

Gene Therapy Coming of Age – Prevention of Acute Pancreatitis in Lipoprotein Lipase Deficiency Through Alipogene Tiparvovec

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Abstract

Lipoprotein lipase deficiency (LPLD) is a rare autosomal recessive disorder caused by loss-of-function mutations in the *LPL* gene. It causes hyperchylomicronaemia and severe hypertriglyceridaemia leading to various clinical manifestations, including attacks of acute pancreatitis, which can cause severe morbidity and even death. Recurrent attacks of acute pancreatitis also put LPLD patients at risk of developing chronic pancreatitis. The psychological and social burden of this disease is substantial. Progressive loss of pancreatic parenchymal tissue leads to exocrine and endocrine insufficiency, each with its own sequelae and associated morbidity and mortality. Concurrent with other causes of hereditary pancreatitis manifesting at young age, LPLD patients most likely have a substantially increased risk of developing pancreatic cancer. Currently, patients with LPLD are instructed to adhere to a diet with severe fat restriction in order to avoid pancreatitis. Such a diet is extremely difficult to maintain, does not abolish hyperchylomicronaemia and hypertriglyceridaemia and does not prevent pancreatitis in all cases. A novel gene therapy approach with the administration of alipogene tiparvovec, containing the human gain-of-function *LPL* gene variant *LPL^{S447X}*, shows promising results with persistent LPL expression and biological activity and reduction in pancreatitis risk.

Keywords

Pancreatitis, lipoprotein lipase (LPL), lipoprotein lipase deficiency (LPLD), gene therapy, alipogene tiparvovec

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This article aims to provide insight into the disease course of pancreatitis in lipoprotein lipase deficiency (LPLD) patients and the burden it puts on these patients, both physically and mentally. As LPLD is such a rare disease and published outcome data are scarce, this is partly accomplished by inference, comparing its disease course with what is known from other causes of (hereditary) pancreatitis.

This article then describes the exciting prospects of recent developments aimed at causally treating LPLD in order to avoid disease symptoms and prevent long-term complications by correcting the underlying defect in lipoprotein lipase (LPL) expression by gene therapy with alipogene tiparvovec.

Acute and Chronic Pancreatitis

Acute pancreatitis is defined as an acute inflammatory process of the pancreas that frequently involves peripancreatic tissues and/or remote organ systems.¹ In 80% of cases the disease course is relatively mild and self-limiting. After a few days of supportive care (nil by mouth, intravenous fluid resuscitation, no total parenteral nutrition [TPN] in cases of LPLD and pain relief), the patient can be discharged from hospital. However, in about 20% of patients a rapidly progressive fulminant illness develops with pancreatic necrosis.²

Development of a systemic inflammatory response syndrome, multi-organ failure (in particular pulmonary insufficiency, renal failure and shock) and septic complications due to infected necrosis requires admission to the intensive care unit.

The overall mortality of acute pancreatitis is approximately 5%. Mortality is high in patients with necrotising pancreatitis (overall 17%) and increases to 30% in those with infected necrosis.³ The incidence of acute pancreatitis varies from 10 to 44 per 100,000, probably reflecting differences in risk factor prevalence, and is rising in western countries.^{4,5} Men are more frequently affected than women and pancreatitis incidence increases with age, with a peak in the sixth decade of life.

The two most common aetiological factors of acute pancreatitis are gallstones (including microlithiasis) and alcohol abuse, which account for more than 80% of cases.⁴ Other causes of acute pancreatitis include:⁶

- structural abnormalities such as pancreas divisum;
- neoplasms;
- metabolic disorders (hypertriglyceridaemia, hypercalcaemia);
- drugs;

- trauma;
- iatrogenic causes (e.g. post-endoscopic retrograde cholangio-pancreatography pancreatitis);
- infections;
- vascular disorders (ischaemia);
- genetic causes (e.g. trypsinogen mutations); and
- a remaining group that cannot be classified, referred to as idiopathic.

Eleven to 27% of patients who suffer from a first attack of acute pancreatitis develop recurrent attacks.⁷⁻⁹ This is dependent on the underlying pathophysiological mechanism responsible for the pancreatic inflammation and the possibility of treatment (biliary stones) or abstaining from its causative agent (alcohol or medication). Evidently, continued alcohol abuse is an important cause of recurrent pancreatitis.⁸

Chronic pancreatitis is characterised by (recurrent episodes of) abdominal pain with ongoing destruction of functional pancreatic parenchyma and progressive fibrosis accompanied by the development of pancreatic exocrine and endocrine insufficiency. Hospitalisation is required in the case of an exacerbation to control pain (e.g. opioid medication, pancreaticojejunostomy) and for the treatment of complications such as pseudocysts. The incidence of chronic pancreatitis in Europe is approximately six to seven per 100,000 population.^{9,10} Besides chronic pain, other medical problems include pseudocyst formation, benign biliary obstruction due to fibrotic stricturing of the intrapancreatic portion of the distal common bile duct and vascular events such as splenic and/or portal vein thrombosis. Exocrine and endocrine insufficiency are late complications of chronic pancreatitis, usually occurring years after the initial diagnosis.

