Novel Approaches to the Treatment of Biliary Atresia

Yang Li, M.D., and Jorge A. Bezerra, M.D.

Biliary atresia remains the most common cause of end-stage cirrhosis in children and the main indication for pediatric liver transplantation. With an estimated incidence of ~1 in 15,000 live births in the United States, the care of infants with biliary atresia represents a remarkable challenge in clinical practice with limited therapeutic options.

PRESENTATION AND DIAGNOSIS

The hallmark of disease is the onset of jaundice in the first 4 to 6 weeks of life in an infant who generally lacks other symptoms. Because physiological and breast milk jaundice is common in neonates (reflected by an increase in serum indirect/unconjugated bilirubin), it is important to fractionate serum bilirubin levels in any jaundiced infant beyond 3 weeks of age so that infants with a direct or conjugated bilirubin ≥ 2 mg/dL or $\geq 20\%$ of the total bilirubin can be evaluated for biliary atresia.¹ The presence of acholic/pale stools and variable levels of hepatosplenomegaly should trigger a prompt evaluation with

comprehensive liver function and injury tests and ultrasound of liver, gallbladder, and spleen. Because of the higher incidence of alpha-1-antitrypsin deficiency in neonates with cholestasis, it is prudent to demonstrate the absence of homozygous Z alleles before pursuing a liver biopsy and intraoperative cholangiogram. However, every effort should be made not to delay a final diagnosis because the outcome of the hepatoportoenterostomy (HPE) is better when the procedure is performed in young infants.²

INFLUENCE OF CLINICAL PHENOTYPES ON OUTCOME

The natural history of the progressive disease can be modified by HPE (Fig. 1). If the procedure does not result in adequate drainage, most patients require liver transplantation within 2 years of age.³ When successful, HPE is followed by normalization of serum bilirubin levels within 3 to 6 months, but even with improved biliary drainage the fibrotic liver disease progresses to end-stage

Abbreviations: BA, biliary atresia; DC, dendritic cell; HPE, hepatoportoenterostomy; IFN- γ , interferon gamma; IL, interleukin; LT, liver transplant; NK, natural killer cell; RRV, rhesus rotavirus type A; SNL, survival with native liver.

From the Liver Care Center and Division of Gastroenterology, Hepatology and Nutrition, Cincinnati Children's Hospital Medical Center; and Department of Pediatrics, University of Cincinnati College of Medicine, Cincinnati, OH.

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FIG 1 Flow diagram on natural history of biliary atresia. Data from different publications are integrated to demonstrate the outcomes at 3 months after HPE and 2, 5, and 10 years of age. The range of percentage is based on different publications and does not necessarily add up to 100%. Abbreviations: BA, biliary atresia; LT, liver transplant; SNL, survival with native liver.

cirrhosis in early childhood. Among key factors influencing the response to HPE are clinical forms and phenotypic variants of disease. Between the two most widely accepted forms, the nonsyndromic (or perinatal) is the most common form and is more likely to have a better outcome.⁴ In contrast, neonates with the syndromic (or embryonic) form are diagnosed at a younger age and have congenital nonhepatic malformation, such as splenic malformations with or without cardiovascular and laterality defects. Despite their younger age at diagnosis, this form of biliary atresia has been linked to worse outcome.

Phenotypic variants that may influence the response to HPE include the cystic biliary atresia, which is characterized by the presence of a cystic malformation of the common bile duct near the site of obstruction. Patients with the cystic variant may have improved bile drainage after HPE, especially when performed in younger infants. A second variant is biliary atresia associated with cytomegalovirus (CMV). Recent reports preliminarily establish a link between CMV detection and poor outcome in patients with biliary atresia, with less than 10% survival with the native liver by 2 years of age.⁵

FIG 2 Cellular and molecular basis of bile duct injury and obstruction in experimental biliary atresia. Upon insult of RRV or a toxic agent, dendritic cells (DCs) release IL-15 to activate natural killer (NK) cells, which in turn release interferon gamma (IFN- γ), perforin, and granzymes that promote epithelial injury of bile ducts. CD8⁺ T lymphocytes also undergo activation, release effector cytokines, and promote an inflammatory obstruction of the duct lumen. These initial events are followed by fibrosis of extrahepatic bile ducts.

