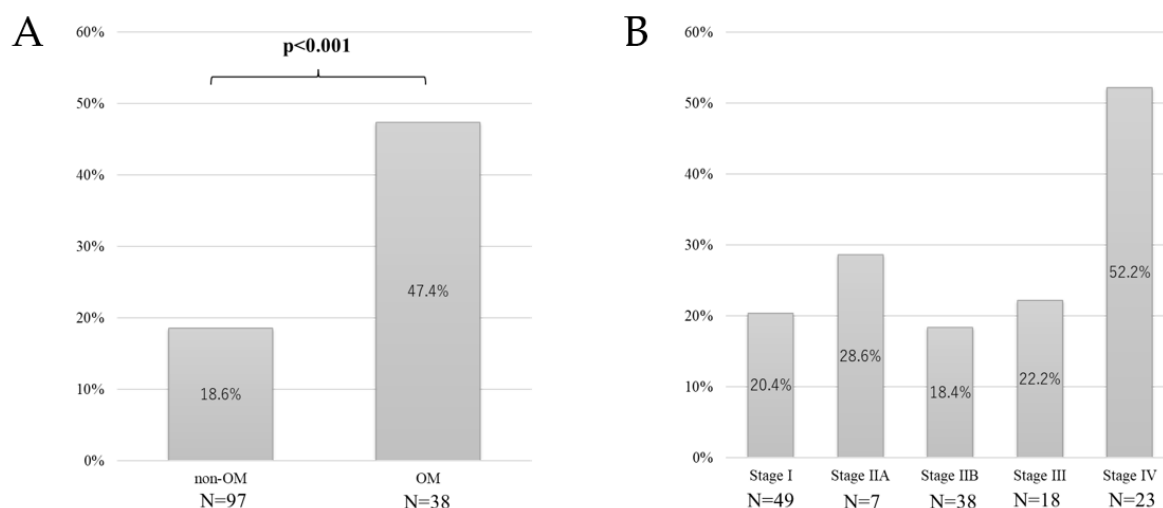
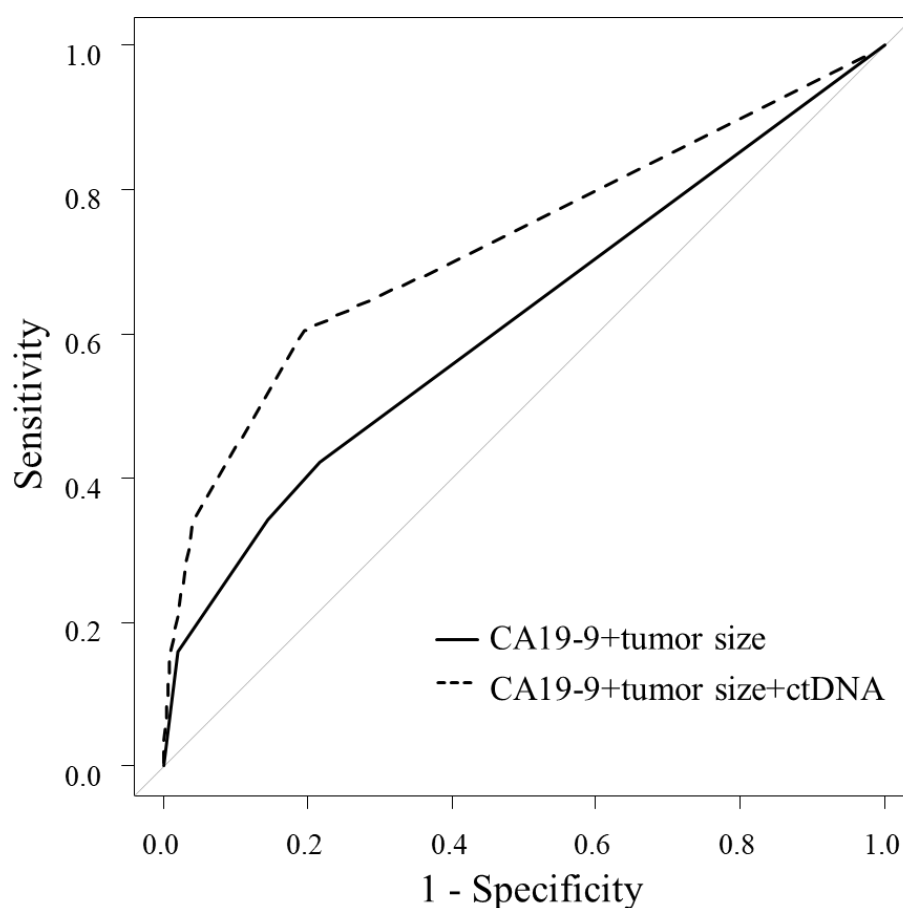




Supplementary Figure 1 ctDNA. A: ctDNA positivity rate by occult metastasis and non-occult metastasis group; B: ctDNA positivity rate by pathological staging.



