CHAPTER 1

Epidemiology and genetics of pancreatitis

David C. Whitcomb

Division of Gastroenterology, Hepatology and Nutrition, Departments of Medicine, Cell Biology & Physiology, and Human Genetics, University of Pittsburgh/UPMC, Pittsburgh, PA, USA

Definition

Chronic pancreatitis (CP) can be defined as “a continuing inflammatory disease of the pancreas, characterized by irreversible morphological change, and typically causing pain and/or permanent loss of function” [1]. This definition is intentionally pragmatic, as developed by the members of the Pancreatic Society of Great Britain and Ireland in March 1983 in Cambridge, England as a pretext to the morphology-based Cambridge classification of CP severity [1]. The definition is vague but has stood the test of time and has been followed in consensus statements by nearly all societies and expert groups for the subsequent two decades.

The pragmatic nature of the Cambridge definition speaks to the challenges in defining a syndrome with multiple etiologies, variable features, unpredictable clinical course, and inadequate treatment [2]. As a morphology-based definition, it also ignores key histologic, clinical, and functional features that dominate the definitions from the Marseilles meetings [3, 4] and ignores the possibility of “minimal change” CP [5a], functional changes such as pancreatitis-associated chronic pain syndrome and/or pancreatic insufficiency, or autoimmune pancreatitis. Furthermore, the definition is independent of etiology, it cannot differentiate progressive disease from old scars from a bout of acute pancreatitis (AP), and it has little prognostic value. A new, two-part mechanistic definition of CP has been proposed that focuses on disruption of the normal injury → inflammation → resolution → regeneration sequence. The definition includes the essence of CP, “Chronic pancreatitis is a pathologic fibro-inflammatory syndrome of the pancreas in individuals with genetic, environmental and/or other risk factors who develop persistent pathologic responses to parenchymal injury or stress,” and the characteristics of CP. “Common features of established and advanced CP include pancreatic atrophy, fibrosis, pain syndromes, duct distortion and strictures, calcifications, pancreatic exocrine dysfunction, pancreatic endocrine dysfunction, and dysplasia.” This new definition opens the door to new diagnostic criteria that distinguishes CP from other disorders with CP-like features, provides a method for diagnosing “early CP,” and may improve methods of mechanism-based therapies – which is the goal of personalized medicine [5b].

Burden of disease

Epidemiologists struggle to determine the incidence and prevalence of CP – in part because of the vague definitions and different detection approaches [6, 7]. Administrative data, such as ICD-9 codes used in the United States, have limited value because the same code, 577.1, is used for recurrent acute pancreatitis (RAP) as well as CP. Indeed, authoritative studies of the burden of digestive diseases in the United States found it impossible to distinguish AP from CP using public records and grouped the two entities into one big problem [8].

Autopsy studies using histologic criteria such as duct ectasia, periductal fibrosis, ductular proliferation, acinar ductular metaplasia, and interstitial inflammation or
Pancreatitis

fibrosis suggest that the incidence of CP is as high as 12–14% [9, 10], with abnormal fibrosis in up to 39% [10]. Histologic changes suggestive of CP are even more prevalent in patients with very common chronic disorders such as renal disease (up to 56%) [9] and diabetes mellitus (DM) (~7% by clinical evaluation but much higher in diabetes autopsy databases such as nPOD [11] – noting the problem of reverse causality [12]). However, it is well recognized that interstitial inflammation and fibrosis alone are not sufficient to make a diagnosis of CP [13].

The emergence and widespread use of sensitive abdominal imaging techniques has helped standardize epidemiological approaches when morphologic criteria are used. While morphology is not the only criteria used in epidemiology studies, it does serve as an equalizing factor. Thus, the burden of CP in terms of disease prevalence from more recent surveys is more useful.

