Original Article

Serum mucin 3A as a potential biomarker for extrahepatic cholangiocarcinoma

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Abstract

Background/Aims: The aim of this study is to evaluate serum mucin 3A (MUC3A) as a candidate biomarker for extrahepatic cholangiocarcinoma (EHCC).

Patients and Methods: 35 Patients with EHCC, 30 patients with pancreatic cancer, 35 patients with gallbladder carcinoma and 78 patients with benign biliary disease were enrolled during January 2015 to January 2016. Serum MUC3A, carbohydrate antigen 19-9 (CA19-9) and carcinoembryonic antigen (CEA) were measured in these patients. Pathology reports of patients with EHCC were collected.

Results: (1) The serum levels of MUC3A (87.3 ± 10.8 ng/ml) in patients with EHCC were higher than in patients with pancreatic cancer (63.2 ± 7.7 ng/ml, $P < 0.001$), patients with gallbladder carcinoma (59.0 ± 10.3 ng/ml, $P < 0.001$) and patients with benign biliary disease (56.6 ± 13.1 ng/ml, $P < 0.001$). (2) ROC analysis showed that using MUC3A could clearly distinguish patients with EHCC from those without EHCC with a threshold of 73.2 ng/ml. (3) According to ROC analysis, the sensitivity, specificity, and accuracy of serum MUC3A for diagnosis of EHCC were 94.3%, 89.5% and 90.4%, respectively, which were all significantly higher than CA19-9 and CEA. (4) The serum levels of MUC3A at 1 month post-operatively in 35 patients with EHCC were decreased compared to pre-operative levels (51.8 ± 5.6 vs. 87.3 ± 10.8 ng/ml, $P < 0.01$). (5) Compared with 20 patients with low MUC3A levels (≤88.8 ng/ml), 15 patients with high MUC3A levels (>88.8 ng/ml) had higher percentage of lymph node metastasis (66.7% vs. 25%, $P = 0.014$), surrounding tissue infiltration (80% vs. 30%, $P = 0.003$), and UICC staging IIa-III (86.7% vs. 35%, $P = 0.002$).

Conclusion: The diagnostic efficiency for EHCC of MUC3A is obviously superior to CA19-9 and CEA, and a high level of serum MUC3A indicates a poor prognosis, therefore, MUC3A can be used as a potential diagnostic and prognostic biomarker for EHCC.

Keywords: Biomarker, diagnosis, extrahepatic cholangiocarcinoma, mucin 3A, prognosis

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INTRODUCTION

Cholangiocarcinoma is a highly lethal malignancy arising from the epithelial cells of the bile, accounting for approximately 3% of the world's annual gastrointestinal cancer and 15% of liver tumors, and the incidence rate has gradually increased

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in recent years.[1-3] Depending on the tumor site, it can be classified as intrahepatic cholangiocarcinoma or extrahepatic cholangiocarcinoma (EHCC). EHCC comprises of tumors arising from the hilar region to the bottom of the common bile duct, not including gallbladder cancer.[4]

EHCC is difficult to diagnose at an early stage due to its special anatomical region. Clinical diagnosis of EHCC is mainly based on the combined algorithm of "painless jaundice + bile duct dilatation + bile duct tumor". Given that operable EHCC are small in diameter and tend to be asymptomatic, the current noninvasive techniques, including computed tomography (CT) and magnetic resonance imaging (MRI), are not able to provide sufficient resolution to reliably detect these small lesions. Despite the use of advanced imaging methods and image-guided biopsy procedures in differentiating the EHCC from benign biliary diseases in recent years, diagnostic limitations still exist. Besides, these procedures have drawbacks since they are invasive and costly. Furthermore, they can only be performed in centers with experienced gastroenterologists.[5] Therefore, more than 2/3 of EHCC patients with definitive diagnoses are in late stage and have missed the opportunity for radical surgery.[6] The survival time of patients with unresectable EHCC was usually only 6-9 months.[2]

