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Orthotopic liver transplantation in an adult with cholesterol ester storage disease

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Abstract

Cholesterol ester storage disease (CESD) is a rare autosomal recessive lipid storage disorder associated with mutations of the gene encoding lysosomal acid lipase, manifestations of which include chronic liver disease and early atherosclerosis. Although normally presenting in childhood, severity is variable and the condition can occasionally remain undetected until middle age. Typical presentation is with asymptomatic hepatosplenomegaly and hyperlipidemia, and the condition is probably underdiagnosed. Treatment is supportive and may include attention to cardiovascular risk factors. Phase I/II trials of enzyme replacement therapy are ongoing, but this approach remains experimental. We present the case of a 42 year-old woman diagnosed with CESD in childhood who ran an indolent course until re-presentation with cirrhotic hydrothorax. She underwent orthotopic liver transplantation but required re-transplantation for hepatic artery thrombosis. She remains well with excellent graft function two years later. Although atherosclerosis was apparent at assessment, and may have contributed to hepatic artery thrombosis, partial correction of the metabolic defect and restoration of liver function by transplantation together with ongoing medical therapy should permit reasonable survival over the longer term from both a liver and vascular perspective. This is the first reported case of orthotopic liver transplantation for CESD in an adult, which was the only available option to improve survival. The case highlights the importance of monitoring patients with CESD through adulthood and suggests that liver replacement at a later stage may yet be indicated and remains of benefit.

Synopsis

Regular follow up of liver function in adults with cholesterol ester storage disease, an uncommon lipid storage disorder, is an important consideration as orthotopic liver transplantation may be required to maintain long term survival.

Introduction

Cholesterol ester storage disease (CESD) is a rare autosomal recessive lipid storage disorder caused by lysosomal acid lipase (LAL) deficiency; this results in chronic liver disease. LAL is responsible for lysosomal cleavage of ester bonds in a range of lipids, including cholesterol esters and triglycerides. Thus, lack of LAL activity leads to intra-lysosomal accumulation of lipid (Anderson et al 1993). In humans, LAL is encoded by the LIPA gene located on chromosome 10. Complete loss-of-function mutations in LIPA lead to development of Wolman's disease; this presents in infancy with failure to thrive and is usually fatal before the age of six months (Anderson and Sando 1991). CESD arises from hypomorphic mutations in the LIPA gene, which impair either expression or function of LAL. The majority of patients described to date possess a homozygous G to A substitution at location -1 of the splice donor site of exon 8 of LIPA (c.894G>A), which causes deletion of exon 8 and production of only 2-4% normal enzyme (Muntoni et al 2007). Heterozygote carrier frequencies have been estimated at 1 in 200 in northern Europe. However, genotypic heterogeneity is evident from reported compound heterozygotes with CESD (Muntoni et al 2007; Pisciotta et al 2009).

CESD usually presents in childhood or adolescence with hypercholesterolemia or hepatomegaly due to hepatic steatosis that eventually progresses to cirrhosis (Chatrath et al 2009). Patients may succumb to complications relating to accelerated atherosclerosis or chronic liver disease, including hepatocellular carcinoma. Historically most patients known to have CESD died before the age of 30 years (Leone et al 1991; McCoy and Yokoyama 1991). However, it is increasingly

recognised that milder forms of CESD can occur raising questions regarding the true natural history of this disorder.

Case history

A 42 year-old woman with a known history of CESD presented with a one-week history of progressive dyspnoea following administration of the H1N1 influenza vaccination. This was accompanied by more gradual abdominal distension. The patient had originally come to medical attention at the age of 6 years with hepatosplenomegaly, whereby CESD was diagnosed following liver biopsy. She defaulted from follow-up and remained well into adulthood. At the age of 39 years, she sought further specialist review during pregnancy. Leukocyte acid esterase (i.e. leukocyte LAL) activity was low at 93 μmol/g/h (350–2000) with an elevated plasma chitotriosidase activity of 222 μmol/L/h (4–120), reaffirming the diagnosis. Lipid profile after overnight fasting demonstrated cholesterol 5.7 mmol/L, triglyceride 1.4 mmol/L, HDL-C 0.93 mmol/L and LDL-C 4.17 mmol/L (Table 1). Pregnancy was uneventful and statin therapy was commenced post-partum. She defaulted from specialist follow-up, though continued statin therapy.

