

Review

Expression Profiles of MUC Mucin Core Protein in the Intrahepatic Biliary System: Physiological Distribution and Pathological SignificanceMotoko Sasaki¹, Hiroko Ikeda¹ and Yasuni Nakanuma¹¹Department of Human Pathology, Kanazawa University Graduate School of Medicine, Kanazawa 920–8640, Japan

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Mucin secreted by mucosal epithelial cells plays a role in the protection of the mucosal surface and also is involved in pathological processes. So far, MUC1–4, 5AC, 5B, 6–8, 11–13 and 15–17 genes coding the backbone mucin core protein have been identified in humans. Their diverse physiological distribution and pathological alterations have been reported. We have studied the expression profiles of MUC genes in the intrahepatic biliary system in developmental, normal and diseased livers using immunohistochemistry and *in situ* hybridization. Fetal intrahepatic bile ducts and ductal plates frequently express MUC1, while intrahepatic large bile ducts after birth express mainly MUC3 mucin. In hepatolithiasis, MUC5AC (gastric foveolar epithelial type) and MUC6 (pyloric gland

type) mucins are newly expressed in surface epithelial cells and proliferated peribiliary glands, respectively, and the expression of gel-forming mucin may play a role in lithogenesis. Most biliary epithelial dysplasias and cholangiocarcinomas associated with hepatolithiasis expressed MUC5AC, suggesting that intrahepatic bile ducts express the gastric foveolar phenotype during carcinogenesis. In addition, intestinal metaplasia, intraductal papillary tumor and mucinous carcinoma, characterized by MUC2 expression, are occasionally associated with hepatolithiasis in a CDX2 homeobox gene-dependent manner. These findings may suggest the presence of intestinal metaplasia-related carcinogenesis in the intrahepatic biliary system as in the stomach.

Key words: MUC mucin core protein, hepatolithiasis, cholangiocarcinoma, biliary epithelial dysplasia, CDX2

I. Introduction

Mucins are heavily glycosylated, generally high molecular weight proteins that are synthesized by the epithelial cells in many organs, such as the gastrointestinal (GI), respiratory and genito-urinary tract, and play a role in the protection of mucosal surface. They consist of protein backbone structures (apomucins) and many carbohydrate side chains. There are two structurally and functionally distinct classes

of mucins: secreted gel-forming mucins (MUC2, MUC5AC, MUC5B and MUC6) and transmembrane mucins (MUC1, MUC3, MUC4, MUC12 and MUC17), although the products of some MUC genes do not fit well into either class (MUC7, MUC8, MUC9, MUC13, MUC15 and MUC16) (Table 1). Each MUC mucin shows a characteristic distribution in organs and cell types [4, 6, 7] (Table 1). In these MUC mucins, MUC1 mucin is widely distributed in epithelial cells, and the epitope of epithelial membrane antigen (EMA), a common immunohistochemical marker to detect epithelial cells and carcinomas, is on the glycosylated form of the MUC1 mucin. Altered mucin gene expression has been reported in inflammatory diseases and carcinomas of the GI tract and breast [4, 6, 7]. Accumulating data suggest

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Table 1. Human MUC genes

	Locus	Type	Expression sites
MUC1	1q21	Membrane	Mammary glands, pancreas, etc.
MUC2	11p15.5	Secreted	Colon, trachea
MUC3	7q22	Membrane*	Small intestine, gallbladder
MUC4	3q29	Membrane*	Trachea, stomach, salivary gland
MUC5AC	11p15.5	Secreted	Stomach (foveolar), trachea
MUC5B	11p15.5	Secreted	Stomach, trachea, gallbladder
MUC6	11p15.5	Secreted	Stomach (pyloric gland), trachea
MUC7	4q13-21	Unclassified	Salivary glands
MUC8	12q24	Unclassified	Trachea (submucosal gland)
MUC11 & 12	7q22	Membrane*	Colon
MUC13	3q13.3	Unclassified*	Colon, trachea, hematopoietic
MUC15	11p14.3	Unclassified	Colon, breast, small intestine
MUC16	19p14.3	Unclassified	Ovarian cancer, ocular surface
MUC17	7q22	Membrane*	Duodenum, transverse colon

Membrane, membrane-binding; *, with EGF-like domain.

that each MUC mucin has different properties, for example, the presence of an EGF-like domain, a transmembrane region and a gel-forming capacity, and may have diverse functions. Therefore, the altered expression of MUC mucin appears to be somehow involved in the pathogenesis of inflammatory diseases and in tumor biology. For instance, in the over-expression of MUC1 in cancer cells, these rigid mucin glycoproteins play a role in metastasis by inhibiting tumor cell adhesion and allowing tumor cells to escape from immune surveillance. MUC4 was recently proved to be a novel intramembrane ligand for receptor tyrosine kinase ErbB2 (HER-2) [9]. Furthermore, MUC4 is also involved in the regulation of p27. MUC4 is considered to be a tumor-associated molecule from the potential role of MUC4 as a marker for adenocarcinoma of the pancreas. The expression of MUC4 in intrahepatic cholangiocarcinoma (ICC) of the mass-forming type is a new independent factor for poor prognosis [27].

