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ORIGINAL ARTICLE – HEPATOBILIARY TUMORS

Higher Tumor Burden Neutralizes Negative Margin Status in Hepatectomy for Colorectal Cancer Liver Metastasis

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ABSTRACT

Objective. The aim of this study was to examine if the prognostic significance of margin status in hepatectomy for colorectal cancer liver metastasis (CRLM) varies for different levels of tumor burden because hepatectomy indications for CRLM have been recently expanded to include patients with a higher tumor burden in whom achieving an R0 resection is difficult.

Methods. Clinicopathological variables in an exploration cohort of 290 patients receiving hepatectomy in Japan for CRLM were investigated. R0 resection was defined as a margin width > 0 mm. Tumor burden was assessed using the recently introduced Tumor Burden Score (TBS), which was calculated as TBS^2 = (maximum tumor diameter in $cm)^2$ + (number of $lesions)^2$. The principal findings were validated using a cohort from the United States.

Results. R1 resection rates significantly increased as TBS increased: 4/86 (4.7%) in patients with TBS < 3, 29/171 (17.0%) in patients with TBS ≥ 3 and < 9, and 9/33 (27.3%) in patients with TBS ≥ 9 ($p < 0.001$). R0 resection was significantly superior to R1 resection in patients with TBS ≥ 5; however, this was not the case for TBS ≥ 6, as confirmed by both univariate and multivariate analyses. Furthermore, prehepatectomy chemotherapy was associated with significantly improved survival for patients with TBS ≥ 8. Analysis of the validation cohort yielded similar results.

Conclusions. R0 resection appeared to have a positive impact on prognosis among patients with low tumor burden; however, this was not the case for patients with high tumor burden. As such, systemic treatment, in addition to surgery, may be central to achieving satisfactory outcomes in the latter patient population.

Although the definition of margin status lacks uniformity, an R0 resection in hepatectomy for colorectal cancer liver metastasis (CRLM) has been considered central to favorable outcomes.[1–14] However, recent changes in CRC management, such as the introduction of highly effective chemotherapy, has expanded the indications for CRLM resection towards patients with markedly advanced disease in whom achieving an R0 resection seems difficult.[15–19] Because of this, the significance of margin status in hepatectomy for CRLM has seemed to change recently[12,15,18–25]; however, the interplay of margin status with tumor burden has not been well studied to date. Achievement of R0 resection in hepatectomy for CRLM seems easier in patients with fewer and/or smaller tumors than in patients with more and/or larger tumors. We recently introduced the 'Tumor Burden Score' (TBS)[26] for accurately assessing tumor burden along a continuum incorporating a wide range of tumor size and number. In the present study, we investigated whether the prognostic significance of margin status in CRLM resection changes or not for different levels of TBS.

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PATIENTS AND METHODS

Exploration Cohort

Between January 2000 and December 2014, primary curative-intent hepatectomy was performed for 434 CRLM patients at Yokohama City University Hospital in Japan. From these 434 patients, 62 patients with extrahepatic disease at the time of index hepatectomy, 56 patients receiving staged hepatectomy (StHx),[27] and 26 patients receiving concomitant ablation were excluded. The majority of the 62 patients with extrahepatic disease did not undergo surgery for the extrahepatic disease. In other words, cancer-free status was not achieved in the majority of patients with extrahepatic disease. Of the 56 StHx patients, 35 received chemotherapy between the first and final procedures of StHx, and 23 of these 35 patients showed partial response (PR) to chemotherapy performed during StHx procedures. Tumor morphology seemed to change considerably during the procedures of StHx in these patients. Regarding concomitant ablation, ablated tumors could not be assessed pathologically; thus, these 144 patients were excluded from the present study. Accordingly, the exploration cohort consisted of 290 patients (Yokohama cohort). The Institutional Review Board of the Yokohama City University approved this study.

Information on clinicopathological variables, such as patient demographics, primary CRC characteristics according to the American Joint Committee on Cancer T stage,[28] CRLM characteristics, history of chemotherapy, and response to chemotherapy assessed by the Response Evaluation Criteria in Solid Tumors (RECIST), version 1.1,[29] were collected for the exploration cohort. Tumor burden was calculated as TBS using the following formula: $(\text{TBS})^2 = (\text{maximum tumor diameter in cm})^2 + (\text{number of lesions})^2$.[26] Tumor size and number were determined from the resected hepatic specimens. Prehepatectomy chemotherapy (PHC) was administered to initially unresectable or marginally resectable CRLM. Hepatectomy was not necessarily performed according to the anatomic principles of resection according to the Brisbane 2000 terminology of the International Hepato-Pancreato-Biliary Association.[30]

In the present study, R0 resection was defined as a resection where each metastasis was entirely covered by non-tumoral parenchyma regardless of its thickness at the liver resection cut surface (margin width > 0 mm), while R1 resection was defined as a resection in which at least one metastasis extended to the liver cut surface (margin width = 0 mm). Postoperative adjuvant chemotherapy was administered to a majority of patients. In patients with PHC, the preoperative chemotherapy was continued postoperatively as adjuvant. Patient follow-up was continued at least 5 years after the most recent surgery or until the patient's death. If recurrence developed, repeat surgery was performed if it was believed to be potentially curative.

