Risk Factors for Cholangiocarcinoma

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Cholangiocarcinoma (CC) is the second most common primary hepatic malignancy after hepatocellular cancer. CC accounts for approximately 10%-25% of all hepatobiliary malignancies. There are considerable geographic and demographic variations in the incidence of CC. There are several established risk factors for CC, including parasitic infections, primary sclerosing cholangitis, biliary-duct cysts, hepatolithiasis, and toxins. Other less-established potential risk factors include inflammatory bowel disease, hepatitis C virus, hepatitis B virus, cirrhosis, diabetes, obesity, alcohol drinking, tobacco smoking, and host genetic polymorphisms. In studies where the distinction between intra- and extrahepatic CC was used, some potential risk factors seem to have a differential effect on CC, depending on the site. Therefore, the consistent use of a more refined classification would allow a better understanding of risk factors for CC. (HEPATOLOGY 2011;54:173-184)

Cholangiocarcinoma (CC) is a malignant neoplasm of the biliary-duct system accounting for 3% of gastrointestinal tumors.[1-3] It is the second most common primary hepatic malignancy, representing 10%-25% of primary hepatic malignancies worldwide.[1,4,5] CC rarely occurs before the age of 40; the typical age at presentation is the seventh decade of life.[3,4] Men have a higher incidence of CC than do women,[3,6-9] with ratios of 1:1.2-1.5. The incidence of CC varies greatly by geographic region secondary to variations in risk factors.[3,5] The prognosis of CC is poor; therefore, mortality and incidence rates are similar. Although there are established risk factors for the development of CC, most patients do not have an identifiable risk aside from age.[1,4]

Anatomically, CC can be classified as intra- or extrahepatic in location.[2] Hilar CC (i.e., Klatskin tumors) is typically considered extrahepatic. The distinction between intrahepatic CC (ICC) and extrahepatic CC (ECC) has become increasingly important, as the epidemiological feature (i.e., incidence and risk factors) associated with each may be different.[1,2] In this review, we will distinguish between ICC and ECC, because epidemiological differences may exist between them. Otherwise, CC will be used when studies do not distinguish between ICC and ECC.

Population-based incidence data on CC are sparse. Most cancer registries combine cases of CC with other hepatobiliary malignancies, such as hepatocellular cancer (HCC) and gallbladder cancer.[6,8] Worldwide, the incidence of CC varies greatly.[3,8] Regions such as Thailand in Southeast Asia have the highest incidence of CC, as high as 113 per 100,000 in men and 50 per 100,000 in women, whereas in Western countries such as Australia, the incidence is low, at 0.2 per 100,000 in men and 0.1 per 100,000 in women.[3,5] Differing exposure to risk factors is thought to account for the varying geographic incidences, with parasitic infections and hepatolithiasis being more prevalent in Asia.[3,5] Several studies published in the early 2000s reported international trends in increased incidence of ICC and decreased incidence of ECC,[6,8] but the role that misclassification of hepatobiliary cancer plays in explaining epidemiological trends may be substantial.

The incidence of CC in the United States is reported from the population-based registries of the Surveillance, Epidemiology, and End Results (SEER) program. The age-adjusted annual incidence of ICC increased from 0.13 per 100,000 persons in 1973 to

Abbreviations: BMI, body mass index; CC, cholangiocarcinoma; CI, confidence interval; ECC, extrahepatic cholangiocarcinoma; HBV, hepatitis B virus; HCC, hepatocellular cancer; HCV, hepatitis C virus; IBD, inflammatory bowel disease; ICC, intrahepatic cholangiocarcinoma; OR, odds ratio; PSC, primary sclerosing cholangitis.

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0.67 per 100,000 in 1997[7] and to 0.85 per 100,000 persons during 1995-1999 and a decline in ECC incidence from 1.08 per 100,000 in 1979 to 0.82 per 100,000 in 1998.[3] However, the recent SEER data from 2000 to 2005 show that the annual incidence of ICC has declined to 0.58 per 100,000 and that of ECC has increased to 0.88 per 100,000.

Differences among studies, registries, and classification of ICC and ECC may account for some of the temporal variations observed in CC (i.e., ICC and ECC). For example, Klatskin tumors were not given a unique code in Version 1 of the ICD-O (International Classification of Diseases for Oncology) (1973-1991); therefore, it could have been characterized topographically as ICC or ECC. In Version 2 of the ICD-O (1992-2000), it was given a unique histology code that could be linked to ICC, rather than ECC. In Version 3 of the ICD-O (2001-present), the histological code could be linked to either ICC or ECC.[10]

In addition to the misclassification of Klatskin tumors, there are other possible reasons for the misclassification of CC, including the detection of CCs at an advanced stage, which makes it difficult to determine the anatomical origin, and the histological variation of CCs, which can result in their classification as other hepatobiliary malignancies. Given that CC is a relatively rare liver cancer in most world regions, misclassifications can substantially impact the findings of epidemiological studies. Consequently, no definitive statement can be made on the temporal trends of CC in most world regions in the absence of striking consistent trends. For example, in the United States, Welzel et al. reported that misclassification of Klatskin tumors had contributed to the temporal trends of increasing ICC and decreasing ECC between 1992 and 2000.[10] Furthermore, recent SEER data (2000-2005) suggest that the temporal trends are reversing, with decreased ICC and increased ECC incidence.[11]

Risk Factors for CC

There are several established risk factors for CC, including parasitic infections, primary sclerosing cholangitis, biliary-duct cysts, hepatolithiasis, and toxins. Other less-established potential risk factors include inflammatory bowel disease (IBD), hepatitis C virus (HCV), hepatitis B virus (HBV), cirrhosis, diabetes, obesity, alcohol, smoking, and host genetic polymorphisms. In studies where the distinction between ICC and ECC was used, some potential risk factors seem to have a differential effect on CC, depending on the site. Therefore, the consistent use of a more refined classification would allow a better understanding of risk factors for CC.