Intrahepatic biliary system

The intrahepatic ducts, which link to the bile canaliculi at the proximal side and to the extrahepatic ducts at the distal site, are a route for the excretion of bile synthesized by hepatocytes. The intrahepatic bile ducts comprise a complex 3-dimensional network of conduits within the liver starting with Hering canals and ending at the hepatic hilum. The intrahepatic bile ducts are lined by specialized epithelial cells called biliary epithelial cells (BECs) or cholangiocytes [10]. Hepatocytes produce primary hepatic bile, which percolates through the intrahepatic bile ducts. During this journey, bile is modified by BECs via a series of secretory and absorptive processes that provide additional bile water (BECs secrete ~40% of the daily bile production in humans) or secrete HCO_3^- to induce the alkaline state [10]. BECs interact with the immune system and microorganisms and are also involved in drug metabolism. To accomplish these functions, BECs display morphological and functional

heterogeneity along the biliary tree. In normal livers, the smallest ductules are rimmed by no more than a few minimally differentiated cuboidal epithelial cells. The BECs lining progressively larger bile ducts gradually become more highly differentiated, mucus-secreting cells.

The intrahepatic biliary system is known to share many physiological and structural characteristics of the GI tract, including the contact of the mucosal surface with toxic substances and the intraluminal secretion of mucin.

Classification of the intrahepatic biliary tree

The intrahepatic biliary tree is divided into intrahepatic large bile ducts and small bile ducts [16]. The large bile ducts are grossly visible, and are characterized by the presence of a fibrous ductal wall and surrounding intrahepatic peribiliary glands, and correspond to the first branches of area ducts and the right and left hepatic ducts, segmental ducts, and their first branches. Intrahepatic peribiliary glands are consistently located around the intrahepatic large bile ducts [28]. Intrahepatic small bile ducts, which are recognizable under light microscopy, correspond to the septal and interlobular bile ducts as well as bile ductules [16]. Intrahepatic peribiliary glands located around the large bile ducts are divided into intramural and extramural glands [28]. Intramural glands are simple tubular mucous glands scattered within the duct wall [28]. Extramural glands are branched tubulo-alveolar sero-mucous glands located in the peribiliary connective tissue that drain into the bile duct lumen by way of their own conduits [28].

In this review, we will focus on the expression profile of MUC mucin in the intrahepatic biliary system in 1) fetal and adult livers, 2) diseased livers, especially in the liver with hepatolithiasis, and 3) pre-neoplastic lesions and cholangiocarcinomas associated with hepatolithiasis.

II. Detection of MUC mRNA and Protein Expression

In situ hybridization is widely used to detect the expression and distribution of MUC mRNA in tissue section. Both radioisotope-labeled and digoxigenin (non-isotope)-labeled oligonucleotides are usually used as probes [1, 23]. Oligonucleotides encompassing the tandem repeat sequence of each MUC gene were synthesized [1, 23]. The specificity of each oligonucleotide probe was confirmed by sequence database search (NCBI). MUC mucin is abundantly produced, when expressed, and multiple tandem repeat regions are present in each MUC mRNA. Positive signals were clearly detected in the MUC expressing cells on tissue sections. Both frozen tissue sections and formalin-fixed, paraffin-embedded tissue sections are available.

The expression of MUC mucins was detected by immunohistochemical methods. For MUC1 mucin expression, several mouse monoclonal antibodies detecting different glycosylated forms are available. DF3 (CA15-3; Toray-Fuji Bionics, Tokyo, Japan), and NCL-MUC1-CORE (Novocastra, New Castle, UK) recognize MUC1 core peptide. DF3 binding to MUC1 mucin was reportedly enhanced by the presence of carbohydrate. EMA (Dako, Santa Barbara, CA) and NCL-MUC-1-GP (Novocastra) recognize the glycosylated form of MUC1. For other types of MUC mucin expression, rabbit or chicken polyclonal antibodies raised against synthetic tandem repeat peptide of each MUC mucin have been used [21–23]. For MUC2, MUC5AC and MUC6 mucin detection, monoclonal antibodies Ccp58, CLH5 and CLH2, respectively, are commercially available (Novocastra) and have been used recently. For MUC3 and MUC5B mucin expression, monoclonal antibody M3.1 and goat polyclonal antibody are available from Santa Cruz Biotech (Santa Cruz, CA).

III. MUC Expression in Fetal and Adult Livers

The expression profile of MUC mucin in fetal and normal postnatal (adult) livers is summarized in Figure 1. In fetal liver, the process of intrahepatic bile duct development is divided into a ductal plate stage, a remodeling ductal plate stage, and a remodeled bile duct stage [32]. The ductal plate is an excess structure consisting of a double-layered cylinder at the peripheral hepatic parenchyma. The ductal plate stage is characterized by the formation of these ductal plates. The remodeling ductal plate stage is characterized by the incorporation of ductal plate cells into the mesenchyma, as well as by the gradual disappearance of the ductal plates [32]. The remodeled bile duct stage is characterized by new bile ducts in the portal tracts as well as the disappearance of the ductal plate. The development of intrahepatic bile ducts proceeded from the hilar to the peripheral portions [32].

