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Graft-versus-host disease

Severe hypercholesterolemia mediated by lipoprotein X in patients with chronic graft-versus-host disease of the liver

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Summary:

We describe a series of cases of extreme hypercholesterolemia mediated by lipoprotein X in patients with chronic graft-versus-host disease of the liver after an allogeneic bone marrow transplant. All of the patients presented with a total cholesterol in excess of 1000 mg/dl (25.9 mmol/l). At the time they were also noted to have pseudohyponatremia. Cholesterol appeared to be predominantly carried by lipoprotein X. Intrahepatic cholestasis leading to reflux of bile lipoproteins into the bloodstream and subsequent formation of protein X appears to be the mechanism underlying this phenomenon. Complications, including retinal cholesterol thromboembolism and cholesteroloma of the lung have been seen in the patient with the highest cholesterol levels. Severe hypercholesterolemia is an important, and likely more common than previously reported, long-term complication of allogeneic hematopoietic stem cell transplantation. It is important for clinicians to familiarize themselves with the diagnostic and therapeutic challenges this condition presents.

While hypercholesterolemia remains a prevalent disorder in the Western world, few conditions have been documented to cause hypercholesterolemia above 1000 mg/dl (25.9 mmol/l). Differential diagnosis of hypercholesterolemia of this magnitude includes homozygous familial hypercholesterolemia,[1] hepatic lipase deficiency,[2] and obstructive biliary cholestasis.[3,4] Such extreme hypercholesterolemia can lead to a number of serious, even fatal

sequelae, ranging from premature coronary artery disease (myocardial infarctions have been documented as early as 18 months of age in patients with homozygous familial hypercholesterolemia[5]) to hyperviscosity syndrome.[6] Its treatment can be very difficult and require multiple medications[7,8] as well as more invasive measures, such as low-density lipoprotein (LDL) apheresis[9] or plasmapheresis.[6]

Allogeneic hematopoietic stem cell transplantation, an increasingly common treatment for a number of malignant and nonmalignant disorders, often leads to graft-versus-host disease (GVHD) of the liver (40% of recipients from HLA-matched related donors[10]). GVHD of the liver frequently presents with a cholestatic picture. However, to our knowledge only two cases of severe hypercholesterolemia in bone marrow transplant patients have been reported in the literature.[11,12] Consequently, there may be insufficient awareness of this important complication among the physicians caring for these frequently very ill patients. From our experience, it appears that it may be more frequent than commonly perceived. In this paper, we describe three cases of extreme hypercholesterolemia developing in patients with chronic GVHD of the liver, and analyze its presentation, possible sequelae, and management.

Methods

Study population

Three patients with severe hypercholesterolemia mediated by lipoprotein X after allogeneic hematopoietic stem cell transplantation, who were seen by us over a 2-month period, are described.

The Institutional Review Boards (IRBs) from the respective hospitals did not require a review of this study. All patients were transplanted on treatment plans approved by the IRB or an equivalent peer-review committee.

Biochemical analysis

Serum was subjected to sequential ultracentrifugation to separate it into three fractions: density <1.006 g/ml (very low density), 1.006–1.063 g/ml (low density), and >1.063 g/ml (high density). The resulting fractions were subjected, together with controls, to 1% agarose gel electrophoresis using barbital

buffer (pH 8.6) in a standard Corning electrophoresis system for 30 min and stained for protein (Amido Black) and lipids (Fat Red 7B). Concentrations of cholesterol, gamma-glutamyltransferase, and alkaline phosphatase were measured in the high- and low-density fractions.

Case I

A 64-year-old woman with history of acute myelogenous leukemia (AML), who underwent allogeneic hematopoietic stem cell transplantation 2 years earlier, presented to the hospital with progressive liver dysfunction due to GVHD (confirmed by liver biopsy). Some details of this patient's course have been previously reported.[13] On admission, the patient's total bilirubin was 9.0 mg/dl (153.9 μmol/l), alkaline phosphatase 449 U/l, AST 101 U/l, and ALT 142 U/l. Her sodium was found to be decreased at 124 mmol/l (as measured by indirect potentiometry). Further investigation showed this to be due to pseudohyponatremia (direct potentiometry measurements showed the sodium level to be 135 mmol/l); the patient's lipid profile was measured and her total cholesterol was found to be 1836 mg/dl (47.6 mmol/l), HDL cholesterol 68 mg/dl (1.76 mmol/l), and triglycerides 208 mg/dl (2.35 mmol/l); LDL cholesterol was not directly measured. Her only known previous measurement of lipid profile was done 2 years prior to admission and showed total cholesterol of 181 mg/dl (4.69 mmol/l), HDL cholesterol 51 mg/dl (1.32 mmol/l), and triglycerides 136 mg/dl (1.54 mmol/l). Dermatological examination did not show any xanthomata. Serum viscosity was slightly elevated at 1.9 centipoise; she had no signs or symptoms consistent with hyperviscosity syndrome. Her medications included high-dose (50 mg/day) prednisone, mycophenolate, and tacrolimus. Screening for other potential causes of hypercholesterolemia revealed elevated TSH (72 μU/ml) and low total T4 (0.6 μg/dl; 7.72 nmol/l) and T3 (41 ng/dl; 0.63 nmol/l).