Established Risk Factors for CC

Parasitic Infections

The hepatobiliary flukes, *Opisthorchis viverrini* and *Clonorchis sinensis*, are associated with the development of CC, particularly in Southeast Asia. They are flat worms that inhabit the bile ducts and, occasionally, the gallbladder and pancreatic duct of mammals. Eggs laid by the adult worms are passed in feces, which may be ingested by snails, where they hatch and then mature into cercariae and, subsequently, penetrate the flesh of freshwater fish, where they develop into metacercariae. Infection in humans occurs via the ingestion of raw, pickled, or undercooked fish.[12,13]

Both parasites increase the susceptibility of cholangiocytes to endo- and exogenous carcinogens via chronic irritation and increased cellular turnover.[12] In 1994, *O. viverrini* was deemed by the International Agency for Research on Cancer as "carcinogenic to humans" secondary to its role in the development of CC. In 2009, the same classification was given to *C. sinensis*.[14] Parasitic infections, particularly *O. viverrini*, are a major public health issue in Thailand, where the incidence of CC is still increasing in some Northeastern regions and is strongly correlated with the prevalence of parasitic infections.[5]

One of the early epidemiological studies (1987-1988) to show a relationship between *O. viverrini* and CC was a hospital-based, case-control study conducted in Thailand by Parkin et al., in which 103 patients with CC were compared with an equal number of age- and sex-matched controls. A strong association was found between elevated *O. viverrini* antibody titers and increased risk of CC (odds ratio [OR] = 5.0; 95% confidence interval [CI] = 2.3-11.0).[15] A more recent (1999-2001) population-based, case-control study from Thailand compared 129 cases of CC with an equal number of age- and sex-matched controls. Elevated *O. viverrini* antibody levels were, again, strongly associated with CC (OR = 27.09; 95% CI = 6.30-116.57). In endemic areas, the population-attributable risk, based on this study, was as high as 88%.[16]

A case-control study by Shin et al. from Korea compared 41 patients with CC with 406 controls and reported a strong association between the presence of *C. sinensis* in the stool and CC (OR = 2.7; 95% CI = 1.1-6.3).[17] A subsequent 2009 meta-analysis, performed by Shin et al., pooled 912 cases and 4909

controls and confirmed the strong association between *C. sinensis* and CC (OR = 4.7; 95% CI = 2.2-9.8). In endemic areas, the population-attributable risk, based on this study, was as high as 27.9% for men and 16.2% for women.[14]

Biliary-Tract Disorders

Bile-Duct Cysts

Bile (i.e., choledochal)-duct cysts are rare congenital disorders characterized by cystic dilatation of the extra- and/or intrahepatic bile ducts. Bile-duct cysts are thought to develop from an abnormal pancreatico-biliary junction, in which the pancreatic and biliary ducts join outside the duodenum and are typically associated with a long common channel (>10 mm).[18] This results in pancreatic enzymes refluxing into the biliary system with subsequent increased intraductal pressure and inflammation, leading to ductal dilatation. With regards to Caroli's disease, the abnormality is attributed to malformation of the ductal plate.[19] It has been postulated that the reflux of pancreatic enzymes, bile stasis, and increased concentration of intraductal bile acids contribute to the formation of malignant cells in patients with bile-duct cysts.[20]

Bile-duct cysts are an established risk factor for CC. Type I (i.e., solitary, extrahepatic) and IV (i.e., extra- and intrahepatic) bile-duct cysts have the higher incidence of CC.[20] The lifetime incidence of CC in these patients ranges from 6%-30%.[4,20] The prevalence of bile-duct cysts is higher in Asian than Western countries.[19-23] The incidence of CC is also higher in Asians with bile-duct cysts, at approximately 18%, with the U.S. incidence closer to 6%.[19,21,23-25] There is an increase in incidence of CC in patients with bile-duct cysts from 0.7% in the first decade of life to >14% after age 20.[26] The average age at malignancy detection has been reported to be 32 years, which is younger than the age at presentation of CC in the general population.[20,24] The risk of malignancy decreases after complete choledochal cyst excision; however, these patients are still at an increased risk of developing CC, compared with the general population.[19-22,25]

Patients with bile-duct cysts are reported to have at least a 10- to 50-fold increased risk of developing CC.[20,27,28] In a Korean, hospital-based, case-control study by Lee et al., there was a strong association between choledochal cysts and ICC, with the OR at 10.7 (95% CI = 1.8-63.9).[27] In a large, SEER-Medicare study by Welzel et al., there was a strong association between choledochal cysts and increased risk of both ICC and ECC, with ORs of 36.9 (95% CI = 22.7-59.7) and 47.1 (95% CI = 30.4-73.2), respectively.[28]

Primary Sclerosing Cholangitis

Primary sclerosing cholangitis (PSC), an autoimmune disease that results in the stricturing of extra- and/or intrahepatic bile ducts, is an established risk factor for CC. Chronic inflammation, proliferation of biliary epithelium, production of endogenous bile mutagens, and bile stasis are postulated mechanisms of carcinogenesis.[2] The lifetime incidence of CC among PSC patients ranges from 6%-36%.[29,30] Although PSC is known to be a strong risk factor for CC, no more than 10% of CC is attributed to PSC.[30] Data on the incidence of PSC suggest either no change or a small increase over time. A recent study by Card et al. showed a nonsignificant rising trend in the incidence of PSC between 1987 and 2002, but the overall incidence estimates in this study were generally lower than most other reports.[31] A subsequent study by Lindkvist et al. reported a significantly increased incidence of PSC between 1992 and 2005.[32] Given that PSC is the most common known risk factor for CC in the West, trending the incidence of PSC is important for monitoring trends in CC.