In the United States the best estimate comes from Minnesota, where the age-adjusted prevalence of CP was estimated at 41.8 cases per 100,000 population [7]. In contrast to earlier studies, the prevalence between males and females was similar, as reported in the North American Pancreatitis Study 2 (NAPS2) reports [14, 15]. In Japan the prevalence of CP was similar to the United States, with 36.9 cases per 100,000 population [16]. In France the prevalence of CP was 26.4 cases per 100,000 population [17], with a strong male predominance. The lowest prevalence was in China, which was only 3 cases of CP per 100,000 population in 1996 but had risen rapidly to 13.5 per 100,000 population by 2003 [18]. The highest rates were in Southern India, where the prevalence of CP is 114–200 per 100,000 population [19]. In addition to difference in prevalence, there are marked differences in rates of the etiologic diagnoses, with alcoholic and idiopathic being the most common causes in all studies. Alcohol etiology is consistently more common in men than in women.

**Clinical features**

The clinical features of CP include recurrent and chronic inflammation, fibrosis, duct distortion, pseudocysts, atrophy, pancreatic exocrine insufficiency, DM, multiple pain patterns, stones, and risk of pancreatic cancer. These features vary with etiology and environmental factors, and none of them are present in all patients – except for when duct distortion is used as the diagnostic criteria as in the Cambridge definition [1].

**Diagnosis**

Using the Cambridge definition of CP, a “clinical” diagnosis of CP can usually be made without ambiguity when significant morphologic features are documented. The problem with the Cambridge definition is the requirement of “irreversible morphological change” in the pancreas, how it is defined, and when it occurs. Indeed, patients may have symptoms of CP for 5–10 years before irreversible morphologic changes are documented, resulting in presumably unnecessary pain, anxiety, uncertainty, suffering, and numerous diagnostic tests. The result of the process is a “diagnosis,” with continued symptomatic treatment. Furthermore, the consequence of classifying CP based on morphologic criteria is that, while all investigators and clinicians agree on what end-stage CP looks like, they continue to sharply disagree on the border between “normal” and “abnormal” and on the minimal required features.

Many experts also deviate from the Cambridge definition, recognizing the limitations of morphology alone and the possibility of minimal change CP with prominent functional features such as pancreatic juice with low bicarbonate concentrations or pancreatitis-like pain syndromes. This view is supported by the clinical improvement in some patients diagnosed with minimal change pancreatitis and pain who find relief with total pancreatectomy and islet autotransplantation (TPIAT) [20–23]. These differences in perspectives on traditional views of CP make a consensus definition of early CP nearly impossible, with a ripple effect of making the criteria for early diagnosis somewhat arbitrary.

**Animal models of early CP**

The use of model organisms to understand human diseases remains a critical component of biomedical research. A good model should be a simplified version of something that reflects its primary components and is useful to study its characteristics under a variety of conditions. In the case of CP, animal models demonstrated that multiple injuries and inflammation resulted in parenchymal pathology, including scaring, but did...
not provide insight into human disease, which appeared stochastic in onset and highly variable in progression, clinical features, and outcomes. Thus, animal models provided insight into downstream pathology but failed to provide insight into etiologies, susceptibility, and variable progression.

Genetic risk factors for CP

In 1996 we discovered that hereditary pancreatitis (HP), a rare, autosomal dominant, highly penetrant, and early-onset syndrome of RAP and CP, was caused by a gain-of-function mutation in the cationic trypsinogen gene (PRSS1) [24–26]. The discovery immediately implicated prematurely activated trypsin as a key factor in the pathogenesis of AP and CP in humans, indicated that RAP can lead to typical CP, and introduced the possibility that other genetic factors associated with trypsin regulation may increase the risk of RAP and/or CP. Further, study of HP families indicated that even with inheritance of the most virulent of pathogenic variants, the age of onset, the progression to CP, DM, pain syndromes, and PDAC were highly variable – even among identical twins [27]. Finally, the high sensitivity of HP patients to alcohol and the strong effect of smoking on the risk of PDAC provided new insights into the role of environmental modifying factors [28].

Since 1996, many additional genetic factors linked to trypsin regulation proved to be strongly associated with susceptibility to and severity of RAP and CP. These include SPINK1 [29, 30], cystic fibrosis transmembrane conductance regulator (CFTR) [31, 32], and CTRC [33–36]. In our US population pathogenic mutations in these four genes are found in 26% of RAP patients and 21% of CP patients [37], not counting the common CTRC G60G risk allele, which is in another 18% of CP patients [36]. Other CP risk genes were also discovered using other candidate gene approaches, including CPA1 [38], and linkage studies including CEL [39] or other approaches such as GGT1 [40].