The patient's serum was sequentially ultracentrifuged to separate it into very low-density (< 1.006 g/ml), low-density (1.006–1.063 g/ml), and high-density (> 1.063 g/ml) fractions as described above. Gel electrophoresis of the fractionated serum showed a broad lipoprotein band in the patient's low-density fraction. Cholesterol as well as liver enzymes were measured in each of these fractions, demonstrating high levels of alkaline phosphatase and gamma-glutamyltransferase associated with the lipoprotein in the low-density fraction. These findings, in conjunction with relatively low triglyceride levels, were consistent with the presence of large amounts of lipoprotein X.[14–18]

The patient was started on colestipol, gradually escalating the dose to a maximum of 30 g/day. A month later, her total cholesterol had decreased to 1114 mg/dl (28.9 mmol/l). However, her liver dysfunction had progressed over the same time. The patient expired from infectious complications related to immunosuppression shortly afterwards.

Case II

The patient was a 47-year-old woman with a history of chronic myelogenous leukemia (CML) who had undergone allogeneic bone marrow transplantation 10 months previously. Her course had been complicated by chronic GVHD of the liver (diagnosed by liver biopsy), chronic renal insufficiency, and glucocorticoid-induced diabetes mellitus. She was found to have total cholesterol of 1236 mg/dl (32.0 mmol/l) 10 months after the transplant. At 3 months after the transplant, her total cholesterol had been measured to be 407 mg/dl (10.5 mmol/l); it then rose to 954 mg/dl (24.7 mmol/l) 9 months after the transplant. Her medications over this period of time included prednisone, mycophenolate, and tacrolimus. Thyroid function tests showed TSH to be 0.16 μU/ml and free thyroxine 0.8 ng/dl (10.3 pmol/l). Most of the cholesterol was demonstrated to be carried by lipoprotein-X using an ultracentrifugation and gel electrophoresis-based method identical to the technique employed for Patient 1. Her dyslipidemia was not treated due to the concomitant progressive hepatic dysfunction. She died from complications of liver failure secondary to chronic GVHD 1 year after the transplant.

Case III

The patient was a 37-year-old man with a history of CML who underwent allogeneic bone marrow transplantation in February of 2002. After 9 months, he developed chronic GVHD of the liver (confirmed by biopsy). Over the next 2 weeks his sodium (as measured by indirect potentiometry) declined from 140 to 129 mmol/l. This was shown to be due to pseudohyponatremia (sodium measured by direct potentiometry was determined to be 135 mmol/l). His total cholesterol was subsequently measured to be 977 mg/dl (25.3 mmol/l), HDL cholesterol 23 mg/dl (0.60 mmol/l), triglycerides 240 mg/dl (2.71 mmol/l), measured LDL cholesterol 218 mg/dl (5.65 mmol/l). The total cholesterol 6 months prior to the transplant was 221 mg/dl (5.72 mmol/l). Most of the cholesterol was demonstrated to be carried by lipoprotein-X using an ultracentrifugation and gel electrophoresis-based method, identical to that described above. His medications included sirolimus, mycophenolate, and prednisone. Thyroid function tests showed the TSH to be 0.56 μU/ml and free thyroxine 0.8 ng/dl (10.3 pmol/l). No antihyperlipidemic treatment was initiated due to the liver disease. His GVHD was successfully treated with immunosuppression and, as his liver function improved, his cholesterol had decreased to 329 mg/dl (8.52 mmol/l) 10 months later without specific intervention.

Discussion

Severe hypercholesterolemia is not a commonly reported complication of allogeneic hematopoietic stem cell transplantation. Several possible etiologies could be identified for the patients we described.

A number of medications used in immunosuppressive regimens have been documented to cause dyslipidemias. These include cyclosporine,[19] tacrolimus,[20] sirolimus,[21,22] mycophenolate,[23] and glucocorticoids.[24] Cholesterol levels between 200 and 300 mg/dl (5.18–7.77 mmol/l), mediated by increased LDL cholesterol levels, and triglyceride levels

between 200 and 350 mg/dl (2.26–3.96 mmol/l) are usually described. All of the patients in this series were taking one or more of these medications. While their contribution to the patients' hypercholesterolemia cannot be ruled out, it must be noted that most of the cholesterol in these patients was not carried by LDL as is characteristic of the medication-induced dyslipidemia, but by lipoprotein X; it is therefore unlikely that the drugs played more than a secondary role in the patients' conditions. However, medication-mediated hypercholesterolemia remains an important complication of allogeneic bone marrow transplantation, which should be monitored.

