Intrahepatic Cholestasis After Liver Transplantation

Ziv Ben-Ari,* Orit Pappo,† and Eytan Mor‡

Cholestasis is a common sequela of liver transplantation. Although the majority of cases remain subclinical, severe cholestasis may be associated with irreversible liver damage, requiring retransplantation. Therefore, it is essential that clinicians be able to identify and treat the syndromes associated with cholestasis. In this review, we consider causes of intrahepatic cholestasis. These may be categorized by time of occurrence, namely, within 6 months of liver transplantation (early) and thereafter (late), although there may be an overlap in their causes. The causes of intrahepatic cholestasis include ischemia/reperfusion injury, bacterial infection, acute cellular rejection, cytomegalovirus infection, small-for-size graft, drugs for hepatotoxicity, intrahepatic biliary strictures, chronic rejection, hepatic artery thrombosis, ABO blood group incompatibility, and recurrent disease. The mechanisms of cholestasis in each category and the clinical presentation, diagnosis, treatment, and outcome are discussed in detail. (Liver Transpl 2003;9:1005-1018.)

Early Intrahepatic Cholestatic Syndrome

Ischemia/Reperfusion Injury

Graft failure can be caused by hypothermic and hypoxic graft storage (cold ischemia), sustained ischemia during graft implantation, and restoration of blood and oxygenation to the graft (reperfusion injury). The cold preservation damages nonparenchymal cells, although parenchymal cells seem to maintain their function.3-6 Specifically, cold preservation leads to sinusoidal endothelial cell impairment7 and sinusoidal accumulation of leukocytes and platelets.8-10 It also activates Kupffer cells,3 thereby inducing the release of interleukin (IL) 8, E-selectin, and other compounds that initiate and perpetuate the injury and impair hepatic microcirculation.11 Initially, the risk of biliary complications after transplantation was attributed to the type of biliary reconstruction, the use of steatotic liver grafts, and the duration of cold ischemic storage.12-15 Studies showed that a storage time in excess of 10 to 12 hours led to biliary strictures and other complications in more than 25% of liver transplant recipients.16-18 When storage was prolonged beyond 13 hours, nonanastomotic strictures developed in 52% of patients, and when it lasted more than 15 hours, the stricture rate...
increased to 69%. There is now evidence, however, that the preservation injury occurs at the level of the bile duct cells. The possible underlying mechanisms of bile duct epithelial cell injury after transplantation are suggested by experimental studies showing their increased detachment from the basement membrane with an increase in preservation time and their greater sensitivity to reperfusion injury compared with hepatocytes. In addition, hydrophobic bile salts can apparently damage intrahepatic bile ducts and amplify the preservation injury. One study of cultured biliary epithelial cells reported that the adenosine triphosphate (ATP) depletion at the end of cold ischemic storage caused a marked delay in functional recovery.

Clinically, reperfusion injury to the graft parenchymal cells is assessed by serum levels of aspartate aminotransferase or alanine aminotransferase. Elevations usually last from several hours to approximately 2 days, and levels normalize within 1 week. By contrast, reperfusion injury to the graft biliary tree, reflected by serum gamma-glutamyl transferase and bilirubin levels, persists for up to 17 days. Histologically, the diagnosis of preservation-reperfusion injury is based on the presence of steatosis, cholestasis, and ballooning degeneration of hepatocytes in early posttransplantation biopsies (Fig. 1).

Although the use of University of Wisconsin solution has permitted an increase in mean preservation time, severe ischemia/reperfusion injury still affects approximately 4.2% of grafts, mainly in the form of primary nonfunction. Up to 25% of patients with graft dysfunction require retransplantation in the first 3 months postoperatively. Promising potential treatments include the replacement of hydrophobic bile salts with hydrophilic ones such as tauroursodeoxycholate to reduce both duct and hepatocyte injury. The modulation of intracellular signaling pathways, such as the activation of nuclear factor KB, which has been shown to inhibit apoptosis by tumor necrosis factor alpha, may also find application in the ischemic liver. Finally, based on the study of Vajdova et al, the inclusion of energy substrates to the perfusion medium may allow for complete restoration of tissue ATP levels, thereby attenuating injury (Table 2).