Less invasive and simpler procedure such as serum markers would be of substantial clinical benefit for diagnosis, monitoring, and predicting outcome for EHCC patients. However, at present, there is no effective tumor marker for EHCC.[7,8] The most commonly used ones are serum carbohydrate antigen 19-9 (CA19-9) and carcinoembryonic antigen (CEA), which only have sensitivities of 50% and 30%, respectively.[9] Therefore, searching for a highly specific and sensitive diagnostic biomarker for EHCC is urgently needed as this could lead to earlier diagnosis and improved outcome.[10]

Proteomic-based approaches are increasingly applied to cancer biomarker discovery as proteins can be qualitatively and quantitatively analyzed. The process of evaluating a biomarker starts from discovery through validation, clinical trial and clinical application. In previous research, we examined the differential proteomics of bile samples collected from EHCC patients and found a significant difference in the level of mucin 3A (MUC3A) expression between EHCC patients and patients with Oddi sphincter dysfunction (SOD).[11] MUC3A has high expression in tumor tissue in patients with EHCC with immunohistochemistry.[12]

Clinically, EHCC needs to be differentiated from other malignant strictures of the bile duct and benign biliary disease. Hepatocellular carcinoma and intrahepatic cholangiocarcinoma are mainly manifested as the space occupying lesions in liver, which are easily differentiated from EHCC by CT and other imaging examinations. Other major causes of malignant bile duct obstruction are pancreatic and gallbladder cancers.[13] To extend our previous findings and apply them to clinical practice, the serum MUC3A was measured in patients with malignant biliary tumors (EHCC, pancreatic cancer, gallbladder carcinoma) and patients with benign biliary disease (common bile duct stones, benign biliary stricture). The diagnostic efficacy of serum MUC3A was evaluated in patients with EHCC and compared with CEA and CA19-9. The correlation between serum MUC3A level with EHCC clinical pathology was also analyzed.

PATIENTS AND METHODS

Study population

35 patients with EHCC admitted to affiliate Hangzhou First People's Hospital, Zhejiang University School of Medicine from January 2015 to January 2016 were enrolled. All patients underwent radical surgery, and postoperative pathology confirmed the diagnosis of cholangiocarcinoma.

Thirty patients with pancreatic cancer and 35 patients with gallbladder carcinoma were enrolled at the same period. All patients underwent ERCP+ cytobrush or biopsy with Spyglass choledochoscopy or endoscopic ultrasonography (EUS)-fine needle aspiration (FNA). The diagnosis of these patients was made on the basis of clinical symptoms, liver function test, spiral computed tomography (CT) or magnetic resonance imaging (MRI), EUS finding, histological or cytological results and follow-up data. Only those patients in whom a confident clinical diagnosis could be established were included in the study.

In addition, 45 patients with common bile duct stones, 33 patients with benign biliary stricture were also enrolled in the study in order to assess the levels of serum tumor markers in the benign biliary disease; these patients were followed-up for at least 1 year and malignancy was eliminated. In the meantime, healthy volunteers (HVs) who were age, and gender matched, were recruited through the recruitment advertisement as control group.

Study design

Serum MUC3A, CA19-9 and CEA were measured in all eligible participants. The clinical pathology data including age, gender, histopathological features, tumor location, lymph node invasion, surrounding tissue infiltration, and tumor stage in patients with EHCC, were collected from the electronic medical record system. All patients were followed up until December 2018.

The study was approved by Ethics Committee of Hangzhou First People's Hospital and conducted in accordance with the Declaration of Helsinki (2014-(12)-0601), and written informed consent was obtained from each participant.

Measurement of serum MUC3A, CA19-9 and CEA

Morning fasting venous blood (10 ml) was drawn using a coagulation tube for biochemical analysis. After centrifugation (3500 g) of the coagulated blood, 0.5 ml supernatant was collected. Serum MUC3A was quantified using enzyme-linked immunosorbent assay kits (ELISA; eBioscience, San Diego, USA) according to the manufacturer's instructions. The test has a sensitivity of 0.39 ng/ml and the detection wave length was 450 nm. The level of serum CA19-9 was measured using chemiluminescence particle immunoassays (ARCHITECT CA19-9 Reagent kit, Abbott) based on the recommended cut-off of 47 U/ml. The level of serum CEA was measured using chemiluminescence particle immunoassays (ARCHITECT CEA Reagent kit, Abbott) based on the recommended cut-off of 5 ng/ml.

