Review Article

# A narrative review of genes associated with liver fibrosis in biliary atresia

**Fangran Liu, Clara Sze Man Tang, Patrick Ho Yu Chung**

Division of Paediatric Surgery, Department of Surgery, School of Clinical Medicine, Li Ka Shing Faculty of Medicine, The University of Hong Kong, Hong Kong, China

*Contributions:* (I) Conception and design: F Liu, PHY Chung; (II) Administrative support: PHY Chung; (III) Provision of study materials or patients: PHY Chung, CSM Tang; (IV) Collection and assembly of data: F Liu; (V) Data analysis and interpretation: F Liu, CSM Tang; (VI) Manuscript writing: All authors; (VII) Final approval of manuscript: All authors.

*Correspondence to:* Patrick Ho Yu Chung, MBBS, MS(HK). Division of Paediatric Surgery, Department of Surgery, School of Clinical Medicine, Li Ka Shing Faculty of Medicine, The University of Hong Kong, Pokfulam, Hong Kong, China. Email: chungphy@hku.hk.

**Background and Objective:** Biliary atresia (BA) is characterized by biliary inflammation and obstruction. In the later phase, liver fibrosis occurs. Although the etiology of BA is believed to be multi-factorial, genetic predisposition has been proposed to play a critical role in the pathogenesis. This review aimed to provide an updated summary of the genes that have been reported to be involved in BA-associated liver fibrosis.

**Methods:** The review was conducted via evaluation of MalaCards (BA disease: MalaCards—research articles, drugs, genes, clinical trials) which is a universally applied website including various human disease database. The database of genes that are involved in liver fibrosis were studied.

**Key Content and Findings:** Thirty-one genes that are associated with BA according to the disease relevance score were reviewed after further evaluations. Eleven genes (*GPT*, *NR1H4*, *TGF-B1*, *MMP7*, *CCN2*, *TIMP1*, *SPP1*, *ADD3*, *KRT7*, *ADD3-AS1*, *SOX9*) that are specific and with a potential association with liver fibrosis were selected for detailed description. Increased expression of *GPT*, *TGF-B1*, *MMP7*, *CCN2*, *TIMP1*, *SPP1*, *ADD3*, *KRT7* and *ADD3-AS1* maybe associated with the development of liver fibrosis in BA patients, while the expression of *NR1H4* and *SOX9* are more likely to suppress liver fibrosis.

**Conclusions:** Current scientific evidence using gene database has revealed a close association between genetic anomalies and the pathogenesis of liver fibrosis in BA. With a better understanding of these anomalies, therapy targeting these related genes may be a new therapeutic approach to alleviate liver fibrosis in BA.

**Keywords:** Biliary atresia (BA); liver; fibrosis; genes; pathogenesis

Submitted Mar 13, 2024. Accepted for publication Jul 31, 2024. Published online Aug 28, 2024.
doi: 10.21037/tp-24-94
**View this article at:** https://dx.doi.org/10.21037/tp-24-94

## Introduction

Biliary atresia (BA) is characterized by progressive obliterative cholangiopathy of the intra- and extrahepatic bile ducts. The pathogenesis mainly starts in the embryonic or perinatal period and often leads to severe and persistent neonatal jaundice (1). BA has an incidence of 1 in 10,000–15,000 live births in the United States (2,3), and an incidence of 1 in 16,700 in the British Isles (4,5). BA is more common in the East Asia, with an incidence up to 1 in 5,000 (6). The cause of BA in most infants is not fully understood and it is postulated that the disease is multifactorial with interaction between genetics and exogenous stimuli. As BA is a disease related to genetic anomalies, gene therapy for fibrosis of BA patients may be a potential new direction. In this review, we performed a review on BA related genes and their role in fibrosis, with a hope to bring an insight to new therapy

**Table 1** The search strategy summary

| Items | Specification |
|---|---|
| Date of search | 10/10/2023 |
| Database searched | MalaCards (https://www.malacards.org) |
| Search terms used | Biliary atresia |
| Timeframe | 1998–2024 |
| Inclusion criteria | All studies related to biliary atresia and liver fibrosis, published in English |
| Selection process | Two independent researchers (F.L. and P.H.Y.C.) performed the selection. When a consensus could not be obtained, C.S.M.T. would make the final decision |

in clinical practice. We present this article in accordance with the Narrative Review reporting checklist (available at https://tp.amegroups.com/article/view/10.21037/tp-24-94/rc).

