World Journal of Gastroenterology

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DOI: 10.3748/wjg.v22.i18.4459

World J Gastroenterol 2016 May 14; 22(18): 4459-4465
ISSN 1007-9327 (print) ISSN 2219-2840 (online)

Combined hepatocellular cholangiocarcinoma: Controversies to be addressed

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Conflict-of-interest statement: There are no potential conflicts of interest relevant to this article.

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Received: February 16, 2016
Peer-review started: February 17, 2016
First decision: March 7, 2016
Revised: March 16, 2016
Accepted: April 7, 2016
Article in press: April 7, 2016
Published online: May 14, 2016

Abstract

Combined hepatocellular cholangiocarcinoma (CHC) accounts for 0.4%-14.2% of primary liver cancer cases and possesses pathological features of both hepatocellular carcinoma and cholangiocarcinoma. Since this disease was first described and classified in 1949, the classification of CHC has continuously evolved. The latest definition and classification of CHC by the World Health Organization is based on the speculation that CHC arises from hepatic progenitor cells. However, there is no evidence demonstrating the common origin of different components of CHC. Furthermore, the definition of CHC subtypes is still ambiguous and the identification of CHC subtype when a single tumor contains many components has remained unresolved. In addition, there is no summary on the newly recognized histopathology features or the contribution of CHC components to prognosis and outcome of this disease. Here we provide a review of the current literature to address these questions.

Key words: Progenitor cell origin; Pathology classification; Combined hepatocellular cholangiocarcinoma

Core tip: This review article focuses on the current views about the histopathology and clinical characteristics of combined hepatocellular cholangiocarcinoma (CHC). Whether the different components of CHC share a common cell origin is still ambiguous. Furthermore, the definition of CHC subtype is still ambiguous and

the identification of CHC subtype when a single tumor contains many components has remained unresolved. The features between hepatocellular carcinoma (HCC) and cholangiocarcinoma (CC) make CHC has better prognosis than CC but poorer than HCC.

Wang AQ, Zhen YC, Du J, Zhu CP, Huang HC, Wang SS, Wu LC, Wan XS, Zhang HH, Miao RY, Sang XT, Zhao HT. Combined hepatocellular cholangiocarcinoma: Controversies to be addressed. *World J Gastroenterol* 2016; 22(18): 4459-4465 Available from: URL: http://www.wjgnet.com/1007-9327/full/v22/i18/4459.htm DOI: http://dx.doi.org/10.3748/wjg.v22.i18.4459

INTRODUCTION

Combined hepatocellular cholangiocarcinoma (CHC) is a rare form of primary liver cancer with pathological features of both hepatocellular carcinoma (HCC) and cholangiocarcinoma (CC). CHC accounts for 0.4%-14.2% of primary liver cancer cases in different regions[1-4]. It was first described and classified by Allen and Lisa in 1949[5], and Goodman updated the pathology classification in 1985[3]. Currently, according to the World Health Organization (WHO), CHC is classified into the classical subtype and subtype with stem-cell features[6].

Despite the extensive study on CHC over the past 60 years, many questions remain unresolved. Although many studies demonstrated that CHC originated from progenitor cells[7-9], whether all subtypes of CHC with stem-cell features share a common origin is unknown. Furthermore, how to classify the newly recognized histopathology features and how many effects on survival for CHC of the diversity of components and the properties of each component remain unclear. In addition, how to distinguish CHC form CK19(+)HCC and how to identify the subtype when a single tumor contains many components are still ambiguous. Finally, the clinical features of CHC including the risk factors, the role of liver transplantation and diagnosis are still controversial.

Therefore, here we review the current literature on CHC to address these issues.

EVOLUTION OF CHC CLASSIFICATION

Allen and Lisa first described CHC in 1949[5]. The authors classified this type of tumor into three subtypes including (1) separate masses composed of either HCC or CC; (2) contiguous but independent masses of HCC and CC; and (3) an intermingling of hepatocellular and glandular elements. In 1985, Goodman[3] updated the classification as (1) collision tumors, a coincidental occurrence of both HCC and CC in the same patient; (2) transitional tumors including areas of intermediate differentiation and an identifiable transition between HCC and CC; and (3) fibrolamellar tumors, which resemble the fibrolamellar variant of HCC but also contain mucin-producing pseudoglands. In 1989, The Liver Cancer Study Group of Japan[10] formulated its own classification of CHCs into three types: double cancer, combined type, and mixed type. In 1994[11], CHC was universally defined by the WHO as a tumor with intimate and unequivocal mixtures of both HCC and CC cells. However, these tumors should be distinguished from cases with separate HCC and CC arising in the same liver and in which HCC and CC are present at adjacent locations.

With the development of medicine, an increasing number of studies have demonstrated that hepatic progenitor cells play an important role in the development of CHC. Therefore, in 2010, the WHO updated the classification of CHC into the classical type and subtypes with stem cell features[6]. The subtypes were further subdivided into the typical subtype, intermediate cell subtype, and cholangiolocellular subtype. The classical type of CHC contains unequivocal components of HCC and CC, whereas subtypes with stem cell features possess special histopathology features (Table 1). This most recent classification system has been widely adopted.

