High-Dose Steroids, Ursodeoxycholic Acid, and Chronic Intravenous Antibiotics Improve Bile Flow After Kasai Procedure in Infants With Biliary Atresia

By Rebecka L. Meyers, Linda S. Book, Molly A. O'Gorman, W. Daniel Jackson, Richard E. Black,
Dale G. Johnson, and Michael E. Matlak
Salt Lake City, Utah

Background/Purpose: Early reports suggest that the use of steroids after Kasai portoenterostomy may improve bile flow and outcome in infants with biliary atresia.

Methods: Of 28 infants with biliary atresia, half received adjuvant high-dose steroids, and half received standard therapy. Infants in the steroid group (n = 14) received intravenous solumedrol (taper of 10, 8, 6, 5, 4, 3, 2 mg/kg/d), followed by 8 to 12 weeks of prednisone (2 mg/kg/d). The steroid protocol also included ursodeoxycholic acid indefinitely and intravenous antibiotics for 8 to 12 weeks followed by oral antibiotic prophylaxis. Infants in the standard therapy group (n = 14) received no steroids, occasional ursodeoxycholic acid, and perioperative intravenous antibiotics followed by oral antibiotic prophylaxis. The infants were not assigned randomly, but rather received standard therapy or adjuvant steroid therapy according to individual surgeon preference.

Results: Eleven of 14 (79%) in the steroid group and 3 of 14 (21%) in the standard therapy group had a conjugated bili-

rubin level less than 1.0 within 3 to 4 months of surgery (P < .001). Fewer patients in the steroid group (21% v 85%) required liver transplantation or died during the first year of life (P < .001). Infants in the steroid group did better despite the fact that this group included 5 infants with biliary atresia-polysplenia-heterotaxia syndrome, a subgroup that might have been expected to have a poor prognosis. Neither bile duct size nor liver histology was a reliable predictor of success or failure in either group.

Conclusions: Adjuvant therapy using high-dose steroids, ursodeoxycholic acid, and intravenous antibiotics may accelerate the clearance of jaundice and decrease the need for early liver transplantation after Kasai portoenterostomy. J Pediatr Surg 38:406-411. Copyright 2003, Elsevier Science (USA). All rights reserved.

INDEX WORDS: Biliary atresia, Kasai portoenterostomy, corticosteroids, liver transplantation, polysplenia-heterotaxia syndrome.

N 1959, Kasai and Susuki developed the hepatic portoenterostomy procedure to drain the abnormal biliary system in children with biliary atresia. Although a successful Kasai portoenterostomy may restore bile drainage, long-term success of the Kasai procedure historically has been achieved in a minority of patients, and recent reports estimate that as many as 70% to 80% ultimately will require a liver transplant or die secondary to progression of their disease. Improvements in pediatric liver transplant outcomes have helped improve the overall prognosis in biliary atresia, and today the strategy of sequential surgical therapy for biliary atresia involv-

ing an initial Kasai procedure followed by liver transplantation, when necessary, is now broadly accepted.

Many previous attempts to improve outcome of the Kasai procedure have centered on technical details of the operation such as the extent of debridement at the biliary plate and variations in the approach to construction of the Roux-en-Y jejunal limb.5 At the 2000 meeting of the American Pediatric Surgical Association it was agreed we have gone about as far as we can by revising, adjusting, and otherwise modifying Dr Kasai's operation. To make an impact on the outcome of infants undergoing portoenterostomy for biliary atresia, efforts to gain insight into the pathogenesis of the disease would help us refine our therapies.⁶ The fundamental problem with biliary atresia is that we do not fully understand the underlying disease process. It appears to be an acquired condition; viral infection, genetic predisposition, abnormal bile acid metabolism, and ductal plate malformations all have been suggested as possible inciting events.7 Immunologic, inflammatory, infectious, and obstructive pathways of progressive intrahepatic biliary epithelilal destruction and portal fibrosis all have been hypothesized.8-14 The Kasai procedure, to the extent possible,

From the Divisions of Pediatric Surgery and Pediatric Gastroenterology, Primary Children's Medical Center, University of Utah School of Medicine, Salt Lake City, UT.

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Address reprint requests to Rebecka L. Meyers, MD, Division of Pediatric Surgery, Primary Children's Medical Center, University of Utah School of Medicine, 100 N Medical Dr, Salt Lake City, UT 84113.

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attempts to address the extrahepatic obstruction. Unfortunately, the transected microscopic ducts on which post-operative bile flow depends are prone to postoperative inflammatory closure.