In 2012 we published the first pancreatitis genome-wide association study (GWAS) [41]. This study identified two major loci, a common PRSS1–PRSS2 haplotype with reduced PRSS1 expression that is protective for multiple etiologies and a common CLDN2 haplotype on the X chromosome, associated with risk of CP, especially in alcoholics. These findings have recently been replicated in a European cohort [42]. These data suggest multiple etiologies and susceptibility factors, with several strong modifying factors that determine the risk of progression and other clinical features of CP. This concept is extended with a recent paper demonstrating that the risk of the common CTRC G60G haplotype is for CP, but not RAP, and is strongly associated with smoking [36].

Mendelian genetic syndromes

An understanding of genetic should begin with simple Mendelian disorders. These disorders are caused by strong pathogenic variants in a single gene that cause well-defined syndromes. In the case of CP, the two most important Mendelian disorders are HP and cystic fibrosis (CF).

Hereditary pancreatitis

HP is defined either by two or more individuals with pancreatitis in two or more generations of the family (i.e., an autosomal dominant pattern of inheritance) or pancreatitis associated with a known disease-causing germ line mutation in the cationic trypsinogen gene PRSS1. The term familial pancreatitis is used when more than one person in the family has RAP or CP – regardless of etiology – since the incidence is above the expected rate in the population by chance alone.

HP has been conclusively linked with gain-of-function mutations in PRSS1 [43–46]. Gain-of-function mutations increase autocatalytic conversion of trypsinogen to active trypsin causing premature, intrapancreatic trypsinogen activation. Trypsin, as the master enzyme regulating activation of the other pancreatic zymogens, is thought to cause widespread enzyme activation, autodigestion of the pancreatic parenchyma, and release of danger-associated molecular pattern (DAMP) molecules that activate the immune system causing AP. Trypsin, chymotrypsin, and other digestive enzymes may also cross-activate the immune system by activating the thrombin pathway or protease-activated receptors [47–51].

Many rare genetic variants in PRSS1 have been reported (see www.pancreasgenetics.org), but the majorities of families either have the PRSS1 N34S or R122H gain-of-function mutation or less commonly, copy number variants (CNV). The other variants may be
loss-of-function variants that cause pancreatic stress and injury signaling through an unfolded protein response [52, 53].

The clinical features of HP have been defined in several large studies [54, 55]. In the European Registry of Hereditary Pancreatitis and Pancreatic Cancer [54], the cumulative risk at 50 years of age for patient with HP for exocrine failure was 37.2%, for endocrine failure 47.6%, and pancreatic resection for pain 17.5%. The cumulative risk of pancreatic cancer was 44.0% at 70 years. In a French study patients with HP reported pancreatic pain (83%), AP (69%), pseudocysts (23%), cholestasis (3%), pancreatic calcifications (61%), exocrine pancreatic insufficiency (34%), DM (26%), and pancreatic adenocarcinoma (5%). In both studies the median age of onset of symptoms was about age 10, with about half the patients developing CP by age 20 years, followed over the next 10 years by pancreatic exocrine insufficiency and DM in up to 40% of patients. The risk of cancer in the fifth to sixth decade of life replicated the studies by Lowenfels [28, 56]. Of note, the incidence of pancreatic cancer is cut in half and delayed by a decade in patients who do not smoke [56].

The diagnosis of HP is made on clinical grounds and genetic testing (see www.pancreas.org). Genetic testing is warranted when there is unexplained documented episode of AP in childhood; recurrent acute attacks of pancreatitis of unknown cause; CP of unknown cause, particularly with onset before age 25 years; and a family history of RAP, CP, or childhood pancreatitis of unknown cause in first-, second-, or third-degree relatives or relatives known to have a mutation in a gene associated with HP [46, 57, 58].

