

Table S1 Clinical characteristics of cholangiocarcinoma patients in this study

Patient	Gender	Age	Diagnosis	HBsAg	HBcAb	AFP (ng/ml)	CEA (ng/ml)	CA125 (U/ml)	CA19-9 (U/ml)
1	F	74	iCCA	N	N	2.6	70.2	825.6	300.1
2	F	71	iCCA	N	N	0.8	58.8	137.7	1000
3	F	48	iCCA	N	P	1.8	3.3	30.15	99.57
4	F	61	iCCA	N	N	4	181.1	207.7	1000
5	M	72	iCCA	N	P	2.2	1.4	14.65	339.9
6	F	58	iCCA	N	P	4.44	121.6	1011	58449
7	M	75	iCCA	N	P	2.5	4.4	47.09	46.58
8	M	60	iCCA	N	P	2.7	44.8	77.06	0.6
9	F	53	iCCA	N	N	2.6	49.3	36.78	1000
10	M	64	iCCA	N	N	5.59	3.58	15.74	4142
11	M	51	iCCA	N	N	3.19	3.93	42.43	48.53
12	F	71	iCCA	N	N	1.6	48	869	6.81
13	M	69	iCCA	N	P	2.06	17.52	687.1	23228
14	M	63	iCCA	N	P	62.6	175.5	12.63	151
15	F	47	iCCA	N	N	7.1	6.3	80.77	126.4
16	F	68	iCCA	N	P	5.5	1.5	526.7	127.8
17	M	59	dCCA	N	P	NA	NA	NA	31.98
18	M	52	pCCA	N	N	2.2	0.9	28	335.2
19	M	76	iCCA	N	N	2.65	4.38	10.71	656.1
20	M	65	pCCA	N	P	2.2	6.4	18.7	1000
21	M	52	pCCA	N	P	4.5	3.3	27.97	952.3
22	M	73	dCCA	N	P	5.6	4.7	NA	72.93
23	M	40	pCCA	N	P	26.3	1.7	81.18	1000
24	M	64	dCCA	N	P	4.1	5.3	NA	1000
25	F	62	dCCA	N	P	4.7	2.3	81.02	1000
26	M	67	iCCA	N	P	209.9	48	68.07	1000
27	F	38	iCCA	N	P	26.5	47.1	11.68	12.48
28	F	66	iCCA	N	P	3.4	3.5	10.32	102.1
29	F	53	iCCA	P	P	1.8	1.3	22.65	192.2
30	M	69	iCCA	P	P	1.4	2.2	15.56	20.89
31	M	64	iCCA	P	P	95.49	3.74	61.07	29925
32	M	55	iCCA	P	P	1.7	2.2	278.2	17.19
33	F	49	iCCA	P	P	NA	NA	22.57	451.6
34	F	72	dCCA	N	P	1.8	2.5	6.06	152.6
35	F	76	pCCA	N	P	3.9	3	14.08	19.18
36	F	35	iCCA	P	P	2.3	1.1	12.21	30.08
37	M	61	pCCA	N	N	1.04	14.29	222.89	387.31
38	M	74	dCCA	N	P	1.77	3.73	80.11	2889
39	F	48	iCCA	P	P	769.1	1.9	20.48	61.09
40	M	79	pCCA	N	P	1.7	4.6	12.31	13.56
41	M	65	iCCA	P	P	6.4	3.8	12.57	56.59
42	F	70	iCCA	N	N	4.7	2.2	5.06	26.43
43	F	61	iCCA	N	N	5.4	2.9	9.2	33.32
44	M	65	pCCA	N	P	2.9	5.2	11.55	44.78
45	F	45	iCCA	N	N	1.76	80.87	25.39	16108
46	F	60	iCCA	N	N	4	93.8	830.6	1000
47	M	66	dCCA	N	P	2.4	5.3	78.13	844.2
48	M	55	dCCA	N	N	4	12.1	12.82	16.75
49	F	67	dCCA	P	P	13.6	14.9	182.27	234.8
50	M	80	dCCA	N	P	5.1	12.9	65.3	1000
51	M	80	dCCA	N	P	2.97	4.24	15.09	807.5
52	M	51	pCCA	N	P	5.97	7.76	38.01	3394
53	M	71	pCCA	N	P	2.6	3.5	60	765.4
54	F	52	iCCA	N	N	20.1	21.9	244.1	9.47
55	F	64	pCCA	N	N	1.9	13.7	17.29	25.48
56	M	66	iCCA	N	N	2.8	10.3	280.2	312.2
57	M	70	dCCA	N	N	3.43	39.64	406.6	1000
58	F	34	dCCA	N	P	3.94	3.96	58.51	175.2
59	M	76	dCCA	N	N	2.36	5.28	9.57	35.14
60	M	54	pCCA	P	P	3.32	7.36	15.02	473.9
61	M	58	pCCA	N	N	2.21	4.73	15.38	59.38
62	M	47	iCCA	P	P	18.8	16.2	79.7	70.38
63	M	58	iCCA	P	P	12.6	4.9	NA	26.96
64	F	51	iCCA	N	N	13.9	1.4	11.76	58.22
65	M	65	iCCA	P	P	8.5	2.3	8.11	27.12
66	M	63	iCCA	P	P	4.3	0.5	9.14	14.29
67	F	50	iCCA	N	P	0.7	2.4	23.58	22.07