Medical therapies must be sought in an attempt to further ameliorate the obstructive process and to address the ongoing immunologic, inflammatory, and infectious components of this disease. In this study we compare the outcome of infants with biliary atresia who either did or did not receive postoperative drug therapy, which included high immunosuppressive and antiinflammatory doses of steroids, a prolonged course of intravenous antibiotics, and the choleretic, ursodeoxycholic acid.

MATERIALS AND METHODS

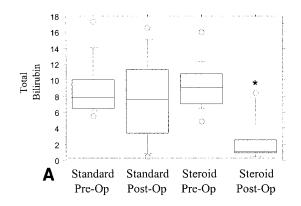
We performed a retrospective review of infants treated for biliary atresia during the past 8 years at University of Utah, Primary Children's Medical Center. Of 28 consecutively treated infants, half received "standard treatment" and half received the "steroid protocol" consisting of high-dose steroids, long-term intravenous antibiotics, and ursodeoxycholic acid. The steroid protocol was used, or not used, based on the personal preference of the attending pediatric surgeon. Starting at the time of surgery, infants in the steroid group (n = 14) were treated with the same initial steroid induction protocol that we have used in our liver transplant patients. This consists of intravenous methylprednisolone (taper of 10, 8, 6, 5, 4, 3, 2 mg/kg/d) followed by 8 to 12 weeks of oral prednisone (2 mg/kg/d). Each dose of intravenous methylprednisolone is given one time q24, so that the initial intravenous "pulse" of tapering doses lasts 7 days. The steroid protocol included intravenous antibiotics for 8 to 12 weeks (either piperacilllan/tazobactam, 300 mg/kg/d, divided q6, plus gentamycin, 5 mg/kg/d q 24, or cefoperazone 150 mg/kg/d divided q8) followed by oral trimethoprim/sulfamethoxazole (10 mg/kg/d divided q12). The steroid protocol also included oral ursodeoxycholic acid indefinitely (20 mg/kg/d divided q12).

Infants in the standard therapy group (n = 14) received no steroids, 3 to 4 days of perioperative intravenous antibiotics (ampicillin, 200 mg/kg/d divided q6, plus gentamicin, 7.5 mg/kg/d divided q8), followed by oral trimethoprim/sulfamethoxazole (10 mg/kg/d divided q12). Two patients in the standard therapy group received a short course of oral prednisone (2 mg/kg/d for 4 days), and a variable course of ursodeoxycholic acid.

Patient charts were reviewed for the presence or absence of the "steroid protocol," age at Kasai portoenterostomy, incidence of perioperative complications, serial determinations of total and conjugated bilirubin levels, incidence of cholangitis, incidence of associated anomalies and subsequent operations, biliary plate and liver biopsy histology (including bile duct proliferation, focal v. bridging hepatic fibrosis, size of remnant biliary radicals seen at the biliary plate), referral for liver transplant, and death. Data for the 2 groups is compared using Student's paired t test and χ^2 analysis. Values are expressed as mean \pm SEM. Significance is set at the 95% confidence interval, P < .05. This study was approved by the Institutional Review Board at the University of Utah, IRB 10259.

RESULTS

A total of 28 infants were treated with Kasai portoenterostomy. Fourteen infants received standard therapy, and 14 received the steroid protocol. One patient in the standard therapy group was lost to follow-up after 6 months and is included in the analysis of the postopera-



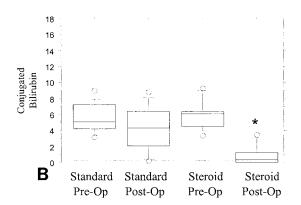


Fig 1. Box-plot distribution of serum (A) total and (B) conjugated bilirubin levels (mg/dL) measured preoperative and 3 to 4 months postoperatively. The postoperative levels in the steroid group are significantly lower than those in all other categories (*P < .001).

tive bilirubin levels but not in the analysis of long term outcome. Adherence to the steroid protocol was very good with the following minor variations: 3 children received equivalent doses of intravenous dexamethasone rather than methylprednisolone; in one child, the steroids were not started until postoperative day 2. All but 2 infants were less than 12 weeks of age at the time of Kasai procedure; one was 13 and one 16 weeks of age. Both of the children operated on after 12 weeks of age were in the steroid protocol group. All surgeons used a similar surgical technique. Six children in the standard therapy group had a Roux-en-Y limb constructed with an intussuscepting valve; the remainder of the patients in both groups received 40 cm retrocolic Roux-en-Y limbs.