Hypothyroidism, which was pronounced in the first patient but absent in all others, is a known complication of allogeneic hematopoietic stem cell transplantation.[25] It is also well documented to cause hyperlipidemia.[26] However, the typical range of cholesterol levels seen in hypothyroidism is well below the levels observed in this patient. On average, total cholesterol levels in patients with both primary and secondary hypothyroidism are between 200 and 300 mg/dl (5.18–7.77 mmol/l), with occasional outliers reaching 600 mg/dl (15.6 mmol/l).[27] Importantly, unlike in this patient, most of the excess cholesterol in hypothyroidism-induced hypercholesterolemia is carried by LDL. Consequently, it is also unlikely to have played a dominant role in these patients' condition.

Agarose gel electrophoresis demonstrated that hypercholesterolemia in all of the patients described in this series was mediated mostly by lipoprotein X rather than LDL. Lipoprotein X is present in patients with intra- or extrahepatic cholestasis, lecithin:cholesterol acyltransferase (LCAT) deficiency, and in newborn infants with immature liver function.[18,28] It is composed primarily of phospholipids and unesterified cholesterol. Reflux of bile into the bloodstream has been hypothesized to be a possible mechanism for its formation. Given that LCAT deficiency and other congenital disorders of lipid metabolism had been excluded by previously documented lower lipid levels in all of these patients, their hyperlipidemia was apparently due to intrahepatic cholestasis caused by chronic GVHD of the liver. Chronic GVHD of the liver is an immune-mediated disorder which can appear anywhere from 2 months to 2 years after transplantation and typically presents with a cholestatic picture.[29] Other cholestatic liver diseases, such as primary biliary cirrhosis, have long been known to occasionally result in severe hypercholesterolemia;[4] however, this degree of hypercholesterolemia is not known to be common in chronic GVHD of the liver.

The data on the three patients presented in this paper as well as two other patients with this condition previously reported in the literature are summarized in Table 1.[11,12] A characteristic presentation involves development of chronic GVHD of the liver within 1 year after allogeneic bone marrow transplantation followed by new onset pseudohyponatremia which typically prompts the measurement of the cholesterol level. While a number of sources have stated in the recent years that pseudohyponatremia is nearly extinct due to the changes in laboratory equipment,[30–32] this in fact is no longer the case, and most modern instruments are susceptible to this error.[13] Apart from the pseudohyponatremia, most patients did not suffer any

Table 1 Patients with extreme hypercholesterolemia secondary to GVHD of the liver after bone marrow transplantation

Patient	I	II	III	IV[a]	V[b]
Gender	F	F	M	M	F
Age	64	47	37	14	22
Highest documented cholesterol level, mg/dl (mmol/l)	1836 (47.6)	1236 (32.0)	1023 (26.5)	2391 (61.9)	2509 (65.0)
Indication for bone marrow transplant	AML	CML	CML	AML	ALL
Time since transplant at diagnosis of hypercholesterolemia, months	24	6	9	9	4
Immunosuppressant regimen at the time of diagnosis	MMF, prednisone, tacrolimus	MMF, prednisone, tacrolimus	MMF, prednisone, sirolimus	Azathioprine, prednisolone	Cyclosporine, prednisone
Presenting symptom of hypercholesterolemia	PHN	Routine screening	PHN	PHN	Not reported
Complications of high cholesterol	None	None	None	None reported	Xanthomata, retinal thromboembolism, cholesteroloma of the lung
Treatment of hypercholesterolemia	Colestipol	Expectant	Expectant	Expectant	Colestipol, lovastatin, plasmapheresis
Outcome	Deceased	Deceased	Spontaneous improvement	Spontaneous improvement	Resolution after treatment

ALL = acute lymphoblastic leukemia; AML = acute myelogenous leukemia; CML = chronic myelogenous leukemia; MMF = mycophenolate mofetil; PHN = pseudohyponatremia.
[a]As reported by Coakley et al.[11]
[b]As reported by Toren and Nagler.[12]

complications of their hypercholesterolemia; however, the one with the highest cholesterol level (2509 mg/dl; 65.0 mmol/l) developed xanthomata, retinal cholesterol thromboembolism, and a cholesteroloma of the lung, all of which resolved after treatment.[12] In addition to this, hyperviscosity syndrome has been reported in at least one patient with severe hypercholesterolemia mediated by lipoprotein X due to primary biliary cirrhosis.[6] None of the patients reported have been observed for a sufficient period of time to allow conclusions about the risk of development of vascular disease.

Given the potential complications of lipoprotein X-mediated hypercholesterolemia, it is important to consider available therapeutic options. Treatment of the underlying GVHD of the liver by adjustment of immunosuppressant regimens remains the mainstay of therapy. This strategy led to resolution or significant improvement of hypercholesterolemia in three out of the five patients. However, GVHD is not always amenable to the currently available treatment, and other approaches may need to be considered to prevent sequelae of extreme hypercholesterolemia. The choice is limited as most of the antihyperlipidemic medications cannot be safely used in patients with significant hepatic damage. In cases with severe complications, plasmapheresis can be employed, as has been reported for one of the patients with GVHD-induced lipoprotein X hypercholesterolemia.[12] As illustrated in this review, extreme hypercholesterolemia, confounded by other comorbidities in these complicated patients, is probably more common than previously reported; it is therefore important that health care providers for this group of patients are aware of the problem, its potential sequelae, and management.

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