Statistical methods

Continuous variables with an apparently Gaussian distribution are presented with their mean and standard deviation as an index of dispersion, whereas the median and the interquartile range (IQR) are used to summarize variables with a skewed distribution. Categorical data were presented as count (percentage). Continuous variables were analyzed using Student's *t*-test or nonparametric tests whereas categorical variables were analyzed using the Chi-square test. Serum levels of MUC3A, CEA and CA19-9 were compared among groups. The optimal cutoff points for MUC3A, CEA and CA19-9 were selected based on the receiver operating characteristic (ROC) curve analysis. The sensitivity, specificity, positive predictive value, and negative predictive value were calculated using a 2 × 2 table of the collected data. The correlation between serum MUC3A levels and age, sex, tumor location, tumor differentiation, lymph node metastasis, surrounding tissue infiltration and tumor stage in EHCC patients undergoing surgery were analyzed using a Chi-square test. Overall survival (OS) in patients with EHCC was compared using the Kaplan-Meier method and a log-rank test. All analyses were conducted using the SPSS version 16.0 statistical package. *P* values <0.05 were considered as significant.

RESULTS

Characteristics of the study population

35 patients with EHCC underwent radical surgery, of which, 16 were males and 19 females, aged 45-70 years, with a mean age of 52.6 ± 8.9 years. Among 30 patients with pancreatic cancer, 15 were males and 15 females, aged 46-71 years, with a mean age of 60.6 ± 10.9 years. Among 35 patients with gallbladder carcinoma, 18 were males and 17 females, aged 50-73 years, with a mean age of 58.6 ± 9.3 years. Among 78 patients with benign biliary disease, 46 were males and 32 females, aged 35-75 years, with a mean age of 50.6 ± 7.5 years. There was no significant difference in gender, age, body mass index (BMI), liver function test between these groups ($P > 0.05$) [Table 1].

Levels of serum MUC3A, CA19-9 and CEA in patients with EHCC and patients without EHCC

The serum levels of MUC3A (87.3 ± 10.8 ng/ml) in patients with EHCC were higher than in patients with pancreatic cancer (63.2 ± 7.7 ng/ml, $P < 0.001$), patients with gallbladder carcinoma (59.0 ± 10.3 ng/ml, $P < 0.001$), patients with benign biliary disease (56.6 ± 13.1 ng/ml, $P < 0.001$) and healthy volunteers (25.1 ± 9.2 ng/ml, $P < 0.001$), the differences were statistically significant ($P < 0.001$). The serum levels of CA19-9 (95.3 ± 52.3) in patients with EHCC were higher than in patients with benign biliary disease (62.2 ± 44.4, $P = 0.004$), and lower than in patients with pancreatic cancer (155.7 ± 94.8, $P < 0.001$), the differences were statistically significant, but similar to patients with gallbladder carcinoma (75.6 ± 34.8, $P = 0.144$), the differences were not statistically significant ($P < 0.01$). The serum levels of CEA (4.8 ± 0.9) in patients with EHCC were higher than in patients with benign biliary disease (3.7 ± 0.8, $P < 0.001$), the differences were statistically significant, and similar to patients with gallbladder carcinoma (5.2 ± 1.5, $P = 0.081$) and patients with pancreatic cancer (4.8 ± 1.3, $P = 0.973$), the differences were not statistically significant [Figure 1].

Predictive value of serum MUC3A, CA19-9 and CEA in EHCC

The predictive value of MUC3A, CA19-9 and CEA was determined by generating a ROC curve [Figure 2]. The optimal cutoff for MUC3A, CA19-9 and CEA for discriminating between EHCC patients and patients without EHCC were 73.2 ng/ml, 78.7 U/L, 4.59 U/L, with an area under the curve (AUC) of 0.981 ($P = 0.000$), 0.627 ($P = 0.020$) and 0.647 ($P = 0.007$), respectively. The AUC of MUC3A was significantly higher than that of CA19-9 and CEA respectively.

