Association Between Specific Mutations in KRAS Codon 12 and Colorectal Liver Metastasis

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IMPORTANCE Currently, one of the most commonly available biomarkers in the treatment of patients with colorectal liver metastases (CRLM) is the Kirsten rat sarcoma viral oncogene homolog (KRAS); however, the prognostic implications of specific mutations of the KRAS gene are still not well defined.

OBJECTIVE To investigate the prognostic impact of specific KRAS mutations on patients undergoing liver resection for CRLM.

DESIGN, SETTING, AND PARTICIPANTS This retrospective single-center study was conducted from January 1, 2003, to December 31, 2013. Data about specific KRAS mutations for 331 patients who underwent hepatic resection for CRLM at Johns Hopkins Hospital between 2003 and 2013 were analyzed. Clinicopathological characteristics, perioperative details, and outcomes were stratified by specific KRAS mutation at codons 12 and 13.

INTERVENTION Resection of CRLM.

MAIN OUTCOMES AND MEASURES Overall survival (OS) and recurrence-free survival.

RESULTS A mutated KRAS (mtKRAS) was identified in 91 patients (27.5%). At a median follow-up of 27.4 months, recurrence was observed in 48 patients (52.7%) with mtKRAS and 130 patients (54.2%) with wild-type KRAS (wtKRAS) (P = .82). Median and 5-year survival among patients with mtKRAS was 32.4 months and 32.7%, respectively, vs 58.5 months and 46.9%, respectively, for patients with wtKRAS (P = .02). Patients with KRAS codon 12 mutations had worse OS (hazard ratio [HR], 1.54; 95% CI, 1.05-2.27; P = .03) vs those with wtKRAS, whereas a KRAS codon 13 mutation was not associated with prognosis (HR, 1.47; 95% CI, 0.83-2.62; P = .19). Among the 6 most common mutations in codons 12 and 13, only G12V (HR, 1.78; 95% CI, 1.00-3.17; P = .05) and G12S (HR, 3.33; 95% CI, 1.22-9.10; P < .02) were associated with worse OS compared with patients with wtKRAS (both P < .05). Among patients who recurred, G12V (HR, 2.96; 95% CI, 1.32-6.61; P = .01), G12C (HR, 6.74; 95% CI, 2.05-22.2; P = .002), and G12S mutations (HR, 4.91; 95% CI, 1.52-15.8; P = .01) were associated with worse OS (both P < .05).

CONCLUSIONS AND RELEVANCE G12V and G12S mutations of codon 12 were independent prognostic factors of worse OS. Among patients who recurred after resection of CRLM, G12V, G12C, and G12S mutations were associated with worse OS. Information on specific KRAS mutations may help individualize therapeutic and surveillance strategies for patients with resected CRLM.
Surgical therapy, often combined with adjuvant systemic chemotherapy, is the best therapeutic option to treat patients with colorectal liver metastasis (CRLM), with postoperative 5-year survival ranging from 40% to 50%. However, while overall survival (OS) has improved, many patients with CRLM will recur and ultimately die of their disease. The factors used to predict outcome following surgical resection of CRLM largely focus on clinicopathological prognostic factors such as preoperative carcinoembryonic antigen (CEA) level, presentation of disease (ie, synchronous vs metachronous disease), disease-free interval between primary tumor and hepatic metastasis, and metastatic tumor number and size. There has been increasing interest in the use of biologic and molecular markers in the prognostic assessment of patients with metastatic colorectal cancer undergoing liver resection. Among patients with colorectal adenocarcinoma, mutated Kirsten rat sarcoma viral oncogene homolog (KRAS) is the most common oncogene of the RAS family, reported in up to 30% to 40% of patients. While the frequency and prognostic impact of KRAS mutation status have been described for both primary and metastatic colorectal cancer, to our knowledge, the role of specific mutations on KRAS codons remains undefined.

Most KRAS mutations are detected in codons 12 and 13, while mutations in codons 61 and 146 are less common. KRAS mutations in codons 12 and 13 include point mutations; the most common are codon 12 Gly → Asp (G12D), codon 12 Gly → Val (G12V), and codon 13 Gly → Asp (G13D) substitutions. Previous evidence has suggested that different biologic characteristics of specific KRAS mutations can lead to variations in epidermal growth factor receptor resistance. In addition, some investigators have suggested that specific KRAS mutations may also be associated with a more aggressive tumor phenotype in patients with unresectable stage IV metastatic colorectal cancer. However, the prognostic implication of different point mutations on survival of patients following curative intent liver resection for CRLM has not been previously investigated. As such, the purpose of the present study was to define the incidence of different specific KRAS mutations among patients with resected CRLM. Specifically, we sought to characterize the prognostic impact of different KRAS point mutations on recurrence and the survival of patients undergoing curative intent liver resection for CRLM.