Validation of Findings Obtained with the Yokohama Cohort

Due to the lengthy time period from which the patient records were drawn, the Yokohama cohort is not fully comparable with contemporary CRLM surgical cohorts. For example, in this cohort, PHC was administered to only one-third of patients. Furthermore, examination of KRAS mutation status was also not performed routinely. By contrast, contemporary surgical cohorts undergo PHC in approximately 60–70% of cases, and KRAS status is now routinely determined for almost all patients.[12–19,23,25,26] To transcend these limitations and ensure the generalizability of our findings to modern treatment settings, we validated the observations obtained with the Yokohama cohort in a cohort of patients who underwent resection for CRLM at Johns Hopkins Hospital, Baltimore, MD, USA (Baltimore cohort).

Statistical Analyses

Summary statistics were presented as whole numbers and percentages for categorical variables, or as median with interquartile ranges (IQRs) or ranges for continuous variables. Categorical variables were compared using the Chi square or Fisher's exact tests, and continuous variables were assessed using either the Mann–Whitney U or Kruskal–Wallis tests. The primary outcomes were disease-free survival (DFS) and overall survival (OS). DFS and OS were estimated using the Kaplan–Meier method, calculated from the date of index surgery; differences in DFS or OS were assessed using the log-rank or Breslow–Gehan–Wilcoxon test, as appropriate. Variables that demonstrated a significant association with the primary outcomes in univariate analysis ($p < 0.1$), or were considered important on biomedical grounds, were incorporated into the multivariate model. Subsequently, multivariate analysis was performed with the aid of the Cox proportional hazard regression model by the forced-entry or backward-elimination manner, as appropriate. A two-tailed p value < 0.05 was considered statistically significant. All analyses were carried out using SPSS statistical software, version 23.0 (IBM SPSS, Chicago, IL, USA).

RESULTS

Clinicopathological Characteristics and Surgical Outcomes of the Exploration Cohort

Patient demographics and clinicopathological variables of the Yokohama cohort are summarized in Table 1. PHC

TABLE 1 Baseline patient demographics and clinical characteristics of the Yokohama cohort

	All patients (n = 290)	
	N	%
Patient characteristics		
Age (years)	66.0 (50.0–73.0)	
Sex		
Male/female	198/92	68.3/31.7
Treatment period		
2000–2004/2005–2009/2010–2014	73/106/111	25.2/36.5/38.3
Primary CRC characteristics		
Tumor site (including multiple cases)		
Right colon/left colon/rectum	75/106/109	25.9/36.5/37.6
T stage		
T1/T2/T3/T4	5/14/247/24	1.7/4.8/85.2/8.3
Nodal metastases		
Negative–positive	101/189	34.8/65.2
Tumor differentiation		
Differentiated/less differentiated	265/25	91.4/8.6
Preoperative variables		
Synchronous/metachronous	142/148	49.0/51.0
DFI $<$ 12 months/DFI $>$ 12 months	219/71	75.5/24.5
Prehepatectomy chemotherapy		
Not received/received	198/92	68.3/31.7
Prehepatectomy modern chemotherapy		
Not received/received	229/61	79.0/21.0
Without/with biologic agents (n = 92)	56/36	58.6/41.4
RECIST response (n = 92)		
SD or PD/PR or rCR	47/45	51.0/49.0
Prehepatectomy CEA level, ng/ml [median (IQR)]	8.1 (3.2–39.0)	
Tumor factors		
Number of CRLMs [median (IQR)]	2 (1–5)	
Size of the largest CRLM, cm [median (IQR)]	3.0 (1.9–4.3)	
Tumor burden score [median (IQR)]	4.4 (2.7–6.7)	
Bilobar disease	111	38.3
KRAS status (n = 88)		
Wild-type/mutated	53/35	60.2/39.8
Operation variables		
Major hepatectomy/minor hepatectomy	80/210	27.6/72.4
Preceding portal embolization	24	8.3
Resection margin width, mm [median (IQR)]	3.0 (0–7.0)	
R0 resection/R1 resection	248/42	85.5/14.5
Postoperative factors		
Postoperative complications		
Clavien–Dindo grade 2 or higher	46	15.8
Clavien–Dindo grade 3 or higher	18	6.2
Bile leakage	6	
Intraperitoneal infection	5	
Intestinal obstruction	3	
Hemoperitoneum	1	
Mortality	2	0.7

TABLE 1 continued

	All patients (n = 290)	
	N	%
Portal venous thrombosis	1	
Non-occlusive mesenteric ischemia	1	
Adjuvant chemotherapy	163	56.2