Although controversial, the center experience with HPE and management of affected patients may influence the outcome. Among the retrospective studies supporting this concept are the report from United Kingdom that showed a better 5-year survival rate with the native liver in centers performing ≥ 5 HPE annually and the report from France that underscored a collaboration among

centers as a potential strategy to achieve better HPE outcome.^{6,7}

Regardless of the clinical form or variant, infants with biliary atresia are at increased risk for malnutrition, ascending cholangitis, and complications of portal hypertension. When these complications are not responsive to treatment and/or progressive cholestasis develops, liver transplantation is the best means to improve long-term outcome, with an overall survival rate beyond 5 years approaching 82%.⁸ This improved posttransplant survival, however, is associated with increased morbidity, long-term exposure of immunosuppression, and notable health care cost. These can be avoided by improved diagnostics and the development of new effective therapies.

LESSONS FROM BASIC RESEARCH

The combination of well-designed observational reports, analyses of diseased tissues, and studies in experimental models of neonatal biliary injury are shedding light into pathogenic mechanisms of disease. Among all proposed mechanisms, inflammation and immune circuits emerge in analyses of liver tissues and in the mouse model of experimental atresia induced by the neonatal infection with rhesus rotavirus type A (RRV).⁹ Mechanistic studies using the genetic inactivation of specific genes and cell depletion strategies uncover a continuum of biological events focused on extrahepatic bile ducts that begin with a viral infection (e.g., rotavirus) that targets the bile duct epithelium and primes macrophages and dendritic cells (“initiating” phase). The secretion of interleukin (IL)-15 and other soluble mediators activates NK cells, which injure cholangiocytes, disrupt the epithelial integrity (phase of epithelial injury), and trigger an adaptive immune response by CD4⁺ and CD8⁺ T cells. What follows is an inflammatory plug that

TABLE 1. FUTURE AREAS OF RESEARCH AND CLINICAL TRIAL DESIGN IN BILIARY ATRESIA

Areas of Interest	Approaches
Molecular signatures of phenotypes	Search for biomarkers that identify individual clinical forms and variants of biliary atresia
Staging of disease	Validate histological and molecular scoring systems to stage liver disease at diagnosis (inflammation and/or fibrosis)
Etiology of disease	Use of novel detection technologies to search for viruses, toxins and genes that cause the disease or influence clinical outcome
Investigation of pathogenesis	Define cellular and molecular immune response, mechanisms of cholangiocyte injury, role of microbiome
Novel clinical trials	Design trials that control for clinical forms and variants, and target the predominant biological makeup at the time of diagnosis

FIG 3 Designing new therapies that target predominant biological processes at diagnosis. Staging of the liver disease at diagnosis using histopathology or molecular profiling might identify subgroups of patients with predominant inflammation (red) or fibrosis (blue), or with mixed features (gray) at diagnosis. Targeting either inflammation or fibrosis pathways may yield better outcomes and block progression of disease.

disrupts bile flow, extends the epithelial injury (Fig. 2), and rapidly evolves to duct fibrosis. A current challenge is to critically review the evidence and select initial best strategies for translational studies.

CORTICOSTEROIDS AND FUTURE CLINICAL TRIALS

If inflammatory cells and immune genes are activated in the liver at the time of diagnosis and experimentally drive epithelial injury and duct obstruction, it would make sense to consider anti-inflammatory agents as an adjuvant treatment to HPE. The most frequently used agent is corticosteroids, but it has not been shown to consistently improve biliary drainage after HPE.¹⁰ In the prospective, placebo-controlled, double-blind START trial (steroid in biliary atresia randomized trial), conducted by the Childhood Liver Disease Research Network (ChIL-DReN), high-dose corticosteroids after HPE did not significantly increase the proportion of patients achieving biliary drainage when compared with placebo (steroid group: 58.6% versus placebo: 48.6%; $P = 0.43$) or transplant-free survival (58.7% versus 59.4%; $P =$

0.99).¹¹ Interestingly, corticosteroids improved short-term outcome in an open-label trial that included patients younger than 70 days and without splenic malformations.¹²

It remains to be determined whether future trials in a large cohort with these clinical characteristics will reproduce these findings (Table 1). In addition, future trials might include a molecular (and perhaps histological) staging of the liver disease at diagnosis,⁹ either targeting predominantly inflammation pathways or antifibrotic strategies (Fig. 3). The evaluation of these innovative, targeted trials may substantially improve the future outcome following HPE in children with biliary atresia.

CORRESPONDENCE

Jorge A. Bezerra, M.D., Division of Gastroenterology, Hepatology and Nutrition, Cincinnati Children's Hospital Medical Center, 3333 Burnet Avenue, Cincinnati, OH 45229. E-mail: jorge.bezerra@ccmc.org

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