Methods

Study Design

Patients who underwent curative intent liver resection for CRLM between January 2003 and August 2013 at Johns Hopkins Hospital with available KRAS data were identified from our institutional review board–approved institutional database. Patients reported to have tumors with KRAS mutations but with unknown specific mutations were excluded from the study. Patients who underwent only an ablative procedure without concurrent hepatic resection and patients who underwent incomplete palliative surgery (R2 resection) were excluded from analysis. Patients who received anti-epidermal growth factor receptor agents in the perioperative period were also excluded. The Johns Hopkins University institutional review board approved the study. No additional patient informed consent that was specific to this study was required given its retrospective nature.

Standard demographic and clinicopathologic data were collected on each patient including sex, age, disease status, tumor characteristics, operative details, perioperative status, type and time of chemotherapy, date of last follow-up, date and type of recurrence, and date of death. Primary tumor characteristics, including tumor location (colon vs rectum), American Joint Committee on Cancer T stage, tumor site (right vs left), and nodal status, were recorded. Number, size, distribution of the hepatic metastases, and presentation (synchronous vs metachronous) were also recorded. Tumor number and size were defined by the resection specimen. The largest lesion was used as the index lesion in the case of patients with multiple tumors. Information on treatment-related variables, such as type of procedure (resection vs resection plus ablation), type of hepatic resection, and margin status were also obtained. A major hepatectomy was defined as a resection of at least 3 Couinaud liver segments.

Data on KRAS mutational status were also recorded. Patients with a KRAS mutation were classified according to the specific KRAS mutation (G12A, G12C, G12D, G12V, G12S, and G13D).

Perioperative mortality was calculated based on the number of patients who died within 90 days of the operation. Long-term clinical outcomes were obtained including data on recurrence and OS at last follow-up. Recurrence was defined as the presence of a biopsy-proven tumor showing colorectal adenocarcinoma cells or a lesion deemed suspicious on follow-up computed tomographic imaging in the setting of an elevated CEA level.

**KRAS Mutation Analysis**

As previously described, the extracted DNA was evaluated for the presence of the most common mutations of the KRAS (codons 12 and 13) genes. These regions of interest were amplified using polymerase chain reaction and the reaction product underwent agarose gel electrophoresis against known positive and negative controls to assess the presence and size of the amplified product. The polymerase chain reaction protocols used were initial denaturation at 95°C for 5 minutes, followed by 40 cycles of amplification at 95°C for 40 seconds, 57°C for 40 seconds, and 72°C for 40 seconds and a final elongation at 72°C for 10 minutes. For amplification of the codon 12/13 region of the KRAS gene, the oligonucleotide primers used were 5′-TCATTATTATTATAAGGCCTGCTG-3′ (sense) and 5′-TTGGATCATATTGCCTCCAAA-3′ (antisense). The amplified products were subsequently column-purified using a GeneJET Polymerase Chain Reaction Purification Kit (Fermentas ThermoScientific) and then sequenced.

**Statistical Analysis**

Demographic, clinicopathologic, and perioperative features of the study population were stratified according to the specific KRAS mutation status. Summary statistics for the population were presented as totals and frequencies for categorical var-
bles or as median values with interquartile ranges (IQRs) for continuous variables. The differences between wild-type and mutant KRAS or between mutant KRAS groups were assessed by the $\chi^2$, $t$, and Mann-Whitney $U$ tests, as appropriate. Overall survival for the study population and recurrence-free survival (RFS) was estimated using the Kaplan-Meier method calculated from the date of surgery, and the differences in OS and RFS were assessed with the log-rank test. The Cox proportional hazards regression model was used to evaluate the association of relevant clinicopathologic variables with prognosis. Clinicopathologic variables of known prognostic importance, such as age, sex, T stage, location of primary tumor (colon or rectum), disease-free interval, regional lymph node status, size of largest liver metastasis, number of lesions, bilateral lesions, preoperative CEA levels, preoperative or adjuvant chemotherapy, use of ablation, and final resection margin status, were tested and a backward stepwise elimination with a threshold of $P = .20$ was used to select variables in the final multivariable analysis model. All analyses were carried out with Stata version 12.0 (StataCorp) and a $P$ value of less than .05 (2-tailed) was considered statistically significant.