## Methods

MalaCards leverages disease associated genes, Gene Cards and Gene Analytics. Each "card" contains a variety of detailed sections. The Gene Cards Knowledgebase integrates more than 190 data sources to provide a gene-centric, disease-centric, pathway-centric, compound-centric database with associated interconnections and rich annotations. This data is available for research purposes, via an academic collaboration agreement, or through commercial licenses through Life Map Sciences. We conducted an in-depth analysis of the following genes according to their correlation with fibrosis of BA according to MalaCards disease database using search strategy shown in *Table 1*. The genes list is composed by: (I) the Gene Cards search mechanism; (II) genetic testing resources supplying specific genetic tests for the disease; (III) genetic variations resources supplying specific causative variations in genes for the disease; (IV) resources that manually curate the association of the disease with genes. A prioritizing algorithm is applied to generate the genes list. *Table 2* shows gene symbols, descriptions, relevance scores, and the context according to which the gene is related to the disease. The relevance score is provided by Elasticsearch and computed by factoring the importance of the different resources to get the association of one gene with one disease. Theory behind relevance score uses the Boolean model to find matching documents and a formula called the practical scoring function to calculate relevance. Thirty-one genes that are associated with BA according to the disease relevance score were reviewed by two authors (F.L. and P.H.Y.C.). After further evaluations by all the authors, 11 genes [glutamic-pyruvic transaminase (*GPT*), nuclear receptor subfamily 1 group H member 4 (*NR1H4*), transforming growth factor beta 1 (*TGF-B1*), matrix metallopeptidase 7 (*MMP7*), cellular communication network factor 2 (*CCN2*), TIMP metallopeptidase inhibitor 1 (*TIMP1*), secreted phosphoprotein 1 (*SPP1*), adducin 3 (*ADD3*), keratin 7 (*KRT7*), ADD3 antisense RNA 1 (*ADD3-AS1*), SRY-box transcription factor 9 (*SOX9*)] that were found to have a closer association with liver fibrosis, were selected for detailed review.

## Gene related to liver fibrosis in BA

### *GPT gene*

*GPT*, also named as alanine aminotransferase (ALT), catalyzes the reversible transamination between alanine and 2-oxoglutarate to form pyruvate and glutamate. *GPT* participates in cellular nitrogen metabolism and involves in liver gluconeogenesis starting with precursors transported from skeletal muscles by similarity. GPT belongs to the class-I pyridoxal-phosphate-dependent aminotransferase family and belongs to ALT subfamily.

BA infants have a significantly higher ALT expression than those suffering from hepatitis and choledochal cyst (7,8). Some researchers used gamma-glutamyl transferase (GGT)/ALT ratio to differentiate biliary obstruction from neonatal hepatitis (9). Among BA patients after Kasai operation, those with persistent cholestasis have a higher ALT expression than those who can achieve jaundice clearance (10). After intervention with certain drugs, such as: herbal medicine Inchinko-to, it has been found that low expression levels of *GPT* are associated with better

**Table 2** Summary of genes involved in biliary atresia-related liver fibrosis

| Rank | BA relevance score | Location | Gene name (abbreviation) | Gene name (description) | Other roles in BA patients besides fibrosis | Related pathway |
|---|---|---|---|---|---|---|
| 1 | 22.81 | Chromosome 8 | *GPT* | Glutamic-pyruvic transaminase | Metabolism | L-alanine biosynthesis II, glycolysis |
| 2 | 20.27 | Chromosome 12 | *NR1H4* | Nuclear receptor subfamily 1 group H member 4 | Metabolism, immune | Synthesis of bile acids and bile salts, gene expression |
| 3 | 19.11 | Chromosome 19 | *TGF-B1* | Transforming growth factor beta 1 | NA | Apoptotic pathways in synovial fibroblasts, GPCR pathway |
| 4 | 18.98 | Chromosome 11 | *MMP7* | Matrix metallopeptidase 7 | NA | Collagen chain trimerization, matrix metalloproteinases |
| 5 | 18.26 | Chromosome 6 | *CCN2* | Cellular communication network factor 2 | NA | Apoptotic pathways in synovial fibroblasts, GPCR pathway |
| 6 | 17.19 | Chromosome X | *TIMP1* | TIMP metallopeptidase inhibitor 1 | NA | Apoptotic pathways in synovial fibroblasts, GPCR pathway |
| 7 | 16.28 | Chromosome 4 | *SPP1* | Secreted phosphoprotein 1 | Immune | Integrin pathway, ERK signaling |
| 8 | 15.81 | Chromosome 10 | *ADD3* | Adducin 3 | Mutation, genetic susceptibility | Activation of cAMP-dependent PKA, signaling by Rho GTPases |
| 9 | 14.08 | Chromosome 12 | *KRT7* | Keratin 7 | NA | Keratinization and nervous system development |
| 10 | 14.06 | Chromosome 10 | *ADD3-AS1* | ADD3 antisense RNA 1 | Mutation, genetic susceptibility | NA |
| 11 | 13.66 | Chromosome 17 | *SOX9* | SRY-box transcription factor 9 | NA | Gene expression (transcription), mammalian disorder of sexual development |

BA, biliary atresia; GPCR, G protein-coupled receptor; cAMP, cyclic adenosine monophosphate; NA, not available.

prognosis and treatment outcomes. Liver function test at puberty for low expression levels of GPT is a good prognostic point for BA children (11,12). Serum *GPT* level as liver fibrosis marker could also indicate drug effect and liver injury for postoperative BA patients (13,14).

### *NR1H4 gene*

*NR1H4* promotes transcriptional activation of target genes *ABCB11/BSEP*, *NR0B2/SHP*, *SLC51B/OSTB* and *FABP6/IBAP*. NR1H4 belongs to the nuclear hormone receptor family and NR1 subfamily. NR1H4 participates in hepatocyte glucose and lipid metabolism. By modulating gene expression, it influences the balance of glucose production, utilization and storage within hepatocytes. Proper glucose metabolism is essential for maintaining blood sugar levels and preventing conditions like diabetes. Additionally, NR1H4 helps regulate lipid metabolism, impacting processes such as fatty acid synthesis, cholesterol homeostasis, and bile acid synthesis (15). The affected glucose and lipid metabolism of hepatocyte in turn inhibit fibrosis, inflammation, and apoptosis.