The sensitivity, specificity, and accuracy of serum MUC3A for diagnosis of EHCC were 94.3%, 89.5% and 90.4%, respectively, which were all significantly higher than CA19-9 (62.9%, 63.6%, 63.5%) and CEA (68.6%, 62.2%, 63.5%) [Table 2].

Table 1: Characteristics of the study population

	Patients with EHCC (*n*=35)	Patients with pancreatic cancer (*n*=30)	Patients with gallbladder carcinoma (*n*=35)	Patients with benign biliary disease (*n*=78)	*P*
Gender (men: women)	16:19	15:15	18:17	46:32	0.46
Age (years)	52.6±8.9	60.6±10.9	58.6±9.3	50.6±7.5	0.36
BMI (kg/m2)	25.2±3.3	23.1±3.5	24.8±3.8	25.0±4.2	0.23
ALT (U/L)	62.1±7.5	69.9±6.5	52.1±5.5	59.2±5.8	0.78
AST (U/L)	70.7±7.6	72.9±6.0	69.7±7.1	62.5±5.2	0.46
r-GT (U/L)	69.9±9.7	71.4±6.7	59.2±6.8	54.4±5.2	0.33
AKP (U/L)	89.4±6.6	81.2±6.8	69.9±5.6	71.2±8.2	0.12
Total bilirubin (μmol/L)	71.0±15.0	86.3±13.9	65.3±10.3	66.4±11.9	0.36
Direct bilirubin (μmol/L)	57.5±9.4	68.6±15.7	50.1±8.4	48.6±9.7	0.59
Albumin (g/L)	38.8±2.6	39.9±3.4	40.8±2.8	41.4±3.3	0.39

Levels of serum MUC3A, CA19-9 and CEA in patients with EHCC at 1 month after surgery

The serum levels of MUC3A, CA19-9 and CEA at 1 month post-operation in 35 patients who underwent surgery were all decreased compared to pre-operative levels (51.8 ± 5.6 vs. 87.3 ± 10.8 ng/ml, 39.6 ± 12.7 vs. 95.3 ± 52.3 U/L and 3.4 ± 0.5 vs. 4.8 ± 0.9 U/L, $P < 0.01$), with the differences being statistically significant [Figure 3].

Correlation between serum MUC3A levels and clinical pathology of cholangiocarcinoma

Of the 35 patients who underwent surgical resection, 13 (37.1%) had tumors in the hilar, and 22 (62.9%) in the distal bile duct. 19 patients were R0 resection, number of patients missing patients were R1 resection. All patients were adenocarcinoma, including well differentiated and moderately differentiated tumors in 18 (51.4%) patients and poorly differentiated and undifferentiated in 17 patients (48.6%). Fifteen (42.9%) patients had lymph node metastasis, 18 (51.4%) patients had invasion of the surrounding tissues, UICC staging I-II was identified in 15 (42.9%) patients, and UICC staging II b-III was identified in 20 (57.1%) patients.

According to median level of serum MUC3A levels, 35 patients were divided in to two groups: 15 patients with high MUC3A (>88.8 ng/ml) and 20 patients with low MUC3A levels (≤88.8 ng/ml). Compared with patients with low MUC3A levels, patients in the high MUC3A levels group had higher percentage of lymph node metastasis (66.7% vs. 25%, $P = 0.014$), surrounding tissue infiltration (80% vs. 30%, $P = 0.003$, and UICC staging II b-III (86.7% vs. 35%, $P = 0.002$) [Table 3]. The mean OS was significantly longer in the low MUC3A group than that in the high MUC3A group (18.4 ± 1.4, 95% CI: 15.6-21.2 months vs. 13.0 ± 0.9, 95% CI: 11.2-14.8 months, $P = 0.000$) [Figure 4].

DISCUSSION

The results of this study showed that serum MUC3A levels in patients with EHCC were significantly higher than in pancreatic cancer, patients with gallbladder carcinoma and patients with benign biliary disease and decreased 1 month postoperatively in those undergoing surgery. Based on the threshold defined from ROC analysis, MUC3A can clearly distinguish patients with EHCC from those without EHCC, including other common malignant biliary obstruction and biliary benign diseases. Patients with high MUC3A levels had high percentage of lymph node metastasis, surrounding tissue infiltration and UICC stages II b-III.

