ESMO GOOD SCIENCE BETTER MEDICINE BEST PRACTICE

ESMO ANNALS OF ONCOLOGY DRIVING INNOVATION IN ONCOLOGY

ORIGINAL ARTICLE

Survival benefit of adjuvant chemotherapy based on molecular residual disease detection in resected colorectal liver metastases: subgroup analysis from CIRCULATE-Japan GALAXY☆

K. Kataoka[1], K. Mori[2], Y. Nakamura[3,4,5], J. Watanabe[6,7], N. Akazawa[8], K. Hirata[9], M. Yokota[10], K. Kato[11], M. Kotaka[12], K. Yamazaki[13], Y. Kagawa[14,15], S. Mishima[3], K. Ando[16], M. Miyo[17], H. Yukami[18], G. Laliotis[19], S. Sharma[19], C. C. Palsuledesai[19], M. Rabinowitz[19], A. Jurdi[19], M. C. Liu[19], A. Aleshin[19], D. Kotani[3], H. Bando[3], H. Taniguchi[20], I. Takemasa[17], T. Kato[21], T. Yoshino[3,22,23] & E. Oki[16]*

[1]Division of Lower GI Surgery, Department of Gastroenterological Surgery, Hyogo Medical University, Nishinomiya; [2]Department of Biostatistics, Clinical Research Center, Shizuoka Cancer Center, Sunto-gun; [3]Department of Gastrointestinal Oncology, National Cancer Center Hospital East, Kashiwa; [4]Translational Research Support Office, National Cancer Center Hospital East, Kashiwa; [5]International Research Promotion Office, National Cancer Center Hospital East, Kashiwa; [6]Department of Colorectal Surgery, Kansai Medical University, Hirakata; [7]Department of Surgery, Gastroenterological Center, Yokohama City University Medical Center, Yokohama; [8]Department of Gastroenterological Surgery, Sendai City Medical Center Sendai Open Hospital, Sendai; [9]Department of Surgery 1, School of Medicine, University of Occupational and Environmental Health, Kitakyushu; [10]Department of General Surgery, Kurashiki Central Hospital, Kurashiki; [11]Department of Surgery, Teine-Keijinkai Hospital, Sapporo; [12]Gastrointestinal Cancer Center, Sano Hospital, Kobe; [13]Division of Gastrointestinal Oncology, Shizuoka Cancer Center, Sunto-gun; [14]Department of Gastroenterological Surgery, Osaka International Cancer Institute, Osaka; [15]Department of Gastroenterological Surgery, Osaka General Medical Center, Osaka; [16]Department of Surgery and Science, Graduate School of Medical Sciences, Kyushu University, Fukuoka; [17]Department of Surgery, Surgical Oncology and Science, Sapporo Medical University, Sapporo; [18]Cancer Chemotherapy Center, Osaka Medical and Pharmaceutical University, Takatsuki, Japan; [19]Natera, Inc., Austin, USA; [20]Department of Clinical Oncology, Aichi Cancer Center Hospital, Nagoya; [21]Department of Surgery, NHO Osaka National Hospital, Osaka; [22]Department of Gastroenterological Surgery/Pediatric Surgery, Graduate School of Medicine, Gifu University, Gifu; [23]Kindai University Faculty of Medicine, Higashiosaka City, Japan

Available online 16 September 2024

Background: The prognostic role of circulating tumor DNA (ctDNA)-based molecular residual disease (MRD) detection and its utility for postsurgical risk stratification has been reported in colorectal cancer. In this study, we explored the use of ctDNA-based MRD detection in patients with colorectal liver metastases (CLM), for whom the survival benefit of adjuvant chemotherapy (ACT) after surgical resection remains unclear.

Methods: Patients with CLM without extrahepatic disease from the GALAXY study (UMIN000039205) were included. The disease-free survival (DFS) benefit of ACT was evaluated in MRD-positive and -negative groups after adjusting for age, gender, number, and size of liver metastases, *RAS* status, and previous history of oxaliplatin for primary cancer. ctDNA was detected using a personalized, tumor-informed 16-plex polymerase chain reaction-next-generation sequencing (mPCR-NGS) assay. ctDNA-based MRD status was evaluated 2-10 weeks after curative surgery, before the start of ACT.

Results: Among 6061 patients registered in GALAXY, 190 surgically resected CLM patients without any preoperative chemotherapy were included with a median follow-up of 24 months (1-48 months). ctDNA positivity in the MRD window was 32.1% (61/190). ACT was administered to 25.1% (48/190) of patients. In the MRD-positive group, 24-month DFS was higher for patients treated with ACT [33.3% versus not reached, adjusted hazard ratio (HR): 0.07, $P < 0.0001$]; whereas no benefit of ACT was seen in the MRD-negative group (24-month DFS: 72.3% versus 62.2%, adjusted HR: 0.68, $P = 0.371$). Multivariate analysis showed that the size of liver metastases (HR: 3.94, $P = 0.031$) was prognostic of DFS in the MRD-positive group. In the MRD-negative group, however, none of the clinicopathological factors were prognostic of DFS.

Conclusions: Our data suggest that ACT may offer notable clinical benefits in MRD-positive patients with CLM. MRD status-based risk stratification could be potentially incorporated in future clinical trials for CLM.

Key words: colorectal liver metastases, adjuvant chemotherapy, prognostic biomarker, disease-free survival, circulating tumor DNA

*Correspondence to: Prof. Eiji Oki, Department of Surgery and Science, Graduate School of Medical Sciences, Kyushu University, 3-1-1, Maidashi, Higashi-ku, Fukuoka City, 812-8582, Japan. Tel: +81-092-642-5462

E-mail: oki.eiji.857@m.kyushu-u.ac.jp (E. Oki).

☆Note: Previous presentation: Abstract accepted for presentation at ESMO 2024.

INTRODUCTION

Colorectal liver metastases (CLM) are observed in approximately one-third of all patients with colorectal cancer (CRC).[1,2] Although surgical resection can be curative, approximately two-thirds of patients with CLM experience recurrence despite hepatectomy.[3]

The benefit of adjuvant chemotherapy (ACT) for patients with CLM remains controversial. Several studies evaluating the efficacy of ACT in resected CLM patients showed some benefit of ACT in improving disease-free survival (DFS) compared with surgery alone, however, statistically significant overall survival (OS) benefits were not consistently observed.[4-9] For example, the phase III JCOG0603 trial, which aimed to confirm the superiority of mFOLFOX6 after liver resection in patients with CLM, showed that while adjuvant mFOLFOX6 conferred a DFS benefit, OS was almost identical between mFOLFOX6 and observation groups.[10] Therefore, new biomarkers to identify a subgroup of patients who are likely to benefit from ACT are needed.

Several groups have recently reported the utility of circulating tumor DNA (ctDNA) as a sensitive and specific biomarker for detection of minimal/molecular residual disease (MRD) and predictor of recurrent risk across several solid tumor types.[11-16] Particularly in CRC, several prospective studies have demonstrated the prognostic significance of ctDNA-positivity. Among them, the GALAXY study within the CIRCULATE-Japan platform trial, the largest ctDNA observational cohort in CRC, demonstrated in an interim analysis that ctDNA positivity was strongly associated with DFS across all stages in resectable CRC.[17] Additionally, high-risk stage II and stage III CRC patients with postsurgical MRD positivity were observed to benefit significantly from ACT, while ctDNA-negative patients did not. Although several recent reports demonstrate ctDNA to be a strong biomarker in surgically resected CLM as well,[18-25] the association between MRD status and the benefit of ACT has never been assessed in this setting. In this study, the benefit of ACT based on MRD status was assessed using data collected in patients with CLM as part of the GALAXY study of CIRCULATE-Japan.

METHODS

GALAXY study design and patient selection

The GALAXY study is a prospective large-scale nationwide registry to monitor ctDNA status for patients with clinical stage II-IV CRC who planned curative surgical resection. It serves to screen patients for assignment to one of the two randomized ctDNA-guided interventional phase III trials within the CIRCULATE-Japan platform: ALTAIR (treatment escalation) and VEGA (treatment de-escalation).[17] The study design, eligibility criteria, and endpoints have been previously published.[17] Blood samples were collected at 4, 12, 24, 36, 48, 72, and 96 weeks after surgery until recurrence. Computed tomography (CT) imaging was carried out every 6 months after surgery. All participants provided written informed consent. This study was approved by the institutional review board of the National Cancer Center Japan and authorized by the head of each participating institution. The study is registered in the Japan Registry of Clinical Trials (UMIN000039205) and was conducted in accordance with the Declaration of Helsinki.

