDR PC*	*Pers. Hx BC&PC*		
		n	*n*	*n*	*n (%, 95% C.I.)*	
Adank ('10) (14)	Netherlands	45	45	0	0 (0%, n.a.)	
Peterlongo ('11) (8)	Italy	62	62	0	3 (4.8%, 0.99-13.29%)	
Stadler ('11) (9)	USA	77	55	22	0 (0%, n.a.)	
Hofstatter ('11)(7)	USA	94	91	3	2 (2.1%, 0.4-6.5%)	
Current study	Netherlands	28	26	2	0/28 (0%)	

Table 3. Overview of literature on role of PALB2-mutations in FPC and in BC-families affected by PC
n.s. not specified; n.a. not applicable

In conclusion, our results provide further evidence for the low prevalence of *PALB2* mutations among non-*BRCA1/2* FPC families and FBC families with PC cases. Therefore, routine analysis of this gene in these families is not warranted. Future research should be directed towards specifying subtypes of FPC/FBC families in which *PALB2* analysis is useful towards identifying other gene(s) involved in the development of PC and BC

REFERENCES

1. Rahman N, Seal S, Thompson D et al: PALB2, which encodes a BRCA2- interacting protein, is a breast cancer susceptibility gene. Nat Genet 2007; 39:165–167.
2. Jones S, Hruban RH, Kamiyama M et al: Exomic sequencing identifies PALB2 as a pancreatic cancer susceptibility gene. Science 2009; 324: 217.
3. Erkko H, Xia B, Nikkila J et al: A recurrent mutation in PALB2 in Finnish cancer families. Nature 2007; 446: 316–319.
4. Tischkowitz MD, Sabbaghian N, Hamel N et al: Analysis of the gene coding for the BRCA2-interacting protein PALB2 in familial and sporadic pancreatic cancer. Gastroenterology 2009; 137: 1183–1186.
5. Casadei S, Norquist BM, Walsh T et al: Contribution of inherited mutations in the BRCA2-interacting protein PALB2 to familial breast cancer. Cancer Res 2011; 71:2222–2229.
6. Hellebrand H, Sutter C, Honisch E et al: Germline mutations in the PALB2 gene are population specific and occur with low frequencies in familial breast cancer. Hum Mutat 2011; 32: E2176–E2188.
7. Hofstatter EW, Domchek SM, Miron A et al: PALB2 mutations in familial breast and pancreatic cancer. Fam Cancer 2011; 10: 225–231.
8. Peterlongo P, Catucci I, Pasquini G et al: PALB2 germline mutations in familial breast cancer cases with personal and family history of pancreatic cancer. Breast Cancer Res Treat 2011; 126: 825–828.
9. Stadler ZK, Salo-Mullen E, Sabbaghian N et al: Germline PALB2 mutation analysis in breast-pancreas cancer families. J Med Genet 2011; 48: 523–525.
10. Slater EP, Langer P, Niemczyk E et al: PALB2 mutations in European familial pancreatic cancer families. Clin Genet 2010; 78: 490–494.
11. Brand RE, Lerch MM, Rubinstein WS et al: Advances in counselling and surveillance of patients at risk for pancreatic cancer. Gut 2007; 56: 1460–1469.
12. Xia B, Dorsman JC, Ameziane N et al: Fanconi anemia is associated with a defect in the BRCA2 partner PALB2. Nat Genet 2007; 39: 159–161.
13. Ameziane N, van den Ouweland AM, Adank MA et al: Lack of large genomic deletions in BRIP1, PALB2, and FANCD2 genes in BRCA1/2 negative familial breast cancer. Breast Cancer Res Treat 2009; 118: 651–653.
14. Adank MA, van Mil SE, Gille JJ, Waisfisz Q, Meijers-Heijboer H: PALB2 analysis in BRCA2-like families. Breast Cancer Res Treat 2011; 127: 357–362.

F. Harinck, S.E. Korsse, M.G.F. van Lier, K. Biermann, G.J.A. Offerhaus, N.C. Krak, C.W.N. Looman, W. van Veelen, E.J. Kuipers, A. Wagner, E. Dekker, E.M.H. Mathus-Vliegen, P. Fockens, M.E. van Leerdam, M.J. Bruno

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CHAPTER 10
PANCREATIC CANCER RISK IN PEUTZ-JEGHERS SYNDROME PATIENTS; A LARGE COHORT STUDY AND IMPLICATIONS FOR SURVEILLANCE

ABSTRACT

Background | Although Peutz-Jeghers syndrome (PJS) is known to be associated with pancreatic cancer (PC), estimates of this risk differ widely. This hampers counselling of patients and implementation of surveillance strategies. We therefore aimed to determine the PC risk in a large cohort of Dutch PJS patients.

Methods | PJS was defined by diagnostic criteria recommended by the WHO, a proven *LKB1* mutation, or both. All patients with a presumptive diagnosis of pancreatic, ampullary or distal bile duct cancer were identified. Cases were reviewed clinically, radiologically and immunohistochemically. Cumulative PC risks were calculated by Kaplan-Meier analysis and relative risks by Poisson regression analysis.

Results | We included 144 PJS patients (49% male) from 61 families (5640 person years follow-up). Seven (5%) patients developed PC at a median age of 54 years. Four patients (3%) were diagnosed with distal bile duct (n=2) or ampullary cancer (n=2) at a median age of 55 years. The cumulative risk for PC was 26% (95% CI 4% to 47%) at age 70 years and relative risk was 76 (95% CI 36 to 160; $p < 0.001$). The cumulative risk for pancreatico-biliary cancer was 32% (95% CI 11% to 52%) at age 70, with a relative risk of 96 (95% CI 53 to 174; $p < 0.001$).

Conclusions | PJS patients have a highly increased risk for pancreatico-biliary cancer. Therefore, patients are eligible for surveillance within well defined research programmes to establish the benefit of such surveillance.

INTRODUCTION

Despite the relative low incidence of pancreatic cancer (PC) (8-10 per 100 000 per year, with an approximate 1% life time risk in western populations (1)), PC is among the top five causes of cancer related deaths in both the USA and Europe (2,3). The mean survival after diagnosis is less than 6 months and the overall 5-year survival is less than 5% (4). This poor prognosis is mainly due to the late onset of symptoms and anatomic location of the disease. Consequently, less than 20% of all patients presents with localised disease and are therefore eligible for curative treatment. Unfortunately, this intended curative treatment proves only to be effective for the minority of patients with an overall 5-year survival after surgical resection of less than 10% (5). Despite recent advantages in the field of surgery and oncology, this dismal prognosis has not significantly changed over the past decades (6).

Detection of precursor lesions or malignancies at an early asymptomatic stage by surveillance with endoscopic ultrasound (EUS) and/or MRI could offer a way to improve the prognosis (7). In particular, when surveillance is directed towards populations of individuals that carry a high risk for developing PC, the potential health gain could be substantial.

One such high-risk population consists of patients with Peutz-Jeghers syndrome (PJS). PJS is an autosomal dominant inherited disorder, caused by germline mutations in the *LKB1* tumour suppressor gene (also known as *STK11*) (8). It is characterised by gastrointestinal hamartomas and mucocutaneous pigmentations. Furthermore, patients with PJS are at risk for developing various types of cancer, including PC (9-11). The actual risk of developing PC for PJS patients is currently unclear. Previous studies reported relative risks ranging from 0 to 132-fold increase and an average age of PC onset ranging from 41 to 60 years of age (10-14). Consequently, this hampers counselling of PJS patients and implementation of surveillance strategies.

These disparate risk estimates were mainly derived from heterogeneous multicentre populations, small single centre cohort studies, and meta-analyses of these same studies. It is therefore key to perform such a study in a large homogenous population. In 2011, our research group reported on the high overall cancer risk in a unique, large pedigree based homogenous cohort of Dutch PJS patients with a substantial prospective period of follow-up (9). For the present study, we performed a thorough re-evaluation of all reported cancers in the pancreatico-biliary region in this patient cohort, including 2 years of extended follow-up. Thus, we aimed to conclude the ongoing debate regarding the true PC risk in PJS, and to provide a more scientific rationale for the implementation of surveillance strategies.

METHODS

Peutz-Jeghers syndrome database

This nationwide cohort study was initiated by two Dutch academic hospitals. Between 1995 and July 2011, PJS patients throughout the Netherlands were included without selection for medical history. All patients had a definite diagnosis of PJS, defined by diagnostic criteria recommended by the World Health Organization (WHO) (Box 1), a proven *LKB1* mutation, or both. Informed consent was obtained from all patients and the study was approved by the Institutional Review Board of both participating hospitals. Patients were followed prospectively between January 1995 and July 2011. Patient information at baseline and during follow-up was obtained by interview and chart review. Clinical data from the period before 1995 as well as data of deceased family members fulfilling the diagnostic criteria for PJS were collected retrospectively.

A. Positive family history of PJS, and 1. Any number of histologically confirmed PJS polyps*, or 2. Characteristic, prominent, mucocutaneous pigmentations.
B. Negative family history of PJS, and 1. Three or more histologically confirmed PJS polyps, or 2. Any number of histologically confirmed PJS polyps and characteristic, prominent, mucocutaneous pigmentations.

Box 1. Diagnostic criteria for Peutz-Jeghers syndrome (PJS) recommended by the World Health Organization (2010). PJS, Peutz-Jeghers syndrome.

** Histology of PJS polyps: a central core of smooth muscle that shows tree-like branching, covered with normal epithelium.*

Case selection and data collection

PC cases were identified from the PJS database. PC was defined according to the most recent WHO classification of tumours of the digestive system (4). In addition, patients with a diagnosis of distal bile duct cancer or ampullary cancer were included. Surveillance of the pancreas might also detect these malignancies and accurate distinction between these three tumours often proves difficult. From all selected cases, medical records were reviewed by two MDs (SEK and FH). The following data were collected: gender, date of birth, cancer diagnosis and death, mutation status and type of mutation, family history of PJS, and family history of PC. The recorded cancer characteristics included tumour type and origin, tumour invasion, data on confirmation (medical record or histology), and presentation (surveillance, accidentally or symptomatic). Radiological images were reviewed by an expert abdominal radiologist (NK). Available formalin-fixed and paraffin-embedded tissue was reviewed by two expert pathologists independently (KB and GJAO).

Immunohistochemical staining for SMAD4, CDX2 and cytokeratins was performed to ascertain the diagnosis (4). Eventually, all available information was re-assessed by an expert panel (KB, GJAO, NK, ED, EMHMV, MEvL and MJB) to determine a definite diagnosis of PC, distal bile duct cancer or ampullary cancer.

Statistical analysis

Data were analyzed using the SPSS V.17.0 statistical software for Windows (IBM, Somers, New York, USA). All risks were calculated for two groups: (1) PC cases; (2) cases of cancer in the pancreatico-biliary region, including cases with PC, distal bile duct cancer or ampullary cancer. Cumulative risks were estimated as a function of time using the Kaplan-Meier method and the Cox regression model. For these cumulative risk analyses, all subjects of the cohort were included. For relative cancer risk calculation, the tumour specific cancer incidence observed in our study population was compared to the age specific and gender specific incidence rates of the Dutch general population from 1960 to 2011 by Poisson regression analysis (log linear analysis) using the package R (15). Subjects were studied with respect to their risk of developing cancer from birth until the date of death, date of last contact or the closing date of the study (1 July 2011). Sociodemographic data and incidence rates of the Dutch general population were derived from the Eindhoven Cancer Registry (1960-2009). These data are representative for the Netherlands. Incidence rates for 2009 were assumed to be representative for 2010 and 2011.

RESULTS

Study population

In total, 144 PJS patients from 61 families were included in the cohort with a total of 5640 person-years of follow-up (including 1757 person-years of prospective follow-up). Forty-nine per cent were male (3050 person-years). At the closing date of the study six patients had been lost to follow-up (4%), and 48 (33%) had died at a median age of 46 years (IQR 32-58 years); the median age of the 90 patients still alive (63%) was 37 years (IQR 21-52 years). The baseline characteristics of the cohort are shown in Table 1.

Pancreatic cancer cases

The case selection process is shown in Figure 1. During follow-up, seven (4.9%) PJS patients from seven families developed PC, six male and one female. None of the cases was detected within the framework of a PC screening/surveillance programme. Adenocarcinoma of the pancreas was found in 6 patients and acinar cell carcinoma in one. Six cases were confirmed by revision of histology, and no histological material was available for the seventh case. Median age at diagnosis was 54 years (IQR 37-62 years). Six patients presented with symptoms. In one patient a tumour mass was incidentally found during a laparotomy because of small bowel polyps. None of the patients could

Total	144
Gender	
Male	70 (49%)
Female	74 (51%)
Families	61
Family history	
Familial PJS	109 (76%)
Sporadic	24 (17%)
Family history unknown	11 (7%)
Fulfilling WHO criteria[1]	142 (99%)
DNA mutation analysis	92 (64%)
LKB1 mutation carrier	82/92 (89%)
Deceased	48 (33%)
Median age at death	*46 years (IQR 32-58 years)*
Lost to follow up	6 (4%)
Cancer	48 (33%)
Median age at diagnosis of first cancer	46 years (IQR 35-55 years)
2 primary cancers	8

Table 1. Baseline characteristics of the Dutch Peutz-Jeghers syndrome cohort.
IQR, interquartile range; PJS, Peutz-Jeghers syndrome; WHO, World Health Organization.
[1] Two patients not fulfilling the WHO-criteria carry a proven LKB1-germline mutation.

be treated curatively. Median survival of patients after diagnosis was 6 months (IQR 4-17 months). Mutation analysis for the *LKB1* gene was performed in five patients, detecting a pathogenic mutation in four of them. For the other two cases, a pathogenic mutation in the *LKB1* gene was detected in affected family members, but mutation analysis was not performed in the PC patients. Two of the patients had been treated curatively for another malignancy (colorectal cancer and liposarcoma) prior to the development of PC. Individual patient characteristics are shown in Table 2A.

Ampullary cancer and distal bile duct cancer cases

In addition to the patients with PC, distal bile duct cancer was diagnosed in a male patient at the age of 57 and in a female patient at the age of 73 years. Both patients only underwent palliative treatment; survival after diagnosis was 3 and 8 months, respectively. Furthermore, ampullary cancer/cancer involving the ampulla was detected in two male patients at the age of 41 and 53 years. Both patients underwent a pylorus preserving pancreaticoduodenectomy as curative treatment. One patient died of metastasized disease 5 years after diagnosis; the other patient is still alive 11 years after diagnosis. Patient characteristics are shown in Table 2B. The median age at diagnosis for the group of patients with pancreatic, distal bile duct or ampullary cancer (n=11) was 54 years (IQR 42-62).