*NR1H4* gene expression is low in BA patients in the Gene Expression Omnibus database (15). Protein and mRNA level experiments also validate the bioanalysis result that there's lower *NR1H4* gene expression in BA

 

patients than in non-BA control group (15). Low *NR1H4* gene expression group have higher immunologically infiltration levels (15). As *NR1H4* has been found to play an important role in modulating hepatocellular transport, bile acid homeostasis as well as metabolic functions, some scholars questioned the differential expression of *NR1H4* in early and late stages of BA. In a study of BA infants with early- and late-stage disease, *NR1H4* expression level was downregulated at early-stage cholestasis, bile salt export pump (BSEP) expression level was similar with its upstream regulator *NR1H4*. At late-stage cholestasis, *NR1H4* and *BSEP* gene expression level returned to normal (16).

### *TGF-B1 gene*

*TGF-B1* is multifunctional protein that controls proliferation, differentiation and other functions in many cell types. Many cells synthesize *TGF-B1* and have specific receptors for its interaction with other growth factors. This gene encodes a secreted ligand of the TGF-beta superfamily of proteins. Ligands of this family bind various TGF-beta receptors leading to recruitment and activation of SMAD family transcription factors that regulate gene expression. SMADs are composed of structurally similar families of proteins that are major signaling sensors for TGF-beta superfamily receptors, which are critical for regulating cell development and growth.

A previous study that evaluated *TGF-B1* divided the cohort into three groups: BA before and after liver transplant as well healthy controls (17). It was revealed that BA patients who were indicated for liver transplant had the lowest plasma *TGF-B1* concentrations and the highest plasma hepatocyte growth factor (HGF)/*TGF-B1* ratios (17), which suggested that low *TGF-B1* expression may represent a worse prognosis. *TGF-B1* expression could be found in activated hepatic stellate cells (HSCs), hepatocytes and bile duct epithelial cells, which also indicated proximity to fibrosis (18). *TGF-B1* regulates proliferation and replication of liver cell that is associated with hepatic fibrosis. *TGF-B1* activates HSCs that is also involved in fibrogenesis (19). *TGF-B1* expressed in both biliary epithelial cells and hepatocytes in the congenital BA group infants and the expression levels of which increased gradually with fibrosis progression (20). In a 3-year follow-up study of patients with successful Kasai portoenterostomy, hepatic *TGF-B1* expression level was significantly decreased, but *TGF-B2* expression level was found to increase (21). This phenomenon signified the potential interaction of TGF-beta family after Kasai portoenterostomy among BA patients (22).

### *MMP7 gene*

*MMP7* degrades casein, gelatins of types I, III, IV, and V, and fibronectin, also activates procollagens, belongs to M10 matrix metallopeptidases (MMPs). Proteins in this family are involved in the breakdown of extracellular matrix (ECM) in normal physiological processes. The encoded preproprotein is proteolytically processed to generate the mature protease. This secreted protease breaks down proteoglycans, fibronectin, elastin and casein and differs from most MMP family members in that it lacks a conserved C-terminal hemopexin domain. The gene is part of a cluster of MMP genes on chromosome 11.

A previous study divided BA patients into three different groups: early-stage BA group at the time of Kasai portoenterostomy, late-stage BA group at the time of liver transplantation with advanced fibrosis and control group without liver fibrosis. *MMP7* expression levels were significantly different between control group and liver transplantation group, which indicated that *MMP7* is associated with disease progression. *MMP7* expression level had a progressive increase from the time at Kasai portoenterostomy to liver transplantation (23). Besides, *MMP7* is mainly expressed in hepatocytes, Kupffer cells and bile ductular epithelial cells of liver. The comparison among different MMPs such as MMP2, MMP7, MMP9 and MMP13 indicated that MMP7 is the MMP with a prominent expression (24), especially in late-stage BA group at the time of liver transplantation with advanced fibrosis. Experiment using mouse model also showed that *MMP7* expressed more in mice with fibrosis, which indicates that *MMP7* is a mediator of hepatic fibrosis between sick BA mice and spontaneously recovered BA mice (25). Many researchers have studied the relation of *MMP7* and BA fibrosis and cirrhosis. *MMP7* plays an important role in BA fibrosis of young patients (24,26-30). *MMP7* express location was in biliary epithelium having ductular proliferation (31). *MMP7* gene is upregulated in BA patients in contrast with control patients (32). *MMP7* may be involved in the pathophysiological process of BA occurrence and development. On the one hand, *MMP7* can promote the migration and activation of macrophages and neutrophils by cutting target proteins such as TNF-α and syndecan-1, thus aggravating inflammation and bile

duct injury. On the other hand, *MMP7* can activate FAS receptors and destroy adhesion molecules between bile duct epithelial cells, thereby mediating cell apoptosis (33). *MMP7* is of great importance both as a prognostic factor and a diagnostic tool for BA.