A hospital-based, retrospective cohort study by Burak et al. from the Mayo Clinic followed 161 patients with PSC for a median of 11.5 years; 11 patients (6.8%) developed CC, with an incidence rate of 0.6% per year. The median time from diagnosis of PSC to diagnosis of CC was 4.1 years (range, 0.8-15.0), and no association was found between the duration of PSC and the risk of CC.[33] Another hospital-based, retrospective cohort study by Claessen et al. followed 211 patients with PSC for a median of 9 years; 7% developed CC. There was no association between the duration of PSC and the risk of CC, with nearly all the cases of CC presenting within 3 years of PSC diagnosis.[29] It is unclear whether the duration of PSC correlates with the risk of developing CC; in fact, most cases present relatively soon after PSC diagnosis. Cohort studies suggest that CC develops within the first 1-2 years of PSC diagnosis. A cohort study by Boberg et al. found that 48 of 394 (12.2%) patients with PSC developed CC, with 24 of them being diagnosed within 1 year of the diagnosis of PSC.[34] In a Swedish cohort study, 14 of 125 (11.2%) patients with PSC developed CC. Eleven of the 14 (~78%) were diagnosed with CC within 2 years of the diagnosis of PSC.[35] Possible explanations for these observations include that early CC may be partly responsible for the patient with PSC seeking medical attention. Given

the difficulty of diagnosing early CC in PSC, the initial presentation may result in a diagnosis of the PSC, but the CC is not diagnosed until later.

Smoking and alcohol consumption have also been examined as risk factors for CC in patients with PSC. A case-control study by Chalasani et al. and a cohort study by Burak et al. did not find smoking to be a significant risk factor, whereas a case-control study by Berquist et al. found a significant association (10 versus 0 patients; $P < 0.0004$).[33,36,37] Chalasani et al. also looked at alcohol consumption and reported a significant association between self-reported present or past alcohol consumption and increased risk of CC in patients with PSC (OR = 2.95; 95% CI = 1.04-8.3).[37] There are no definitive data to suggest that smoking and/or alcohol consumption confer an increased risk of CC in PSC patients.

Hepatolithiasis

Hepatolithiasis is the presence of calculi or concretions located proximal to the confluence of the right and left hepatic ducts. Hepatolithiasis is found mainly in Southeast Asia (e.g., up to 20% in Taiwan) and is rare in the West (1%-2%). It has been postulated that prolonged irritation and inflammation of the biliary epithelium by the calculi, bile stasis, and bacterial infections predispose to malignancy.[38,39] In addition, infestation with parasites, such as *C. sinensis* and *Ascaris lumbricoides*, has been shown in up to 30% of patients with hepatolithiasis.[40]

Hepatolithiasis is an established risk factor for ICC in Asian countries, with 2-10% of patients with hepatolithiasis developing ICC.[4,38,39] The Korean, hospital-based, case-control study by Lee et al. found a strong association between hepatolithiasis and ICC, with an OR of 50.0 (95% CI = 21.2-117.3).[27] A Chinese, hospital-based, case-control study by Zhou et al. also showed a significant association, with the OR at 5.8 (95% CI = 1.97-16.9).[41] There are less data on the relationship between hepatolithiasis and ICC in Western countries, but an Italian, hospital-based, case-control study also showed a significant association between hepatolithiasis and ICC, with an OR of 6.7 (95% CI = 1.3-33.4).[42]

Toxins

The currently banned carcinogenic agent, Thorotrast, a radiographic contrast agent used primarily from 1930 to 1960, has been strongly associated with an increased risk of developing CC. The estimated latency period between exposure and malignancy diagnosis ranges between 16 and 45 years; this is because the biological half-life of Thorotrast is 400 years.[43] The association between Thorotrast and CC was best shown in a Japanese study that followed 241 patients exposed to Thorotrast during World War II. The study found a more than 300-fold increased risk of CC in exposed patients, compared with nonexposed controls.[44] Other large studies from Germany and Denmark have also shown a significantly increased risk of CC among patients exposed to Thorotrast.[43,45,46]

Possible Risk Factors for Cholangiocarcinoma

IBD

Most data describing the association between IBD and CC pertains to patients with IBD and PSC. In the cohort study by Boberg et al., there was a significantly longer duration of IBD in PSC patients with CC than in those without CC (17.4 versus 9.0 years, respectively).[34] Yet, the cohort studies by Burak et al. and Claessen et al. did not find a significant association between the presence of IBD and CC in patients with PSC.[29,33] In the Swedish cohort study, the cumulative risk of developing CC in PSC patients with IBD for more than 20 years did not differ from that of those with a disease duration of less than 20 years (7% versus 8%).[35] The presence and magnitude of association between IBD and CC is likely to be affected by the presence of PSC and by the duration of observation in each study. This is related to the unpredictable onset point for each of PSC and IBD during the course of the other condition. This complexity makes the associations among PSC, IBD, and CC difficult to define.

However, there are studies that evaluate IBD, both ulcerative colitis and Crohn's disease, as risk factors independent of PSC for CC (Table 1). Two SEER-Medicare studies showed a positive association of ICC with ulcerative colitis, but not with Crohn's disease.[28,47] One of the studies showed that Crohn's disease was significantly associated with ECC.[28] A Danish, population-based study by Welzel showed that IBD, type not specified, was significantly associated with ICC.[48] A different Danish, population-based cohort study also found a positive association between UC and CC, but no association with Crohn's disease. There were no reported differences in those data for ICC versus ECC.[49] In these studies, PSC was not controlled for in the analysis of IBD; therefore, it remains unclear whether IBD is an independent risk factor for CC. Although IBD may be a risk factor for CC, likely via

