# Precursor Lesions of Cholangiocarcinoma: A Clinicopathologic Review

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**ABSTRACT:** Cholangiocarcinoma (CCA) develops through multistep carcinogenesis. During the past decades, 2 precursors have been proved to evolve to CCA. The 2 main precursor lesions of CCA are biliary intraepithelial neoplasia and intraductal papillary neoplasm of the bile duct. It is an interesting and relatively novel entity for the hepatobiliary surgeons, radiologists, oncologists, and pathologists. It worth being familiar with these 2 entities for better communication between pathologists, oncologists, and surgeons to improve the treatment and follow-up of these lesions, which can definitely decrease their evolvement to CCA as an aggressive, poor prognostic, and life-threatening cancer. In this narrative review, I collected and discussed all published studies about these 2 precursor lesions of CCA including radiologic, clinical, and pathological manifestation.

**KEYWORDS:** Biliary intraepithelial neoplasia, CHolangiocarcinoma

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## Introduction

Cholangiocarcinoma (CCA) is an aggressive cancer of intra- and extrahepatic bile ducts which has been proved to develop through a multistep sequence (multistep carcinogenesis). Precursor neoplastic lesions of bile ducts were used to be called as biliary dysplasia or atypical biliary epithelium.[1] However, in 2005, Zen et al proposed the entity of biliary intraepithelial neoplasia (BilIN) as precursors of CCA in multistep carcinogenesis.[2] Another preinvasive lesion of biliary epithelium which is also considered as precursor of CCA is intraductal papillary neoplasm of the bile duct (IPNB) which was introduced as distinct pathologic entities in World Health Organization classification of hepatobiliary tumors in 2010.[3]

Recently, pathologists and clinicians are exposed to more cases of precursor lesions of biliary tracts diagnosed by imaging techniques, and it is very important for them to be familiar with these 2 entities and start reporting them for early diagnosis of biliary tract lesions before the occurrence of invasion to become malignant and aggressive CCA which has poor prognosis.[4,5] This subject is especially very important in the patients with primary sclerosing cholangitis which causes chronic recurring inflammation and destruction of biliary epithelium of bile ducts,[6,7] although recently there are some reports of the occurrence of BilIN in nonbiliary cirrhotic patients.[8]

In this review, we will discuss about all the clinicopathologic and molecular aspects of precursors of CCA by reviewing all the published studies in English literature.

### *Normal histology of biliary epithelium*

Epithelial cells that line biliary tracts are called cholangiocytes which progressively continue into a system of interlobular, septal, major ducts, and final extrahepatic bile ducts continuing to ampulla of Vater in the duodenum.[9] Biliary epithelium is flat with a single layer of columnar cells with basal oval nuclei and eosinophilic cytoplasm.[10]

### *Histologic subtypes of CCA*

Cholangiocarcinoma is classified as intrahepatic cholangiocarcinoma (ICC), perihilar, and distal. They can be conventional, bile ductular, and intraductal. Conventional can be small or large bile duct type. Each of them can be well-, moderately, and poorly differentiated adenocarcinoma.[11,12]

## Precursors of CCA

There are 2 main types of premalignant lesions which are considered as precursors of CCA:

1. Biliary intraepithelial neoplasia (BilIN)
2. Intraductal papillary neoplasm of the bile duct (IPNB).

It is believed that these are more important in the development of ICC and perihilar CCA rather than extrahepatic CCA.[12] Other rare, less well-defined and controversial precursor lesions of CCA are peribiliary glands, von Meyenburg complex, and bile duct adenoma.[13-18]

### *Predisposing factors*

There is not significant association between cirrhosis and precursors of CCA; however, primary sclerosing cholangitis has been proved to have association with them. It is the overwhelming driver of most hilar/extrahepatic CCA in the

Western world.[6,7] There is significant difference in the predisposing factors in different geographic areas of the world.[19] There are significantly higher rates of ICC seen in Eastern Asia when compared with Western countries which is mostly attributed to hepatolithiasis and parasitic infestation by liver flukes.[20] Recently, association between these lesions and hepatitis viruses, especially hepatitis C virus (HCV), has been described.[8,21] The association with chemical agents such as environmental and occupational exposure in printing plant has been reported from Japan as well as thorotrast.[22]