### *CCN2 gene*

*CCN2* is hypertrophic chondrocyte-specific protein 24 that belongs to the CCN family. It is a mitoattractant of connective tissue and secreted by vascular endothelial cells. *CCN2* promotes proliferation and differentiation of chondrocytes and mediates heparin- and divalent cation-dependent cell adhesion in many cell types including fibroblasts, myofibroblasts, endothelial and epithelial cells. It also enhances fibroblast growth factor-induced DNA synthesis. *CCN2* is related to cell matrix induction, mitosis and chemotaxis, which is specifically induced in by TGF-beta in fibroblast cell.

*CCN2* serum expression level was associated with fibrosis in BA. This result indicated that *CCN2* is a potential indicator to monitor fibrosis in BA patients (34). One study evaluated the expression of *CCN2* mRNA in BA patients and normal controls. It was reported that BA patients had significantly higher *CCN2* and collagen type IV expression than normal control group. *CCN2* expression also positive associated with collagen type IV expression. As collagen type IV is used as an indicator of fibrosis severity, this study concluded that *CCN2* may affect BA pathogenesis and fibrosis progression (35). *CCN2* expression is mainly observed in biliary epithelial cells and vascular endothelial cells of BA patients and related to biliary epithelial cell fibrogenesis (36). Thirty-six infants with BA and neonatal hepatitis were divided into two different disease groups and their *CCN2*, *TGF-B1* expression levels and liver fibrosis were compared. The result showed that BA patients have a higher expression of *CCN2* and *TGF-B1* expression as well as fibrosis than patients suffering from hepatitis. Hepatic *CCN2* expression level was significantly decreased in a 3 years' follow-up study of successful Kasai portoenterostomy patients. In addition, taking into consideration of *CCN2* role in fibroblasts senescence, it can be expected that cholangiocyte senescence-associated secretory phenotype paracrine communication plays an essential role in the prognosis of BA (37).

### *TIMP1 gene*

*TIMP1* is a metalloproteinase inhibitor that functions by forming one to one complex with target metalloproteinases, such as collagenases, and irreversibly inactivates them by binding to their catalytic zinc cofactor. It also functions as a growth factor that regulates cell differentiation, migration and cell death, and activates cellular signaling cascades via CD63 and ITGB1. This gene belongs to the TIMP gene family. The proteins encoded by this gene family are natural inhibitors of the MMPs. In addition to its inhibitory role against most of the known MMPs, the encoded protein is able to promote cell proliferation in a wide range of cell types and may also have an anti-apoptotic function. Transcription of this gene is highly inducible in response to many cytokines and hormones. In addition, the expression from some but not all inactive X chromosomes suggests that this gene inactivation is polymorphic in human females. This gene is located within intron 6 of the synapsin I gene and is transcribed in the opposite direction.

In the process of liver fibrosis, activated HSC are a key source of collagen production. At the same time, the activated HSC secretes a large amount of *TGF-B1*, which upregulates the gene expression of *TIMP1* and downregulates the expression of MMPs. This leads to an imbalance of MMPs/TIMPs, which in turn leads to an imbalance of ECM synthesis and degradation, which promotes liver fibrosis (38). The imbalance of *TIMP1* activity is an important factor leading to the progression of liver fibrosis. One study found that the positive expression of *TIMP1* protein in liver fibrosis tissues increased with the aggravation of fibrosis degree, while the positive expression of MMP1 protein did not change significantly (39). This further supports the role of *TIMP1* in liver fibrosis. Many studies have revealed that *TIMP1* is associated with liver fibrosis in BA (23,25,40-42). Some studies found that vitamin D deficiency could increase the expression of *TIMP1*, which in turn promote hepatic fibrosis in BA children (43). Furthermore, *TIMP1* expression is associated with persistent jaundice and portal hypertension among patients among Kasai operation (10).

### *SPP1 gene*

*SPP1* is also known as osteopontin (*OPN*). Among the related pathways of SPP1 are integrin pathway and ERK

signaling. Gene Ontology (GO) annotations related to this gene include cytokine activity and ECM binding.

Compared with other cholestatic diseases, the expression levels of osteopontin mRNA and protein in the liver of BA patients are significantly increased, and the expression of osteopontin is limited to the proliferating biliary epithelium/bile duct blockade epithelium. Osteopontin expression was not found in the portal biliary tract of normal liver. According to previous study, the expression of osteopontin in biliary epithelial cells is related to the expression of IL-2 and TNF-α, and the increased expression of osteopontin in biliary interlobular epithelium in BA patients is related to portal fibrosis and biliary hyperplasia, suggesting the role of osteopontin in the mechanism of BA (44).

Another study found that osteopontin was expressed in BA intrahepatic bile duct epithelial cells, but only in small amounts in normal intrahepatic bile duct epithelial cells. The expression of osteopontin in BA liver tissue is positively correlated with the degree of liver fibrosis. In the process of forming fibrosis in BA, *MMP7* and osteopontin can form a positive feedback loop, whereby *MMP7* amplifies fibrosis and inflammation through osteopontin and TNF-α (45). The expression level of osteopontin was positively correlated with the degree of BA fibrosis and the hardness of BA liver (46). Plasma osteopontin level is not only positively correlated with serum ALT and serum total bilirubin level, but also can be used as a biomarker to predict portal hypertension and liver dysfunction (47).