The distribution of the total and conjugated bilirubin levels for each treatment group is shown in the box-plots in Fig 1. These bilirubin levels were measured preoperatively and 3 to 4 months postoperatively. Eleven of 14 (79%) in the steroid group, and 3 of 14 (21%) in the standard therapy group, had a conjugated bilirubin levels less than 1.0 within 3 to 4 months of surgery (P < .001).

Fewer patients in the steroid group (21% v 85%)

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Table 1. Outcome of Infants Receiving Standard Therapy Versus the Steroid Protocol

	Standard Therapy	Steroid Protocol
No. of infants	13	14
Liver transplant, death, or both		
<1 yr of age	11 (85%)	3 (21%)*
Overall survival rate	5 (38%)	13 (92%)†
Death <1 yr of age	7 (53%)	1 (7%)†
Mean follow-up of survivors	6.9 yr	3.8 yr
Liver transplant		
<1 yr of age	8 (61%)	3 (21%)*
>1 yr of age	1 (8%)	1 (7%)
Jaundice free with native liver	1 (8%)	10 (71%)†

NOTE. One patient in the standard-therapy group was lost to follow-up at 6 months.

required liver transplantation or died during the first year of life (P < .001). One child in the standard therapy group was lost to follow-up at 6 months of age when the family moved out of state; mean follow-up for all other children was 3.8 years. Ten children (71%) in the steroid group are jaundice free with their native liver, whereas only one child (8%) in the standard therapy group has undergone a truly successful Kasai procedure (Table 1). There were no reoperations in either group, although it is clear, in retrospect, that one child in each group might have benefited from reoperation when good initial bile drainage ceased after an episode of cholangitis.3,15 Infants in the steroid group did better despite the fact that this group included 5 infants with biliary atresiapolysplenia-heterotaxia syndrome, a subgroup that might have been expected to have a poor prognosis. Additional diagnoses included one child with choledochal cyst in the standard therapy group and one child with a preoperative subdural hemorrhage in the steroid group. Inguinal hernias were present in 3 children in each group.

Age at operation did not seem to have a significant effect on outcome (Fig 2). Neither bile duct size nor liver histology was an independent predictor of success or failure in either group (Table 2). Cholangitis developed in 6 patients (43%) in the standard therapy group and 4 (28%) in the steroid protocol group. Despite the very high doses of steroids used, we did not identify any specific steroid complications other than fluid retention and increased appetite. Surgical complications in the standard therapy group included one wound infection, one drain site hernia, and 2 children with prolonged postoperative ascitic drainage. Surgical complications in the steroid group included one child with line sepsis and 3 children with prolonged postoperative ascitic drainage.

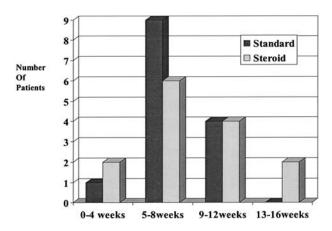


Fig 2. Age at time of Kasai portoenterostomy. There were no significant differences in age at operation between the 2 groups.

DISCUSSION

Steroids have been used to augment antibiotic treatment of refractory cases of postoperative cholangitis in biliary atresia patients for more than 20 years. ¹⁵ Karrer and Lilly ¹⁶ subsequently proposed using very high-dose "blast"-type steroids, citing potential choleretic and antiinflammatory properties that might benefit the child with cholangitis. The choleretic effect of steroids involves induction of Na⁺-K⁺ ATPase, which increases canalicular electrolyte transport and stimulates bile flow independent of the bile salt concentration. ¹⁷ In addition, when given in high doses, steroids have pronounced antiinflammatory and immunosuppressive effects decreasing edema and collagen deposition, inhibiting scarring, and arresting migration of infiltrating monocytes and lymphocytes.

Although the etiology of neonatal biliary atresia is not known, the disease is characterized by a progressive sclerosing and inflammatory process causing atresia of all or part of the extrahepatic biliary system and rapidly extending to involve the intrahepatic ducts. It has been hypothesized that this inflammatory process is part of an immunologic reaction, based on a liver histologic appearance similar to that seen in graft versus host disease,

Table 2. Histology of Liver and Biliary Plate

	Jaundice Free With Native Liver		Death or Liver Transplant	
	Standard	Steroid	Standard	Steroid
No. of infants	1	10	11	3
Size of ductule at				
biliary plate*	50 μ m	49 \pm 8 μ m	80 \pm 15 μ m	66 \pm 16 μ m
Hepatic histology				
Bridging fibrosis	_	3	2	1
Focal fibrosis	1	7	9	2
Giant cell				
transformation	_	5	4	2

^{*(}smallest ductule + largest ductile)/2, mean \pm SEM.