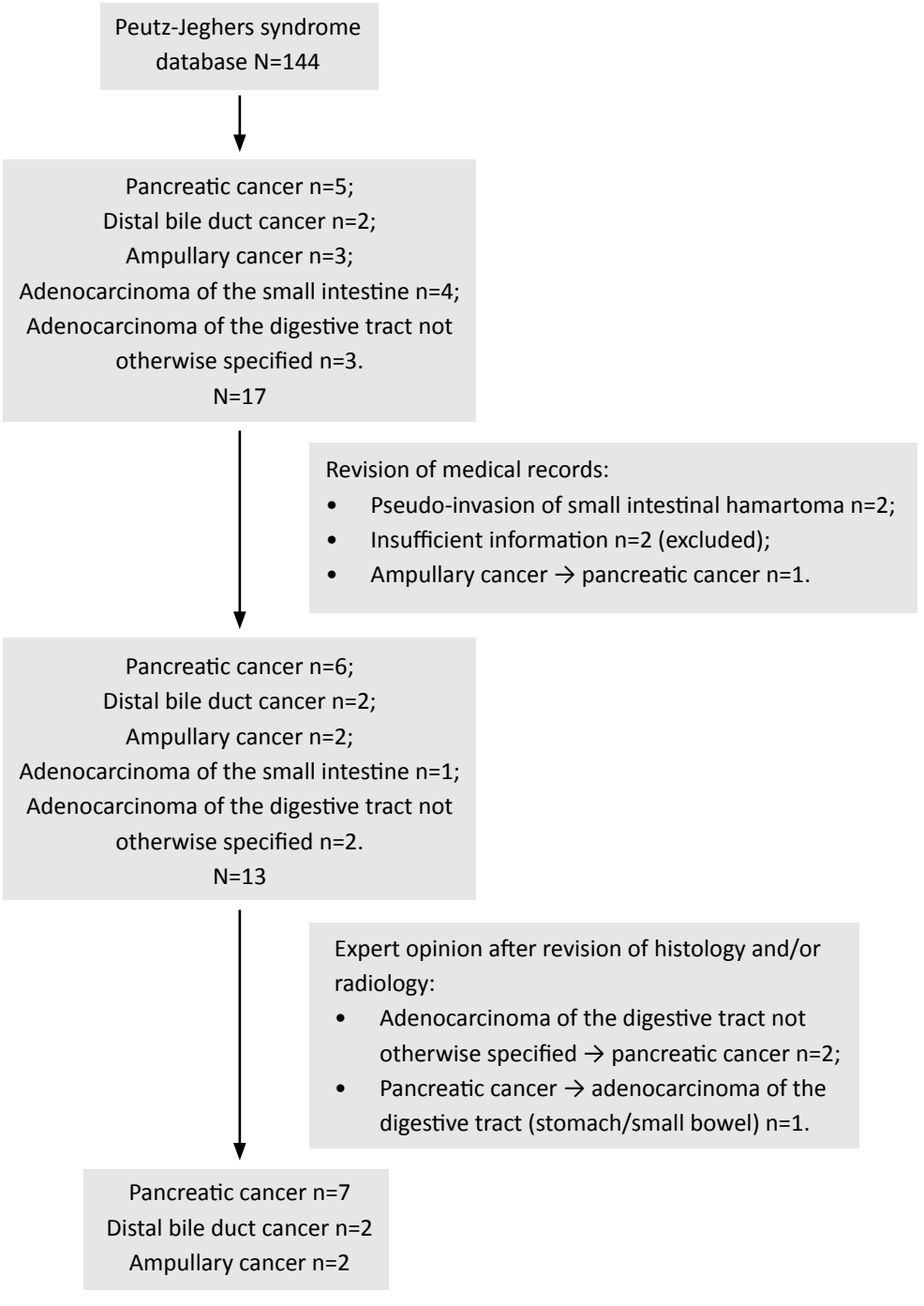


Figure 1. Case selection

A	Germline *LKB1* mutation in patient (Germline *LKB1* mutation in family)	Age at diagnosis (year of diagnosis)	Age at death	Type of cancer	Confirmation	Location tumour	Distant metastases	Other malignancy in medical history (year of diagnosis)
1.	+ (+)[1]	35 (1998)	35	Adenocarcinoma	HI	Head	Yes	No
2.	NT (c.370A>T, p.Lys124X)	57 (1994)[2]	58	Adenocarcinoma	HI	Head	Yes	Unknown
3.	NT (c.468C>G, p.Tyr156X)	66 (1998)	68	Adenocarcinoma	HI	Head	Yes	Colon cancer pT2NxMx (1996)
4.	c.991dupC, p.Arg331fs	36 (1997)	36	Adenocarcinoma	HI	Head	Yes	No
5.	c.582C>A, p.Asp194Glu	45 (2008)	46	Acinar cell carcinoma	HI	Tail	Yes	Liposarcoma (1992)
6.	Deletion exon 6, 7 and 8	54 (2004)	54	Adenocarcinoma	MR	Head	Yes	No
7.	- (-)	62 (2005)	62	Adenocarcinoma	HI	Head	Yes	No
B								
1.	c.156_157dupGG, p.Asp53fs	57 (2006)	57	Bile duct cancer	HI	Distal bile duct	Yes	No
2.	NT (unknown)	73 (2008)	74	Bile duct cancer	HI	Distal bile duct	Yes	No
3.	c.291-2A>G, p.98_155del	53 (2006)	58	Ampullary NET	HI	Ampulla	Yes	Non-Hodgkin Lymphoma grade 3 (2001)
4.	c.735-1G>A, p.?	41 (2000)	-	Dysplastic hamartoma near the ampulla with invasive growth in the ampulla	HI	Ampulla	No	No

Table 2. Characteristics of Peutz-Jeghers syndrome patients with pancreatic cancer (Table 2A) and with distal bile duct cancer and ampullary cancer (Table 2B)
F, female; HI, histology; M, male; MR, medical record; NET, neuro-endocrine tumor; NT, not tested; +, proven germline LKB1 mutation; -, tested but no germline mutation detected. [1]Specific LKB1 germline mutation unknown. [2]Occurred in retrospective follow-up period.

Cumulative cancer risk

Pancreatic cancer (n=7)

The Kaplan-Meier estimate for the cumulative PC risk was 2.4% (SE 1.7%; 95% CI -0.9% to 5.7%) at age 40; 3.9% (SE 2.2%; 95% CI -0.4% to 8.2%) at age 50; 11.1% (SE 5.3%; 95% CI 0.7% to 21.5%) at age 60; and 25.6% (SE 10.8%; 95% CI 4.4% to 46.8%) at age 70 (Figure 2A). There was no significant difference in risk between males and females (p=0.272).

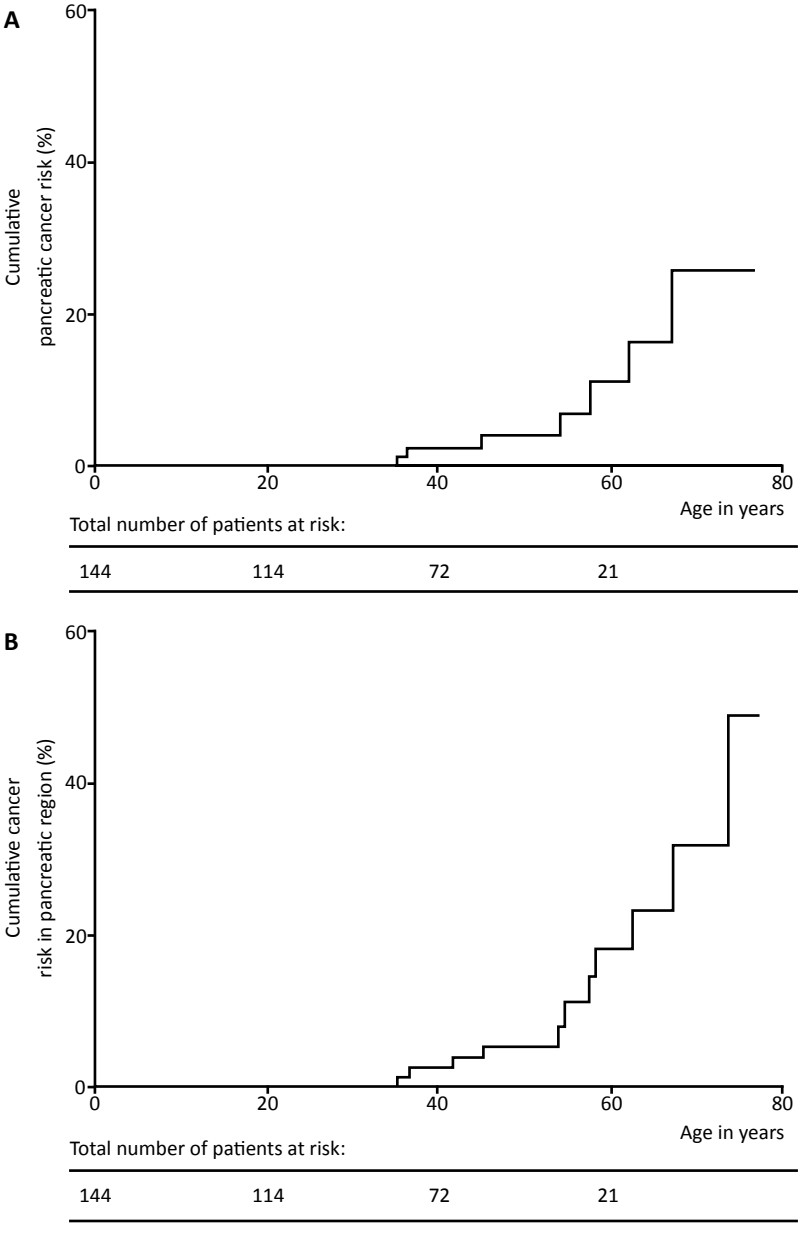


Figure 2. Cumulative pancreatic cancer risk (A) and cancer risk in pancreatic region (B) in Peutz-Jeghers syndrome patients according to age.

Pancreatic, ampullary or distal bile duct cancer (n=11)
For pancreatic, distal bile duct or ampullary cancer the cumulative risk was 2.4% (SE 1.7%; 95% CI -0.9% to 5.7%) at age 40; 5.2% (SE 2.6%; 95% CI 0.1% to 10.3%) at age 50; 18.2% (SE 6.5%; 95% CI 5.5% to 30.9%) at age 60; and 31.6% (SE 10.6%; 95% CI 10.8% to 52.4%) at age 70 (figure 2B). There was no significant difference in risk for these malignancies between males and females (p=0.248)

Relative cancer risk
From 1960, 131 patients contributed to 4430 person-years at risk (males 2259 person-years, females 2171 person-years). Poisson regression analysis showed that the relative risk for PC (HR 76.2, 95% CI 36.3 to 160.0) as well as for pancreatic, ampullary or distal bile duct cancer (HR 95.8, 95% CI 52.8 to 173.7) was significantly higher in PJS patients than in the general population (p<0.001).

DISCUSSION

This nationwide, long term follow-up cohort study shows that patients with PJS have a highly increased PC risk. We found a cumulative cancer risk of more than 25% at the age of 70 years and a 76-fold increased risk compared to the general population. The cumulative risk for developing any type of malignancy in the pancreatico-biliary region, including pancreatic, distal bile duct, or ampullary cancer, is as high as 29% at the age of 70 years and the relative risk for these cancers is 96. These data emphasise the relevance and clinical potential of surveillance of the pancreas for PJS patients, provided a suitable and effective surveillance program is available.
Estimates on the risk of PC in patients with PJS vary widely within the literature. Our data are most in line with those from Giardiello *et al.* (11). In a meta-analysis in which 210 PJS patients from six American and European studies were included, six cases of PC were identified which amounted to a cumulative risk of 36% by the age of 64 years and a relative risk of 132. The mean age at PC onset was 40.8 years (SD 16.2). Because the source data were contributed by multiple centres worldwide, the authors were not able to give extensive information about the intricacies of case selection or confirmation of the cancer diagnosis, including revision of pathology specimens and potential confounding issues relating to the problematic distinction between pancreatic, distal bile duct, and ampullary cancer. Furthermore, incidence rates of the US population were used for relative risk calculation, while the study population consisted of both US and European (UK and Dutch) patients. This might have led to biased relative cancer risks, as differences in cancer risk between PJS and control populations could exist due to variations in geography, race, culture and diet.
The extremely elevated risk found by Giardiello *et al.* has not been reproduced by more recent studies. An international collaborative study concerning 419 PJS patients from

eight centres worldwide found a cumulative risk for developing PC of 11% by the age of 70 (12). Relative risk was not reported. Another collaborative study found no PC in a total of 149 PJS cases (13). Interpretation of the results of these two series is difficult, since no information is provided on the average age of the cohort or follow-up period. Albeit speculative, the lower risk found in these series could be the result of the participants being too young or the lack of sufficient follow-up time. The same data on age and follow-up period is missing in most current nationwide or single centre studies, which are often limited by a small sample size.

To our knowledge this is the first study to investigate the PC risk within a large, nationwide PJS patient cohort with long term follow up. This cohort goes back to the original family described by Jan Peutz in 1921 (16), and encompasses a substantial period of prospective follow-up time amounting to 5640 person-years including 1757 person-years of prospective follow-up. Because it is well known that differentiation between pancreatic, distal bile duct and ampullary cancer is a diagnostic challenge, we attempted to address the issue of case selection by careful expert revision of clinical, radiological, and histological materials. This enabled us to provide reliable risk estimates for PC alone and for cancers of the pancreas and pancreatico-biliary region including distal bile duct and ampullary cancer. The latter are sometimes misclassified or impossible to differentiate from PC. As such, these numbers could be looked upon as absolute minimum and maximum risk estimates. Another important clinical consideration when making such a separate risk assessment is that distal bile duct and ampullary cancer, just like PC, have a potential for early detection in surveillance programs of the pancreas.

A few limitations of our study warrant consideration. Firstly, because this PJS patient cohort was initiated by two tertiary referral centres, selection bias could potentially have led to an overestimated incidence of PC. Secondly, we were unable to gather reliable information about the smoking behaviour of our patients. This is unfortunate because smoking is one of the most important risk factors for the development of PC (17, 18) and therefore a probable confounding factor between different PJS populations.

The evidence is slowly accumulating that surveillance of high risk individuals leads to the detection of high grade precursor lesions and asymptomatic early stage PC (19-27). However, we currently still lack definite evidence that surveillance has a net benefit in terms of mortality reduction of PC related mortality and gain in life years, and whether this benefit outweighs the potential negative side effects of overtreatment, including associated complications and costs. We and others therefore suggest that surveillance of PJS patients should only be performed within the framework of well established research protocols (28-30). Results of the international Cancer of the Pancreas screening (CAPS) summit meeting in 2011 indicate that surveillance in high risk individuals should be regarded as a promising development, though more evidence is needed to address its real value (30). During this meeting, 49 experts in the field of PC voted on statements with respect to PC surveillance. This resulted in a number of outstanding questions that still

need to be addressed, including questions with respect to who to screen, when to start screening, the optimal frequency of screening, and particularly the optimal management of the asymptomatic pancreatic lesions detected.

Based on the results of our current study, we recommend that PJS patients should be offered surveillance regardless of family history for PC, since all subjects with PC in our series had a negative family history of PC. Although the median age of PC onset in our cohort was 54 years, we propose that surveillance starts at the age of 30 years. This suggestion is based on the fact that two patients in our series developed cancer in the pancreatico-biliary region at a very young age. If screening had started 10 years earlier than the median age of PC onset, these cases would have been missed.

It has been noted that some patients with PJS develop intestinal-type intraductal papillary mucinous neoplasms (IPMNs) (31). IPMNs are well defined premalignant lesions of PC. One pancreatic adenocarcinoma in our study showed histological indication for development out of an IPMN lesion. Future research should be directed towards unravelling the molecular pathway of PC development in PJS patients. Such knowledge may tailor surveillance recommendations even more. Furthermore, the efficacy and cost effectiveness of PC surveillance must be further studied.

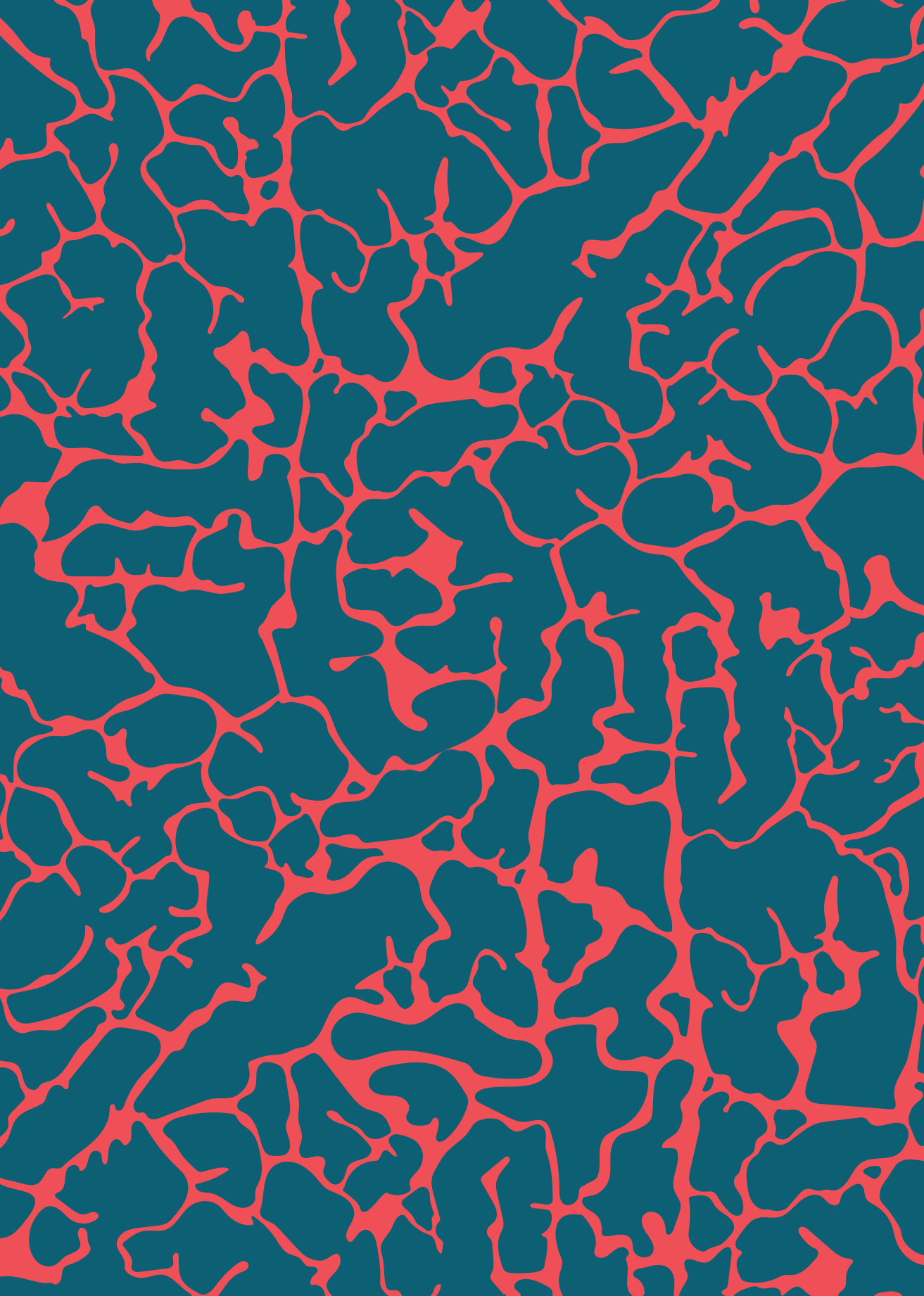
In conclusion, absolute and relative risks of developing pancreatic, distal bile duct and ampullary cancer are very high in patients with PJS. This observation, and the prospect that detection of these malignancies, or preferably their precursor lesions, might be possible at an early and potentially curable point in time, render PJS patients eligible for surveillance by yearly EUS and/or MRI within well defined research protocols.