### *ADD3 gene*

Adducins are heteromeric proteins composed of different subunits referred to as adducin alpha, beta and gamma. The three subunits are encoded by distinct genes and belong to a family of membrane skeletal proteins involved in the assembly of spectrin-actin network in erythrocytes and at sites of cell-cell contact in epithelial tissues. Structurally, each subunit is comprised of two distinct domains. *ADD3* is membrane-cytoskeleton-associated protein that promotes the assembly of the spectrin-actin network and regulates expression of profibrotic markers MMP2, MMP9, *TGF-B1*, tubular tight junction protein E-cadherin, and mesenchymal markers vimentin and alpha-SMA.

*ADD3* gene is associated with cell damage repair, cycle and apoptosis in patients with BA. Single nucleotide polymorphisms in *ADD3* were associated with BA, with the strongest association detected for rs17095355, which was associated with BA susceptibility in Asians and Caucasians (48,49). In addition, many other studies have confirmed the link between the function/mutation of *ADD3* gene and the occurrence of BA (49-51). Deletion of *ADD3* gene can cause excessive deposition of actin and myosin, resulting in biliary fibrosis (52). Long non-coding RNA (lncRNA) *ADD3*-AS1 is involved in the occurrence of liver fibrosis in BA children and can be used as a potential biomarker for the diagnosis of liver fibrosis in BA patients (53). Studies using zebrafish as experimental subjects found that *ADD3* affects the occurrence of BA through Hedgehog signaling pathway (54).

### *KRT7 gene*

The protein encoded by *KRT7* gene is a member of the keratin gene family. The type II cytokeratins consist of basic or neutral proteins which are arranged in pairs of heterotypic keratin chains co-expressed during differentiation of simple and stratified epithelial tissues. This type II cytokeratin is specifically expressed in the simple epithelia lining the cavities of the internal organs and in the gland ducts and blood vessels. The genes encoding the type II cytokeratins are clustered in a region of chromosome 12q12–q13. Alternative splicing may result in several transcript variants; however, not all variants have been fully described.

The positive percentage of *KRT7* has been reported to be useful as a marker for liver fibrosis and cirrhosis (55). Increased *KRT7* immunopositivity in periportal hepatocytes after jaundice clearance can predict hepatic fibrosis during follow-up (56,57). *KRT7* is associated with epithelial mesenchymal transformation and is localized in intrahepatic biliary duct epithelial cells (58). The absence of interlobular biliary ducts in BA patients results in abnormal expression of *KRT7* in hepatocytes, which represents phenotypic changes in stem cells due to loss of contact with the biliary tree (59).

### *ADD3-AS1 gene*

*ADD3-AS1* is an RNA gene and is affiliated with the lncRNA class. Diseases associated with *ADD3-AS1* include BA. This gene increases susceptibility to BA, plays a role in the mechanism of liver fibrosis in BA patients, and can be used as an important indicator to monitor liver fibrosis in BA patients (53,60).

Although the specific mechanism of action of *ADD3-AS1* in liver fibrosis in BA patients is unclear, several molecular

pathways related to *ADD3-AS1* may play a key role in the development of liver fibrosis in BA patients, which are listed as follows. MicroRNAs (miRNAs) are a class of small RNA molecules that have been shown to play an important role in liver fibrosis caused by various chronic liver diseases. They can interfere with the development of liver fibrosis by regulating the expression of genes related to liver fibrosis. Chitinase-3-like protein 1 (CHI3L1) is a protein that plays a role in the interaction between liver macrophages and myofibroblasts, which further activates HSCs, forming a cascade that leads to liver fibrosis. Intrahepatic cell and epithelial mesenchymal transformation may also play an important role in liver fibrosis in BA patients. Persistent inflammatory factors, such as infiltration of inflammatory cells and mediators, may damage bile duct epithelial cells and cause the occurrence of BA liver fibrosis.

### *SOX9 gene*

*SOX9* is a protein coding gene. Among its related pathways are gene expression (transcription) and mammalian disorder of sexual development. GO annotations related to this gene include DNA-binding transcription factor activity and protein kinase activity. An important paralog of this gene is *SOX10*. The protein encoded by this gene recognizes the sequence CCTTGAG along with other members of the high mobility group (HMG)-box class DNA-binding proteins. It acts during chondrocyte differentiation and, with steroidogenic factor 1, regulates transcription of the anti-Muellerian hormone (*AMH*) gene. In addition to the role in cartilage development, *SOX9* also acts as a regulator of proliferation and differentiation in epithelial stem/progenitor cells. Furthermore, *SOX9* has vital role in Hirschsprung disease as well, not specific with only liver fibrosis.

*SOX9* is mainly expressed in the fibrotic bridge and portal vein space, regulating liver fibrosis, bile duct development and liver regeneration process. In BA patients, *SOX9* is mainly expressed in reactive ductal epithelium, and the expression pattern is significantly different from that of normal people, *SOX9* may play a role in BA diagnosis (61). Other studies have also found an association between abnormal expression of *SOX9* in ductular reaction form and liver fibrosis in BA patients (62,63). *SOX9* may play a role in BA mechanisms by affecting reactive duct cells and hepatocytes (64). *SOX9* plays a protective role in the progression of catheter response and can be used as a prognostic indicator to predict natural liver survival in BA patients after Kasai surgery, and patients with high *SOX9* expression of BA have a better prognosis (65).