### Table 1. Syndromes of Intrahepatic Cholestasis After Liver Transplantation (Early and Late)

<table>
<thead>
<tr>
<th>Early (≤6 mo After Transplantation)</th>
<th>Late (&gt;6 mo After Transplantation)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ischemia/reperfusion injury</td>
<td>Intrahepatic biliary strictures</td>
</tr>
<tr>
<td>Bacterial infection</td>
<td>Chronic rejection</td>
</tr>
<tr>
<td>Small-for-size graft</td>
<td>Hepatic artery thrombosis and stenosis</td>
</tr>
<tr>
<td>Acute cellular rejection</td>
<td>ABO blood type incompatibility</td>
</tr>
<tr>
<td>CMV infection</td>
<td>Recurrent disease</td>
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<tr>
<td>Drugs</td>
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</table>

### Bacterial Infection

Septic complications are responsible for the majority of the morbidity in liver transplant recipients during the first 3 months after transplantation. Bacteremia occurs in 19% to 25% of patients within 30 days of transplantation and accounts for 22% to 36% of all major infections in this patient group. By source, the most common infections are catheter-related, nosocomial, and intra-abdominal. However, the spectrum of infections seems to be evolving, and those caused by gram-positive bacteria (e.g., Enterococcus and Staphylococcus) are now more frequent (40% to 59% of all cases of bacteremia) than those caused by gram-negative bacteria. More than half of all staphylococcal bacteremias are catheter-related, whereas gram-negative bacteremias originate primarily from an intra-abdominal or pulmonary source. Enteric gram-negative bacteria (Klebsiella and Enterobacter) and Pseudomonas aeruginosa are the most frequently encountered pathogens.

Severe preservation-reperfusion injury associated with cholestasis may precede the development of septic complications. The liver plays a major role in the clearance of systemic toxemia, and probably acts as a regulatory organ in the host defense system through interactions between liver macrophages (mainly Kupffer cells) and the immune system.

**Figure 1. Preservation injury: hepatocyte ballooning and steatosis, small aggregates of neutrophils.***
cells) and hepatocytes. The activated liver simultaneously produces and releases various cytokines. Moseley et al.\(^4^5\) reported that intrahepatic cholestasis in the setting of extrahepatic bacterial infection was attributable to the direct effects of endotoxin and/or endotoxin-induced lipopolysaccharide-induced proinflammatory cytokines (tumor necrosis factor alpha, IL-1, IL-6, and IL-8) on bile acid transport at the sinusoidal and canicular membrane domains.\(^4^0\) These findings were supported by a study in an endotoxemic rat model showing marked impairment of basolateral and canicular bile acid and organic anion transport contributing to the cholestasis of sepsis.\(^4^3\) Clinically, bacterial infection may present as high fever and increased levels of serum cholestatic liver enzymes and positive blood cultures.

Liver biopsy specimens from patients with bacteria-induced intrahepatic cholestasis typically show biliary tract inflammation with neutrophil infiltrates, bile duct proliferation, and bile plugs.\(^4^6\) The definitive diagnosis is made only after biliary tract obstruction is excluded by imaging studies of the biliary tree (percutaneous transhepatic cholangiography or endoscopic retrograde cholangiopancreatography). Lefkowitch\(^4^8\) described an unusual form of intrahepatic cholestasis in patients with gram-negative intra-abdominal infection that was manifested by inspissated bile within dilated and proliferated portal and periporal bile ductules. He called this disorder cholangitis lenta. Antibiotic treatment of the underlying infection leads to resolution of clinical and pathologic cholestasis. More recent studies have confirmed the relationship between systemic infection and a striking cholangitic response in patients after liver transplantation.\(^5^5\) Biopsy specimens displayed prominent cholangitis with "obstructive"-type features in the presence of consistently normal cholangiograms. Treatment of the underlying infection

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### Table 2. Characteristics of Early Intrahepatic Cholestatic Syndromes After Liver Transplantation