In the fetal liver, new bile ducts in the portal tracts, either at the hilar level or the peripheral level, frequently expressed MUC1 mucin, in both the unglycosylated (detected by monoclonal antibody (MAb) DF3) and glycosylated form (detected by anti-EMA) on their luminal surface [21] (Fig. 2). Ductal plates also focally expressed MUC1 mucin. In addition, the remodeled bile ducts in the fetal liver focally express MUC6 mucin. By contrast, in the adult liver, the BECs of intrahepatic large bile ducts constantly expressed MUC3 mucin, whereas those of small bile ducts did not (Fig. 2) [21, 23, 31]. Although MUC3 is a membrane-binding type mucin, the immunohistochemical MUC3 expression is usually observed in the supranuclear cytoplasm in BECs and carcinomas. This may reflect the soluble form of MUC3 produced by alternative splicing [2]. The unglycosylated form of MUC1 mucin (detected by DF3) is not expressed in adult intrahepatic bile ducts, whereas the glycosylated form of MUC1 mucin (detected by EMA) is expressed in intrahepatic large bile ducts. These

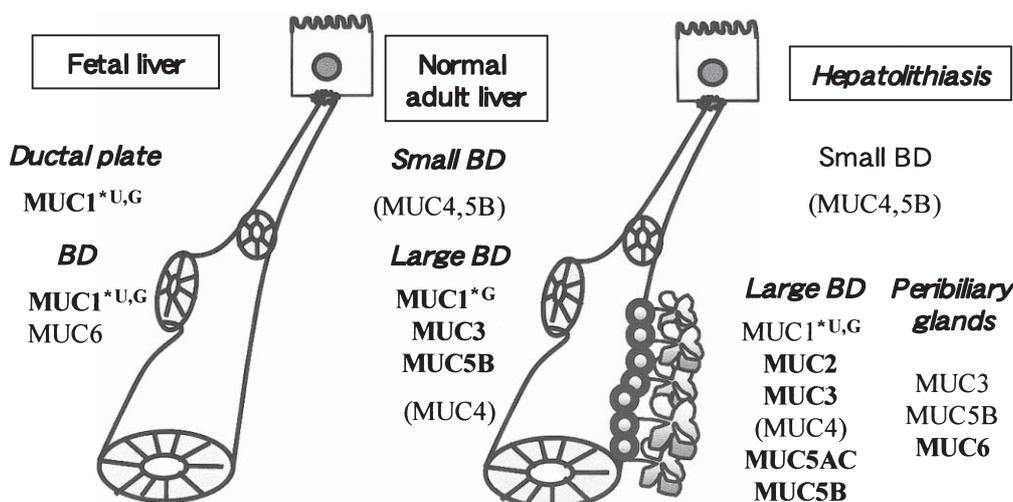


Fig. 1. Expression profile of MUC mucin in fetal and normal adult livers and hepatolithiasis. BD, bile duct. (.), weak and focal expression; *U, G, unglycosylated and glycosylated form; *G, glycosylated form.

