ORIGINAL ARTICLE

Local tumour control after radiofrequency or microwave ablation for colorectal liver metastases in relation to histopathological growth patterns

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Abstract

Background: Regrowth after ablation is common, but predictive factors for local control are scarce. This study investigates whether histopathological growth patterns (HGP) can be used as a predictive biomarker for local control after ablation of colorectal liver metastases (CRLM).

Methods: Patients who received simultaneous resection and ablation as first treatment for CRLM between 2000 and 2019 were considered eligible. HGPs were determined on resected CRLM according to international guidelines and were classified as desmoplastic or non-desmoplastic. As minimal intertumoural heterogeneity has been demonstrated, the HGP of resected and ablated CRLM were presumed to be identical. Local tumour progression (LTP) was assessed on postoperative surveillance imaging. Uni- and multivariable competing risk methods were used to compare LTP.

Results: In total 221 patients with 443 ablated tumours were analysed. A desmoplastic HGP was found in 60 (27.1%) patients who had a total of 159 (34.7%) ablated lesions. Five-year LTP [95%CI] was significantly higher for ablated CRLM with a presumed non-desmoplastic HGP (37% [30–43] vs 24% [17–32], Gray's-test p = 0.014). On multivariable analysis, a non-desmoplastic HGP (adjusted HR [95% CI]; 1.55 [1.03–2.35]) was independently associated with higher LTP rates after ablation.

Conclusion: HGP is an independent predictor of local tumour progression following ablation of CRLM.

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Introduction

Colorectal cancer (CRC) is one of the most prevalent solid malignancies in the world, with an estimated 1,8 million people affected each year.[1] CRC frequently metastasizes to the liver, with approximately 50% of patients developing colorectal liver metastases (CRLM).[2–4] There are multiple treatment modalities, such as chemotherapy, resection, and ablation which can prolong survival outcomes in patients with unresectable CRLM.[5] Local treatment with either ablation or surgical resection, however, offers the only potential for cure in patients with CRLM.[6] After surgical treatment for CRLM, 5-year survival rates vary widely between 20% and 70%.[6–11]

When compared to resection, ablation of CRLM is associated with lower complication rates.[12,13] On the other hand, survival outcomes (e.g. overall survival and disease free survival) after ablation may be inferior to surgical resection.[6,12,13] The impaired survival outcomes can possibly be explained by the higher rate of local disease progression.[14] Local regrowth occurs in 10%–40% of patients after ablation and in 4%–17% of patients after resection of CRLM.[9,10,14–19] It must be noted, however, that there is a substantial risk of selection bias as most of these studies compare hepatectomy for resectable CRLM with ablation for unresectable

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CRLM.[9,10,14–19] Several studies indicate that local tumour control is similar between ablation and resection when sufficient resection and ablation margins are reached.[20,21] A randomised controlled trial, comparing ablation and resection for upfront resectable CRLM, is currently running in the Netherlands to prove non-inferiority of thermal ablation compared to resection.[22] Few factors, such as ablation margin, lesion size, KRAS mutational status or predictive models combining these factors are available to predict local regrowth after ablation.[23–31] As ablation provides important benefits over resection in terms of complications, a better insight into the factors associated with local disease recurrence is needed.

Histopathological growth patterns (HGP) have been described as a robust and independent prognostic factor in patients with CRLM and may thus be useful in the search for better predictors of outcome after ablation.[32,33] HGPs are assessed on haematoxylin and eosin (H&E) stained tissue sections of resected CRLM and specifically describe the tumour to liver boundary.[33] Based on their morphology and prognostic impact HGPs are classified into a desmoplastic and a non-desmoplastic subtype.[32,34] CRLM with a desmoplastic HGP are separated from the liver tissue by a rim of desmoplastic tissue, without direct contact between cancer cells and hepatocytes.[33] This in contrast to non-desmoplastic HGP, in which tumour cells infiltrate the normal liver parenchyma.[33] Better survival outcomes and lower rates of positive resection margins are observed in patients with desmoplastic CRLM.[32,34,35] As such, a desmoplastic phenotype might also be associated with lower rates of local tumour progression (LTP) after ablation.[35]

This study therefore aims to analyse the potential of HGP as a predictive risk factor for LTP in patients undergoing ablation for CRLM.