### Results

#### Demographic, Clinicopathologic, and Perioperative Characteristics

A total of 331 patients who underwent curative intent liver resection for CRLM at Johns Hopkins Hospital and who met the inclusion criteria were identified. Baseline characteristics of the population, stratified for presence of KRAS mutation and type of mutation, are summarized in Table 1. Overall, the median patient age was 50 years (IQR, 42–61 years) and most patients were men ($n = 206$; 62.2%). Most patients had a pri-
mary colonic tumor (n = 241; 72.8%), while about one-fourth of patients (n = 90; 27.2%) had a primary rectal tumor. Most patients had T3-T4 colorectal tumors (n = 239; 72.2%) and nodal metastasis (n = 203; 61.3%). The median preoperative CEA level was 7.6 ng/mL (IQR, 3.1-27.0 ng/mL; to convert to micrograms per liter, multiply by 1.0). Most patients had a synchronous CRLM presentation (n = 184; 55.6%). Regarding the extent of CRLM, most patients (n = 241; 72.8%) had unilateral disease with an average tumor burden of 2 metastases (IQR, 1-3). The median size of the largest metastatic liver lesion was 2.5 cm (IQR, 1.8-3.8 cm). At the time of surgery, liver resection involved either a minor (n = 189; 57.1%) or major (n = 142; 42.9%) hepatectomy; 62 (18.7%) underwent a concurrent ablative procedure. Most patients (n = 256; 77.3%) received an R0 resection.

**KRAS Mutations**

Among the 331 patients evaluated, the overall incidence of any KRAS mutations was 27.5% (n = 91). No correlation was found between the presence of a KRAS mutation and any specific clinicopathologic characteristic (Table 1). Among all patients with CRLM (n = 331), KRAS codon 12 and 13 mutations were detected in 51 (15.5%); KRAS codon 12 mutations were detected in 67 patients (20.2%) and KRAS codon 13 mutations were detected in 24 patients (7.3%). Among patients with KRAS codon 12 mutations (n = 67), G12V was observed in 22 cases (32.8%), G12D was observed in 25 cases (37.3%), G12C was observed in 6 cases (9.0%), G12S in 7 cases (10.4%), and G12A in 4 cases (6.0%). Among patients with KRAS codon 13 mutations (n = 24), G13D was observed in 23 cases (95.8%). Various clinicopathologic characteristics were assessed and compared according to KRAS status (Table 1). The presence of KRAS codon 13 mutations was more common among men and patients with rectal cancer compared with patients who had KRAS codon 12 mutations (both P < .05). No other differences were observed between patients with KRAS codon 12 mutations vs KRAS codon 13 mutations.

**Recurrence-Free Survival**

At a median follow-up of 27.4 months, most patients (n = 178; 53.8%) developed a recurrence. The median RFS for the entire cohort was 20.9 months; 1-, 3-, and 5-year RFS were 65.9%, 34.4%, and 29.2%, respectively. Median and 5-year RFS among patients with mutated KRAS were 18.9 months and 30.6%, respectively, compared with 21.3 months and 33.2%, respectively, for patients with wild-type KRAS (wtKRAS) (P = .57). Median and 5-year RFS among patients with mutations in codon 12 were 22.0 months and 33.9%, respectively, compared with 18.9 months and 21.1%, respectively, for patients with codon 13 mutations (P = .45). Among the 6 most common KRAS codon 12 and 13 mutations, no mutation was associated with worse RFS compared with wtKRAS cases.

**Overall Survival**

Median OS for the entire cohort was 51.8 months; 1-, 3-, and 5-year OS were 93.3%, 61.4%, and 43.1%, respectively. Median and 5-year OS among patients with wtKRAS were 32.4 months and 32.7%, respectively, compared with 58.5 months and 46.9%, respectively, for patients with wtKRAS (P = .02) (eFigure in the Supplement).