The currently prevailing theory that explains how chronic pancreatitis develops is the necrosis–fibrosis sequence, which starts off with a so-called ‘sentinel’ acute pancreatitis event.¹¹ Whether chronic pancreatitis eventually develops is closely related to the number and severity of acute attacks.¹² In one report, 10.9% of patients with a first attack of acute pancreatitis developed one or more attacks within five years, with 6.4% of patients progressing to chronic pancreatitis.⁷ Another study reported a cumulative incidence of chronic pancreatitis of 13% at 10 years and 16% at 20 years.¹³ After surviving a second attack, the incidence of chronic pancreatitis increased to 38% after only two years of follow-up. These data show that patients who suffer from repeated acute pancreatitis attacks, such as LPLD patients, are likely to develop chronic pancreatitis.

Health Issues and Socioeconomic Impact of Pancreatitis

There are no reliable data on the disease burden of patients who have had an attack of severe pancreatitis or those suffering from recurrent attacks. Obviously, the prospect of facing the risk of developing a new painful attack and not knowing if, when and how severe this will be scares patients, affecting their lives both socially and professionally. Frequent absence from work for indefinite periods of time puts stress on the patient’s relationship with their employer. Not being regarded as a reliable co-worker by colleagues is disappointing and frustrating. Patients with established chronic pancreatitis have an impaired quality of life compared with age- and gender-adjusted control populations with significantly lower scores of physical functioning, psychological status and social wellbeing.^{14,15} Patients suffering from

chronic pancreatitis also have a significantly higher degree of financial difficulties, disease-related absence from work or unemployment compared with controls.¹⁵⁻¹⁸

Exocrine pancreatic insufficiency, which is likely to develop after a few years, causes steatorrhea, abdominal complaints and weight loss. To maintain a healthy nutritional status, patients are required to use lifelong pancreatic enzyme supplementation with each meal.¹⁹ Endocrine pancreatic insufficiency puts patients at risk of all the well-known complications associated with diabetes, such as nephropathy, retinopathy, cardiovascular disease and neuropathy.

Lastly, patients with chronic pancreatitis have an increased risk of developing pancreatic cancer compared with the normal population (relative risk 13.3, 95% confidence interval [CI] 6.1–28.9).²⁰ This is explained by the cumulative effect of ongoing chronic inflammation with increasing DNA damage and progressive development of pre-cancerous precursor lesions such as pancreatic intraepithelial neoplasms (PanINs). The longer inflammation is present, obviously related to the age at which inflammation starts, the higher the risk.²¹ This is demonstrated by a classic genetic condition, such as hereditary pancreatitis (*PRSS1* mutation), in which patients develop recurrent attacks of acute pancreatitis at a very early age (median age 10 years), soon progressing into chronic pancreatitis.²² In these patients, the risk of developing pancreatic cancer is increased 69-fold (95% CI 56.4–84.4) compared with the normal population.²⁰

Pancreatitis Due to Lipoprotein Lipase Deficiency Epidemiology, Clinical Manifestation and Conventional Management

LPLD is a rare autosomal recessive disorder caused by loss-of-function mutations in the *LPL* gene. Its prevalence is estimated at one to two per million. It is the most common genetic cause of hyperchylomicronaemia, a condition in which triglycerides are not offloaded from chylomicrons because of dysfunctional *LPL*, causing continuous and excessively high levels of plasma chylomicrons and severe hypertriglyceridaemia. This can be seen when looking at a blood sample with one’s bare eyes because the plasma appears lactescent and turbid.