Origin

Since the identification of CHC occurred, the debate on the origin of CHC has been ongoing. It is theoretically reasonable for CHC to originate from hepatocytes, cholangiocytes or hepatic progenitor cells. Many published studies have aimed to validate the origin of CHC at genetic and protein levels.

Genetic level

Multiple studies have been conducted to verify whether the HCC and CC components of CHC share the same origin and to identify the origin of the components. In 1996, Imai *et al*[12] investigated alterations of p53, K-ras and Rb-1 genes in seven CHC patients. The authors found that both components of CHC have the same genetic characteristics and speculated that they thus share the same origin. In 2000, Fujii *et al*[13] studied the allelic status of chromosome arms 1p, 1q, 3p, 4q, 5q, 6q, 8p, 9p, 10q, 11q, 13q, 16q, 17p, 17q, 18q, and 22q in HCC and CC foci of CHC. According to genetic patterns, they classified CHC origin into three possibilities: (1) a collision tumor in which two independent neoplastic clones develop at close proximity; (2) a single clonal tumor with divergent potential; and (3) a single clonal process in which genetic heterogeneity occur in the process of clonal evolution. This was the first unique classification of the origin of CHC at the genetic level. The authors considered that different types of CHCs had the same or different origins. Cazals-Hatem[14] screened for loss of heterozygosity (LOH) and for p53 and beta-catenin

Table 1 Evolution of classification of combined hepatocellular cholangiocarcinoma

Ref.	Classification
Allen *et al*[5], 1949	Separate masses
	Contiguous but independent masses
	Intimate intermingling of hepatocellular and glandular element
Goodman *et al*[3], 1985	Collision tumors, a coincidental occurrence of both HCC and CC in same patient
	Transitional tumors including areas of intermediate differentiation
	Fibrolamellar tumors, having features of fibrolamellar HCC and CC
Liver Cancer Study Group of Japan[10] 1989	Double cancer
	Combined type
	Mixed type
WHO[6] 2010	CHC-classical: typical HCC and typical CC
	CHC-SC
	CHC-SC-typical: nests of mature looking hepatocytes with peripheral clusters of small cells that have a high nucleus:cytoplasm ratio and hyperchromatic nuclei
	CHC-SC-int: tumor cells show features intermediate between hepatocytes and cholangiocytes. These tumor cells show strands, solid nests and/or trabeculae of small, uniform cells with scant cytoplasm and hyperchromatic nuclei
	CHC-SC-CLC: admixtures of small monotonous glands, antler-like anastomosing patterns. Each tumor cell is cuboidal, smaller in size than normal hepatocytes, with a high nucleus: cytoplasm ratio, and distinct nucleoli

CHC: Combined hepatocellular-cholangiocarcinoma; CHC-SC-typical: Combined hepatocellular-cholangiocarcinoma, stem cell features, typical subtype; CHC-SC-int: Combined hepatocellular-cholangiocarcinoma, stem cell features, intermediate cell subtype; CHC-SC-CLC: Combined hepatocellular-cholangiocarcinoma, stem cell features, cholangiolocellular subtype; HCC: Hepatocellular carcinoma; CC: Cholangiocarcinoma.

mutations in 9 CC, 15 CHC and 3 collision tumors compared with 137 HCC cases. Recurrent specific LOH was identified at 3p and 14q with significant differences among CHC, CC and HCC. The authors concluded that CHC is genetically more similar to CC compared with HCC. CHC shared frequent +1q (71%), +8q (57%), and 8p (57%) with HCC and higher numbers of imbalance with CC. The similar chromosomal changes with HCC and CC suggested that both share characteristics of CHC. These data also supported the same origin of different components of CHC. Although the genetic studies of CHC have provided some preliminary findings, whether HCC and CC components within CHC share the same origin should be further explored.

Protein level

Currently, the widely accepted origin of CHC is hepatic progenitor cells. However, it really takes a long time to get the conclusion. Many studies have used immunohistochemistry analyses to evaluate protein expression in CHC. A study by Okada *et al*[15] in 1987 reported the same levels of ABH, Lewis, and sialyl Lea antigens in CHC and HCC, indicating that CHC might have a hepatocellular origin. Imai *et al*[12] studied the expression patterns of carcinoembryonic antigen and keratin in seven CHC patients and demonstrated the same phenotypic characteristics in both components of CHC, suggesting a common origin of the hepatocellular and cholangiocarcinoma components. Zhang *et al*[8] performed immunohistochemical analysis of twelve CHC cases for hepatocytic (hepPar1, alpha-fetoprotein), cholangiocytic cytokeratin (CK7, CK19), hepatic progenitor cell (OV-6), hematopoietic stem cell (c-kit, CD34), as well as CD45 and chromogranin-A markers. The results suggested that CHC has the same hepatic progenitor cell origin. The same conclusion was also obtained by Akiba *et al*[16] who performed immunohistochemical analyses of biliary markers (CK7, CK19, and EMA), hepatocyte paraffin (hepPar-1), hepatic progenitor cell markers[17] (CD56, c-kit, CD133, and EpCAM), and vimentin. Kim *et al*[18] studied the expression of Yes-associated protein 1 (YAP1), a potential oncogene that can promote stem cell proliferation, among three groups comprising 36 HCCs with stem characteristics, 64 HCCs without stemness and 58 CHCs. Higher expression of YAP1 was observed in CHCs and HCCs with stemness than in HCCs without stemness. From the results of the expression of hepatocellular, cholangiocellular and progenitor cell markers, we speculate that the origin of CHC is hepatic progenitor cell.