F, female; M, male; iCCA, intrahepatic cholangiocarcinoma; pCCA, perihilar cholangiocarcinoma; dCCA, distal cholangiocarcinoma; HBsAg, Hepatitis B Surface Antigen; HBcAb, Hepatitis B Core Antibody; P, Positive; N, Negative; NA, not available.

Table S2 Clinical characteristics of hepatocellular carcinoma patients in this study

Patient	Gender	Age	Diagnosis	HBsAg	HBcAb	AFP (ng/ml)	CEA (ng/ml)	CA125 (U/ml)	CA19-9 (U/ml)
1	M	62	HCC	N	P	126.6	2.5	32.51	40.8
2	F	54	HCC	N	P	>1210	1.9	12.58	14.18
3	M	60	HCC	P	P	27.4	2.7	6.46	31.39
4	M	31	HCC	P	P	>1210	1.2	26.6	17.42
5	M	49	HCC	P	P	>1210	4.4	54.96	146.7
6	M	59	HCC	N	P	71.5	3.8	13.9	11.21
7	M	62	HCC	P	P	4.5	5.7	16.5	10.88
8	M	63	HCC	P	P	>1210	3.3	13.66	19.34
9	M	70	HCC	P	P	84.91	3.69	12.44	67.66
10	F	49	HCC	P	P	>1210	1.7	13.12	25.82
11	M	75	HCC	P	P	1.1	2.9	22.61	29.71
12	F	52	HCC	P	P	26.3	0.9	25.77	52.85
13	M	39	HCC	P	P	2.2	1.2	8.45	12.56
14	M	50	HCC	P	P	39.1	1.3	311.1	23.36
15	M	50	HCC	N	N	10.5	1.9	3.75	33.69
16	F	69	HCC	P	P	9145	1.07	NA	27.46
17	M	50	HCC	P	P	>1210	2.6	73.68	11.49
18	M	54	HCC	P	P	423.1	2.4	74.54	32.85
19	F	69	HCC	N	N	7.85	6.71	NA	34.95
20	F	78	HCC	N	N	37.2	1.3	14.87	8.33
21	M	63	HCC	P	P	>1210	8.7	22.08	1.91
22	M	55	HCC	P	P	1.3	5.3	13.16	24.95
23	F	54	HCC	P	P	7.7	2.7	18.71	25.27
24	F	66	HCC	P	P	116.1	2.7	7.23	9.93
25	M	47	HCC	P	P	5	3.4	6.49	21.09
26	M	60	HCC	P	P	8.5	5.1	12.94	97.4
27	M	31	HCC	P	P	>1210	2	6.02	8.55
28	M	61	HCC	P	P	>1210	4.5	9.12	29.5
29	M	66	HCC	N	P	>1210	3.4	20.95	18.35
30	M	52	HCC	P	P	>1210	4.3	21.86	12.6
31	M	73	HCC	P	P	1136	1.8	7.22	8.93
32	F	67	HCC	P	P	403.8	2.75	9.3	33.93
33	M	82	HCC	P	P	3.6	3.4	10.43	24.97
34	M	54	HCC	N	P	2.3	0.8	8.8	7.56
35	M	56	HCC	P	P	1210	2.7	19.61	5.43
36	M	53	HCC	N	P	6.11	2.22	7.09	6.21
37	M	79	HCC	P	P	249.5	1.22	9	7.94
38	M	54	HCC	N	N	1210	3	78.92	17.61
