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Secretion of the Human Exocrine Pancreas in Health and Disease

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1. Introduction

Secretion from the human exocrine pancreas is essential for nutrient digestion and regulated through complex neurohormonal processes. These secretions are coordinated with responses occurring in digestive and interdigestive periods, and affected by meal composition and patient-related factors. In diseases affecting the exocrine pancreas the amount and/or composition of pancreatic secretion can change and cause maldigestion. Although our understanding of secretion in the healthy state has been well-characterized, knowledge of the changes that occur during disease states remains limited. Management of pancreatic insufficiency is primarily focused on replacement of digestive enzymes, particularly lipase, with exogenous pancreatic enzymes. Advances in techniques for the collection and analysis of pancreatic secretions have renewed interest in the study of pancreatic secretion for studies of both pancreatic physiology and disease. In this chapter we review various aspects of human pancreatic secretion in health and disease. The treatment of exocrine pancreatic insufficiency, including rationale for dosing of pancreatic enzyme replacement therapy (PERT) and evaluation of ongoing steatorrhea despite pancreatic enzyme replacement therapy are not within the scope of the current chapter, and have been reviewed elsewhere (12).

2. Anatomy of the exocrine pancreas

The anatomy of the exocrine pancreas is reviewed in detail elsewhere (43). In brief, the functional exocrine unit of the pancreas consists of acinar cells and a network of pancreatic ductules and ducts. The production of the vast majority of pancreatic enzymes occurs in the acinar cells. Of the various pancreatic enzymes, proteases (e.g., trypsin, carboxypeptidases, chymotrypsin, and elastase) are the most abundant (according to weight) comprising around 90% of the enzymes found in human pancreas fluid; amylase, lipase, and nucleases are relatively less abundant (54). Following production and storage of inactive proenzymes (termed zymogens) and active enzymes in the acinar cells, they are secreted into a network of pancreatic ductules and ducts in response to a meal. These conduits are lined with ductal epithelial cells, which facilitate transport of enzymes in a bicarbonate-rich fluid. Secretion from acinar and ductal cells is highly regulated, primarily in response to nutrient ingestion and neural stimulation (8). Diseases that interfere with either the function of the ductal cells or the continuity of the ductal network will lead to exocrine pancreatic insufficiency, as discussed below.

3. Normal secretion of the exocrine pancreas

During the interdigestive (i.e., fasting) state, exocrine secretion is somewhat cyclic and

associated with the three phases of gastrointestinal motility (32). Pancreatic secretion within the digestive period is much greater than the interdigestive period. This secretion is highly regulated by neurohormonal responses and occurs in three primary phases – cephalic, gastric, and intestinal (8, 33). The initial cephalic phase is primarily controlled by the vagal nerve and results in levels of enzyme secretion that account for 20-25% of the total elicited by a meal (8). Next, gastropancreatic reflexes are activated by gastric distention, but only produce a small increase in pancreatic enzyme secretion. Finally, the intestinal phase is activated with transit of the chyme into the duodenum, resulting in the majority (50-80%) of the pancreatic stimulus associated with a meal (8). Together these responses cause a rapid increase in enzyme secretion to a maximal output about 1 hour after meal ingestion (15, 21). The enzyme output generally remains elevated for approximately 3-4 hours, but can vary depending on multiple factors (15, 21, 44). The cessation of pancreatic enzyme secretion is believed to be primarily the result of nutrient exposure to the distal small intestine, based on experiments demonstrating the inhibition of endogenously stimulated secretion following ileal perfusion of carbohydrates or lipids (38). Hormones believed to be involved in this feedback inhibition include peptide YY and glucagon-like peptide-1(33).

Several meal-related factors influence exocrine pancreatic secretion, including caloric content, nutrient composition, and physical properties of the meal. In regards to caloric content, there appears to be a minimal and maximal threshold of pancreas stimulation, with the maximal enzyme response occurring after consumption of approximately 500 kcal (7). Additionally, the meal composition of fats, carbohydrates, and proteins influences enzyme release. Specifically, chronic exposure to dietary fat appears to be a stronger stimulant of pancreatic enzyme secretion than other nutrients. For example, healthy subjects exposed to a high fat diet for two weeks had an enzyme output that was 2 to 4 times greater than those on a high carbohydrate diet in the immediate postprandial and interdigestive periods, respectively (5). Although temporary exposure to a high fat diet 24 hours also increased enzyme output in this study, the observation was not replicated with intraduodenal infusion of different nutrient compositions (28). Another key observation is that ingestion of a solid meal results in a more sustained enzyme response when compared to an identical homogenized meal (5, 44).

In addition to the secretion of proenzymes and activated pancreatic enzymes, the secretion of a bicarbonate rich fluid has an important role. The transport of bicarbonate by duct cells into the pancreatic duct lumen is mediated by the cystic fibrosis transmembrane conductance regulator (CFTR) chloride secretory pathway. This transporter can either secrete chloride to drive a chloride-bicarbonate exchanger on duct cells or directly transport bicarbonate into the duct lumen. While the primary dietary factors that stimulate acinar cell enzyme secretion are the hydrolysis of proteins and fats into amino acids and fatty acids, duodenal pH appears to be the primary driving force for ductal fluid and electrolyte secretion. After ingestion of a meal in a healthy subject, the duodenal pH changes from a baseline near 7.0 to a nadir of 5.0-4.5 in the early postprandial phase (15, 44). When a threshold of about pH 4.5 is reached in the duodenum, secretin is released and secretion of bicarbonate-rich fluids into the duodenum follows. The alkaline fluid in the duodenum has several functions including: inactivating pepsin, increasing the solubility of fatty acids and bile acids, maintaining an optimal pH (>4.0) for pancreatic and brush border enzymes, and preventing intestinal mucosal damage.