<table>
<thead>
<tr>
<th>Syndrome</th>
<th>Mechanism of Cholestasis</th>
<th>Clinical Presentation</th>
<th>Diagnosis</th>
<th>Treatment</th>
<th>Outcome</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ischemia/reperfusion injury</td>
<td>Detachment of bile-duct epithelium, ATP depletion, hydrophobic bile salt injury</td>
<td>Increased liver enzymes, nonanatomistic biliary strictures</td>
<td>Histology: cholestasis, ballooning</td>
<td>Supportive, hydrophilic bile salts</td>
<td>Mortality 20% per year, 30% retransplantation with graft dysfunction</td>
</tr>
<tr>
<td>Bacterial infection</td>
<td>Cytokine release (TNF-alpha, IL-1, IL-6, IL-8), impaired bile acid transport</td>
<td>Increased cholestatic liver enzymes, fever</td>
<td>Positive cultures Histology: biliary tract inflammation, neutrophil infiltrates, bile plugs, normal ERCP/PTC</td>
<td>Bacterial infection</td>
<td>Early</td>
</tr>
<tr>
<td>Small-for-size graft</td>
<td>Portal hypertension Sinusoidal and hepatocyte cell damage</td>
<td>Increased liver enzymes, jaundice, coagulopathy, ascites</td>
<td>Clinical and histology: diffuse ischemic patterns, ballooning and cholestasis</td>
<td>Retransplantation when not resolved</td>
<td>Either resolved or retransplantation</td>
</tr>
<tr>
<td>Acute cellular rejection</td>
<td>Lymphocytic cholangitis (CD8), cytokine release, loss of Na+K+ ATPase</td>
<td>Increased liver enzymes</td>
<td>Histology: portal inflammation, lymphocytic cholangitis, endothelialitis</td>
<td>Immunosuppressive regimen</td>
<td>Usually resolves Seldom requires retransplantation</td>
</tr>
<tr>
<td>CMV infection</td>
<td>Hepatitis and cholangitis</td>
<td>Increased liver enzymes, jaundice</td>
<td>Positive viral cultures Histology: intranuclear inclusion bodies, Immunohistochemistry In-situ DNA hybridization</td>
<td>Ganciclovir</td>
<td>Usually resolves</td>
</tr>
<tr>
<td>Drugs</td>
<td>Cyclosporine: impaired bile formation, reduction in bile flow, inhibited bile salt export pump</td>
<td>Increased cholestatic liver enzymes, bilirubin, biliary sludge</td>
<td>Histology: cholestasis, increased cyclosporin trough level</td>
<td>Dose reduction, Conversion to tacrolimus, UDCA</td>
<td>Usually resolves</td>
</tr>
<tr>
<td>Azathioprine: damage to hepatic sinusoidal and venular endothelial cells</td>
<td>Increased cholestatic liver enzymes</td>
<td>Histology: veno-occlusive disease, peliosis hepatitis, nodular regenerative hyperplasia</td>
<td>Withdrawal</td>
<td>Usually resolves when identified early</td>
<td></td>
</tr>
<tr>
<td>Sulfonamides: idiosyncratic</td>
<td>Increased liver enzymes</td>
<td>Histology: vanishing bile ducts</td>
<td>Withdrawal</td>
<td>Usually resolves when identified early</td>
<td></td>
</tr>
</tbody>
</table>

Abbreviations: ATP, adenosine triphosphate; ERCP, endoscopic retrograde cholangiopancreatography; PTC, percutaneous transhepatic cholangiography; UDCA, ursodeoxycholic acid.
leads to resolution of the clinical and histologic changes, but the prognosis is poor in patients who do not respond to antibiotics (Table 2). 45

**Small-for-Size Graft**

The shortage of organs for transplantation has prompted physicians on some occasions to use a pediatric liver for an adult recipient. More recently, the innovative technique of transplanting a segmental liver allograft from a living donor has been widely used. If the hepatocyte cellular mass is inadequate when these small-for-size grafts are used, graft function is impaired. This is indicated clinically by features of cholestasis associated with hyperbilirubinemia, increased prothrombin time, and profuse ascites. 46, 47 The diagnosis is based on histologic findings of diffuse ischemic patterns with cellular ballooning, which progress to cholestasis in subsequent biopsies. In a series of 70 livers from pediatric donors (age <13 years) transplanted in adult recipients, the incidence of arterial thrombosis and graft loss was high when the ratio of donor-liver weight to estimated recipient-liver weight was less than 0.4. 46 In the whole group, cholestasis with bilirubin levels >5 mg/dL on postoperative day 7 was associated with prolonged cold and warm ischemic times.

In the living donor liver graft scenario, the ratio of minimal graft weight to recipient standard liver volume has been defined as 40%. 47 Below that, there is a high risk for the development of a small-for-size syndrome with subsequent graft loss. Therefore, when the recipient of a living donor transplant is an adult, the use of a left liver lobe supplying only 40% of the total liver volume has been abandoned, and a right-lobe transplantation is preferred. According to one recent report, patients implanted with a small-for-size graft had transient portal hypertension early after reperfusion. 48 This was associated with intragraft upregulation of endothelin-1 and ultrastructural evidence of sinusoidal damage. A downregulation of heme oxygenase-1 and heat shock protein 70 may also account for hepatocyte injury and subsequent cholestasis. 48 In patients in whom the clinical picture is not resolved, retransplantation should be considered (Table 2).