39	M	77	HCC	P	P	277.4	4.4	7.65	36.71
40	M	66	HCC	P	P	3	2.3	15.54	11.32
41	F	67	HCC	N	P	2.77	1.91	6.33	19.68
42	F	64	HCC	P	P	47.8	4	15.43	30.78
43	M	46	HCC	P	P	139.41	5.55	3.51	86.39
44	M	48	HCC	P	P	398.5	4.5	18.82	17.93
45	M	51	HCC	P	P	1210	0.9	16.96	5.17
46	M	56	HCC	P	P	1210	6	12.67	13.75
47	M	50	HCC	P	P	1188	4.6	8.04	59.26
48	F	50	HCC	P	P	0.97	1.54	9.07	25.67
49	M	67	HCC	N	P	3.6	2.2	5.33	9.21
50	M	55	HCC	P	P	1065	1.61	14.64	11.2
51	M	55	HCC	P	P	1.9	2.6	12.47	14.84
52	F	50	HCC	P	P	23.81	3.09	11.55	6.5
53	M	44	HCC	P	P	1210	2	34.23	18.38
54	M	55	HCC	P	P	1210	2.6	6.17	20.24
55	M	69	HCC	N	P	3.2	2.2	23.47	20.34
56	M	73	HCC	P	P	1.5	3.5	21.4	15.43
57	M	50	HCC	P	P	79.5	2.5	82.77	21.51
58	F	46	HCC	P	P	8	2.7	16.75	0.77
59	M	66	HCC	P	P	4.3	1.5	10.22	28.54
60	M	67	HCC	P	P	1210	3.8	44.65	51.02
61	M	77	HCC	N	P	632.9	2.2	26.17	9.72
62	M	77	HCC	N	N	77	2.8	NA	6.52
63	M	48	HCC	P	P	1210	2.3	22.46	7.08
64	M	80	HCC	N	P	1	5.23	15.59	26.65
65	M	55	HCC	P	P	4.5	7.1	10.09	14.99
66	M	58	HCC	P	P	12.9	4.5	10.6	24.76
67	M	51	HCC	P	P	125.2	1.3	4.95	0.6
68	M	54	HCC	P	P	5.3	2.4	13	21.04
69	M	65	HCC	N	P	1.17	2.32	5.84	6.5
70	M	82	HCC	P	P	5.14	1.45	45.3	4.78
71	M	61	HCC	N	N	3.3	1.9	20.59	10.23
72	M	52	HCC	N	P	14.7	1.6	NA	6.47
73	F	64	HCC	N	P	13.8	1.6	20.75	6.84
74	M	63	HCC	N	P	3.9	3.8	8.89	15.77
75	M	58	HCC	N	P	2.2	6.3	39.18	2.87
76	M	55	HCC	N	P	955.1	2.1	27.42	71.58
77	M	62	HCC	N	P	0.8	1	12.44	13.2
78	M	51	HCC	P	P	11.3	3.7	14.32	12.86
79	M	52	HCC	P	P	70.5	2.9	11.46	45.32
80	M	37	HCC	P	P	3.2	2.4	58.21	18.08
81	M	67	HCC	P	P	1301	2.54	6.66	16.96
82	M	68	HCC	P	P	7.8	1.6	10.69	4.03
83	M	47	HCC	P	P	2.9	1.8	10.88	17.17

F, female; M, male; HCC, hepatocellular carcinoma; HBsAg, Hepatitis B Surface Antigen; HBcAb, Hepatitis B Core Antibody; P, Positive; N, Negative; NA, not available.