Mucin is widely distributed in the body, mainly in the gastrointestinal tract, respiratory tract, urogenital surface cells and mucus secretions. Currently, 17 proteins have been found and divided into secretory or membrane-bound classes. In normal tissue, epithelium mucin plays a role in cell protection and lubrication. In tumor cells, it contributes to adhesion to normal cells and facilitates the escape of tumor cells from immune recognition through the steric hindrance phenomenon. Studies have shown that expression of mucin genes is tissue- and cell-specific, and abnormal expression is important in tumorigenesis and cell differentiation, which can promote the malignant progression of tumors.[14,15]

The main mucin genes expressed in cholangiocytes were MUC3, MUC6 and MUC5B. There is no previous report about MUC protein used as a potential diagnostic and/or prognostic marker for EHCC. MUC3 is a type of mucin located in the mucin cluster of the chromosome 7q22. It is classified as a membrane-associated mucin and can be divided into 2 subtypes: MUC3A and MUC3B.[16] Studies have shown that hepatobiliary epithelial cells mainly express MUC1 before birth, and switch to the expression of MUC3 after birth.[17] In normal gallbladder and extrahepatic bile duct, the expressed mucin gene is mainly MUC3.[18] Vilkin *et al*.[19] reported that high levels of MUC3, MUC5AC and MUC5B are expressed in bile aspirated during ERC examinations.

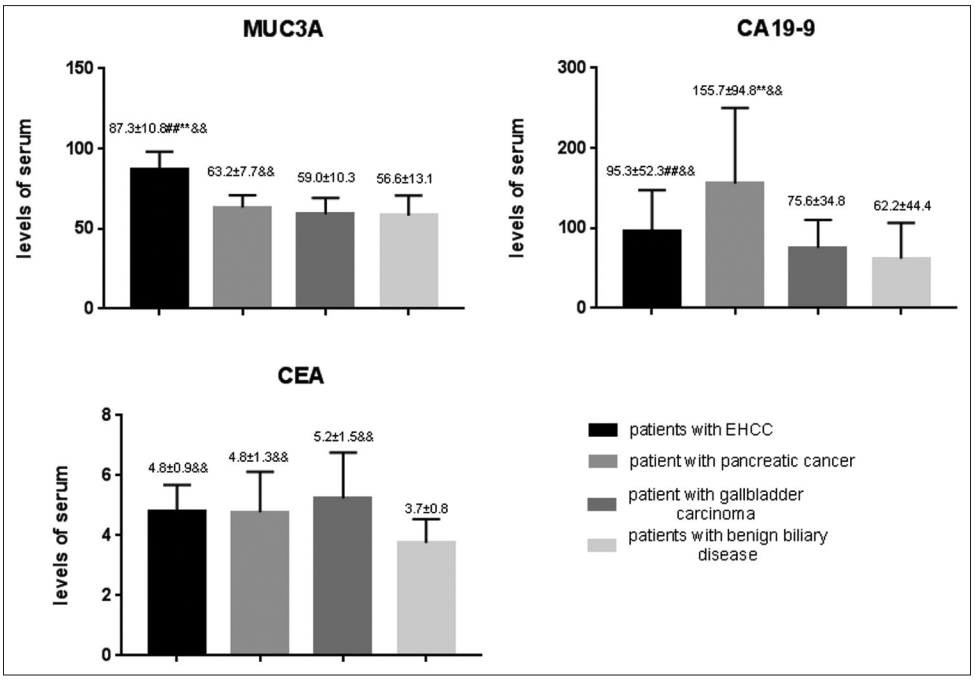

Figure 1: Levels of serum MUC3A, CA19-9 and CEA in patients with EHCC and patients without EHCC. EHCC: extrahepatic cholangiocarcinoma (n = 35) Compared with patients with pancreatic cancer (n = 30), $^{\#\#}P < 0.01$; Compared with patients with gallbladder carcinoma (n = 35), $^{**}P < 0.01$; Compared with patients with benign biliary disease (n = 78), $^{\&\&}P < 0.01$