No additional risk factors for chronic liver disease could be elicited at time of admission. Examination and chest radiography demonstrated the presence of a moderate right pleural effusion. Laboratory values were as follows: serum bilirubin 45 μmol/L (< 17), prothrombin time (PT) 16.2 seconds (9.8–12.6), serum creatinine 57 μmol/L (35–125), serum sodium 138 mmol/L (135–145), urine sodium <5 mmol/L.

Her Model of End-stage Liver Disease score was 14 (Kamath et al., 2001), and United Kingdom End-stage Liver Disease (UKELD) score 50.1 (Lewsey et al., 2006).

Pleural aspiration revealed a transudative effusion consistent with cirrhotic hydrothorax. Abdominal ultrasonography (US) demonstrated an enlarged nodular liver with patent portal and hepatic veins, gross ascites and 22 cm splenomegaly. Diuretics did not eliminate the requirement for regular thoracocentesis and a transjugular intrahepatic portosystemic shunt was felt unlikely to improve survival. Patients with end-stage liver disease and a UKELD score greater than 49 have previously been shown to benefit from liver transplantation in terms of their predicted survival at one year (Barber et al 2007), so this together with the patient's diuretic resistant ascites lead us to proceed with a formal liver transplant assessment.

Computed tomography (CT) confirmed the previous findings at US (Figure 1, panel A). Multiple high-density calcified nodules were also noted. Further, there was significant calcification of the descending aorta and aortic arch (Figure 1, panels B and C). Transthoracic echocardiography demonstrated mild left ventricular diastolic dysfunction but was otherwise normal; nine minutes of exercise treadmill testing were completed without electrocardiographic changes. Psychiatric evaluation was satisfactory and the patient was listed for liver transplantation on the grounds of the accepted indication of diuretic-resistant ascites in the form of hydrothorax, further supported by a UKELD score greater than 49.

At operation, a mildly fatty, deceased donor allograft was implanted using duct-to-duct biliary anastomosis and cavo-cavoplasty; an infra-renal aortic conduit was fashioned as initial arterial inflow was impaired. Cold and warm ischemic times were 9 hours 10 minutes and 36 minutes, respectively. Standard post-operative immunosuppression with prednisolone, azathioprine and tacrolimus was administered. Explant histology showed cirrhosis and was consistent with the diagnosis of CESD. Numerous lipid-laden macrophages and Kupffer cells were present within sinusoids that were CD68 positive and diastase periodic acid Schiff resistant (Figure 2). No additional form of chronic liver disease was apparent.

Initial recovery post transplantation was complicated by an episode of acute cellular rejection that resolved with intravenous methyl-prednisolone and the patient was discharged home three weeks following transplantation. Repeat enzyme analysis at discharge showed normalisation of plasma chitotriosidase activity (42 μmol/L/h) and persistent leukocyte acid esterase deficiency (39 μmol/g/h). Repeated fasting lipid profile revealed a significant rise in HDL cholesterol (Table 1).

Ten weeks post-transplant, during investigation for asymptomatic deranged liver function tests, imaging revealed unexpected hepatic artery thrombosis. Following clinical deterioration, the patient was subsequently re-listed and received a second transplant three months following her initial graft. Explant histology revealed an ischemic liver with organised thrombus of the hepatic artery at the hilum. The thrombosed arterial conduit was left in situ and not examined histologically. Subsequent progress has been uncomplicated; the patient was formally

anticoagulated with low molecular weight heparin for three months following re-transplant to reduce the risk of further hepatic artery thrombosis and is currently well on aspirin and statin therapy with excellent graft function almost two years following her second transplant.