CEA carcinoembryonic antigen, *CRC* colorectal cancer, *CRLM* colorectal cancer liver metastasis, *DFI* disease-free interval, *IQR* interquartile range, *KRAS* Kirsten rat sarcoma viral oncogene homolog, *PD* progressive disease, *PR* partial response, *rCR* radiologic complete response, *RECIST* Response Evaluation Criteria in Solid Tumors, *SD* stable disease

was administered to 92 patients (31.7%). The median number of CRLMs was 2 (IQR 1–5), maximum tumor diameter 3.0 cm (IQR 1.9–4.3), and TBS 4.4 (IQR 2.7–6.7) (electronic supplementary Fig. 1a). At a median follow-up of 43.7 months, 108 patients died. Median DFS and OS were 15.8 and 91.2 months, respectively. Furthermore, 1-, 5-, and 10-year DFS and OS rates were 57.4% and 98.6%, 33.9% and 62.5%, and 28.2% and 40.2%, respectively.

Prognostic Implication of Tumor Burden Score (TBS) and Independent Prognostic Factors for Overall Survival in the Exploration Cohort

Dividing the Yokohama cohort into three groups according to TBS (Group 1, < 3; Group 2, 3–9, Group 3, ≥ 9) appeared to provide adequate discrimination of survival outcomes (electronic supplementary Fig. 2). Importantly, DFS and OS rates were significantly different across these three TBS groups ($p < 0.001$). Median OS and 5-year OS rate decreased significantly in the order of Groups 1, 2, and 3. In the Yokohama cohort, cumulative DFS and OS rates were significantly higher in the R0 resection group ($n = 248$) compared with the R1 resection group ($n = 42$). Median DFS values and 5-year DFS rates were 17.5 months and 36.7%, and 11.8 months and 16.1% for the R0 and R1 groups, respectively ($p < 0.007$). Median OS values and 5-year OS rates were 98.8 months and 68.0%, and 43.5 months and 31.5% for the R0 and R1 groups, respectively ($p < 0.001$) (electronic supplementary Fig. 2).

Multivariate analysis identified the following independent prognostic factors for DFS: TBS [hazard ratio (HR) 1.077, 95% confidence interval (CI) 1.048–1.106, $p < 0.001$], disease-free interval < 12 months (HR 1.824, 95% CI 1.217–2.733, $p = 0.004$), margin status (HR 1.493, 95% CI 1.024–2.686, $p = 0.008$), and prehepatectomy serum carcinoembryonic antigen (CEA) level ≥ 10 ng/ml (HR 1.529, 95% CI 1.129–2.071, $p = 0.006$). Furthermore, the following were determined as independent prognostic factors for OS: TBS (HR 1.082, 95% CI 1.050–1.115, $p < 0.001$), primary tumor lymph node status (HR 2.098, 95% CI 1.283–3.433, $p = 0.004$), margin status (HR 1.593, 95% CI 1.027–2.606, $p = 0.011$), and prehepatectomy serum CEA level ≥ 10 ng/ml (HR 1.934, 95% CI 1.299–2.880, $p = 0.001$) (electronic supplementary Table 1).

Prognostic Significance of Margin Status for Different Levels of TBS in the Exploration Cohort

R1 resection rates were significantly greater among patients with a higher TBS: 4/86 (4.7%) in Group 1, 29/171 (17.0%) in Group 2, and 9/33 (27.3%) in Group 3 ($p < 0.001$). When patients were dichotomized by using a TBS cut-off value of 6, 7, 8, or 9, DFS and OS were significantly worse for the R1 resection group than R0 for those with TBS less than the cut-off. By contrast, neither DFS nor OS was different between the R1 and R0 resection groups for those with TBS more than the cut-off (electronic supplementary Fig. 3, and Fig. 1). With an increasing TBS cut-off value, from 6 to 9, the survival difference decreased. A difference of survival between the R0 and R1 resection groups was observed for TBS cut-off values of 5, 4, and 3 (data not shown).

Regarding the impact of PHC, DFS was not different between patients who received PHC and those who did not, among patients with TBS beyond 6, 7, or 8 (electronic supplementary Fig. 4). However, in patients with TBS ≥ 9, DFS was significantly better in patients who received PHC than in those who did not. Furthermore, OS was similar between patients who received PHC and those who did not among those with TBS of ≥ 6 or ≥ 7. However, in patients with TBS ≥ 8 or ≥ 9, OS was significantly better in patients who received PHC than in those who did not. Of note, there were no long-term survivors among patients with TBS ≥ 9 who did not receive PHC (electronic supplementary Fig. 4).