Main predisposing factors for ICC are choledochal cysts, choledocholithiasis, cirrhosis, cholelithiasis, hepatitis B virus (HBV), HCV, alcohol, cholecystocholithiasis, inflammatory bowel disease, and smoking. These predisposing factors for extrahepatic CCA are mainly choledochal cysts, choledocholithiasis, cirrhosis, cholelithiasis, HBV, alcohol, cholecystocholithiasis, inflammatory bowel disease, and smoking.[23]

### *Clinical manifestation*

These precursor lesions are more common in male patients after 50 years of age; however, there are very little published articles about the clinical manifestations of them; most of the cases of BilIN have been incidentally discovered in the cases with other problems such as hepatolithiasis, biliary strictures, biliary cysts, papillary adenoma, or primary sclerosing cholangitis.[24-29] Some others have been found in the nontumoral biliary tracts of the patients with CCA such as surgical margins.[27] Rare presentations such as hemobilia have also been reported in individual case reports.[25] Also, there are reports of the discovery of BilIN in nonbiliary conditions such as alcoholic cirrhosis and hepatitis C–related cirrhosis without producing any related clinical findings.[30] Biliary intraepithelial neoplasia has been considered as a flat type of intraepithelial neoplasia or flat dysplasia, which unlike the tumoral counterpart of intraepithelial neoplasia such as colonic adenoma will not produce significant clinical manifestations.[31]

Intraductal papillary neoplasm of the bile duct is a tumoral type of precursor lesions of CCA which causes papillary ingrowth within the biliary tract and can produce mass lesions and clinical manifestations related to the mass and biliary dilatation.[31] These lesions have been reported to produce symptoms such as abdominal discomfort, abdominal pain, pyrexia, anorexia, and even jaundice.[32,33] Therefore, although the main precursors of the CCA are the lesions of BilIN, however, BilINs are mostly asymptomatic but less common IPNB cases can produce clinical sign and symptoms related to tumoral dysplasia.[29-33]

### *Imaging findings*

Biliary intraepithelial neoplasias are flat and microscopic epithelial lesions, so they cannot be detected by the image analysis, and the diagnosis completely depends on pathological examination.[31] The utility of conventional imaging modalities as a diagnostic method is very limited. However, techniques such as diffusion-weighted imaging or magnetic resonance cholangiopancreatography can detect papillary lesions within the duct, ie, IPNB. Uncommon mucin-producing variant of IPNB can cause biliary dilatation which is visible by magnetic resonance imaging. Endoscopic procedures such as endoscopic retrograde cholangiopancreatography or cholangioscopy have also been considered as modalities which can be useful for the diagnosis of BilINs.[34,35,36] Although cross-sectional imaging studies are necessary for the diagnosis of early biliary tract neoplasia, so far, these are not recommended as screening test in high-risk patients such as primary sclerosing cholangitis or hepatolithiasis.[4]

### *Molecular pathogenesis*

It has been proved that development of CCA occurs through multistep carcinogenesis. As it has been mentioned in previous paragraphs, there are 2 main types of precursors in CCA (BilIN and IPNB). This multistep carcinogenesis seems to be different in these 2 types of precursors, but there are many controversial issues regarding the difference between these two. In some studies, different molecules which are involved in cell cycle and cell proliferation have been compared in these 2 lesions, ie, P21, P53, and cyclin D1. Studies have shown that all the 3 are upregulated in both lesions.[37] However, mucin profile of BilIN and IPNB is different, for example, MUC-1 shows overexpression in BilIN, but it is negative in IPNB. This will cause more tubular type of CCA which is more aggressive than colloid carcinoma which occurs most commonly secondary to IPNB.[38] This mucin and cytokeratin profile will be described in more detail in the next section of immunohistochemical findings in precursor lesions.

Another study has shown that KRAS mutation occurs in more than 30% of cases with BilIN and seems to be an early molecular event during the progression of BilIN to CCA in patients with hepatolithiasis, whereas p53 overexpression is a late molecular event.[39]

Recently, the role of autophagy has been proposed in precursor lesions of CCA, ie, proteins related to autophagy are upregulated in early stages of BilIN (ie, BilIN 1, 2) in the patients with hepatolithiasis.[40]

### *Pathologic and immunohistochemical findings of BilIN*

Biliary intraepithelial neoplasia is a microscopic finding and grossly the pathologic changes are very subtle such as mild granularity and thickening of the mucosa.[4] Microscopically, according to the differences in cellular and nuclear features as well as architectural findings, BilIN is divided into 3 types: BilIN I, II, III.