Methods

Patient cohort

Patients who received simultaneous ablation and resection as first treatment for CRLM between January 2000 and January 2019 at the Erasmus MC Cancer Institute, Rotterdam, the Netherlands, were eligible for inclusion. HGPs were determined on H&E-stained tissue sections of resected CRLM. Patients for whom the HGP could not be determined were excluded, as were patients who did not complete potentially curative treatment, defined as the complete local treatment by either resection or ablation of all known (metastatic) disease at time of surgery. Patient information and clinicopathological data were obtained from a prospectively maintained database. Medical records and radiologic imaging were reviewed to obtain information on time to LTP and size of ablated CRLM. The current study was performed according to the REMARK guidelines and the reporting standards of ablation.[36,37] Institutional review was obtained from the medical ethics committee of the Erasmus University Medical Centre, which granted a waiver for (renewed) informed consent (MEC-2018-1743).

All patients received a treatment with curative intent, consisting of simultaneous ablation and resection, as it was considered to be the best choice of treatment at the time of surgery. The decision to perform ablation was made after consultation between the treating physicians and was mainly based on tumour size, number of tumours, remnant vital liver tissue, anatomical locations and patient age and condition.[6,38] The use of either radiofrequency (RFA) or microwave ablation (MWA) was left at the discretion of the treating physicians and was primarily dependent on location and number.[39] Lesions were assessed using intra-operative ultrasound, after which ablation was performed by a dedicated interventional radiologist together with the surgeon. The ablative time, power output, and type of electrode were determined by the interventional radiologist, based on tumour characteristics and tumour localisation. Overlapping ablation was used in certain patients to ensure an adequate tumour free margin of the ablation zone. In general, ablation is considered technically successful if ablative margins of at least 5 mm are achieved.[37] Although recent studies indicate that a minimal ablative margin of 10 mm should always be the procedural goal, as LTP is significantly reduced or even non-existent when these margins are achieved.[23,26,27,40] For the majority of patient in our cohort technical success was evaluated with the use of intraoperative ultrasound. Perioperative chemotherapy is not considered standard of care in the Netherlands, but pre-operative chemotherapy is administered to increase resectability and treatment options.[6,38,41] Clinical follow up is crucial for assessing technical effectiveness of the ablative procedure.[37] Immediate post-operative radiology evaluation, usually within 1 week after ablation, was performed to assess for technical effectiveness of ablation. Patients subsequently received computer tomography (CT) or magnetic resonance imaging (MRI) in line with both the ESMO and Dutch guidelines.[6,41]

Assessment of HGPs

HGPs were determined according to international consensus guidelines.[33] All available H&E-stained tissue sections of resected CRLM were assessed.[33] The entire interface between tumour and liver tissue was evaluated for each type of HGP, estimating its relative presence in percentage, since different HGPs can appear in conjunction. Patients were classified as desmoplastic only when all available slides of all resected CRLM displayed only the desmoplastic HGP (i.e. 100% desmoplastic, Fig. 1a), and as non-desmoplastic if any other type of HGP was observed in any slide of any resected CRLM (i.e. <100% desmoplastic, Fig. 1b).[32] Minimal intra- and intertumoural heterogeneity in HGP has been reported in patients with multiple CRLM, with 90–94% concordance between metastases.[42] Based on this high concordance, the HGP of ablated and resected CRLM, which were treated simultaneously, were assumed to be identical in this study. A similar methodology as previously described was used to determine the between metastases HGP concordance for the

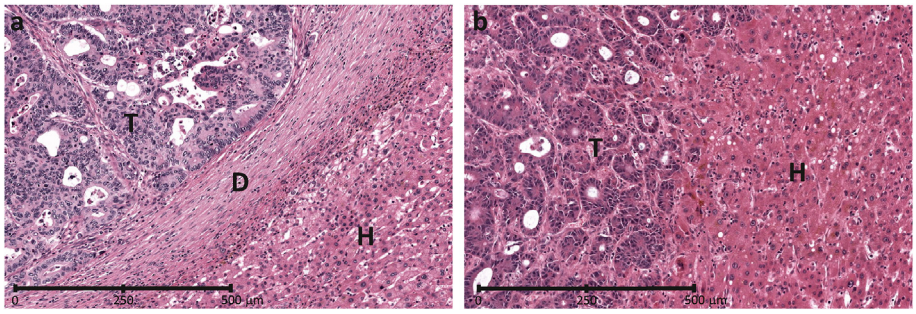

Figure 1 H&E stained tissue sections of resected colorectal liver metastases. Example of the desmoplastic (a) and non-desmoplastic histopathological growth pattern (b). *T* tumour, *D* desmoplastic rim, *H* hepatocytes

specific cohort of patients in this study, as it present a different cohort of those previously described.[42]