Taking the above-stated findings into account, repeated multivariate analyses, including variables such as treatment era, TBS, primary node status, disease-free interval, margin status, serum CEA level, and PHC, were performed for patients with TBS ≥ 6, ≥ 7, ≥ 8 or ≥ 9, in a backward-elimination manner. Results of the final step of each analysis are shown in Table 2. Among patients with

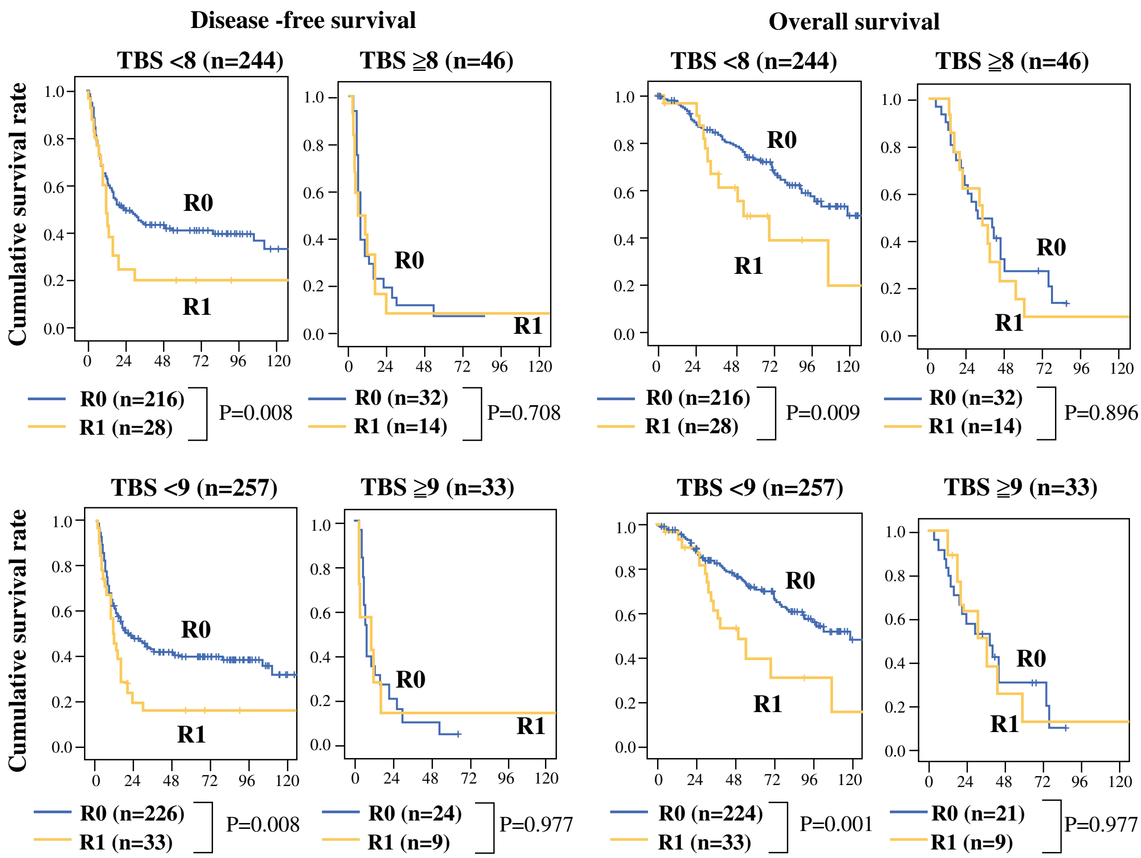

FIG. 1 Eradication of advantage of R0 resection according to increased TBS in the Yokohama cohort. Because the 75th percentile value of TBS was approximately 6, the cohort was repeatedly dichotomized by TBS cut-off values of 6, 7, 8, or 9. When the cohort was dichotomized by using TBS cut-off values of 6, 7, 8, or 9, DFS and OS were significantly worse for the R1 resection group than the R0 group for patients with TBS less than the cut-off. By contrast, neither DFS nor OS were different between the R1 and R0 groups for patients with TBS more than the cut-off. As the TBS cut-off value is increased from 6 to 9, the prognostic benefit of R0 resection gets eradicated (results for TBS cut-off values of 6 and 7 are reported in electronic supplementary Fig. 3). *TBS* Tumor Burden Score, *DFS* disease-free survival, *OS* overall survival

TBS $\geq$ 6, independent prognostic factors were identified as TBS, margin status, and serum CEA level $\geq$ 10 ng/ml for DFS, and TBS, primary node status, and serum CEA level for OS (Table 2a). However, TBS and margin status were not independent predictors of prognosis in patients with TBS $\geq$ 7. For these patients, primary node status and/or serum CEA level were independently prognostic for both DFS and OS (Table 2b). Among patients with TBS $\geq$ 8, prehepatectomy serum CEA level was determined to be the sole independent prognostic factor for both DFS and OS (Table 2c). Among patients with TBS $\geq$ 9, PHC and/or serum CEA level were independently prognostic for both DFS and OS (Table 2d).