In this retrospective subset analysis, patients with surgically resected CLM from the GALAXY study with ctDNA status available between 2 to 10 weeks after curative-intent hepatectomy were included. Patients with extrahepatic metastases, those treated with neoadjuvant therapy, and those enrolled in associated interventional phase II/III trials in the CIRCULATE-Japan platform (ALTAIR, VEGA, and FANTASTIC) were excluded from the analysis.[26,27]

Tumor-informed ctDNA testing

A clinically validated, personalized, tumor-informed 16-plex polymerase chain reaction (mPCR)-next-generation sequencing (NGS) assay (Signatera™; Natera, Inc., Austin, Texas, USA) was used for the detection and quantification of ctDNA in blood samples as previously described.[17] Briefly, formalin-fixed, paraffin-embedded tumor tissue from surgical resection or biopsy samples and matched normal DNA extracted from peripheral blood samples were processed for whole-exome sequencing to identify and track up to 16 patient- and tumor-specific somatic single nucleotide variants in the associated patient's plasma using a multiplex PCR-based NGS approach.[13] Cell-free DNA was extracted from patient plasma (median 9.6 ml, range 3.0-12.2 ml) at a given time point and was used to detect ctDNA. Plasma samples with at least two tumor-specific variants detected above a pre-defined threshold were defined as ctDNA positive. ctDNA concentration was reported as mean tumor molecules/ml of plasma.

Statistical analyses

In this ancillary analysis, benefit of ACT on DFS was assessed according to MRD status in patients with resected CLM. DFS was defined as the time between the date of landmark and the date of diagnosis with recurrence or death due to any cause or the latest radiological assessment. Recurrence was determined based on diagnostic imaging or any other diagnostic procedure if imaging was not confirmative (e.g. colonoscopy to diagnose local recurrence). The chi-square test was used to compare categorical variables, while Fisher's exact test was used to compare the proportion of DFS or OS in specified times during patient follow-up. To control for multiple hypothesis testing, we applied the Bonferroni correction to the *P* values obtained from the chi-square and Fisher's exact tests conducted in our study. Survival analyses were carried out using R software v4.4.0 using packages survival, survminer and coxphf. The Kaplan—Meier method was used to estimate the survival distribution. Differences between the groups were tested using the log-rank test. A multivariable Cox proportional hazards model was used to assess prognostic factors associated with DFS (coxph and cox.zph). Major hepatectomy was defined as the resection of at least three Couinaud hepatic

segments, whereas minor hepatectomy was defined as the resection of fewer than three such segments. Presence of post-operative complication was defined as Clavien—Dindo II or greater.

The MRD window was defined as 2-10 weeks after surgery before the start of any adjuvant therapy; MRD analyses were landmarked at the date of MRD time point to account for immortal time bias. The surveillance window was defined as the time from 4 weeks after ACT or the end of the MRD window if the patient had no ACT, until the last follow-up or recurrence. The surveillance analyses were landmarked at 10 weeks after surgery. Regarding analysis of ctDNA clearance, Cox regression was used to compare cumulative incidence function differences between the ACT and observation groups, was landmarked at 2 months after surgery, and was adjusted for age (<70 or >70 years), sex, *RAS* status (wild versus mutant), size (<50 mm versus ≥ 50 mm), and number of liver metastases (1 versus ≥ 2), history of oxaliplatin, and synchronicity (synchronous versus metachronous). ctDNA clearance at 3- or 6-months analyses in ACT-treated patients were landmarked from the dates of blood collection at 3 month and 6 month time points, respectively. Blood was collected for 3- and 6-month time points between 70-112 days and 160-200 days after surgery, respectively. P values < 0.05 were considered statistically significant.

RESULTS

Patient characteristics

Of 6061 patients enrolled in the GALAXY study between May 2020 and July 2024, 190 patients with CLM and no extrahepatic disease undergoing upfront curative-intent surgery (no preoperative chemotherapy) were included in this analysis (Figure 1). Median follow-up was 24.0 months (1-48 months). ctDNA was detected in 98.41% (186/189) of the patients with CLM before surgery. Patient characteristics along with ctDNA status during the MRD window are detailed in Table 1.

ctDNA status in MRD and surveillance windows is prognostic of survival outcomes

Of the 190 patients included in this study, 32.11% (61/190) were ctDNA-positive in the MRD window, i.e. MRD-positive. ctDNA detection rates were higher in patients with synchronous tumors (48.44%, 31/64) versus metachronous tumors (23.81%, 30/126; $P = 0.002$). Frequency of multiple liver metastases were significantly higher in the MRD-positive group than in the MRD-negative group (Table 1). ACT for CLM was administered in 24.6% [15/61; 5-fluorouracil (5-FU) with oxaliplatin, $N = 13$, and oral 5-FU/capecitabine, $N = 2$] of the MRD-positive patients and 25.6% (33/129; 5-FU with oxaliplatin, $N = 28$; and oral 5-FU/capecitabine, $N = 5$) of the MRD-negative patients, respectively.

Biomarker status such as *RAS*/*BRAF*/MSI was similar between the two groups. Frequency of grade II or higher post-operative complications was numerically higher in the MRD-positive group than in the MRD-negative group. R0 resection was carried out in 183 patients and R1 resection in the rest of 7 patients.

Compared with MRD-negative patients, MRD-positive patients were almost six times more likely to recur

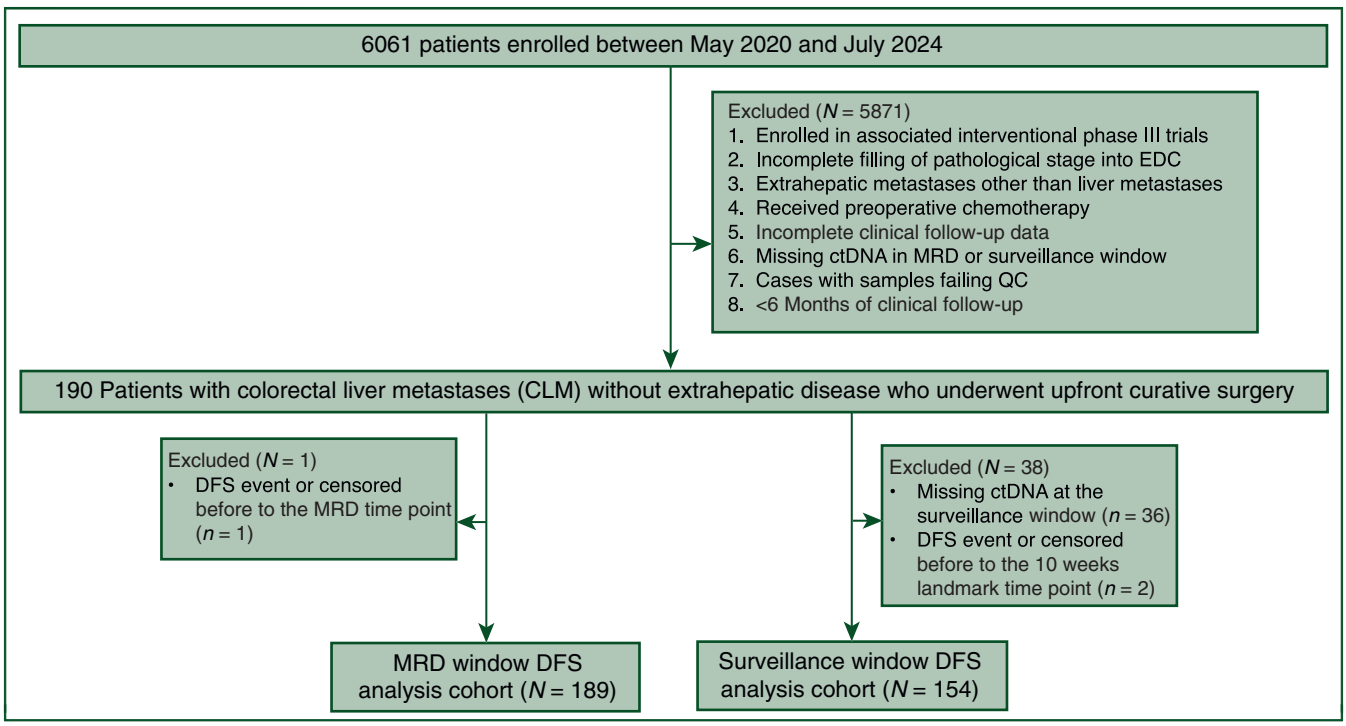

Figure 1. CONSORT diagram depicting the inclusion of patients in sub-analyses in this study.
ctDNA, circulating tumor DNA; DFS, disease-free survival; EDC, Electronic Data Capture; MRD, molecular residual disease.