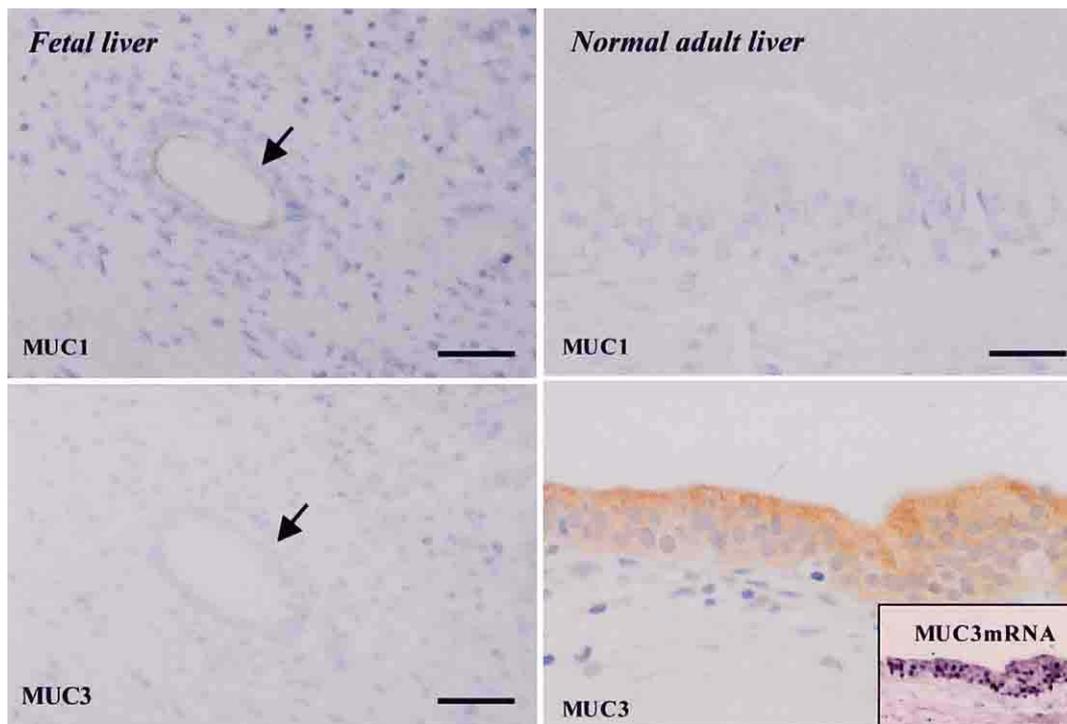


Fig. 2. MUC1 and MUC3 expression in fetal and normal adult livers. Fetal intrahepatic bile ducts (left upper) and ductal plates frequently express MUC1 mucin (unglycosylated form), but not MUC3 (left lower). In contrast, intrahepatic large bile ducts after birth mainly express MUC3 mucin (right lower), whereas the unglycosylated form of MUC1 mucin is not expressed (right upper). Immunohistochemical staining for MUC1 and MUC3 was detected using monoclonal antibody, DF3, and rabbit polyclonal antibody, M3P [18]. Inset, MUC3mRNA expression detected by *in situ* hybridization using digoxigenin-labeled oligonucleotide probe [19] corresponds to MUC3 protein expression. Bars=50 μ m.

findings suggest that the glycosylation status of MUC1 mucin is altered before and after birth. BECs of intrahepatic large and small bile ducts express MUC4 mucin focally and weakly (unpublished data). BECs of intrahepatic large bile ducts express MUC5B constantly, while MUC5B expression is focal and weak in BECs of intrahepatic small bile ducts (unpublished data). MUC2 and MUC5AC mucin were absent in the intrahepatic biliary elements of the fetal and adult livers. These data suggest that BECs switch MUC1 mucin expression before birth to that of MUC3 after birth [21]. The characteristic transition may be similar to the changes in the hepatocellular expression of alpha-fetoprotein and albumin during the perinatal period. The molecular mechanism involved in the induction of MUC3 mucin and the altered glycosylation of MUC1 mucin after birth remain to be addressed.

There are several studies reporting MUC expression in the gallbladder, extrahepatic bile ducts, and the ampulla of Vater [21]. In the fetal gallbladder, MUC3, MUC6, MUC5B and MUC1 are detected by *in situ* hybridization [1]. In the adult gallbladder, the strong expression of MUC3, the moderate expression of MUC5B and MUC6, and the weak expression of MUC1, MUC2 and MUC5AC were detected by *in situ* hybridization [1]. MUC4 mucin was not detected in fetal and adult gallbladder [1]. In the adult extrahepatic bile duct and the ampulla of Vater, the focal expression of

MUC1 and MUC5AC were noted, but MUC2 expression was absent [35]. The expression of MUC3 and MUC5B has not been reported in the extrahepatic bile duct and the ampulla of Vater, so far. The constant expression of MUC3 and MUC5B expression in adult intrahepatic large bile ducts is similar to that in the adult gallbladder (Table 1, Fig. 1).

IV. MUC Expression in the Biliary Tract with Hepatolithiasis

Hepatolithiasis is not rare in East Asia [14, 17], and chronic proliferative cholangitis of the intrahepatic large bile duct with mucus hypersecretion is a key lesion in the process of stone formation [15] (Fig. 3). Histologically, biliary mucosa in the liver with hepatolithiasis shows a marked proliferation of intramural and extramural peribiliary glands accompanied by chronic inflammation and fibrosis (chronic proliferative cholangitis) [15] (Fig. 3). The majority of these stones are of the calcium-bilirubinate type [17], which differs from pure cholesterol and black pigment stones in their composition and etiology [30]. Bacterial infection, bile stasis and an alteration of the bile composition are thought to be responsible for the nucleation, formation and maturation of intrahepatic stones [17]. In the biliary tract with hepatolithiasis, surface epithelial cells express MUC5AC mucin (gastric surface type) and proliferative peribiliary glands express