Validation of Prognostic Significance of Margin Status for Different Levels of TBS in the Baltimore Cohort

Between January 2000 and December 2014, 582 patients with CRLM underwent hepatectomy at Johns Hopkins Hospital (Baltimore, MD, USA). One-hundred and eighty-nine of these patients for whom KRAS mutation status was unavailable or concomitant ablation had been performed were excluded. In addition, 36 patients with extrahepatic disease at the time of index hepatectomy were excluded. The Baltimore cohort, thus consisting of the remaining 357 patients, of whom 222 (62.9%) received PHC, was used to validate the findings on prognostic significance of margin status for different TBS levels that had been obtained with the exploration (Yokohama) cohort. Because comparison of TBS between the Yokohama and Baltimore cohorts showed that TBS was significantly lower in the latter (electronic supplementary Fig. 1), the TBS cut-off values were set lower for the Baltimore cohort compared with the Yokohama cohort, i.e. 4, 5, 6, or 7. Although the TBS cut-offs were set one unit lower than for the Yokohama cohort, the varying significances of R0/1 resection with different TBS cut-off values were also observed in the validation cohort, similar to the findings with the Yokohama cohort.

TABLE 2 Multivariate analysis of the Yokohama cohort for independent prognostic factors for disease-free or overall survival with tumor burden score ≥ 6, ≥ 7, ≥ 8, or ≥ 9

Variables	HR	95% CI	*p* value
(a) Tumor Burden Score ≥ 6 (*n* = 89)			
Disease-free survival			
Tumor Burden Score (continuous value)	1.055	1.006–1.107	0.028
Margin status (R0 resection, 0; R1 resection, 1)	1.576	1.100–2.257	0.013
Serum CEA level (< 10 ng/ml, 0; > 10 ng/ml, 1)	1.671	1.027–2.720	0.039
Overall survival			
Tumor Burden Score (continuous value)	1.078	1.025–1.135	0.004
Primary node status (negative, 0; positive, 1)	2.436	1.087–5.458	0.031
Serum CEA level (< 10 ng/ml, 0; ≥ 10 ng/ml, 1)	2.088	1.087–4.013	0.027
(b) Tumor Burden Score ≥ 7 (*n* = 65)			
Disease-free survival			
Tumor Burden Score (continuous value)	1.044	0.984–1.207	0.154
Serum CEA level (< 10 ng/ml, 0; > 10 ng/ml, 1)	2.215	1.007–4.872	0.048
Overall survival			
Primary node status (negative, 0; positive, 1)	1.065	1.007–1.126	0.027
Serum CEA level (< 10 ng/ml, 0; ≥ 10 ng/ml, 1)	2.462	1.172–5.173	0.017
(c) Tumor Burden Score ≥ 8 (*n* = 46)			
Disease-free survival			
Serum CEA level (< 10 ng/ml, 0; ≥ 10 ng/ml, 1)	3.338	1.255–8.879	0.016
Overall survival			
Prehepatectomy chemotherapy (not received, 0: received, 1)	1.452	0.968–1.100	0.080
Serum CEA level (< 10 ng/ml, 0; ≥ 10 ng/ml, 1)	3.338	1.956–7.266	0.022
(d) Tumor Burden Score ≥ 9 (*n* = 33)			
Disease-free survival			
Prehepatectomy chemotherapy (not received, 0; received, 1)	0.137	0.039–0.479	0.002
Serum CEA level (< 10 ng/ml, 0; ≥ 10 ng/ml, 1)	3.082	0.885–10.734	0.077
Overall survival			
Prehepatectomy chemotherapy (not received, 0; received, 1)	0.186	0.051–0.679	0.011
Serum CEA level (< 10 ng/ml, 0; ≥ 10 ng/ml, 1)	5.313	1.278–22.090	0.022

CEA carcinoembryonic antigen, *CI* confidence interval, *HR* hazard ratio

In other words, when the cohort was dichotomized by using TBS cut-off values of 4, 5, 6, or 7, DFS and OS were significantly worse in the R1 resection group than in the R0 group in patients with TBS less than the cut-off. By contrast, neither DFS nor OS was different between the R0 and R1 groups in patients with TBS more than the cut-off (electronic supplementary Fig. 5). The prognostic benefit derived from an R0 resection thus appeared to weaken as TBS increased. Multivariate analyses by TBS were also performed (electronic supplementary Table 2). Independent prognostic factors observed for the validation cohort were similar to those seen for the exploration cohort. Specifically, TBS, primary node status, disease-free interval, margin status, and serum CEA level were prognostic for DFS, and TBS, primary node, margin status, mutant KRAS status, and serum CEA level were prognostic for OS in the validation cohort.

Regarding the impact of PHC, DFS was not different between patients who received PHC and those who did not, among those with TBS ≥ 4, ≥ 5, or ≥ 6. In patients with *TBS ≥ 7 or ≥ 8, DFS tended to be better, and in those with TBS ≥ 9, DFS was significantly better for those patients who received PHC than those who did not. Furthermore, OS was similar between patients who received PHC and those who did not among those with TBS ≥ 4. However, in patients with TBS ≥ 8, OS was significantly better in patients who received PHC than in those who did not. Of note, there were no long-term survivors among patients with TBS ≥ 9 who did not receive PHC.