Table 1. Patient characteristics

Characteristic	All patients	ctDNA at the MRD window		
	$N = 190$[a]	ctDNA (−), $N = 129$[a]	ctDNA (+), $N = 61$[a]	P value[b]
Age	68 (34-85)	69 (37-85)	68 (34-84)	>0.9
Sex				>0.9
Male	118 (62)	83 (64)	35 (57%)	
Female	72 (38)	46 (36)	26 (43%)	
Performance status				>0.9
0	186 (98)	127 (98)	59 (97%)	
1	4 (2.1)	2 (1.6)	2 (3.3%)	
Tumor location				0.6
Right-sided	50 (26)	40 (31)	10 (16%)	
Left-sided	140 (74)	89 (69)	51 (84%)	
Synchronicity				0.015
Synchronous	64 (34)	33 (26)	31 (51%)	
Metachronous	126 (66)	96 (74)	30 (49%)	
Hepatectomy				>0.9
Minor	159 (84)	112 (87)	47 (77%)	
Major	31 (16)	17 (13)	14 (23%)	
Number of liver metastasis				0.010
1	121 (64)	93 (72)	28 (46%)	
≥2	69 (36)	36 (28)	33 (54%)	
Size of liver metastasis (mm)				>0.9
<50	180 (95)	123 (95)	57 (93%)	
≥50	10 (5)	6 (5)	4 (7%)	
Pathological T stage				>0.9
T1-T2	23 (12)	16 (12)	7 (11%)	
T3-T4	167 (88)	113 (88)	54 (89%)	
Pathological N stage				0.3
N0	77 (41)	60 (47)	17 (28%)	
N1-N2	113 (59)	69 (53)	44 (72%)	
Post-operative treatment				>0.9
Chemotherapy	48 (25)	33 (26)	15 (25%)	
Observation	142 (75)	96 (74)	46 (75%)	
Prior L-OHP history	50 (26)	39 (30)	11 (18%)	>0.9
Post-operative complication	23 (12)	10 (7.8)	13 (21%)	0.2
BRAF				>0.9
$BRAF^{wt}$	188 (99)	127 (98)	61 (100%)	
$BRAF^{V600E}$	2 (1)	2 (1.6)	0 (0%)	
RAS				>0.9
RAS^{wt}	106 (56)	74 (57)	32 (52%)	
RAS^{mut}	84 (44)	55 (43)	29 (48%)	
MSI				>0.9
MSS	188 (99)	128 (99)	60 (98%)	
MSI-high	2 (1)	1 (0.8)	1 (1.6%)	

ctDNA, circulating tumor DNA; L-OHP, trans-/-diaminocyclohexane oxalatoplatinum; MSI, microsatellite instability; MSS, microsatellite stability.
[a]Median (range); *n* (%).
[b]Wilcoxon rank sum test; Pearson's Chi-square test; Fisher's exact test for proportions with the Bonferroni correction.

[hazard ratio (HR): 5.74, 95% confidence interval (CI) 3.81-8.64; $P < 0.0001$; 24-month DFS: 10.80%, 95% CI 4.45% to 20.3% for MRD-positive versus 64.50%, 95% CI 54.80% to 72.60% for MRD-negative patients, $P < 0.0001$; Figure 2A] and also exhibited significantly shorter OS (HR: 6.44, 95% CI 2.22-18.72; $P = 0.0006$; 24-month OS: 83.60%, 95% CI 67.40% to 92.1% versus 98.40%, 95% CI 93.80% to 99.60%, $P = 0.016$; Figure 2B). We further evaluated the prognostic value of ctDNA compared with other known clinicopathologic risk factors by performing a multivariate analysis. We observed that MRD positivity was the most significant prognostic factor associated with inferior DFS (HR: 7.32, 95% CI 4.46-12, $P < 0.001$, Figure 2C). The only other factor prognostic of poor DFS was larger (≥50 mm) liver metastases (HR: 2.44, 95% CI 1.02-5.9, $P = 0.046$; Figure 2C).

The multivariate analyses of DFS among MRD-positive and MRD-negative subgroups are depicted in Figure 3. The size of liver metastases (HR: 3.94, 95% CI 1.14-13.7 $P = 0.031$) and *RAS* mutation status (HR: 2.91, 95% CI 1.38-6.2, $P = 0.005$) were found to be prognostic of DFS in the MRD-positive group (Figure 3A). In the MRD-negative group, however, no clinicopathological factor was prognostic of DFS (Figure 3B).

During the surveillance window, ctDNA status was available for 154 patients, 35.71% (55/154) of whom were ctDNA-positive. ctDNA detection rates were higher in patients with synchronous tumors (48.08%, 25/52), versus metachronous tumors (29.41%, 30/102; $P = 0.07$). Compared with patients who remained serially ctDNA-negative in the surveillance window, those who were ctDNA-positive at any point had significantly inferior DFS (HR: 10.64, 95% CI 5.93-19.09; $P < 0.0001$; 24-month DFS: 8.59%, 95% CI 2.0% to 21.42% versus 81.50%, 95% CI 71.10% to 88.40%, $P < 0.0001$; Figure 2D) and OS (HR: 5.15, 95% CI 1.26-21.08; $P = 0.023$; 24-month OS: 95.5%, 95% CI 82.7% to 98.9% versus 99.0%, 95% CI 92.80% to 99.90%, $P = 0.368$; Figure 2E). Furthermore, the multivariate analysis indicated that ctDNA positivity during surveillance was an independent factor significantly associated with worse DFS (HR: 12.71, 95% CI 6.68-24.20, $P < 0.001$; Figure 2F), followed by multiple liver metastases (HR: 2.81, 95% CI 1.60-4.90, $P < 0.001$; Figure 2F).

ctDNA status and dynamics are predictive of ACT benefit in patients with resected CLM

We first examined whether ctDNA status in the MRD window is predictive of ACT benefit in postsurgical patients with CLM. A landmark at 2 months after surgery was implemented to address the immortal time bias, and HR was adjusted for the following confounding factors: age, sex, number and size of metastases, synchronicity, *RAS* mutational status, and prior oxaliplatin exposure. No statistically significant benefit from ACT was observed among patients who were MRD-negative [adjusted HR: 0.68, 95% CI 0.29-1.58, $P = 0.371$; recurrence rate: 24.24% (8/33) for ACT group versus 35.87% (33/92) for the observation group; Figure 4A], regardless of the synchronicity (Supplementary Figure S1, available at https://doi.org/10.1016/j.annonc.2024.08.2240) or number of liver metastases (Supplementary Figure S2, available at https://doi.org/10.1016/j.annonc.2024.08.2240). On the contrary, MRD-positive patients derived statistically significant benefit from ACT (adjusted HR: 0.07, 95% CI 0.02-0.26, $P < 0.0001$), with a recurrence rate of 66.67% (10/15) for the ACT group versus 93.55% (29/31) for the observation group (Figure 4B).

Next, we evaluated whether ctDNA clearance on ACT in MRD-positive patients was predictive of ACT benefit and