The HGP on tissue slide level, CRLM level, and patient level were considered to be either desmoplastic (i.e. 100% desmoplastic) or non-desmoplastic (i.e. <100% desmoplastic). For the between metastasis analysis, CRLM were considered to be concordant if CRLM HGP and patient HGP were identical. Between metastasis concordance was defined as the proportion of concordant CRLM.

Local tumour progression

The primary outcome of this study was LTP, which was calculated from the date of ablation to the date of first local regrowth on follow-up imaging. Local tumour progression was defined as tumour foci within or in direct contact (0 mm) with the post-ablation zone (Fig. 2). Analyses on LTP were performed on an individual lesion level, with sub-group analyses for tumour size and ablative technology used. Several reasons for censoring were considered. First, in case of no visible LTP on last available follow-up imaging. Second, if hepatic resection was performed removing the post-ablation zone. And last, if a new lesion originating elsewhere in the liver progressed into the post-ablation zone.

For those patients treated for LTP with surgical resection, the HGPs were additionally determined on the resection specimen and compared to the presumed HGP at first treatment of CRLM (i.e. concomitant resection and ablation).

Definitions

In this study, R1 resection was defined as tumour cells at the resection margin (0 mm). Primary tumour location was divided into right sided (caecum to splenic flexure), left-sided (splenic flexure to rectum) and rectum. The size of the ablated CRLM was measured in millimetres using last-available pre-operative imaging (i.e. post-chemotherapy when applicable), which is usually performed no longer than 6 week prior to surgical ablation. Vanishing metastases were defined as CRLM with a complete radiological response following pre-operative chemotherapy. For patients with post-chemotherapy vanishing metastases, anatomical landmarks and intraoperative ultrasound were used to achieve adequate ablation of all vanishing CRLM. Vanishing CRLM ablated by anatomical landmarks were excluded from analyses on individual lesion level, as it was unknown if there were any vital tumour cells at the time of ablation.

Statistical analysis

Continuous variables are presented as median with interquartile range (IQR), and categorical variables as absolute counts with corresponding percentages. Continuous variables were compared with the Kruskal–Wallis test, and categorical variables using the chi-squared test. Median follow-up for survivors was calculated using the reverse Kaplan–Meier method. Overall survival (OS) was estimated using Kaplan–Meier analysis and compared by means of the log rank test. Competing risk analyses on LTP were performed with death as a competing risk. The Gray's-test was used to compare cumulative incidence functions.[43] Uni- and multivariable Fine and Gray models were computed to investigate risk factors for LTP and to correct for potential confounding.[44] Size of CRLM ablated, pre-operative chemotherapy, and ablative technology used were considered to be potentially related to LTP following ablation of CRLM and were entered in the multivariable Fine and Gray models. Outcomes of the Fine and Gray regression analyses are reported as hazard ratio (HR) with corresponding 95% confidence interval (CI). No imputation of missing data was applied. All statistical tests were two-sided and p-values below 0.05 were considered statistically significant. All analysis were performed using R version 4.0.3 (http://cran.r-project.org) with dplyr, survival, survminer, cmprsk and tableone packages.

Results

Baseline characteristics

A total of 306 patients who received combined ablation and resection as surgical treatment for CRLM were evaluated in this study. Sixty-five (21.2%) did not complete potentially curative surgery and were excluded. For 19 (6.2%) patients the HGP could not be determined. One patient was lost to follow-up (Supplemental Fig. 1). In total 221 patients were considered eligible for analysis, 60 (27.1%) of which had desmoplastic CRLM (Table 1).