MUC3A is a member of the Mucin family, a group of glycoproteins with oligosaccharide chains with high molecular weight (120-400 kd). Yoo *et al*.[20] reported that the mucin genes with the highest expression levels in gallbladder tissue in cholesterol-associated diseases were MUC3 and MUC5B, and cholesterol stones and gallbladder infections were associated with increased MUC3 and MUC5B expression. Sierzega *et al*.[21] investigated mucin expression, as examined by immunohistochemistry, in surgical specimens resected from 101 patients with pancreatic ductal cell adenocarcinoma, 33 with chronic pancreatitis, and 40 normal pancreatic tissue specimens. The three-MUC diagnostic model (MUC3, MUC5AC, and MUC6) allowed excellent discrimination of pancreatic cancer from non-malignant tissues. High expression of MUC3A in tumor tissue specimen was also reported in several types of cancer such as renal cell carcinoma,[22] breast,[23] and colorectal cancer.[24]

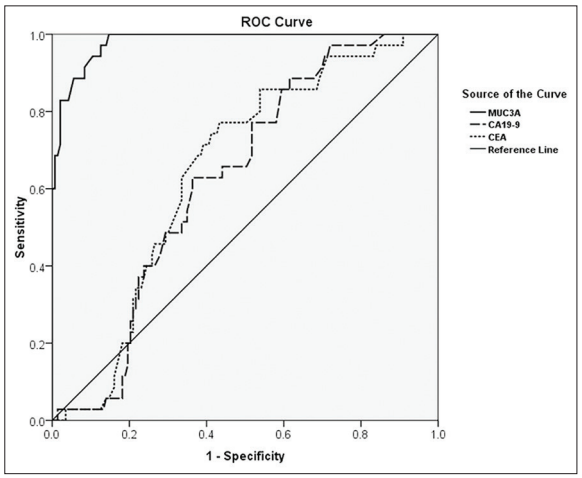

Figure 2: The diagnostic value of MUC3A, CA19-9, CEA in extrahepatic cholangiocarcinoma (EHCC) using receiver operating characteristic (ROC) curves. The optimal cutoff for MUC3A, CA19-9 and CEA for discriminating between EHCC patients and patients without EHCC were 73.2 ng/ml, 78.7 U/L, 4.59 U/L, with an area under the curve (AUC) of 0.981 (P = 0.000), 0.627 (P = 0.020) and 0.647 (P = 0.007), respectively

However, the practical diagnostic value of mucin expression in tumor tissue specimen examined by immunohistochemistry is limited clinically.

Our study shows that serum MUC3A was significantly higher in EHCC patients than in those patients without EHCC, such as pancreatic cancer, gallbladder carcinoma and benign biliary disease (including common bile duct stones, benign biliary

Table 2: Predictive value of serum MUC3A, CA19-9 and CEA in EHCC

	Sensitivity	Specificity	PPV	NPV	FPR	Accuracy
MUC3A	94.3%	89.5%	68.8%	98.5%	10.5%	90.4%
CA19-9	62.9%	63.6%	29.7%	87.5%	36.4%	63.5%
CEA	68.6%	62.2%	30.8%	89.0%	37.8%	63.5%

EHCC: Extrahepatic cholangiocarcinoma. PPV: Positive predictive value, NPV: Negative predictive value, FPR: False positive rate. MUC3A: Cutoff : 73.2 ng/ml, AUC: 0.981 (95% CI: 0.965-0.997). CA19-9: Cutoff : 78.7 U/L, AUC: 0.627 (95% CI: 0.539-0.715). CEA: Cutoff : 4.59 U/L, AUC: 0.647 (95% CI: 0.559-0.736)