UNSOLVED HISTOPATHOLOGY PROBLEMS

Although the classification of CHC has been widely accepted, there are still some special histopathology features that have not been classified. Nakajima[19] first described CHC with sarcomatous transformation in 1988, and several similar cases have since been reported[20-22]. In 2013, Terada[23] reported a CHC case with ductal plate malformation features that was characterized with CC cells forming markedly irregular tubules with intraluminal cell projections, bridge formations, and intraluminal tumor biliary cells. Jung *et al*[24] reported cholangiocellular carcinoma with satellite nodules showing intermediate differentiation.

Current studies have established that CHC contains various components. However, which component plays a more important role in the prognosis of CHC and whether the number of components could affect the recurrence and survival remain ambiguous. Akiba *et al*[16] investigated 54 CHC cases according to the WHO classification. The pathology type was defined by predominant histologic pattern (≥ 50%). CHC has wide histologic diversity, which poses a challenge for classification of CHC. Ikeda *et al*[25] divided 24 CHC cases into two groups: group A with less than 5% stem cell areas, and group B with more than 5% stem cell features. The expression level of delta-like 1 homolog was higher in group B than in group A. The postoperative overall survival rate was better in group A than in group B. These results suggest that the contribution of different components in CHC might be a significant factor that affects outcome. Sasaki *et al*[26] examined 62 CHC patients and found that the intermediate cell subtype was significantly associated with gender, tumor size, and histological grade of HCC and inversely correlated with the degree of stromal fibrosis. Significant associations were observed between cholangiocellular carcinoma and degree of fibrosis and inflammation, and an inverse association was observed with histological grade of HCC. The proportion of typical subtype was significantly inversely correlated with the degree of inflammation. Furthermore, the histological diversity score was also associated with vascular invasion. These data demonstrate a correlation between the proportion of each stem cell subtype and the histological diversity with clinicopathological factors, suggesting various properties of each component in the development of CHC. Although the WHO provided a clear definition and classification of CHC, the roles of newly recognized histopathology features, properties of each component, and the functions of diversity of components in recurrence and survival still need further exploration. In addition, the definition of CHC subtypes is still ambiguous, and how to distinguish CHC from CK19(+)HCC and how to identify the subtype of CHC when a single tumor contains many kinds of components require further research.

CLINICAL CHARACTERISTICS

Risk factors

Similar to other primary liver cancers, CHC could be induced by various factors impairing liver parenchyma. Compared with HCC, the relationship between HBV or HCV and CHC incidence is relatively weak[27]. A Japanese study revealed that the anti-HCV-positive rate was high in CHC as well as in HCC[28]. A hospital-based case-control study in China found that HBV infection and heavy alcohol intake might contribute to the development of CHC[29]. The percent of HCV-HBC infection was 37.3% and chronic liver disease was 38.3%, which suggested that viral infection and cirrhosis may be risk factors for ICC and CHC.

Clinical features

Whether the clinical features of CHC are similar to those of HCC or CCC is controversial. Compared with CC, the clinicopathological features of CHC include more advanced histological differentiation, increased prevalence in males, and lower levels of serum bilirubin and ALP. These features may make it possible to diagnose CHC in patients with suspected CC[30]. CHC is a hypovascular liver cancer with striking elevation of serum AFP and multiple regional lymphadenopathy[31]. These features are similar to both HCC and CC. The prevalence of hepatitis B positivity and cirrhosis in CHC was intermediate between HCC and CC[2,32]. CHCs were more likely to occur in males than CC[33]. Compared with HCC, CHC had lower incidence in Asian or Pacific patients with less distant spread. While clinical characteristics of CHC are similar to those of HCC, overall survival is more similar to that of ICC. Ng *et al*[34] investigated 21 cases of CHC and found that invasive characteristics with venous permeation, direct invasion into liver parenchyma and microsatellite formation were similar to those of HCC. The hypovascular features and regional lymphadenopathy features of CHC resemble those of CC. The elevations of serum AFP, venous permeation, and direct invasion and microsatellite formation in CHC are similar to those of HCC. The 75% positive rate of hepatitis B surface antigen and 61.5% AFP elevation rate detected in CHC were also closer to levels in HCC[34]. The features of both HCC and CC make CHC be a typical cancer having no special characters.