## Discussion

Based on our work, we believe that by regulating certain genes can potentially suppress or even reverse liver fibrosis in BA (66). This study discussed 11 genes correlated to liver fibrosis in BA, with an aim to provide a new treatment strategy to BA related liver fibrosis. The roles of each gene in alleviating or aggravating BA liver fibrosis have been discussed in detail. Even though the modulating process of some genes are uncertain, these findings can still demonstrate their ability in reversing liver fibrosis and improving the treatment outcome. According to MalaCards, we can identify many pathways that are mostly associated with each fibrosis related gene in BA. The limitations of this study include a lack of detailed literature on some genes and their influence on liver fibrosis cannot be fully determined.

## Conclusions

According to detailed literature search, we found 11 genes that are closely related to liver fibrosis in BA. Increased expression of *GPT*, *TGF-B1*, *MMP7*, *CCN2*, *TIMP1*, *SPP1*, *ADD3*, *KRT7* and *ADD3-AS1* maybe associated with the development of liver fibrosis in BA patients, while the expression of *NR1H4* and *SOX9* are more likely to suppress liver fibrosis. Future works that explore gene therapy is recommended for its potential in alleviating fibrosis in BA. Drugs targeting fibrosis related genes can be a new research area in BA.

## Acknowledgments

*Funding:* This study was supported by the Theme-based Research Scheme, Research Grant Council, Hong Kong (Project code: T12-712/21-R 'Translating Disease-Mechanism Discoveries to Improve Treatment of Biliary Atresia, an Intractable Newborn Liver Disease').

## Footnote

*Reporting Checklist:* The authors have completed the Narrative Review reporting checklist. Available at https://tp.amegroups.com/article/view/10.21037/tp-24-94/rc

*Peer Review File:* Available at https://tp.amegroups.com/

article/view/10.21037/tp-24-94/prf

*Conflicts of Interest:* All authors have completed the ICMJE uniform disclosure form (available at https://tp.amegroups.com/article/view/10.21037/tp-24-94/coif). P.H.Y.C. serves as an unpaid editorial board member of *Translational Pediatrics* from August 2022 to July 2024. The other authors have no conflicts of interest to declare.

*Ethical Statement:* The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.



## References

1. Chardot C. Biliary atresia. Orphanet J Rare Dis 2006;1:28.
2. Suchy FJ. Anatomy, histology, embryology, developmental anomalies, and pediatric disorders of the biliary tract. In: Feldman M, Friedman LS, Brandt LJ. editors. Sleisenger and Fordtran's Gastrointestinal and Liver Disease. 9th edition. Elsevier; 2010:1045-1066.e5.
3. Pithawa AK. Sleisenger and Fordtran's Gastrointestinal and Liver Disease: pathophysiology, diagnosis, management. Med J Armed Forces India 2007;63:205.
4. McKiernan PJ, Baker AJ, Kelly DA. The frequency and outcome of biliary atresia in the UK and Ireland. Lancet 2000;355:25-9.
5. Hartley JL, Davenport M, Kelly DA. Biliary atresia. Lancet 2009;374:1704-13.
6. Al-Hussaini A, Abanemai M, Alhebbi H, et al. The Epidemiology and Outcome of Biliary Atresia: Saudi Arabian National Study (2000-2018). Front Pediatr 2022;10:921948.
7. Chu B, Jiang LR, Zhou S, et al. Value of the liver function test in differential diagnosis of infantile hepatitis syndrome and biliary atresia. Zhongguo Dang Dai Er Ke Za Zhi 2009;11:953-6.
8. Chen YT, Gao MJ, Zheng ZB, et al. Comparative analysis of cystic biliary atresia and choledochal cysts. Front Pediatr 2022;10:947876.
9. Tang KS, Huang LT, Huang YH, et al. Gamma-glutamyl transferase in the diagnosis of biliary atresia. Acta Paediatr Taiwan 2007;48:196-200.
10. Honsawek S, Chongsrisawat V, Vejchapipat P, et al. Association of serum levels of tissue inhibitors of metalloproteinase-1 with clinical outcome in children with biliary atresia. Asian Pac J Allergy Immunol 2006;24:161-6.
11. Kuroda T, Saeki M, Morikawa N, et al. Biliary atresia and pregnancy: puberty may be an important point for predicting the outcome. J Pediatr Surg 2005;40:1852-5.
12. Iinuma Y, Kubota M, Yagi M, et al. Effects of the herbal medicine Inchinko-to on liver function in postoperative patients with biliary atresia--a pilot study. J Pediatr Surg 2003;38:1607-11.
13. Kobayashi H, Horikoshi K, Yamataka A, et al. Beneficial effect of a traditional herbal medicine (inchin-ko-to) in postoperative biliary atresia patients. Pediatr Surg Int 2001;17:386-9.
14. Yeung F, Fung ACH, Chung PHY, et al. Long-term follow-up of biliary atresia using liver transient elastography. Pediatr Surg Int 2022;38:1013-8.
15. Ma Y, Lu L, Tan K, et al. Reduced peroxisome proliferator-activated receptor-α and bile acid nuclear receptor NR1H4/FXR may affect the hepatic immune microenvironment of biliary atresia. Front Immunol 2022;13:875593.
16. Chen HL, Liu YJ, Chen HL, et al. Expression of hepatocyte transporters and nuclear receptors in children with early and late-stage biliary atresia. Pediatr Res 2008;63:667-73.
17. Cheng CH, Tsau YK, Tsai IJ. Kidney volume and plasma hepatocyte growth factor-transforming growth factor beta1 ratio among children with biliary atresia before and after liver transplantation: the reversibility of nephromegaly. Am J Kidney Dis 2005;46:830-6.
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 1998;153:527-35.
19. Kobayashi H, Horikoshi K, Yamataka A, et al. Are stable postoperative biliary atresia patients really stable? Pediatr Surg Int 2001;17:104-7.
20. Li FB, Zhao H, Peng KR, et al. Expression of transforming growth factor-beta1 and connective tissue growth factor in