In order to further characterise this patient's disorder, sequencing of LIPA was performed (Figure 3). This revealed compound heterozygosity: c.[894G>A];[599T>C] (the common c.894G>A substitution described above and a T to C substitution at location 599 causing leucine to proline substitution). The second mutation is thought to disrupt alpha helical structure (Anderson et al 1994). This compound heterozygous combination has been previously reported in siblings with CESD (Maslen et al 1995).

Discussion

We have presented the first report of adult liver transplantation for CESD. Our subject developed diuretic-resistant ascites with hydrothorax at the age of 42 years, having been diagnosed in childhood and run an indolent course during early adulthood. CESD has been previously diagnosed *de novo* in adulthood: Elleder et al (1990) described CESD in two women aged 43 and 56 years respectively, both of whom had asymptomatic hepatomegaly. One of these patients subsequently died of cerebrovascular disease. The same group subsequently described a 51 year-old man with a long-standing history of dyslipidemia who died of cholangiocarcinoma (Elleder et al 2000). Autopsy findings demonstrated hepatomegaly with micronodular

cirrhosis and disseminated cholangiocarcinoma; extensive severe atherosclerosis was also observed (Elleder et al 2000). Chatrath et al (2009) reported a 43 year-old man diagnosed with CESD on liver biopsy performed with a view to staging hepatitis C infection. Further, they summarised the features of an additional 18 cases previously described. Reported deaths were either due to vascular disease or hepatic disease (liver failure or hepatobiliary malignancy).

No drug therapy exists for this condition, although normalisation of lipid profiles using cholestyramine or HMG-CoA reductase inhibitors has been reported (Leone et al 1991; McCoy and Yokoyama 1991). Normalisation of the lipid profile does not necessarily prevent progression to hepatic failure in CESD, however, as seen in the case reported by Leone et al (1995). Enzyme replacement therapy has shown promise in reducing lipid deposition in peripheral tissues in mouse and rat models of complete LAL deficiency analogous to Wolman's disease, and is now being examined in phase I/II clinical trials for patients with CESD (Du et al 2008; Enns 2012). Although this is not currently routine treatment and is unlikely to be of significant benefit once irreversible liver damage has occurred this is an important research area, as is detection of CESD in adults with abnormal liver function.

Although successful liver transplantation for CESD has previously been reported in children and young adolescents (Arterburn et al 1991; Ferry et al 1991; Leone et al 1995), this patient is far older than any of the previously reported cases. As a result of the increased cardiovascular mortality seen in CESD, and given that the patient had been untreated for the vast majority of her life, a major concern was the degree

of established atherosclerosis and associated cardiovascular risk present. Although our patient had no overt signs of vascular disease, CT imaging had revealed extensive aortic calcification (Figure 1). Furthermore, doppler ultrasound imaging of the carotid arteries revealed dense plaques associated with mild stenoses. These findings of established atherosclerosis would be in keeping with her known CESD and may have been a factor in the development of post-transplant hepatic artery thrombosis; arterial anastomosis is technically more challenging in the presence of calcified atherosclerotic plaques and plaque rupture around the site of anastomosis can occur. Despite these findings, we felt that there remained a clear benefit for liver transplantation.

Initial fasting lipid profiling demonstrated hypercholesterolemia with low HDL cholesterol (Table 1). Although statin therapy led to a reduction in total cholesterol, the low HDL cholesterol reversed only following liver transplantation. Multiple studies in healthy subjects have demonstrated the predictive importance of low HDL cholesterol in future cardiac and cerebrovascular outcome (Natarajan et al 2010). Indeed a recent consensus statement on defining the metabolic syndrome has used a cut-off of less than 1.3 mmol/L as abnormal in a female population (Alberti et al 2009). Normalisation of HDL cholesterol levels in CESD following commencement of HMG-CoA reductase inhibitors has previously been reported in pre-cirrhotic cases in the paediatric population (Leone et al 1991), so it is likely that the low HDL cholesterol level observed here was related to impaired liver synthetic function. Liver transplantation may therefore be effective in reducing long-term vascular risk although this may be counter-balanced by the side-effects of calcineurin inhibitors

and the ongoing peripheral defect as evidenced by persistent leukocyte acid esterase deficiency.