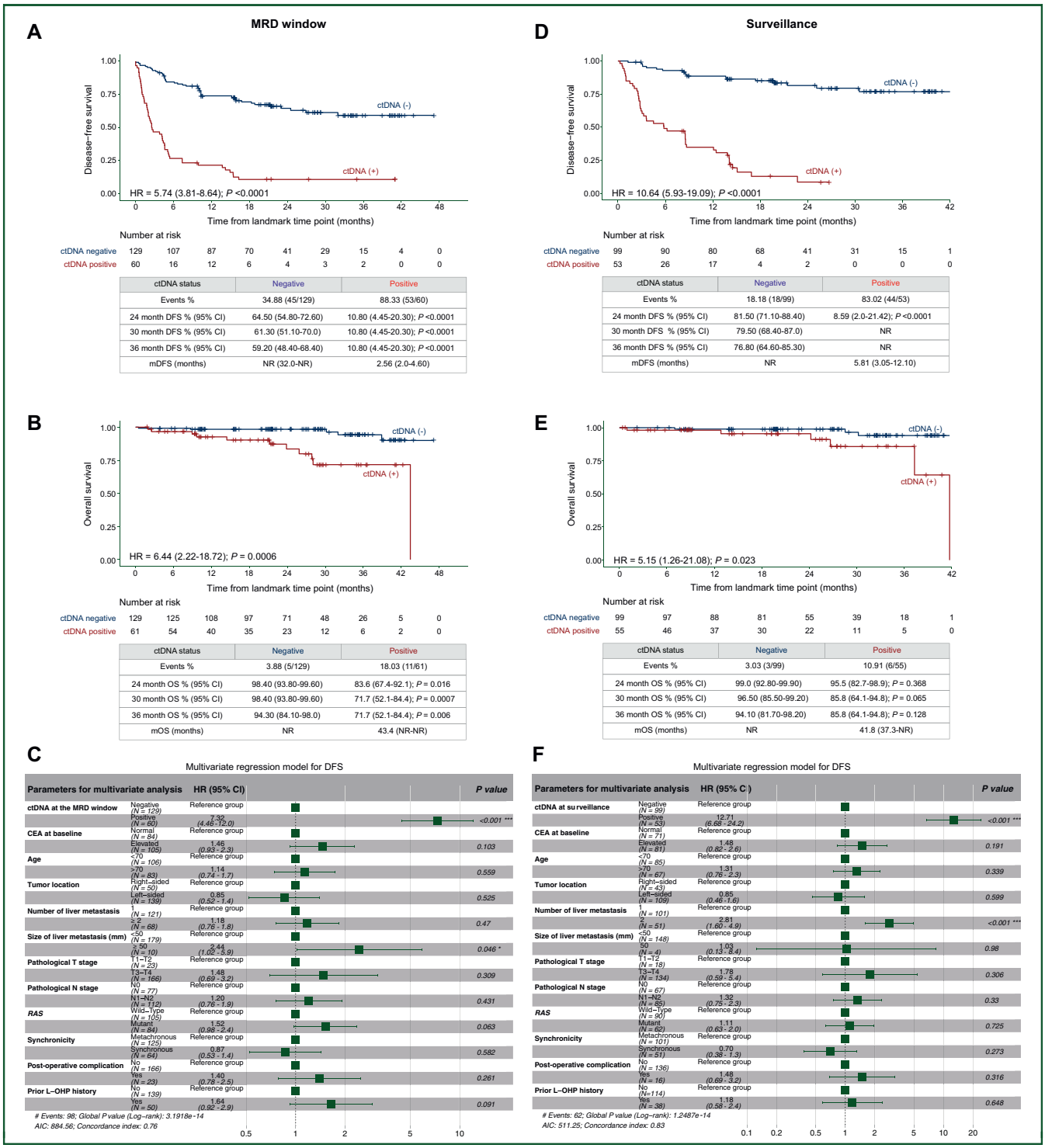

Figure 2. ctDNA status in the MRD and surveillance windows are predictive of survival outcomes in postsurgical patients CLM. Kaplan−Meier estimates for (A) DFS and (B) OS stratified by ctDNA status during the MRD window (MRD-negative versus MRD-positive). (C) Forest plot showing the multivariate analysis for DFS including ctDNA status during MRD window and other clinicopathologic factors. Kaplan−Meier estimates for (D) DFS and (E) OS stratified by ctDNA status during the surveillance window (negative versus positive). (F) Forest plot showing the multivariate analysis for DFS including ctDNA status during surveillance window and other clinicopathologic factors. The DFS and OS analyses in the MRD window were landmarked from the date of the MRD time point; analyses in the surveillance window were landmarked at 10 weeks after surgery. Median DFS/OS and percentage DFS and OS at 24, 30, and 36 months were estimated from the landmark time point; *P* value was calculated using Fisher's exact test for proportions with the Bonferroni correction.

CEA, carcinoembryonic antigen; CI, confidence interval; CLM, colorectal liver metastases; ctDNA, circulating tumor DNA; DFS, disease-free survival; HR, hazard ratio; L-OHP, trans-/-diaminocyclohexane oxalatoplatinum; MRD, molecular residual disease; NR, not reached; OS, overall survival.

* Indicates *P* value <0.05.

*** Indicates *P* value <0.001.

https://doi.org/10.1016/j.annonc.2024.08.2240

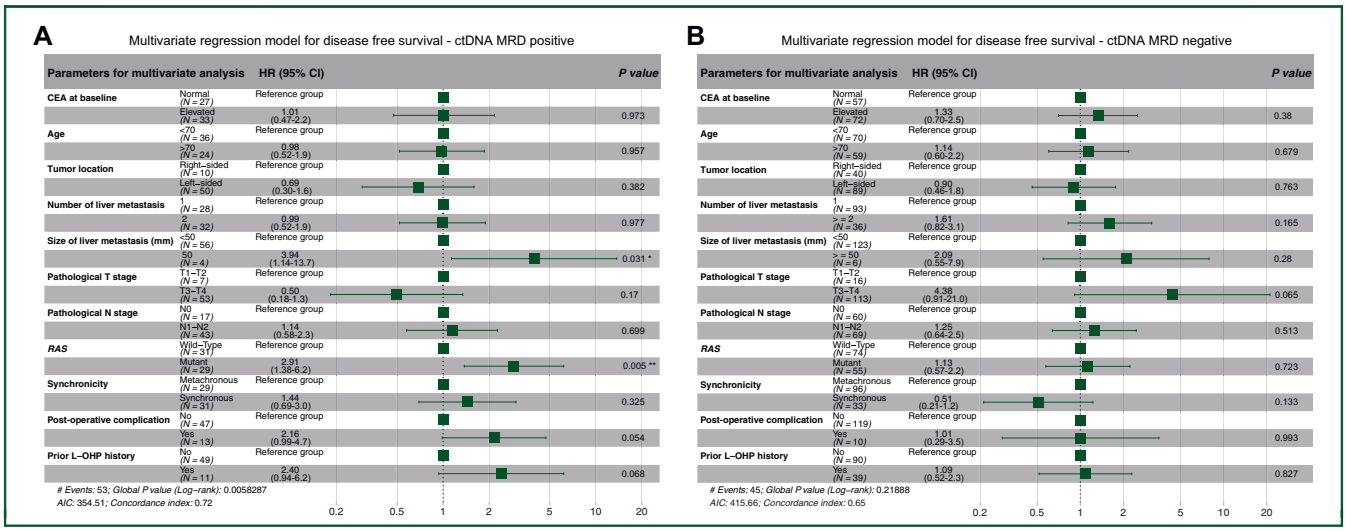

Figure 3. Forest plot showing the multivariate analysis for DFS postsurgical patients with CLM who were MRD-positive (A) and MRD-negative (B). These analyses were landmarked from the date of the MRD time point.
CEA, carcinoembryonic antigen; CI, confidence interval; CLM, colorectal liver metastases; ctDNA, circulating tumor DNA; MRD, molecular residual disease.
* Indicates *P* value <0.05.
** Indicates *P* value <0.001.

patient outcomes. Compared with patients who remained ctDNA-positive after receiving ACT, those who achieved ctDNA clearance from MRD time point to 3- and 6-month time point in response to ACT had significantly longer DFS (clearance at 3 months: HR: 15.13, 95% CI 1.69-135.08, $P = 0.015$; clearance at 6-months: HR: 6.84, 95% CI 1.09-42.68, $P = 0.04$; Figure 4C and D).

ctDNA detection and site of recurrence in patients with radiological recurrence

Of the 189 patients who were recurrence free at the MRD time point, 50% (95/189) experienced radiological recurrence later during the follow-up. Among 95 patients who recurred, 55.8% (53/95) were MRD-positive, while the remaining 44.2% (42/95) were MRD-negative. Among the 42 MRD-negative patients, ctDNA results during the surveillance window were available for 35, 57.1% (20/35) of whom turned ctDNA positive before their radiological relapse. Notably, among patients with radiological recurrence, MRD positivity was associated with significantly shorter OS (HR: 6.08, 95% CI 1.28-28.8, $P = 0.023$; 36-month OS: 68.0%, 95% CI 45.90% to 82.70% versus 93.30%, 95% 61.30% to 99.00%, $P = 0.078$), when compared with MRD negativity (Figure 5A). We then assessed the correlation between the ctDNA detection rate in the MRD window and the site of recurrence. Of the 53 recurrences observed in the MRD-positive group, the site of recurrence was known for 52 patients (Figure 5B, Supplementary Table S1, available at https://doi.org/10.1016/j.annonc.2024.08.2240), 75.0% (39/52) of whom had metastases in the liver, while only 28.6 (12/42) recurrences in the MRD-negative group were in the liver ($P < 0.0001$; Figure 5C). Of those 12 patients, 58.3% (7/12) had fewer than two liver metastatic lesions and 91.7% (11/12) had metastatic liver lesions >50 mm. We noted that while most of the MRD-positive patients with radiological recurrence had liver involvement (73.6%, 39/53, $P = 0.0004$), a majority of the MRD-negative patients who had recurrence, had metastatic disease in the lung (61.9%, 26/42, $P = 0.0021$) (Figure 5D and E).

DISCUSSION

ctDNA as a tool to detect MRD has emerged as one of the strongest biomarkers to predict recurrence in CRC. Several groups have reported its prognostic relevance in CRC and suggested the potential role of ctDNA as a tool to guide personalized treatment after curative resection.[28,29] Recent data in GALAXY indicated that MRD-positive patients with stage II/III CRC derived significant benefit from ACT, but MRD-negative patients did not. In this subgroup analysis of GALAXY, similar findings were noted among patients with CLM without extrahepatic metastases. While the previously published interim analysis of the GALAXY study and this subgroup analysis are limited by their retrospective, observational nature, to our knowledge, this is the first report to show the potential utility of postsurgical ctDNA monitoring for risk stratification and identifying the subpopulation of CRC patients with liver-only metastases who are likely to benefit from ACT.