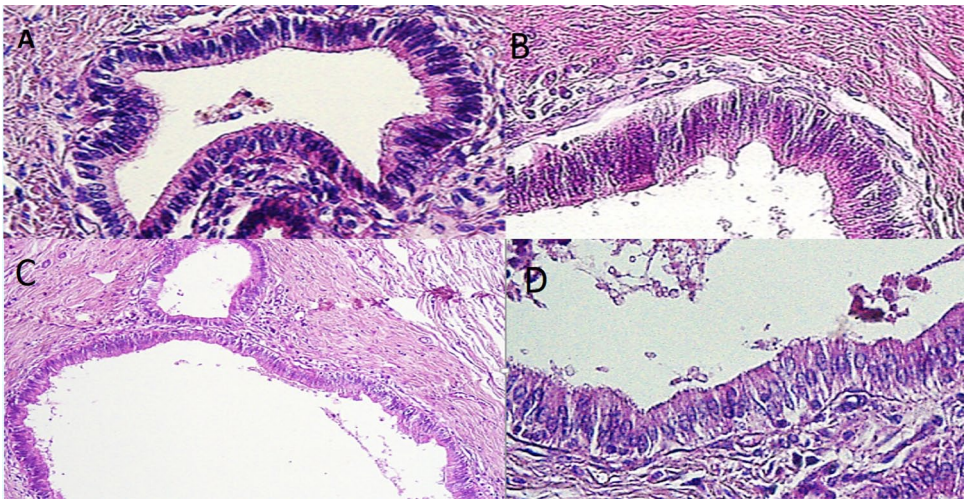


**Figure 1.** Sections from bile duct (A, B) with BilIN I and (B, C) BilIN II. (H&EX250)

*BilIN I.* This lesion can be flat or micropapillary. This can be counterpart of low-grade dysplasia in gastrointestinal epithelial cells, ie, the nuclei are located within the lower two-thirds of the epithelium. There is focal nuclear and mild nuclear pseudostratification with increased nuclear to cytoplasmic ratio as well as mild irregularities of nuclear membrane and nuclear elongation. The number of mitosis is very low and rare.[4,5] Nuclear sizes and shapes are almost uniform, and there is no pleomorphism. The presence of large nuclei is mostly in favor of higher grades of BilIN, ie, BilIN-Ii or BilIN-III[1] (Figure 1A and B).

*BilIN II.* This lesion can also be seen in flat or micropapillary configuration. There is loss of cellular polarity and nuclear pseudostratification; however, the nuclei are hyperchromatic and enlarged and can reach the luminal surface. Some nuclear membrane irregularities and variations in nuclear sizes are present. Mitosis is not very common; however, there is more mitosis compared with BilIN I.[4,5,41] (Figure 1C and D).

*BilIN III.* This lesion can also be seen in flat or micropapillary shape. This lesion is the counterpart of carcinoma in situ in other mucosal parts of the body. There is marked loss of cellular polarity and nuclear stratification as well as cribriform pattern is present. The nuclei can reach the luminal surface with enlargement and hyperchromasia. Nuclear membrane irregularities are also present.[4,5,41]

Immunohistologically, MUC5AC, as an early-phase protein, and Ki67, as a "late-phase" expression, were identified with increasing degrees of dysplasia. Early-phase cell cycle proteins, p16 (decrease) and p21 (increase), altered with increasing degrees of dysplasia.[41]

It has been shown that S100P, which is negative in normal bile duct epithelial cells, can be positive in preinvasive BilIN as well as CCA. This can be used for confirmation of premalignant nature of BilIN.[42,43]

As a whole it seems that characteristic immunohistochemical pattern in BilIN is reactive with CK7, CK19, CK20, MUC5AC+, MUC1 and negative with MUC2 and MUC6.[44]