DIAGNOSIS

Imaging methods

The development of imaging methods has led to their important role in evaluating the properties of primary liver cancer. Dynamic computed tomography for CHC diagnosis has three enhancement patterns, including type Ⅰ hyper-enhancement in the early phase and hypo-enhancement due to washout of contrast medium in the late phase, resembling HCC; type Ⅱ peripheral enhancement in the early phase; and type Ⅲ late phases and an area of hyper-enhancement in the early phase and an area of slight delayed enhancement in the late phase. Type Ⅲ can be identified on the presentation of computed tomography[35]. Contrast-enhanced computed tomography could also predict the dominant component of CHC, which could optimize the treatment strategy for CHC patients[36]. Sensitivities and specificities for diagnosis of CHC range from 33% to 34% and 81% to 100%, respectively[37]. Therefore, tumor markers and risk factors should be used to improve the accuracy of diagnosis. Magnetic resonance imaging also contributes to the diagnosis of CHC[4].

Pathology

Before operation, it is difficult to accurately diagnose CHC. Although it is not entirely possible to confirm reliable diagnosis of CHC on cytologic preparations alone, cellblock or core biopsy for histochemical and immunohistochemical studies could be helpful for diagnosis[38,39]. Analysis of operative specimens is the gold standard for the diagnosis of CHC.

Treatment

Although the prognosis of CHC is poor, the development of hepatectomy, liver transplantation, and adjuvant chemotherapy and radiation therapy has improved the survival of patients.

Hepatectomy is the most important treatment for CHC, and can prolong the life of patients and even cure disease. Eguchi *et al*[40] reported successful hepatectomy for the intrahepatic recurrence of a CHC case and showed that resection of recurrent tumors could improve the poor prognosis of CHC.

Liver transplantation is another treatment for CHC. In CHC patients with liver transplantation, the 5-year survival was better than liver transplantation for intrahepatic CC but poorer than that of HCC meeting the Milan criteria[41-44]. The overall survival rates of CHC patients with liver transplantation at 1, 3 and 5 years were 79%, 66% and 16%, respectively[43]. Groeschl *et al*[45] showed that the survival benefit for localized CHC transplantation is similar to liver resection for CHC, but inferior to transplantation for HCC. Itoh *et al*[46] showed that better survival could be achieved after the living-donor liver transplantation for CHC meeting the Milan criteria or the Kyushu University criteria, similar to that of HCC.

Apart from operation, several adjuvant treatments are available, such as chemotherapy and radiation therapy. Disease-free survival of 42 mo after operation for a CHC case with lymph node metastasis was achieved through adjuvant chemotherapy and radiation therapy[47]. The case suggested an improved prognosis using multimodal therapy for CHC. Oral administration of UFT[48] effectively treated CHC with lymph node metastases. Systematic chemotherapy with fluorouracil, doxorubicin and cisplatin effectively suppressed the progression of CHC[49].

Together these findings suggest that for treatment of CHC, we should combine surgery with multimodal therapy to improve the survival of patients.

PROGNOSIS

The prognosis of primary liver cancer is poor. How about the prognosis of CHC? Tickoo investigated 27 CHCs with regard to their clinical features, which were different from those of pure HCC[50]. CHC showed disappointing prognosis, with overall 3- and 5-year survival rates of 30% and 18%, respectively. The 1- and 3-year survival rates in the CHC group (81.9% and 47%, respectively) were higher than those of the CC group and lower than those of the HCC group, which suggested a better prognosis of CHC compared with CC but worse compared with HCC[2]. In another center[4], the overall survival rates of CHC at 1, 3 and 5 years were 53%, 26% and 12%, respectively. Thus, the survival of CHC appears to be better than that of CC but worse than that of HCC[51]. Many factors could affect recurrence and survival after treatment. In a study of 30 CHC patients, Lin *et al*[4] found that major vascular branch invasion, regional organ invasion, nodal and distant metastases could affect prognosis. Park *et al*[52] concluded that the presence of portal vein thrombosis, distant metastasis and cholangiocellular component might be a poor prognosis indicator. Age, gender, transarterial chemoembolization and T stage were significant prognosis factors in CHC patients. The prognosis is poorer once the cholangiocellular components occur in liver cancer[52]. Sex, tumor-related factors (tumor number, major thrombus, and microvascular thrombus), serum gamma-glutamyl transpeptidase (GGT), and carbohydrate antigen 19-9 level seemed to be independent prognosis factors of long-term surgical survival for HBV-related CHC[53]. Disease-free survival could be affected by tumor size, major thrombus and serum GGT. The overall survival for CHC was associated with tumor size and lymph node metastasis. Poorer disease-free survival and similar overall survival rates were observed between CHCs and CCs[54]. The presence of cirrhosis, percent of serum hepatitis B or C marker positivity and the level of serum alpha-fetoprotein may be prognosis factors. Together these data indicate that CHC has a poor prognosis and many factors affecting the survival of HCC and CC could also be important prognostic factors for CHC.