The utility of genetic testing is in making an early diagnosis of a high-risk condition that may explain early functional symptoms and signal the likelihood that the person may develop some or all of the complications of CP. A positive result, in the context of pancreatitis-like symptoms, has a very high likelihood of the symptoms coming from the pancreas. No further diagnostic testing for the etiology of CP-like symptoms is needed. A negative genetic testing result for HP suggests that the etiology is not pathogenic PRSSI variants, although many other pathogenic genetic variants in other loci are also possible (see Chapter 12). Genetic testing, in the future, may also provide guidance on likelihood of specific syndromes, such as constant pain or diabetes, although these ideas currently remain at a research stage.

**Cystic fibrosis**

CF refers to an autosomal recessive disorder affecting secretory epithelial cells of glands, respiratory mucosa, and the digestive system. The term “cystic fibrosis” refers to the CP (with pseudocysts and fibrosis) that occurs in all affected individuals, beginning *in utero*.

The disease is caused by mutations in the *CFTR* gene [59–61]. The CFTR protein forms a regulated anion channel that facilitates transport of chloride and bicarbonate across the apical membrane of epithelial cells during active secretion and/or absorption. CFTR is the most important molecule for the function of the pancreatic duct cell – there are no significant alternate molecules for physiologic anion secretion. Loss of CFTR results in failed flushing of digestive zymogens out of the pancreas and into the intestine. Thus, dysfunction of CFTR results in retention of zymogens in the duct where they can become active and begin digesting the surrounding pancreas, leading to AP. Since the pancreas is so strongly dependent on CFTR function, the severity of pathogenic *CFTR* variants can be estimated from the effects on the pancreas. Furthermore, pancreatic injury can typically be detected at birth, justifying CF screening using serum trypsinogen measurements, and end-stage CP with pancreatic exocrine insufficiency often occurs during the first year of life. Thus, the disease was characterized by failure to thrive and salty sweat with death in infancy until pancreatic enzyme replacement therapy was developed. Only after surviving pancreatic exocrine insufficiency will a child begin developing respiratory failure.

The organs that are most strongly affected by *CFTR* mutations include the pancreas, sweat glands, sinuses, respiratory system, gastrointestinal track, male reproductive system, and liver. The features of *CFTR*-associated diseases depend on the functional consequences of specific mutations on the two *CFTR* alleles [62, 63], as well as mutations in modifier genes and effects of environmental factors. CF is caused by two severe mutations (*CFTR*+/−/*CFTR*−/−). Residual CFTR function can occur with some milder mutations, and the severity of CF is linked to the *least* severe mutation. The milder forms of CF can be referred to as atypical CF (aCF) and are caused by mild-variable mutations with two possible genotypes: (*CFTR*+/−/*CFTR*−/−) or
SNPs that were classified personalized medicine SPINK1 CFTR 5 genotypes is determined by the least variable genotypes. We found that CFTR as a medical model that variants [31, 32]. In many CP cases it appeared that heterozygous pathogenic CFTR variants were found in individuals who also harbored SPINK1 variants as \((CFTR^{wt}/CFTR^{sev})\) or \((SPINK1^{N34S}/SPINK1^{wt})\) genotypes [65–67], a phenomenon called epistasis. Thus, these cases of idiopathic CP were clearly examples of complex trait genetics.

In 2011 we reported that a common \(CFTR\) variant, R75Q, affected bicarbonate conductance while maintaining chloride conductance and had major effects on the pancreas but minimal effects on the lungs, presumably because the pancreas uses CFTR as a bicarbonate channel [67]. Since the functional effect of \(CFTR\) genotypes is determined by the least severe mutation, either two bicarbonate defective (BD) variants \((CFTR^{BD}/CFTR^{BD})\) or one BD and one severe variant \((CFTR^{BD}/CFTR^{sev})\) can result in a monogenic pancreatitis-predominant disorder. We then made a screening panel of 81 previously reported CFTR single-nucleotide polymorphisms (SNPs) and screened nearly a thousand patients with pancreatitis from the North American Pancreatitis Study 2 (NAPS2) cohort [68]. We identified nine \(CFTR\) SNPs that were classified as benign by pulmonologists but were associated with pancreatitis: R74Q, R75Q, R117H, R170H, L967S, L997F, D1152H, S1235R, and D1270N. When these variants were cloned into wild-type CFTR genes and expressed in experimental cells, they had normal chloride conductance but failed to transform into bicarbonate-conducting channels when CFTR was activated with WNK1/SPAK [68]. Molecular modeling demonstrated that four different mechanisms were involved in this transformation and/or regulation of bicarbonate conductance.