REFERENCES

1. Jemal A, Bray F, Center MM, Ferlay J, Ward E, Forman D. Global cancer statistics. CA Cancer J Clin. 2011;61:69-90.
2. Malvezzi M, Arfe A, Bertuccio P, Levi F, La Vecchia C, Negri E. European cancer mortality predictions for the year 2011. Ann Oncol. 2011;22:947-56.
3. U.S. Cancer Statistics Working Group. United States Cancer Statistics: 1999–2007 Incidence and Mortality Web-based Report. Atlanta: U.S. Department of Health and Human Services, Centers for Disease Control and Prevention and National Cancer Institute. Available at: www.cdc.gov/uscs. 2010;
4. U.S. Cancer Statistics Working Group. Available at http://seer.cancer.gov/statfacts/html/pancreas.html. 2010; Available from: http://seer.cancer.gov/statfacts/html/pancreas.html.
5. Kuhlmann KF, de Castro SM, Wesseling JG, ten Kate FJ, Offerhaus GJ, Busch OR, van Gulik TM, Obertop H, Gouma DJ. Surgical treatment of pancreatic adenocarcinoma; actual survival and prognostic factors in 343 patients. Eur J Cancer. 2004;40:549-58.
6. Vincent A, Herman J, Schulick R, Hruban RH, Goggins M. Pancreatic cancer. Lancet. 2011;378:607-20.
7. Harinck F, Poley JW, Kluijt I, Fockens P, Bruno MJ, Dutch Research Group of Pancreatic Cancer Surveillance in High-Risk I. Is early diagnosis of pancreatic cancer fiction? Surveillance of individuals at high risk for pancreatic cancer. Dig Dis. 2010;28:670-8.
8. Beggs AD, Latchford AR, Vasen HF, Moslein G, Alonso A, Aretz S, Bertario L, Blanco I, Bulow S, Burn J, Capella G, Colas C, Friedl W, Moller P, Hes FJ, Jarvinen H, Mecklin JP, Nagengast FM, Parc Y, Phillips RK, Hyer W, Ponz de Leon M, Renkonen-Sinisalo L, Sampson JR, Stormorken A, Tejpar S, Thomas HJ, Wijnen JT, Clark SK, Hodgson SV. Peutz-Jeghers syndrome: a systematic review and recommendations for management. Gut. 2010;59:975-86.
9. van Lier MG, Westerman AM, Wagner A, Looman CW, Wilson JH, de Rooij FW, Lemmens VE, Kuipers EJ, Mathus-Vliegen EM, van Leerdam ME. High cancer risk and increased mortality in patients with Peutz-Jeghers syndrome. Gut. 2011;60:141-7.
10. Giardiello FM, Welsh SB, Hamilton SR, Offerhaus GJ, Gittelsohn AM, Booker SV, Krush AJ, Yardley JH, Luk GD. Increased risk of cancer in the Peutz-Jeghers syndrome. N Engl J Med. 1987;316:1511-4.
11. Giardiello FM, Brensinger JD, Tersmette AC, Goodman SN, Petersen GM, Booker SV, Cruz-Correa M, Offerhaus JA. Very high risk of cancer in familial Peutz-Jeghers syndrome. Gastroenterology. 2000;119:1447-53.
12. Hearle N, Schumacher V, Menko FH, Olschwang S, Boardman LA, Gille JJ, Keller JJ, Westerman AM, Scott RJ, Lim W, Trimbath JD, Giardiello FM, Gruber SB, Offerhaus GJ, de Rooij FW, Wilson JH, Hansmann A, Moslein G, Royer-Pokora B, Vogel T, Phillips RK, Spigelman AD, Houlston RS. Frequency and spectrum of cancers in the Peutz-Jeghers syndrome. Clin Cancer Res. 2006;12:3209-15.

13. Mehenni H, Resta N, Park JG, Miyaki M, Guanti G, Costanza MC. Cancer risks in LKB1 germline mutation carriers. Gut. 2006;55:984-90.
14. You YN, Wolff BG, Boardman LA, Riegert-Johnson DL, Qin R. Peutz-Jeghers syndrome: a study of long-term surgical morbidity and causes of mortality. Fam Cancer. 2010;9:609-16.
15. Team RDC. R: A language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing; 2008.
16. Peutz JLA. Over een zeer merkwaardige, gecombineerde familiaire polyposis van de slijmliezen van den tractus intestinalis met die van de neuskeelholte en gepaard met eigenaardige pigmentaties van huid en slijmvliezen. Ned Maandschr v Geneesk. 1921;10:134-46.
17. Gold EB, Goldin SB. Epidemiology of and risk factors for pancreatic cancer. Surg Oncol Clin N Am. 1998;7:67-91.
18. Bosetti C, Lucenteforte E, Silverman DT, Petersen G, Bracci PM, Ji BT, Negri E, Li D, Risch HA, Olson SH, Gallinger S, Miller AB, Bueno-de-Mesquita HB, Talamini R, Polesel J, Ghadirian P, Baghurst PA, Zatonski W, Fontham E, Bamlet WR, Holly EA, Bertuccio P, Gao YT, Hassan M, Yu H, Kurtz RC, Cotterchio M, Su J, Maisonneuve P, Duell EJ, Boffetta P, La Vecchia C. Cigarette smoking and pancreatic cancer: an analysis from the International Pancreatic Cancer Case-Control Consortium (Panc4). Ann Oncol. 2011;
19. Al-Sukhni W, Borgida A, Rothenmund H, Holter S, Semotiuk K, Grant R, Wilson S, Moore M, Narod S, Jhaveri K, Haider MA, Gallinger S. Screening for Pancreatic Cancer in a High-Risk Cohort: An Eight-Year Experience. J Gastrointest Surg. 2011;
20. Canto MI, Goggins M, Hruban RH, Petersen GM, Giardiello FM, Yeo C, Fishman EK, Brune K, Axilbund J, Griffin C, Ali S, Richman J, Jagannath S, Kantsevoy SV, Kalloo AN. Screening for early pancreatic neoplasia in high-risk individuals: a prospective controlled study. Clin Gastroenterol Hepatol. 2006;4:766-81; quiz 665.
21. Canto MI, Hruban RH, Fishman EK, Kamel IR, Schulick R, Zhang Z, Topazian M, Takahashi N, Fletcher J, Petersen G, Klein AP, Axilbund J, Griffin C, Syngal S, Saltzman JR, Mortele KJ, Lee J, Tamm E, Vikram R, Bhosale P, Margolis D, Farrell J, Goggins M. Frequent detection of pancreatic lesions in asymptomatic high-risk individuals. Gastroenterology. 2012;142:796-804; quiz e14-5.
22. Kimmey MB, Bronner MP, Byrd DR, Brentnall TA. Screening and surveillance for hereditary pancreatic cancer. Gastrointest Endosc. 2002;56:S82-6.
23. Langer P, Kann PH, Fendrich V, Habbe N, Schneider M, Sina M, Slater EP, Heverhagen JT, Gress TM, Rothmund M, Bartsch DK. Five years of prospective screening of high-risk individuals from families with familial pancreatic cancer. Gut. 2009;58:1410-8.
24. Ludwig E, Olson SH, Bayuga S, Simon J, Schattner MA, Gerdes H, Allen PJ, Jarnagin WR, Kurtz RC. Feasibility and yield of screening in relatives from familial pancreatic cancer families. Am J Gastroenterol. 2011;106:946-54.

25. Poley JW, Kluijt I, Gouma DJ, Harinck F, Wagner A, Aalfs C, van Eijck CH, Cats A, Kuipers EJ, Nio Y, Fockens P, Bruno MJ. The yield of first-time endoscopic ultrasonography in screening individuals at a high risk of developing pancreatic cancer. Am J Gastroenterol. 2009;104:2175-81.
26. Vasen HF, Wasser M, van Mil A, Tollenaar RA, Konstantinovski M, Gruis NA, Bergman W, Hes FJ, Hommes DW, Offerhaus GJ, Morreau H, Bonsing BA, de Vos tot Nederveen Cappel WH. Magnetic resonance imaging surveillance detects early-stage pancreatic cancer in carriers of a p16-Leiden mutation. Gastroenterology. 2011;140:850-6.
27. Verna EC, Hwang C, Stevens PD, Rotterdam H, Stavropoulos SN, Sy CD, Prince MA, Chung WK, Fine RL, Chabot JA, Frucht H. Pancreatic cancer screening in a prospective cohort of high-risk patients: a comprehensive strategy of imaging and genetics. Clin Cancer Res. 2010;16:5028-37.
28. Bartsch DK, Gress TM, Langer P. Familial pancreatic cancer-current knowledge. Nat Rev Gastroenterol Hepatol. 2012;
29. Brand RE, Lerch MM, Rubinstein WS, Neoptolemos JP, Whitcomb DC, Hruban RH, Brentnall TA, Lynch HT, Canto MI. Advances in counselling and surveillance of patients at risk for pancreatic cancer. Gut. 2007;56:1460-9.
30. Canto MI, Harinck F, Hruban RH, Offerhaus GJ, Poley JW, Fockens P, Kamel IR, Nio CY, Schulick RD, Bassi C, Kluijt I, Goggins MG, Bruno MJ. International consensus recommendations on the management of patients with increased risk for familial pancreatic cancer: Cancer of the Pancreas Screening Consortium (CAPS) 2011 Summit. Gastroenterology 142:S-135, 2012 (abstr 764). 2012;
31. Sato N, Rosty C, Jansen M, Fukushima N, Ueki T, Yeo CJ, Cameron JL, Iacobuzio-Donahue CA, Hruban RH, Goggins M. STK11/LKB1 Peutz-Jeghers gene inactivation in intraductal papillary-mucinous neoplasms of the pancreas. Am J Pathol. 2001;159:2017-22.



PART III

PSYCHOLOGICAL ASPECTS RELATED TO SCREENING

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CHAPTER 11
FEASIBILITY OF A PANCREATIC CANCER SURVEILLANCE PROGRAM FROM A PSYCHOLOGICAL POINT OF VIEW

ABSTRACT

Purpose | The success of any surveillance program depends not solely on its technological aspects but also on the commitment of participants to adhere to follow-up investigations, which is influenced by the psychological impact of surveillance. This study investigates the psychological impact of participating in a pancreatic cancer surveillance program.

Methods | High-risk individuals participating in an endoscopic ultrasonography-magnetic resonance imaging-based pancreatic cancer surveillance program received a questionnaire assessing experiences with endoscopic ultrasonography and magnetic resonance imaging, reasons to participate, psychological distress, and benefits and barriers of surveillance. High-risk individuals were individuals with a strong family history of pancreatic cancer or carriers of pancreatic cancer-prone gene mutations.

Results | Sixty-nine participants (85%) completed the questionnaire. Surveillance was reported as "very to extremely uncomfortable" by 15% for magnetic resonance imaging and 14% for endoscopic ultrasonography. Most reported reason to participate was that pancreatic cancer might be detected in a curable stage. Abnormalities were detected in 27 respondents, resulting in surgical resection in one individual and a shorter follow-up interval in five individuals. Surveillance outcomes did not influence cancer worries. Overall, 29% was "often" or "almost always" concerned about developing cancer. Six respondents (9%) had clinical levels of depression and/or anxiety. According to 88% of respondents, advantages of surveillance outweighed disadvantages.

Conclusions | Although endoscopic ultrasonography is more invasive than magnetic resonance imaging, endoscopic ultrasonography was not perceived as more burdensome. Despite one third of respondents worrying frequently about cancer, this was not related to the surveillance outcomes. Anxiety and depression levels were comparable with the general population norms. Advantages of participation outweighed disadvantages according to the majority of respondents. From a psychological point of view, pancreatic cancer surveillance in high-risk individuals is feasible and justified.

INTRODUCTION

With a median survival of less than 6 months and a 5-year survival of <5%, pancreatic cancer (PC) is one of the most fatal of human malignancies (1,2). The poor prognosis is mainly due to the late onset of symptoms and the aggressiveness of this tumor type, such that the majority of patients presents with incurable disease. A way to improve the prognosis of this disease would be to diagnose precursor lesions or a malignancy at an early asymptomatic stage when resection offers the best chance for cure. PC surveillance of the general population is not feasible because of the relatively low incidence of PC (10/100,000 in the Western World) (3,4) and the lack of a noninvasive, reliable, and cheap surveillance tool. However, surveillance might be worthwhile when offered to subpopulations of individuals who are at high risk of developing PC.

Currently, several groups of individuals at high risk of developing PC have been identified. These include (1) mutation carriers of PC-prone hereditary syndromes (syndromic PC) and (2) individuals with a strong family history of PC but without a known underlying genetic defect (familial PC [FPC]) (5). The lifetime risk of developing PC in these inherited and familial syndromes is strongly increased compared with the general population. This lifetime risk is estimated to be >10% in mutation carriers of *BRCA1*, *BRCA2*, mismatch repair genes, and *TP53* from families affected by at least two PC cases, up to 17% for *CDKN2A* mutation carriers, up to 36% for patients with the Peutz Jeghers syndrome, and can exceed 40% in FPC family members with three affected first-degree relatives (FDR) (6). Some studies have already provided information about the effectiveness of surveillance (7–12). In these studies, the effectiveness of different surveillance techniques, such as computed tomography, magnetic resonance imaging (MRI), and endoscopic ultrasonography (EUS), were investigated. Preliminary results of these studies are promising, although it is still unclear whether surveillance will actually improve survival. When assessing the success of a surveillance program, it is important not only to focus on technological aspects such as test performance but also to focus on the psychological aspects related to surveillance (13). For example, a surveillance tool might be technologically successful in detecting cancer in a curable stage. However, if individuals do not participate in the surveillance program because the psychological burden of surveillance is too high, a surveillance program will ultimately not be successful. To date, knowledge about the psychological aspects of PC surveillance is limited. Thus far, only three articles have been published that address the psychological aspects of PC surveillance (14–16). These studies provide relevant information on patient views regarding the value of genetic counseling for FPC in the absence of predictive genetic testing (16), psychological well-being of high-risk individuals participating in PC surveillance (15), and intentions of high-risk individuals to participate in PC surveillance (14). Knowledge is limited about the specific experiences of high-risk individuals with PC surveillance, their perceived burden, and expectations of such a surveillance program.

These are important topics that should to be taken into account when studying the feasibility of a PC surveillance program from a psychological point of view (17). Therefore, this study was undertaken to investigate the psychological impact of an EUS-MRI-based PC surveillance among high-risk individuals and to evaluate whether PC surveillance is psychologically feasible. Specific aims of this study were to (1) investigate participants' experienced burden of a PC surveillance program, (2) investigate their motivations to participate in such a program, (3) investigate general levels of distress and, and (4) identify factors associated with anxiety experienced during an EUS-MRI-based surveillance program.