*Histologic differential diagnosis of BilIN.* Differential diagnosis of BilIN from reactive atypia is very important, especially in the patients with primary sclerosing cholangitis and diseases of biliary tract such as hepatolithiasis. Inflamed bile duct epithelial cells can reveal mild nuclear atypia and hyperchromasia (reactive atypia). So, it can be very difficult to differentiate BilIN I from reactive atypia. However, in reactive atypia, variable degree of acute inflammation is present in the stromal background. Therefore, diagnosis of BilIN should be made with caution in the presence of acute inflammation.[5]

## *Pathologic and immunohistochemical findings of IPNB*

Intraductal papillary neoplasm of the bile duct has been defined in 2010 classification as the biliary counterpart of the intraductal papillary mucinous neoplasm (IPMN) of the pancreas. This lesion is currently considered as an important precursor of CCA, comprising 9% to 38% of CCAs in eastern countries.[12]

Grossly it is seen connected to a dilated bile ducts as white- to red-colored soft, papillary lesions without invasion covered by epithelial cells which can be pancreatobiliary, intestinal, gastric, and oncocytic.[3] In the eastern countries, intestinal subtype is more common which is mostly secondary to hepatolithiasis and clonorchiasis. In Western countries, pancreaticobiliary histologic subtype is more common and oncocytic and gastric types are very rare.[12] These types of epithelial linings are very similar to IPMN of pancreas; however, in contrast to IPMN, mucin production is rarely seen in IPNB.[12] According to the degree of dysplasia of the epithelial lining, IPNB is subdivided into IPNB with low, intermediate, and high grade of dysplasia which is very

similar to histologic classification of BilIN.[3] It seems that the biliary tumors, used to be called as papillomatosis and papillary adenocarcinoma, should now be described as an IPNB.[45]

Immunohistochemical studies of IPNB shows that cyclin D1 and p21 expression increase in IPNB just as it does in BilIN. Another protein of cell cycle is p16 which shows aberrant expression in IPNB.[12] Studies regarding inactivation of p53 in different grades of dysplasia in IPNB are controversial; however, most of them has shown increased expression with increasing grade of dysplasia and with invasion. This seems to be different for KRAS mutations which are more common in lower grades of dysplasia and early lesions.[12] There seems to be a correlation between expression of MUC antigens and patient survival. Previous studies on the expression of MUC1 and MUC2 in bile duct tumors indicated that patients with invasive CCA and poor outcome were MUC1-positive and MUC2-negative. Accordingly, many patients with noninvasive IPNB and favorable outcome have been MUC1-negative and MUC2-positive.[46]

*Histologic differential diagnosis of IPNB.* Biliary intraepithelial neoplasia is a microscopic lesion of bile ducts without grossly visible lesion, so it could be differentiated from the IPNB.[46]

### *Treatment of BilIN and IPNB*

There are very few reports about the treatment of these 2 precursors and currently no guideline is available. Most of the recommendations have been based on individual cases and experiences. In one study, presence of BilIN in the surgical margin of a case of CCA has not been resected and no complication has occurred in the course of the disease[28]; however, in most of the previous studies, lesion excision by surgery or laparoscopy has been recommended as the treatment of choice with successful results.[27,47,48] Definitely, more cases and cohort studies are necessary for better evaluation and understanding of these precursor lesions.

## Conclusions

There are increasing literature regarding precursor lesions in different organs such as cervix, gastrointestinal tract, and genitourinary tract. However, there are not so many studies about the precursor lesions of CCA which is an aggressive, poor prognostic, and life-threatening cancer. Surgeons, oncologists, hepatologists, radiologists, and pathologists should be familiar with precursors of CCA, ie, BilIN and IPNB, especially in the patients with ongoing inflammation, fibrosis, and destruction of bile duct epithelial cells such as primary sclerosing cholangitis, hepatolithiasis, and parasitic infestations.

## Author Contributions

Bita Geramizadeh: Idea, coceot, literature search and writing the manuscript.