Taking the above-stated findings into account, repeated multivariate analyses, including variables such as treatment era, TBS, primary node status, disease-free interval, margin status, serum CEA level, and PHC, were performed. Results of the final step of each analysis are shown in Table 3. In patients with TBS ≥ 4, independent prognostic factors were determined as TBS, margin status, and KRAS mutation status for DFS. For OS, independent prognostic factors were TBS, primary node status, and KRAS

TABLE 3 Multivariate analysis of the Baltimore cohort for independent prognostic factors for disease-free or overall survival with tumor burden score ≥ 4, ≥ 5, ≥ 6, or ≥ 7

Variables	HR	95% CI	*p* value
(a) Tumor Burden Score ≥ 4 (*n* = 166)			
Disease-free survival			
Tumor Burden Score (continuous value)	1.112	1.012–1.211	0.011
Margin status (R0 resection, 0; R1 resection, 1)	2.560	1.105–5.935	0.028
KRAS status (wild-type, 0; mutated, 1)	2.087	1.182–3.683	0.011
Overall survival			
Tumor Burden Score (continuous value)	1.104	1.010–1.312	0.042
Primary node status (negative, 0; positive, 1)	1.306	1.113–2.378	0.021
KRAS status (wild-type, 0; mutated, 1)	3.645	2.070–6.418	< 0.001
(b) Tumor Burden Score ≥ 5 (*n* = 105)			
Disease-free survival			
KRAS status (wild-type, 0; mutated, 1)	2.087	1.182–3.683	0.011
Overall survival			
Serum CEA level (< 50 ng/ml, 0; ≥ 50 ng/ml, 1)	4.371	2.125–8.992	0.029
KRAS status (wild-type, 0; mutated, 1)	3.645	2.070–6.418	< 0.001
(c) Tumor Burden Score ≥ 6 (*n* = 65)			
Disease-free survival			
KRAS status (wild-type, 0; mutated, 1)	2.842	1.152–7.010	0.023
Overall survival			
Prehepatectomy chemotherapy (not received, 0; received, 1)	0.912	0.891–1.235	0.071
KRAS status (wild-type, 0; mutated, 1)	3.645	2.070–6.418	< 0.001
(d) Tumor Burden Score ≥ 7 (*n* = 44)			
Disease-free survival			
Prehepatectomy chemotherapy (not received, 0; received, 1)	0.885	0.645–0.984	0.035
KRAS status (wild-type, 0; mutated, 1)	1.825	1.152–6.478	0.039
Overall survival			
Prehepatectomy chemotherapy (not received, 0; received, 1)	0.812	0.578–0.912	0.011
KRAS status (wild-type, 0; mutated, 1)	3.645	2.070–6.418	< 0.001

KRAS Kirsten rat sarcoma viral oncogene homolog, *CEA* carcinoembryonic antigen, *CI* confidence interval, *HR* hazard ratio

mutation status (Table 3a). However, TBS and margin status were not independent predictors of prognosis in patients with TBS ≥ 5, although KRAS mutation status and/or serum CEA level remained prognostic for DFS or OS (Table 3b). In patients with TBS ≥ 6, KRAS mutation status was determined to be the sole independent prognostic factor for both DFS and OS (Table 3c). In patients with TBS ≥ 7, PHC and KRAS mutation status were independent prognostic factors for both DFS and OS (Table 3d).

Impact of the Effect of Prehepatectomy Chemotherapy on Margin Status

To evaluate the impact of PHC, we evaluated the associations among margin status, TBS, PHC regimens, number of administered PHC courses, effect of PHC, and outcome of surgery. PHC regimens, lines, and course numbers are summarized in electronic supplementary Table 3. In the Yokohama cohort, negative impact of R1 resection on OS was eradicated in patients with TBS ≥ 8 or ≥ 9 for those who exhibited PR to PHC irrespective of regimen or number of administered courses. In contrast, the disadvantageous effect of an R1 resection remained in those who had stable disease (SD) or progressive disease (PD) in response to PHC (electronic supplementary Fig. 6). Although a prognostic difference between modern and other regimens was not observed in the Yokohama cohort, the interesting findings regarding the association between the impact of margin status and the use of modern regimens were observed in the Baltimore cohort. In this cohort, in which modern cytotoxic regimens were used for most of the patients receiving PHC, the disadvantageous effect of an R1 resection was eradicated in patients exhibiting PR in response to PHC using modern regimens, but not in those exhibiting SD or PD. In other words, a difference in DFS or