METHODS

Sample

Eligible for this psychological questionnaire study were all participants of a Dutch PC surveillance study. This is a multicenter prospective study investigating the effectiveness of PC surveillance in high-risk individuals. High-risk individuals were defined as (1) FDR of patients with FPC and (2) carriers of a PC-prone gene mutation. FPC kindreds are defined as families with (1) at least two FDR with PC, (2) at least three relatives with PC (FDR or second-degree relative [SDR]), or (3) at least two SDR relatives with PC of which one was <50 years at time of diagnosis. PC-prone gene mutations include *CDKN2A* (familial atypical multiple mole melanoma syndrome), *LKB1* (Peutz Jeghers syndrome), *BRCA1* (hereditary breast and ovarian cancer syndrome), BRCA2 (hereditary breast and ovarian cancer syndrome), mismatch repair genes (Lynch syndrome), and *TP53* (Li-Fraumeni syndrome). Carriers of a *BRCA1/2* mutation, mismatch repair gene, or *TP53* mutation are only eligible when at least two family members are affected by PC. The minimal age for inclusion is 45 years or at least 10 years younger than the age of the youngest relative with PC. Patients with Peutz Jeghers syndrome have to be at least aged 30 years. Before inclusion, all high-risk individuals were extensively evaluated by a clinical geneticist. This evaluation included (1) obtaining a detailed personal and family medical history, (2) verification of clinical diagnoses reported by patients and family members, by review of medical and pathologic records, and by revision of histological slides whenever available, and (3) based on the medical information, genetic testing for the suspected gene mutation(s). Clinical geneticists informed all high-risk individuals that EUS and MRI surveillance was offered as part of a research protocol and that the effectiveness of PC surveillance has not been proven yet. In this counseling, the possibility of false-positive and false-negative outcomes of the PC screening was also explained, as well as a possible cancer diagnosis, or findings of undetermined significance.

Procedure of PC surveillance study

Enrolment in the PC surveillance study started in October 2006 and is currently ongoing in four Dutch medical centers (Erasmus MC-University Medical Center in Rotterdam, University Medical Center Groningen, Academic Medical Center in Amsterdam and The Netherlands Cancer Institute-Antoni van Leeuwenhoek Hospital in Amsterdam). Surveillance entails EUS and MRI. Both tests are scheduled on different days, at maximum 2 weeks apart. EUS is performed under conscious sedation (midozalam/fentanyl). Individuals without pancreatic abnormalities and individuals with a small cystic lesion without malignant features are scheduled for annual follow-up. Whenever EUS and/or MRI detect an abnormality, management is based on consensus agreement of an expert panel (experienced endosonographers, surgeons, and radiologists). This management strategy can either be (1) surgical resection in case of a highly suspicious lesion (solid lesion, main duct intraductal papillary mucinous neoplasm [IPMN], or branch type IPMN >30 mm and/or with malignant features) or (2) shortening of the follow-up interval to 3 months.

Psychological questionnaire study

Since October 2008, a questionnaire study was added to this PC surveillance study. The institutional review boards of the participating hospitals approved the psychosocial questionnaire study. All participants of the PC surveillance study received a letter of invitation by their gastroenterologist and a questionnaire 4 weeks after receiving their surveillance results. Those who did not respond to the initial letter of invitation were sent a reminder letter and a copy of the questionnaire approximately 2 weeks later.

Measurements

Sociodemographic and clinical data. Data were obtained by medical records and our questionnaire on age, sex, marital status, offspring, level of education, personal cancer history, family cancer history, genetic background, surveillance results, and surveillance follow-up policy.

Family history of PC. Participants were asked whether and, if so, how many FDR (i.e., parents, siblings, or children) and SDR (i.e., uncles, aunts, grandparents, nieces, and nephews) ever had cancer. Parallel questions were posed regarding the death of a FDR and/or SDR due to cancer (i.e., At what age did a close relative die of (pancreatic) cancer?).

Participants' view on surveillance

Motivations to undergo PC surveillance. Participants were asked to select from a checklist their motive(s) for undergoing PC surveillance. Space was also provided for additional reasons not listed in the checklist (18).

Attitudes toward, and experiences with, participation in PC surveillance. A 16-item questionnaire comprising four subscales was used, assessing communication (with the physician), reassurance, nervous anticipation, and specific perceived disadvantages (19). Furthermore, specific questions about experiences with each of the surveillance interventions (EUS and MRI) were developed by our group, for example: "How did you experience undergoing a MRI? Was this experience: not uncomfortable, slightly uncomfortable, very uncomfortable or extremely uncomfortable."
Benefits and barriers. The perceived benefits and barriers to PC surveillance were assessed with six questions adapted from previous work (20,21).
Perceived risk. Respondents were asked to report their perceived risk of developing cancer (again) compared with that of an average person in the Dutch population of their age (item adapted from Lerman et al. (22)). Response categories ranged from "lower" to "much higher." Furthermore, participants were asked on a scale from 0 to 100 what they thought their chance was of developing PC with and without undergoing yearly PC surveillance.

Psychological distress
Cancer-related worries. Cancer-related worries were assessed with the eight-item Cancer Worry Scale (23–25). Scores range from 8 to 32, with higher scores indicating more frequent worries about cancer. Internal consistency in this study was indicated by a Cronbach's alpha of 0.84, which is considered high.
Anxiety and depression. Anxiety and depression levels were measured with the 14-item Hospital Anxiety and Depression Scale (HADS) (26–28). Generalized anxiety (HADS-A) and depression (HADS-D) were measured with two seven-item subscales. Response options range from 0 (not at all) to 3 (very much), adding to a maximum score of 21 for each subscale. A score of >11 on a subscale reflects a high level of anxiety or depression and is considered a clinically significant disorder. A score between 8 and 10 is defined as a "moderate level of distress," suggesting a mild disorder. Cronbach's alpha in this study was 0.82 for the Anxiety Subscale and 0.80 for the Depression Subscale.

Data analyses
Descriptive statistics was generated to describe the study sample in terms of sociodemographic and clinical backgroundvcharacteristics, to report on the experiences with the surveillancevinterventions, and to document the prevalence of psychologicalvdistress.
Depending on the level of measurement, χ^2 test or Student's t test was used to identify sociodemographic (i.e., age, gender, education, marital status, and offspring), clinical (i.e., history of cancer and surveillance result), or psychological (i.e., risk perception and experiences with MRI and EUS) variables significantly associated with anxiety at the univariate level, using "a low level of anxiety" (scores between 0 and 7 on the anxiety

scale of the HADS) and "moderate to high levels of anxiety" (scores >8) as dependent variable. All analyses were conducted using the Statistical Package for the Social Sciences (version 17.0; SPSS Institute, Chicago, IL).

RESULTS

Response

Of the 81 eligible individuals, 69 (85%) returned a completed questionnaire. None of the nonrespondents had had cancer, whereas 20 (29%) of the respondents had been treated for cancer (P=0.03). No statistically significant differences were found between the respondents and nonrespondents with respect to any other sociodemographic (age and gender) and clinical (genetic background, personal cancer history, surveillance technique undergone, baseline or follow-up surveillance, surveillance results, and type of abnormality found) variables. The 69 respondents stemmed from 50 families.

Characteristics of the study sample

As listed in Table 1, the mean age of the sample was 52 years (range=20–71 years). Men and women were equally represented. Thirty-eight respondents (55%) carried a proven PC-prone gene mutation. Twenty respondents (29%) had been treated for any type of cancer (Table 1). The mean number of relatives with cancer (including FDR, SDR, and third-degree relatives) was 6.8 (range=0–22, SD 3.6). The mean number of relatives with PC was 1.9 (range=0–5, SD 1.10). Nearly all respondents (96%) had undergone both EUS and MRI surveillance investigations. Three individuals (4%) did not undergo MRI, two because of claustrophobia, and in one, MRI was contraindicated (because of a metallic expander in the breast). Twenty-eight respondents (41%) completed the questionnaire after they had undergone their first-time (baseline) surveillance; all others had already undergone at least one surveillance investigation before. Thus far, there are no dropouts in the surveillance program. In 27 respondents (39%), EUS and/or MRI detected an abnormality. The most frequent detected abnormalities were cystic lesions. In 20 individuals, a total of 35 cystic lesions were detected by EUS and/or MRI. The median size of the cystic lesions was 5.5 mm (SD: 3.6, range: 2–18 mm). None of the cysts showed malignant features. In one individual, a solid lesion was detected, which was morphological suspicious for a malignancy and, therefore, surgically resected. Pathologic examination did not reveal a malignancy, but premalignant lesions (PanIN-2 and an incipient IPMN) were detected. Five individuals were rescheduled for interval investigations after 3 months, four because of the detection of a lesion of undetermined significance and one because of the suspicion of a newly developed cyst found during follow-up investigations.

	Respondents (N=69)		Nonrespondents (N=12)		p
	Mean (range)	**SD**	**Mean (range)**	**SD**	
Age:	52 (20-71)	9.6	49 (34-63)	8.0	.26
	N	**%**	**N**	**%**	
Gender:					
Male	32	46	6	50	.82
Female	37	54	6	50	
Level of education:					
Primary school	3	4			
High school	39	56	-	-	-
College or university	27	39			
Marital status:					
Married/partner	58	84	-	-	-
Single/divorced	11	16			
Genetic background individuals:					
Familial Pancreatic Cancer	31	45	6	50	.75
Hereditary Tumor Syndromes	38	55	6	50	
Hereditary Tumor Syndromes - individuals:					
CDKN2A/CDK4	21	30	1	8	
STK11	4	6	0	0	-
BRCA1	1	1	3	25	
BRCA2	10	15	2	17	
p53	2	3	0	0	
Personal cancer history:					
No	49	71	12	100	.03
Yes	20	29	0	0	
Cancer types:	N=20		N=0		
Breast cancer	4	20			
Ovarian cancer	0	0			
Cervical cancer	1	5			
Melanoma*	13	65	-	-	-
Other skin cancers	6	30			
Colon cancer	0	0			
Lung cancer	0	0			
* Three individuals had melanoma and another type of skin cancer and one individual had melanoma and breast cancer					

Table 1. Characteristics of the respondents (n=69) and the nonrespondents (n=12)

	Respondents (N=69)		Nonrespondents (N=12)		p
	N	**%**	**N**	**%**	
Genetic background families:	N=50		N=10		
Familial Pancreatic Cancer families	22	44	5	50	-
Hereditary Tumor Syndrome families	28	56	5	50	
Hereditary Tumor Syndromes - families:	N=28		N=5		
CDKN2A/CDK4	16	32	1	20	
STK11	4	8	0	0	
BRCA1	2	4	2	40	
BRCA2	5	10	2	40	
p53	1	2	0	0	
	Mean (range)	**SD**	**Mean (range)**	**SD**	**p**
Number of relatives with cancer:					
Total (1st, 2nd and 3rd degree)	6.8 (0-22)	3.6	-	-	-
1st degree	2.6 (0-7)	1.5			
Number of relatives with pancreatic cancer:					
Total (1st, 2nd and 3rd degree)	1.9 (0-5)	1.10	-	-	-
1st degree	1.2 (0-3)	1.0			
	N	**%**	**N**	**%**	**p**
Surveillance technique undergone:					
EUS and MRI	66	96	12	100	.46
EUS only	3	4	0	0	
MRI only	0	0	0	0	
Baseline or follow-up PC-surveillance:					
Baseline (first surveillance)	28	41	5	42	.99
Follow-up (underwent surveillance before)	41	59	7	58	
Results PC-surveillance:					
No abnormality	42	61	8	67	.58
Abnormality → no consequence	21	30	2	17	
Abnormality → interval-EUS	5	7	2	17	
Abnormality → surgical resection	1	1	0	0	
Type of abnormality found in surveillance:					
Cystic lesion	20	74	2	50	.40
Solid lesion	1	4	0	0	
Lesion of undetermined significance	4	15	2	50	
> 3 out of 9 chronic pancreatitis features	2	7	0	0	

Continuation Table 1. Characteristics of the respondents (n=69) and the nonrespondents (n=12)

Motivations to participate

As listed in Table 2, all respondents reported that a reason to participate in the PC surveillance program was that surveillance might lead to early detection of PC in a stage when it is still curable. Contributing to scientific research was the second most frequently reported motivation. When asked for their opinion about the effectiveness of PC surveillance, 43 respondents (62%) reported that a tumor in the pancreas can "certainly" be detected by EUS and MRI, and 25 (36%) reported that a tumor in the pancreas can "probably" be detected by EUS and MRI (not in the table).

	n	%
Cancer might be detected early and still be treatable	69	100
To contribute to scientific research	53	77
Because of surveillance my fear of cancer decreases	13	19
Gives me a sense of control over my body	10	15
I was referred by a physician	7	10
A family member asked me to undergo surveillance	7	10
Self reported other reasons: Because relatives died of pancreatic cancer For their children	8 5 2	12 7 3

Table 2. Motivations to participate in the pancreatic cancer surveillance program (n=69) (more than one answer was allowed)

Experiences with EUS and MRI

Seventeen respondents (25%) had experienced the EUS and/or the MRI investigation as very to extremely uncomfortable. Of these respondents, three experienced both EUS and MRI as very to extremely uncomfortable. Seven respondents (10%) experienced only EUS as very to extremely uncomfortable, mostly because the sedation was experienced as inadequate or related to postsedation effects as prolonged drowsiness. Seven other respondents (11%) reported MRI to be very or extremely uncomfortable, predominantly because of claustrophobia. There was no statistically significant difference in the frequency that respondents were dreading one of the two procedures. In Table 3, detailed information on experiences and attitudes toward PC surveillance is presented. One fifth of the respondents reported to be nervous before a follow-up visit and to dread the follow-up visits. However, only five respondents (7%) preferred follow-up visits less frequently. With respect to the general disadvantages, only 10 (14%) of the respondents experienced the investigations as burdensome and 12 (17%) reported that the follow-up visits reminded them of PC while they would rather think less often about it. Approximately 70% of the respondents reported that the surveillance investigations gave them a sense of security and that they would worry more about the disease if there were no follow-up visits. Approximately 90% said that perceived advantages of follow-up outweighed perceived disadvantages.

	Rather/Very much	
	n	%
Communication		
Can you ask about things at follow-up?	56	81
At follow-up, can you discuss with your doctor matters that are of concern to you or worries you?	54	78
Do people in the hospital pay attention to what you say?	65	94
Do the physicians at follow-up in the hospital have enough time for you?	59	86
Nervous anticipation		
Are you nervous before a follow-up visit?	14	20
Do you sleep less well in the week before follow-up?	8	11
Do you postpone plans till after the follow-up visit?	4	6
Do you normally dread the follow-up visits?	13	19
Would you rather have follow-up visits less frequently?	5	7
Reassurance		
Do the follow-up visits convey you a sense of security?	47	68
Are you reassured after the follow-up visit?	55	80
Do the advantages of follow-up outweigh the disadvantages?	61	88
Would you worry more about your disease if there was no follow-up?	50	72
General disadvantages		
Would you prefer, if possible, to have follow-up visits in a hospital closer by?	17	25
Do you think the investigations at follow-up burdensome?	10	14
Does the follow-up remind you each time of your disease, while you'd rather think less often about it?	12	17

Table 3. Experiences with, and attitudes towards pancreatic cancer surveillance (n=69)

PC risk perception

Forty respondents (58%) perceived their risk of developing PC as moderately to extremely elevated compared with the general population. Thirty-seven respondents (54%) reported a lower personal risk percentage if participating in surveillance compared with not participating in surveillance, whereas 30 respondents (43%) reported the same personal risk to develop PC with or without surveillance.

Psychological distress

Cancer worries

Respondents worried most about the possibility of getting cancer (n = 20, 29%), and 17 individuals (25%) worried about the chance of family members developing cancer (Table 4). There was no correlation between PC surveillance results and cancer worries. Even those individuals in whom the positive PC surveillance results led to a change of management (n= 1 surgical resection, n=5 shortening of follow-up interval) did not experience more concerns about cancer. In the majority of respondents (99%), cancer worries did not affect their mood and did not interfere with their daily activities.