Supplementary Figure 2 Prediction accuracy of occult metastases using ROC curves. Adding the ctDNA status to the CA19-9 levels and tumor size significantly increased

the area under the curve from 0.617 to 0.724 ($p = 0.013$), improving the accuracy of the diagnosis of occult metastases.

Supplementary Table 1 Gene list of Oncomine Pan-Cancer Cell-Free assay

Gene list of Oncomine Pan-Cancer Cell-Free assay									
<i>AKT1</i>	<i>ALK</i>	<i>AR</i>	<i>ARAF</i>	<i>BRAF</i>	<i>CHEK</i>	<i>CTNN</i>	<i>DDR2</i>	<i>EGFR</i>	<i>ERBB</i>
					2	B1			2
<i>ERBB</i>	<i>ESR1</i>	<i>FGFR</i>	<i>FGFR</i>	<i>FGFR</i>	<i>FGFR</i>	<i>FLT3</i>	<i>GNA1</i>	<i>GNA</i>	<i>GNAS</i>
3		1	2	3	4		1	Q	
<i>HRAS</i>	<i>IDH1</i>	<i>IDH2</i>	<i>KIT</i>	<i>KRAS</i>	<i>MAP2</i>	<i>MAP2</i>	<i>MET</i>	<i>MTO</i>	<i>NRAS</i>
					K1	K2		R	
<i>NTRK</i>	<i>NTRK</i>	<i>PDGF</i>	<i>PIK3C</i>	<i>RAF1</i>	<i>RET</i>	<i>ROS1</i>	<i>SF3B1</i>	<i>SMA</i>	<i>SMO</i>
1	3	RA	A					D4	
<i>ERG</i>	<i>ETV1</i>	<i>CCN</i>	<i>CCN</i>	<i>CCN</i>	<i>CDK4</i>	<i>CDK6</i>	<i>MYC</i>	<i>APC</i>	<i>FBXW</i>
		D1	D2	D3					7
<i>PTEN</i>	<i>TP53</i>								

Supplementary Table 2 Metastatic sites of occult metastases

Metastatic sites	Overall cohort (N = 135)	ctDNA positive (N = 35)	ctDNA negative (N = 100)
Stage IV	23	12	11
Not resected	10	5	5
Liver	3	2	1
Peritoneum	2	1	1
Peritoneum + Peritoneal cytology positive	4	2	2
Para-aortic lymph node	1	0	1
Resected	13	7	6
Peritoneum + Peritoneal cytology positive	1	1	0
Para-aortic lymph node	7	4	3
Peritoneal cytology positive	5	2	3

Recurrence within 6 months	15	6	9
Liver	9	4	6
Peritoneum	1	0	1
Lung	1	0	1
Local	2	0	1
Liver + Peritoneum	1	1	0
Lung + Local	1	1	0

Supplementary Table 3 ctDNA detected before surgery

I D	Group	Sta ge	Mutat ion type	Locus	Gene	Mutat ion ID	Molec ular covera ge	Alle le cou nt	MAF (%)
5	After NAT	IIB	SNV	chr15:6643 6839	MAP2 K1	<i>p.F129 L</i>	8,009	11	0.14%
6	After NAT	IB	SNV	chr17:7577 105	TP53	<i>p.P278 R</i>	2,850	2	0.07%
8	After NAT	IA	SNV	chr7:14045 3146	BRAF	<i>p.L597 V</i>	2,581	3	0.12%
14	After NAT	IIB	SNV	chr20:5748 4596	GNAS	<i>p.Q227 R</i>	5,718	4	0.07%
14	After NAT	IIB	SNV	chr17:7577 126	TP53	<i>p.E271 G</i>	2,259	2	0.09%
14	After NAT	IIB	SNV	chr17:7577 105	TP53	<i>p.P278 L</i>	2,259	4	0.18%
14	After NAT	IIB	SNV	chr17:7579 884	TP53	<i>p.V10 A</i>	6,131	11	0.18%
21	After NAT	IIB	SNV	chr12:2538 0275	KRAS	<i>p.Q61 H</i>	2,247	3	0.13%