Several recent studies have shown ctDNA to be a strong prognostic biomarker in surgically resected CLM.[18-24] These studies have reported ctDNA positivity after surgery,[18-24] post-ACT,[18,22-24] and during surveillance[18,22] to be associated with significantly higher risk of recurrence. Furthermore, patients with postsurgical ctDNA positivity have also been shown to have worse OS.[20,23] Consistent with these reports, we observed that patients with ctDNA positivity after surgery in the MRD window as well as during surveillance experienced a significantly inferior DFS and OS. Additionally, in multivariate analyses, ctDNA positivity in the

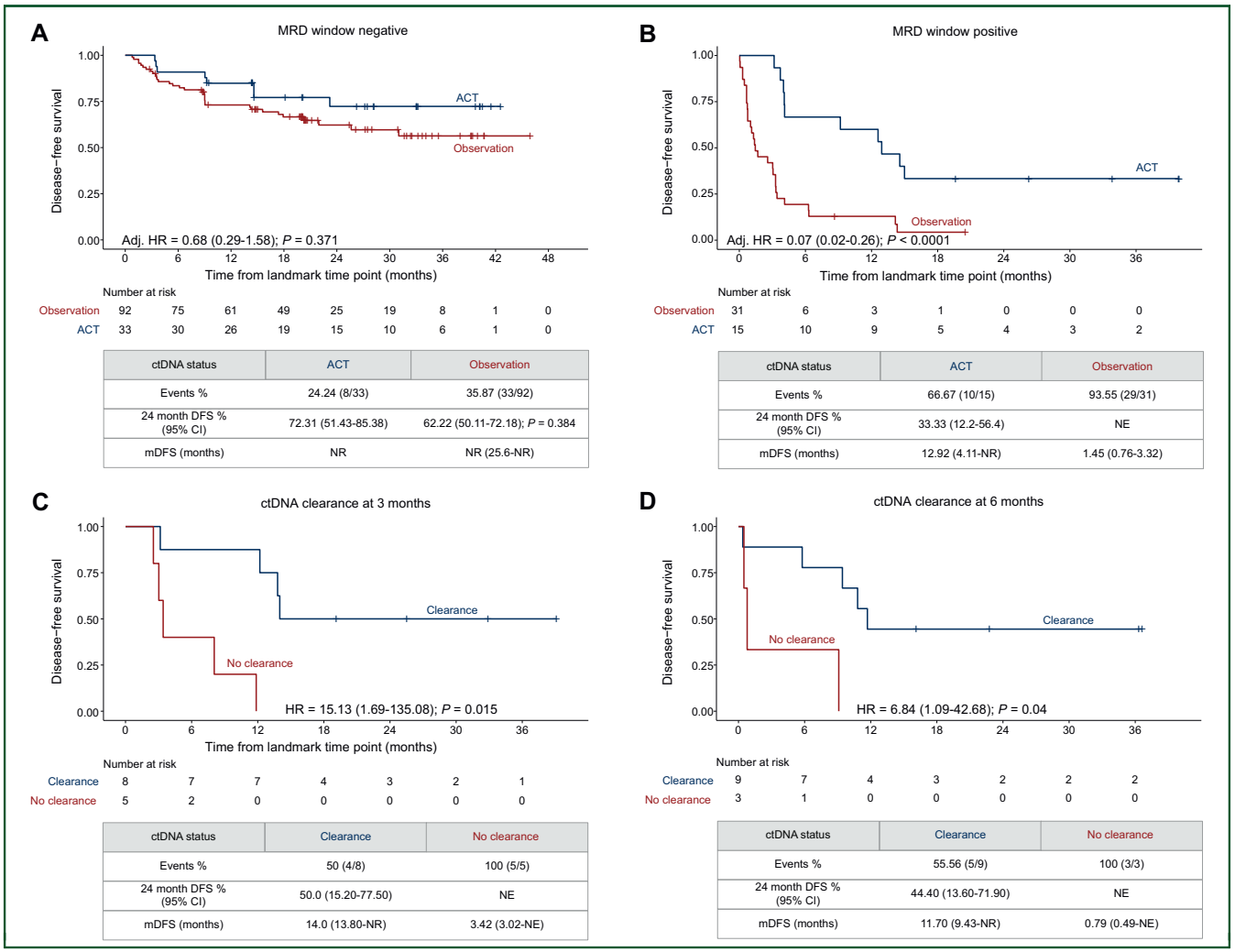

Figure 4. ctDNA status in the MRD window and ctDNA dynamics in response to ACT are predictive of ACT benefit in patients with CLM. Kaplan—Meier estimates for DFS stratified by adjuvant treatment (observation versus ACT) in (A) MRD-negative and (B) MRD-positive patients. HRs were adjusted for age, sex, number, and size of metastases, synchronicity, *RAS* mutational status, and prior oxaliplatin history. Kaplan—Meier estimates for DFS stratified by ctDNA clearance status from MRD window to (C) 3-month time point or (D) 6-month time point in MRD-positive patients receiving ACT. Analyses in (A) and (B) were landmarked at 2 months after surgery, analyses in (C) and (D) were landmarked at 3- and 6-month time point dates, respectively. HRs and 95% CIs were calculated using the Cox proportional hazard model and the corresponding *P* values were calculated using the two-sided log-rank test. Median DFS and percent DFS at 24 months were estimated from the landmark time point, and the corresponding *P* value was calculated using Fisher's exact test for proportions with the Bonferroni correction.
ACT, adjuvant chemotherapy; CI, confidence interval; CLM, colorectal liver metastases; ctDNA, circulating tumor DNA; HR, hazard ratio; mDFS, median disease-free survival; MRD, molecular residual disease; NE, not evaluable; NR, not reached.

MRD or surveillance window and number of liver metastases were found to be the only factors prognostic of DFS.

In this cohort, the frequency of post-operative complications was numerically higher in the MRD-positive (21%) group than in the MRD-negative group (7.8%). Several registry data indicated the association of post-operative complications with recurrence due to the invasive nature of hepatectomy procedures.[30,31] Additionally, subgroup analysis of JCOG0603 indicated that more aggressive hepatectomy negatively affected the compliance to ACT.[32] Upon experiencing severe complications, patients with resected CLM are expected to have delayed recovery and non-compliance to ACT. This may indicate that there is a potential bias of receiving ACT; number and size of CLM may be related with aggressiveness of surgery, which was associated with post-operative complication rate and administration of ACT. Therefore, when interpreting the difference of DFS and OS in MRD-positive patients with or without ACT, we need to take into account that this difference may be partly affected by the extent of surgery and post-operative complication. To avoid severe complications, when more aggressive hepatectomy is anticipated, safe procedures should be planned by combining with radiofrequency ablation for small tumors located deep within the liver and preoperative chemotherapy to shrink the tumor size.[33]

In this analysis, more than half of the patients had only one liver metastasis, and approximately one-third of these patients exhibited post-operative MRD positivity. These patients' characteristics and MRD positivity rate were comparable with previous reports.[23] There are no reports, however, in which benefit of ACT was assessed based on post-operative ctDNA status. Our results indicated benefit of ACT in MRD-positive patients: while most of the MRD-