4. Alterations in exocrine secretion in disease

Pancreatic enzyme secretion is a highly controlled process during health, so it is not surprising that varying degrees of dysfunction are observed in disorders of the exocrine pancreas. Although enzyme response to different nutrients has been well characterized in a healthy state, much less is known regarding the extent of change in disease. Rather, most studies have focused on the clinical ramifications of inadequate enzyme production and/or secretion.

Mechanisms of abnormal exocrine function

Pancreatic disorders can decrease the gland's digestive activities by decreasing enzyme production, impairment of enzyme secretion, and/or reduced mixing of enzymes with food. Reduced acinar cell mass leads to decreased enzyme production and arises from disorders of primary and secondary exocrine pancreatic insufficiency (EPI). Primary disorders are illustrated by Shwachman-Diamond syndrome and Johanson-Blizzard syndrome and secondary causes include chronic pancreatitis, acute severe inflammation or pancreatic surgical resection. However, for most diseases an isolated parenchymal insult is not adequate to cause clinically manifest exocrine pancreatic insufficiency. Patients with reduced enzyme secretion can have impairment at the cellular (e.g., impaired duct cell function in cystic fibrosis) or macroscopic level (e.g., obstruction of the main pancreatic duct secondary to a stricture, intraductal stone, or mass). Impaired mixing of pancreatic enzymes with food often occurs in the postoperative setting, such as following a gastrojejunostomy. Additionally, reduced fluid and bicarbonate secretion by ductal cells can lead to impaired mixing and also result in a more acidic pH than is optimal for enzyme activities (2, 33).

Chronic pancreatitis

Chronic pancreatitis is one of the most common causes of exocrine pancreatic insufficiency (EPI), with insufficiency observed in 30 to 90% depending on the clinical severity of chronic pancreatitis (39). In a study examining the natural history of chronic pancreatitis, EPI was observed to develop slowly and was often first manifest 10-15 years after symptom onset (the most common is pain) (39). This delay is attributable to the dramatic functional reserve of the exocrine pancreas, requiring loss of more than 90% of exocrine pancreatic function before developing malabsorption. Whether or not the reduction of the various pancreatic enzymes occurs in parallel remains unclear. However, the clinical abnormalities are predominantly related to fat maldigestion. This is primarily a consequence of minimal redundancy of lipase production from non-pancreatic sources. In contrast, 5% of normal amylase content is sufficient to maintain starch digestion; this amount can typically be reached by a combination of salivary gland and brush border oligosaccharidase production of amylase (40). In a landmark study, DiMagno and colleagues demonstrated that steatorrhea (>7gram fat/24 hours) develops when the lipase output is <10% of normal (13). The reported lipase output in chronic pancreatitis may range from 1-60% of levels found in healthy controls (33). The wide variability is in part due to methodologic differences in the studies (e.g., method of pancreas stimulation and collection methods), but also likely reflects that subjects were studied at various stages of disease.

Chronic pancreatitis can alter all aspects of pancreatic enzyme secretion, so EPI is typically multifactorial. First, parenchymal fibrosis can lead to acinar cell injury causing atrophy and resultant loss of enzyme production. Second, duct cell function is impaired in chronic pancreatitis leading to decreased bicarbonate secretion (10). This causes postprandial duodenal pH to be lower in chronic pancreatitis than in healthy subjects; this value can exceed the pH threshold (pH 4.5) for irreversible lipase inactivation (15). Obstruction of the main pancreatic duct can also occur due to fibrotic strictures and intraductal calculi and likely contributes to reduced duct pancreatic secretion. However, neither endoscopic nor surgical

correction of main pancreatic duct obstruction has been convincingly demonstrated to restore pancreatic enzyme secretion. Since the functional and tissue loss found in chronic pancreatitis is currently not reversible, clinical treatment of exocrine insufficiency is limited to oral supplementation with pancreatic enzyme replacement therapy.

Cystic fibrosis

The CFTR gene codes for a chloride channel, which is central to the production of fluid and electrolyte secretion in various exocrine glands. In the pancreas, the ultimate pathophysiologic result of CFTR mutations is decreased electrolyte and fluid secretion into the pancreatic duct lumen. The consequence of its reduced function or loss is a more viscous and relatively acidic pancreatic fluid. Although acinar cells may not be directly affected, both pancreatic enzyme secretion and function are impaired. In addition to impaired flow, the acidic pancreatic duct lumen can result in precipitation of mucins, secretory protein aggregation, and premature activation of digestive enzymes (48). Furthermore, the decreased bicarbonate secretion leads to a more acidic intraluminal pH in the duodenum. It has been observed that the pH is lower by 1-2 units in both the interdigestive and digestive periods in cystic fibrosis compared to controls (24). Collectively, EPI is prevalent in classic CF with estimates that 80% of patients with CF will develop EPI by 2 years of life (18). Importantly, EPI appears to be mostly limited to those subjects with two severe deleterious gene mutations, though pancreatic disease can rarely occur in those with less severe mutations (18)

As in other diseases, it has been demonstrated that the impaired lipase secretion may at least partially be compensated by increased activity of gastric lipase (3). Otherwise, duodenal output of pancreatic enzymes has not been well studied in cystic fibrosis. There are multiple methodologic challenges to completing this, but probably most limiting is that EPI manifests at a very young age, which poses several technical and ethical issues for research study design. In these patients with EPI, lifelong oral pancreatic enzyme replacement is generally required (11, 55).