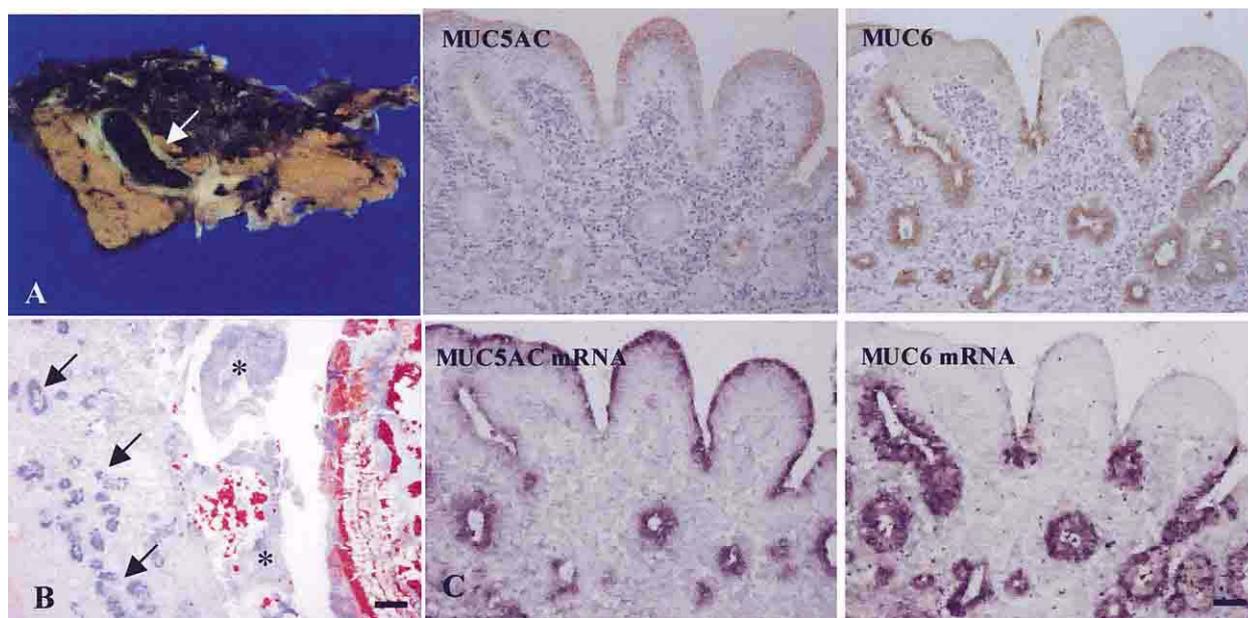


Fig. 3. Gross appearance, mucin histochemistry, and MUC expression in hepatolithiasis. **A)** The gross appearance of hepatolithiasis. An arrow indicates calcium-bilirubinate stones compacted in the intrahepatic large bile ducts. **B)** The intrahepatic large bile duct containing hepatoliths includes proliferated peribiliary glands (arrows) and abundant secreted mucin in the lumen of large bile ducts (astrisk). Alcian blue (pH 2.5)/PAS staining. **C)** In hepatolithiasis, surface epithelial cells and a part of proliferated peribiliary glands express MUC5AC mucin (left). MUC6 mucin (right) is mainly expressed in proliferated peribiliary glands. Upper, immunohistochemical staining for MUC5AC (left) and MUC6 (right) mucin detected with rabbit polyclonal antibody, M5P-b1 and chicken polyclonal antibody, M6P [19]. Lower, MUC5AC (left) and MUC6 (right) mRNA expression detected with *in situ* hybridization using digoxigenin-labelled oligonucleotide probe [19]. Bars=100 μ m (**B**), 50 μ m (**C**).

MUC6 mucin (gastric pyloric gland type) [24] (Fig. 3). The expression profile of MUC mucin in the biliary tract with hepatolithiasis resembles that of the gastric mucosa. The altered MUC expression may play a role in the initiation and progression of hepatolithiasis, since newly-expressed MUC5AC and MUC6 mucin have the capacity for viscous gel-formation [23]. MUC2, colon-type mucin, is also newly expressed in the biliary tract with hepatolithiasis at the site of intestinal metaplasia and hyperplastic epithelia. As shown in Figures 4A and 4D, nuclear expression of the CDX2 homeobox gene, which induces intestinal differentiation, is co-localized with MUC2 expression at the site of intestinal metaplasia. This finding suggests that CDX2 induces the expression of MUC2 mucin in intrahepatic large bile duct in hepatolithiasis [5, 23]. Furthermore, the focal expression of MUC1 and increased expression of MUC3 and MUC5B are also noted in large bile ducts and peribiliary glands in hepatolithiasis. Although the significance of the increased expression of MUC3 mucin remains unclear, the increased expression of MUC5B, which is one of the gel-forming mucins, may contribute to the increased viscosity of the bile and lithogenesis.

The regulatory factors of increased and altered MUC expression have not as yet been fully clarified. Lipopolysaccharide (LPS), a component of cell wall of gram-negative bacteria, up-regulates MUC2 and MUC5AC immediately in cultured mouse BECs via TNF- α and COX-2 related path-

ways [34]. In our preliminary study, the chronic exposure of mouse cultured BECs to LPS and an acidic condition induced the expression of the CDX2 homeobox gene and the following MUC2 expression. This finding may suggest that chronic inflammatory conditions are important for the alteration and over-expression of MUC mucin.

V. Coordinated Expression of Trefoil Factor Family (TFF) and MUC Mucin in the Biliary Tract with Hepatolithiasis

The trefoil factor family (TFF) are mucin-associated proteins that are important for mucosal defense and the repair of the gastrointestinal epithelia [11, 18, 19]. In humans, three types of TFF (TFF1–3) and their characteristic and coordinated distribution together with MUC mucin have been reported. That is, a combination of TFF1 with MUC5AC and that of TFF2 with MUC6 are generated in gastric surface mucous cells and gastric pyloric glands, respectively. TFF3 is co-expressed with MUC2 in intestinal goblet cells, and also co-expressed with MUC5B and MUC8 [33]. The expression of TFF1, TFF2 and TFF3 are augmented markedly in the biliary mucosa in hepatolithiasis in coordination with gel-forming mucin [24, 26]. That is, the expression of TFF1 and TFF2 is almost parallel to the expression of MUC5AC and MUC6, respectively. TFF3 is co-expressed with MUC2 at the site of intestinal metaplasia [16]. Furthermore, TFF3 is