LPLD usually manifests during infancy or childhood, with episodes of severe pain and failure to thrive.²³ LPLD has various clinical manifestations including lipaemia, hepatosplenomegaly, lipaemia retinalis, recurrent acute pancreatitis, (premature) atherosclerosis and eruptive xanthomas.²⁴ The latter are transiently present in 50% of patients and appear as deposits of lipid in the skin resulting from the extravascular phagocytosis of chylomicrons by macrophages.²³

Psychoneurological manifestations are also frequently seen, including paresthesias of the hands, peripheral neuropathies and a reversible form of dementia manifesting as memory loss.²⁵ Twenty-five per cent of diagnosed individuals develop symptoms before reaching one year of age and the majority develop symptoms before 10 years of age.²⁶

The severity of symptoms correlates with the degree of chylomicronaemia, which varies with dietary fat intake. As for management of LPLD, there is currently no effective drug therapy available.²⁷

Acute pancreatitis is a frequent and feared complication of LPLD.²⁶ Attacks are often recurrent and may develop into chronic pancreatitis, with the possible development of exo- and endocrine insufficiency.²⁸ Hyperchylomicronaemia-induced pancreatitis rarely occurs unless triglyceride levels exceed 10–20mmol/l.^{29,30} This relationship seems less clear the other way around: there are patients with triglyceride concentrations much higher than 10–20mmol/l who for many years do not develop overt pancreatitis (personal communication).

The only means by which patients can limit pancreatitis attacks is to lower chylomicronaemia by strict adherence to a diet severely restricted in (non-medium-chain triglyceride [MCT]) fat content. Although dietary restriction with very low total fat content (as low as 10–20g/day) positively affects pancreatitis incidence, it does not fully correct lipid abnormalities or prevent pancreatitis in all cases. Moreover, a very-low-fat diet is extremely unpalatable, which makes it very difficult to adhere to, particularly for younger patients. Fat is also an important calorie source, as it is twice as dense in energy as proteins or carbohydrates. By severely restricting fat intake without compensating for calorie losses by means of an energy-enriched diet, children may fail to thrive and adults may become malnourished.

Currently available drugs including fibrates are not able to lower triglyceride levels or alter the disease course.³¹ Acute plasmapheresis has been utilised to remove chylomicrons from the blood in acute situations.³² However, this is not a preventative therapy and it does not prevent inflammatory damage from occurring. Chronic plasmapheresis has been proposed as a prophylactic therapy in patients with recurrent pancreatitis due to severe primary hypertriglyceridaemia that is unresponsive to other measures.³³ Even though such therapy would abolish or reduce the number of acute pancreatitis attacks, lifelong chronic plasmapheresis every four weeks is expensive and is likely to negatively affect quality of life in a similar way to chronic haemodialysis.³⁴

The clinical manifestations of acute pancreatitis caused by the hyperchylomicronaemia syndrome are largely indistinguishable from those in acute pancreatitis of other causes.²⁷ Importantly, one should realise that in patients with LPLD, triglyceride levels in plasma can be misleadingly normal or only moderately elevated when there is a delay in presentation and patients have ceased their food intake to cope with the pain and nausea.³⁵ This may prevent the correct diagnosis being made. It has been reported that pancreatic calcifications do not appear to be common.²⁷

Pathophysiology of Pancreatitis Associated with Lipoprotein Lipase Deficiency

The exact pathophysiological mechanism that explains how hyperchylomicronaemia causes pancreatitis is unclear. A plausible hypothesis is that the largest of the chylomicrons may aggregate and impair blood flow in the pancreatic capillaries, resulting in ischaemia disrupting acinar cells, causing exposure of triglycerides to pancreatic lipase.^{36,37} The free fatty acids that result from the enzymatic degradation of triglycerides could then lead to further damage of acinar cells, reinforced by the release of inflammatory mediators and free radicals.^{38,39} Indeed, isolated pancreatic perfusion of triglycerides in an *ex vivo* animal model caused oedema, haemorrhage and elevated amylase levels. This was attenuated when free fatty acids were added to the perfusate.⁴⁰ In rats, inducing hypertriglyceridaemia in various pancreatitis models by repeated

subcutaneous injections of cerulean and retrograde duct injection of sodium taurocholate caused a marked dose-dependent elevation of pancreatic enzymes.⁴¹

Complications of Pancreatitis in Lipoprotein Lipase Deficiency Patients

With regard to the acute and chronic complications of pancreatitis in LPLD patients, there is no apparent reason to assume that these are any different from or less severe than in other types of pancreatitis. There is one important exception: attacks start at a much younger age compared with non-hereditary forms of pancreatitis. It therefore seems only logical to compare the probable disease course of LPLD pancreatitis with hereditary pancreatitis that is caused by a mutation in the trypsinogen gene (*PRSS1*).²¹

For example, in a large series of patients with hereditary pancreatitis in France, the prevalence of exocrine pancreatic insufficiency was 34% (median age at occurrence 29 years) and of diabetes was 26% (median age at occurrence 38 years).²² In another report from the same research group, the cumulative risk of developing pancreatic cancer at 50 and 75 years of age was, respectively, 11 and 49% for men and 8 and 55% for women.⁴² Apart from inflammation, smoking and diabetes were the main associated risk factors. There was no correlation with the type of *PRSS1* mutation.

