OUTLINE OF THE THESIS
The studies in this thesis discuss a wide range of different developments in the diagnosis and treatment of pancreatic cancer. Therefore, this thesis is divided into two parts. In PART I - Molecular Markers, the emphasis is laid on possible ways to detect periampullary and particularly pancreatic carcinomas in an early stage to increase the chance for curative resection. Since pancreatic cancer is basically a disease of the genes, the optimal marker needs to be sought in the molecular changes that occur relatively early in the stepwise tumor progression from normal epithelium to infiltrating carcinoma. Such molecular changes may ultimately prove to be sensible targets for different treatment options. In particular for early detected lesions optimal surgical management with low mortality offers the only possibility for cure. Therefore, in PART II - Management, several aspects of the management of the different periampullary cancers are evaluated. Surgical procedures with curative as well as palliative intention are discussed.

PART I - Molecular Markers

In Chapter 1 the detection of micrometastases in bone marrow of patients with suspected pancreatic and ampullary cancer is prospectively evaluated. The predictive value of positive micrometastases on the overall survival is determined.

Fine needle aspirations of periampullary tumors are often not conclusive in their diagnosis when only conventional cytology is performed. Since K-ras, p53 and DPC4 are genes frequently mutated in the development of periampullary cancers, we used this genetic panel in Chapter 2 to explore whether its use could improve the diagnostic yield of cytological samples interpreted as atypical, i.e., suspicious but not conclusive for cancer.

Standard staging procedures of periampullary tumors do not identify all patients with an adverse outcome despite apparently successful resection. The purpose of Chapter 3 was to determine whether cytologic examination and K-ras mutation analysis of intraoperative abdominal washings obtained from patients with periampullary carcinoma could identify a subset of patients with a worse prognosis.

Although K-ras mutation analysis is a valuable adjunct to brush cytology alone in diagnosing a malignant cause of extrahepatic biliary stenosis, its specificity remains a concern. The aim of Chapter 4 was to determine whether the finding of a K-ras mutation in endobiliary brush cytology of patients with a clinically benign extrahepatic biliary stenosis represent an early or false-positive diagnosis of cancer after long-term follow-up.

In order to estimate the prevalence of K-ras codon 12 mutations in brush cytology and bile from patients without clinically manifest periampullary cancer, and also not "at risk", i.e. without advanced precursor lesions, the "epidemiologic necropsy" study design was used in Chapter 5. In this design necropsies without the disease under evaluation and without risk factors for that
diseases are used for study. In this way it is possible to determine the prevalence of mutations in the general population and therefore the specificity of the K-ras mutation analysis when used as a screening test in subjects without manifest disease.

Recently, a novel test for K-ras mutations called the Amplification Refractory Mutation System (ARMS) assay has been developed. This is a quantitative assay that in principal increases the specificity, and specificity is of major importance when screening for malignant disease (see chapter 4). The aim of the study described in Chapter 6 was to assess the test characteristics and additional value of the ARMS assay in diagnosing the cause of extrahepatic biliary stenosis.

Telomeric dysfunction is a major mechanism for the generation of chromosomal instability (CIN). The timing of telomeric dysfunction has not yet been examined in the context of the multistage progression model (PanINs) of pancreatic cancer (General introduction, figure 1). In Chapter 7 we examined the telomere repeat lengths by means of a novel fluorescence in situ hybridization protocol using tissue microarrays with a series of 82 PanIN lesions of all histological grades.

Novel tumor markers for pancreatic cancer are needed for early detection and development of promising treatment modalities. The aim of Chapter 8 was to identify genes differentially expressed in pancreatic cancer. cDNA from normal pancreas and pancreatic cancer tissue was hybridized to the Affymetrix Human Genome U95 GeneChip set for simultaneous analysis of 60,000 cDNA fragments. Genes expressed at levels at least fivefold greater in the pancreatic cancers as compared to normal tissues were identified and a selection was validated.

The molecular profiles of ampullary carcinoma, which has the best prognosis from all periampullary carcinomas, are not yet well understood. In Chapter 9 we utilized high-throughput gene expression analysis to identify the genes differentially expressed in ampullary carcinoma. The identified genes might serve as diagnostic markers or a lead for further research to therapeutic targets.

PART II - Management

Ampullary tumors are relatively uncommon and differentiation between adenoma and carcinoma is often difficult. Furthermore there is still discussion on the extent and type of surgery needed in these tumors. Short-term results and long-term survival after local and radical resection of ampullary adenomas and adenocarcinomas were analyzed in Chapter 10. The aim was to identify those tumors that may be treated by local resection.

Resection is the only curative treatment option for pancreatic cancer. Given the small number of long-term survivors, minimization of surgery-related mortality and morbidity is imperative. Numerous studies have shown that several high-risk surgical procedures can be performed with a lower postoperative mortality rate in high volume centers compared to low volume centers. Nevertheless, centralization of pancreatic surgery is still under debate. In Chapter 11 the effect of hospital volume on hospital mortality after pancreatic resection was determined by performing
a systematic review of the available data.
The Netherlands is one of the few countries with an independent nationwide registry of the mortality data after pancreaticoduodenectomy. During the last decade a plea for centralization of pancreaticoduodenectomy in high-volume centers has been taken place in the Netherlands. In Chapter 12 the effect of this plea is analyzed by the evaluation of changes in mortality after pancreaticoduodenectomy as well as changes in referral pattern between 1994 and 2003.
At time of presentation around 80% of patients with periampullary cancer is unresectable. In these patients palliation of the major symptoms such as jaundice, duodenal obstruction and pain, is of major importance. Chapter 13 summarizes the methods of surgical and non-surgical palliative treatment in pancreatic cancer.
A minority of the patients undergoing an exploratory laparotomy with the intention to perform a pancreaticoduodenectomy appears to have unresectable disease during operation. Controversy still exists whether or not a double bypass should be performed routinely in these patients. The randomized controlled trial in Chapter 14 compares the outcome after a double bypass consisting of a hepaticojejunostomy and a prophylactic gastrojejunostomy with the outcome after a single bypass existing of a hepticojejunostomy alone. The aim was to evaluate the effect of a prophylactic gastrojejunostomy on the development of gastric outlet obstruction and quality of life in patients with unresectable periampullary cancer found during exploratory laparotomy.