Prospective

The obvious features between HCC and CC make CHC have better prognosis than CC but poorer than HCC. Owing to the limited therapy, the improvement of prognosis could be achieved by the development of basic science. With the development of sequencing techniques, it will soon be possible to demonstrate whether the different components of CHC share the same origin at the DNA level[55]. In addition, we could further explore whether CHC originates from hepatic progenitor cells at the RNA and protein levels. Finally, to address the unsolved pathology questions of CHC, many more studies are needed to clearly distinguish CHC from CK19(+)HCC and define the correct subtypes when a single mass has many different components. Owing to the variation of function in deciding the survival of different cancer components, perhaps a score system on the properties of each component and the numbers of components to predict the prognosis

could be established.

REFERENCES

1 **Kassahun WT**, Hauss J. Management of combined hepatocellular and cholangiocarcinoma. *Int J Clin Pract* 2008; **62**: 1271-1278 [PMID: 18284443 DOI: 10.1111/j.1742-1241.2007.01694.x]

2 **Koh KC**, Lee H, Choi MS, Lee JH, Paik SW, Yoo BC, Rhee JC, Cho JW, Park CK, Kim HJ. Clinicopathologic features and prognosis of combined hepatocellular cholangiocarcinoma. *Am J Surg* 2005; **189**: 120-125 [PMID: 15701504 DOI: 10.1016/j.amjsurg.2004.03.018]

3 **Goodman ZD**, Ishak KG, Langloss JM, Sesterhenn IA, Rabin L. Combined hepatocellular-cholangiocarcinoma. A histologic and immunohistochemical study. *Cancer* 1985; **55**: 124-135 [PMID: 2578078]

4 **Lin G**, Toh CH, Wu RC, Ko SF, Ng SH, Chou WC, Tseng JH. Combined hepatocellular cholangiocarcinoma: prognostic factors investigated by computed tomography/magnetic resonance imaging. *Int J Clin Pract* 2008; **62**: 1199-1205 [PMID: 17537192 DOI: 10.1111/j.1742-1241.2007.01291.x]

5 **Allen RA**, Lisa JR. Combined liver cell and bile duct carcinoma. *Am J Pathol* 1949; **25**: 647-655 [PMID: 18152860]

6 **Theise ND**, Park YN, Nakanuma Y. Combined hepatocellular-cholangiocarcinoma. In: Bosman FT, Carneiro F, Hruban RH, Theise ND, editors. WHO Classifica- tion of Tumours of the Digestive System, 4th ed. Lyon: IARC Press, 2010: 225-227

7 **Kim H**, Park C, Han KH, Choi J, Kim YB, Kim JK, Park YN. Primary liver carcinoma of intermediate (hepatocyte-cholangiocyte) phenotype. *J Hepatol* 2004; **40**: 298-304 [PMID: 14739102]

8 **Zhang F**, Chen XP, Zhang W, Dong HH, Xiang S, Zhang WG, Zhang BX. Combined hepatocellular cholangiocarcinoma originating from hepatic progenitor cells: immunohistochemical and double-fluorescence immunostaining evidence. *Histopathology* 2008; **52**: 224-232 [PMID: 18184271 DOI: 10.1111/j.1365-2559.2007.02929.x]

9 **Cai X**, Zhai J, Kaplan DE, Zhang Y, Zhou L, Chen X, Qian G, Zhao Q, Li Y, Gao L, Cong W, Zhu M, Yan Z, Shi L, Wu D, Wei L, Shen F, Wu M. Background progenitor activation is associated with recurrence after hepatectomy of combined hepatocellular-cholangiocarcinoma. *Hepatology* 2012; **56**: 1804-1816 [PMID: 22684921 DOI: 10.1002/hep.25874]

10 The general rules for the clinical and pathological study of primary liver cancer. Liver Cancer Study Group of Japan. *Jpn J Surg* 1989; **19**: 98-129 [PMID: 2659865]

11 **Ishak KG**, Anthony PP, Sobin LH. Histological Typing of the Liver. International Histological Classification of Tumours. WHO. 2nd ed. Hong Kong: Springer, 1994

12 **Imai Y**, Oda H, Arai M, Shimizu S, Nakatsuru Y, Inoue T, Ishikawa T. Mutational analysis of the p53 and K-ras genes and allelotype study of the Rb-1 gene for investigating the pathogenesis of combined hapatocellular-cholangiocellular carcinomas. *Jpn J Cancer Res* 1996; **87**: 1056-1062 [PMID: 8957064]

13 **Fujii H**, Zhu XG, Matsumoto T, Inagaki M, Tokusashi Y, Miyokawa N, Fukusato T, Uekusa T, Takagaki T, Kadowaki N, Shirai T. Genetic classification of combined hepatocellular-cholangiocarcinoma. *Hum Pathol* 2000; **31**: 1011-1017 [PMID: 11014564 DOI: 10.1053/hupa.2000.9782]

14 **Cazals-Hatem D**, Rebouissou S, Bioulac-Sage P, Bluteau O, Blanché H, Franco D, Monges G, Belghiti J, Sa Cunha A, Laurent-Puig P, Degott C, Zucman-Rossi J. Clinical and molecular analysis of combined hepatocellular-cholangiocarcinomas. *J Hepatol* 2004; **41**: 292-298 [PMID: 15288479 DOI: 10.1016/j.jhep.2004.04.030]