Alipogene Tiparvec to Prevent Pancreatitis in Lipoprotein Lipase Deficiency

As pointed out above, the medical and (psycho)social consequences of acute and chronic pancreatitis are substantial. The ultimate goal when treating patients suffering from LPLD is to prevent pancreatitis and avoid short- and long-term complications. With this intention, a novel gene therapy by means of alipogene tiparvec has been developed. Alipogene tiparvec (Glybera; AMT-011; AAV1–*LPL*^{S447X}) contains the human *LPL* gene variant *LPL*^{S447X}, a gain-of-function variant found in 20% of Caucasians. It is associated with enhanced removal of proatherogenic apoB100-containing particles including low-density lipoprotein (LDL) cholesterol,⁴³ lower plasma triglyceride levels, higher high-density lipoprotein (HDL) cholesterol concentrations and lower rates of cardiovascular disease compared with the general population.^{43,44}

The therapeutic gene in alipogene tiparvec – *LPL*^{S447X} – is delivered by a non-replicating and non-integrating vector that comprises a protein shell derived from adeno-associated virus serotype 1 (AAV1), the cytomegalovirus (CMV) promoter, a woodchuck hepatitis virus post-transcriptional regulatory element and AAV2-derived inverted terminal repeats.⁴⁵ Alipogene tiparvec is produced using insect cells and recombinant baculovirus technology. It is formulated in a solution for intramuscular injection.

Clinical Development Programme

After proof-of-concept and pre-clinical safety studies had been conducted⁴³ and *ex vivo* expression of *LPL* had been demonstrated in the muscle cells of LPLD subjects,⁴² a clinical development programme was undertaken for alipogene tiparvec.

Human phase I/II and II/III clinical trials in which AAV1–*LPL*^{S447X} was administered by a single series of intramuscular injections have been performed in The Netherlands and Canada. Three open, non-controlled intervention studies have been completed, designated

Table 1: Summary of Clinical Studies with Alipogene Tiparvec

Study	Phase	Number of Patients	Duration of Monitoring*	Duration of Follow-up [†]	Status
PREP-01	Observational	18	41 (13–78) weeks	–	Completed
CT-AMT-010-01	Phase I/II	8	12 weeks; reported ⁴³	5 years	Active phase completed, follow-up ongoing between 4 and 5 years
PREP-02	Observational	22	32 (2–83) weeks	–	Completed
CT-AMT-011-01	Phase II/III	14	12 weeks	5 years	Active phase completed, follow-up ongoing between 2 and 3 years
CT-AMT-011-02 (EXT)	Phase II/III	5	14 weeks	1 year	Ongoing

*Average with minimal and maximal duration for the PREP studies. [†]After completion, patients will be included in the lipoprotein lipase deficiency registry.

the study numbers 010-01,⁴⁶ 011-01 and 011-02. The latter two studies have not yet been reported as full papers, but preliminary data are available.^{47,48} The first two studies were preceded by prospective observational studies; the third had a run-in period.

In all studies safety was a key objective, including vector biodistribution and shedding and immune response to vector capsid and transgene (LPL protein). Efficacy objectives included whole-plasma triglyceride lowering. In addition, various lipid/lipoprotein parameters and clinical symptoms (including pancreatitis) were followed. Muscle biopsies, local vector presence, gene expression and biologic activity were measured.

During all studies, a fat-restricted diet was employed in which fat content did not exceed 20% of the total daily calorie intake and this percentage contained no more than 55g/day. Patients received regular counselling by a dietician during the pre-study prospective observational phase (PREP) and main studies. Only LPLD subjects with severe hypertriglyceridaemia, <20% of normal post-heparin plasma LPL activity, homozygous or compound heterozygosity for deleterious LPL mutations and a history of pancreatitis were eligible for the intervention studies. A short immunosuppressant regimen was added in the second and third intervention studies to attempt to limit T-cell responses to AAV1 capsid (seen in half of the subjects in the first study). At the time of the second study initiation, such regimens were suspected to potentially attenuate the efficacy of AAV1–LPL^{S447X} gene therapy. Further follow-up did not provide evidence for neutralising effects. A further improved regimen was concomitantly administered in the third study in order to reduce any potential immune response to the vector or the construct that might limit future re-administration.

Tables 1 and 2 provide a summary of all studies and their designs.

Conducted in The Netherlands, CT-AMT-010-01 was the first *in vivo* human experiment of AAV1–LPL^{S447X}, with 12 weeks of observation in the main study post-dosing.⁴⁶ A total of eight LPLD subjects were administered AMT-010, the predecessor of alipogene tiparvec. This contained the same LPL^{S447} gene construct in the same vector, but was produced in a mammalian instead of an insect cell-based production system.