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## REFERENCES


1. Zen Y, Adsay NV, Bardadin K, et al. intraepithelial neoplasia: an international interobserver agreement study and proposal for diagnostic criteria. *Mod Pathol*. 2007;20:701-709.
2. Naito Y, Kusano H, Nakashima O, et al. Intraductal neoplasm of the intrahepatic bile duct: clinicopathological study of 24 cases. *World J Gastroenterol*. 2012;18:3673-3680. doi:10.3748/wjg.v18.i28.3673.
3. Schlitter AM, Born D, Bettstetter M, et al. Intraductal papillary neoplasms of the bile duct: stepwise progression to carcinoma involves common molecular pathways. *Mod Pathol*. 2014;27:73-86. doi:10.1038/modpathol.2013.112.
4. Ainechi S, Lee H. Updates on precancerous lesions of the biliary tract: biliary precancerous lesion. *Arch Pathol Lab Med*. 2016;140:1285-1289.
5. Sato Y, Sasaki M, Harada K, et al. Pathological diagnosis of flat epithelial lesions of the biliary tract with emphasis on biliary intraepithelial neoplasia. *J Gastroenterol*. 2014;49:64-72. doi:10.1007/s00535-013-0810-5.
6. Geramizadeh B, Ghavvas R, Kazemi K, Shamsaeefar A, Nikeghbalian S, Malekhosseini SA. Cholangiocarcinoma secondary to primary sclerosing cholangitis in explanted livers: a single-center study in the south of Iran. *Hepat Mon*. 2015;15:e33626. doi:10.5812/hepatmon.33626.
7. Akbarzadeh L, Geramizadeh B, Kazemi K, Nikeghbalian S, Malekhosseini S. Biliary intraepithelial neoplasia (BilIN) in primary sclerosing cholangitis: the first report from Iran. *Hepat Mon*. 2016;16:e38726. doi:10.5812/hepatmon.38726.
8. Rougemont AL, Genevay M, McKee TA, Gremaud M, Mentha G, Rubbia-Brandt L. Extensive biliary intraepithelial neoplasia (BilIN) and multifocal early intrahepatic cholangiocarcinoma in non-biliary cirrhosis. *Virchows Arch*. 2010;56:711-717. doi:10.1007/s00428-010-0899-3.
9. Strazzabosco M, Fabris L. Functional anatomy of normal bile ducts. *Anat Rec (Hoboken)*. 2008;291:653-660. doi:10.1002/ar.20664.
10. Nakanuma Y. A novel approach to biliary tract pathology based on similarities to pancreatic counterparts: is the biliary tract an incomplete pancreas. *Pathol Int*. 2010;60:419-429. doi:10.1111/j.1440-1827.2010.02543.x.
11. Nakanuma Y, Kakuda Y. Pathologic classification of cholangiocarcinoma: new concepts. *Best Pract Res Clin Gastroenterol*. 2015;29:277-293. doi:10.1016/j.bpg.2015.02.006.
12. Ettel M, Eze O, Xu R. Clinical and biological significance of precursor lesions of intrahepatic cholangiocarcinoma. *World J Hepatol*. 2015;7:2563-2570. doi:10.4254/wjh.v7.i25.2563.