https://doi.org/10.1016/j.annonc.2024.08.2240

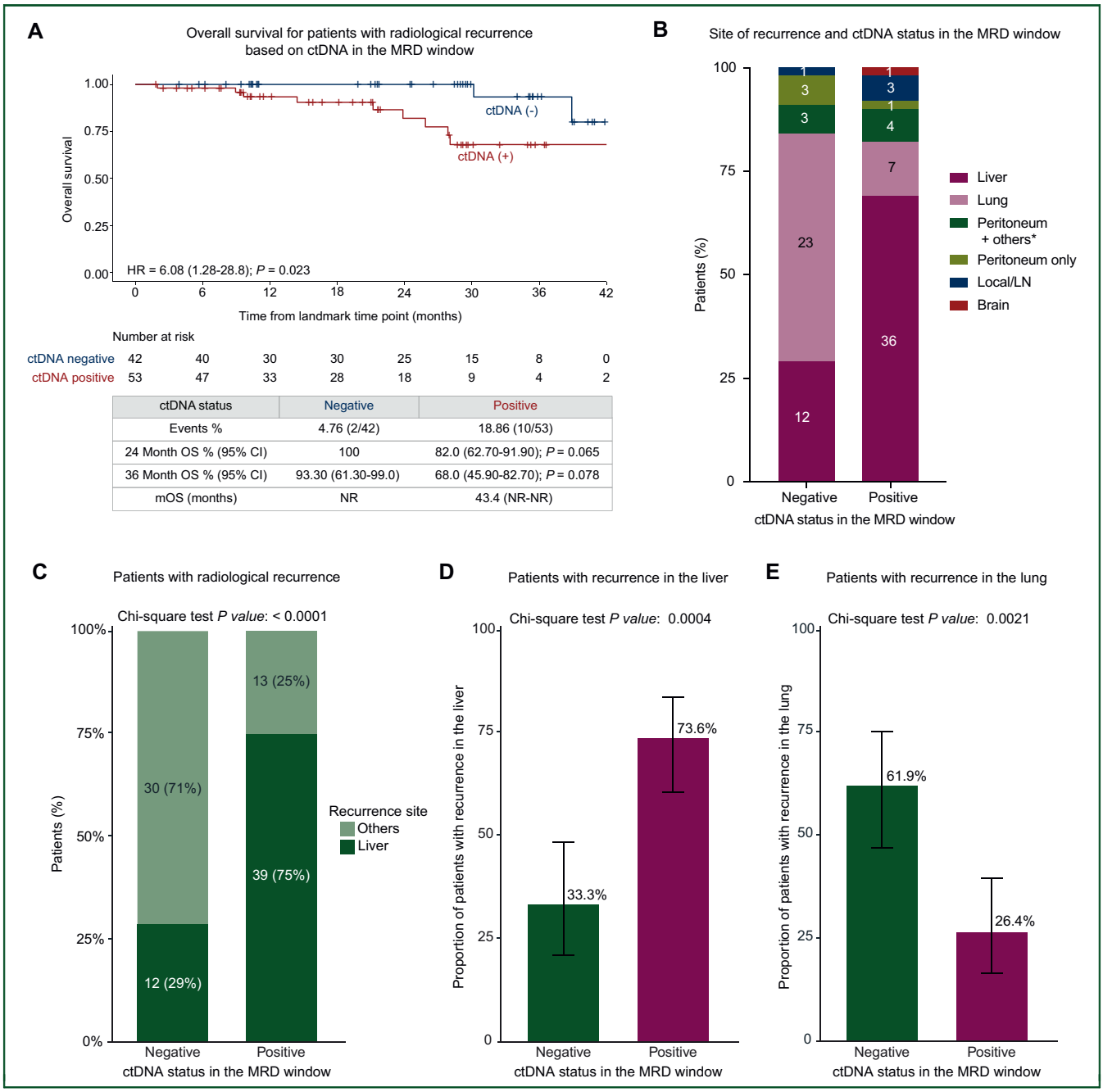

Figure 5. (A) Kaplan–Meier estimates for OS stratified by ctDNA status during the MRD window among patients with CLM who had a radiological recurrence during follow-up. This analysis was landmarked from the date of the MRD time point. HRs and 95% CIs were calculated using the Cox proportional hazard model and the corresponding *P* values were calculated using the two-sided log-rank test. Median and percent OS were estimated from the landmark time point and the corresponding *P* value was calculated using Fisher's exact test for proportions with the Bonferroni correction. (B) Bar plot showing ctDNA detection status in the MRD window among patients with radiological recurrence based on site of recurrence. *A total of 39/52 (75%) MRD-positive patients with radiological recurrence had metastatic involvement in the liver; 3 of whom had metastatic disease in both liver and peritoneum and were listed under the 'peritoneum + others' category. For 1 out of 95 patients with radiological recurrence, the site of recurrence was unknown. (C) Bar plot showing the percentage of liver involvement in MRD-positive versus -negative patients that recurred. (D) Bar plot showing the percentage of liver involvement in MRD-positive versus -negative patients that recurred. (E) Bar plot showing the MRD detection rate among CLM patients with recurrence in the lung.

CI, confidence interval; CLM, colorectal liver metastases; ctDNA, circulating tumor DNA; HR, hazard ratio; LN, lymph node; mDFS, median disease-free survival; MRD, molecular residual disease; NR, not reached; OS, overall survival.

positive patients on observation experienced clinical recurrence, ACT achieved ctDNA clearance in 61.5% (8/13) of MRD-positive patients resulting in 24-month DFS of 50%. ctDNA clearance rate in response to ACT was higher in our cohort than that reported in the retrospective study by Tie et al.[23] (3/11, 27.2%). While it is difficult to draw a definitive conclusion given the small cohort sizes and inadequate information regarding the regimen and intensity of ACT, both these cohorts indicate that the lack of ctDNA clearance on ACT was associated with markedly shorter DFS. Overall, these data suggest that ctDNA clearance on adjuvant therapy is indicative of ACT efficacy and that patients not achieving ctDNA clearance could be considered for treatment escalation or other therapies. The ongoing

randomized, double-blind, phase III ALTAIR study will evaluate the superiority of trifluridine/tipiracil compared with placebo in patients with resected CRC with positive ctDNA status at any time after standard-of-care ACT.[34]

Although our study suggests the benefit of ACT in MRD-positive patients, appropriate ACT duration and regimen in resectable CRC is still controversial. Currently, a non-randomized, phase II study is assessing ctDNA-directed ACT among patients with resected CLM, wherein ctDNA-positive patients will receive multiagent chemotherapy (FOLFOX or FOLFIRI), whereas ctDNA-negative patients will receive single-agent 5-FU or capecitabine or surveillance per provider judgment.[29] 'The first-strike strategy' using intensive chemotherapy may also be effective to eradicate MRD. Currently, several clinical trials to compare more intensive ACT such as FOLFOXIRI are ongoing in MRD-positive CRC patients after curative surgery (CIRCULATE-US, NCT05174169; AFFORD, NCT05427669; DYNAMIC-III, ACTRN126170015, and CLAUDIA, NCT05534087). In Japan, phase II studies to evaluate the efficacy of more intensive chemotherapy such as modified-FOLFOXIRI and FOLFOXIRI plus bevacizumab in resected oligometastatic disease are ongoing (AURORA and FANTASTIC).[35,36]

In a subgroup analysis in the MRD-negative group, ACT did not result in a meaningful DFS improvement in patients with either synchronous or metachronous CLM. Although not statistically significant, ACT seemed to be associated with superior DFS in MRD-negative patients with two or more liver metastases, compared with those with a single metastasis. Surprisingly, in this cohort, recurrence pattern was different in MRD-positive and -negative groups: among MRD-positive patients, only 25% of recurrences occurred in sites other than the liver, while among the MRD-negative group, 71% of recurrences were located in sites such as lung or peritoneum. This observation may be related to differences in ctDNA shedding according to the recurrence site.[37] Several series mentioned the low concordance of tissue-derived DNA and ctDNA in lung and peritoneal metastases when number and size of these metastases are small, which indicated that sensitivity of ctDNA is lower in lung or peritoneum than in liver or lymph node.[38,39] Typically, liver is the most frequent site of metastases after liver resection. Our data suggest that ctDNA analysis for this population may detect the potential liver metastases at earlier time points i.e. MRD positive, thus the frequency of potential lung or peritoneum micrometastases, which may be detected at the MRD window, will relatively increase in the MRD-negative group. This underscores the importance of longitudinal ctDNA testing to capture later recurrences.

Our study is associated with the following limitations: the sample size and duration of follow-up are still insufficient from the aspect of statistical power to draw definitive conclusions. Given the limited occurrence of CLM without extrahepatic metastases, previous phase III trials were also conducted in cohorts $<$300 patients. Furthermore, despite small sample size, we observed that MRD positivity could potentially identify patients who are likely to benefit from ACT, an observation that is consistent with other studies in CRC and other cancers.[17,31,40] OS data from MRD-positive and -negative patients stratified by adjuvant treatment (ACT versus observation) were not shown in this analysis because OS event data are not yet mature enough. No phase III randomized controlled trial (RCT) has yet demonstrated the OS benefit by ACT in this setting, therefore updated analysis with OS data will be awaited. The ACT regimen in this cohort was heterogeneous. Nevertheless, most of the patients received CAPOX as their ACT regimen for 3 to 6 months in accordance with Japanese treatment guidelines.[41] Finally, the study design was retrospective and observational in nature, since a randomized trial of ACT versus observation in patients with postsurgical ctDNA positivity was not feasible in Japan when CIRCULATE-Japan was initiated. A randomized study design comparing ACT versus observation with stratification according to ctDNA status is essential to validate our findings and support clinical adoption of ctDNA-guided adjuvant treatment decision-making. Such studies, although theoretically optimal, could be difficult to carry out in practice.

In conclusion, we observed a potential benefit of ACT for prolonging DFS in MRD-positive patients with CLM after surgical resection, while no statistically significant benefit of ACT was observed among MRD-negative patients. Our results indicate that ctDNA could be a useful biomarker to refine a subgroup of patients who would benefit from ACT in this setting. Further phase III, ctDNA-guided RCTs in CLM are warranted to further validate these findings.

ACKNOWLEDGEMENTS

We thank all the patients and their families for their participation, as well as the study investigators and study teams for their contributions.

FUNDING

This work was supported by the Japan Agency for Medical Research and Development [grant number 19ck0106447h0002, 21ck0106711h0001, 21ck0106710h0001, 22lk0201164h0001, 22lk0201148h0002] and the National Cancer Center Research and Development Fund [grant number 2021-A-6].