Acute pancreatitis

In contrast to chronic pancreatitis and cystic fibrosis, which are chronic, progressive diseases, acute pancreatitis is a multiphasic disease. Thus, impairment of pancreas enzyme secretion is variable and depends on the clinical severity of acute pancreatitis and chronologic proximity to the inciting insult. Knowledge of pancreatic enzyme secretion during the early phase of acute pancreatitis is limited. Since exogenous pancreatic stimulation would potentially worsen underlying clinical disease, studies have not been performed in this context. However, there does appear to be some impaired exocrine function during the first month after disease onset for most patients (59). The prevalence of early onset EPI (diagnosed using fecal elastase-1 level) is particularly high (>80%) in those with clinically severe acute pancreatitis or pancreatic necrosis (6). The impairment is mild and temporary for the vast majority of patients, but can potentially be irreversible (46). The prevalence remains uncharacterized, however patients with severe acute pancreatitis may also develop EPI following clinical resolution of the episode due parenchymal atrophy or fibrosis.

Pancreatic cancer

The changes in pancreas enzyme secretion are likewise not well characterized in pancreatic cancer. Although the prevalence of EPI in pancreatic cancer has not been characterized in a large clinical cohort, estimates from smaller series suggests this ranges from 50 to 60% (4, 51, 58). In addition to tumor-mediated parenchymal destruction, abnormal secretion can develop as a consequence of pancreatic duct obstruction, and following surgical resection. The location of the tumor is an important factor in determining whether pancreatic secretion remains adequate. In one study, a shorter length of unaffected

pancreatic duct (i.e., a tumor located closer to the pancreatic head) was associated with decreased bicarbonate and enzyme output in response to CCK stimulation (14).

Pancreatic surgery

Alterations in exocrine secretion following pancreatic surgery are influenced by the type of surgery, extent of resection, and health of the remnant pancreatic parenchyma. The exocrine function remains largely unchanged, and occasionally improves, in those undergoing a drainage procedure with or without minor resection of the parenchyma (29). Conversely, clinically diagnosed EPI is observed in approximately 25-50% of those following a pancreaticoduodenectomy (29, 30, 45, 58, 62). In patients undergoing surgery for a diffuse disease, such as chronic pancreatitis, the risk of abnormal secretion following surgery is increased. For example, in a clinical series limited to subjects with chronic pancreatitis, over 50% developed clinically overt steatorrhea following pancreaticoduodenectomy and up to 40% following distal pancreatectomy (20, 31, 58).

Non-pancreatic etiologies of impaired exocrine pancreatic function

In addition to the preceding pancreatic etiologies, there are a few non-pancreatic diseases that can indirectly impair exocrine pancreatic function. For example, in those undergoing a partial or total gastrectomy there are multiple factors that decrease intraluminal enzyme activity. The greatest contributing factor is post-cibal asynchrony, which describes disordered mixing of meal contents and pancreatic enzymes in the small intestine (33). The asynchrony is further compounded by pancreatic denervation during surgical dissection and the loss of compensatory gastric lipase production. Asynchrony is also a contributing factor to the EPI observed following pancreaticoduodenectomy. Although steatorrhea is not universally present in patients with Zollinger-Ellison syndrome (ZES), it is important to consider the effect on exocrine pancreatic function due to its unique pathophysiologic mechanism (53). In ZES there is excessive gastrin production which leads to inappropriate gastric acid secretion. This lowers the duodenal pH beyond the threshold at which normally secreted pancreatic enzymes are are inactivated (23) The relatively acidic pH in the duodenal lumen in ZES also leads to bile salt precipitation. Lastly, there is evidence to demonstrate the presence of impaired exocrine function in patients without clinically evident pancreatic disease. For example, patients with type 2 diabetes mellitus can develop fibrosis that is histologically similar to chronic pancreatitis (47). This condition, currently referred to as diabetic exocrine pancreatopathy, differs from chronic pancreatitis in that chronic inflammatory cells are absent and overt EPI is rare. Exocrine pancreatic insufficiency may also occur in celiac disease (when measured using fecal elastase levels) despite the absence of structural changes in the pancreas (52, 60). The EPI in celiac disease is generally attributed with mucosal injury, but may also be due to disruption of enteric-mediated cholecystokinin stimulation of the pancreas and gallbladder emptying. In children and some adults, short-term support with oral pancreatic enzymes is often considered. Pancreatic secretion usually resolves with mucosal healing on a gluten free diet (52). EPI has also been reported to occur in Crohn's disease. Additional studies are needed to further characterize the potential mechanisms of exocrine dysfunction in each of these diseases (33).

5. Measurement of human pancreatic secretion (direct pancreatic function testing)

Our knowledge of normal human pancreatic secretion primarily comes from human studies using tube(s) to collect intestinal fluids following

Table 1. Comparison of direct and indirect pancreatic function tests. Estimates of sensitivity and specificity are assigned semi-quantitatively due to extreme heterogeneity in study designs, which prevents accurate pooling (9, 26).