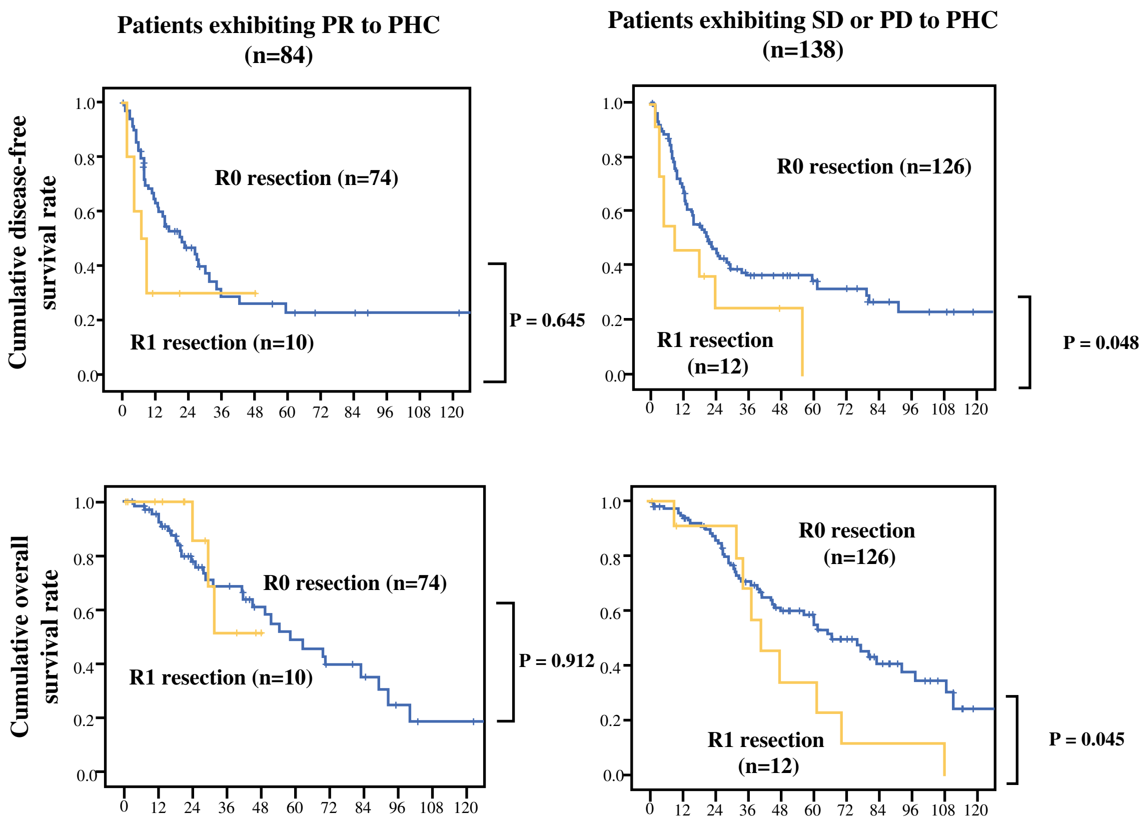

FIG. 2 Eradication of the disadvantageous effect of an R1 resection in patients exhibiting partial response to prehepatectomy modern cytotoxic chemotherapy. In the validation (Baltimore) cohort in which modern cytotoxic regimens were used for most patients receiving PHC, the disadvantageous effect of an R1 resection was eradicated in patients exhibiting PR to PHC using modern regimen(s), but not in patients exhibiting SD or PD. *PHC* prehepatectomy chemotherapy, *PR* partial response, *SD* stable disease, *PD* progressive disease

OS was not observed among the R0 and R1 resection groups in patients showing PR to PHC. By contrast, in patients with SD or PD in response to PHC, the disadvantageous effect of an R0 resection remained, i.e. either DFS or OS was significantly better for the R0 group than the R1 group (Fig. 2).

DISCUSSION

In many studies of the prognostic significance of hepatectomy margin status, the number and size of tumors is evaluated dichotomously, such as whether there was one or multiple tumors, or if tumor size was less or more than 5 cm.[1,2,4–7,9,12,13,15,20–23] Such binary evaluation of the tumor number and size variables is inappropriate for determining tumor burden. In the present study, we use a more accurate metric for tumor burden (TBS), which quantifies tumor burden along a continuum that incorporates a wide range of tumor size and number as continuous variables,[26] to evaluate if the prognostic significance of margin status changes according to tumor burden. We found that R0 resection is significantly important for gaining better outcomes up to a certain level of tumor burden, beyond which the advantage of R0 compared with R1 resection is eradicated. In addition, we show that there have been no long-term survivors among patients with TBS > 9 who did not receive PHC (electronic supplementary Fig. 4). This suggests that disease cure cannot be accomplished through surgery alone, and thus treatment additional to surgery, such as preoperative chemotherapy, is required for patients with high tumor burden.