15 **Okada Y**, Jinno K, Moriwaki S, Morichika S, Torigoe S, Arima T, Nagashima H, Koprowski H. Expression of ABH and Lewis blood group antigens in combined hepatocellular-cholangiocarcinoma. Possible evidence for the hepatocellular origin of combined hepatocellular-cholangiocarcinoma. *Cancer* 1987; **60**: 345-352 [PMID: 3036328]

16 **Akiba J**, Nakashima O, Hattori S, Tanikawa K, Takenaka M, Nakayama M, Kondo R, Nomura Y, Koura K, Ueda K, Sanada S, Naito Y, Yamaguchi R, Yano H. Clinicopathologic analysis of combined hepatocellular-cholangiocarcinoma according to the latest WHO classification. *Am J Surg Pathol* 2013; **37**: 496-505 [PMID: 23388123 DOI: 10.1097/PAS.0b013e31827332b0]

17 **Zhu CP**, Wang AQ, Zhang HH, Wan XS, Yang XB, Chen SG, Zhao HT. Research progress and prospects of markers for liver cancer stem cells. *World J Gastroenterol* 2015; **21**: 12190-12196 [PMID: 26576103 DOI: 10.3748/wjg.v21.i42.12190]

18 **Kim GJ**, Kim H, Park YN. Increased expression of Yes-associated protein 1 in hepatocellular carcinoma with stemness and combined hepatocellular-cholangiocarcinoma. *PLoS One* 2013; **8**: e75449 [PMID: 24086533 DOI: 10.1371/journal.pone.0075449]

19 **Nakajima T**, Kubosawa H, Kondo Y, Konno A, Iwama S. Combined hepatocellular-cholangiocarcinoma with variable sarcomatous transformation. *Am J Clin Pathol* 1988; **90**: 309-312 [PMID: 2458034]

20 **Jeong BJ**, Hyun DH, Lee KW, Ryu ST, Lee JW, Lee JI, Jeong S, Lee DH, Kim PS, Kim HG, Kim YS, Kim JM. [A case of sarcomatoid combined hepatocellular-cholangiocarcinoma]. *Korean J Gastroenterol* 2004; **43**: 56-60 [PMID: 14745254]

21 **Kim JH**, Lee YG, Lee J, Jung CK, Kim HT, Kang H, Kho KH, Hong SP, Hwang SG, Park PW, Rim GS. [A case of combined hepatocellular-cholangiocarcinoma with sarcomatous transformation and second primary colon cancer]. *Korean J Hepatol* 2004; **10**: 142-147 [PMID: 15218348]

22 **Xiang S**, Chen YF, Guan Y, Chen XP. Primary combined hepatocellular-cholangiocellular sarcoma: An unusual case. *World J Gastroenterol* 2015; **21**: 7335-7342 [PMID: 26109824 DOI: 10.3748/wjg.v21.i23.7335]

23 **Terada T**. Combined hepatocellular-cholangiocarcinoma with stem cell features, ductal plate malformation subtype: a case report and proposal of a new subtype. *Int J Clin Exp Pathol* 2013; **6**: 737-748 [PMID: 23573322]

24 **Jung W**, Kim BH. Cholangiolocellular carcinoma with satellite nodules showing intermediate differentiation. *Clin Mol Hepatol* 2015; **21**: 183-186 [PMID: 26157757 DOI: 10.3350/cmh.2015.21.2.183]

25 **Ikeda H**, Harada K, Sato Y, Sasaki M, Yoneda N, Kitamura S, Sudo Y, Ooi A, Nakanuma Y. Clinicopathologic significance of combined hepatocellular-cholangiocarcinoma with stem cell subtype components with reference to the expression of putative stem cell markers. *Am J Clin Pathol* 2013; **140**: 329-340 [PMID: 23955451 DOI: 10.1309/ajcp66avbanvntqj]

26 **Sasaki M**, Sato H, Kakuda Y, Sato Y, Choi JH, Nakanuma Y. Clinicopathological significance of 'subtypes with stem-cell feature' in combined hepatocellular-cholangiocarcinoma. *Liver Int* 2015; **35**: 1024-1035 [PMID: 24712771 DOI: 10.1111/liv.12563]

27 **Yang X**, Li M, Liu Q, Zhang Y, Qian J, Wan X, Wang A, Zhang H, Zhu C, Lu X, Mao Y, Sang X, Zhao H, Zhao Y, Zhang X. Dr.VIS v2.0: an updated database of human disease-related viral integration sites in the era of high-throughput deep sequencing. *Nucleic Acids Res* 2015; **43**: D887-D892 [PMID: 25355513 DOI: 10.1093/nar/gku1074]

28 **Tomimatsu M**, Ishiguro N, Taniai M, Okuda H, Saito A, Obata H, Yamamoto M, Takasaki K, Nakano M. Hepatitis C virus antibody in patients with primary liver cancer (hepatocellular carcinoma, cholangiocarcinoma, and combined hepatocellular-cholangiocarcinoma) in Japan. *Cancer* 1993; **72**: 683-688 [PMID: 8192727]