During the past week	Often/always worried	
	n	%
How often have you thought about your chance of getting cancer (again)?	8	12
Have these thoughts affected your mood?	1	1
Have these thoughts interfered with your ability to do daily activities?	1	1
How concerned are you about the possibility of getting cancer one day?	20	29
How often do you worry about developing cancer?	9	13
How much of a problem is this worry?	5	7
How often do you worry about the chance of family members developing cancer?	17	25
How concerned are you about the possibility that you will ever need surgery (again)?	7	10

Table 4. Cancer worries. Items of adapted Cancer Worry Scale (n=69)

Anxiety and depression

The mean scores of the HADS subscales were 4.2 on the anxiety scale (range: 0–14, SD = 3.7) and 3.0 on the depression scale (range: 0–13, SD = 3.2). Scores above 10 on the HADS subscales indicate a significant clinical level of anxiety or depression and were represented in six respondents (9%). One of them scored above cutoff on the depression subscale, two of them scored above the cutoff on the anxiety subscale, and three respondents scored above the cutoff on both the anxiety and the depression subscales.

In Table 5, the anxiety scale of the HADS is divided into two groups: (1) low-anxiety levels (scores 0 –7) and (2) moderate to high anxiety levels (score =/> 8). Fifty-eight participants had low-anxiety levels, and 11 participants had moderate to high anxiety levels. None of the sociodemographic (i.e., age, gender, education, marital status, and offspring), clinical (i.e., history of cancer, surveillance result, and approaching the age at which a close relative died of cancer), or psychological (i.e., risk perception and experiences with MRI and EUS) variables were significantly associated with moderate to high levels of anxiety except for "worrying about follow-up investigations" (P = 0.04). Having worries about the next MRI was significantly associated with higher levels of anxiety.

Furthermore, individuals with a positive surveillance result (abnormalities found during surveillance) were not more anxious, depressed, and did not have more worries about developing cancer, than individuals with a negative surveillance result.

DISCUSSION

For a surveillance program to be effective, it is not only important to use sensitive screening techniques but it is also crucial that participants adhere to the program. Adherence to a surveillance program is influenced by one's experiences with the program, and therefore, insight in the psychological experiences with this surveillance program is of great importance. Insight in these experiences is particularly relevant for this high-risk group as most individuals have experienced multiple losses due to PC, contributing

	HADS anxiety low (*N* = 58), N(%)	HADS-anxiety moderate- high (*N* = 11), N(%)	*p*
Sociodemographics			
Age			
20-29 years	1 (100)	0 (0)	.31
30-39 years	4 (80)	1 (20)	
40-49 years	15 (71)	6 (29)	
50-59 years	21 (84)	4 (16)	
60-69 years	16 (100)	0 (0)	
70-79 years	1 (100)	0 (0)	
Age; approaching age close relative died of PC:			
0-5 years	18 (90)	2 (10)	.40
5 years	25 (86)	4 (14)	
No first degree relative with PC	15 (75)	5 (25)	
Gender:			
Female	29 (78)	8 (22)	.17
Male	29 (91)	3 (9)	
Level of education:			
Primary school	8 (80)	2 (20)	.93
High school	27 (85)	5 (16)	
College/university	23 (85)	4 (15)	
Marital status:			
Married/common-law	50 (86)	8 (14)	.26
Single/divorced/separated	8 (73)	3 (27)	
Offspring:			
Yes	51 (82)	11 (18)	.22
No	7 (100)	0 (0)	
Mutation status:			
Carrier PC-associated gene mutation	32 (84)	6 (16)	.97
No underlying gene-mutation	26 (84)	5 (16)	
Personal history of cancer			
Yes	15 (75)	5 (25)	.19
No	43 (88)	6 (12)	
Family history of cancer (first degree)			
Yes; one or more	40 (87)	6 (13)	.35
No	18 (78)	5 (22)	
Family history of cancer (total; $1^{st} - 3^{rd}$ degree)			
Yes; one or more	57 (84)	11 (16)	.66
No	1 (100)	0 (0)	
Family history of PC (first degree)			
Yes; one or more	44 (88)	6 (12)	.15
No	14 (74)	5 (26)	

Table 5. Associations of psychological and clinical data with anxiety (n = 69)

	HADS anxiety low (*N* = 58), N(%)	HADS-anxiety moderate- high (*N* = 11), N(%)	*p*
Surveillance result			
Normal test result	34 (81)	8 (19)	.84
Abnormality without further consequences	19 (90)	2 (11)	
Interval-EUS because of positive test result	4 (83)	1 (17)	
Abnormality; surgery	1 (100)	0 (0)	
Experiences surveillance methods			
MRI-experiences (n=65; 1 missing and 3 didn't undergo MRI)			
Not to slightly uncomfortable	48 (87)	7 (13)	.16
Very to extremely uncomfortable	7 (70)	3 (30)	
EUS-experiences (n=68; 1 missing)			
Not to slightly uncomfortable	50 (86)	8 (14)	.20
Very to extremely uncomfortable	7 (70)	3 (30)	
Worrying about/dreading follow-up investigations			
MRI (n=69)			
Not at all to a little	54 (87)	8 (13)	.04
Much to very much	4 (57)	3 (43)	
EUS (n=69)			
Not at all to a little	53 (87)	8 (13)	.08
Much to very much	5 (63)	3 (38)	
Risk perception (n=69)			
Chance of getting cancer compared to general population			
Lower risk	4 (100)	0 (0)	.63
Same risk	7 (88)	1 (13)	
Slightly tot extremely elevated risk	47 (84)	10 (16)	
	Mean (SD)	Mean (SD)	
Chance of getting cancer with regular surveillance (0-100)	32 (28)	37 (29)	.62

Continuation Table 5. Associations of psychological and clinical data with anxiety (n=69)

to a higher psychological burden of undergoing these procedures. Second, because of their lifelong PC risk, they should adhere to the screening on a lifelong regimen. Because of this repetitive nature of surveillance, it is of great importance that the burden of the procedures in this high-risk group is studied in detail, allowing possible adaptations in the procedure in a way that they are well tolerated by high-risk individuals. To our knowledge, this is the first study to investigate the specific experiences, such as the perceived burden, of high-risk individuals with a PC surveillance program, consisting of annualsurveillance by MRI and EUS. Our results show that EUS-MRI-based PC surveillance among individuals at high risk for developing PC is feasible from a psychological point of view. This is supported by the fact that the majority of respondents did not experience surveillance by EUS (with sedation) and MRI as psychologically too burdensome.

One of the aims of this study was to investigate the motivation of high-risk individuals to participate in a PC surveillance program. Although the effectiveness of PC surveillance seems promising based on theoretical reasoning and preliminary (pre) clinical data, we currently lack longterm results that indicate that PC surveillance will actually prevent people from dying of PC. This unproven efficacy was extensively discussed with all potential participants before they decided whether to participate in the surveillance study. Nevertheless, the most frequently reported reason to participate was that surveillance might lead to early detection of PC at a stage when it is still curable. All respondents indicated this reason as being one of their motivations to participate. It is important to realize that because of the posttest design of our study, these results are based on information from individuals who had decided to participate in the PC surveillance program. Nonparticipants may not believe in the ability of early detection of PC. Future results of our ongoing prospective psychological study that includes both participants and nonparticipants of the surveillance study will provide information about reasons not to participate. Preliminary results of this ongoing prospective study show that only a small proportion (14%) of the high-risk individuals decline participation in the PC surveillance program (unpublished data). We, therefore, expect that our current data are not severely biased with respect to those reporting a favorable attitude. Furthermore, this study has a high response rate (85%), suggesting that the results are representative for the total group of high-risk individuals participating in the PC surveillance program.

At present, EUS is the most promising PC surveillance technique (7,9,29). Compared with MRI, EUS is an invasive technique, and for this reason, we hypothesized that the acceptability of EUS would be lower compared with MRI. Remarkably, we found that EUS and MRI were regarded as equally burdensome. One explanation might be the routine use of conscious sedation for EUS. Another perspective was given by Lewis *et al.* (14), who stated that individuals with a family history of PC or a personal history of cancer often prefer the more invasive surveillance techniques.

Another aim of this study was to investigate the general psychological distress participants might experience and the extent to which levels of anxiety are related to participating

in the surveillance program. With respect to cancer-specific worries, Maheu *et al.* (15) reported that pretest cancer worries remained the most important predictor for cancer worries after undergoing PC surveillance. This suggests that the frequency of cancer worries reported in this study by high-risk individuals after undergoing PC surveillance may have been the same before undergoing surveillance. This is in concordance with the finding that the surveillance result itself did not have an impact on the level of cancer worries. Despite the fact that approximately a quarter of respondents worried about the possibility of getting cancer themselves or worried about their relatives developing cancer, these worries did not interfere with their daily activities, suggesting that respondents seem to cope well with these worries.

Anxiety and depression scores at a level that does indicate a need for professional psychosocial care were present in approximately 10% of the respondents. This is comparable with the proportion of individuals in the general Dutch population (30) and suggests that the levels of distress found in respondents are not a result of participation in the surveillance program but may have other causes.

In contrast to our expectations, we did not find a statistically significant association between surveillance results and levels of anxiety. The only factor that was significantly associated with a higher level of anxiety was anticipating worries about undergoing follow-up MRI.

In this study, respondents completed questionnaires after receiving their surveillance results. Therefore, it is not possible to investigate possible changes in distress levels and risk perception as a result of participation in the PC surveillance program. Future results of our ongoing prospective psychological study, which includes both participants and nonparticipants, will provide more information on possible causal relationships between the surveillance program and participants' psychological well-being. These results in combination with results of additional studies will hopefully shed greater light on perceived burden of PC surveillance, the perceived distress of surveillance, motivations to participate, and the emotional response to the test results.

In summary, results of our study indicate that PC surveillance by EUS and MRI is feasible from a psychological point of view. Although EUS is more invasive than MRI, there is no significant difference in the percentage of respondents who perceived one of the surveillance methods as more burdensome. Although almost one third of respondents worry frequently about cancer and a minority of respondents actually have anxiety and depression levels that indicate clinically significant disorder, there was no association with surveillance results, and the large majority of participants expressed a positive attitude toward the PC surveillance program.

REFERENCES

1. Edwards BK, Brown ML, Wingo PA, et al. Annual report to the nation on the status of cancer, 1975–2002, featuring population-based trends in cancer treatment. J Natl Cancer Inst 2005;97:1407–1427.
2. Jemal A, Siegel R, Ward E, et al. Cancer statistics, 2009. CA Cancer J Clin 2009;59:225–249.
3. National Cancer Institute. Surveillance epidemiology and end results. Available at: http://seer.cancer.gov/statfacts/html/pancreas. Accessed March 15, 2011.
4. Integrale Kanker Centra N. Kankerregistratie. Available at: www.ikcnet.nl. Accessed March 15, 2011.
5. Brand RE, Lerch MM, Rubinstein WS, et al. Advances in counselling and surveillance of patients at risk for pancreatic cancer. Gut 2007;56:1460–1469.
6. Klein AP, Brune KA, Petersen GM, et al. Prospective risk of pancreatic cancer in familial pancreatic cancer kindreds. Cancer Res 2004;64:2634–2638.
7. Canto MI, Goggins M, Hruban RH, et al. Screening for early pancreatic neoplasia in high-risk individuals: a prospective controlled study. Clin Gastroenterol Hepatol 2006;4:766 –781.
8. Langer P, Kann PH, Fendrich V, et al. Five years of prospective screening of high-risk individuals from families with familial pancreatic cancer. Gut 2009;58:1410 –1418.
9. Poley JW, Kluijt I, Gouma DJ, et al. The yield of first-time endoscopic ultrasonography in screening individuals at a high risk of developing pancreatic cancer. Am J Gastroenterol 2009;104:2175–2181.
10. Rulyak SJ, Brentnall TA. Inherited pancreatic cancer: surveillance and treatment strategies for affected families. Pancreatology 2001;1:477– 485.
11. Verna EC, Hwang C, Stevens PD, et al. Pancreatic cancer screening in a prospective cohort of high-risk patients: a comprehensive strategy of imaging and genetics. Clin Cancer Res 2010;16:5028 –5037.
12. Vasen HF, Wasser M, van Mil A, et al. Magnetic resonance imaging surveillance detects early-stage pancreatic cancer in carriers of a p16-Leiden mutation. Gastroenterology 2011;140:850–856.
13. Wilson JM, Jungner YG. [Principles and practice of mass screening for disease] Principios y metodos del examen colectivo para identificar enfermedades. Bol Oficina Sanit Panam 1968;65:281–393.
14. Lewis ZK, Frost CJ, Venne VL. Pancreatic cancer surveillance among high-risk populations: knowledge and intent. J Genet Couns 2009;18:229–238.
15. Maheu C, Vodermaier A, Rothenmund H, et al. Pancreatic cancer risk counselling and screening: impact on perceived risk and psychological functioning. Fam Cancer 2010;9:617– 624.

16. Axilbund JE, Brune KA, Canto MI, et al. Patient perspective on the value of genetic counselling for familial pancreas cancer. Hered Cancer Clin Pract 2005;3:115–122.
17. Bleiker EM, Menko FH, Taal BG, et al. Screening behavior of individuals at high risk for colorectal cancer. Gastroenterology 2005;128:280 –287.
18. Bleiker EM, Aaronson NK, Menko FH, et al. Genetic counseling for hereditary cancer: a pilot study on experiences of patients and family members. Patient Educ Couns 1997;32:107–116.
19. Stiggelbout AM, de Haes JC, Vree R, et al. Follow-up of colorectal cancer patients: quality of life and attitudes towards follow-up. Br J Cancer 1997; 75:914 –920.
20. Champion VL. Instrument development for health belief model constructs. Adv Nurs Sci 1984;6:73– 85.
21. Kash KM, Holland JC, Osborne MP, Miller DG. Psychological counseling strategies for women at risk for breast cancer. J Natl Cancer Inst 1995;17:73–79.
22. Lerman C, Kash K, Stefanek M. Younger women at increased risk for breast cancer: perceived risk, psychological well-being, and surveillance behavior. Monogr Natl Cancer Inst 1995;16:171–176.
23. Lerman C, Daly M, Masny A, et al. Attitudes about genetic testing for breast-ovarian cancer susceptibility. J Clin Oncol 1994;12:843– 850.
24. Douma KF, Aaronson NK, Vasen HF, et al. Psychological distress and use of psychosocial support in familial adenomatous polyposis. Psychooncology 2010;19:289 –298.
25. Lammens CR, Aaronson NK, Wagner A, et al. Genetic testing in Li-Fraumeni syndrome: uptake and psychosocial consequences. J Clin Oncol 2010;28:3008 –3014.
26. Bjelland I, Dahl AA, Haug TT, et al. The validity of the Hospital Anxiety and Depression Scale. An updated literature review. J Psychosom Res 2002;52:69 –77.
27. Zigmond AS, Snaith RP. The hospital anxiety and depression scale. Acta Psychiatr Scand 1983;67:361–370.
28. Spinhoven P, Ormel J, Sloekers PP, et al. A validation study of the Hospital Anxiety and Depression Scale (HADS) in different groups of Dutch subjects. Psychol Med 1997;27:363–370.
29. Canto MI, Goggins M, Yeo CJ, et al. Screening for pancreatic neoplasia in high-risk individuals: an EUS-based approach. Clin Gastroenterol Hepatol 2004;2:606–621.
30. de Graaf R, Ten Have M, van Gool C, et al. Prevalence of mental disorders and trends from 1996 to 2009. Results from the Netherlands Mental Health Survey and Incidence Study-2. Soc Psychiatry Psychiatr Epidemiol [published online ahead of print January 1, 2011]. doi: 10.1007/s00127-010-0334-8.

CHAPTER 12
SUMMARY, GENERAL DISCUSSION & FUTURE PERSPECTIVES

This thesis deals with inherited and familial pancreatic cancer. In this final chapter we will discuss results and novel insights obtained from our research projects, highlight areas requiring further investigation, and give a forecast on the future prospects of surveillance of pancreatic cancer in high risk individuals

NOVEL INSIGHTS

This thesis starts with appraising the validity of pancreatic screening of high-risk individuals by applying the principles of screening and practice for disease as proposed by Wilson and Jungner (1) (**Chapter 2**). Through this appraisal, we conclude that screening these high-risk individuals by endoscopic ultrasound (EUS) and magnetic resonance imaging (MRI) is a promising approach. However, this appraisal also clearly high-lights important questions that need to be answered in order to make a final judgement whether the benefits of screening (reduction of pancreatic cancer related mortality and gains in life years) outweigh the negative side-effects of over-diagnosis, over-treatment and costs. In this thesis, answers to some of these questions are provided.