Recently, some studies showed that biological behavior of CRLM is more decisive for treatment outcome rather than technical aspects of surgery, which seemed reflected by margin status to some extent, in the era of modern chemotherapy.[15,17–19] CRLM presenting with TBS > 9 should be considered to have high malignant potential. For such cases, aggressive tumor biology is an important determinant of outcomes. Increased serum CEA level and mutated KRAS have been reported as important surrogates of aggressive tumor biology,[17–19,25,26,31,32] and these two factors were determined to represent the independent prognostic factors in patients with higher tumor burden in this study. Hence, ingenuity in intensifying the effect of preoperative chemotherapy, including through the use of biologic agents, may be more important for gaining better outcomes rather than efforts to obtain negative margin status.[25] In contrast, an advantage of PHC was not observed among patients with TBS up to 6 or 7 in the present study for either the exploration or the validation cohort (electronic supplementary Fig. 4). Therefore, TBS may be useful as an indicator for PHC.

TBS is very easy to calculate based on radiological findings such as computed tomography (CT) imaging, and thus chronological changes in imaging TBS during chemotherapy can be more easily assessed compared with RECIST[28] or the morphologic response that has been proposed by the MD Anderson Group.[12] Responses to PHC have been reportedly significantly correlated with outcomes of hepatectomy for CRLM.[11–13,15–19,23–25] Very recently, imaging TBS proved to be significantly correlated with pathological TBS and to have excellent prognostic discriminatory ability.[33] Therefore, we consider that chronological changes of imaging TBS can substitute for these response criteria, and we are currently constructing a prospective database to examine this.

The advantageous effect of PHC to hepatectomy for CRLM may be explained by two mechanisms: neutralization of invasive front of the CRLM lesions, and eradication of micrometastases. The results of the present study, as well as some previous studies,[12,13,15–20,24] demonstrating that the disadvantageous effect of an R1 resection is eradicated by the effect of PHC, support the first mechanism. As for the second mechanism, some authors have concluded that a wider margin width was optimal for CRLM surgery.[5,9,10] In other words, they believed that the wider the margin width, the larger the chance of cure. Notably, Wakai et al.[5] clearly demonstrated that 1 cm or more of margin width was required for eradicating micrometastases surrounding the CRLM lesions. However, the majority of patients did not undergo PHC, and the size and number of lesions were evaluated dichotomously in these studies. Contemporary surgical cohorts underwent PHC in approximately 60–70% of cases.[12–19,23,25,26] In addition, recent highly effective chemotherapy has expanded the indications for CRLM resection towards patients with markedly advanced disease.[15–19] Previous studies showing that a better response to PHC was significantly associated with a better survival after hepatectomy for CRLM [11–13,15–19,23–25] support the idea that effective PHC is considered to be beneficial for neutralizing the invasive front of lesions, as well as eradicating micrometastases. Obtaining a wider margin width is considered more difficult in patients with a high tumor burden than in those with a lower burden. Therefore, although wider margin width seems desirable, the effort to obtain wider margin is considered less important than effective PHC in patients with high tumor burden. Conversely, obtaining wider margin may be more essential than PHC in patients with a low tumor burden.

We recognized that there were two critical drawbacks in the exploration cohort of our study: lack of evaluation of KRAS mutation status, and scarce use of PHC. KRAS mutation status has been considered to reflect biological behavior of tumors and has reportedly significant impact on outcomes of hepatectomy for CRLM.[17–19,32] In addition, PHC using modern cytotoxic agents, such as oxaliplatin and irinotecan, has been used for the majority of CRLM patients receiving hepatectomy in the current global trend. To overcome these drawbacks, we validated the findings obtained with the exploration cohort using a second patient cohort, for which KRAS status had been routinely evaluated and PHC provided to a majority of patients. The validation supported the findings of the main analysis of the exploration cohort, that the effect of R0 resection was significant up to a certain level of tumor burden but was eradicated beyond that level. Furthermore, treatment additional to surgery, such as PHC, seemed necessary for gaining better outcomes of surgery for CRLM patients with higher tumor burden. Of note, the latter was strongly supported by the fact that no long-term survivor existed among patients with TBS > 9 who did not receive PHC in either of the two cohorts.

Several drawbacks still remain in the present study. TBS used for the present analyses were calculated from pathological parameters of resected specimens. Namely, in patients receiving PHC, their TBS was calculated using resultant tumor morphology from PHC. Thus, the comparison of TBS between patients who did and did not receive PHC is not fair because the response rate of the modern cytotoxic regimens has reportedly been $> 60\%$,[11–13,15,17–19,23–25] and PHC can significantly alter TBS for patients receiving such regimens. In fact, the exploration (Yokohama) and validation (Baltimore) cohorts of our study differed significantly for distribution of TBS probably because of this reason (electronic supplementary Fig. 1). We recognize that significance and patient selection criteria of PHC cannot be appropriately assessed with the present study.

CONCLUSION

R0 resection was important for gaining better outcomes of hepatectomy for CRLM to a certain level of tumor burden; however, beyond this level, the advantage of R0 resection was eradicated. Treatment additional to surgery, such as PHC, may be central to favorable outcomes of surgery for CRLM patients with high tumor burden.

DISCLOSURE The authors declare that they have no conflict of interest.

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