DISCLOSURE

KK reports speakers' bureau from Merck Biopharma Co., Ltd, Takeda Pharmaceutical Co., Ltd., Eli Lilly Japan Co., Ltd, and Guardant Health Japan Corp; research funding from Sysmex. KM reports speakers' bureau from Ono Pharmaceutical outside the submitted work. YN reports advisory role from Guardant Health Pte Ltd, Natera, Inc., Roche Ltd, Seagen, Inc., Premo Partners, Inc., Daiichi Sankyo Co., Ltd, Takeda Pharmaceutical Co., Ltd, Exact Sciences Corporation, Gilead Sciences, Inc.; speakers' bureau from Guardant Health Pte Ltd, MSD K.K., Eisai Co., Ltd, Zeria Pharmaceutical Co., Ltd, Miyarisan Pharmaceutical Co., Ltd, Merck Biopharma Co., Ltd, CareNet, Inc., Hisamitsu Pharmaceutical Co., Inc., Taiho Pharmaceutical Co., Ltd, Daiichi Sankyo Co.,

Ltd, Chugai Pharmaceutical Co., Ltd, Becton, Dickinson and Company, Guardant Health Japan Corp; research funding from Seagen, Inc., Genomedia Inc., Guardant Health AMEA, Inc., Guardant Health, Inc., Tempus Labs, Inc., Roche Diagnostics K.K., Daiichi Sankyo Co., Ltd, Chugai Pharmaceutical Co., Ltd. JW reports receiving honoraria for lectures from Johnson and Johnson, Medtronic, Eli Lilly, and Takeda Pharmaceuticals, and receiving research funding from Medtronic, AMCO, Terumo, and Stryker Japan outside the submitted work. MY reports receipt of personal fees and honoraria for lectures from Medtronic, Johnson & Johnson, Intuitive, Eli Lilly, Taiho, Merck KGaA, Takeda, Daiichi Sankyo. MK reports speakers' bureau from Taiho Pharmaceutical Co., Ltd, Chugai Pharmaceutical Co., Ltd, Takeda Pharmaceutical Co., Ltd, Eli Lilly, Yakult Honsha. KY reports speakers' bureau from Chugai Pharmaceutical Co., Ltd., Daiichi Sankyo Co., Ltd, Yakult Honsha Co., Ltd, Takeda Pharmaceutical Co., Ltd, Merck BioPharma Co., Ltd, Taiho Pharmaceutical Co., Ltd, Eli Lilly Japan K.K., Ono Pharmaceutical Co., Ltd, MSD Co., Ltd, Bristol Myers Squibb Co., Ltd; research funding from Taiho Pharmaceutical Co., Ltd. YK reports speakers' bureau from Guardant Health Pte Ltd, MSD K.K., Merck Biopharma Co., Ltd, Taiho Pharmaceutical Co. Ltd, Chugai Pharmaceutical Co., Ltd, Takeda Pharmaceutical Co., Ono Pharmaceutical Co., Ltd, and Eli Lilly Japan K.K. SM reports honoraria from Taiho Pharmaceutical Co., Ltd, Chugai Pharmaceutical Co., Ltd, and Eli Lilly Co., Ltd. GL, SS, CCP, and AJ are employees at Natera, Inc. with stock or options to own stock. MR reports employment, leadership position, stock and other ownership interests, consulting and advisory role at Natera Inc., MyOme, and Marble Therapeutics; and research funding, patents/royalties/other intellectual property, and travel/accommodations/expenses from Natera, Inc. and MyOme. MCL is an employee at Natera, Inc. with stock or options to own stock, and further reports grants/contracts (funding to Institution; Mayo Clinic) from Eisai, Exact Sciences, Genentech, Genomic Health, GRAIL, Menarini Silicon Biosystems, Merck, Novartis, Seattle Genetics, Tesaro; travel support reimbursement from AstraZeneca, Genomic Health, Ionis; and *ad hoc* advisory board meetings (all funds to Mayo Clinic; no personal compensation) from AstraZeneca, Celgene, Roche/Genentech, Genomic Health, GRAIL, Ionis, Merck, Pfizer, Seattle Genetics, Syndax. AA is an employee at Natera, Inc. with a leadership position and stock or options to own stock. DK reports honoraria from Takeda, Chugai, Lilly, MSD, Ono, Seagen, Guardant Health, Eisai, Taiho, Bristol Myers Squibb, Daiichi Sankyo, Pfizer, Merck Biopharma, and Sysmex: research funding from Ono, MSD, Novartis, Servier, Janssen, IQVIA, Syneos Health, CIMIC, and Cimic Shift Zero. HB reports research funding from Ono Pharmaceutical and honoraria from Ono Pharmaceutical, Eli Lilly Japan, and Taiho Pharmaceutical. HT reports speakers' bureau from MSD K.K, Merck Biopharma, Takeda, Taiho, Lilly Japan, Bristol Myers Squibb Japan, Chugai Pharmaceutical, Ono Yakuhin, Amgen; research funding from Takeda, Daiichi Sankyo. IT reports speakers' bureau from Medtronic, Johnson & Johnson, Intuitive, Medicaroid, Eli Lilly and research funding from Medtronic, Sysmex. TY reports honoraria from Chugai Pharmaceutical, Takeda Pharmaceutical, Merck Biopharma, Bayer Yakuhin, Ono Pharmaceutical and MSD K.K; consulting fee from Sumitomo Corp.; and research funding from Amgen, Bristol Myers Squibb, Chugai Pharmaceutical, Daiichi Sankyo, Eisai, FALCO Biosystems, Genomedia, Medical & Biological Laboratories, Merus N.V., Molecular Health GmbH, MSD, Nippon Boehringer Ingelheim, Ono Pharmaceutical, Pfizer Japan, Roche Diagnostics, Sanofi, Sysmex, Taiho Pharmaceutical, and Takeda Pharmaceutical. EO reports speakers' bureau from Chugai Pharmaceutical Co., Ltd, Bristol Myers, Ono Pharmaceutical Co., Ltd, Eli Lilly, Takeda Pharmaceutical Co., Ltd; research funding from Guardant Health, Inc.; advisory role from GlaxoSmithKline plc. All other authors have declared no conflicts of interest.

DATA SHARING

The authors declare that all relevant, non-proprietary data used to conduct the analyses are available within the article. To protect the privacy and confidentiality of patients in this study, clinical data are not made publicly available in a repository or the supplementary material of the article, but can be requested at any time from the corresponding author. Any requests will be reviewed within a time frame of 2 to 3 weeks by the CIRCULATE-Japan study steering committee to verify whether the request is subject to any intellectual property or confidentiality obligations. All data shared will be de-identified. Raw data of patient characteristics, outcomes, carcinoembryonic antigen (CEA) levels before surgery, and ctDNA results before surgery, at the MRD window, 3- and 6-months timepoints, and surveillance windows are included in Supplementary Table S2, available at https://doi.org/10.1016/j.annonc.2024.08.2240.

CODE AVAILABILITY

The fully documented code for the R statistical computing environment for analyses related to this manuscript are deposited at the github repository and can be accessed at https://github.com/Natera-TMED/Kataoka-et-al_CIRCULATE-Galaxy-Liver-Mets.git.

REFERENCES

1. Bray F, Laversanne M, Sung H, et al. Global cancer statistics 2022: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *CA Cancer J Clin*. 2024;74(3):229-263.
2. Adam R, de Gramont A, Figueras J, et al. Managing synchronous liver metastases from colorectal cancer: a multidisciplinary international consensus. *Cancer Treat Rev*. 2015;41(9):729-741.
3. Beppu T, Sakamoto Y, Hasegawa K, et al. A nomogram predicting disease-free survival in patients with colorectal liver metastases treated with hepatic resection: multicenter data collection as a Project Study for Hepatic Surgery of the Japanese Society of Hepato-Biliary-Pancreatic Surgery. *J Hepatobiliary Pancreat Sci*. 2012;19(1):72-84.
4. Georgilis E, Gavriatopoulou M, Tsilimigras DI, Malandrakis P, Theodosopoulos T, Ntanasis-Stathopoulos I. Optimizing adjuvant therapy after surgery for colorectal cancer liver metastases: a systematic review. *J Clin Med*. 2023;12(6):2401.
5. Mitry E, Fields AL, Bleiberg H, et al. Adjuvant chemotherapy after potentially curative resection of metastases from colorectal cancer: a