**Acute Hepatic Allograft Cellular Rejection**

Despite major advances in immunosuppression, acute hepatic allograft rejection occurs in the majority of transplant recipients (64%), 49 usually within 7 to 10 days after transplantation. However, mild rejection is reported in 73% of patients in whom acute cellular rejection developed, and moderate or severe acute rejection developed in only 17% of them. 49 Acute hepatic cellular rejection may present with fever and increased liver enzyme levels, although clinical and laboratory tests are unreliable for diagnosis, and histopathologic analysis remains the gold standard. 49 Acute rejection is associated with lymphocytic cholangitis, a cytotoxic T-cell-mediated nonsuppurative destructive cholangitis of the small bile ducts that can induce cholestasis. 50, 51 Recently, Angermuller et al 52 showed that activated Kupffer cells migrate into the rejecting liver and release cytokines, resulting in the loss of Na+K+ATPase activity. Because Na+K+ATPase is the cotransporter for hepatocyte taurocholate uptake, these data may contribute to our understanding of the mechanism of cholestasis during acute allograft rejection. 52

The treatment of acute moderate-severe cellular rejection includes intravenous high-dose corticosteroids. Nonresponders may require OKT3 administration; conversion to tacrolimus in patients with cyclosporine-based immunosuppression is also effective, as is the addition of mycophenolate mofetil.

Although the cholestatic findings subside as the rejection resolves, in rare cases they can progress to severe and irreversible bile duct damage requiring retransplantation (Table 2). 53, 54

**Cytomegalovirus Infection**

Cytomegalovirus (CMV) is the most common viral infection after liver transplantation, 55, 56 usually occurring 1 to 4 months postoperatively. 55-57 Overall, it develops in 30% to 50% of liver transplant recipients. Approximately half of these cases are associated with symptomatic CMV disease. Clinically, CMV hepatitis presents with fever and jaundice and increased values on cholestatic liver chemistry tests. Diagnosis is based on positive viral cultures and CMV antigenemia assay. Histologically, cholestatic features are often present, in addition to microabcesses and typical intranuclear inclusion bodies. 58 A study of CMV-infected rat liver allografts undergoing acute rejection showed a significant increase in portal inflammation and more severe bile duct damage compared with uninfected grafts. 59 Indeed, CMV infection may be a risk factor for vanishing bile duct syndrome 59 and chronic liver graft rejection. 60, 61 The latter suggestion is supported by in situ hybridization and immunohistochemistry findings of CMV in patients with chronic graft rejection. 62 However, the CMV may not be a causal factor, but rather a consequence of graft injury or an “innocent bystander.” 63 The drug of choice for treating established CMV infection is ganciclovir (Table 2).
Drug-Induced Cholestasis
Advances in immunosuppressive therapy have played an important role in the evolution and success of liver transplantation. Agents that selectively target various cellular activation pathways involved in the immune response have become increasingly available. This has resulted in lower rates of graft rejection, thereby minimizing drug toxicity and morbidity, and in tailoring of regimens to meet specific patient requirements. There may be difficulties in diagnosing drug toxicity related to immunosuppressive agents in liver allograft because of the multifactorial possibilities of liver damage in this setting. However, several other hepatotoxic medications (antibiotics, antifungal, antiviral) may also be prescribed for prophylaxis against opportunistic infections, manifested clinically as an asymptomatic increase in liver enzymes. Careful consideration of the characteristic features of drug-induced cholestasis after liver transplantation is crucial to improving patient management.

Cyclosporine
Cyclosporine has potential hepatotoxic and/or cholestatic effects with either hyperbilirubinemia or with associated formation of biliary sludge. It interferes with several hepatic processes, mainly bile formation, inducing a cholestatic syndrome in rats and humans. The cholestasis is caused by a reduction in canalicular bile flow via inhibition of the ATP-dependent export carriers of bile salts in the hepatocyte canalicular membrane, impairment of bile formation dependent on the biliary secretion of glutathione, and inhibition of the canalicular bile salt export pump. Therefore, cyclosporine should be considered a cause of unexplained cholestasis in the transplant recipient. In affected patients, dose reduction or conversion to tacrolimus is optional. Several investigators have noted that coadministration of taurodeoxycholate and S-adenosyl-L-methionine to rats improved bile flow and bile acid secretion. These findings merit further investigation in cyclosporine-treated liver transplant recipients.

Tacrolimus
The influence of tacrolimus on aspects of hepatobiliary physiology has not been fully investigated. Kadmon et al found that like cyclosporine, tacrolimus inhibits canalicular ATP-dependent transport of bile acids, and Sanchez-Campos et al noted that at high doses, tacrolimus induces cholestasis by inhibiting primarily biliary excretion of glutathione. However, others have reported that tacrolimus improves recovery of bile secretion.