Table 1. IBD as a Potential Risk Factor for Cholangiocarcinoma

First Author	Country	Study Dates	Study Design	Risk Factor	CC Type	Cases (% With Risk Factor)	Controls (% With Risk Factor)	Risk Estimate (95% CI)	Selected Adjusted Variables
Welzel[48]	Denmark	1978-1991	Case-control	IBD	ICC	764 (0.92%)	3056 (0.20%)	4.67 (1.6-13.9)	Age, sex
Erichsen[49]	Denmark	1978-2003	Cohort	UC	ECC/ICC	Incidence rate 8.2	Incidence rate 2.0	4.1 (2.4-6.8)	Age, sex
				Crohn's disease	ECC/ICC	Incidence rate 4.3	Incidence rate 1.4	3.0 (0.9-8.6)	
Shaib[47]	United States	1993-1999	Case-control	UC	ICC	625	90,834	2.2 (1.2-3.9)	Age, sex, race, geographic location
				Crohn's disease	ICC	625	90,834	2.0 (0.6-6.3)	
Welzel[28]	United States	1993-1999	Case-control	UC	ICC	535 (2.4%)	102,782 (0.6%)	4.5 (2.6-7.9)	Age, sex, race, geographic location
				Crohn's disease	ICC	535 (0.9%)	102,782 (0.4%)	2.4 (1.0-5.9)	
				UC	ECC	549 (0.9%)	102,782 (0.6%)	1.7 (0.7-4.0)	
				Crohn's disease	ECC	549 (1.1%)	102,782 (0.4%)	2.8 (1.3-6.4)	

Abbreviations: CC, cholangiocarcinoma; CI, confidence interval; ECC, extrahepatic cholangiocarcinoma; IBD, inflammatory bowel disease; ICC, intrahepatic cholangiocarcinoma; UC, ulcerative colitis.

PSC, it is not clear that IBD confers any added risk for CC in PSC patients.

Choledocholithiasis and Cholangitis

Given that proposed mechanisms for CC formation involve chronic inflammation and bile stasis, studies have examined choledocholithiasis and cholangitis as risk factors for CC (Table 2). The two large SEER-Medicare studies showed a strong positive association of CC with choledocholithiasis and cholangitis, with risk estimates ranging from 4 to 64.[28,47] In the Danish, population-based, case-control study conducted by Welzel et al., choledocholithiasis and cholangitis were, again, significantly associated with ICC.[48] These studies could not definitively exclude PSC-associated cholangitis; therefore, it is unclear whether choledocholithiasis and/or cholangitis are independent risk factors for ICC or ECC.

Chronic Viral Hepatitis and Cirrhosis

HCV, HBV, and liver cirrhosis, regardless of etiology, have been postulated as risk factors for CC (Tables 3-5). Tobersenson et al. reviewed the pathology of more than 1000 explanted livers and found bile-duct dysplasia, a precursor lesion to CC, in approximately 2% of the livers. All affected livers were from patients with underlying cirrhosis caused by HCV, alcohol, or both.[50] The study supports the biologic plausibility of chronic viral hepatitis and cirrhosis as potential risk factors for CC.

Asian Studies. Several case-control studies, all hospital based, examined viral hepatitis in relation to CC. A Korean case-control study by Shin et al. that

Table 2. Cholangitis and Choledocholithiasis as Potential Risk Factors for Cholangiocarcinoma

First Author	Country	Study Dates	Study Design	Risk Factor	CC Type	Cases (% With Risk Factor)	Controls (% With Risk Factor)	Risk Estimate (95% CI)	Selected Adjusted Variables
Welzel[48]	Denmark	1978-1991	Case-control	Cholangitis	ICC	764 (1.3%)	3056 (0.23%)	6.32 (2.3-17.5)	Age, sex
				Choledocholithiasis	ICC	764 (0.79%)	3056 (0.03%)	23.97 (2.9-198.9)	
Shaib[47]	United States	1993-1999	Case-control	Cholangitis	ICC	625 (3.4%)	90,834 (0.2%)	8.8 (4.9-16.0)	Age, sex, race, geographic location
				Choledocholithiasis	ICC	625 (1.1%)	90,834 (0.3%)	4.0 (1.9-8.5)	
Welzel[28]	United States	1993-1999	Case-control	Cholangitis	ICC	535 (12.5%)	102,782 (0.2%)	64.2 (47.7-86.5)	Age, sex, race, geographic location
				Choledocholithiasis	ICC	535 (11%)	102,782 (0.5%)	22.5 (16.9-30.0)	
				Cholangitis	ECC	549 (9.1%)	102,782 (0.2%)	45.7 (32.9-63.6)	
				Choledocholithiasis	ECC	549 (15.8%)	102,782 (0.5%)	34.0 (26.6-43.6)	

Abbreviations: CC, cholangiocarcinoma; CI, confidence interval; ECC, extrahepatic cholangiocarcinoma; ICC, intrahepatic cholangiocarcinoma.

Table 3. Hepatitis B Virus as a Potential Risk Factor for Cholangiocarcinoma

First Author	Country	Study Date	Risk Factor	CC Type	Cases (% With Risk Factor)	Controls (% With Risk Factor)	Risk Estimate (95% CI)	Selected Adjusted Variables
Shin[17]	Korea	1990-1993	$HBsAg^+$	NS	41 (12.5%)	406 (3.5%)	1.3 (0.3-5.3)	Age, sex, socioeconomic status
Yamamoto[51]	Japan	1991-2002	$HBsAg^+$	ICC	50 (4%)	205 (2%)	Not calculated	Age, sex
Shaib[53]	United States	1992-2002	$HBsAg^-/Anti\text{-}HBc^+$	ICC	83 (9.6%)	236 (0%)	28.6 (3.9-1268.1)	Age, sex, race
			$HBsAg^-/Anti\text{-}HBc^+$	ECC	163 (0%)	236 (0%)	3.2 (0.6-382)	
Shaib[47]	United States	1993-1999	^HBV	ICC	625 (0.2%)	90,834 (0.2%)	0.8 (0.1-5.9)	Age, sex, race, geographic location
Donato[42]	Italy	1995-2000	$HBsAg^+$	ICC	26 (11.5%)	824 (5.5%)	2.7 (0.4-18.5)	Age, sex, residence
Lee[27]	Korea	2000-2004	$HBsAg^+$	ICC	622 (13.5%)	2,488 (5.0%)	2.3 (1.6-3.3)	Age, sex
Zhou[41]	China	2004-2006	$HbsAg^+$	ICC	312 (48.4%)	438 (9.6%)	8.9 (5.97-13.2)	Age, sex, date of admission