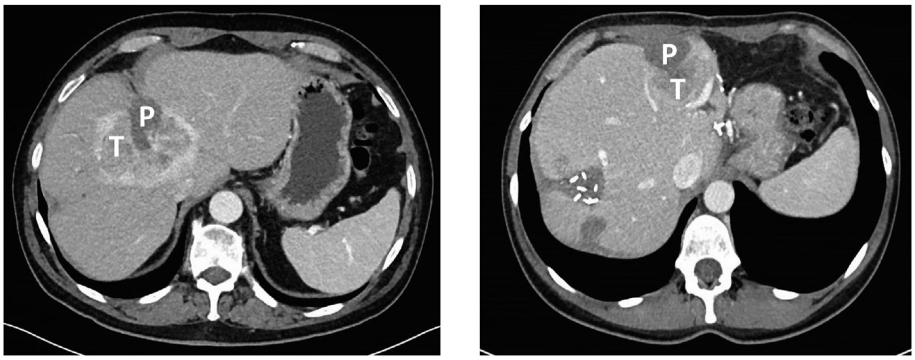

Figure 2 Local tumour progression within or in direct contact with the post-ablation zone. Examples of computed tomography follow-up imaging after combined resection and ablation for colorectal liver metastases. *P* post-ablation zone, *T* local tumour progression

Of these patients, 158 (71.5%) received pre-operative chemotherapy. Patient baseline characteristics stratified by HGP are provided in Table 1. Both the total number of CRLM and the number of ablated lesions did not differ significantly between patients with a desmoplastic and non-desmoplastic HGP. Positive resection margins (29.2% versus 13.3% p = 0.015) and extrahepatic disease (12.4% compared to 1.7%, p = 0.015) were more frequent in patients with a non-desmoplastic HGP (Table 1).

Four hundred fifty-eight lesions were ablated, of which 15 (3.3%) were considered vanishing metastases and were therefore excluded from further analyses. Amongst the 443 remaining ablated lesions, 149 (33.6%) were considered desmoplastic. The majority of ablated lesions were treated with RFA (N = 359, 81.0%). Ablated desmoplastic CRLM were more often treated with MWA (27.5% vs 14.6%, p = 0.001) and were slightly smaller than ablated non-desmoplastic lesions (median [IQR] of 1.0 [0.7–1.4] vs 1.1 [0.8–1.7], p = 0.01) (Table 2).

Between metastases concordance

Of all included patients, 153 (69.2%) underwent resection for multiple CRLM. Mean between metastases HGP concordance in these patients was 85,7%.

Overall survival

The median (IQR) follow-up for survivors was 80 (45–106) months, during which 84 (38.0%) patients developed LTP and 146 (66.1%) died. Eighty-four (38.0%) patients died without LTP, 65 (77.4%) with a non-desmoplastic and 19 (22.6%) with a desmoplastic HGP. Five years OS [95%CI] was inferior for patients with a non-desmoplastic compared to a desmoplastic HGP (44% [32–60] vs 30% [23–40], p = 0.044, Fig. 3a).

Local tumour progression

During follow-up, LTP was observed in 126 (28.4%) of the 443 ablated CRLM. Local tumour progression occurred in 31 (20.8%) of the 149 desmoplastic and in 95 (32.3%) of the 294 non-desmoplastic lesions. For all CRLM with observed LTP, median (IQR) time to local recurrence was 14 (8–27) months for desmoplastic versus 12 (7–20) months for non-desmoplastic lesions. When accounting for death as competing risk, the cumulative incidence of LTP for individual lesions was significantly lower for desmoplastic ablated CRLM (5-year [95%CI]: 24% [17–32] vs 37% [30–43], Gray's-test p = 0.014, Fig. 3b). On univariable analysis, increasing tumour size (adjusted HR [95% CI]; 1.70 [1.40–2.06], p < 0.001) and a non-desmoplastic HGP (adjusted HR [95%CI]; 1.65 [1.10–2.47], p = 0.015) were both associated with a higher rate of LTP after ablation of CRLM. On multivariable analysis, increasing tumour size (adjusted HR [95% CI]; 1.64 [1.34–2.00], p < 0.001) and a non-desmoplastic HGP (adjusted HR [95%CI]; 1.55 [1.03–2.35], p = 0.036) remained independent predictors for a higher rate of LTP (Table 3).