Azathioprine
Azathioprine is a purine analogue that is metabolized in the liver to 6-mercaptopurine by the enzyme glutathione S-transferase. Azathioprine causes inhibition of nucleic acid synthesis during the S-phase of the cell cycle. It suppresses T-cell-mediated immune reactions and, in large doses, suppresses B-cell functions. Azathioprine hepatotoxicity is mediated by damage to the hepatic sinusoidal and venular endothelial cells. There are numerous case reports of possible azathioprine-related liver damage, with a variety of clinical, biochemical, and histologic manifestations, including an asymptomatic increase in serum transaminase activity, veno-occlusive disease with portal hypertension, peliosis hepatis, and cholestasis. Azathioprine hepatotoxicity can present after liver transplantation as veno-occlusive disease and as nodular regenerative hyperplasia. In the latter case, all reported patients had cholestatic liver biochemistry, emphasizing the need for early identification of azathioprine-induced hepatotoxicity and consequent withdrawal of the agent.

Sulfonamides
Trimethoprim sulfamethoxazole is used after liver transplantation to prevent Pneumocystis carinii pneumonia. It rarely induces liver injury, and this reaction is usually considered idiosyncratic. The pattern of trimethoprim sulfamethoxazole hepatotoxicity is variable, but it is usually characterized by cholestasis or a mixed hepatocellular-cholestatic reaction. Vanishing bile duct syndrome has also been recently reported. Aerosolized pentamidine is an alternative for patients who are intolerant to trimethoprim sulfamethoxazole.

Late Intrahepatic Cholestatic Syndrome

Intrahepatic Biliary strictures
Intrahepatic biliary strictures develop several months to years after grafting. Main etiologic factors that should be considered are (1) Arterial occlusion (late
(2) Biliary injury from prolonged cold preservation: ischemic times in excess of 12 hours significantly increase the risk. (3) Rejection may be associated with strictures caused either directly by immunologic injury of the biliary tree or by arteriopathy (chronic rejection). (4) CMV infection in combination with rejection has been implicated in the development of intrahepatic biliary strictures. (5) Diffuse biliary strictures often present as part of the picture of biliary tree damage after an ABO-incompatible transplantation. However, intrahepatic strictures may occur in the absence of arterial thrombosis, rejection, or ABO-incompatible grafting, and the late appearance of intrahepatic strictures may indicate disease recurrence in patients who received a transplant for primary sclerosing cholangitis. The diagnosis is based on dilated intrahepatic bile ducts on ultrasound or computed tomography and cholangiography (percutaneous transhepatic cholangiography or endoscopic retrograde or magnetic resonance cholangiopancreatography; Fig. 2). However, ultrasound Doppler and arteriography should be performed to rule out hepatic artery thrombosis, and a liver biopsy should be performed to rule out chronic rejection, recurrent primary sclerosing cholangitis, and CMV infection. Diffuse strictures may represent either with the development of abnormal liver enzyme levels or, on occasion, with accompanying features of acute cholangitis. The management of intrahepatic biliary stricture is difficult, requiring balloon dilation and stenting and frequent endoscopic retrograde cholangiopancreatography, and has a success rate of only approximately 25%. The long-term outcome is relatively poor, and patients may require retransplantation (Table 3).

**Chronic Rejection**

Chronic or ductopenic rejection generally occurs within the first year after transplantation. In exceptional cases, it can be seen as early as 2 to 3 weeks postoperatively. Clinically, chronic rejection is suggested by the combination of progressively increasing serum levels of alkaline phosphatase, gamma-glutamyl transferase, and bilirubin. The current incidence rate is 2% to 5%. In most cases, ductopenic rejection is preceded by one or more episodes of acute rejection. Chronic ductopenic rejection is more likely in patients who undergo transplantation for primary sclerosing cholangitis, primary biliary cirrhosis, and autoimmune hepatitis. Other risk factors include lack of azathioprine (in the cyclosporine era), few histocompatibility leukocyte antigen A and B matches, and grafting a liver from a male donor into a female recipient. A close relationship has also been found between CMV infection and chronic rejection, as previously mentioned; one study reported a persistent CMV genome in the bile ducts of patients with chronic rejection.

Ductopenic rejection is characterized histologically by progressive destruction of the small (interlobular and septal) intrahepatic bile ducts. It has often been assumed that progressive intimal and subintimal infiltration of the second- and third-order branches of the hepatic artery with foamy macrophages accompanied by foam cells or obliterative arteritis (Fig. 3) results in arterial stenoses and, ultimately, in ischemic injury to...
the interlobular bile ducts and hepatocytes in zone 3 of the parenchyma. However, it was recently reported that direct immune-mediated damage is more likely responsible for the zone 3 necroinflammatory lesions frequently seen in the early stages of chronic rejection in association with the typical portal inflammatory infiltrates of acute cellular rejection.93 Treatment of chronic rejection involves conversion from cyclosporine-based immunosuppression to tacrolimus and the administration of mycophenolate mofetil and sirolimus. However, retransplantation may be required (Table 3).