Table S3 Clinical characteristics of benign patients in this study

Patient	Gender	Age	Diagnosis	HBsAg	HBcAb	AFP (ng/ml)	CEA (ng/ml)	CA125 (U/ml)	CA19-9 (U/ml)
1	M	48	Hepatic hemangioma	N	P	3.4	3.7	7.84	1.67
2	F	53	Hepatic hemangioma	N	P	2.5	2.1	14.81	15.57
3	F	50	Hepatic hemangioma	N	N	3	0.5	15.58	10.15
4	M	51	Hepatic hemangioma	N	P	4.3	2.2	4.74	6.58
5	F	48	Hepatic hemangioma	N	N	0.96	0.81	16.57	5.8
6	F	53	Hepatic hemangioma	N	N	3.3	2.2	4.03	4.28
7	F	32	Hepatic hemangioma	N	N	1.3	1.3	12.33	3.55
8	F	53	Hepatic hemangioma	N	N	2.7	0.5	9.22	5.51
9	F	40	Hepatic hemangioma	N	N	1.65	0.49	9.98	6.19
10	F	56	Hepatic hemangioma	N	P	5.4	2.3	19.29	29.94
11	F	54	Hepatic hemangioma	N	N	1.4	2	13.56	25.91
12	F	72	Hepatic hemangioma	N	P	2.82	0.85	11.92	6.99
13	M	60	Hepatic hemangioma	N	P	0.97	3.59	9.73	205.3

Patient	Gender	Age	Diagnosis	HBsAg	HBcAb	AFP (ng/ml)	CEA (ng/ml)	CA125 (U/ml)	CA19-9 (U/ml)
1	M	67	Intrahepatic stone	P	P	2.9	4.2	10.26	171
2	M	66	Intrahepatic stone	N	P	2.7	3.6	7.8	25.4
3	F	51	Intrahepatic stone	P	P	1.96	3.59	9.46	26.77
4	F	52	Intrahepatic stone	N	N	3.1	3.5	5.53	30.34
5	F	66	Intrahepatic stone	N	N	3.7	2.2	7.6	17.15
6	F	64	Intrahepatic stone	N	P	3.7	2.6	130.6	94.31
7	F	48	Intrahepatic stone	N	P	1.4	3.2	27.64	136.8
8	F	63	Intrahepatic stone	N	P	1.6	5.5	47.82	1000
9	F	63	Intrahepatic stone	N	N	3.2	3.98	9.8	806
10	F	64	Intrahepatic stone	N	P	7.4	7.4	267.5	349.2
11	F	56	Intrahepatic stone	N	N	1.8	6	10.4	56.26
12	F	60	Intrahepatic stone	N	N	2.64	1.06	33.62	106.7
13	F	58	Intrahepatic stone	N	N	26.16	1.01	9.24	7.27
14	M	77	Intrahepatic stone	N	N	3.3	4.12	83.85	295.3
15	F	48	Intrahepatic stone	N	P	3.54	0.74	8.81	4.4
16	F	50	Intrahepatic stone	N	N	4.22	1.1	13.96	9.18
17	F	69	Intrahepatic stone	N	N	2.7	3.76	15.5	800.6
18	F	76	Intrahepatic stone	N	N	3.86	3.48	15.14	22.46
19	F	52	Intrahepatic stone	N	P	3.81	3.49	134.6	1000
20	F	64	Intrahepatic stone	N	N	1.6	1	3.15	21.86

F, female; M, male; HBsAg, Hepatitis B Surface Antigen; HBcAb, Hepatitis B Core Antibody; P, Positive; N, Negative.

Table S4 Clinical characteristics of chronic hepatitis B patients and healthy individuals in this study