Sub-group analyses revealed similar effect size estimates (adjusted HR [95%CI]) for a non-desmoplastic HGP and LTP for lesions treated with RFA (1.46 [0.92–2.32]) and MWA (1.97 [0.74–5.26]) (Supplemental Table 1), although these did not reach statistical significance. Sub-group analyses for smaller lesions revealed similar associations (adjusted HR [95% CI]) between a non-desmoplastic HGP and LTP for lesions smaller than one (1.47 [0.68–3.15]) and two (1.56 [0.99–2.46]) centimetres only (Supplemental Table 2). Although statistical significance was not reached.

Change of HGP

One hundred twenty-six lesions showed LTP, of which 17 (13.5%) received re-ablation as treatment for LTP. A percutaneous approach was chosen for the majority of these lesions (N = 12, 70.6%) when compared to an open approach (N = 5, 29.4%). Surgical resection as treatment for LTP was used in 15 (11.7%) lesions, twelve with a presumed non-desmoplastic, and three with a presumed desmoplastic HGP of the first ablated CRLM. Amongst the presumed non-desmoplastic lesions, three lesions changed to a desmoplastic HGP after surgical resection of LTP. The majority of lesions remained non-desmoplastic. All presumed desmoplastic lesions changed to a non-desmoplastic HGP after surgical resection of LTP (Supplemental Table 3).

Table 1 Patient and tumour characteristics stratified by histopathological growth pattern

		Total n = 221	Desmoplastic n = 60 (27.1%)	Non-desmoplastic 161 (72.9%)	P-value
Age At Resection (median [IQR])		63.0 [56.0–69.0]	66.0 [55.8–71.0]	63.0 [56.0–68.0]	0.194
Gender	Male	142 (64.3%)	34 (56.7%)	108 (67.1%)	0.151
	Female	79 (35.7%)	26 (43.3%)	53 (32.9%)	
ASA class	ASA Class I–II	199 (90.0%)	54 (90%)	145 (90.1%)	0.989
	ASA Class > II	22 (10%)	6 (10%)	16 (9.9%)	
Location primary tumour	right-sided	45 (20.7%)	12 (20.7%)	33 (20.8%)	0.992
	left-sided	106 (48.8%)	28 (48.3%)	78 (49.1%)	
	rectum	66 (30.4%)	18 (31.0%)	48 (30.2%)	
	Missing	4 (1.8%)			
T-stage	pT0-2	31 (14.4%)	10 (17.2%)	21 (13.3%)	0.463
	pT3-4	185 (85.6%)	48 (82.8%)	137 (86.7%)	
	Missing	5 (2.3%)			
N-stage	N0	70 (32.4%)	17 (29.3%)	53 (33.5%)	0.556
	N+	146 (67.6%)	41 (70.7%)	105 (66.5%)	
	Missing	5 (2.3%)			
KRAS mutational status	Wild type	35 (59.3%)	8 (57.1%)	27 (60.0%)	0.849
	Mutant	24 (40.7%)	6 (42.9%)	18 (40.0%)	
	Missing	162 (73.3%)			
preoperative CEA - μg/l (median [IQR])		15.0 [4.4–51.4]	9.6 [4.0–38.5]	17.2 [4.8–55.9]	0.176
	Missing	22 (10%)			
preoperative chemotherapy	chemo-naive	63 (28.5%)	15 (25.0%)	48 (29.8%)	0.481
	pre-treated	158 (71.5%)	45 (75.0%)	113 (70.2%)	
resection margin	R0	166 (75.1%)	52 (86.7%)	114 (70.8%)	0.015
	R1	55 (24.9%)	8 (13.3%)	47 (29.2%)	
Extrahepatic disease	no	200 (90.5%)	59 (98.3%)	141 (87.6%)	0.015
	yes	21 (9.5%)	1 (1.7%)	20 (12.4%)	
RFA/MWA	RFA	196 (88.7%)	53 (88.3%)	143 (88.8%)	0.919
	MWA	25 (11.3%)	7 (11.7%)	18 (11.2%)	
Total lesions treated (median [IQR])		5.0 [3.0–7.0]	5.0 [3.0–7.0]	5.0 [3.0–7.0]	0.624
Total lesions ablated (median [IQR])		2.0 [1.0–3.0]	2.0 [1.0–4.0]	2.0 [1.0–2.0]	0.060
Diameter of largest CRLM (median [IQR])		2.8 [1.9–4.0]	2.2 [1.5–3.5]	3.0 [2.0–4.3]	0.006
	Missing	2 (0.9%)			
Diameter of largest ablated CRLM (median [IQR])		1.4 [1.0–2.0]	1.3 [1.0–1.9]	1.4 (1.0–2.0]	0.422
	<3 cm[a]	207 (95.4%)	59 (98.3%)	148 (94.3%)	0.201
	≥ 3 cm	10 (4.6%)	1 (1.7%)	9 (5.7%)	
	Missing	4 (1.8%)			