In summary, we present the first published case of successful liver transplantation in an adult with CESD. The report highlights the challenges of concomitant vascular disease and the potential indolent nature of CESD in adulthood. The patient described here was lost to follow up twice before presenting with severe decompensated liver disease. Our case therefore also emphasises the need for educating patients with asymptomatic but potentially progressive liver disease and for continued awareness amongst clinicians overseeing adult metabolic disorders. Two years following transplantation, the patient has remained free of vascular events, with excellent liver function and a normal lipid profile. Liver transplantation for CESD may therefore yield a significant survival advantage even well into adulthood. There is little evidence to support liver replacement other than in the context of liver failure. We can only speculate as to whether pre-emptive liver transplantation might ameliorate the mortality associated with vascular disease in CESD, particularly if HMG-CoA reductase inhibition might already offset this. Liver transplantation is therefore a viable option for adults with liver failure related to CESD; close monitoring of patients with CESD and liaison with tertiary hepatology services are important for the optimal timing of this intervention.

Conflicts of interest and financial disclosures

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References

Alberti KG, Eckel RH, Grundy SM et al (2009) Harmonizing the metabolic syndrome: a joint interim statement of the International Diabetes Federation Task Force on Epidemiology and Prevention. *Circulation* 120: 1640–1645.

Anderson RA, Byrum RS, Coates PM, et al (1994) Mutations at the lysosomal acid cholesteryl ester hydrolase gene locus in Wolman disease. *P Natl Acad Sci USA* 91: 2718–2722.

Anderson RA, Rao N, Byrum RS, et al (1993) In situ localization of the genetic locus encoding the lysosomal acid lipase/cholesteryl esterase (LIPA) deficient in Wolman disease to chromosome 10q23.2-q23.3. *Genomics* 15: 245– 247.

Anderson RA and Sando GN (1991) Cloning and expression of cDNA encoding human lysosomal acid lipase/cholesteryl ester hydrolase. Similarities to gastric and lingual lipases. *J Biol Chem* 266: 22479–84.

Arterburn JN, Lee WM, Wood RP, et al (1991) Orthotopic liver transplantation for cholesteryl ester storage disease. *J Clin Gastroenterol* 13: 482–485.

Barber KM, Pioli SE, Blackwell JE, et al (2007) Development of a UK score for patients with end-stage liver disease. *Hepatology* 46: 510A.

Chatrath H, Keilin S, Attar BM (2009) Cholesterol ester storage disease (CESD) diagnosed in an asymptomatic adult. Dig Dis Sci 54: 168–173.

Du H, Cameron TL, Garger SJ, et al (2008) Wolman disease/cholesteryl ester storage disease: efficacy of plant-produced human lysosomal acid lipase in mice. *J Lipid Res* 49: 1646–1657.

Elleder M, Chlumska A, Hyanek J, et al. (2000) Subclinical course of cholesteryl ester storage disease in an adult with hypercholesterolemia, accelerated atherosclerosis, and liver cancer. *J Hepatol* 32: 528–534.

Elleder M, Chlumska A, Ledvinova J, et al (2000) Testis - a novel storage site in human cholesteryl ester storage disease. Autopsy report of an adult case with a long-standing subclinical course complicated by accelerated atherosclerosis and liver carcinoma. *Virchows Arch* 436: 82–87.

Elleder M, Ledvinova J, Cieslar P, et al (1990) Subclinical course of cholesterol ester storage disease (CESD) diagnosed in adulthood. Report on two cases with remarks on the nature of the liver storage process. *Virchows Arch A* 416: 357–365.