Chronic Hepatitis B	Gender	Age	HBsAg	HBcAb	CA19-9 (U/ml)
1	M	55	P	P	8.4
2	M	32	P	P	18.42
3	M	35	P	P	17.69
4	M	42	P	P	32.66
5	M	46	P	P	26.95
6	M	28	P	P	17.93
7	M	29	P	P	18.27
8	F	34	P	P	15.85
9	F	51	P	P	22.09
10	F	55	P	P	22.35
11	M	27	P	P	19.46
12	F	22	P	P	29.85
13	F	38	P	P	7.2
14	M	34	P	P	2.91
15	F	38	P	P	12.95
16	M	45	P	P	9.94
17	M	42	P	P	9.85
18	M	26	P	P	10.94
19	M	31	P	P	22.84
20	M	24	P	P	6.35
21	M	48	P	P	18.95
22	M	26	P	P	13.85
23	F	29	P	P	1.4
24	M	32	P	P	15.4
25	M	46	P	P	25.35
26	F	42	P	P	18.82
27	F	48	P	P	12.91
28	M	41	P	P	25.36
29	F	27	P	P	19.98
30	M	44	P	P	13.71
31	M	41	P	P	15.17
32	M	40	P	P	27.93
33	M	29	P	P	18.56
34	M	30	P	P	19.71
35	F	37	P	P	16.17
36	F	41	P	P	15.75
37	F	25	P	P	14.84
38	M	38	P	P	26.77
39	F	32	P	P	29.43
40	F	29	P	P	28.39
41	M	27	P	P	20.08
42	M	24	P	P	28.93
43	M	46	P	P	19.21
44	F	44	P	P	14.93
45	M	36	P	P	7.5

Healthy individuals	Gender	Age	HBsAg	HBcAb	CA19-9 (U/ml)
1	M	70	N	N	30.8
2	F	51	N	N	15.18
3	M	38	N	N	24.39
4	M	42	N	N	18.56
5	F	49	N	N	24.7
6	F	49	N	N	20.12
7	F	45	N	N	17.88
8	M	54	N	N	19.82
9	M	44	N	N	22.68
10	M	54	N	N	6.6
11	F	45	N	N	6.5
12	F	61	N	N	18.9
13	F	68	N	N	15.71
14	F	82	N	N	26.89
15	M	76	N	N	19.91
16	M	54	N	N	25.95
17	M	61	N	N	9.4
18	F	50	N	N	15.85
19	F	56	N	N	25.95
20	M	69	N	N	10.93
21	F	60	N	N	15.85
22	M	68	N	N	22.09
23	M	49	N	N	25.55
24	F	55	N	N	19.56
25	F	70	N	N	18.93
26	F	55	N	N	15.43
27	M	47	N	N	7.21
28	M	53	N	N	29.94
29	M	64	N	N	5.61
30	F	46	N	N	8.2
31	F	52	N	N	17.75
32	M	62	N	N	30.75
33	M	75	N	N	25.93
34	F	57	N	N	12.78
35	F	81	N	N	18.43
36	F	53	N	N	30.17
37	M	86	N	N	24.84
38	F	61	N	N	25.75
39	F	61	N	N	15.2
40	M	67	N	N	10.72
41	F	64	N	N	28.56
42	M	51	N	N	28.99
43	M	71	N	N	19.23
44	M	64	N	N	30.76
45	M	53	N	N	16.52

F, female; M, male; HBsAg, Hepatitis B Surface Antigen; HBcAb, Hepatitis B Core Antibody; P, Positive; N, Negative.

Table S5 Primer of gene corresponding to each biomarker for PCR

Gene	Forward	Reverse	Amplicon
PI15-1	5'GGCATTGTGCAGAGCAAAGTA	5'TATTGGTTGGCGGGGATGAG	160bp
PI15-2	5'GGCATTGTGCAGAGCAAAGTA	5'AGGTCCATGGTCCCAAATGC	396bp
AFP	5'AGGGAGCGGCTGACATTATT	5'GCAGCATTTCTCCAACAGGC	329bp
CEACAM5	5'AATAACGGGACCTATGCCTGTTT	5'CCCAACCAGCACTCCAAT	151bp
MUC16	5'GCCTCTACCTAACGGTTACAATGAA	5'GGTACCCCATGGCTGTTGTG	114bp
KLK3	5'GAGGAGTTCTTGACCCCAA	5'CAGAATCACCCGAGCAGGT	154bp
GH1	5'GGCGCCTCTGACAGCAACGTCT	5'GCACGATGCGCAGGAATGTCTC	232bp
β -actin	5'TTGCCGACAGGATGCAGAA	5'GCCGATCCACACGGAGTACTT	101bp

PI15-1 and PI15-2 are the two primers corresponding to the gene PI15. PI15, peptidase inhibitor 15; AFP, alpha fetoprotein; CEACAM5, carcinoembryonic antigen related cell adhesion molecule 5; MUC16, mucin 16; KLK3, kallikrein related peptidase 3; GH1, growth hormone 1.