Name	Sensitivity	Specificity	Advantages	Disadvantages	Cost
Direct Pancreatic Function Tests:					
CCK-stimulated pancreatic function test	High	High	• Provides the most direct measure of pancreatic enzyme output	• Tubes are no longer available to measure enzyme output • Invasive	$$$$
Secretin-stimulated pancreatic function test	High	Moderate	• Highly sensitive in early stages of disease • High negative predictive value to rule out chronic pancreatitis	• Invasive • False positives: CFTR mutations and cigarette smoking • Does not directly assess acinar cell function	$$$$
Indirect Pancreatic Function Tests:					
Coefficient of fat absorption (CFA)	High	Moderate	• Highly accurate for fat malabsorption • Can be used to monitor PERT • Noninvasive	• Requires 3 day stool collection • False positives: any cause of fat malabsorption (which must be excluded before diagnosing EPI)	$
Fecal elastase (FE-1)	Mild - Moderate	Moderate-High	• Convenient collection • Noninvasive	• Lower accuracy in those with mild EPI or history of pancreatic resection • Cannot be used to monitor PERT • False positives: bacterial overgrowth, watery stool	$
^{13}C-mixed triglyceride breath test (MTBT)	Moderate	Moderate	• Can be used to monitor PERT • Noninvasive	• Limited availability • Time consuming test	$$

EPI, exocrine pancreatic insufficiency; PERT, pancreatic enzyme replacement therapy

pancreatic stimulation with either a standardized meal or an intravenous secretagogue such as secretin or cholecystokinin. The use of exogenous stimulation to measure pancreatic function is referred to as direct pancreatic function testing (PFT) (**Table 1**).

Although different types of tubes have been used, one of the most recognized is the Dreiling tube, which permitted simultaneous aspiration from the gastric and duodenal lumens. One drawback of this approach was that the enzyme output could not be determined because the flow rate could not be calculated. To resolve this issue, a method was developed using a second tube placed with ports in the both proximal and distal duodenum. A non-absorbable marker (typically polyethylene glycol) was infused through the proximal port and aspirated with pancreatic secretions through the distal port (22). Using this marker-perfusion method permitted calculation of pancreatic enzyme output as well as a correction coefficient

for the amount of fluid not collected distally. A variety of endogenous and exogenous stimuli were administered, and the changes in pancreatic secretions could be characterized. Significant drawbacks to these techniques included patient discomfort and prolonged fluoroscopy time to maintain proper tube location. Despite these challenges, the vast majority of the data supporting our understanding of human pancreatic secretory physiology was acquired using this technique.

Further studies regarding the mechanisms of human pancreatic secretion became possible following adaptation of early work demonstrating the ability to isolate functioning pancreatic acini from rodents (61). The ability to isolate acini from human pancreases and study them in a controlled environment has led to further understanding of the cellular mechanisms of pancreatic secretion in health and disease; however, these findings are beyond the scope of this review (42).

More than a decade ago, an endoscopic-based pancreatic function test (ePFT) was developed that greatly improved patient tolerance and eliminated the need for fluoroscopy (10). Although the stimuli for pancreatic secretion remained similar (i.e., cholecystokinin (CCK) and/or secretin), the method of fluid collection was through the suction channel of the endoscope rather than an enteric tube. Since there is only one "port" for collecting samples through the endoscope, this technique permits determination of enzyme and analyte concentration, but not enzyme output (or flow rate). The most common use of the ePFT is for evaluation of suspected chronic pancreatitis. However, there is emerging utilization of this methodology for translational science, including the study of disease biomarkers (27).

Currently, the most commonly used measurement made using ePFT is the peak bicarbonate concentration following secretin stimulation. Although this measurement is more directly a measure of pancreatic duct-cell function than acinar-cell function, previous studies have demonstrated that peak bicarbonate corresponds to peak lipase concentrations in the pancreatic fluid of healthy subjects as well as chronic pancreatitis (56). For the evaluation of chronic pancreatitis, measurement of peak bicarbonate concentration (following secretin stimulation) has improved discrimination compared to peak lipase or amylase concentrations (following CCK stimulation) (37). Importantly, the negative predictive value (97%) of a normal bicarbonate response to secretin stimulation to rule out chronic pancreatitis is very good, however the positive predictive value (45%) is only fair (35). Thus, it is currently more effective to use endoscopic function tests to "rule out" a diagnosis of chronic pancreatitis.

In addition to the previously mentioned electrolytes and pancreatic enzymes, there is a large number of other potential analytes in pancreatic fluid. During an ePFT there is typically an abundant volume of fluid collected, which provides the opportunity for other studies. Investigators have begun to explore various molecular targets in pancreas fluid, including: protein expression, cytokines, DNA methylation markers, microRNAs, and genetic mutations (27). Various markers are being examined for the purposes of identifying diagnostic or disease biomarkers, as well as observations that may lead to novel therapeutic approaches. Although early studies were primarily limited to proteomics, there has been a recent resurgence in other areas with exciting preliminary findings (49, 50). For example, Abu Dayyeh et al. recently demonstrated that prostaglandin E2 (PGE2) is a promising disease biomarker for the various stages of chronic pancreatitis (1). Levels are differentially expressed in early and advanced chronic pancreatitis in contrast to healthy controls, with areas under the curve (AUC) of 0.62 and 0.9, respectively. When used in combination with the pancreatic fluid bicarbonate, the AUC for diagnosis of early and advanced chronic

pancreatitis were 0.94 and 1.0, respectively. Another recent example of biomarker discovery includes the identification of a series of DNA hypermethylation markers that identify patients with pancreatic cancer compared to controls with AUCs ranging from 0.62-0.92 (36).