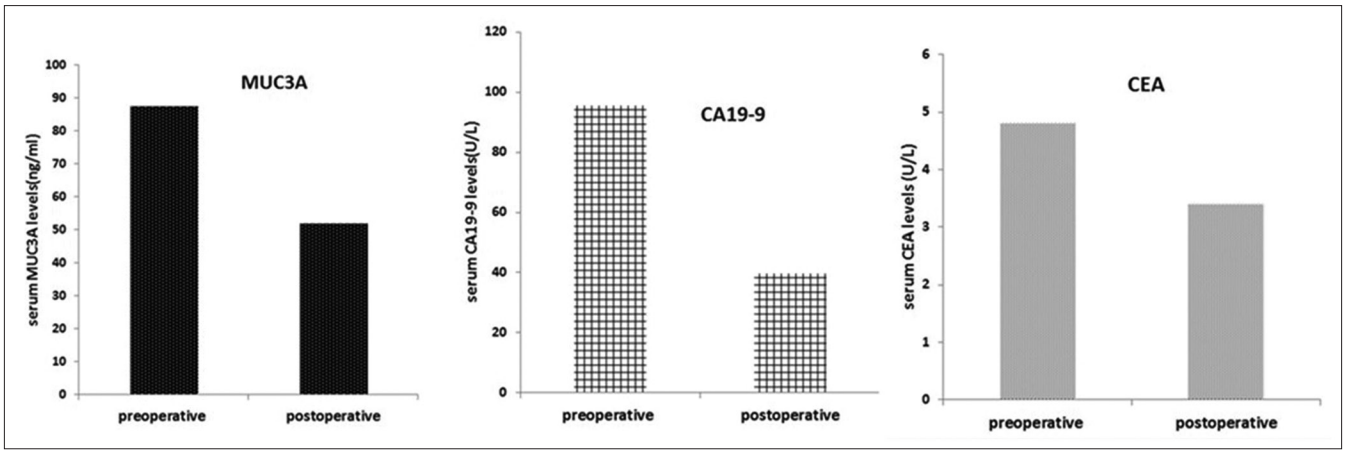

Figure 3: The serum levels of MUC3A, CEA and CA19-9 in EHCC patients before and postoperative 1 month. Compared to preoperative level, the serum levels of MUC3A, CA19-9 and CEA were decreased significantly postoperative 1 month (51.8 ± 5.6 vs. 87.3 ± 10.8 ng/ml, 39.6 ± 12.7 vs. 95.3 ± 52.3 U/L and 3.4 ± 0.5 vs. 4.8 ± 0.9 U/L, $P < 0.01$)

stricture). These diseases often need differential diagnosis with EHCC in clinical practice. MUC3A could clearly distinguish EHCC patients from these diseases with the cut-off value of 73.2 ng/ml according to ROC analysis. Using pathology examination as the gold standard, the sensitivity, specificity and accuracy of MUC3A in the diagnosis of EHCC were 94.3%, 89.5% and 90.4%, respectively, which is superior to both CA19-9 and CEA. Moreover, compared to pre-operation levels, the level of serum MUC3A in patients who underwent radical resection surgery had a significant decline at 1 month post-operation. Therefore, our study shows that MUC3A could play a role in EHCC diagnosis and assessment for responses to treatment.

MUC3A is a membrane-associated mucin that recent evidence reveals has a role in pathogenesis and progression of cancers. High MUC3A expression is an adverse prognostic biomarker for overall survival (OS) and recurrence-free survival (RFS) in postoperative localized clear-cell renal cell carcinoma (ccRCC) patients.[22] Sotoudeh M *et al*.[25] reported that MSLN (Mesothelin), ANTXR1 (TEM8), and MUC3A are the probable targets of CAR T cell therapy in gastric adenocarcinoma. It has been reported that the expression of MUC3A is closely related to the depth of infiltration, lymph node metastasis and tumor stage in gastric cancer and colon cancer.[26,27] Shibahara *et al*.[28] reported intravenous infiltration (HR: 6.93, 95% CI: 1.93-24.96, $P = 0.003$), non-curative resection (HR: 10.19, 95% CI: 3.05-34.07, $P < 0.001$) and positive MUC3 expression (HR: 3.37, 95% CI: 1.13-10.03,