Abbreviations: ^, diagnostic code; CC, cholangiocarcinoma; CI, confidence interval; ECC, extrahepatic cholangiocarcinoma; HBc, hepatitis B core; HBsAg, hepatitis B surface antigen; HBV, hepatitis B virus; ICC, intrahepatic cholangiocarcinoma; NS, not specified.

compared 41 cases of CC with 406 noncancer controls did not find a significant association between HBV or HCV seropositivity and CC.[17] In another Korean case-control study by Lee et al. that compared 622 cases of ICC with 2488 controls, there was a significant association between ICC and HBV as well as cirrhosis of any etiology. There was no significant association between HCV seropositivity and ICC.[27] A case-control study from China by Zhou et al. compared 312 ICC cases with 438 controls and reported a strong association between ICC and HBV seropositivity, but no significant association with HCV seropositivity.[41] Lastly, a case-control study from Japan by Yamamoto et al. reported that HCV was a significant risk factor for ICC. The presence of cirrhosis merely trended toward significance, whereas HBV infection was not a significant risk factor for ICC.[51]

European Studies. Few Western European studies reported an association between CC and both HCV and cirrhosis. A large, population-based cohort study from Denmark by Sorensen et al. examined cancer risk in 11,605 patients with cirrhosis over a mean follow-

Table 4. Hepatitis C Virus as a Potential Risk Factor for Cholangiocarcinoma

First author	Country	Study Dates	Study Design	Risk Factor	CC Type	Cases (% With Risk Factor)	Controls (% With Risk Factor)	Risk Estimate (95% CI)	Selected Adjusted Variables
El-Serag[54]	United States	1988-2004	Cohort	^HCV	ICC	4/100,000 person-years	1.6/100,000 person-years	2.55 (1.3-4.9)	Age, sex
				^HCV	ECC	4.3/100,000 person-years	4.2/100,000 person-years	1.05 (0.6-1.9)	
Shin[17]	Korea	1990-1993	Case-control	$Anti\text{-}HCV^+$	NS	41 (13.8%)	406 (2.3%)	3.9 (0.9-17.1)	Age, sex, SES
Yamamoto[51]	Japan	1991-2002	Case-control	$Anti\text{-}HCV^+$	ICC	50 (36%)	205 (3%)	6.02 (1.5-24.1)	Age, sex
Shaib[53]	United States	1992-2002	Case-control	$Anti\text{-}HCV^+$	ICC	83 (6%)	236 (0.8%)	7.9 (1.3-84.5)	Age, sex, race
				$Anti\text{-}HCV^+$	ECC	163 (3.7%)	236 (0.8%)	2.8 (0.3-35.1)	
Shaib[47]	United States	1993-1999	Case-control	^HCV	ICC	625 (0.8%)	90,834 (0.2%)	5.2 (2.1-12.8)	Age, sex, race, geographic location
Welzel[28]	United States	1993-1999	Case-control	^HCV	ICC	535 (<0.9%)	102,782 (0.1%)	4.4 (1.4-14.0)	Age, sex, race, geographic location
				^HCV	ECC	549 (<0.9%)	102,782 (0.1%)	1.5 (0.2-11.0)	
Donato[42]	Italy	1995-2000	Case-control	$Anti\text{-}HCV^+$	ICC	26 (23%)	824 (6%)	9.7 (1.6-58.9)	Age, sex, residence
Lee[27]	Korea	2000-2004	Case-control	$Anti\text{-}HCV^+$	ICC	622 (1.9%)	2488 (1.9%)	Not calculated	Age, sex
Zhou[41]	China	2004-2006	Case-control	$Anti\text{-}HCV^+$	ICC	312 (2.9%)	438 (1.4%)	0.93 (0.3-3.1)	Age, sex, date of admission

Abbreviations: ^, diagnostic code; CC, cholangiocarcinoma; CI, confidence interval; ECC, extrahepatic cholangiocarcinoma; HCV, hepatitis C virus; ICC, intrahepatic cholangiocarcinoma;NS, not specified.

Table 5. Cirrhosis as a Potential Risk Factor for Cholangiocarcinoma

First Author	Country	Study Dates	Study Design	Risk Factor	CC Type	Cases (% With Risk Factor)	Controls (Case-Control Study)/ Individuals at Risk (Cohort Study)	Risk Estimate (95% CI)	Selected Adjusted Variables
Sorensen[52]	Denmark	1977-1993	Cohort	Cirrhosis	NS	21	11,605	10.0 (6.2-15.2)	
Yamamoto[51]	Japan	1991-2002	Case-control	Cirrhosis	ICC	50 (10%)	205 (1%)	5.03 (.045-56.82)	Age, sex
Shaib[47]	United States	1993-1999	Case-control	^Cirrhosis	ICC	625 (8.5%)	90,834 (0.4%)	27.2 (19.9-37.1)	Age, sex, race, geographic location
Welzel[28]	United States	1993-1999	Case-control	^Cirrhosis	ICC	535 (3.2%)	102,782 (0.3%)	10.0 (6.1-16.4)	Age, sex, race, geographic location
					ECC	549 (1.8%)	102,782 (0.3%)	5.4 (2.9-10.2)	
Lee[27]	Korea	2000-2004	Case-control	Cirrhosis	ICC	622 (7.8%)	2488 (0.4%)	13.6 (6.5-28.5)	Age, sex