AMT-010 was well tolerated at both doses and neither drug-related serious adverse events nor dose-limiting toxicity were noted. In 25% of subjects in the lower-dose group and in 50% of the higher-dose group, triglyceride levels fell below the target level of 10mmol/l or were reduced by >40%, both of which were pre-defined efficacy end-points.

Table 2: Number of Patients on Dosing Regimens in the Alipogene Tiparvec Clinical Development Programme

AAV1–LPL ^{S447X} Dose (gc/kg bodyweight)	1x10 ¹¹	3x10 ¹¹	3x10 ¹¹	1x10 ¹²	1x10 ¹²
Immunosuppressants*			+ A	+ A	+ B
Study					
CT-AMT-010-01	4	4	–	–	–
CT-AMT-011-01	–	2	4	8	–
CT-AMT-011-02	–	–	–	–	5

*Immunosuppressant regimens: A: Cyclosporine oral dose of 3mg/kg/day + 2x1g/day of mycophenolate mofetil orally, starting one day after alipogene tiparvec, for 12 weeks; B: Cyclosporine oral dose of 3mg/kg/day + 2x1g/day of mycophenolate mofetil orally, both starting three days before alipogene tiparvec, for 12 weeks + bolus methyl-prednisolone 1mg/kg IV pre-alipogene tiparvec.

In a recent conference publication, the investigators report the long-term follow-up results (two to three years) of this study.⁴⁹ The pancreatitis incidence decreased despite an increase in triglycerides over time. Vector DNA and LPL^{S447X} expression was present in (injected) muscle half a year after one-time AMT-010 administration. No safety issues with AMT-010 were noted during this follow-up. The authors concluded that a single dose of AMT-010 was safe and effective to prevent recurrent pancreatitis, most likely through continued expression of LPL^{S447X}.

In a more recent communication focusing on the safety of AMT-010 after three to four years of follow-up, the investigators report a *post hoc* analysis of pancreatitis events. They compared the incidence with PREP-01 prospective observation data. Data from these patients (still) indicate a clinically relevant decline in pancreatitis frequency.⁵⁰

The second intervention study (CT-AMT-011-01) was conducted in Quebec and was based on the results of the Dutch study. Here it was decided to study a further dose increase compared with the highest dose employed in CT-AMT-010-01. The investigators confirmed the inability of the controlled diet to lower triglycerides and prevent pancreatitis according to observations made during PREP-02.⁵¹ Alipogene tiparvec was well tolerated, without dose-limiting toxicity. Thus far, during long-term follow-up this therapy has had an excellent safety profile.

During the 2009 meeting of the International Society for Atherosclerosis, the investigators presented results of the one-year follow-up.⁵² An important reduction (>40%) in fasting triglyceride levels was observed in the majority of subjects beginning two weeks post-injection and was maintained for the duration of the main study

(12 weeks). From week two after alipogene tiparvec injection to the end of year one, no pancreatitis episode had been reported and several clinical features of LPLD had significantly improved. Approximately four months post-injection, total fasting plasma triglycerides tended to drift back towards pre-treatment values. Despite this, the increase was associated with important changes in the characteristics of lipoproteins and reduction of triglycerides in the chylomicron stratum. The authors concluded that their data suggest that gene therapy with a single dose of alipogene tiparvec is associated with significant clinical improvement for at least one year after the dose is given.

Up to two-year follow-up was recently reported. Pancreatitis incidence was still reduced in this study. Reversal to baseline hypertriglyceridaemia after transient reduction of plasma levels was confirmed, but reduced chylomicronaemia did not similarly revert.⁵³

A third open-label clinical trial, the CT-AMT-011-02 study, was started in two centres in Quebec to collect more information on the mechanism of action of alipogene tiparvec and to capture additional efficacy and safety data. In this study's protocol, 24-hour post-prandial testing was repeated and lipid and lipoprotein distribution kinetic assessment was included. Short-term follow-up results in this study are reported to be similar to those observed in the previous study.⁵³ The results of the post-prandial and other lipid lipoprotein analyses were also recently reported.⁵⁴

During the run-in phase of the study, prior to one-time intramuscular alipogene tiparvec administration, post-prandial handling of chylomicrons in LPLD patients was grossly abnormal and hugely different from normal post-prandial chylomicron clearance in control subjects. Total plasma tracer (for chylomicrons produced *de novo* after a standard test meal) accumulated over the first nine hours following dietary intake. There was no or only a moderate decline in the subsequent period up to 24 hours. The majority of the tracer was found in the chylomicron fraction. This hampered clearance of tracer from plasma and chylomicron fraction is in line with the absence of LPL activity and chylomicronaemia.