 

congenital biliary atresia and neonatal hepatitis liver tissue. Genet Mol Res 2016. doi: 10.4238/gmr.15017217.
21. Kerola A, Lohi J, Heikkilä P, et al. Divergent expression of liver transforming growth factor superfamily cytokines after successful portoenterostomy in biliary atresia. Surgery 2019;165:905-11.
22. Vejchapipat P, Theamboonlers A, Poomsawat S, et al. Serum transforming growth factor-beta1 and epidermal growth factor in biliary atresia. Eur J Pediatr Surg 2008;18:415-8.
23. Hsieh CS, Chuang JH, Huang CC, et al. Evaluation of matrix metalloproteinases and their endogenous tissue inhibitors in biliary atresia-associated liver fibrosis. J Pediatr Surg 2005;40:1568-73.
24. Huang CC, Chuang JH, Chou MH, et al. Matrilysin (MMP-7) is a major matrix metalloproteinase upregulated in biliary atresia-associated liver fibrosis. Mod Pathol 2005;18:941-50.
25. Nadler EP, Li X, Onyedika E, et al. Differential expression of hepatic fibrosis mediators in sick and spontaneously recovered mice with experimental biliary atresia. J Surg Res 2010;159:611-7.
26. Jiang J, Wang J, Shen Z, et al. Serum MMP-7 in the Diagnosis of Biliary Atresia. Pediatrics 2019;144:e20190902.
27. Irvine KM, Wockner LF, Hoffmann I, et al. Multiplex Serum Protein Analysis Identifies Novel Biomarkers of Advanced Fibrosis in Patients with Chronic Liver Disease with the Potential to Improve Diagnostic Accuracy of Established Biomarkers. PLoS One 2016;11:e0167001.
28. Yang L, Zhou Y, Xu PP, et al. Diagnostic Accuracy of Serum Matrix Metalloproteinase-7 for Biliary Atresia. Hepatology 2018;68:2069-77.
29. Wu JF, Jeng YM, Chen HL, et al. Quantification of Serum Matrix Metallopeptide 7 Levels May Assist in the Diagnosis and Predict the Outcome for Patients with Biliary Atresia. J Pediatr 2019;208:30-37.e1.
30. Lertudomphonwanit C, Mourya R, Fei L, et al. Large-scale proteomics identifies MMP-7 as a sentinel of epithelial injury and of biliary atresia. Sci Transl Med 2017;9:eaan8462.
31. Kerola A, Lampela H, Lohi J, et al. Increased MMP-7 expression in biliary epithelium and serum underpins native liver fibrosis after successful portoenterostomy in biliary atresia. J Pathol Clin Res 2016;2:187-98.
32. Bezerra JA, Tiao G, Ryckman FC, et al. Genetic induction of proinflammatory immunity in children with biliary atresia. Lancet 2002;360:1653-9.
33. Kong F, Dong R, Chen G, et al. Progress in Biomarkers Related to Biliary Atresia. J Clin Transl Hepatol 2024;12:305-15.
34. Tamatani T, Kobayashi H, Tezuka K, et al. Establishment of the enzyme-linked immunosorbent assay for connective tissue growth factor (CTGF) and its detection in the sera of biliary atresia. Biochem Biophys Res Commun 1998;251:748-52.
35. Kobayashi H, Hayashi N, Hayashi K, et al. Connective tissue growth factor and progressive fibrosis in biliary atresia. Pediatr Surg Int 2005;21:12-6.
36. Narkewicz MR, Kasaragod A, Lucia MS, et al. Connective tissue growth factor expression is increased in biliary epithelial cells in biliary atresia. J Pediatr Surg 2005;40:1721-5.
37. Meadows V, Baiocchi L, Kundu D, et al. Biliary Epithelial Senescence in Liver Disease: There Will Be SASP. Front Mol Biosci 2021;8:803098.
38. Dang SS, Li YP. Advances in understanding the role of transforming growth factor-β1 in the pathogenesis of liver fibrosis. World Chinese Journal of Digestology 2010;18:1631-6.
39. Yang CQ, Hu GL, Tan DM, et al. Relativity of Expression of MMP-1, TIMP-1 and Variability of Type I, III Collagen During Experimental Liver Fibrosis. Journal of Clinical Hepatology 2000;16:222-4.
40. Baba H, Ohtsuka Y, Fujii T, et al. Immunological investigation of the hepatic tissue from infants with biliary atresia. Pediatr Surg Int 2009;25:157-62.
41. Nadler EP, Patterson D, Violette S, et al. Integrin alphavbeta6 and mediators of extracellular matrix deposition are up-regulated in experimental biliary atresia. J Surg Res 2009;154:21-9.
42. Wiśniewska J, Sadowska A, Wójtowicz A, et al. Perspective on Stem Cell Therapy in Organ Fibrosis: Animal Models and Human Studies. Life (Basel) 2021;11:1068.
43. Sun S, Xu M, Zhuang P, et al. Effect and mechanism of vitamin D activation disorder on liver fibrosis in biliary atresia. Sci Rep 2021;11:19883.
44. Whitington PF, Malladi P, Melin-Aldana H, et al. Expression of osteopontin correlates with portal biliary proliferation and fibrosis in biliary atresia. Pediatr Res 2005;57:837-44.
45. Nomden M, Beljaars L, Verkade HJ, et al. Current Concepts of Biliary Atresia and Matrix Metalloproteinase-7: A Review of Literature. Front Med (Lausanne) 2020;7:617261.
46. Honsawek S, Chayanupatkul M, Chongsrisawat V, et al.