Abbreviations: ^, diagnostic code; CC, cholangiocarcinoma; CI, confidence interval; ECC, extrahepatic cholangiocarcinoma; ICC, intrahepatic cholangiocarcinoma; NS, not specified.

up period of 6 years and reported a 10-fold increased risk of CC among patients with cirrhosis, compared with the expected cancer cases in the general population (standardized incidence ratio of 21 versus 2).[52] A hospital-based, case-control study in Italy by Donato et al. compared 26 ICC cases with 824 controls. Both HCV and HBV seropositivity were analyzed, but only HCV was significantly associated with ICC.[42]

U.S. Studies. Several U.S. studies have shown an association between the presence of HCV and/or cirrhosis and increased risk of ICC. From the M.D. Anderson Cancer Center (The University of Texas, Houston, TX), a hospital-based, case-control study by Shaib et al. compared 83 patients with ICC and 163 with ECC to 236 controls. HCV was a significant risk factor for ICC. Cirrhosis was not analyzed as a separate variable, but 80% of HCV-positive patients had cirrhosis. For ECC, neither HCV nor HBV status was a significant risk factor.[53] A large, population-based, case-control study by Shaib et al. of Medicare-enrolled patients compared 625 cases of ICC with 90,834 controls. In a multivariate analysis, HCV was significantly associated with ICC. It was unclear whether patients with HCV also had a recorded diagnostic code for cirrhosis. However, nonspecific cirrhosis was strongly associated with ICC. The prevalence of HBV infection was similar in cases and controls.[47] A similar population-based, case-control study by Welzel et al. of Medicare-enrolled patients examined risk factors for both ICC and ECC. There were 549 cases of ECC and 535 cases of ICC, compared with 102,782 controls. Significant risk factors for ICC included HCV and nonspecific cirrhosis. Regarding ECC, nonspecific cirrhosis was also a risk factor, but HCV infection was not significant.[28] A large cohort study of U.S. veterans by El-Serag et al. examined the association between HCV and both ICC and ECC in a cohort of 146,394 HCV-infected veterans and 572,293 uninfected controls. The risk for ICC in the HCV-infected cohort, though low at 4 per 100,000 person-years, was more than double that in the controls. The risk of ECC did not differ between the HCV-infected and uninfected veterans.[54]

The association of these risk factors with CC is not entirely clear, as studies have differing conclusions, and there is a paucity of population-based or prospective cohort studies. In countries such as Korea and Thailand, where both HBV and CC are endemic, data show HBV, but not HCV, as a risk factor for ICC. On the other hand, countries such as Japan and Western nations, including the United States, where HCV is more prevalent, were more likely to show an association between HCV and ICC.[27,55]

Diabetes and Obesity

Diabetes and obesity have been examined as possible risk factors for CC. Most studies presented in this section were previously discussed in the section on viral hepatitis and cirrhosis (Table 6).

The two SEER-Medicare studies showed a significant positive association between diabetes and CC.[28,47] Another large, population-based, case-control study from the United Kingdom by Grainge et al. found a significant association between diabetes and CC.[56] Conversely, a population-based study by Welzel conducted in Denmark did not find a significant association between diabetes and ICC.[48] Additionally, one hospital-based, case-control study showed a significant association between diabetes and ICC,[27] whereas at least three others failed to show a signification association (Table 6).[41,51,53] The data on diabetes as a risk

Table 6. Obesity and Diabetes as Potential Risk Factors for Cholangiocarcinoma

First Author	Country	Study Dates	Study Design	Risk Factor	CC Type	Cases (% With Risk Factor)	Controls (% With Risk Factor)	Risk Estimate (95% CI)	Selected Adjusted Variables
Welzel[48]	Denmark	1978-1991	Case-control	^Diabetes	ICC	764 (1.96%)	3056 (1.41%)	1.43 (0.8-2.6)	Age, sex
				^Obesity	ICC	764 (0.79%)	3056 (0.39%)	2.05 (0.7-5.6)	
Grainge[56]	United Kingdom	1987-2002	Case-control	^Diabetes	NS	372 (9.4%)	5760 (5.9%)	1.48 (1.0-2.2)	Age, sex, practice group
				Obesity (BMI $\geq$30)	NS	372 (19.6%)	5760 (15.7%)	1.52 (1.0-2.2)	
Yamamoto[51]	Japan	1991-2002	Case-control	Diabetes	ICC	50 (22%)	205 (12%)	1.95 (0.6-5.8)	Age, sex
Shaib[53]	United States	1992-2002	Case-control	Diabetes	ICC	83 (14.5%)	236 (8.5%)	Not calculated	Age, sex, race
				Diabetes	ECC	163 (11.7%)	236 (8.5%)	Not calculated	
Shaib[47]	United States	1993-1999	Case-control	^Diabetes	ICC	625 (26.4%)	90,834 (15.6%)	2.0 (1.6-2.4)	Age, sex, race, geographic location
Welzel[28]	United States	1993-1999	Case-control	^Diabetes	ICC	535 (33.1%)	102,782 (22.1%)	1.8 (1.5-2.1)	Age, sex, race, geographic location
				^Diabetes	ECC	549 (30.1%)	102,782 (22.1%)	1.5 (1.3-1.8)	
				^Obesity	ICC	535(4.3%)	102,782 (3.1%)	1.7 (1.1-2.6)	
				^Obesity	ECC	549 (2.9%)	102,782 (3.1%)	1.1 (0.7-1.8)	
Lee[27]	Korea	2000-2004	Case-control	Diabetes	ICC	622 (15.4%)	2,488 (5.6%)	3.2 (2.3-4.3)	Age, sex
Zhou[41]	China	2004-2006	Case-control	Diabetes	ICC	312 (4.2%)	438 (2.5%)	1.5 (0.6-3.8)	Age, sex, date of admission

Abbreviations: ^, diagnostic code; BMI, body mass index; CC, cholangiocarcinoma; CI, confidence interval; ECC, extrahepatic cholangiocarcinoma; ICC, intrahepatic cholangiocarcinoma.

factor for CC, especially ICC, are mostly indicative of a modest association, but are inconsistent.