Fourteen weeks after alipogene tiparvec administration, a lower peak level of the tracer was observed in total plasma. Importantly, the amount of tracer recovered from the chylomicron fraction was greatly reduced, time to peak was reduced and in each case the tracer was completely eliminated over the 24-hour period following the meal, indicating improved chylomicron clearance (in all five patients). The authors concluded that a one-time administration of alipogene tiparvec results in enhanced post-prandial clearance of chylomicron, which may well explain the long-term reduction observed in the incidence of pancreatitis.

Summary

LPLD is a rare genetic disorder caused by loss-of-function mutations in the *LPL* gene. This leads to severe hyperchylomicronaemia and hypertriglyceridaemia, which causes patients to suffer from (recurrent) attacks of acute pancreatitis and puts them at risk of developing acute and chronic complications.

The development of alipogene tiparvec aims to offer LPLD patients the prospect of controlling the symptoms of hyperchylomicronaemia, including those associated with pancreatitis, and preventing long-term complications. Interventional studies have consistently shown that alipogene tiparvec is well tolerated without dose-limiting toxicity. A decrease in triglyceride levels for three months post-dosing were noted in virtually all patients. In tested patients, a persistent decrease in chylomicronaemia was seen, with markedly improved metabolism of post-prandial chylomicron fraction. Muscle biopsies taken half a year post-dosing show persistent gene expression and biologic activity.

At short- and longer-term follow-up, prevention or reduction of pancreatitis attacks is noticed and the picture starts to emerge that this effect is persistent over time. Continued follow-up for longer periods of time is essential to document the natural course of this disease after therapy with alipogene tiparvec, including the monitoring of continued gene expression and biologic activity.

When alipogene tiparvec gene therapy should be administered needs to be determined: should it be given to all LPLD patients or only to those who have suffered at least one acute pancreatitis attack? Beyond the scope of gastroenterology, but of great importance and interest, is the long-term impact of alipogene tiparvec on other complications of LPLD, including cardiovascular events. Future studies are needed to determine the long-term impact of this therapy. ■



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- Bradley EL, A clinically based classification system for acute pancreatitis. Summary of the International Symposium on Acute Pancreatitis, Atlanta, Ga, September 11 through 13, 1992, *Arch Surg*, 1993;128(5): 586–90.
- Whitcomb DC, Clinical practice. Acute pancreatitis, *N Engl J Med*, 2006;354:2142–50.
- Banks PA, Freeman ML, Practice Parameters Committee of the American College of Gastroenterology, Practice guidelines in acute pancreatitis, *Am J Gastroenterol*, 2006;101:2379–2400.
- Spanier BW, Dijkgraaf MG, Bruno MJ, Epidemiology, aetiology and outcome of acute and chronic pancreatitis: An update, *Best Pract Res Clin Gastroenterol*, 2008;22:45–63.
- Yadav D, Lowenfels AB, Trends in the epidemiology of the first attack of acute pancreatitis: a systematic review, *Pancreas*, 2006;33:323–30.
- Pandolfi SJ, Saluja AK, Imrie CW, et al., Acute pancreatitis: bench to the bedside, *Gastroenterology*, 2007;132:1127–51.
- Eland IA, Sturkenboom MJ, Wilson JH, et al., Incidence and mortality of acute pancreatitis between 1985 and 1995, *Scand J Gastroenterol*, 2000;35:1110–16.
- Gullo L, Migliori M, Pezzilli R, et al., An update on recurrent acute pancreatitis: data from five European countries, *Am J Gastroenterol*, 2002;97:1959–62.
- Lankisch PG, Assmus C, Maisonneuve P, et al., Epidemiology of pancreatic diseases in Luneburg County. A study in a defined German population, *Pancreatology*, 2002;2:469–77.
- Lévy P, Barthet M, Mollard BR, et al., Estimation of the prevalence and incidence of chronic pancreatitis and its complications, *Gastroenterol Clin Biol*, 2006;30:838–44.
- Schneider A, Whitcomb DC, Hereditary pancreatitis: a model for inflammatory diseases of the pancreas, *Best Pract Res Clin Gastroenterol*, 2002;16:347–63.
- Ammann RW, Muellhaupt B, Progression of alcoholic acute to chronic pancreatitis, *Gut*, 1994;35:552–6.
- Lankisch PG, Breuer N, Bruns A, et al., Natural history of acute pancreatitis: a long-term population-based study,