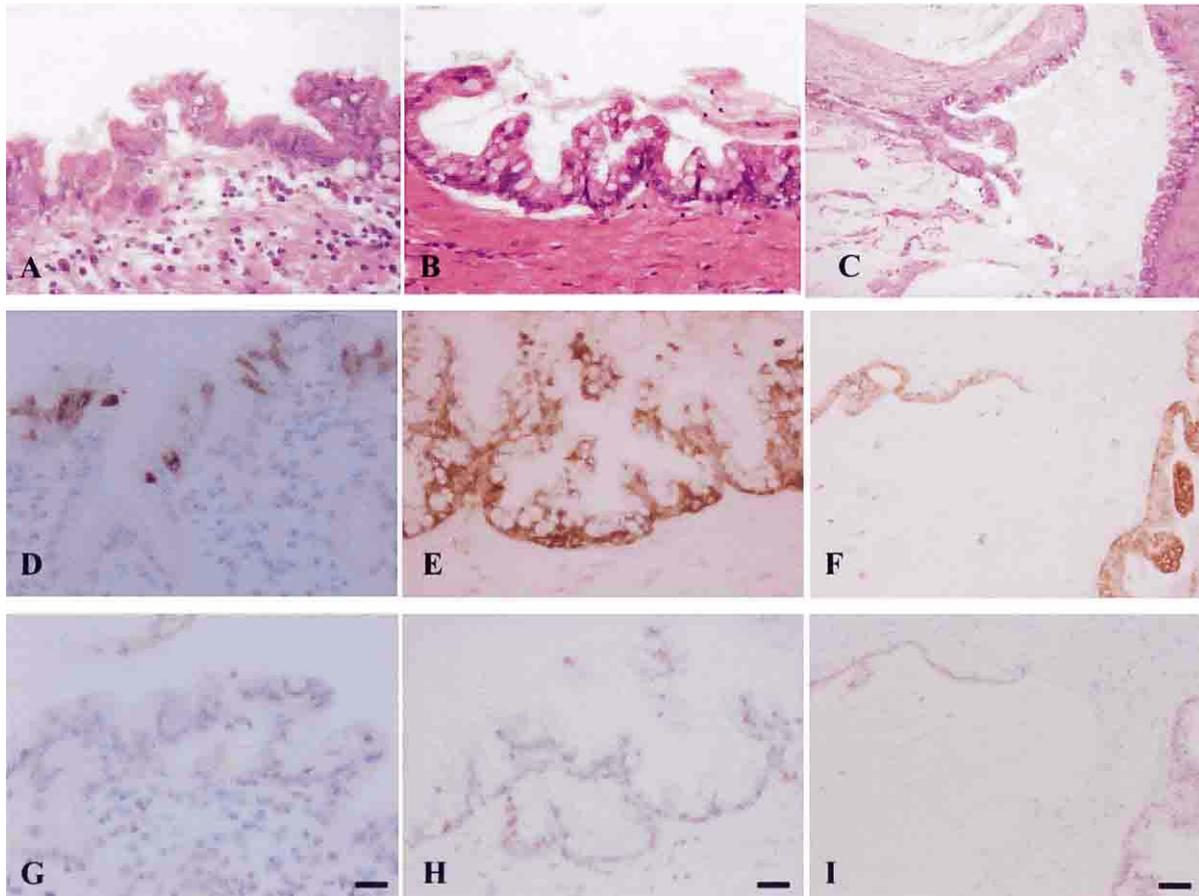


Fig. 4. CDX2-dependent MUC2 expression in intestinal metaplasia, intraductal papillary tumor and mucinous carcinoma associated with hepatolithiasis. Intestinal metaplasia (A), intraductal papillary tumors (B) and mucinous carcinoma (C). MUC2 is expressed in the cytoplasm of goblet cells at the intestinal metaplasia (D), intraductal papillary tumors (E) and mucinous carcinoma (F). CDX2 is expressed in the nuclei of goblet cells at intestinal metaplasia (G), intraductal papillary tumors (H) and mucinous carcinoma (I) corresponding to MUC2 expression. Hematoxylin and eosin (A–C). Immunohistochemical staining for MUC2 (D–F) and CDX2 (G–I). Bars=50 μ m (A, B, D, E, G, H), 100 μ m (C, F, I).

more widely distributed in BECs in large bile ducts and peribiliary glands [16]. It is conceivable that widely distributed TFF3 may be co-expressed with MUC5B. Since TFFs are known to interact with MUC2 and MUC5AC and possibly raise the viscosity of mucin [29], it is likely that over-secreted TFFs in the biliary tract with hepatolithiasis may couple with MUC5AC and MUC2 mucin, increase the viscosity of secreted biliary mucin, and contribute to the formation of hepatoliths [24, 26]. Furthermore, we have reported the augmented expression of mucin-associated molecule that is deleted in malignant brain tumor-1 (DMBT1) [12, 13], which is a candidate of the TFF receptor, in BECs in the biliary tract with hepatolithiasis, and suggested the possible participation of this molecule in lithogenesis [25]. Bovine gallbladder mucin, which has scavenger receptor cysteine-rich (SRCR) domains and accelerates cholesterol crystallization, was identified as an alternative splicing form of DMBT1. In our previous report, DMBT1 protein was frequently detected in hepatic bile samples of hepatolithiasis (50%), but not in the other bile samples [14]. The percent-

age of cholesterol in the intrahepatic calculi was significantly higher in the patients with DMBT-1 positive bile [14]. It is conceivable that the increased expression of MUC mucin, TFFs and DMBT1 cooperate to protect biliary mucosa and participate in lithogenesis in hepatolithiasis.