22	Upfront surgery	IA	SNV	chr7:11642 3475	<i>MET</i>	<i>p.M12 68I</i>	3,154	3	0.10%
24	After NAT	IB	SNV	chr17:7577 138	<i>TP53</i>	<i>p.R267 Q</i>	2,539	3	0.12%
35	After NAT	IIB	SNV	chr17:7578 518	<i>TP53</i>	<i>p.A138 R</i>	3,815	3	0.08%
37	After NAT	IV	SNV	chr7:55249 092	<i>EGFR</i>	<i>p.C797 Y</i>	2,650	2	0.08%
37	After NAT	IV	SNV	chr6:15233 2832	<i>ESR1</i>	<i>p.E380 Ter</i>	1,091	2	0.18%
37	After NAT	IV	SNV	chr17:7577 559	<i>TP53</i>	<i>p.S241 C</i>	3,696	4	0.11%
39	After NAT	III	SNV	chr2:19826 6834	<i>SF3B1</i>	<i>p.K700 E</i>	4,661	4	0.09%
41	Upfront surgery	IB	SNV	chr12:2539 8284	<i>KRAS</i>	<i>p.G12 V</i>	2,683	2	0.07%
41	Upfront surgery	IB	SNV	chr17:7578 263	<i>TP53</i>	<i>p.R196 *</i>	3,235	3	0.09%
55	After NAT	IV	SNV	chr3:17891 7478	<i>PIK3C A</i>	<i>p.G118 D</i>	5,295	4	0.08%
56	Upfront surgery	IB	SNV	chr17:7578 517	<i>TP53</i>	<i>p.A138 V</i>	2,149	2	0.09%

56	Upfront surgery	IB	SNV	chr17:7577 561	TP53	p.S240 R	2,209	3	0.14%
57	Upfront surgery	III	SNV	chr12:2539 8281	KRAS	p.G13 D	2,553	2	0.08%
59	After NAT	IV	SNV	chr20:5748 4420	GNAS	p.R201 C	5,072	7	0.14%
63	After NAT	IB	SNV	chr7:14045 3145	BRAF	p.L597 R	4,653	4	0.09%
63	Upfront surgery	IB	SNV	chr4:18035 64	FGFR 3	p.R248 C	3,305	3	0.09%
63	Upfront surgery	IB	SNV	chr17:7577 096	TP53	p.D281 G	2,187	2	0.09%
64	After NAT	IIB	SNV	chrX:6694 3600	AR	p.E894 K	2,929	2	0.07%
65	After NAT	IIB	SNV	chr17:7578 265	TP53	p.I195 T	5,213	4	0.08%
79	After NAT	IA	SNV	chr17:7578 457	TP53	p.R158 H	6,914	5	0.07%
80	Upfront surgery	IA	SNV	chr17:7578 461	TP53	p.V157 I	1,831	3	0.16%

81	After NAT	IIB	SNV	chr17:7576 897	TP53	p.Q317 *	4,303	4	0.09%
86	After NAT	IV	SNV	chr17:7578 454	TP53	p.A159 G	4,428	3	0.07%
87	After NAT	IIA	SNV	chr17:7577 144	TP53	p.L265 Q	3,195	4	0.13%
88	After NAT	IV	SNV	chr3:17893 6074	PIK3C A	p.P539 R	3,217	3	0.09%
88	After NAT	IV	SNV	chr18:4859 1918	SMA D4	p.R361 C	3,912	4	0.10%
96	Upfron t surger y	IIA	SNV	chr7:55249 071	EGFR	p.T790 G	2,479	2	0.08%
96	Upfron t surger y	IIA	SNV	chr7:11641 2022	MET	p.Y102 1D	3,763	3	0.08%
10 2	Upfron t surger y	III	SNV	chr17:7578 412	TP53	p.V173 A	5,577	5	0.09%
10 6	After NAT	IV	SNV	chr12:2539 8284	KRAS	p.G12 D	2,471	2	0.08%
10 6	After NAT	IV	SNV	chr17:7578 260	TP53	p.V197 M	2,415	2	0.08%
10 9	After NAT	IV	SNV	chr12:2538 0275	KRAS	p.Q61 H	3,005	81	2.70%
11 5	After NAT	IV	SNV	chr12:2539 8284	KRAS	p.G12 D	4,462	4	0.09%