Enns G (2012) Initial human experience with sbc-102, a recombinant enzyme replacement therapy in adults with lysosomal acid lipase deficiency. Presented at the 8th Annual Lysosomal Disease Network (LDN) World Symposium.

Ferry GD, Whisennand HH, Finegold MJ, et al (1991) Liver transplantation for cholesteryl ester storage disease. *J Pediatr Gastroenterol Nutr* 12: 376–378.

Kamath PS, Wiesner RH, Malinchoc M, et al (2001) A model to predict survival in patients with end-stage liver disease. *Hepatology* 33: 464–470.

Leone L, Ippoliti PF, Antonicelli R (1991) Use of simvastatin plus cholestyramine in the treatment of lysosomal acid lipase deficiency. *J Pediatr* 119: 1008–1009.

Leone L, Ippoliti PF, Antonicelli R, et al (1995) Treatment and liver transplantation for cholesterol ester storage disease. *J Pediatr* 127: 509–510.

Lewsey JD, Dawwas M, Copley LP, et al (2006) Developing a prognostic model for 90-day mortality after liver transplantation based on pretransplant recipient factors. *Transplantation* 82: 898–907.

Maslen CL, Babcock D, Illingworth DR (1995) Occurrence of a mutation associated with Wolman disease in a family with cholesteryl ester storage disease. *J Inherit Metab Dis* 18: 620–623.

McCoy E and Yokoyama S (1991) Treatment of cholesteryl ester storage disease with combined cholestyramine and lovastatin. *Annals NY Acad Sci* 623: 453–454.

Muntoni S, Wiebusch H, Jansen-Rust M, et al (2007) Prevalence of cholesteryl ester storage disease. *Arterioscler Thromb Vasc Biol*, 27: 1866–1868.

Natarajan P, Ray KK, Cannon CP (2010) High-density lipoprotein and coronary heart disease: current and future therapies. *J Am Coll Cardiol* 55: 1283–1299.

Pisciotta L, Fresa R, Bellocchio A, et al (2009) Cholesteryl Ester Storage Disease (CESD) due to novel mutations in the LIPA gene. *Mol Genet Metab* 97: 143–148.

Figure legends

Figure 1. CT images of a 42 year-old woman with end-stage liver disease due to CESD. Imaging obtained prior to liver transplantation. Post-contrast axial CT image of the upper abdomen showing a nodular liver containing multiple tiny foci of calcification in association with splenomegaly, ascites and a moderate right pleural effusion (A). Non-contrast sagittal CT images of the major arteries showing extensive calcification in the abdominal and thoracic aorta (B and C respectively).

Figure 2. Liver explant histology following transplantation for end-stage liver disease due to CESD. Chromotrope Aniline Blue staining (A) demonstrating established cirrhosis with complete nodules surrounded by broad fibrous septae (x40). Haematoxylin and Eosin (H&E) stained section (B) demonstrating numerous foamy macrophages with tan-coloured cytoplasm within residual portal areas and fibrous septae (x200). H&E stained section (C) demonstrating lipid laden, foamy hypertrophic Kupffer cells and sinusoidal macrophages (arrow) (x400). CD68 staining (D) showing strong positivity for intrasinusoidal macrophages and Kupffer cells (x400) which are periodic acid Schiff positive and diastase resistant (E) (x400).

Figure 3. DNA sequence chromatograms of a section of the *LIPA* gene from a 42 year-old woman with CESD. The grey boxes highlight heterozygous substitutions found in both forward and reverse directions at locations 599 (top, C substituted for wild-type T) and 894 (bottom, A substituted for wild-type G). The former is thought to result in disruption of alpha-helical structure, while the latter is found at a splice junction and usually causes deletion of exon 8, resulting in less than 10% expression of normal protein (r.[894g>t,822_894delinsu]).