6. Indirect pancreatic function testing

Tests measuring pancreatic function without the use of hormonal stimulation are referred to as indirect PFTs. Since the indirect tests are typically less accurate for detecting early stages of exocrine dysfunction they are more helpful for quantifying the degree of insufficiency in those with known pancreatic disease, rather than diagnosis. Indirect PFTs are non-invasive and typically less expensive, so the selection of the pancreatic function test to be employed in the clinical setting requires considering the tradeoff between diagnostic test performance, invasiveness, and costs (**Table 1**). For example, indirect PFTs are generally adequate to identify EPI in patients with overt morphological changes (e.g. calcifications and/or main pancreatic duct dilation). In contrast, a direct PFT would be preferred to identify the presence of EPI in a patient with clinical suspicion of chronic pancreatitis and normal or equivocal imaging tests (9). Poorly studied indirect tests (including serum trypsin, fecal chymotrypsin, and qualitative fecal fat analysis) are not reviewed here.

Coefficient of fat absorption

Among indirect testing methods, the coefficient of fat absorption (CFA) is considered the gold standard to diagnose fat malabsorption from any cause, and to document and quantify EPI in those with pancreatic disease (19, 51, 63). This test involves consumption of a high-fat diet (100 grams/day) for at least 5 days with stool collection during the final three days of the diet. The daily dietary fat intake is recorded, and factored into the final CFA calculation, using the following equation:

CFA (%) = 100 x [(mean daily fat intake – mean daily stool fat) / mean daily fat intake]

A CFA of <0.93 (which corresponds to >7 grams of stool fat per 24 hour period while on a 100 g fat diet) is considered abnormal (26). Although this test is accurate when carefully performed, the three-day collection period can be inconvenient for patients and errors may occur when performed in a non-controlled environment with either collection or processing. This test is also useful to assess adequacy of PERT dosing, but follow-up testing is rarely performed outside of the research setting

Fecal elastase-1

In contrast to the CFA, a fecal elastase-1 (FE-1) level can be determined from a single formed stool sample. The test's convenience makes it the most widely used indirect PFT in clinical practice. Pancreatic elastase is resistant to degradation as it passes through the gut, so it can be measured in the stool (57). Although early studies demonstrated strong correlation with pancreatic enzyme output during direct PFT, more recent studies have demonstrated only fair accuracy in mild EPI (26). It is important to recognize that false positives will occur if a liquid stool specimen is analyzed. Also, low levels are commonly observed in diabetes mellitus (both type 1 and type 2 diabetes) but it is uncertain if this truly represents EPI (25, 41). Lastly, since FE-1 levels detected by monoclonal (but not polyclonal) assays are unaffected by PERT this test is not useful for monitoring the adequacy of therapy.

^{13}C-mixed triglyceride breath test

The ^{13}C-mixed triglyceride breath test (MTBT) is another indirect test that measures the intraluminal lipolytic activity as an estimate of exocrine pancreatic function. The test involves ingesting a standardized meal that includes triglycerides with radiolabelled carbon tracers (26). The triglycerides are hydrolyzed by

pancreatic lipase releasing ^{13}C, which is absorbed, then transported to the liver. In the liver lipolysis and beta-oxidation occur leading to the formation of $^{13}CO_2$. These molecules are exhaled by the lungs and measured in serial breath samples. A decrease in the recovery of $^{13}CO_2$ is associated with decreased pancreatic lipase secretion and fat malabsorption as measured by CFA (16, 17, 34). Although this is a non-invasive test, it lasts approximately 6 hours and the radiotracers are not universally available, so the clinical utility of this test worldwide is limited.

7. Summary

Pancreas exocrine secretion is a complex response to a meal which involves the coordinated release and transport of enzymes from acinar cells and fluid and electrolytes into the pancreatic ductal and ultimately the duodenal lumen where they are required for normal digestion. Essentially all pancreatic disorders may alter this process to different degrees, and do so through a variety of mechanisms. There are various methods for determination of enzyme output and bicarbonate secretion in response to endogenous and exogenous pancreatic stimulation. These tools have shaped our current understanding of pancreas physiology, and hold significant potential for biomarker discovery and identification of novel therapeutic targets.

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8. References

1. **Abu Dayyeh BK, Conwell D, Buttar NS, Kadilaya V, Hart PA, Baumann NA, Bick BL, Chari ST, Chowdhary S, Clain JE, Gleeson FC, Lee LS, Levy MJ, Pearson RK, Petersen BT, Rajan E, Steen H, Suleiman S, Banks PA, Vege SS, and Topazian M.** Pancreatic juice prostaglandin e2 concentrations are elevated in chronic pancreatitis and improve detection of early disease. *Clinical and Translational Gastroenterology* 6: e72, 2015. PMID: 25630864.
2. **Andersen JR, Bendtsen F, Ovesen L, Pedersen NT, Rune SJ, and Tage-Jensen U.** Pancreatic insufficiency. Duodenal and jejunal pH, bile acid activity, and micellar lipid solubilization. *International Journal of Pancreatology* 6: 263-270, 1990. PMID: 2212745.
3. **Balasubramanian K, Zentler-Munro PL, Batten JC, and Northfield TC.** Increased intragastric acid-resistant lipase activity and lipolysis in pancreatic steatorrhoea due to cystic fibrosis. *Pancreas* 7: 305-310, 1992. PMID: 1594551.
4. **Bartel MJ, Asbun H, Stauffer J, and Raimondo M.** Pancreatic exocrine insufficiency in pancreatic cancer: A review of the literature. *Digestive and Liver Disease*, 2015. PMID: 26211872.
5. **Boivin M, Lanspa SJ, Zinsmeister AR, Go VL, and DiMagno EP.** Are diets associated with different rates of human interdigestive and postprandial pancreatic enzyme secretion? *Gastroenterology* 99: 1763-1771, 1990. PMID: 2227289.
6. **Boreham B and Ammori BJ.** A prospective evaluation of pancreatic exocrine function in patients with acute pancreatitis: correlation with extent of necrosis and pancreatic endocrine insufficiency. *Pancreatology* 3: 303-308, 2003. PMID: 12890992.
7. **Brunner H, Northfield TC, Hofmann AF, Go VL, and Summerskill WH.** Gastric emptying and secretion of bile acids, cholesterol, and pancreatic enzymes during digestion. Duodenal perfusion studies in healthy subjects. *Mayo Clinic Proceedings* 49: 851-860, 1974. PMID: 4214308.
8. **Chandra R, Liddle RA.** Regulation of Pancreatic Secretion. *Pancreapedia: Exocrine Pancreas Knowledge Base*, 2015. DOI:10.3998/panc.2015.38