ASA American Society of Anaesthesiologists, *CEA* carcinoembryonic antigen, *CRLM* colorectal liver metastases, *IQR* interquartile range, *KRAS* Kirsten rat sarcoma viral oncogene, *MWA* microwave ablation, *RFA* radiofrequency ablation.
[a] Vanishing metastases included.

Discussion

To the best of our knowledge, this study is the first to assess the effect of HGP on local tumour control after ablation of CRLM. Local tumour progression was observed in 38% of patients which is on the high side when compared to the 10–40% reported in the current literature.[9,10,16–19,31,45] The high LTP rates could, in part, be explained by the fact that all patients received simultaneous ablation and resection, which implies that surgical resection alone was not possible or preferable due to poor anatomical location or a high metastatic tumour load. The consequence of

Table 2 Ablated lesions characteristics stratified by histopathological growth pattern

		Total n = 443	Desmoplastic n = 149 (33.6%)	Non-desmoplastic n = 294 (66.4%)	P-value
pre-operative chemotherapy	Yes	342 (77.2%)	120 (80.5%)	222 (75.5%)	0.233
	No	101 (22.8%)	29 (19.5%)	72 (24.5%)	
Type Ablation	RFA	359 (81.0%)	108 (72.5%)	251 (85.4%)	0.001
	MWA	84 (19.0%)	41 (27.5%)	43 (14.6%)	
Diameter of largest ablated CRLM (median [IQR])		1.0 [0.7–1.6]	1.0 [0.7–1.4]	1.1 [0.8–1.7]	0.01
	<3	420 (97.7%)	148 (99.3%)	272 (96.8%)	0.097
	≥3	10 (2.3%)	1 (0.7%)	9 (3.2%)	
	Missing	13 (2.9%)			

CRLM colorectal liver metastases, *IQR* interquartile range, *MWA* microwave ablation, *RFA* radiofrequency ablation.

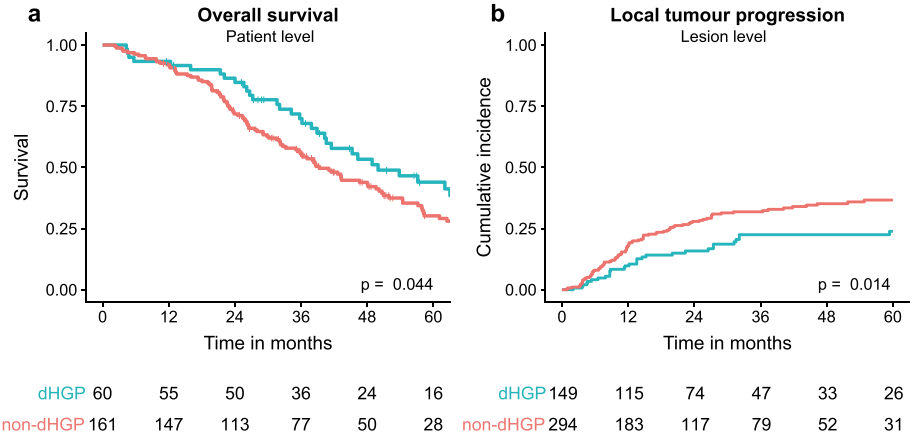

Figure 3 Overall survival curve (a) and cumulative incidence of local tumour progression per individual lesion (b) stratified by histopathological growth pattern. The p value represents the results of the overall log rank test and the Gray's-test for overall survival and local tumour progression, respectively. The number of at risk per point in time is provided in the table below. *dHGP* desmoplastic histopathological growth pattern, *non-dHGP* non-desmoplastic histopathological growth pattern

this is two-fold. First, the median (IQR) number of CRLM treated per patient in this patient subgroup was 5 (3–7), which is considerably higher when compared to the literature.[10,16,17,19,45] Second, as ablation was mainly performed as an auxiliary treatment modality for lesions not amenable for surgical resection and not as a primary treatment modality, the baseline risk for LTP may be higher when compared to most literature.[9,10,16–19,31,45] Lesions with a presumed non-desmoplastic HGP were found to be independently associated with a higher risk of LTP. This study therefore indicates that a non-desmoplastic HGP, as determined on concomitant resection specimens, may be associated with impaired local tumour control following ablation of CRLM.