**Hepatic Artery Thrombosis and Stenosis**

Hepatic artery thrombosis (HAT) occurs in approximately 7% of adult recipients of orthotopic liver transplants94,95 and 10% to 40% of pediatric recipients of primary liver grafts.96,97 The clinical presentation varies from a mild elevation of serum transaminase and bilirubin levels to fulminant hepatic necrosis. Early HAT results in massive injury to hepatocytes and ischemic bile duct epithelial cell injury, as the biliary branch of the hepatic artery provides the main blood supply to the biliary tree. In particular, ischemic damage to bile ducts may lead to dehiscence of the biliary anastomosis, secondary bile duct strictures, and intrahepatic bilomas or abscesses.98 HAT is associated with a high rate of allograft loss and patient mortality. Emergency revascularization is sometimes an effective means of hepatic allograft salvage; nevertheless, many patients eventually

**Table 3. Characteristics of Late Intrahepatic Cholestatic Syndromes After Liver Transplantation**

<table>
<thead>
<tr>
<th>Syndrome</th>
<th>Mechanism of Cholestasis</th>
<th>Clinical Presentation</th>
<th>Diagnosis</th>
<th>Treatment</th>
<th>Outcome</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intrahepatic biliary strictures</td>
<td>Hepatic artery thrombosis, preservation injury, ABO-incompatible, CMV infection, rejection, recurrent PSC</td>
<td>Increased cholestatic liver enzymes or cholangitis</td>
<td>Cholangiography</td>
<td>Balloon dilation and stenting, retransplantation</td>
<td>Significantly compromises graft survival, retransplantation</td>
</tr>
<tr>
<td>Chronic rejection</td>
<td>T-cell mediated nonsuppurative destructive cholangitis and ischemic interaction of inflammatory cytokines and atherosclerosis</td>
<td>Increased cholestatic liver enzymes and bilirubin</td>
<td>Histology: destruction of small intrahepatic bile ducts, obliterator arthritis</td>
<td>Conversion to tacrolimus, mycophenolate mofetil, sirolimus</td>
<td>Replantation</td>
</tr>
<tr>
<td>Hepatic artery thrombosis or stenosis</td>
<td>Ischemic injury to interlobular bile ducts and hepatocytes</td>
<td>Asymptomatic or cholangitis caused by biliary strictures and intrahepatic abscesses</td>
<td>Duplex ultrasound, angiography, cholangiography</td>
<td>Retransplantation</td>
<td>Death caused by sepsis/replantation</td>
</tr>
<tr>
<td>ABO blood type incompatibility</td>
<td>Donor ABH antigens expression on biliary epithelium, susceptible to immunologic injury</td>
<td>Cholangitis increased liver enzymes</td>
<td>ABO mismatch, cholangiography</td>
<td>Retransplantation</td>
<td>Decreased graft survival (44%)</td>
</tr>
<tr>
<td>Recurrent disease</td>
<td>Fibrosing cholestatic hepatitis B &amp; C: direct cytopathic role of the virus</td>
<td>Cholestatic liver enzymes, bilirubin, hepatic failure</td>
<td>Histology: ballooning, high nucleocapsid antigen load, cholestasis, fibrosis, viral load quantitation</td>
<td>HBV: lamivudine or adefovir, HCV: interferon and ribavirin, reduce immuno-suppression</td>
<td>Death caused by graft failure, Replantation Stable graft function</td>
</tr>
<tr>
<td>Primary biliary cirrhosis: immunologic: aberrant expression of E2 on bile ducts</td>
<td>Slightly increased cholestatic liver enzymes, or normal biochemistry</td>
<td>Histology: granulomatous bile duct damage</td>
<td>UDCA, conversion to cyclosporine</td>
<td>Retransplantation</td>
<td>Benign, may require retransplantation</td>
</tr>
<tr>
<td>Primary sclerosing cholangitis: immunologic</td>
<td>Slightly increased cholestatic liver enzymes, abnormal biochemistry</td>
<td>Cholangiography: histology fibrous cholangitis</td>
<td>No specific treatment</td>
<td>Benign, may require retransplantation</td>
<td></td>
</tr>
</tbody>
</table>