 

Increased osteopontin and liver stiffness measurement by transient elastography in biliary atresia. World J Gastroenterol 2010;16:5467-73.
47. Honsawek S, Vejchapipat P, Chongsrisawat V, et al. Association of circulating osteopontin levels with clinical outcomes in postoperative biliary atresia. Pediatr Surg Int 2011;27:283-8.
48. Bai MR, Niu WB, Zhou Y, et al. Association of common variation in ADD3 and GPC1 with biliary atresia susceptibility. Aging (Albany NY) 2020;12:7163-82.
49. Cheng G, Tang CS, Wong EH, et al. Common genetic variants regulating ADD3 gene expression alter biliary atresia risk. J Hepatol 2013;59:1285-91.
50. Zeng S, Sun P, Chen Z, et al. Association between single nucleotide polymorphisms in the ADD3 gene and susceptibility to biliary atresia. PLoS One 2014;9:e107977.
51. Tsai EA, Grochowski CM, Loomes KM, et al. Replication of a GWAS signal in a Caucasian population implicates ADD3 in susceptibility to biliary atresia. Hum Genet 2014;133:235-43.
52. Mack CL, Feldman AG, Sokol RJ. Clues to the etiology of bile duct injury in biliary atresia. Semin Liver Dis 2012;32:307-16.
53. Ye Y, Wu W, Zheng J, et al. Role of long non-coding RNA-adducin 3 antisense RNA1 in liver fibrosis of biliary atresia. Bioengineered 2022;13:6222-30.
54. Tang V, Cofer ZC, Cui S, et al. Loss of a Candidate Biliary Atresia Susceptibility Gene, add3a, Causes Biliary Developmental Defects in Zebrafish. J Pediatr Gastroenterol Nutr 2016;63:524-30.
55. Redkar R, Raj V, Chigicherla S, et al. Risk Prediction Scoring System to Predict the Postsurgical Outcomes of Biliary Atresia. J Indian Assoc Pediatr Surg 2020;25:280-5.
56. Hukkinen M, Kerola A, Lohi J, et al. Treatment Policy and Liver Histopathology Predict Biliary Atresia Outcomes: Results after National Centralization and Protocol Biopsies. J Am Coll Surg 2018;226:46-57.e1.
57. Lampela H, Kosola S, Heikkilä P, et al. Native liver histology after successful portoenterostomy in biliary atresia. J Clin Gastroenterol 2014;48:721-8.
58. Xiao Y, Zhou Y, Chen Y, et al. The expression of epithelial-mesenchymal transition-related proteins in biliary epithelial cells is associated with liver fibrosis in biliary atresia. Pediatr Res 2015;77:310-5.
59. Ernst LM, Spinner NB, Piccoli DA, et al. Interlobular bile duct loss in pediatric cholestatic disease is associated with aberrant cytokeratin 7 expression by hepatocytes. Pediatr Dev Pathol 2007;10:383-90.
60. Sato K, Glaser S, Francis H, et al. Concise Review: Functional Roles and Therapeutic Potentials of Long Non-coding RNAs in Cholangiopathies. Front Med (Lausanne) 2020;7:48.
61. Arboleda-Bustan JE, Ribalta T, Albert A, et al. Expression of Protein SOX9 in Biliary Atresia. J Pediatr Gastroenterol Nutr 2022;74:e21-6.
62. El-Araby HA, Saber MA, Radwan NM, et al. SOX9 in biliary atresia: New insight for fibrosis progression. Hepatobiliary Pancreat Dis Int 2021;20:154-62.
63. Yin C. Molecular mechanisms of Sox transcription factors during the development of liver, bile duct, and pancreas. Semin Cell Dev Biol 2017;63:68-78.
64. Suda H, Yoshii D, Yamamura K, et al. New insight into reactive ductular cells of biliary atresia provided by pathological assessment of SOX9. Pediatr Surg Int 2014;30:481-92.
65. Yoshii D, Shimata K, Yokouchi Y, et al. SOX9 contributes to the progression of ductular reaction for the protection from chronic liver injury. Hum Cell 2022;35:721-34.
66. Dunbar CE, High KA, Joung JK, et al. Gene therapy comes of age. Science 2018;359:eaan4672.

**Cite this article as:** Liu F, Tang CSM, Chung PHY. A narrative review of genes associated with liver fibrosis in biliary atresia. Transl Pediatr 2024;13(8):1469-1478. doi: 10.21037/tp-24-94