9. **Conwell DL, Lee LS, Yadav D, Longnecker DS, Miller FH, Mortele KJ, Levy MJ, Kwon R, Lieb JG, Stevens T, Toskes PP, Gardner TB, Gelrud A, Wu BU, Forsmark CE, and Vege SS.** American Pancreatic Association practice guidelines in chronic pancreatitis: evidence-based report on diagnostic guidelines. *Pancreas* 43: 1143-1162, 2014. PMID: 25333398.
10. **Conwell DL, Zuccaro G, Jr., Vargo JJ, Trolli PA, Vanlente F, Obuchowski N, Dumot JA, and O'Laughlin C.** An endoscopic pancreatic function test with synthetic porcine secretin for the evaluation of chronic abdominal pain and suspected chronic pancreatitis. *Gastrointestinal Endoscopy* 57: 37-40, 2003. PMID: 12518128.
11. **Cystic Fibrosis F, Borowitz D, Robinson KA, Rosenfeld M, Davis SD, Sabadosa KA, Spear SL, Michel SH, Parad RB, White TB, Farrell PM, Marshall BC, and Accurso FJ.** Cystic Fibrosis Foundation evidence-based guidelines for management of infants with cystic fibrosis. *The Journal of Pediatrics* 155: S73-93, 2009. PMID: 19914445.
12. **DiMagno EP and DiMagno MJ.** Chronic Pancreatitis: Landmark Papers, Management Decisions, and Future. *Pancreas* 45: 641-650, 2016. PMID: 27077713.
13. **DiMagno EP, Go VL, and Summerskill WH.** Relations between pancreatic enzyme ouputs and malabsorption in severe pancreatic insufficiency. *N Engl J Med* 288: 813-815, 1973. PMID: 4693931.
14. **DiMagno EP, Malagelada JR, and Go VL.** The relationships between pancreatic ductal obstruction and pancreatic secretion in man. *Mayo Clinic Proceedings* 54: 157-162, 1979. PMID: 431121.
15. **DiMagno EP, Malagelada JR, Go VL, and Moertel CG.** Fate of orally ingested enzymes in pancreatic insufficiency. Comparison of two dosage schedules. *N Engl J Med* 296: 1318-1322, 1977. PMID: 16213.
16. **Dominguez-Munoz JE, Iglesias-Garcia J, Castineira Alvarino M, Luaces Regueira M, and Larino-Noia J.** EUS elastography to predict pancreatic exocrine insufficiency in patients with chronic pancreatitis. *Gastrointestinal Endoscopy* 81: 136-142, 2015. PMID: 25088920.
17. **Dominguez-Munoz JE, Iglesias-Garcia J, Vilarino-Insua M, and Iglesias-Rey M.** 13C-mixed triglyceride breath test to assess oral enzyme substitution therapy in patients with chronic pancreatitis. *Clin Gastroenterol Hepatol* 5: 484-488, 2007. PMID: 17445754.
18. **Durno C, Corey M, Zielenski J, Tullis E, Tsui LC, and Durie P.** Genotype and phenotype correlations in patients with cystic fibrosis and pancreatitis. *Gastroenterology* 123: 1857-1864, 2002. PMID: 12454843.
19. **Forsmark CE.** Management of chronic pancreatitis. *Gastroenterology* 144: 1282-1291 e1283, 2013. PMID: 23622138.
20. **Frey CF, Child CG, and Fry W.** Pancreatectomy for chronic pancreatitis. *Annals of Surgery* 184: 403-413, 1976. PMID: 1015887.
21. **Fried M, Mayer EA, Jansen JB, Lamers CB, Taylor IL, Bloom SR, and Meyer JH.** Temporal relationships of cholecystokinin release, pancreatobiliary secretion, and gastric emptying of a mixed meal. *Gastroenterology* 95: 1344-1350, 1988. PMID: 3169498.
22. **Go VL, Hofmann AF, and Summerskill WH.** Simultaneous measurements of total pancreatic, biliary, and gastric outputs in man using a perfusion technique. *Gastroenterology* 58: 321-328, 1970. PMID: 5437985.
23. **Go VL, Poley JR, Hofmann AF, and Summerskill WH.** Disturbances in fat digestion induced by acidic jejunal pH due to gastric hypersecretion in man. *Gastroenterology* 58: 638-646, 1970. PMID: 4315422.
24. **Gregory PC.** Gastrointestinal pH, motility/transit and permeability in cystic fibrosis. *Journal of Pediatric Gastroenterology and Nutrition* 23: 513-523, 1996. PMID: 8985838.
25. **Hahn JU, Kerner W, Maisonneuve P, Lowenfels AB, and Lankisch PG.** Low fecal elastase 1 levels do not indicate exocrine pancreatic insufficiency in type-1 diabetes mellitus. *Pancreas* 36: 274-278, 2008. PMID: 18362841.
26. **Hart PA and Conwell DL.** Diagnosis of Exocrine Pancreatic Insufficiency. *Current Treatment Options in Gastroenterology* 13: 347-353, 2015. PMID: 26077487.
27. **Hart PA, Topazian M, Raimondo M, Cruz-Monserrate Z, Fisher WE, Lesinski GB, Steen H, and Conwell DL.** Endoscopic Pancreas Fluid Collection: Methods and Relevance for Clinical Care and Translational Science. *Am J Gastroenterol* 111: 1258-1266, 2016. PMID: 27481304.