Data on obesity are limited (Table 6). Obesity was reported as a significant, but weak, risk factor for CC in two population-based, case-control studies. In the study by Grainge et al., a body mass index $\geq$30 was significantly associated with CC, type not specified.[56] The U.S. study by Welzel et al. reported a significant association between obesity and ICC, but not between obesity and ECC.[28] However, in the Danish, population-based study by Welzel et al., there was no significant association between obesity and ICC.[48] The data available on obesity are too limited to make any conclusions.

Alcohol Drinking

Several cohort studies, population- and hospital-based, case-control studies, have reported a strong association between heavy alcohol use, typically >80 g/day, and CC (Table 7). A cohort study by Sorensen et al. that examined 11,605 patients with cirrhosis found a significantly increased CC risk in individuals with alcoholic cirrhosis.[52] The two SEER-Medicare studies also found alcoholic liver disease to be significantly associated with CC (both ICC and ECC).[28,47] However, the population-based, case-control study by Grainge et al. did not find alcohol use to be a risk factor for CC.[56] Few hospital-based, case-control studies have shown a significant association between alcohol intake and CC,[17,27,53] whereas others have not.[41,42,51] Based on the strong magnitude of association (risk estimate range from 2 to 15) and studies with different designs, heavy alcohol use is likely to be a risk factor for CC.

Smoking

Data on smoking are not consistent (Table 7). Three large, population-based, case-control studies found smoking to be weakly, but significantly, associated with CC, with risk estimates from 1.38 to 1.8. For these studies, the frequency and/or duration of smoking was not quantified. In several hospital-based, case-control studies, there was no significant association between smoking and CC.[17,27,41,51,53] Some of these studies quantified smoking, but there was no consistency among studies in terms of smoking frequency or duration. Smoking may be a weak risk factor for CC, but, given the conflicting data, a firm conclusion cannot be made.

Genetic Polymorphisms

Host genetic factors, either alone or interacting with environmental factors, have been examined as possible risk factors for CC. Genes coding for enzymes responsible for metabolism of carcinogens, DNA repair, and inflammation have been examined for polymorphic

Table 7. Alcohol Drinking and Tobacco Smoking as Potential Risk Factors for Cholangiocarcinoma

First Author	Country	Study Dates	Study Design	Risk Factor	CC Type	Cases (% With Risk Factor)	Controls (% With Risk Factor)	Risk Estimate (95% CI)	Selected Adjusted Variables
Sorensen[52]	Denmark	1977-1993	Cohort	Alcoholic cirrhosis	NS	17	11,605	15.3 (8.9-24.5)	
Grainge[56]	United Kingdom	1987-2002	Case-control	Smoking (current exposure)	NS	372 (27.5%)	5760 (20.9%)	1.38 (1.0-1.9)	Age, sex, practice group
				^Alcohol (problem drinker)	NS	372 (0.3%)	5760 (0.7%)	Not calculated	
Shin[17]	Korea	1990-1993	Case-control	Heavy smoking (>1 pack/day, >10 years)	NS	41 (36.6%)	406 (46.8%)	0.8 (0.2-2.5)	Age, sex, socioeconomic status
				Heavy alcohol (>80 g/day, >10 years)	NS	41 (22%)	406 (11.1%)	4.6 (1.4-15.2)	
Yamamoto[51]	Japan	1991-2002	Case-control	Smoking (any previous exposure)	ICC	50 (34%)	205 (44%)	Not calculated	Age, sex
				Heavy alcohol (>5 g sake/day, >10 years)	ICC	50 (2%)	205 (5%)	0.97 (0.5-1.9)	
Shaib[53]	United States	1992-2002	Case-control	Smoking (>25 pack years)	ICC	83 (24.1%)	236 (15.7%)	Not calculated	Age, sex, race
				Smoking (>25 pack years)	ECC	163 (20.9%)	236 (15.7%)	Not calculated	
				Heavy alcohol (>80 g/day)	ICC	83 (21.7%)	236 (3.8%)	5.9 (2.1-17.4)	
				Heavy alcohol (>80 g/day)	ECC	163 (17.8%)	236 (3.8%)	3.6 (1.5-9.4)	
				Mild/moderate alcohol (80 g/day)	ICC	83 (33.7%)	236 (48.3%)	Not calculated	
				Mild/moderate alcohol (80 g/day)	ECC	163 (26.9%)	236 (48.3%)	0.5 (0.3-0.8)	
Shaib[47]	United States	1993-1999	Case-control	^Smoking	ICC	625 (3.8%)	90,834 (2.1%)	1.8 (1.2-2.70)	Age, sex, race, geographic location
				^Alcoholic liver disease	ICC	625 (2.2%)	90,834 (0.3%)	7.4 (4.3-12.8)	
Welzel[28]	United States	1993-1999	Case-control	^Smoking	ICC	535 (2.2%)	102,782 (1.2%)	1.8 (1.0-3.2)	Age, sex, race, geographic location
				^Smoking	ECC	549 (2.2%)	102,782 (1.2%)	1.7 (1.0-3.0)	
				^Alcoholic liver disease	ICC	535(0.9%)	102.782 (0.3%)	3.1 (1.3-7.5)	
				^Alcoholic liver disease	ECC	549 (1.5%)	102,782 (0.3%)	4.5 (2.2-9.1)	
Donato[42]	Italy	1995-2000	Case-control	Heavy alcohol (>80 g/day)	ICC	26 (23.1%)	824 (33%)	0.4 (0.2-1.6)	Age, sex, residence
Lee[27]	Korea	2000-2004	Case-control	Smoking (any prior exposure)	ICC	622 (47.1%)	2,488 (45.6%)	Not calculated	Age, sex
				Heavy alcohol (>80g/day)	ICC	622 (18%)	2,488 (3.1%)	6.6 (4.8-9.2)	
Zhou[41]	China	2004-2006	Case-control	Smoking (≥4 day/week, ≥6 months)	ICC	312 (13.8%)	438 (15.3%)	1.23 (0.7-2.2)	Age, sex, date of admission
				Alcohol (≥1 drink/week, ≥ 6 months)	ICC	312 (12.5%)	438 (9.4%)	0.80 (0.5-1.3)	

Abbreviations: ^, diagnostic code; CC, cholangiocarcinoma; CI, confidence interval; ECC, extrahepatic cholangiocarcinoma; ICC, intrahepatic cholangiocarcinoma; NS, not specified.