29 **Zhou YM**, Zhang XF, Wu LP, Sui CJ, Yang JM. Risk factors for combined hepatocellular-cholangiocarcinoma: a hospital-based case-control study. *World J Gastroenterol* 2014; **20**: 12615-12620 [PMID: 25253966 DOI: 10.3748/wjg.v20.i35.12615]

30 **Lee CC**, Wu CY, Chen JT, Chen GH. Comparing combined hepatocellular-cholangiocarcinoma and cholangiocarcinoma: a clinicopathological study. *Hepatogastroenterology* 2002; **49**: 1487-1490 [PMID: 12397714]

31 **Toh CH**, Cheung YC, Ng SH, Lin CY, Chan SC, Ng KK.

Combined hepatocellular-cholangiocarcinoma: a case report. *Int J Clin Pract* 2004; **58**: 1170-1173 [PMID: 15646418]
32 **Wan XS**, Xu YY, Qian JY, Yang XB, Wang AQ, He L, Zhao HT, Sang XT. Intraductal papillary neoplasm of the bile duct. *World J Gastroenterol* 2013; **19**: 8595-8604 [PMID: 24379576 DOI: 10.3748/wjg.v19.i46.8595]
33 **Wachtel MS**, Zhang Y, Xu T, Chiriva-Internati M, Frezza EE. Combined hepatocellular cholangiocarcinomas; analysis of a large database. *Clin Med Pathol* 2008; **1**: 43-47 [PMID: 21876650]
34 **Ng IO**, Shek TW, Nicholls J, Ma LT. Combined hepatocellular-cholangiocarcinoma: a clinicopathological study. *J Gastroenterol Hepatol* 1998; **13**: 34-40 [PMID: 9737569]
35 **Sanada Y**, Shiozaki S, Aoki H, Takakura N, Yoshida K, Yamaguchi Y. A clinical study of 11 cases of combined hepatocellular-cholangiocarcinoma Assessment of enhancement patterns on dynamics computed tomography before resection. *Hepatol Res* 2005; **32**: 185-195 [PMID: 15978872 DOI: 10.1016/j.hepres.2005.04.003]
36 **Shin CI**, Lee JM, Kim SH, Choi JY, Lee JY, Han JK, Jo SY, Choi BI. Recurrence patterns of combined hepatocellular-cholangiocarcinoma on enhanced computed tomography. *J Comput Assist Tomogr* 2007; **31**: 109-115 [PMID: 17259842 DOI: 10.1097/01.rct.0000235072.34808.9b]
37 **Fowler KJ**, Sheybani A, Parker RA, Doherty S, M Brunt E, Chapman WC, Menias CO. Combined hepatocellular and cholangiocarcinoma (biphenotypic) tumors: imaging features and diagnostic accuracy of contrast-enhanced CT and MRI. *AJR Am J Roentgenol* 2013; **201**: 332-339 [PMID: 23883213 DOI: 10.2214/ajr.12.9488]
38 **Dusenbery D**. Combined hepatocellular-cholangiocarcinoma. Cytologic findings in four cases. *Acta Cytol* 1997; **41**: 903-909 [PMID: 9167724]
39 **Kilpatrick SE**, Geisinger KR, Loggie BW, Hopkins MB. Cytomorphology of combined hepatocellular-cholangiocarcinoma in fine needle aspirates of the liver. A report of two cases. *Acta Cytol* 1993; **37**: 943-947 [PMID: 7504382]
40 **Eguchi H**, Nagano H, Sakon M, Miyamoto A, Kondo M, Arai I, Morimoto O, Dono K, Umeshita K, Nakamori S, Wakasa K, Monden M. A successful resection and long-term survival of a patient with intrahepatic recurrences of combined hepatocellular-cholangiocarcinoma: report of a case. *Surg Today* 2002; **32**: 742-746 [PMID: 12181730 DOI: 10.1007/s005950200140]
41 **Chan AC**, Lo CM, Ng IO, Fan ST. Liver transplantation for combined hepatocellular cholangiocarcinoma. *Asian J Surg* 2007; **30**: 143-146 [PMID: 17475587 DOI: 10.1016/s1015-9584(09)60149-4]
42 **Perkins JD**. Transplanted patients with combined hepatocellular-cholangiocarcinoma: now what? *Liver Transpl* 2007; **13**: 1465-1466 [PMID: 17972421]
43 **Panjala C**, Senecal DL, Bridges MD, Kim GP, Nakhleh RE, Nguyen JH, Harnois DM. The diagnostic conundrum and liver transplantation outcome for combined hepatocellular-cholangiocarcinoma. *Am J Transplant* 2010; **10**: 1263-1267 [PMID: 20420633 DOI: 10.1111/j.1600-6143.2010.03062.x]