Morphological differences between the desmoplastic and non-desmoplastic HGP could potentially explain these results. In CRLM with a desmoplastic phenotype, cancer cells are separated from the normal liver by a rim of fibrous tissue.[33] The large majority of non-desmoplastic HGP consists of the replacement subtype, which mimics the pre-existing liver parenchyma with cancer cells occupying the place of hepatocytes, resulting in an irregular tumour–liver interface and aggressive tumour behaviour.[32] The presence of a physical barrier between CRLM and normal liver in tumours exhibiting the desmoplastic HGP may result in greater contrast on intraoperative ultrasound imaging, enabling better margin assessment. These results are in line with the previous observed association between positive resection margins and a non-desmoplastic HGP.[35] In addition to this morphological difference, desmoplastic CRLM are vascularized by neo-angiogenesis, while CRLM with a non-desmoplastic HGP have the ability to co-opt the pre-existent hepatic sinusoidal blood vessels.[33,46] Given this difference in vascularization and the ability of non-desmoplastic tumour cells to occur within a little distance from metastases, the intact hepatic sinusoidal vessels of the liver parenchyma surrounding the post-ablation zone may hypothetically better facilitate non-desmoplastic rather than desmoplastic regrowth.[47]

Sub-group analyses in lesions treated with RFA and MWA revealed similar associations (i.e. comparable effect-size

Table 3 - Univariable and Multivariable Fine and Gray regression analysis for local tumour progression of individual ablated lesions

Variable	Univariable	P-value	Multivariable	P-value
	Hazard ratio [95% CI]		Hazard ratio [95% CI]	
Diameter of CRLM ablated (cont.) - cm	1.70 [1.40–2.06]	<0.001	1.64 [1.34–2.00]	<0.001
Preoperative chemotherapy - yes vs no	0.98 [0.64–1.49]	0.910	1.19 [0.77–1.85]	0.430
Ablation technique - MWA vs RFA	1.38 [0.90–2.12]	0.140	1.40 [0.90–2.19]	0.130
Non-desmoplastic HGP - yes vs no	1.65 [1.10–2.47]	0.015	1.55 [1.03–2.35]	0.036

Cont. continuous, *CRLM* colorectal liver metastases, *HGP* histopathological growth pattern, *LTP* local tumour progression, *MWA* microwave ablation, *RFA* radiofrequency ablation.

estimates) between non-desmoplastic HGP and a higher LTP risk, although these associations did not reach statistical significance. Given the reduced sample size for these sub-group analyses there is a high likelihood for a type 2 statistical error, as also evidenced from the wide confidence intervals. Taken together with our observation that the HGP was an independent predictor for LTP in the entire cohort when correcting for ablative technology used via multivariable regression, these results suggest that this effect is independent of ablative technology used. Similar results were seen when sub-analyzed for smaller lesions. Although not statistical significant, effect size of HGP for LTP (adjusted HR [95%CI]) was more or less similar for CRLM smaller than one (1.47 [0.68–3.15]) and two (1.56 [0.99–2.46] centimeters when compared to the analyses of the combined cohort (1.55 [1.03–2.35]). The high possibility for a type 2 statistical error, as evidenced by the considerably reduced sample size and the wide confidence intervals, combined with the observation that HGP was an independent predictor for LTP in the combined cohort when correcting for tumour size after multivariable analyses, suggest that the predictive value of HGP is independent of tumour size. Thus, it can be concluded that HGP is of potential predictive value for LTP for smaller lesions (less than one and two cm in size) also.