^{*}*P* < .01.

[†]*P* < .001.

after bone marrow transplant, and in acute cellular rejection after liver transplant.8 Liver biopsy in biliary atresia classically shows bile duct proliferation, canalicular stasis, swelling and vacuolization of bile duct epithelial cells, portal tract edema and fibrosis, and monocytic and lymphocytic cell infiltration of the portal tracts.8,18 A number of cellular inflammatory markers have been studied including CD14-positive macrophages in the monocytic infiltrate, which, when activated by endotoxin, secrete a number of inflammatory cytokines into the periductular tissue.11 Chemokines including interleukin-8 and monocyte chemoattractant protein-1 have been identified in the inflammatory infiltrate of cholangitis.¹⁹ Expression of intracellular adhesion molecule-1 (ICAM-1) by the bile duct epithelium may play a role in the recruitment of lymphocytes.²⁰ Human leukocyte antigen (HLA-DR) expression is increased around the ductal epithelium,21 which induces a cytotoxic lymphocytic attack of the bile duct epithelial cells.²² The inflamed bile duct epithelial cells become a rich source of transforming growth factor (TGF)- β , thereby stimulating transcription of collagen type 1 genes in the surrounding hepatic stellate (Ito) cells and leading to periductular fibrosis.¹⁸

In an attempt to ameliorate the progressive intraheptic periportal inflammatory process, Muraji and Higashimoto¹² reported the use of steroids as an adjuvant medical treatment after Kasai portoenterostomy. An initial steroid dose of 4 mg/kg/d was tapered to 2 mg/kg/d over one to 2 weeks, and repeat "pulse" dose steroids were given if bile drainage tapered or cholangitis ensued. Ten of 14 patients (71%) became jaundice free without liver transplantation compared with a 30% survival rate of the 10 patients treated before implementation of the adjuvant steroid regimen. Extending on this work, Dillon et al⁶ were the first to advocate long-term high-dose steroids and achieved equally impressive results reporting 19 of 25 patients (76%) to be jaundice free with their native liver at a mean follow-up of 4 years. In the current study, our initial "pulse" dose of steroids is higher than that used by Dillon et al, with a similar 2 mg/kg/d dose continued for several weeks. Our results are comparable with 10 of 14 patients (71%) achieving excellent and sustained bile flow. The safety of steroid therapy in this patient population has been shown clearly in all 3 of these studies.^{6,12}

However, proof that the improved outcome is caused primarily by the steroids remains elusive, because none of these studies have been randomized or rigorously controlled for a host of potential confounding variables, and none of the studies are of sufficient size to be empowered to definitively answer the question of causality. Although the surgical technique used by all surgeons in this study is reportedly the same, the majority of operations in each group were performed by different

surgeons, and subtle differences in technique cannot be excluded.

Although the similarity of the results of these 3 studies using adjuvant steroids is striking, namely success of the Kasai procedure in 71%, 76%, and 71%, respectively, there are some key differences in the methodology. Like Dillon et al⁶ our steroid doses are higher and used longer than those used by Muraji and Higashimoto. Unlike both Dillon et al and Muraji and Higashimoto, we used a more aggressive long-term approach to intravenous antibiotics, continuing them for the duration of the high-dose steroid protocol. Comparison of the outcomes suggests that our aggressive use of antibiotics probably did not yield superior results; the results of all 3 studies are equivalent. Because none of these studies are large, controlled, or randomized, the conclusion can only be inferred.

Steroids are felt to offer both choleretic and antiin-flammatory benefits. Many other choleretic agents have been tried in post-Kasai patients including intravenous 10% dehydrocholic acid, glucagon, prostaglandin E2, and ursodeoxycholic acid.^{5,23} In the current study, we used a dose and duration of ursodeoxycholic acid similar to the method of Dillon et al.⁶ Because all children in the steroid group received both steroids and ursodeoxycholic acid, we do not know if the 2 agents had an additive effect. Ursodeoxycholic acid has been shown to significantly improve essential fatty acid deficiencies in infants after Kasai portoenterostomy and has shown a tendency toward decreasing bilirubin levels.²³