28. **Holtmann G, Kelly DG, and DiMagno EP.** Nutrients and cyclical interdigestive pancreatic enzyme secretion in humans. *Gut* 38: 920-924, 1996. PMID: 8984034.
29. **Jalleh RP and Williamson RC.** Pancreatic exocrine and endocrine function after operations for chronic pancreatitis. *Annals of Surgery* 216: 656-662, 1992. PMID: 1466619.
30. **Kachare SD, Fitzgerald TL, Schuth O, Vohra NA, and Zervos EE.** The impact of pancreatic resection on exocrine homeostasis. *American Surgeon* 80: 704-709, 2014. PMID: 24987904.
31. **Kalser MH, Leite CA, and Warren WD.** Fat assimilation after massive distal pancreatectomy. *The New England Journal of Medicine* 279: 570-576, 1968. PMID: 5667467.
32. **Keller J, Groger G, Cherian L, Gunther B, and Layer P.** Circadian coupling between pancreatic secretion and intestinal motility in humans. *American Journal of Physiology Gastrointestinal and Liver Physiology* 280: G273-278, 2001. PMID: 11208550.
33. **Keller J and Layer P.** Human pancreatic exocrine response to nutrients in health and disease. *Gut* 54 Suppl 6: vi1-28, 2005. PMID: 15951527.
34. **Keller J, Meier V, Wolfram KU, Rosien U, and Layer P.** Sensitivity and specificity of an abbreviated (13)C-mixed triglyceride breath test for measurement of pancreatic exocrine function. *United European Gastroenterology Journal* 2: 288-294, 2014. PMID: 25083286.
35. **Ketwaroo G, Brown A, Young B, Kheraj R, Sawhney M, Mortele KJ, Najarian R, Tewani S, Dasilva D, Freedman S, and Sheth S.** Defining the accuracy of secretin pancreatic function testing in patients with suspected early chronic pancreatitis. *Am J Gastroenterol* 108: 1360-1366, 2013. PMID: 23711627.
36. **Kisiel JB, Raimondo M, Taylor WR, Yab TC, Mahoney DW, Sun Z, Middha S, Baheti S, Zou H, Smyrk TC, Boardman LA, Petersen GM, and Ahlquist DA.** New DNA Methylation Markers for Pancreatic Cancer: Discovery, Tissue Validation, and Pilot Testing in Pancreatic Juice. *Clinical Cancer Research*, 2015. PMID: 26023084.
37. **Law R, Lopez R, Costanzo A, Parsi MA, and Stevens T.** Endoscopic pancreatic function test using combined secretin and cholecystokinin stimulation for the evaluation of chronic pancreatitis. *Gastrointestinal endoscopy* 75: 764-768, 2012. PMID: 22046082.
38. **Layer P, Peschel S, Schlesinger T, and Goebell H.** Human pancreatic secretion and intestinal motility: effects of ileal nutrient perfusion. *Am J Physiol* 258: G196-201, 1990. PMID: 1689548.
39. **Layer P, Yamamoto H, Kalthoff L, Clain JE, Bakken LJ, and DiMagno EP.** The different courses of early- and late-onset idiopathic and alcoholic chronic pancreatitis. *Gastroenterology* 107: 1481-1487, 1994. PMID: 7926511.
40. **Layer P, Zinsmeister AR, and DiMagno EP.** Effects of decreasing intraluminal amylase activity on starch digestion and postprandial gastrointestinal function in humans. *Gastroenterology* 91: 41-48, 1986. PMID: 2423408.
41. **Leeds JS, Oppong K, and Sanders DS.** The role of fecal elastase-1 in detecting exocrine pancreatic disease. *Nature reviews Gastroenterology & Hepatology* 8: 405-415, 2011. PMID: 21629239.
42. **Lerch MM and Gorelick FS.** Models of acute and chronic pancreatitis. *Gastroenterology* 144: 1180-1193, 2013. PMID: 23622127.
43. **Longnecker DS.** Anatomy and Histology of the Pancreas. *Pancreapedia: Exocrine Pancreas Knowledge Base*, 2014. DOI: 10.3998/panc.2014.3
44. **Malagelada JR, Go VL, and Summerskill WH.** Different gastric, pancreatic, and biliary responses to solid-liquid or homogenized meals. *Dig Dis Sci* 24: 101-110, 1979. PMID: 371939.
45. **Matsumoto J and Traverso LW.** Exocrine function following the whipple operation as assessed by stool elastase. *Journal of Gastrointestinal Surgery* 10: 1225-1229, 2006. PMID: 17114009.
46. **Mitchell CJ, Playforth MJ, Kelleher J, and McMahon MJ.** Functional recovery of the exocrine pancreas after acute pancreatitis. *Scandinavian Journal of Gastroenterology* 18: 5-8, 1983. PMID: 6609418.
47. **Mohapatra S, Majumder S, Smyrk TC, Zhang L, Matveyenko A, Kudva YC, and Chari ST.** Diabetes Mellitus Is Associated With an Exocrine Pancreatopathy: Conclusions From a Review of Literature. *Pancreas*, 2016. PMID: 26918874.