PART I - SURVEILLANCE OF INDIVIDUALS AT HIGH RISK OF DEVELOPING PANCREATIC CANCER

When work on this thesis started, both EUS and MRI were considered the most accurate tests for pancreatic imaging within a screening setting (2-4). However, it was unknown whether one of these two tests was sufficient, and/or superior or whether both tests were complementary. None of the trials that were conducted were blinded (2-5). Consequently, a direct comparison was not impossible. **Chapter 3** presents the results of our prospective head-to-head blinded comparison of EUS and MRI for the detection of clinically relevant pancreatic lesions at first time screening in individuals at high risk for developing pancreatic cancer. These individuals include mutation carriers of pancreatic cancer prone gene mutations and first degree relatives (FDR) of patients with familial pancreatic cancer (FPC) (see **Chapter 7**). A total of 11 clinically relevant lesions (two solid lesions and nine cystic lesions larger than 10 millimeter) were detected in nine out of 139 high-risk individuals (6%). Six of these 11 lesions (55%) were detected by both tests. EUS detected a total of 8 (73%) and MRI detected a total of 9 (82%) clinically relevant lesions. EUS proved to be particularly sensitive for the detection of small solid lesions. Two solid lesions detected by EUS, including a stage I pancreatic cancer were missed by MRI. MRI was particularly sensitive for the detection of cystic lesions. All 9 cystic lesions sized ≥10 mm were detected by MRI, whereas EUS detected 6 (66%). Based on our results, EUS and MRI are complementary rather than interchangeable tests in the setting of surveillance of individuals at high risk of developing pancreatic cancer. However, longer term follow-up studies are required to validate the potential of EUS and MRI to detect lesions that progress to advanced neoplasia or early cancer.

Chapter 4 beholds the consensus of an international meeting attended by 50 experts in the fields of gastroenterology, surgery, radiology, pathology and genetics. By organizing this international meeting, we have formulated international consensus statements and recommendations regarding surveillance and management of high-risk individuals with an inherited or familial predisposition for pancreatic cancer. This should help to harmonize current surveillance efforts and serve as a platform for the development and refinement of future multidisciplinary research protocols and guidelines.

It is well-known that decision analytic models are useful to predict the effectiveness of screening programs (6). Through modelling it is possible to scrutinize strategies for prevention and early detection and to highlight key issues and uncertainties to optimize current and future screening strategies. In **Chapter 5** we developed a microsimulation model in order to explore the uncertainties of early detection of pancreatic cancer in high-risk individuals, to analyse the impact of these uncertainties on the effect of screening and consequently highlight the areas for further research. Our explorative modelling study showed that there is potential for pancreatic screening to be effective. Parameters that turned out to be of most importance to influence the outcome of screening were (1) the follow-up strategy of screen positives, (2) the duration of the preclinical stage and (3) the level of pancreatic cancer risk. Our results indicate that screening is most efficient if patients are identified and treated before the disease becomes invasive. Furthermore, an increased duration of the preclinical stage and a higher level of pancreatic cancer risk positively influence the outcome of screening by lowering the mortality rate and interval cancer rate. Interestingly, the effect of the sensitivity of the screen test showed to be of less influence; a 10% increased probability to detect (preinvasive) pancreatic cancer decreased the incidence and mortality rate by <4%, and lowered the number of tests to prevent one pancreatic cancer related death by 6%.

Detection and surgical treatment of high-grade precursor lesions are defined as success of a screening program (**Chapter 4**). Precursor lesions of pancreatic ductal carcinoma include Pancreatic Intraepithelial Neoplasia (PanIN) and Intraductal Papillary Mucinous Neoplasm (IPMN). In **Chapter 6** we focused on these lesions and studied the prevalence of precursor lesions in resection specimen of patients with inherited and familial pancreatic cancer in comparison to sporadic cases. Our data showed that the number of patients with presence of high-grade precursor lesions was significantly higher in the inherited or familial cases compared to sporadic cases. Interestingly, all of these high-grade precursor lesions were PanINs-lesions as high-grade (incipient) IPMNs were not detected. Furthermore, more precursor lesions were detected in inherited or familial cases. Next important step is to investigate how these pathological lesions correlate with morphological features on EUS and MRI. For this, prospectively collected data is needed in which individuals have undergone (repeated) imaging by EUS and MRI prior to resection.

PART II - WHO ARE AT RISK?

An overview of which individuals have an inherited or familial increased risk for developing pancreatic cancer is provided in **Chapter 7**. Briefly, this includes carriers of pancreatic cancer prone gene mutations and first degree relatives of familial pancreatic cancer (FPC) cases. The effectiveness and impact of surveillance will be highest when directed towards individuals who are at high risk for developing pancreatic cancer since the prevalence of high-grade precursor lesions and pancreatic cancer is highest in these individuals. It is therefore of great importance to increase our knowledge about pancreatic cancer susceptibility genes and the actual risk of developing pancreatic cancer within the known pancreatic cancer associated hereditary cancer syndromes.

Therefore, we investigated the usefulness of testing for *CDKN2A* mutations in FPC families not affected by melanomas in **Chapter 8**. The current recommendation is to test for *CDKN2A* mutations in FPC-families only when at least one melanoma case is present (7). It is not recommended to search for *CDKN2A* mutations in FPC-families without melanomas. However, we identified *CDKN2A* mutations in six of 28 FPC families (21.4%), in which three (10.7%) had no melanomas and/or dysplastic nevi reported at the time of DNA analysis. These findings emphasise the need to include *CDKN2A*-mutation analysis in genetic testing for FPC families, even in the absence of reported melanomas.

It recently became clear that the Fanconi gene *FANCN/PALB2* (Partner and localizer of *BRCA2*) should not only be considered as a susceptibility gene for breast cancer (8) but also as a susceptibility gene for pancreatic cancer (9). In **Chapter 9** we therefore aimed to determine the prevalence of *PALB2* in Dutch non-*BRCA1/2* familial pancreatic cancer patients (n=31) and non-*BRCA1/2* familial breast cancer patients with a personal or family history of pancreatic cancer (n=34). We did not identify any *PALB2* mutations in any of the tested patients. These data suggest that there is only a limited role for *PALB2* mutations in both familial pancreatic cancer and familial breast cancer.

Accurate risk estimates for pancreatic cancer in PJS have been lacking in the literature. The reported risks vary widely from a 0 to 132-fold increase (10-13). Consequently, this hampers the counselling of PJS patients and the implementation of surveillance strategies. We therefore aimed in **Chapter 10** to calculate a reliable risk estimate for developing pancreatic cancer in PJS. Using a large, nationwide Dutch PJS patient cohort in which long-term follow-up is available, we calculated a cumulative risk for developing pancreatic cancer of 26% at the age of 70 years, and a relative risk of 76 compared to the general population. The risk for pancreatic-biliary cancer (including pancreatic, distal bile duct and ampullary cancer) was even higher with a cumulative risk of 32% at age 70 years, and a relative risk of 96. These data provide strong evidence to include PJS patients in pancreatic cancer surveillance programs.

PART III - PSYCHOLOGICAL ASPECTS RELATED TO SCREENING

When individuals do not participate in or adhere to a surveillance program because of the perceived high psychological burden of surveillance, the program will ultimately not be successful, even if its yield and outcome from a scientific viewpoint is beneficial. We therefore measured the psychological impact of joining a pancreatic cancer surveillance program in **Chapter 11** by conducting a retrospective questionnaire-study among participants of a EUS-MRI based screening study (discussed in **Chapter3**) (14). In total, 69 individuals (85%) completed the questionnaire. The most frequently reported reason to participate in the program was that screening might lead to early detection of pancreatic cancer or its precursor lesion at a stage when the disease is still curable. Despite the fact that EUS is an invasive technique and MRI is not, both imaging modalities were regarded equally burdensome. Interestingly, those individuals in whom positive screening results led to a change of management (n=1 surgical resection, n=5 shortening of follow-up interval) were not statistically more worried, anxious or depressed than respondents who had imaging results that not prompted a change in management. Based on these results, we concluded that psychologically surveillance in high-risk individuals is both feasible and justified.

In order to further evaluate the psychological burden of pancreatic cancer surveillance we initiated a prospective study to detect possible changes in cancer worries and levels of anxiety and depression over time (15). Preliminary results of this prospective study indicate that: (1) the expected burden of EUS is higher than the perceived burden; and that (2) levels of anxiety, depression and cancer worries are not significantly influenced by participating in the pancreatic cancer screening program.

MY CURRENT VISION ON PANCREATIC SURVEILLANCE AND FUTURE PERSPECTIVES

After having been involved in research on topics related to surveillance of individuals at high risk for developing pancreatic cancer for the past few years, I believe that currently we still cannot definitely answer the ultimate question whether surveillance is effective and if so, by which investigational modality. Although our knowledge on pancreatic cancer and surveillance of high-risk individuals has expanded over the past few years, in particular with regard to pathobiology and molecular biology, there are still large gaps in our knowledge. This pertains the individual risk of persons affected which is most evident in familial pancreatic cancer in which the underlying condition and causal mutation is unknown, the transition risks of early lesions including PanIN and IPMN into advanced neoplasia or early cancer, and the detection and characterisation of such lesions, in particular with regard to high-risk features indicative for malignant transformation, by imaging investigations or molecular tools. For these reasons future studies should focus on the following topics.

Future studies should focus on improving our knowledge which individuals are truly at high risk for developing pancreatic cancer. Efforts should be made to discover additional pancreatic cancer susceptibility genes. Relatively new techniques, such as exomic sequencing (which previously identified *PALB2* as a pancreatic cancer gene (9)), could be of help in such endeavour. In addition, research should focus on determining the absolute pancreatic cancer risk within the different inherited cancer syndromes.

Our understanding of the natural history of pancreatic cancer and its precursor lesions needs to be deepened. For both PanINs and IPMNs it is currently unclear what the probability is for progression to invasive cancer and within which interval/time frame this occurs. Furthermore, it is unknown whether lesions that arise in high-risk individuals have the same biological behavior as lesions seen in sporadic cases. Expanding our knowledge in these areas will provide more insight regarding the 'window of opportunity' for early detection and resection of high-risk lesions and thus the potential for screening and the optimal screening interval.

In addition, we need to acquire more insight into the correlation between morphological features detected on imaging investigations (EUS and MRI) and pathology findings. These insights will lead to an improved ability to determine which lesions can be safely observed with continued surveillance and which lesions justify a timely resection. We still face difficulties to distinguish high-grade lesions and early cancers from low-grade and non-neoplastic lesions. Previous studies have shown that surveillance may also result in the resection of benign lesions that, in hindsight, did not justify resection. (3, 4, 16). For one, currently it is not possible to correctly identify and stage PanIN lesions based on morphological features. Although data have shown that PanIN lesions are possibly correlated with EUS features of chronic pancreatitis (17), it is unclear whether the number and/or type of feature correlates with the degree of dysplasia. Future studies should focus on which of the available imaging tests is superior in detecting clinical relevant lesions. Focus should also be on whether serum markers (18-20), stool markers (21) and/or markers in pancreatic juice (22-25) are of additional value.

Another important issue that needs to be resolved is whether the Sendai criteria also apply to IPMN-like lesions found in high-risk individuals rather than in sporadic IPMN (26, 27). The Sendai criteria are international guidelines for management of IPMNs. The majority of surveillance studies apply the Sendai criteria in order to decide whether or not to resect a cystic lesion. However, high-grade PanIN lesions have been found in pancreata of high-risk individuals who had resections of cystic lesions that did not meet these criteria (4, 28, 29). If these findings are confirmed in larger studies, the Sendai criteria have to be reconsidered in high-risk individuals.

Lastly, I believe that follow-up of affected individuals should be performed only within well-defined research programmes. Pooling data from various patients cohorts around the world is required to reach sufficient numbers for meaningful statistical analysis and accurate estimation of risk reduction and survival benefit. For this purpose we have developed an international web-based registry, which will be launched in the nearby future. Most importantly, sufficient follow-up time is required in order to judge the true potential of surveillance of high-risk individuals to prevent pancreatic cancer. Given an average age of 65 at which time inherited pancreatic cancer develops, a median age of high risk individuals of 52 at the time of inclusion in surveillance programs, and an estimated 15 years it takes from the first mutation to progress to metastatic pancreatic cancer, it is predictable that clinically relevant lesions will develop with increasing frequency in these cohorts in the upcoming years. To achieve our final goals we need to exercise patience, be persistent in our research efforts and remember that it took twenty years to prove that screening for colorectal cancer improves survival.

CONCLUSION

Pancreatic cancer is one of the most fatal malignancies known to mankind. Of all human cancers its incidence ranks 10th, but from a viewpoint of the fatality pancreatic cancer ranks 4th. A well-defined group of individuals are at a particular high risk of developing this disease. In the last decade, surveillance programmes in these high-risk individuals have been initiated in order to detect precursor lesions or early asymptomatic pancreatic cancer.

Early detection in individuals at high risk to develop pancreatic cancer is a promising approach to fight the tremendous burden and high death toll of this devastating disease. Although appealing and supported by novel pathophysiological insights into the development of pancreatic cancer, ongoing research efforts are needed to determine whether pancreatic cancer surveillance is truly effective and ultimately lowers mortality.

REFERENCES

1. Wilson JM, Jugner G. Principles and Practice of Screening for Disease. Geneva:World Health Organisation. 1968.
2. Brentnall TA, Bronner MP, Byrd DR, Haggitt RC, Kimmey MB. Early diagnosis and treatment of pancreatic dysplasia in patients with a family history of pancreatic cancer. Ann Intern Med. 1999;131(4):247-55. Epub 1999/08/24.
3. Canto MI, Goggins M, Hruban RH, Petersen GM, Giardiello FM, Yeo C, et al. Screening for early pancreatic neoplasia in high-risk individuals: a prospective controlled study. Clin Gastroenterol Hepatol. 2006;4(6):766-81; quiz 665. Epub 2006/05/10.
4. Canto MI, Goggins M, Yeo CJ, Griffin C, Axilbund JE, Brune K, et al. Screening for pancreatic neoplasia in high-risk individuals: an EUS-based approach. Clin Gastroenterol Hepatol. 2004;2(7):606-21. Epub 2004/06/30.
5. Kimmey MB, Bronner MP, Byrd DR, Brentnall TA. Screening and surveillance for hereditary pancreatic cancer. Gastrointestinal endoscopy. 2002;56(4 Suppl):S82-6. Epub 2002/09/26.
6. van Oortmarssen GJ, Boer R, Habbema JD. Modelling issues in cancer screening. Stat Methods Med Res. 1995;4(1):33-54. Epub 1995/03/01.
7. Brand RE, Lerch MM, Rubinstein WS, Neoptolemos JP, Whitcomb DC, Hruban RH, et al. Advances in counselling and surveillance of patients at risk for pancreatic cancer. Gut. 2007;56(10):1460-9.
8. Rahman N, Seal S, Thompson D, Kelly P, Renwick A, Elliott A, et al. PALB2, which encodes a BRCA2-interacting protein, is a breast cancer susceptibility gene. Nature genetics. 2007;39(2):165-7. Epub 2007/01/04.
9. Jones S, Hruban RH, Kamiyama M, Borges M, Zhang X, Parsons DW, et al. Exomic sequencing identifies PALB2 as a pancreatic cancer susceptibility gene. Science. 2009;324(5924):217. Epub 2009/03/07.
10. Giardiello FM, Brensinger JD, Tersmette AC, Goodman SN, Petersen GM, Booker SV, et al. Very high risk of cancer in familial Peutz-Jeghers syndrome. Gastroenterology. 2000;119(6):1447-53. Epub 2000/12/13.
11. Hearle N, Schumacher V, Menko FH, Olschwang S, Boardman LA, Gille JJ, et al. Frequency and spectrum of cancers in the Peutz-Jeghers syndrome. Clin Cancer Res. 2006;12(10):3209-15. Epub 2006/05/19.
12. Mehenni H, Resta N, Park JG, Miyaki M, Guanti G, Costanza MC. Cancer risks in LKB1 germline mutation carriers. Gut. 2006;55(7):984-90. Epub 2006/01/13.
13. You YN, Wolff BG, Boardman LA, Riegert-Johnson DL, Qin R. Peutz-Jeghers syndrome: a study of long-term surgical morbidity and causes of mortality. Fam Cancer. 2010;9(4):609-16. Epub 2010/06/24.
14. Harinck F, Nagtegaal T, Kluijt I, Aalfs C, Smets E, Poley JW, et al. Feasibility of a pancreatic cancer surveillance program from a psychological point of view. Genet Med. 2011;13(12):1015-24. Epub 2011/08/23.