13. Blanc JF, Bernard PH, Carles J, Le Bail B, Balabaud C, Bioulac-Sage P. Cholangiocarcinoma arising in Von Meyenburg complex associated with hepatocellular carcinoma in genetic haemochromatosis. *Eur J Gastroenterol Hepatol*. 2000;12:233-237. doi:10.1097/00042737-200012020-00016.
14. Jain D, Sarode VR, Abdul-Karim FW, Homer R, Robert ME. Evidence for the neoplastic transformation of Von-Meyenburg complexes. *Am J Surg Pathol*. 2000;24:1131-1139. doi:10.1097/00000478-200008000-00011.
15. Song JS, Lee YJ, Kim KW, Huh J, Jang SJ, Yu E. Cholangiocarcinoma arising in von Meyenburg complexes: report of four cases. *Pathol Int*. 2008;58:503-512. doi:10.1111/j.1440-1827.2008.02264.x.
16. Pinho AC, Melo RB, Oliveira M, et al. Adenoma-carcinoma sequence in intrahepatic cholangiocarcinoma. *Int J Surg Case Rep*. 2012;3:131-133. doi:10.1016/j.ijscr.2012.01.002.
17. Sato H, Nakanuma Y, Kozaka K, Sato Y, Ikeda H. Spread of hilar cholangiocarcinomas via peribiliary gland network: a hither-to-unrecognized route of periductal infiltration. *Int J Clin Exp Pathol*. 2013;6:318-322.
18. Nakanishi Y, Zen Y, Hirano S, et al. Intraductal oncocytic papillary neoplasm of the bile duct: the first case of peribiliary gland origin. *J Hepatobiliary Pancreat Surg*. 2009;16:869-873.
19. Chinchilla-Lopez P, Aguilar-Olivos NE, Garcia-Gomez J, et al. Risk factors, and survival of patients with intrahepatic cholangiocarcinoma. *Ann Hepatol*. 2017;16:565-568. doi:10.5604/01.3001.0010.0293.
20. Goral V. Cholangiocarcinoma: new insights. *Asian Pac J Cancer Prev*. 2017;18:1469-1473.
21. Gupta A, Dixon E. Epidemiology and risk factors: intrahepatic cholangiocarcinoma. *Hepatobiliary Surg Nutr*. 2017;6:101-104. doi:10.21037/hbsn.2017.01.02.
22. Yamada K, Kumagai S, Endo G. Chemical exposure levels in printing workers with cholangiocarcinoma (second report). *J Occup Health*. 2015;57:245-252. doi:10.1539/joh.14-0239-OA.
23. Clements O, Eliahoo J, Kim JU, Taylor-Robinson SD, Khan SA. Risk factors for intrahepatic and extrahepatic cholangiocarcinoma: a systematic review and meta-analysis. *J Hepatol*. 2020;72:95-103.
24. Ohtsuka M, Shimizu H, Kato A, Yoshitomi H, Furukawa K, Tsuyuguchi T. Intraductal papillary neoplasms of the bile duct. *Int J Hepatol*. 2014;2014:459091. doi:10.1155/2014/459091.
25. Umeda R, Nakamura Y, Masugi Y, et al. Hemobilia due to biliary intraepithelial neoplasia associated with Zollinger-Ellison syndrome. *Clin J Gastroenterol*. 2012;5:189-194. doi:10.1007/s12328-012-0296-3.
26. Jung W, Kim BH. Biliary intraepithelial neoplasia: a case with benign biliary stricture. *Korean J Hepatol*. 2011;17:328-330. doi:10.3350/kjhep.2011.17.4.328.