There were some baseline differences to be noted in this study. Ablated desmoplastic CRLM were more often treated with MWA compared to non-desmoplastic CRLM. The underlying reason for the difference in ablative modality used between the different types of HGP is unknown. Of the other clinicopathological risk factors assessed in our study, size of CRLM ablated was found to be a strong predictor for LTP after ablation of CRLM. These results are in line with previous studies reporting tumour size as a predictive risk factor for LTP after ablation of CRLM (HR 1.2–3.7).[23–25,27,40,48]

Absence of viable tumour cells on post-ablative biopsies can be used to assess complete ablation and have been described as a predictor for LTP after ablation of CRLM.[49,50] Ki-67 determination, Fluorescent tissue imaging, software assisted evaluation of the ablation zone and intraoperative spectroscopy have also been described as novel tools to predict successful ablation.[50–54] Unfortunately, these techniques were not applied in our retrospective cohort of patients. Total lesions ablated, primary tumour origin, prior hepatectomy, and chemotherapy have also been mentioned as predictors for LTP after ablation of CRLM.[18,23,25,40,54–58] Evidence regarding these factors is however inconclusive as these factors are rarely described or results are inconsistent.[18,23,25,40,48,55–59]

For HGPs to be implemented as potential decision criterion for ablation, reliable preoperative determination is necessary. Promising results from radiomics models applied to pre-operative imaging have been obtained, showing that such models may effectively predict different HGPs of CRLM on CT or MRI.[60–62] All these studies however used a 50% cut-off to categorise desmoplastic and non-desmoplastic HGP, which limits their applicability. This because recent studies have indicated that any percentage of non-desmoplastic HGP (i.e. <100% desmoplastic HGP) is associated with impaired survival outcomes, and that a 100% cut-off would therefore by more relevant from a clinical perspective.[32] In order to achieve such cut-off, future radiomics models have to be capable to detect even small areas of non-desmoplastic HGP. Further development of such radiomics approaches is therefore crucial for the implementation of HGPs in pre-treatment clinical decision making.

Ultrasound imaging may also be useful in the pre-operative determination of HGPs. It could be hypothesized that the presence of a physical barrier between CRLM and normal liver in tumours exhibiting the desmoplastic HGP results in greater contrast on intraoperative imaging. Future research should focus on the possible association between HGPs and ultrasound imaging. In order to do so, a prospective study in which patients receive an additional pre-operative ultrasound before surgical resection of CRLM would be advisable.

This study is limited in several ways. First, histopathological assessment of HGPs was not possible after ablation of CRLM as HGP determination requires vital cells at the tumour liver interface of a resection specimen.[33] For the purpose of this study, HGPs of ablated CRLM were determined on the assumption that all CRLM within patients exhibit a high between metastases concordance. Actual mean between metastases HGP concordance of resected CRLM in our cohort of patients was 85,7%, which is in line with the 90% found in a previous publication by Höppener et al., leaving us with a minor uncertainty regarding the actual HGPs of ablated CRLM.[42] Another key and important

limitation of this study was the lack of stratifying or assessing the impact of ablation margins on local tumour progression. Ablation margins, measured at the first post-ablative imaging, are one of the most important factors affecting local tumour control after ablation of CRLM.[26,27,30] The high variability between patients in time to first post-operative imaging and last pre-operative imaging and the subsequent risk of post-ablative tissue involution or change in tumour size, which could both affect the minimal ablation margin, prevented us from reliably measuring the minimal ablation margin for this cohort of patients.[63,64] Another limitation of our study was the high rate of pre-operative chemotherapy. The majority (71.3%) of patients in our cohort received pre-operative chemotherapy. This is relevant as there is evidence that suggests pre-operative chemotherapy alters the HGP of CRLM, resulting in a higher proportion of desmoplastic patients.[32] Last, the retrospective nature of this study may have predisposed for selection bias regarding KRAS, as mutational status was only determined to assess eligibility for anti-EGFR therapy within the palliative setting. Due to the high rate of missing data (73.3%) for KRAS mutational status and subsequent risk of selection bias, we were unable to analyse and correct for the potential predictive value of KRAS mutation on LTP after ablation of CRLM. A recent study, however, assessing the predictive value of HGP for survival after resection of CRLM in 780 patients, found that the prognosis was independent of KRAS mutation status.[34] Based on these results, it can be concluded that the predictive value of HGP for LTP would not have been affected significantly by KRAS mutational status.

In conclusion, this study suggests that a non-desmoplastic HGP is an independent predictor of LTP following ablation of CRLM.

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Conflicts of interest

None to declare.

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Appendix A. Supplementary data

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