- Am J Gastroenterol*, 2009;104:2797–2805.
14. Fitzsimmons D, Kahl S, Butturini G, et al., Symptoms and quality of life in chronic pancreatitis assessed by structured interview and the EORTC QLQ-C30 and QLQ-PAN26, *Am J Gastroenterol*, 2005;100:918–26.
 15. Wehler M, Nichterlein R, Fischer B, et al., Factors associated with health-related quality of life in chronic pancreatitis, *Am J Gastroenterol*, 2004;99:138–46.
 16. Gastard J, Joubaud F, Farbos T, et al., Etiology and course of primary chronic pancreatitis in Western France, *Digestion*, 1973;9:416–28.
 17. Pezzilli R, Morselli-Labate AM, Fantini L, et al., Assessment of the quality of life in chronic pancreatitis using Sf-12 and EORTC QLQ-C30 questionnaires, *Dig Liver Dis*, 2007;39:1077–86.
 18. Lankisch PG, Löhr-Happe A, Otto J, et al., Natural course in chronic pancreatitis. Pain, exocrine and endocrine pancreatic insufficiency and prognosis of the disease, *Digestion*, 1993;54:148–55.
 19. Sikkens EC, Cahen DL, Kuipers EJ, et al., Pancreatic enzyme replacement therapy in chronic pancreatitis, *Best Pract Res Clin Gastroenterol*, 2010;24:337–47.
 20. Raimondi S, Lowenfels AB, Morselli-Labate AM, et al., Pancreatic cancer in chronic pancreatitis; aetiology, incidence, and early detection, *Best Pract Res Clin Gastroenterol*, 2010;24:349–58.
 21. Greer JB, Whitcomb DC, Inflammation and pancreatic cancer: an evidence-based review, *Curr Opin Pharmacol*, 2009;9:411–18.
 22. Rebours V, Boutron-Ruault MC, Schnee M, et al., The natural history of hereditary pancreatitis: a national series, *Gut*, 2009;58:97–103.
 23. Brunzell JD, Deeb SS, Familial lipoprotein lipase deficiency, Apo C-II deficiency and hepatic lipase deficiency, In: Scriver CR, Beaudet AL, Sly WS, et al., eds., *The Metabolic Basis of Inherited Disease*, New York, McGraw-Hill, 2001:2789–2816.
 24. Benlian P, De Gennes JL, Foubert L, et al., Premature atherosclerosis in patients with familial chylomicronemia caused by mutations in the lipoprotein lipase gene, *N Engl J Med*, 1996;335:848–54.
 25. Mas JL, Bousser MG, Lacombe C, et al., Hyperlipidemic dementia, *Neurology*, 1985;35:1385–87.
 26. Kawashiri MA, Higashikata T, Mizuno M, et al., Long-term course of lipoprotein lipase (LPL) deficiency due to homozygous LPL(Arita) in a patient with recurrent pancreatitis, retained glucose tolerance, and atherosclerosis, *J Clin Endocrinol Metab*, 2005;90:6541–4.
 27. Chait A, Brunzell JD, Chylomicronemia syndrome, *Adv Intern Med*, 1992;37:249–73.
 28. Truninger K, Schmid PA, Hoffmann MM, et al., Recurrent acute and chronic pancreatitis in two brothers with familial chylomicronemia syndrome, *Pancreas*, 2006;32:215–19.
 29. Lithell H, Vessby B, Walldius G, et al., Hypertriglyceridemia—acute pancreatitis—ischemic heart disease. A case study in a pair of monozygotic twins, *Acta Med Scand*, 1987;221:311–16.
 30. Fortson MR, Freedman SN, Webster PD 3rd, Clinical assessment of hyperlipidemic pancreatitis, *Am J Gastroenterol*, 1995;90:2134–9.
 31. Paradis G, *Effect of a Nutritional Supplement Rich Containing Medium-chain Triglyceride (MCT) and the Adjunction of Fibrates on Lipid-lipoprotein Profile in Humans with Homozygous LPL Deficiency*, Quebec, Université Laval, 2001.
 32. Routy JP, Smith GH, Blank DW, et al., Plasmapheresis in the treatment of an acute pancreatitis due to protease inhibitor-induced hypertriglyceridemia, *J Clin Apher*, 2001;16:157–9.
 33. Piolat A, Nadler F, Cavallero E, et al., Prevention of recurrent acute pancreatitis in patients with severe hypertriglyceridemia: value of regular plasmapheresis, *Pancreas*, 1996;13:96–9.
 34. Kimmel PL, Patel SS, Quality of life in patients with chronic kidney disease: focus on end-stage renal disease treated with hemodialysis, *Semin Nephrol*, 2006;26:68–79.