15. Harinck F, Nagtegaal T, Kluijt I, Aalfs CM, Smets E, Poley J-W, et al. Su1816 Prospective Evaluation of Psychological Impact of Pancreatic Cancer Surveillance in High-Risk Individuals. Gastroenterology. 2012;142(5, Supplement 1):S-511.
16. Langer P, Kann PH, Fendrich V, Habbe N, Schneider M, Sina M, et al. Five years of prospective screening of high-risk individuals from families with familial pancreatic cancer. Gut. 2009;58(10):1410-8. Epub 2009/05/28.
17. Brune K, Abe T, Canto M, O'Malley L, Klein AP, Maitra A, et al. Multifocal neoplastic precursor lesions associated with lobular atrophy of the pancreas in patients having a strong family history of pancreatic cancer. Am J Surg Pathol. 2006;30(9):1067-76. Epub 2006/08/26.
18. Brand RE, Nolen BM, Zeh HJ, Allen PJ, Eloubeidi MA, Goldberg M, et al. Serum biomarker panels for the detection of pancreatic cancer. Clin Cancer Res. 2011;17(4):805-16. Epub 2011/02/18.
19. Faca VM, Song KS, Wang H, Zhang Q, Krasnoselsky AL, Newcomb LF, et al. A mouse to human search for plasma proteome changes associated with pancreatic tumor development. PLoS medicine. 2008;5(6):e123. Epub 2008/06/13.
20. Herreros-Villanueva M, Gironella M, Castells A, Bujanda L. Molecular markers in pancreatic cancer diagnosis. Clinica chimica acta; international journal of clinical chemistry. 2013;418:22-9. Epub 2013/01/12.
21. Kisiel JB, Yab TC, Taylor WR, Chari ST, Petersen GM, Mahoney DW, et al. Stool DNA testing for the detection of pancreatic cancer: assessment of methylation marker candidates. Cancer. 2012;118(10):2623-31. Epub 2011/11/16.
22. Gronborg M, Bunkenborg J, Kristiansen TZ, Jensen ON, Yeo CJ, Hruban RH, et al. Comprehensive proteomic analysis of human pancreatic juice. Journal of proteome research. 2004;3(5):1042-55. Epub 2004/10/12.
23. Kanda M, Knight S, Topazian M, Syngal S, Farrell J, Lee J, et al. Mutant GNAS detected in duodenal collections of secretin-stimulated pancreatic juice indicates the presence or emergence of pancreatic cysts. Gut. 2013;62(7):1024-33. Epub 2012/08/04.
24. Kanda M, Sadakari Y, Borges M, Topazian M, Farrell J, Syngal S, et al. Mutant TP53 in duodenal samples of pancreatic juice from patients with pancreatic cancer or high-grade dysplasia. Clin Gastroenterol Hepatol. 2013;11(6):719-30 e5. Epub 2012/12/04.
25. Park JY, Kim SA, Chung JW, Bang S, Park SW, Paik YK, et al. Proteomic analysis of pancreatic juice for the identification of biomarkers of pancreatic cancer. Journal of cancer research and clinical oncology. 2011;137(8):1229-38. Epub 2011/06/22.
26. Tanaka M, Chari S, Adsay V, Fernandez-del Castillo C, Falconi M, Shimizu M, et al. International consensus guidelines for management of intraductal papillary mucinous neoplasms and mucinous cystic neoplasms of the pancreas. Pancreatology. 2006;6(1-2):17-32. Epub 2005/12/06.
27. Tanaka M, Fernandez-del Castillo C, Adsay V, Chari S, Falconi M, Jang JY, et al. International consensus guidelines 2012 for the management of IPMN and MCN of the pancreas. Pancreatology. 2012;12(3):183-97. Epub 2012/06/13.

28. Bartsch DK, Dietzel K, Bargello M, Matthaei E, Kloeppel G, Esposito I, et al. Multiple small "imaging" branch-duct type intraductal papillary mucinous neoplasms (IPMNs) in familial pancreatic cancer: indicator for concomitant high grade pancreatic intraepithelial neoplasia? Fam Cancer. 2013;12(1):89-96. Epub 2012/11/28.
29. Canto MI, Hruban RH, Fishman EK, Kamel IR, Schulick R, Zhang Z, et al. Frequent detection of pancreatic lesions in asymptomatic high-risk individuals. Gastroenterology. 2012;142(4):796-804; quiz e14-5. Epub 2012/01/17.

SAMENVATTING

ACHTERGROND

Per jaar krijgen in Nederland ongeveer 2.000 mensen te horen dat zij alvleesklierkanker (pancreascarcinoom) hebben. Ongeveer hetzelfde aantal patiënten overlijdt jaarlijks aan de gevolgen van deze ziekte. Hiermee is alvleesklierkanker een van de meest dodelijke vormen van kanker. De gemiddelde overleving na het stellen van de diagnose is minder dan zes maanden en na vijf jaar is minder dan zes procent van alle patiënten nog in leven. Deze slechte overleving wordt voornamelijk veroorzaakt doordat alvleesklierkanker in een vroeg stadium meestal geen klachten geeft. Om deze reden wordt deze ziekte in meer dan 80% van de patiënten pas in een (te) vergevorderd stadium ontdekt. Genezing is dan vaak niet meer mogelijk.

Om alvleesklierkanker te genezen, moet de kanker chirurgisch verwijderd worden. Echter, zelfs voor de selecte groep patiënten die voor deze, in opzet, genezende operatie in aanmerking komt, blijkt deze behandeling in de meerderheid van de patiënten niet toereikend. Vijf jaar na de operatie is minder dan 10% van de patiënten nog in leven. De beste behandelresultaten worden gezien bij patiënten bij wie een kleine kanker verwijderd is.

Voor het verbeteren van de overleving lijkt het dus noodzakelijk alvleesklierkanker op te sporen als de tumor nog klein is en nog geen klachten geeft (asymptomatisch). Of beter nog, indien er alleen maar sprake is van een goedaardig voorstadium. Screening heeft als doel een ziekte eerder op te sporen, voordat deze klachten geeft, om op deze manier de kans op genezing te vergroten.

Met name door het ontbreken van een eenvoudige screeningstest (bijvoorbeeld een bloed- of ontlastingstest) is screenen van de algemene bevolking niet zinvol. Echter, screening lijkt mogelijk wel zinvol wanneer dit gericht wordt op personen met een verhoogd risico op het krijgen van alvleesklierkanker. Personen met een verhoogd risico op alvleesklierkanker zijn onder andere (1) personen met een foutje (mutatie) in hun erfelijk materiaal (DNA) waardoor ze een verhoogd risico hebben om alvleesklierkanker te ontwikkelen en (2) eerstegraads verwanten van patiënten met familiair alvleesklierkanker. Bij familiair alvleesklierkanker komt alvleesklierkanker veel voor binnen een familie maar is het onduidelijk welke DNA-mutatie of mutaties dit heeft/hebben veroorzaakt. Men spreekt van eerstegraads verwanten wanneer er een directe lijn is tussen twee verwanten, bijvoorbeeld ouder-kind of broer-zus. Voor deze hoog-risico personen is de kans dat zij tijdens hun leven alvleesklierkanker ontwikkelen enorm hoog en bedraagt 10% tot 40%. Naar schatting speelt bij ongeveer 10% van alle gevallen van alvleesklierkanker een erfelijke of familiaire factor een rol bij het ontstaan van de ziekte.

ONDERZOEKSDOELEN VAN DIT PROEFSCHRIFT

Dit proefschrift gaat over erfelijke en familiaire alvleesklierkanker. De doelen waren:
(1) Onderzoeken welke screeningstechniek en –aanpak het meest geschikt zijn voor het opsporen van vroeg-stadium alvleesklierkanker en goedaardige voorloper stadia.
(2) Meer inzicht krijgen de hoog-risico populatie waarbij wij ons gefocust hebben op (A) welke DNA-mutaties van belang zijn en (B) het bepalen van het daadwerkelijke alvleesklierkanker risico voor bepaalde hoog-risico populaties.
(3) Onderzoeken wat de psychische impact is van deelname aan een alvleesklierkanker screeningsprogramma.

BEHAALDE RESULTATEN

1. Screenen van personen met een verhoogd risico op alvleesklierkanker

Niet elke ziekte is geschikt voor screening. Om vast te kunnen stellen of screening naar een bepaalde ziekte verantwoord is, hebben Wilson en Jungner in 1968 tien criteria opgesteld. In **hoofdstuk 2** hebben wij deze criteria getoetst op alvleesklierkanker screening. Voor deze toetsing hebben wij gebruik gemaakt van de op dat moment aanwezige literatuur. Deze toetsing toonde dat screening van personen met een erfelijk of familiair verhoogd risico op alvleesklierkanker de potentie heeft zinvol te zijn aangezien aan het merendeel van de tien criteria werd voldaan. Het toonde echter ook dat aanvullend onderzoek noodzakelijk is om te beoordelen of deze vorm van screening daadwerkelijk aan alle criteria kan voldoen. Alleen dan wegen de voordelen van screenen (vermindering van alvleesklierkanker gerelateerde sterfte en winst in levensjaren), op tegen de nadelen van screening zoals overdiagnosiek, overbehandeling en kosten.
Een van de criteria van Wilson en Jungner waar op basis van de op dat moment aanwezige literatuur nog niet aan voldaan werd, had betrekking op de screeningtest. Bij aanvang van dit promotietraject was het niet duidelijk welke test het meest geschikt is voor screening van de alvleesklier. Eerder onderzoek had al laten zien dat het jaarlijks screenen van hoog-risico personen resulteert in het opsporen van asymptomatische kankers en voorloper stadia van alvleesklierkanker. Ook was op basis van eerder onderzoek duidelijk dat zowel inwendige echografie (endoscopic ultrasonography; EUS) en Magnetic Resonance Imaging (MRI) de twee beste testen hiervoor waren. Het was echter niet duidelijk hoe deze twee testen zich ten opzichte van elkaar verhouden; is een van de twee testen beter of vullen de twee testen elkaar aan en is het dus noodzakelijk om beiden uit te voeren? Om een antwoord te vinden op deze vragen, hebben wij een multicenter studie uitgevoerd waarin wij EUS en MRI geblindeerd met elkaar hebben vergeleken. De resultaten van deze studie worden gepresenteerd in **hoofdstuk 3**. Voor deze analyse hebben wij gekeken naar de uitkomsten van de eerste screeningsronde. Op basis van deze eerste analyse, lijken EUS en MRI elkaar aan te vullen in plaats van dat een test beter is dan de andere. EUS lijkt

het meest geschikte onderzoek voor het opsporen van kleine solide afwijkingen inclusief vroeg-stadium alvleesklierkanker, daarentegen lijkt MRI geschikter voor de detectie van cysteuze (vochtblazen) afwijkingen. Deze vochtblazen zijn potentiële voorlopers van alvleesklierkanker.

Wereldwijd zijn er ongeveer tien centra die onderzoek doen naar de zinvolheid van het screenen naar alvleesklierkanker in hoog-risico personen. Omdat alvleesklierkanker een betrekkelijk zeldzame ziekte is (per jaar wordt deze ziekte in Nederland in ongeveer 10 per 100.000 inwoners gediagnosticeerd, voor dikke darm kanker is dit getal 60), is het onmogelijk dat één centrum op zichzelf voldoende resultaten zal verzamelen om een antwoord te geven op de vraag of deze vorm van screening verantwoord is. Om deze reden hebben wij in samenwerking met het John Hopkins Ziekenhuis in Baltimore (VS) in 2011 een internationaal congres georganiseerd waarbij van alle betrokken centra meerdere vertegenwoordigers aanwezig waren. Het doel van dit congres was een platform te creëren om zo wereldwijde alvleesklierkanker screening te verbeteren en standaardiseren. Als eerste opzet hiervoor, werden er tijdens dit congres aanbevelingen opgesteld die betrekking hadden op deze vorm van screening. Zo werden er aanbevelingen gedaan met betrekking tot wie er in aanmerking komen voor screening, wat de beste aanpak is voor screening (welke testen, screening interval, wat te doen wanneer er een afwijking gevonden wordt), wanneer er overgegaan moet worden naar chirurgische verwijdering van de afwijkingen en wat een succesvolle uitkomst van screening zou zijn. Deze aanbevelingen staan beschreven in **hoofdstuk 4**.

Naast een klinische studie (beschreven in twee paragrafen hierboven), hebben wij ook een project gestart waarbij wij met behulp van computermodellen een eerste opzet gedaan hebben te onderzoeken onder welke voorwaarden het screenen naar alvleesklierkanker mogelijk en zinvol is. Het voordeel van het gebruiken van dergelijke computermodellen is dat alle informatie die er is over (screening op) alvleesklierkanker, samengevoegd kan worden in deze modellen. Tevens is het mogelijk om met deze modellen te variëren in veel variabelen die de uitkomsten van een screeningsprogramma beïnvloeden (bijvoorbeeld de gevoeligheid van de test, het risico op het krijgen van alvleesklierkanker, de duur van het ontstaan van kanker en de behandeling na het stellen van de diagnose). Hierdoor kan bepaald worden wat de meest belangrijke variabelen zijn, en waar in vervolgonderzoek meer op gefocust moet worden. **Hoofdstuk 5** geeft de resultaten van onze eerste analyse. Deze analyse toonde dat variabelen met betrekking tot follow-up beleid, het risiconiveau en natuurlijk gedrag het meest van belang zijn.

Om meer inzicht te krijgen in het natuurlijk gedrag van erfelijk/sporadisch alvleesklierkanker, hebben wij ons in **hoofdstuk 6** gericht op de voorloperstadia van alvleesklierkanker. In het operatieweefsel van patiënten met erfelijk/familiair en sporadisch pancreascarcinoom hebben wij onderzocht hoe vaak voorloperstadia voorkwamen. De twee voorstadia die het meest gezien worden, ook bij erfelijke en familiaire vormen van alvleesklierkanker, zijn Pancreatic Intraepithelial Neoplasia (PanIN) en Intraductal Papillairy Mucinous

Neoplasms (IPMN). Binnen de voorloperstadia zijn er zowel voor PanINs als voor IPMNs drie verschillende gradaties te onderscheiden; hoe hoger de gradatie, hoe onrustiger het weefsel is. Onze analyse toonde dat bij patiënten met erfelijke of familiair alvleesklierkanker vaker voorlopers gezien werden met de hoogste gradering. Ook zagen wij in deze groep meer voorstadia t.o.v. de sporadische patiënten. Opvallend was, dat er enkel PanINs gezien werden en geen IPMNs. Op basis van deze uitkomsten lijkt het met name van belang om in de nabije toekomst onderzoek te doen naar hoe deze PanIN lesies het beste opgespoord kunnen worden.

2. Wie zijn er 'at-risk'?

In het tweede deel van dit proefschrift ligt de nadruk op de hoog-risico populatie. **Hoofdstuk 7** geeft een overzicht van de beschikbare literatuur over erfelijk en familiair alvleesklierkanker. Hierin bespreken wij de verschillende kanker syndromen die gerelateerd zijn aan alvleesklierkanker en geven wij een overzicht van de geschatte risico op alvleesklierkanker voor de verschillende hoog-risico groepen.

In **hoofdstuk 8** en **hoofdstuk 9** richten we ons op de familiair alvleesklierkanker families. Eerder in de achtergrond staat al vermeld dat het verschil tussen erfelijk en familiair alvleesklierkanker is dat bij familiair alvleesklierkanker het niet duidelijk is welke DNA-mutatie(s) verantwoordelijk is (zijn) voor de clustering van alvleesklierkanker binnen een familie. Deze onduidelijkheid met betrekking tot het verantwoordelijke gen, maakt het onmogelijk om binnen een familiair alvleesklierkanker familie precies aan te geven welke familieleden *at risk* zijn en welke niet. Los van het feit dat dit voor een familielid lastig en belastend kan zijn (hij/zij weet niet zeker of hij/zij een verhoogd alvleesklierkanker risico heeft) is dit vanuit een screeningssetting ook complex. Op basis van de erfelijkheidsleer, is het voorhand bekend dat bij een dergelijke familie 50% van de familieleden ten onrechte gescreend worden; het is echter onmogelijk om aan te tonen welke 50% dit is. Het is dus zowel voor het individu als voor de uiteindelijke effectiviteit van een screeningsprogramma (deze is immers het hoogst wanneer deze alleen gericht wordt op personen met een daadwerkelijk verhoogd risico) van belang om meer inzicht te krijgen in de genen die een rol spelen bij het ontstaan van familiair alvleesklierkanker. Zowel **hoofdstuk 8** als **hoofdstuk 9** presenteren data waarin wij ons gefocust hebben op het beter identificeren van de hoog-risico persoon. In **hoofdstuk 8** laten wij zien dat het van meerwaarde is om bij families waar gedacht wordt aan familiair alvleesklierkanker te testen op een *CDKN2A* mutatie. Deze mutatie is met name bekend om zijn sterk verhoogde risico op melanomen (huidkanker). Het is echter ook duidelijk dat deze mutatie geassocieerd is met andere kankertypes, waaronder alvleesklierkanker. Als het gaat om familiair alvleesklierkankerfamilies dan wordt er op dit moment geadviseerd om alleen te testen voor een *CDKN2A* mutatie als er naast alvleesklierkanker bij ten minste een familielid ook melanomen voorkomen. Met onze studie toonden wij aan dat het zinvol is om ook te testen voor deze mutatie wanneer er alleen maar alvleesklierkanker binnen een familie voorkomt.