VI. MUC Expression in Biliary Epithelial Dysplasia and Cholangiocarcinoma

In the patients with hepatolithiasis, intrahepatic cholangiocarcinoma (ICC) is known to develop in approximately 10% of more than 1000 patients with hepatolithiasis patients [8, 14] and the relative risk is at least 30-fold greater when compared with usual ICC without any background liver diseases. Gastric mucosal metaplasia and intestinal metaplasia are common in the intrahepatic biliary tracts, but these changes of the intrahepatic biliary tracts are rare. Biliary epithelial dysplasia is being accepted as a precursor lesion of ICC in the biliary tract with hepatolithiasis. The stepwise development and progression through biliary epithelial

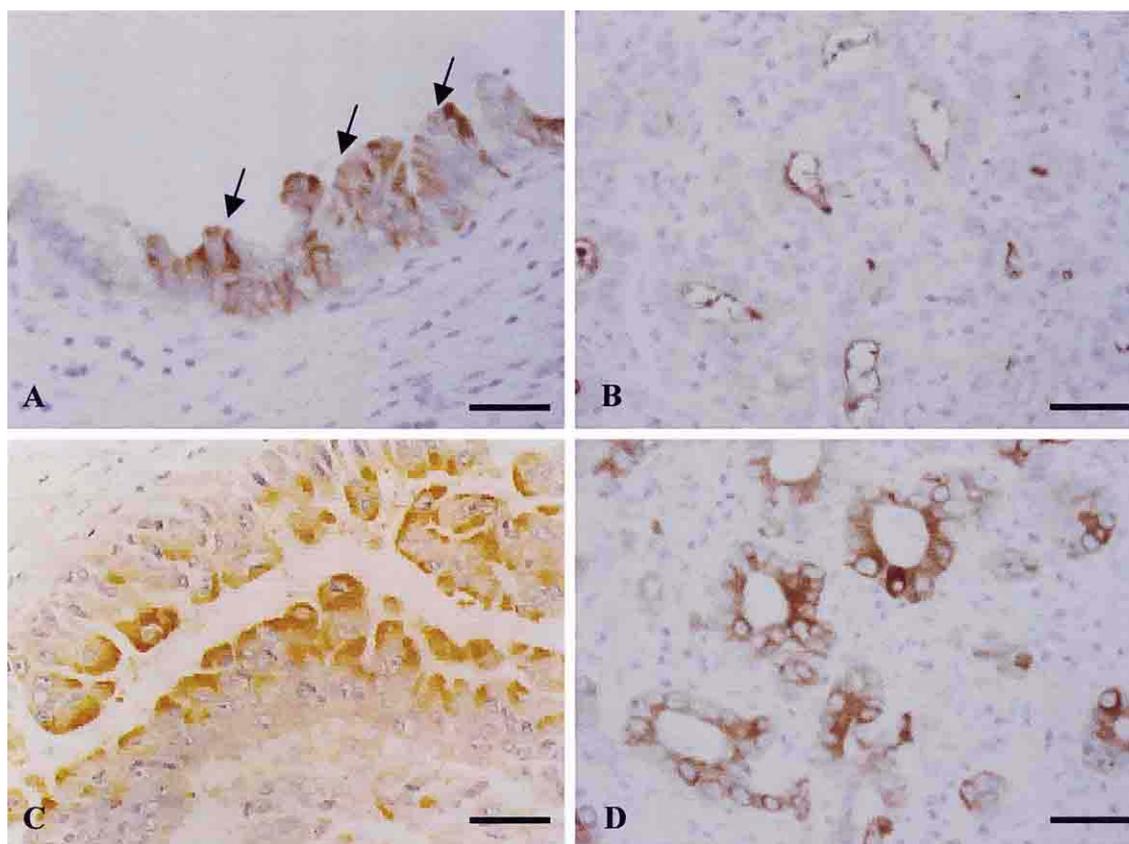


Fig. 5. MUC expression in biliary epithelial dysplasia and intrahepatic cholangiocarcinoma associated with hepatolithiasis. **A)** Biliary epithelial dysplasia strongly express MUC5AC mucin (arrows). **B–D)** Intrahepatic cholangiocarcinoma associated with hepatolithiasis express MUC1 (**B**), MUC3 (**C**) and MUC5AC (**D**) mucin. Immunohistochemical staining for MUC1, MUC3 and MUC5AC. Bars=50 µm.