pooled analysis of two randomized trials. *J Clin Oncol*. 2008;26(30):4906-4911.
6. Portier G, Elias D, Bouche O, et al. Multicenter randomized trial of adjuvant fluorouracil and folinic acid compared with surgery alone after resection of colorectal liver metastases: FFCD ACHBTH AURC 9002 trial. *J Clin Oncol*. 2006;24(31):4976-4982.
7. Ychou M, Hohenberger W, Thezenas S, et al. A randomized phase III study comparing adjuvant 5-fluorouracil/folinic acid with FOLFIRI in patients following complete resection of liver metastases from colorectal cancer. *Ann Oncol*. 2009;20(12):1964-1970.
8. Nordlinger B, Sorbye H, Glimelius B, et al. Perioperative FOLFOX4 chemotherapy and surgery versus surgery alone for resectable liver metastases from colorectal cancer (EORTC 40983): long-term results of a randomised, controlled, phase 3 trial. *Lancet Oncol*. 2013;14(12):1208-1215.
9. Hasegawa K, Saiura A, Takayama T, et al. Adjuvant oral uracil-tegafur with leucovorin for colorectal cancer liver metastases: a randomized controlled trial. *PLoS One*. 2016;11(9):e0162400.
10. Kanemitsu Y, Shimizu Y, Mizusawa J, et al. Hepatectomy followed by mFOLFOX6 versus hepatectomy alone for liver-only metastatic colorectal cancer (JCOG0603): a phase II or III randomized controlled trial. *J Clin Oncol*. 2021;39(34):3789-3799.
11. Coombes RC, Page K, Salari R, et al. Personalized detection of circulating tumor DNA antedates breast cancer metastatic recurrence. *Clin Cancer Res*. 2019;25(14):4255-4263.
12. Tie J, Cohen JD, Lahouel K, et al. Circulating tumor DNA analysis guiding adjuvant therapy in stage II colon cancer. *N Engl J Med*. 2022;386(24):2261-2272.
13. Reinert T, Henriksen TV, Christensen E, et al. Analysis of plasma cell-free DNA by ultradeep sequencing in patients with stages I to III colorectal cancer. *JAMA Oncol*. 2019;5(8):1124-1131.
14. Huffman BM, Aushev VN, Budde GL, et al. Analysis of circulating tumor DNA to predict risk of recurrence in patients with esophageal and gastric cancers. *JCO Precis Oncol*. 2022;6:e2200420.
15. Martin TK, Dinerman A, Sudhaman S, et al. Early real-world experience monitoring circulating tumor DNA in resected early-stage non-small cell lung cancer. *J Thorac Cardiovasc Surg*. 2024. https://doi.org/10.1016/j.jtcvs.2024.01.017.
16. Botta GP, Abdelrahim M, Drengler RL, et al. Association of personalized and tumor-informed ctDNA with patient survival outcomes in pancreatic adenocarcinoma. *Oncologist*. 2024. https://doi.org/10.1093/oncolo/oyae155.
17. Kotani D, Oki E, Nakamura Y, et al. Molecular residual disease and efficacy of adjuvant chemotherapy in patients with colorectal cancer. *Nat Med*. 2023;29(1):127-134.
18. Li Y, Xu J, Hu X, et al. Personalized circulating tumor DNA monitoring improves recurrence surveillance and management after curative resection of colorectal liver metastases: a prospective cohort study. *Int J Surg*. 2024;110(5):2776-2787.
19. Liu W, Jin KM, Zhang MH, et al. Recurrence prediction by circulating tumor DNA in the patient with colorectal liver metastases after hepatectomy: a prospective biomarker study. *Ann Surg Oncol*. 2023;30(8):4916-4926.
20. Loupakis F, Sharma S, Derouazi M, et al. Detection of molecular residual disease using personalized circulating tumor DNA assay in patients with colorectal cancer undergoing resection of metastases. *JCO Precis Oncol*. 2021;5:PO.21.00101.
21. Marmorino F, Prisciandaro M, Giordano M, et al. Circulating tumor DNA as a marker of minimal residual disease after radical resection of colorectal liver metastases. *JCO Precis Oncol*. 2022;6:e2200244.
22. Ogaard N, Reinert T, Henriksen TV, et al. Tumour-agnostic circulating tumour DNA analysis for improved recurrence surveillance after resection of colorectal liver metastases: a prospective cohort study. *Eur J Cancer*. 2022;163:163-176.
23. Tie J, Wang Y, Cohen J, et al. Circulating tumor DNA dynamics and recurrence risk in patients undergoing curative intent resection of colorectal cancer liver metastases: a prospective cohort study. *PLoS Med*. 2021;18(5):e1003620.
24. Wang DS, Yang H, Liu XY, et al. Dynamic monitoring of circulating tumor DNA to predict prognosis and efficacy of adjuvant chemotherapy after resection of colorectal liver metastases. *Theranostics*. 2021;11(14):7018-7028.
25. Wullaert L, van Rees JM, Martens JWM, et al. Circulating tumour DNA as biomarker for colorectal liver metastases: a systematic review and meta-analysis. *Cells*. 2023;12(21):2520.
26. Taniguchi H, Nakamura Y, Kotani D, et al. CIRCULATE-Japan: circulating tumor DNA-guided adaptive platform trials to refine adjuvant therapy for colorectal cancer. *Cancer Sci*. 2021;112(7):2915-2920.
27. Kataoka K, Yamada T, Taniguchi H, Ikeda M, Yamazaki K, Kanemitsu Y. A ctDNA-driven multidisciplinary treatment strategy for resectable colorectal cancer -what surgical oncologists should know. *Eur J Surg Oncol*. 2022;48(1):1-2.
28. Folprecht G, Reinacher-Schick A, Weitz J, et al. The CIRCULATE trial: circulating tumor DNA based decision for adjuvant treatment in colon cancer stage II evaluation (AIO-KRK-0217). *Clin Colorectal Cancer*. 2022;21(2):170-174.
29. Sato S, Nakamura Y, Oki E, Yoshino T. Molecular residual disease-guided adjuvant treatment in resected colorectal cancer: focus on CIRCULATE-Japan. *Clin Colorectal Cancer*. 2023;22(1):53-58.
30. Schiesser M, Chen JW, Maddern GJ, Padbury RT. Perioperative morbidity affects long-term survival in patients following liver resection for colorectal metastases. *J Gastrointest Surg*. 2008;12(6):1054-1060.
31. Kasi PM, Aushev VN, Ensor J, et al. Circulating tumor DNA (ctDNA) for informing adjuvant chemotherapy (ACT) in stage II/III colorectal cancer (CRC): interim analysis of BESPOKE CRC study. *J Clin Oncol*. 2024;42(suppl 3):9.
32. Takamizawa Y, Kataoka K, Mizusawa J, et al. The impact of surgical invasiveness on the efficacy of mFOLFOX6 in resected colorectal liver metastasis: an exploratory analysis of JCOG0603. *Ann Oncol*. 2023;34(suppl 2):S438.
33. Meijerink MR, van der Lei S, Dijkstra M, et al. Surgery versus thermal ablation for small-size colorectal liver metastases (COLLISION): an international, multicenter, phase III randomized controlled trial. *J Clin Oncol*. 2024;42(suppl 17):LBA3501.
34. ClinicalTrials.gov [Internet]. ctDNA-directed post-hepatectomy chemotherapy for patients with resectable colorectal liver metastases. Bethesda, MD, USA: National Library of Medicine. Identifier NCT05062317. Last Updated: April 19, 2024. Available at https://clinicaltrials.gov/study/NCT05062317. Accessed July 23, 2024.
35. Ikeda M, Kataoka K, Yamada T, et al. A phase II study of mFOLFOXIRI after metastasectomy in patients with oligometastatic colorectal cancer (FANTASTIC study). *Ann Oncol*. 2024;35(suppl 1):S72.
36. Oki E, Nakanishi R, Ando K, et al. Recurrence monitoring using ctDNA in patients with metastatic colorectal cancer: COSMOS-CRC-03 and AURORA studies. *ESMO Gastrointest Oncol*. 2024;3:100034.
37. Cohen SA, Liu MC, Aleshin A. Practical recommendations for using ctDNA in clinical decision making. *Nature*. 2023;619(7969):259-268.
38. Kagawa Y, Elez E, Garcia-Foncillas J, et al. Combined analysis of concordance between liquid and tumor tissue biopsies for RAS mutations in colorectal cancer with a single metastasis site: the METABEAM study. *Clin Cancer Res*. 2021;27(9):2515-2522.
39. Bando H, Nakamura Y, Taniguchi H, et al. Effects of metastatic sites on circulating tumor DNA in patients with metastatic colorectal cancer. *JCO Precis Oncol*. 2022;6:e2100535.
40. Powles T, Assaf ZJ, Degaonkar V, et al. Updated overall survival by circulating tumor DNA status from the phase 3 IMvigor010 trial: adjuvant atezolizumab versus observation in muscle-invasive urothelial carcinoma. *Eur Urol*. 2024;85(2):114-122.
41. Hashiguchi Y, Muro K, Saito Y, et al. Japanese Society for Cancer of the Colon and Rectum (JSCCR) guidelines 2019 for the treatment of colorectal cancer. *Int J Clin Oncol*. 2020;25(1):1-42.