44 **Vilchez V**, Shah MB, Daily MF, Pena L, Tzeng CW, Davenport D, Hosein PJ, Gedaly R, Maynard E. Long-term outcome of patients undergoing liver transplantation for mixed hepatocellular carcinoma and cholangiocarcinoma: an analysis of the UNOS database. *HPB* (Oxford) 2016; **18**: 29-34 [PMID: 26776848 DOI: 10.1016/j.hpb.2015.10.001]
45 **Groeschl RT**, Turaga KK, Gamblin TC. Transplantation versus resection for patients with combined hepatocellular carcinoma-cholangiocarcinoma. *J Surg Oncol* 2013; **107**: 608-612 [PMID: 23386397 DOI: 10.1002/jso.23289]
46 **Itoh S**, Ikegami T, Yoshizumi T, Wang H, Takeishi K, Harimoto N, Yamashita Y, Kawanaka H, Aishima S, Shirabe K, Maehara Y. Long-term outcome of living-donor liver transplantation for combined hepatocellular-cholangiocarcinoma. *Anticancer Res* 2015; **35**: 2475-2476 [PMID: 25862916]
47 **Hayashi H**, Beppu T, Ishiko T, Mizumoto T, Masuda T, Okabe K, Baba Y, Okabe H, Takamori H, Kanemitsu K, Hiroto M, Baba H. [A 42-month disease free survival case of combined hepatocellular-cholangiocarcinoma with lymph node metastases treated with multimodal therapy]. *Gan To Kagaku Ryoho* 2006; **33**: 1941-1943 [PMID: 17212153]
48 **Shimizu J**, Hayashi S, Dono K, Yasumoto T, Zenitani M, Munakata K, Watanabe N, Takamoto K, Kagawa Y, Hata T, Kawanishi K, Ikeda K, Fujita J, Akagi K, Kitada M, Shimano T. [A case report of combined hepatocellular-cholangiocarcinoma whose lymph node recurrence effectively treated with UFT]. *Gan To Kagaku Ryoho* 2009; **36**: 2380-2382 [PMID: 20037429]
49 **Kim GM**, Jeung HC, Kim D, Kim JH, Yoon SH, Jung ES, Shin SJ. A case of combined hepatocellular-cholangiocarcinoma with favorable response to systemic chemotherapy. *Cancer Res Treat* 2010; **42**: 235-238 [PMID: 21253326 DOI: 10.4143/crt.2010.42.4.235]
50 **Tickoo SK**, Zee SY, Obiekwe S, Xiao H, Koea J, Robiou C, Blumgart LH, Jarnagin W, Ladanyi M, Klimstra DS. Combined hepatocellular-cholangiocarcinoma: a histopathologic, immunohistochemical, and in situ hybridization study. *Am J Surg Pathol* 2002; **26**: 989-997 [PMID: 12170085]
51 **Zhou L**, Rui JA, Wang SB, Chen SG, Qu Q, Chi TY, Wei X, Han K, Zhang N, Zhao HT. Outcomes and prognostic factors of cirrhotic patients with hepatocellular carcinoma after radical major hepatectomy. *World J Surg* 2007; **31**: 1782-1787 [PMID: 17610113 DOI: 10.1007/s00268-007-9029-z]
52 **Park SE**, Lee SH, Yang JD, Hwang HP, Hwang SE, Yu HC, Moon WS, Cho BH. Clinicopathological characteristics and prognostic factors in combined hepatocellular carcinoma and cholangiocarcinoma. *Korean J Hepatobiliary Pancreat Surg* 2013; **17**: 152-156 [PMID: 26155231 DOI: 10.14701/kjhbps.2013.17.4.152]
53 **Chu KJ**, Lu CD, Dong H, Fu XH, Zhang HW, Yao XP. Hepatitis B virus-related combined hepatocellular-cholangiocarcinoma: clinicopathological and prognostic analysis of 390 cases. *Eur J Gastroenterol Hepatol* 2014; **26**: 192-199 [PMID: 24370644 DOI: 10.1097/MEG.0b013e3283625df9]
54 **Kim SH**, Park YN, Lim JH, Choi GH, Choi JS, Kim KS. Characteristics of combined hepatocelluar-cholangiocarcinoma and comparison with intrahepatic cholangiocarcinoma. *Eur J Surg Oncol* 2014; **40**: 976-981 [PMID: 24909336 DOI: 10.1016/j.ejso.2014.04.016]
55 **Miao R**, Luo H, Zhou H, Li G, Bu D, Yang X, Zhao X, Zhang H, Liu S, Zhong Y, Zou Z, Zhao Y, Yu K, He L, Sang X, Zhong S, Huang J, Wu Y, Miksad RA, Robson SC, Jiang C, Zhao Y, Zhao H. Identification of prognostic biomarkers in hepatitis B virus-related hepatocellular carcinoma and stratification by integrative multi-omics analysis. *J Hepatol* 2014; **61**: 840-849 [PMID: 24859455 DOI: 10.1016/j.jhep.2014.05.025]

P- Reviewer: Avalos-Gonzalez J **S- Editor**: Qi Y
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ISSN 1007-9327