We did not find any correlation between the microscopic size of the remnant biliary ductules or the degree of hepatic fibrosis and the probability of postoperative bile flow,^{24,25} nor did we find any significant correlation between the age at which the Kasai was performed and the probability of success. Larger studies, more appropriately empowered to answer such questions, have shown that the outcome of the Kasai procedure usually is improved when performed before 10 to 12 weeks of age.6,10,26,27 In this study, we performed a Kasai on 2 children greater than 12 weeks of age, both of whom received the adjuvant steroid protocol. Both had excellent early bile flow. The 13-week-old patient eventually lost bile flow several months later. The 16-week-old patient remains jaundice free with his native liver in follow-up of more than 2 years. Others have suggested that the outcome may be worse for children who undergo the Kasai before 4 weeks of age²⁸ and have postulated a more severe form of the disease in the youngest infants. Unfortunately, it remains difficult to predict accurately which children will benefit from the Kasai procedure. The latest predictive attempts advocate the measurement of serum markers such as hyaluronic acid and plasma endothelin-1.29,30

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The steroid group in this study includes 5 children with biliary atresia-polysplenia-heterotaxia syndrome, 4 of whom were treated in a cluster of 6 months raising interesting epidemiologic questions. There has been a widely held, but largely unsubstantiated, belief that the Kasai operation is likely to fail in infants with this syndrome. However, a recent series reported 44% actuarial survival rate at 5 years in the patients with biliary atresia-polysplenia-heterotaxia syndrome, a survival rate that was slightly lower, but not significantly different, than the remainder of the biliary atresia patients in that study.³¹ In this study, 4 of the 5 children with this syndrome had excellent bile flow for several months. Two of 5 ultimately required liver transplant, and the remaining 3 continue to do well with their native livers and without jaundice.

The clinical success of the Kasai procedure often is judged by the requirement for liver transplantation, but a more elusive yardstick is the overall health of the child. Although in the current study the overall survival rate of the standard therapy group was disturbingly low, liver transplant outcomes in these patients have since improved remarkably. It is now generally accepted that the overall survival rate of a patient with a failed Kasai procedure followed by transplantation should approach

80% in experienced centers.^{2,3,10} It is interesting to speculate on the potential relative "health" of a child with a failed Kasai procedure and subsequent successful liver transplant compared with a child with a successful Kasai and retention of the native, but fibrotic, liver with the attendant possibility of progressive liver failure and portal hypertension. Miyano et al³² has shown that although about 70% of his patients had evidence of bile secretion immediately after surgery, only 32% remained that way after 5 years.32 Yet, the successfully treated adjuvant steroid patients of Dillon et al⁶ seemed to maintain their bile flow at 5 years. Average follow-up of successfully treated patients in this study is 3.8 years, and no patient has had significant growth retardation or portal hypertensive bleeding to date. The longer-term potential salutary effects of the steroid treatment on progression of hepatic fibrosis and portal hypertension are not yet known.

We do not know whether the beneficial effects of the adjuvant steroid therapy are caused by the choleretic, immunosuppressive, or antiinflammatory effects of the steroids. But given the consistently promising results of the adjuvant steroid studies published to date, a multicenter, randomized, controlled clinical trial is badly needed.

REFERENCES

- 1. Aronson DC, de Ville de Goyet J, Francois D, et al: Primary management of biliary atresia: don't change the rules. Br J Surg 82:672-673, 1995
- 2. Davenport M, Kerker N, Mieli G, et al: Biliary atresia: The King's College hospital experience (1974-1995). J Pediatr Surg 32: 479-485, 1997
- 3. Ryckman FC, Alonso MH, Cucuvalas JC, et al: Biliary atresia— Surgical management and treatment options as they relate to outcome. Liver Transplant Surg 4:S24-33, 1998
- 4. Karrer FM, Hall RJ, Lilly JR: Biliary atresia and the polysplenia syndrome. J Pediatr Surg 26:524-527, 1991
- 5. Ohi R, Hanamatsu M, Mochizuki I, et al: Progress in the treatment of biliary atresia. World J Surg 9:285-293, 1985
- 6. Dillon PW, Owings E, Cilley R, et al: Immunosuppression as adjuvant therapy for biliary atresia. J Pediatr Surg 36:80-85, 2001
- 7. Nio M, Ohi R: Biliary atresia. Semin Pediatr Surg 9:177-186, 2000
- 8. Ohya T, Fujimoto T, Shimormura H, et al: Degeneration of intrahepatic bile duct with lymphocyte infiltration into biliary epithelial cells in biliary atresia. J Pediatr Surg 30:515-518, 1995
- 9. Haas JE: Bile duct and liver pathology in biliary atresia. World J Surg 2:561-569, 1978
- 10. Altman RP, Lilly JR, Greenfield J, et al: A multivariate risk analysis of the portoenterostomy (Kasai) procedure for biliary atresia. Ann Surg 226:348-355, 1997
- 11. Tracy TF, Dillon P, Fox ES, et al: The inflammatory response in pediatric biliary diseases: Macrophage phenotype and distribution. J Pediatr Surg 31:121-126, 1996
- 12. Muraji T, Higashimoto Y: The improved outlook for biliary atresia with corticosteroid therapy. J Pediatr Surg 32:1103-1107, 1997