27. Matthaei H, Lingohr P, Strasser A, et al. Biliary intraepithelial neoplasia (BilIN) is frequently found in surgical margins of biliary tract cancer resection specimens but has no clinical implications. *Virchows Arch*. 2015;466:133-141. doi:10.1007/s00428-014-1689-0.
28. Umemura A, Suto T, Sasaki A, et al. Pure laparoscopic left hemihepatectomy for hepatic peribiliary cysts with biliary intraepithelial neoplasia. *Case Rep Surg*. 2016;2016:7236427. doi:10.1155/2016/7236427.
29. Horn A, Meves V, Behrens A, Jenssen C, Vieth M, Pohl J. Papillary adenoma arising from an extensive biliary low-grade intraepithelial neoplasia. *Gastrointest Endosc*. 2016;83:836-837. doi:10.1016/j.gie.2015.10.007.
30. Wu TT, Levy M, Correa AM, Rosen CB, Abraham SC. Biliary intraepithelial neoplasia in patients without chronic biliary disease: analysis of liver explants with alcoholic cirrhosis, hepatitis C infection, and noncirrhotic liver diseases. *Cancer*. 2009;115:4564-4575. doi:10.1002/cncr.24471.
31. Jang KT, Ahn S. Tumoral versus flat intraepithelial neoplasia of pancreatobiliary tract, gallbladder, and ampulla of Vater. *Arch Pathol Lab Med*. 2016;140:429-436. doi:10.5858/arpa.2015-0319-RA.
32. Terada T. Non-invasive intraductal papillary neoplasms of the common bile duct: a clinicopathologic study of six cases. *Int J Clin Exp Pathol*. 2012;5:690-697.
33. Kim HJ, Kim JS, Kim BH, Bak YT. Clinicopathologic study of biliary intraepithelial neoplasia in cholangiocarcinoma. *Dig Surg*. 2018;35:116-120. doi:10.1159/000475848.
34. Kubo S, Takemura S, Sakata C, Urata Y, Nishioka T, Nozawa A. Changes in laboratory test results and diagnostic imaging presentation before the detection of occupational cholangiocarcinoma. *J Occup Health*. 2014;56:317-322.
35. Joo I, Lee JM. Imaging bile duct tumors: pathologic concepts, classification, and early tumor detection. *Abdom Imaging*. 2013;38:1334-1350.
36. Chapman R, Fevery J, Kalloo A, et al. Diagnosis and management of primary sclerosing cholangitis. *Hepatology*. 2010;51:660-678.
37. Nakanishi Y, Zen Y, Kondo S, Itoh T, Itatsu K, Nakanuma Y. Expression of cell cycle-related molecules in biliary premalignant lesions: biliary intraepithelial neoplasia and biliary intraductal papillary neoplasm. *Hum Pathol*. 2008;39:1153-1161. doi:10.1016/j.humpath.2007.11.018.
38. Hsu M, Sasaki M, Igarashi S, Sato Y, Nakanuma Y. KRAS and GNAS mutations and p53 overexpression in biliary intraepithelial neoplasia and intrahepatic cholangiocarcinomas. *Cancer*. 2013;119:1669-1674. doi:10.1002/cncr.27955.
39. Zen Y, Sasaki M, Fujii T, Chen TC, Chen MF, Yeh TS. Different expression patterns of mucin core proteins and cytokeratins during intrahepatic cholangiocarcinogenesis from biliary intraepithelial neoplasia and intraductal papillary neoplasm of the bile duct—an immunohistochemical study of 110 cases of hepatolithiasis. *J Hepatol*. 2006;44:350-358.
40. Sasaki M, Nitta T, Sato Y, Nakanuma Y. Autophagy may occur at an early stage of cholangiocarcinogenesis via biliary intraepithelial neoplasia. *Hum Pathol*. 2015;46:202-209. doi:10.1016/j.humpath.2014.09.016.
41. Sato Y, Harada K, Sasaki M, Nakanuma Y. Histological characteristics of biliary intraepithelial neoplasia-3 and intraepithelial spread of cholangiocarcinoma. *Virchows Arch*. 2013;462:421-427. doi:10.1007/s00428-013-1384-6.
42. Aishima S, Iguchi T, Fujita N, et al. Histological and immunohistological findings in biliary intraepithelial neoplasia arising from a background of chronic biliary disease compared with liver cirrhosis of non-biliary aetiology. *Histopathology*. 2011;59:867-875. doi:10.1111/j.1365-2559.2011.04011.x.
43. Nakanuma Y, Uchida T, Sato Y, Uesaka K. An S100P-positive biliary epithelial field is a preinvasive intraepithelial neoplasm in nodular-sclerosing cholangiocarcinoma. *Hum Pathol*. 2017;60:46-57. doi:10.1016/j.humpath.2016.10.003.
44. Aishima S, Nishihara Y, Tsujita E, et al. Biliary neoplasia with extensive intraductal spread associated with liver cirrhosis: a hitherto unreported variant of biliary intraepithelial neoplasia. *Hum Pathol*. 2008;39:939-947. doi:10.1016/j.humpath.2007.10.031.
45. Ohtsuka M, Kimura F, Shimizu H, et al. Similarities and differences between intraductal papillary tumors of the bile duct with and without macroscopically visible mucin secretion. *Am J Surg Pathol*. 2011;35:512-521. doi:10.1097/PAS.0b013e3182103f36.
46. Kloek JJ, van der Gaag NA, Erdogan D, et al. A comparative study of intraductal papillary neoplasia of the biliary tract and pancreas. *Hum Pathol*. 2011;42:824-832. doi:10.1016/j.humpath.2010.09.017.
47. Wang W, Chen W, Li K, Wang J. Successful treatment of biliary intraepithelial neoplasia in the common bile duct via local excision: a case report. *Oncol Lett*. 2016;11:3142-3144.
48. Zarei M, Shamsaeefar A, Kazemi K, Dehghani M, Malekhosseini SA, Geramizadeh B. Biliary inraepithelial neoplasia in non-biliary cirrhosis—report from 100 explanted livers: a single center experience. *Clinical Pathology*. 2019;12:1-4.