11	After	IV	SNV	chr17:3786	<i>ERBB</i>	<i>p.E321</i>	5,130	8	0.16%
7	NAT			8241	2	<i>G</i>			
11	After	IV	SNV	chr4:55593	<i>KIT</i>	<i>p.N564</i>	2,273	5	0.22%
7	NAT			624		<i>D</i>			
11	After	IV	SNV	chr12:2539	<i>KRAS</i>	<i>p.G12</i>	4,397	28	0.64%
7	NAT			8284		<i>V</i>			
11	After	IV	SNV	chr17:7576	<i>TP53</i>	<i>p.E326</i>	4,456	7	0.16%
7	NAT			869		<i>G</i>			
11	After	IV	SNV	chr17:7577	<i>TP53</i>	<i>p.D281</i>	2,314	5	0.22%
7	NAT			096		<i>G</i>			
11	After	III	SNV	chr19:4117	<i>MAP2</i>	<i>p.Q60</i>	5,146	4	0.08%
8	NAT			541	<i>K2</i>	<i>L</i>			
11	After	III	SNV	chr7:11641	<i>MET</i>	<i>p.Y102</i>	3,070	6	0.20%
8	NAT			2022		<i>1H</i>			
11	After	III	SNV	chr17:7577	<i>TP53</i>	<i>p.G245</i>	4,102	6	0.15%
8	NAT			548		<i>S</i>			
11	After	III	SNV	chr17:7577	<i>TP53</i>	<i>p.S240</i>	4,102	6	0.15%
8	NAT			563		<i>G</i>			
12	After	IV	SNV	chr10:8969	<i>PTEN</i>	<i>p.D92</i>	2,839	2	0.07%
5	NAT			2792		<i>E</i>			
12	After	IV	SNV	chr17:7577	<i>TP53</i>	<i>p.R267</i>	2,549	2	0.08%
5	NAT			138		<i>Q</i>			
12	Upfront	IV	SNV	chr12:2539	<i>KRAS</i>	<i>p.G12</i>	4,349	52	1.20%
6	t			8284		<i>D</i>			
	surger								
	y								
12	Upfront	IV	SNV	chr17:7578	<i>TP53</i>	<i>p.Q192</i>	4,850	34	0.70%
6	t			275		*			
	surger								
	y								

126	Upfront surgery	IV	SNV	chr18:48591919	SMA D4	p.R361 H	4,227	10	0.24%
126	Upfront surgery	IV	SNV	chr17:7577557	TP53	p.C242 S	3,791	4	0.11%
130	After NAT	IA	SNV	chr12:25398284	KRAS	p.G12 D	3,645	3	0.08%
133	After NAT	IV	SNV	chr7:55259485	EGFR	p.P848 L	2,616	2	0.08%
133	After NAT	IV	SNV	chr18:48575151	SMA D4	p.A118 V	4,248	3	0.07%

Supplementary Table 4 ctDNA detected in subgroup with samples collected before and after neoadjuvant therapy

I D	Group	Sta ge	Mutati on type	Locus	Gen e	Mutati on ID	Molec ular covera ge	Allel e coun t	MAF (%)
10	Before NAT	IV	SNV	chr17:7577091	TP5 3	p.R283 C	2,711	3	0.11%
17	Before NAT	IIB	SNV	chr7:55242475	EGF R	p.E749 Q	4,530	5	0.11%
19	Before NAT	IIB	SNV	chr7:140453137	BR AF	p.V600 L	4,426	3	0.07%
24	After NAT	IB	SNV	chr17:7577138	TP5 3	p.R267 Q	2,539	3	0.12%
35	After NAT	IIB	SNV	chr17:7578518	TP5 3	p.A138 R	3,815	3	0.08%

28	Before NAT	IA	SNV	chr17:7578 508	TP5 3	p.C141 Y	4,553	3	0.07%
39	Before NAT	III	SNV	chr2:19826 6834	SF3 B1	p.K700 E	3,950	12	0.30%
39	After NAT	III	SNV	chr2:19826 6834	SF3 B1	p.K700 E	4,661	4	0.09%
42	Before NAT	IB	SNV	chr17:7578 532	TP5 3	p.M133 T	1,969	8	0.41%
50	Before NAT	IA	SNV	chr12:2539	KR	p.G12V	2,060	3	0.15%
			SNV	8284	AS	p.C634	2,656	3	0.11%
				chr10:4360 9949	RET	Y			
58	Before NAT	IB	SNV	chr2:19826 6834	SF3 B1	p.K700 E	1,777	9	0.51%
76	Before NAT	IIA	SNV	chr12:2539	KR	p.G12V	3,817	3	0.08%
			SNV	8284	AS	p.Y220	3,229	3	0.09%
				chr17:7578 190	TP5 3	C			
79	Before NAT	IA	SNV	chr12:2539	KR	p.G12C	4,688	31	0.66%
			SNV	8285	AS	p.Y103	3,150	14	0.44%
			SNV	chr17:7579 379	TP5 3	S p.R273	2166	11	0.51%
				chr17:7577 120	TP5 3	H			
79	After NAT	IA	SNV	chr17:7578 457	TP5 3	p.R158 H	6,914	5	0.07%
85	Before NAT	IA	MNV	chr7:14048 1402	BR AF	p.G469 L	2,025	2	0.10%
10	Before	IB	SNV	chr7:14045	BR	p.N581	1,729	3	0.17%
9	NAT			3193	AF	S			

11	Before	III	SNV	chr12:2539	KR	<i>p.G12V</i>	4,951	4	0.08%
4	NAT			8284	AS				
13	Before	IA	SNV	chr7:14045	BR	<i>p.K601</i>	3,895	3	0.08%
2	NAT			3134	AF	<i>E</i>			