- 13. Nietgen GW, Vacanti JP, Perez-Atayde AR: Intrahepatic bile duct loss in biliary atresia despite portoenterostomy: A consequence of ongoing obstruction. Gastroenterol 102:2126-2133, 1992
- 14. Lunzmann K, Schweizer P: The influence of cholangitis on the prognosis of biliary atresia. Eur J Pediatr Surg 9:19-23, 1999
- 15. Altman RP, Anderson KD: Surgical management of intractable cholangitis following successful Kasai procedure. J Pediatr Surg 17: 894-900, 1982
- 16. Karrer FM, Lilly JR: Corticosteroid therapy in biliary atresia. J Pediatr Surg 20:693-695, 1985
- 17. Miner PB, Gaito JM: Bile flow in response to pharmacologic agents: Hepatic DNA as a reference standard. Biochem Pharmacol 28:1063-1066, 1979
- 18. Ramm GA, Nair VG, Bridle KR, et al: Contribution of hepatic parenchymal and nonparenchymal cells to hepatic fibrogenesis in biliary atresia. Am J Pathol 153:527-535, 1998
- 19. Hsieh CS, Huang CC, Wu JJ, et al: Ascending cholangitis provokes IL-8 and MCP-1 expression and promotes inflammatory cell infiltration in the cholestasis rat liver. J Pediatr Surg 36:1623-1628, 2001
- 20. Dillon PW, Belchis D, Tracy T, et al: Increased expression of intercellular adhesion molecules in biliary atresia. Am J Pathol 145: 263-267. 1994
- 21. Muraji T, Hashimoto Y, Ifuku H, et al: Increased expression of HLA-DR antigens on biliary epithelial cells in biliary atresia. J Jpn Soc Pediatr Surg 24:793-796, 1988
- 22. Endo N, Ohi R, Sawai T, et al: Change of infiltrating pattern of inflammatory cells in the liver of biliary atresia. J Jpn Pediatr Surg 25:213-223, 1989
 - 23. Yamashiro Y, Ohtsuke Y, Shimizu T, et al: Effects of ur-

sode-oxycholic acid treatment on essential fatty acid deficiency in patients with biliary atresia. J Pediatr Surg 29:425-428, 1994

- 24. Ohi R, Shikes RH, Stellin GP, et al: In biliary atresia duct histology correlates with bile flow. J Pediatr Surg 19:467-469, 1984
- 25. Azarow KS, Phillips MJ, Sandler AD, et al: Biliary atresia: Should all patients undergo a portoenterostomy? J Pediatr Surg 32:168-174, 1997.
- 26. Schoen BT, Lee H, Sullivan K, et al: The Kasai portoenterostomy: When is it too late? J Pediatr Surg 36:97-99, 2001
- 27. Tagge DU, Tagge EP, Drongowski RA, et al: A long-term experience with biliary atresia. Ann Surg 214:590-598, 1991
 - 28. Volpert D, White F, Finegold MJ, et al: Outcome of early

- hepatic portoenterostomy for biliary atresia. J Pediatr Gastroenterol and Nutr $32:265-269,\ 2000$
- 29. Hasegawa T, Sasaki T, Kimura T, et al: Measurement of serum hyaluronic acid as a sensitive marker of liver fibrosis in biliary atresia. J Pediatr Surg 35:1643-1646, 2000
- 30. Hasegawa T, Kimura T, Sasaki, et al: Plasma endothelin-1 level as a marker reflecting the severity of portal hypertension in biliary atresia. J Pediatr Surg 36:1609-1612, 2001
- 31. Karrer FM, Price MR, Bensard DD, et al: Long-term results with the Kasai operation for biliary atresia. Arch Surg 131:493-496, 1996
- 32. Miyano T, Fujimoto T, Ohya T, et al: Current concept of treatment of biliary atresia. World J Surg 17:332-336, 1993