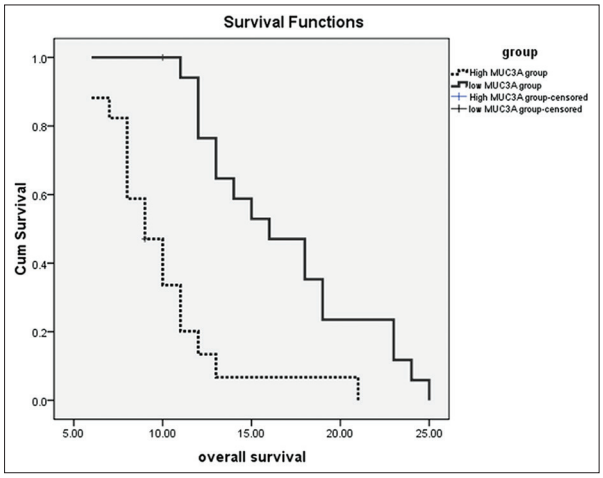

Figure 4: Comparison of mean overall survival (OS) between the low MUC3A group and the high MUC3A group. The mean OS was significantly longer in the low MUC3A group than that in the high MUC3A group (18.4 ± 1.4, 95% CI: 15.6-21.2 months vs. 13.0 ± 0.9, 95% CI: 11.2-14.8 months, $P = 0.000$)

Table 3: Serum MUC3A level and clinical pathological features of extrahepatic cholangiocarcinoma

Clinical Pathology	Low MUC3A (*n*=20)	High MUC3A (*n*=15)	*P*
Sex			
Male (*n*=16)	9 (45%)	7 (46.7%)	0.922
Female (*n*=19)	11 (55%)	8 (53.3%)	
Age			
<60 (*n*=24)	13 (65.0%)	11 (73.3%)	0.598
≥60 (*n*=11)	7 (35.0%)	4 (26.7%)	
Tumor site			
Hilar (*n*=13)	8 (40%)	5 (33.3%)	0.686
Distal (*n*=22)	12 (60%)	10 (66.7%)	
Differentiation			
Well-moderately (*n*=18)	10 (50%)	8 (53.3%)	0.845
Poor- undifferentiated (*n*=17)	10 (50%)	7 (46.7%)	
Lymph node metastasis			
No (*n*=20)	15 (75%)	5 (33.3%)	0.014
Yes (*n*=15)	5 (25%)	10 (66.7%)	
Surrounding tissue infiltration			
No (*n*=17)	14 (70%)	3 (20%)	0.003
Yes (*n*=18)	6 (30%)	12 (80%)	
UICC staging			
I a-II a (*n*=15)	13 (65%)	2 (13.3%)	0.002
II b-III (*n*=20)	7 (35%)	13 (86.7%)	

$P = 0.03$) were independent risk factors for poor prognosis of appendicular adenocarcinoma. Our study also showed that patients with high MUC3A level had a higher likelihood of lymph node metastasis, peripheral infiltration and UICC staging IIa-III. These results indicated that higher MUC3A is closely correlated to more late stage tumors and poor prognosis in patient with EHCC.

However, the exact mechanism of MUC3A expression is yet to be clarified. Kitamoto *et al*.[27] reported that DNA hypomethylation in the 5'-flanking region of the MUC3A gene plays an important role in MUC3A expression in carcinomas of various organs.

However, this study included a relatively small sample size and employed a non-multicenter design, and some of the patients with EHCC in this study were locally advanced cholangiocarcinoma. Therefore, a large-scale multicenter study to investigate the diagnostic potential of MUC3A for clinical use is still necessary.

In summary, serum MUC3A is a potential diagnostic biomarker for EHCC which could further improve diagnostic accuracy. Moreover, a high level of serum MUC3A indicates a poor prognosis. The functions of MUC3A need to be further investigated in order to better understand the tumor biology and use it as targets for future therapeutic agents. Detection of serum MUC3 can improve the diagnostic rate of EHCC and evaluate prognosis in a more accurate manner.

CONCLUSION

MUC3A can be used as a potential diagnostic and prognostic biomarker for extrahepatic cholangiocarcinoma.

Declaration of patient consent

The authors certify that they have obtained all appropriate patient consent forms. In the form the patient(s) has/have given his/her/their consent for his/her/their images and other clinical information to be reported in the journal. The patients understand that their names and initials will not be published and due efforts will be made to conceal their identity, but anonymity cannot be guaranteed.

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Conflicts of interest

There are no conflicts of interest.

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