Abbreviations: CMV, cytomegalovirus; HBV, hepatitis B virus; HCV, hepatitis C virus; PSC, primary sclerosing cholangitis; PTC, percutaneous transhepatic cholangiography; UDCA, ursodeoxycholic acid.
require retransplantation.99,100 Late-onset HAT has a more benign course.101 Although often asymptomatic because of adequate portal vein flow or collateral circulation, in about 20% to 67% of patients, HAT is associated with progressive graft failure that requires retransplantation.16,102 In the latter patients, HAT is suspected because of worsening cholestatic biochemistry, or episodes of cholangitis because of nonanastomotic biliary strictures, intrahepatic abscesses, or bile leaks. Ischemic cholestasis may be found also in patients with hepatic artery stenosis. These patients may present with minimal biochemical liver enzyme elevations or with the development of nonanastomotic bile duct strictures associated with intrahepatic cholestasis.103 In whole liver allografts, the reported incidence of hepatic artery stenosis is rather low (5%). In segmental liver allografts, a higher incidence may be expected because of the smaller diameter of the arteries. Characteristic tardus parvus wave form on Doppler ultrasound is suspicious for hepatic artery stenosis.104 The final diagnosis is confirmed by arteriography (Table 3).

Donor-Recipient ABO Blood Type Incompatibility

Despite early reports suggesting that ABO-incompatible liver grafts could be transplanted without adverse results,105 there is increasing evidence of diminished graft survival, with rates varying from 19% to 75%.106-109 Clinically, affected patients show an increase in serum transaminase levels, hepatic failure, and coagulopathy during the first weeks after transplantation.106 The blood group–related ABH antigens are expressed in the epithelial cells of the intrahepatic biliary system.109 One study reported donor ABH antigen expression up to 150 days after transplantation, in association with a high incidence of late, severe, extensive biliary strictures (82%), hepatic artery complications (24%), decreased graft survival (44%), and acute cellular rejection.107 Thus, after transplantation across the ABO barrier, the biliary epithelium of hepatic allografts may be particularly susceptible to immunologic injury, with subsequent bile duct damage. Therefore, most centers restrict the use of ABO-incompatible grafts to emergencies when no other donor is available, although transplantation of blood group A2 livers into blood group O recipients is apparently safe and not associated with graft loss.110 The majority of patients who develop extensive biliary strictures require retransplantation (Table 3).

Recurrent Disease

Recurrent Viral Hepatitis B and C

In liver transplantation performed for chronic end-stage hepatitis B virus (HBV) infection, posttransplantation immunoprophylaxis with polyclonal antihepatitis B surface antigen immunoglobulin will prevent HBV reinfection in the graft in 64% to 80% of patients. The remainder will have aggressive HBV reinfection111 with rapid progression to cirrhosis.112 Fibrosing cholestatic hepatitis (FCH) is a severe form of HBV hepatitis, occurring as either a primary allograft reinfection after liver transplantation or as a severe HBV reactivation induced by immunosuppression in patients with a previously latent infection. Clinically, it presents as high serum bilirubin and liver enzyme levels and rapidly developing hepatic failure. Histologically, FCH is characterized by hepatocyte ballooning with cell loss and a mixed inflammatory infiltrate, high nucleocapsid antigen load, ductular proliferation, cholestasis, and immature fibrous tissue.113,114 It has been postulated that the cause of FCH is a high cytoplasmic expression of viral antigens, including hepatitis B surface antigen and hepatitis B core antigen.113 It is very rarely seen now thanks to improved strategies for preventing HBV reinfection in liver allografts. Treatment with lamivudine yielded good results,115 although progressively increasing rates of resistance to lamivudine have been reported over time, with rates of 27%, 40%, and >50% at 1, 2, and 3 years of therapy, respectively.116 Studies suggested that lamivudine-resistant posttransplantation patients have a relatively milder course, but the emergence of HBV mutants may lead to rapid development of liver failure in some cases. Adefovir, a nucleotide analogue of adenosine, seems to be a promising new anti-HBV agent: Adefovir treatment in a patient with FCH and lamivudine
dine resistance after liver transplantation was associated with a remarkable clinical, virologic, and histologic improvement. Its effectiveness in the transplant setting is currently under evaluation. Without treatment, FCH causes rapid graft failure and is always fatal within a few months of diagnosis.

Hepatitis C virus (HCV) infection is the leading indication for liver transplantation. Recurrence is almost universal and may be more aggressive than originally believed. A subgroup of patients with HCV develop FCH after liver transplantation, which is clinically and histologically similar to the FCH after liver transplantation for HBV, except that the inflammatory component is histologically detected in cholestatic HCV only. Factors that may influence disease progression and development of FCH include high serum levels of HCV RNA before transplantation, HCV genotype 1, severe damage detected in the first posttransplantation biopsy, and immunosuppression, which enhances divergence of the HCV quasispecies and results in the emergence of new variants. However, other reports showed that the posttransplantation quasispecies pattern remains stable over time in patients with recurrent cholestatic hepatitis, suggesting that viral escape from immune pressure may play a pathogenic role. As for HBV, a very high expression of HCV proteins in the liver graft was recently postulated to be linked to the development of FCH. Other causes of cholestasis (biliary strictures, CMV, sepsis) should be excluded. Treatment of FCH is difficult in patients with HCV. Early use of a combination of interferon and ribavirin might be considered, but data are limited. Retransplantation may be life-saving.