 35. Domínguez-Muñoz JE, Malfertheiner P, Ditschuneit HH, et al., Hyperlipidemia in acute pancreatitis. Relationship with etiology, onset, and severity of the disease, *Int J Pancreatol*, 1991;10:261–7.
 36. Uchida T, Tsuchiya R, Harada N, et al., Ischemic changes in the pancreas of Watanabe heritable hyperlipidemic (WHHL) rabbits, *Int J Pancreatol*, 1988;3:261–71.
 37. Maeda N, Cicha I, Tateishi N, et al., Triglyceride in plasma: prospective effects on microcirculatory functions, *Clin Hemorheol Microcirc*, 2006;34:341–6.
 38. Havel RJ, Pathogenesis, differentiation and management of hypertriglyceridemia, *Adv Intern Med*, 1969;15:117–54.
 39. Nordstoga K, Christophersen B, Ytrehus B, et al., Pancreatitis associated with hyperlipoproteinaemia type I in mink (*Mustela vison*): earliest detectable changes occur in mitochondria of exocrine cells, *J Comp Pathol*, 2006;134:320–28.
 40. Saharia P, Margolis S, Zuidema GD, et al., Acute pancreatitis with hyperlipemia: studies with an isolated perfused canine pancreas, *Surgery*, 1977;82:60–67.
 41. Kimura W, Mössner J, Role of hypertriglyceridemia in the pathogenesis of experimental acute pancreatitis in rats, *Int J Pancreatol*, 1996;20:177–84.
 42. 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;103:111–19.
 43. Rip J, van Dijk KW, Sierts JA, et al., AAV1-LPL(S447X) gene therapy reduces hypertriglyceridemia in apoE2 knock in mice, *Biochim Biophys Acta*, 2006;1761:1163–8.
 44. Wittrup HH, Tybjaerg-Hansen A, Nordestgaard BG, Lipoprotein lipase mutations, plasma lipids and lipoproteins, and risk of ischemic heart disease. A meta-analysis, *Circulation*, 1999;99:2901–7.
 45. Rip J, Nierman MC, Sierts JA, et al., Gene therapy for lipoprotein lipase deficiency: working toward clinical application, *Hum Gene Ther*, 2005;16:1276–86.
 46. Stroes ES, Nierman MC, Meulenberg JJ, et al., Intramuscular administration of AAV1-lipoprotein lipase S447X lowers triglycerides in lipoprotein lipase-deficient patients, *Arterioscler Thromb Vasc Biol*, 2008;28:2303–4.
 47. Safety and Efficacy in LPL-Deficient Subjects of AMT-011, an Adeno-Associated Viral Vector Expressing Human Lipoprotein Lipase [S447X], US National Institutes of Health, ClinicalTrials.gov, 2010. Available at: clinicaltrials.gov/ct2/show/NCT01109498?term=LPLD&rank=1 (accessed 22 June 2010).
 48. Efficacy and Safety of Human Lipoprotein Lipase (LPL)[S447X] Expressed by an Adeno-Associated Viral Vector in LPL-deficient Subjects, US National Institutes of Health, ClinicalTrials.gov, 2009. Available at: clinicaltrials.gov/ct2/show/NCT00891306?term=LPLD&rank=2 (accessed 22 June 2010).
 49. Stroes E, Kuivenhoven J, van Deventer S, et al., Safety and efficacy of AMT-010 gene therapy in lipoprotein lipase deficiency (LPLD), *Atheroscler Suppl*, 2009;10:e786.
 50. Stroes ES, Sonnemans M, van den Bulk N, et al., Patient and environmental safety of AMT-010 gene therapy for up to 4 years in lipoprotein lipase deficiency (LPLD), *Atheroscler Suppl*, 2010;11:75.
 51. Gaudet D, de Wal J, Tremblay K, et al., Review of the clinical development of alipogene tiparovec gene therapy for lipoprotein lipase deficiency, *Atheroscler Suppl*, 2010;11:55–60.
 52. Gaudet D, Brisson D, Methot J, van Deventer S, An open-label, dose escalation study to assess the Safety and efficacy of AAV1-LPLS447X gene therapy with alipogene tiparovec (AMT-011) for patients with severe hypertriglyceridemia due to lipoprotein lipase Deficiency (LPLD), *Atheroscler Suppl*, 2009;10(2):e286.
 53. Gaudet D, Methot J, Brisson D, et al., Alipogene tiparovec may prevent pancreatitis from chylomicronaemia in lipoprotein lipase deficiency (LPLD), *Atheroscler Suppl*, 2010;11:73–4.
 54. Gaudet D, Frisch F, Methot J, et al., Gene therapy with Glybera® results in enhanced post-prandial clearance of chylomicrons in LPLD patients, *Atheroscler Suppl*, 2010;11:74.