dysplasia, non-invasive ICC, and invasive ICC has been proposed in hepatolithiasis [14]. In biliary epithelial dysplasia, the up-regulation of MUC5AC mucin coupled with TFF1 has been reported [22, 24] (Fig. 4). This indicates that gastric metaplasia is an early event during the stepwise carcinogenesis of ICC associated with hepatolithiasis. In ICC associated with hepatolithiasis, MUC1, MUC3, MUC4 and MUC5AC mucin are frequently over-expressed [20, 22, 24] (Fig. 4). Especially, the expression of MUC1 is evident in invasive ICC, when compared with non-invasive ICC, and MUC1 expression is associated with poor patient outcome [3]. MUC1 expression on carcinoma cells contributes to the inhibition of cell-cell and cell-substratum interaction and may play a role in the metastasis of carcinoma cells. Rigid MUC1 molecules may be involved in enabling carcinoma cells to escape from immune surveillance. MUC3 expression is seen 57% of ICC associated with hepatolithiasis [18], and the expression is rather frequent in non-invasive ICC. MUC5AC expression is seen in most ICC [26]. The distribution of MUC3 and MUC5AC expression varies between each ICC. The focal expression of MUC6 and MUC5B has also been reported. Figure 6 summarizes the expression profile of MUC mucin during stepwise development and progression through biliary epithelial dysplasia, non-invasive ICC, and invasive ICC in the biliary tract with hepatolithiasis. The

molecular mechanisms involved in the altered regulation of mucin genes and the significance of the altered expression of MUC mucin in cancer cells are not well understood and further studies are needed.

MUC2 mucin is also expressed in 29% of ICC with hepatolithiasis and this frequency is a little higher compared to the usual ICC [22]. Recently, a specific group of intraductal papillary tumors (including pre-neoplastic lesions) and associated mucinous carcinoma, which are categorized as counterparts of pancreatic intraductal papillary-mucinous neoplasm [27], is occasionally associated with hepatolithiasis, and the characteristic expression of MUC2 mucin has been noted in these tumors [5] (Fig. 4). MUC2 itself has the property of a tumor suppressor gene. In contrast to MUC1 expression indicating a poorer prognosis, MUC2 expression is higher in the mucinous type of tumors with a favorable outcome in biliary and pancreatic tumors [3]. The MUC2 expression in the intrahepatic biliary system, including intestinal metaplasia, intraductal papillary tumors and mucinous carcinoma, is dependent on the CDX2 homeobox gene, which induces intestinal differentiation [5] (Fig. 4). Accumulating data suggest that CDX2 functions as a tumor suppressor and CDX2-dependent regulation of cell proliferation may be an important factor in determining the prognosis of the patient. CDX2 is a key molecule in intraductal papillary

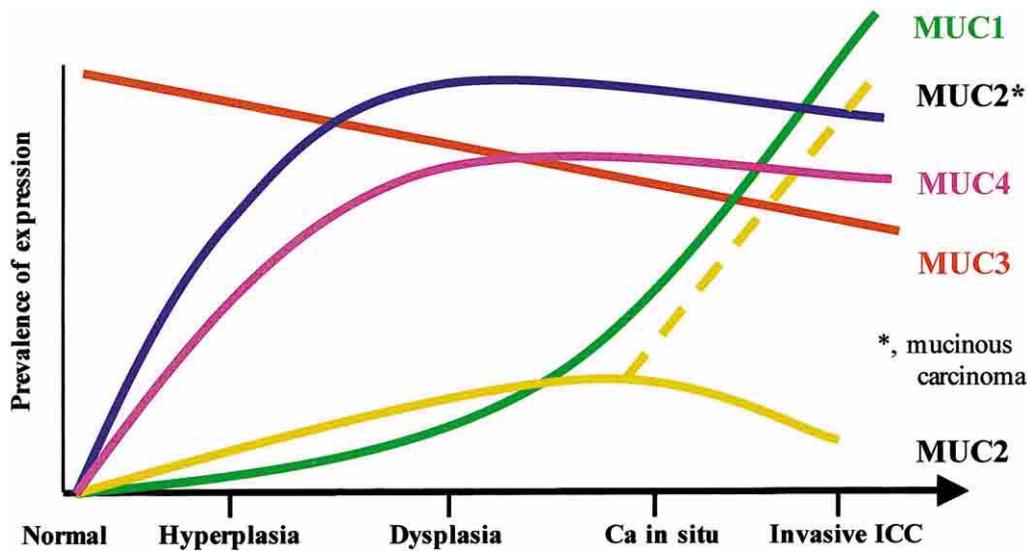


Fig. 6. The expression profile of MUC mucin during stepwise development and progression through biliary epithelial dysplasia, non-invasive and invasive intrahepatic cholangiocarcinoma in hepatolithiasis. ICC, intrahepatic cholangiocarcinoma.

tumors associated with hepatolithiasis, gastric carcinoma and Barrett's adenocarcinoma, in which transient or constant intestinal metaplasia is important in the carcinogenesis. Therefore, it is conceivable that a common factor which promotes or regulates CDX2 expression is closely related to the pathway of stepwise progression of these carcinomas.

VII. Summary

The expression profiles of MUC mucin may be closely related to the physiological functions of the intrahepatic biliary tree, and their alterations in the biliary tract with hepatolithiasis, pre-neoplastic lesions, and ICC may be associated with lithogenesis and carcinogenesis of the intrahepatic biliary system. The expression profiles of MUC mucins of cholangiocarcinoma thus reflect the aggressiveness of carcinoma cells and also the prognosis of patients.

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