Recurrent Cholestatic Disease

Primary biliary cirrhosis. The recurrence of primary biliary cirrhosis (PBC) after transplantation is now well recognized, with rates of 8% to 16% at 1 to 6 years postoperatively and approaching 50% at 10 years. The diagnosis of PBC recurrence is based on histologic graft findings of bile duct damage, ductopenia, bile duct proliferation, and portal granuloma (Fig. 4). Clinically, it can be asymptomatic; total serum bilirubin level and hepatic biochemistry are normal, although serum alkaline phosphatase may be slightly increased. Risk factors remain unclear but probably include histocompatibility leukocyte antigen or sex mismatching between donor and recipient and immunosuppression. Dmitrewski et al reported that histologic features characteristic of PBC were found more commonly and earlier after transplantation in patients receiving FK506 than in those given cyclosporine. Because UDCA has been found to be effective in the management of patients with symptomatic PBC, it seems sensible to offer it also to patients with evidence of recurrent PBC to slow disease progression (Table 3).

Primary sclerosing cholangitis. The recurrence rate for primary sclerosing cholangitis (PSC) after liver transplantation is higher than for PBC, ranging from 8.6% to 25%. Clinically, it may present as an increase in liver enzymes or fever and cholangitis. The diagnosis is based on cholangiographic findings of intrahepatic and/or extrahepatic biliary stricturing, beading, and irregularity, and on histologic findings of fibrous cholangitis and/or fibro-obliterative lesions with or without ductopenia, biliary fibrosis, or biliary cirrhosis (Fig. 5). Both cholangiography and histology are necessary owing to the histologic similarity of recurrent PSC and chronic rejection. Biliary strictures, both

Figure 4. Recurrent primary biliary cirrhosis: damaged medium-sized bile duct surrounded by dense mononuclear cell infiltrates, accompanied by epithelioid macrophages forming an ill-defined granuloma. There was intraepithelial invasion by lymphocytes.

Figure 5. Recurrent primary sclerosing cholangitis: large bile duct showing inflammation and fibrosis.
anastomotic and nonanastomotic, are frequent, occurring in 16.2% and 27.2% of patients, respectively.\textsuperscript{136} No specific clinical risk factors for recurrent PSC have been identified, and studies report no difference in overall patient and graft survival between those with and without recurrent PSC.\textsuperscript{134} No specific treatment is indicated in patients with PSC. Retransplantation is sometimes required (Table 3).

**Summary**

In summary, cholestasis is a common sequela of liver transplantation. Various processes occurring after the transplantation interfere with bile flow, uptake, transfer, and secretion or directly cause epithelial ductular and bile duct damage. The histologic picture is characterized by the accumulation of bile in liver cells and biliary passages. Cholestasis may be categorized by time of occurrence: early (within 6 months of liver transplantation), caused by ischemia/reperfusion injury, bacterial infection, small-for-size-graft, acute cellular rejection, CMV infection, and drugs; and late (after 6 months), caused by intrahepatic biliary strictures, chronic rejection, hepatic artery thrombosis, ABO incompatibility, and recurrent disease. The causes may overlap. However, the mechanisms of cholestasis in each category differ, as do the clinical presentation, diagnosis, treatment, and outcome.

Once risk factors are identified, prompt action is needed—either retransplantation or initiation of pharmacological agents such as UDCA, which appears to displace endogenous bile acids and thus reverses their suspected toxicity.\textsuperscript{137} Moreover, according to preliminary trials, prophylactic UDCA administration in the early postoperative period may increase bile flow, decrease total bile acid output, increase the composition of bile increases by the portion of UDCA, and reduce ALT level.\textsuperscript{138} When added to a cyclosporine-based immunosuppression regimen, UDCA significantly decreased rejection episodes and improved 90-day and 1-year survival.\textsuperscript{139} However, these findings have not been confirmed. More controlled multicenter studies are needed to assess the role of UDCA in cholestatic syndromes after liver transplantation.

Given that more than one mechanism is often involved, recognition of the primary cause for cholestasis with appropriate treatment might help reverse this process. In some cases, however, progressive hepatocyte and epithelial cell damage may lead to irreversible functional loss of bile formation and bile flow, requiring retransplantation.

**References**