Table 8. Genetic Polymorphisms as Potential Risk Factors of Cholangiocarcinoma

First author	Country	Polymorphism	CC Type	Cases	Controls	Risk Estimate (95% CI)	Selected Adjusted Variables
Ko[59]	Korea	MTHFR 677CC + TSER 2R+	NS	47	204	5.38 (1.2-23.6)	
Marahatta[60]	Thailand	GSTO1*A140D	NS	30	98	8.5 (2.1-37.8)	Sex, race
Hoblinger[57]	Germany	MRP2/ABCC2 variant c.3972C>T	ICC/ECC	60	73	1.83 (1.1-3.1)	
Melum[61]	Scandinavia	NKG2D rs11053781 + PSC	NS	49	368	2.08 (1.3-3.3)	Sex
		NKG2D rs2617167 + PSC	NS	49	368	2.32 (1.5-3.7)	
		MICA 5.1 + PSC	NS	49	368	0.43 (0.2-0.9)	
Prawan[62]	Thailand	CYP1A2*1A/*1A†	NS	216	233	0.28 (0.1-0.9)	Age, sex
		NAT2* 13,*6B,*7A	NS	216	233	~0.23-0.38 (0.1-0.9)	
Huang[58]	China	XRCCI 194W	NS	127	786	1.9 (1.1-3.5)	Age, sex, hospital
		XRCC1 R280H	NS	127	786	0.5 (0.3-0.9)	
Sakoda[63]	China	PTGS2 Ex 10 +837 C	NS	127	786	1.8 (1.2-2.7)	Age, sex, hospital

†In males. Range based on allelic combinations.

Abbreviations: MTHFR, 5,10-methylenetetrahydrofolate reductase is a key enzyme in folate metabolism and provides methyl groups for DNA methylation; TS, thymidylate synthase is a rate-limiting enzyme in the synthesis of dTMP and DNA repair; GST, glutathione S-transferases are a family of detoxification enzymes; MRP2/ABC2, multidrug resistance-associated protein 2 is one of the ATP-binding cassette transporters that is involved in biliary clearance of endogenous and exogenous toxic compounds; NKG2D, natural killer cell receptor is involved in activation of natural killer (NK) cells, which are important for tumor surveillance; MICA, major histocompatibility complex class I chain-related molecule A is a ligand to NKG2D; CYP1A2, member of cytochrome P450 involved in activation of carcinogens; NAT, *N*-acetyltransferases are involved in the metabolism and detoxification of amines; BER, base excision repair corrects DNA damage caused by oxidative stress, which encompass the X-ray repair cross-complementing group 1 (XRCC1); PTGS2, prostaglandin-endoperoxide synthase 2 is induced by inflammation; NS, not specified.

variants that may be associated with increased susceptibility to CC. In several hospital-based, case-control studies, different gene polymorphisms have been associated with increased, as well as decreased, risk of developing CC (Table 8).[57-63] Given the varying study populations and lack of study replication in independent cohorts, it is difficult to draw firm conclusions regarding these findings.

The Possible Effect of CC Classification on Risk Factor Epidemiology

A significant limitation to exploring risk factors of CC resides in the classification systems that have been used. (1) Most cancer registries combine CC with other hepatobiliary malignancies; therefore it is unclear whether CC also includes HCC and gallbladder cancer.[6,8] (2) When ICC and ECC are reported separately, sometimes HCC is included with ICC and gallbladder cancer is included with ECC.[6,8] (3) Misclassification of Klatskin tumor as ICC has been shown to result in an overestimation of the incidence of ICC and an underestimation of ECC.[10] (4) Most CC studies do not distinguish site (e.g., ductal, hilar, and peripheral) or histology. Specific risk factors for different types of CC are, therefore, likely to be missed, depending on the distribution of these types in a given study. (5) In studies where the distinction between ICC and ECC was used, some potential risk factors seem to have a differential effect on CC, depending on the site. The consistent use of a more refined classification would allow a better understanding of risk factors for CC.

Summary

CC is a rare malignancy in Western countries, but is more common in Asia. This difference is mostly attributed to the higher prevalence of established risk factors, such as parasitic infections, bile-duct cysts, and hepatolithiasis. However, most cases of CC are not associated with established risk, except in areas endemic for liver flukes. The established risk factors for CC include parasitic infections, biliary-duct cysts, hepatolithiasis, and PSC. Less-established risk factors include IBD, HCV, HBV, cirrhosis, obesity, diabetes, alcohol, smoking, and genetic polymorphisms. There are not enough consistent data to support that IBD independent of PSC, obesity, smoking, or specific genetic polymorphisms confer an increased risk for CC. Available data suggest that diabetes and heavy alcohol drinking may confer an increased risk for CC. The data also suggest that in Western countries, HCV is consistently associated with ICC and not ECC. In Asian countries, it appears that HBV may be associated with ICC. Cirrhosis is the most consistently illustrated risk factor for ICC, but not ECC. The lack of an accurate, consistent CC classification system may have hindered the conduct and interpretation of risk factors in epidemiological studies.

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