In **hoofdstuk 9** hebben wij onderzocht of het zinvol is om standaard te testen voor *PALB2* mutaties in families met familiair alvleesklierkanker. Deze mutatie werd in 2009 ook gelinkt aan een verhoogd risico op alvleesklierkanker, waar hij eerder alleen geassocieerd was met een risico op borstkanker. In geen van de door ons onderzochten families vonden wij een *PALB2* mutaties. Op basis van deze bevindingen concludeerden wij dat *PALB2* slechts een beperkte bijdrage heeft binnen het ontstaan van familiair alvleesklierkanker en dat het niet zinvol is deze mutatie mee te nemen in de standaard genetische work-up van familiair alvleesklierkanker families

Hoofdstuk 10 focust op de erfelijk alvleesklierkanker groep. In dit hoofdstuk geven wij een risicoschatting voor alvleesklierkanker in patiënten met het syndroom van Peutz Jeghers. Het Peutz Jeghers syndroom is een zeldzame erfelijke aandoening die gekarakteriseerd wordt door pigmentaties (verkleuring) van slijmvliezen en de huid, poliepen in het maagdarmkanaal en een verhoogd risico op verschillende vormen van kanker. Bij aanvang van dit promotietraject was er nog veel onduidelijkheid over hoe hoog het risico op het krijgen van alvleesklierkanker was voor patiënten met dit syndroom. De getallen liepen uiteen van gèèn risico tot een risico van wel 36% op de leeftijd van 64 jaar (het geschatte risico op het krijgen van alvleesklierkanker voor de algehele bevolking is <1%). Door gebruik te maken van de Nederlandse Peutz-Jeghers Syndroom database waarin nagenoeg alle patiënten met Peutz-Jeghers in Nederland zijn opgenomen, zijn wij erin geslaagd een betrouwbare risicoschatting te geven. Het risico op het krijgen van alvleesklierkanker was 26% op de leeftijd van 70 jaar.

3. Psychologische impact van screenen

Naast het beantwoorden van de vraag of screening bijdraagt aan een verbeterde overleving is het ook van groot belang om meer inzicht te krijgen in de psychologische impact van screening; in welke mate worden de screeningsmethodes als fysiek of psychisch belastend ervaren, in hoeverre worden de zorgen om alvleesklierkanker positief of negatief beïnvloed door de screening, en in hoeverre worden vervolgafspraken nagekomen? De resultaten zullen helpen bij het verbeteren van zowel de medische als de psychische zorg bij mensen met en verhoogd risico op alvleesklierkanker. **Hoofdstuk 11** beschrijft de resultaten van onze studie waarbij wij ons gefocust hebben op deze psychologische impact. Een opmerkelijke bevinding was dat, ondanks dat EUS een invasief onderzoek is en MRI niet, beide onderzoeken als even belastend ervaren werden. Tevens had het vinden van een afwijking bij beide of een van de testen geen invloed op het niveau van angsten en zorgen over kanker. Het niveau van zowel angsten als zorgen was voorafgaand aan de eerste screeningsronde al vergelijkbaar met wat gezien wordt in de algehele bevolking en bleef na deze eerste ronde op een gelijk niveau. Op basis van deze gegevens, concludeerden wij dat vanuit een psychologisch oogpunt, screening gerechtvaardigd is.

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LIST OF PUBLICATIONS

Harinck F, Bruno MJ. Endosonography in the management of biliopancreatic disorders. Best Pratice & Research: Clinical Gastroenterology 2009;23 (5): 703-10.

Poley JW, Kluijt I, Gouma DJ, **Harinck F**, Wagner A, Aalfs C, van Eijck CH, Cats A, Kuipers EJ, Nio CY, Fockens P, Bruno MJ. The yield of first-time endosonograpic ultrasonography in screening individuals at high risk of developing pancreatic cancer. American Journal of Gastroenterology 2009; 104 (9): 2175-81.

Harinck F, Canto MI, Schulick RD, Goggins M, Poley J-W, Fockens P, Kluijit I, Bruno MJ. Surveillance in individuals at high-risk of pancreatic cancer, too early to tell? Gut. 2010;59(7):1005.

Harinck F, Poley J-W, Bruno MJ. Screening and surveillance of individuals at high risk for pancreatic cancer, where are we now? Annals of Gastroenterology and Hepatology.

Harinck F, Kluijt I, Bleiker EMA, Poley J-W, Fockens P, Bruno MJ. Erfelijk pancreascarcinoom en de mogelijkheid tot vroege detectie en preventie. Kankerbreed, 2010; 2 (2):14-17.

Harinck F, Poley J-W, Kluijt I, Fockens P, Bruno MJ. Is early diagnosis of pancreatic cancer fiction? Surveillance of individuals at high risk for pancreatic cancer. Digestive Diseases. 2010;28(4-5):670-8.

Harinck F, Nagtegaal T, Kluijt I, Aalfs CA, Smets E, Poley J-W., Wagner A, van Hooft JE, Fockens P, Bruno MJ, Bleiker EMA. Feasibility of a pancreatic cancer surveillance program from a psychological point of view. Genetics in Medicine. 2011;13(12):1015-24

Harinck F, Kluijt I, van der Stoep N, Oldenburg R, Wagner A, Aalfs CA, Sijmons RH, Poley J-W., Kuipers EJ, Fockens P, van Os TAM, Bruno MJ. Indication for CDKN2A mutation analysis in familial pancreatic cancer families without melanomas. Journal of Medical Genetics. 2012;49(6):362-5.

Harinck F, Kluijt I, Wagner A, Bruno MJ. Hereditary and familial forms of pancreatic cancer. European Gastroenterology & Hepatology Review. 2012;8(1):50-53.

Harinck F, Kluijt I, van Mil SE, Waisfisz Q, van Os TAM, Aalfs CA, Wagner A, Olderode-Berends M, Sijmons RH, Kuipers EJ, Poley J-W, Fockens P, Bruno MJ. Routinely testing for PALB2 mutations in familial pancreatic cancer families and familial breast cancer families with pancreatic cancer is not indicated. European Journal of Human Genetics. 2012;20(5):577-9.

Harinck F, Poley J-W, van Eijck CHJ, Busch OR, Bruno MJ. Richtlijn Pancreascarcinoom: landelijk en multidisciplinair. Magna 2012 Jun;2(18):55-7

Harinck F, Korsse SE, van Lier MGF, Biermann K, Offerhaus GJ, Krak NC, Looman CWN, van Veelen W, Kuipers EJ, Wagner A, Dekker E, Mathus-Vliegen EMH, Fockens P, van Leerdam ME, Bruno MJ. Pancreatic cancer risk in Peutz-Jeghers syndrome patients; results of a large Dutch cohort study and implications for surveillance. Journal of Medical Genetics. 2013 Jan;50(1):59-64.

Canto MI, **Harinck F**, Hruban RH, Offerhaus GJ, Poley J-W, Fockens P, Kamel I, Nio CY, Schulick RD, Bassi C, Kluijt I, Goggins M, Bruno MJ. International Cancer of the Pancreas Screening (CAPS) Consortium Summit on the Management of Patients with Increased Risk for Familial Pancreatic Cancer. Gut. 2013 Mar;62(3):339-47.

CURRICULUM VITAE
& PHD PORTFOLIO

CURRICULUM VITAE

Femme Harinck werd geboren op 3 september 1980 in Leiden. In 1998 haalde zij haar VWO-diploma op het Aquino college in Leiden. Hierna vertrok zij voor een jaar naar Amerika om hier aan het Allegheny College te studeren. Vervolgens verhuisde zij naar Amsterdam waar zij in 2000 haar propedeuse Medische Biologie behaalde aan de Universiteit van Amsterdam. In 2000 startte zij met de opleiding Geneeskunde aan dezelfde universiteit. Het artsenexamen behaalde zij in 2008. In april 2008 begon zij aan haar promotieonderzoek op de afdeling Maag-, Darm- en Leverziekten van zowel het Erasmus Medisch Centrum in Rotterdam en de afdeling Maag-, Darm- en Leverziekten van het Academisch Medisch Centrum in Amsterdam onder supervisie van prof.dr. M.J. Bruno en prof.dr. P. Fockens. In mei 2012 is zij gestart met de opleiding tot Maag-Darm-Leverarts (opleider dr. R.A. de Man). De vooropleiding Interne Geneeskunde volgde zij in het Sint Lucas Andreas ziekenhuis in Amsterdam (opleider dr. C.E.H. Siegert). Eind oktober jl. is ze begonnen in het Erasmus MC aan het Maag-Darm-Leverziekten opleidingsonderdeel. Femme is getrouwd met Thomas Dirksmeier met wie zij in juli 2013 een dochter (Ymke) kreeg.

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	Biostatistics for clinicians Netherlands Institute for Health Sciences
	Biomedical English Writing and Communication Erasmus Medisch Centrum Rotterdam
	Basiscursus Regelgeving en Organisatie van Klinisch Onderzoek Erasmus Medisch Centrum Rotterdam
	Workshop: Feedback geven en ontvangen Erasmus Medisch Centrum Rotterdam, Nu91
	Workshop: Haal meer uit je team Erasmus Medisch Centrum Rotterdam, Nu91
2011	Workshop Writing Successful Grant Proposals Erasmus Molecular Medicine Postgraduate School
	Genetics for Dummies Erasmus Molecular Medicine Postgraduate School
	CPO minicursus - Methodologie van Patiëntgebonden Onderzoek en Voorbereiding van Subsidieaanvragen Erasmus Medisch Centrum Rotterdam

Oral presentations at (inter)national conferences

2009	Nederlandse Vereniging voor Gastroenterologie, najaarscongres, Veldhoven, Nederland Surveillance of individuals at high-risk of pancreatic cancer; preliminary results of a multicentre prospective study

2009	International Society for Gastrointestinal Hereditary Tumours, Düsseldorf, Duitsland Comparative yield of EUS and MRI in the surveillance of individuals at high risk for pancreatic cancer
	Digestive Disease Week, Chicago, Verenigde Staten Comparative yield of EUS and MRI in the surveillance of individuals at high risk for pancreatic cancer
2010	European Bridging Meeting in Gastroenterology, Berlijn, Duitsland The use of endoscopic ultrasound and magnetic resonance imaging in surveillance of individuals at high risk for developing pancreatic cancer
	Joint Genetics Meeting, Amsterdam, Nederland Burden of undergoing pancreatic cancer surveillance in high-risk individuals; first experiences
2011	Digestive Disease Week, Chicago, Verenigde Staten Indication for CDKN2A mutation analysis in familial pancreatic cancer families without melanomas
	Informatiebijeenkomst Erfelijk Alvleesklierkanker, Utrecht, Nederland De alvleesklier: haar ligging en functies
	International Society for Gastrointestinal Hereditary Tumours, San Antonio, Verenigde Staten Indication for CDKN2A mutation analysis in familial pancreatic cancer families without melanomas
	Nederlandse Vereniging voor Gastroenterologie, voorjaarscongres, Veldhoven, Nederland Indication for CDKN2A mutation analysis in familial pancreatic cancer families without melanomas
	CAPS International Consortium Summit, Baltimore, Verenigde Staten The Dutch experience of pancreatic cancer surveillance in high-risk individuals

2012 The 1st meeting of the Hungarian Pancreatic Club, Szeged, Hongarije
Invited lecture: The Dutch experience on Surveillance of individuals at high risk of developing pancreatic cancer

European Pancreatic Club conference, Praag, Tsjechië
Invited lecture: Pro-contra session on surveillance of individuals at high risk for developing pancreatic cancer

Nederlandse Vereniging voor Gastroenterologie, voorjaarscongres, Veldhoven, Nederland
(1) Prospective evaluation of psychological impact of pancreatic cancer surveillance in high-risk individuals
(2) Pancreatic cancer risk in Peutz-Jeghers patients; results of a large Dutch cohort study and implications for surveillance

Nederlands Endoscopie Symposium (NES2012), Utrecht, Nederland
Alvleesklierkanker screening bij personen met een verhoogd risico op deze ziekte en de psychosociale belasting

2014 Digestive Disease Week, Chicago, Verenigde Staten
A comparative prospective blinded analysis of the effectiveness of EUS and MRI as screening tools for pancreatic cancer

Nederlandse Vereniging voor Gastroenterologie, voorjaarscongres, Veldhoven, Nederland
(1) A comparative prospective blinded analysis of the effectiveness of EUS and MRI as screening tools for pancreatic cancer
(2) Clinicopathological characteristics of pancreatic resection specimen of inherited/familial versus sporadic pancreatic ductal adenocarcinoma

Poster presentations at (inter)national conferences

2008 United European Gastroenterology Week, Wenen, Oostenrijk
Endoscopic Ultrasonography is a valuable tool in screening individuals at high risk for pancreatic cancer

2009 Digestive Disease Week, Chicago, Verenigde Staten
Features of chronic pancreatitis in individuals at high risk for developing pancreatic cancer

2009	International Society for Gastrointestinal Hereditary Tumours, Düsseldorf, Duitsland Features of chronic pancreatitis in individuals at high risk for developing pancreatic cancer
	United European Gastroenterology Week, Londen, Engeland (1) Yield of endosonography and magnetic resonance imaging in the surveillance of individuals at high risk of pancreatic cancer (2) Features of chronic pancreatitis in individuals at high risk for developing pancreatic cancer (3) EUS surveillance of individuals at high risk of pancreatic cancer; focal areas of hypoechogenicity suspicious for a mass lesion with spontaneous resolvement
2010	Digestive Disease Week, New Orleans, Verenigde Staten (1) Spontaneous resolvement of focal pancreatic lesions in EUS screening of individuals at high risk of pancreatic cancer (2) Burden of undergoing pancreatic cancer surveillance in high-risk individuals; first experiences
	Joint Genetics Meeting, Amsterdam, Nederland Comparative yield of EUS and MRI in the surveillance of individuals at high risk for pancreatic cancer
2011	United European Gastroenterology Week, Stockholm, Zweden (1) Indication for CDKN2A mutation analysis in familial pancreatic cancer families without melanomas (2) PALB2 seems not to be involved in pancreatic cancer and/or breast cancer development in a Dutch cohort of familial pancreatic cancer families and families with clustering of both pancreatic cancer and breast cancer (3) Feasibility of a pancreatic cancer surveillance program from a psychological point of view
	European Human Genetics Conference, Amsterdam, Nederland Indication for CDKN2A mutation analysis in familial pancreatic cancer families without melanomas

2011	Digestive Disease Week, Chicago, Verenigde Staten PALB2 seems not to be involved in pancreatic cancer and/or breast cancer development in a Dutch cohort of familial pancreatic cancer-families and families with clustering of both pancreatic cancer and breast cancer
2012	European Human Genetics Conference, Nürnberg, Duitsland The psychological impact of pancreatic cancer surveillance in high-risk individuals
	Digestive Disease Week, San Diego, Verenigde Staten (1) Prospective evaluation of psychological impact of pancreatic cancer surveillance in high-risk individuals (2) Pancreatic cancer risk in Peutz-Jeghers patients; results of a large Dutch cohort study and implications for surveillance
2014	Digestive Disease Week, Chicago, Verenigde Staten (1) Clinicopathological characteristics of pancreatic resection specimen of inherited/familial versus sporadic pancreatic ductal adenocarcinoma (2) Exploring the effects of factors associated with the outcome of pancreatic cancer screening in high-risk individuals

Tutoring

2009-11	Tutoring bachelor students Faculty of Medicine, Academic Medical Center, University Medical Center, Amsterdam
2011-12	Supervision student F. Boersma – Academic internship

Others

2011	Utrecht, The Netherlands Organizing an information day on inherited and familial pancreatic cancer Target population: Individuals with an increased inherited or familial risk for developing pancreatic cancer

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