48. **Pallagi P, Hegyi P, and Rakonczay Z, Jr.** The Physiology and Pathophysiology of Pancreatic Ductal Secretion: The Background for Clinicians. *Pancreas* 44: 1211-1233, 2015. PMID 26465950.
49. **Paulo JA, Kadiyala V, Banks PA, Steen H, and Conwell DL.** Mass Spectrometry-Based (GeLC-MS/MS) Comparative Proteomic Analysis of Endoscopically (ePFT) Collected Pancreatic and Gastroduodenal Fluids. *Clinical and Translational Gastroenterology* 3: e14, 2012. PMID: 23238231.
50. **Paulo JA, Kadiyala V, Lee LS, Banks PA, Conwell DL, and Steen H.** Proteomic analysis (GeLC-MS/MS) of ePFT-collected pancreatic fluid in chronic pancreatitis. *Journal of Proteome Research* 11: 1897-1912, 2012. PMID: 22243521.
51. **Pezzilli R, Andriulli A, Bassi C, Balzano G, Cantore M, Delle Fave G, Falconi M, and Exocrine Pancreatic Insufficiency collaborative G.** Exocrine pancreatic insufficiency in adults: a shared position statement of the Italian Association for the Study of the Pancreas. *World Journal of Gastroenterology : WJG* 19: 7930-7946, 2013. PMID: 24307787.
52. **Rana SS, Dambalkar A, Chhabra P, Sharma R, Nada R, Sharma V, Rana S, and Bhasin DK.** Is pancreatic exocrine insufficiency in celiac disease related to structural alterations in pancreatic parenchyma? *Annals of Gastroenterology* 29: 363-366, 2016. PMID: 27366039.
53. **Roy PK, Venzon DJ, Shojamanesh H, Abou-Saif A, Peghini P, Doppman JL, Gibril F, and Jensen RT.** Zollinger-Ellison syndrome. Clinical presentation in 261 patients. *Medicine* 79: 379-411, 2000. PMID: 11144036.
54. **Scheele G, Bartelt D, and Bieger W.** Characterization of human exocrine pancreatic proteins by two-dimensional isoelectric focusing/sodium dodecyl sulfate gel electrophoresis. *Gastroenterology* 80: 461-473, 1981. PMID: 6969677.
55. **Stallings VA, Stark LJ, Robinson KA, Feranchak AP, Quinton H, Clinical Practice Guidelines on G, Nutrition S, and Ad Hoc Working G.** Evidence-based practice recommendations for nutrition-related management of children and adults with cystic fibrosis and pancreatic insufficiency: results of a systematic review. *Journal of the American Dietetic Association* 108: 832-839, 2008. PMID: 18442507.
56. **Stevens T, Dumot JA, Zuccaro G, Jr., Vargo JJ, Parsi MA, Lopez R, Kirchner HL, Purich E, and Conwell DL.** Evaluation of duct-cell and acinar-cell function and endosonographic abnormalities in patients with suspected chronic pancreatitis. *Clin Gastroenterol Hepatol* 7: 114-119, 2009. PMID: 18955165.
57. **Sziegoleit A, Krause E, Klor HU, Kanacher L, and Linder D.** Elastase 1 and chymotrypsin B in pancreatic juice and feces. *Clinical Biochemistry* 22: 85-89, 1989. PMID: 2720968.
58. **Tseng DS, Molenaar IQ, Besselink MG, van Eijck CH, Borel Rinkes IH, and van Santvoort HC.** Pancreatic Exocrine Insufficiency in Patients With Pancreatic or Periampullary Cancer: A Systematic Review. *Pancreas* 45: 325-330, 2016. PMID: 2649577.
59. **Vujasinovic M, Tepes B, Makuc J, Rudolf S, Zaletel J, Vidmar T, Seruga M, and Birsa B.** Pancreatic exocrine insufficiency, diabetes mellitus and serum nutritional markers after acute pancreatitis. *World Journal of Gastroenterology* 20: 18432-18438, 2014. PMID: 25561813.
60. **Vujasinovic M, Tepes B, Volfand J, and Rudolf S.** Exocrine pancreatic insufficiency, MRI of the pancreas and serum nutritional markers in patients with coeliac disease. *Postgraduate Medical Journal* 91: 497-500, 2015. PMID: 26253920.
61. **Williams JA, Korc M, and Dormer RL.** Action of secretagogues on a new preparation of functionally intact, isolated pancreatic acini. *Am J Physiol* 235: 517-524, 1978. PMID: 215042.
62. **Wollaeger EE, Comfort MW, and et al.** Efficiency of gastrointestinal tract after resection of head of pancreas. *JAMA* 137: 838-848, 1948. PMID: 18870005.
63. **Wollaeger EE, Comfort MW, and Osterberg AE.** Total solids, fat and nitrogen in the feces; a study of normal persons taking a test diet containing a moderate amount of fat: comparison with results obtained with normal persons taking a test diet containing a large amount of fat. *Gastroenterology* 9: 272-283, 1947. PMID: 20264640.