The pancreas is susceptible to variants that impair CFTR-mediated bicarbonate conductance because of the way it makes bicarbonate-rich pancreatic juice [68, 69]. Since other organs also use CFTR to secrete bicarbonate, we evaluated the risk of rhinosinusitis and male infertility in patients with CP, with or without the \((CFTR^{BD}/CFTR^{other})\) genotypes. We found that \(CFTR^{BD}\) significantly increased the risk of rhinosinusitis (OR 2.3, \(P<0.005\)) and male infertility (OR 395, \(P<0.001\)). Thus, a variant subtype of CF has been defined that is characterized by CP and dysfunction of other organs that utilize CFTR for bicarbonate secretion, but without lung disease.

A new paradigm of personalized medicine

To advance our understanding of CP, we require a paradigm shift. It is recognized that CP is a complex disorder. It is useful to understand a complex disorder in contrast to a simple disorder [70]. A simple disorder is when a specific microorganism invades a host and causes a specific clinical syndrome. Modern Western medicine has been built on the germ theory of disease, which organizes the study of simple disorders using Koch’s postulates to test a defined hypothesis. In simple diseases the pathologic agent is sufficient to cause the disease syndrome. In contrast, complex disorders typically include acquired conditions caused by complex gene–environment, gene–gene, or multiple gene–environmental interactions where the pathologic agents are neither necessary nor sufficient to cause the disorder. Further complexity occurs if a sequence of pathologic events is needed before enough qualifying features of the syndrome emerge to meet diagnostic criteria. In complex disorders the “scientific method” used in medical research to identify the etiology of disease by applying Koch’s postulates fail, since none of the hypothesized pathogenic agents will meet the four criteria. The challenges of evaluating and managing a complex disorder include developing a new way of thinking about the diagnosis and management of these disorders, integration of complex genetic risk into the paradigm, and developing new tools to assist the practitioner. Specifically, personalized medicine demands going beyond a simple Boolean operator of the germ theory (is a pathologic agent present, yes or no?) to more sophisticated disease modeling and outcome simulation where the influence of multiple variables of different effects can be assessed under different conditions.

The terms personalized medicine and precision medicine are used interchangeably. We will use the term personalized medicine as a medical model that
utilizes genetic information and biomarkers of disease activity to define the specific mechanism of disease within a subject from among multiple possibilities and target disease management at the specific mechanism. In contrast, we use the term precision medicine to define a medical model that optimizes the treatment of the patient within a disease mechanism. Thus, in our view, personalized medicine defines the underlying problem, whereas precision medicine defines the optimal treatment for the problem.

Driven by multiple genetic discoveries and environmental risk assessments on the one hand and a failure to effectively define and treat pancreatic diseases on the other, the CP disease model shifted from “germ theory” (a single agent causing a stereotypic disorder) to a complex genetic disorder with individual patients harboring different combinations of pathogenic factors that alone are neither necessary nor sufficient to cause pancreatic disease [70]. This approach may have profound implications for both early detection and disease management. The new and exciting opportunity is to define the specific risk complex in individual patients, to monitor disease activity and to target pathogenic pathways so that the pathologic endpoints are never reached (see Chapter 12b). This is personalized medicine [70, 71], and this must be the future direction for the pancreatic diseases management since the end stages are irreversible.

References

Chapter 1: Epidemiology and genetics of pancreatitis

42 Derikx MH, Kovacs P, Scholz M, Masson E, Chen JM, Ruffert C, et al. Polymorphisms at PRSS1-PRSS2 and CLDN2-MORC4 loci associate with alcoholic and


64 Kerem E. Atypical CF and CF related diseases. Paediatric Respiratory Reviews 2006;7 Suppl 1:S144-S146. PMID: 16798544


69 Whitcomb DC, Ermentrout GB. A mathematical model of the pancreatic duct cell generating high bicarbonate concentrations in pancreatic juice. Pancreas. 2004;29(2):E30-E40. PMID: 15257112