Patients with KRAS mutations had worse OS relative to patients with wtKRAS (32.4 months vs 58.5 months, respectively; P = .02). Of note, 5-year OS of patients with KRAS codon 12 and 13 mutations were 34.4% and 29.2%, respectively, compared with 46.9% for patients who had wtKRAS (P = .05 for codon 12) (Figure 1). On both Kaplan-Meier analysis (log-rank P = .03) and Cox regression analysis (univariate hazard ratio [HR], 1.47; 95% CI, 1.05-2.27; P = .03 and multivariate HR, 1.70; 95% CI, 1.13-2.55; P = .01; Table 2) patients with KRAS codon 12 mutations had a worse OS compared with patients with wtKRAS. In contrast, KRAS codon 13 mutations were not associated with a worse prognosis compared with wtKRAS (HR, 1.47; 95% CI, 0.83-2.62) (Table 2). On univariable analysis, in addition to tumor and operative factors, such as primary tumor nodal metastasis (HR, 1.80; 95% CI, 1.24-2.62; P < .001), concurrent use of ablation (HR, 1.54; 95% CI, 1.08-2.20; P = .02), and R1 surgical margin status (HR, 1.55; 95% CI, 1.09-2.21; P = .02), codon 12 KRAS mutation (HR, 1.54; 95% CI, 1.05-2.27; P = .03) was associated with a worse long-term survival (all P < .05) (Table 2). On multivariable analysis, after controlling for other competing risk factors, codon 12 KRAS muta-
Mutations (HR, 1.7; 95% CI, 1.13-2.55; \( P = .01 \)) remained independently associated with a worse OS (Table 2).

On further analysis of the 6 most common \( \text{KRAS} \) codon 12 and 13 mutations, the \( G12V \) and \( G12S \) mutations were the point mutations most associated with worse long-term prognosis. Specifically, \( G12V \) (\( n = 22 \)) (HR, 1.78; 95% CI, 1.00-3.17; \( P = .05 \)) and \( G12S \) (\( n = 7 \)) (HR, 3.33; 95% CI, 1.22-9.10; \( P = .02 \)) mutations were associated with a roughly 2- to 3-fold increased risk for long-term death compared with wt \( \text{KRAS} \) (Figure 2; Table 3). In addition, in the subgroup of patients who recurred after the curative intent liver resection, the presence of the \( \text{KRAS} \) mutations \( G12V \) (HR, 2.96; 95% CI, 1.32-6.61; \( P = .01 \)), \( G12S \) (HR, 4.91; 95% CI, 1.52-15.8; \( P = .01 \)), and \( G12C \) (HR, 6.74; 95% CI, 2.05-22.2; \( P = .002 \)) were associated with an increased risk for death after recurrence compared with patients with wt \( \text{KRAS} \) who recurred (Table 3).

### Discussion

Various scoring systems that include different clinicopathologic variables have been proposed to stratify patient prognosis after hepatic resection for CRLM.\(^{25,26}\) However, owing to inconsistent predictive power and lack of reproducibility, these scoring systems are becoming less relevant in clinical practice.\(^{27}\) Attention has turned to the use of biological and molecular markers to predict the long-term outcome of patients undergoing surgery for CRLM.\(^4\) Currently, one of the most commonly available biomarkers in the treatment of patients with CRLM is \( \text{KRAS} \). The frequency and prognostic impact of \( \text{KRAS} \) mutation status have been described in both primary and metastatic colorectal cancer\(^4\); however, the prognostic implications of specific mutations of the \( \text{KRAS} \) gene are still not well defined. Given this, we sought to evaluate the impact of different \( \text{KRAS} \) point mutations on recurrence and survival of patients undergoing curative intent liver resection for CRLM. To our knowledge, this is the first study to evaluate the prognostic effects of different point mutations in \( \text{KRAS} \) codons among patients who have undergone CRLM resection. Using a comprehensive analysis of 331 patients who underwent liver resection for CRLM, we reported worse OS among patients with \( \text{KRAS} \) codon 12 mutations and, specifically, those patients with either a \( \text{KRAS} \) \( G12V \) or a \( G12S \) mutation. In contrast, patients with a \( \text{KRAS} \) codon 13 mutation did not have a worse prognosis. These findings suggest that \( \text{KRAS} \) mutation status has prognostic implications. More importantly, stratification of patients undergoing liver resection for CRLM based on specific \( \text{KRAS} \) mutations may be even more informative with regard to long-term outcome.

### Table 2. Univariable and Multivariable Cox Proportional Hazard Analysis for Overall Survival

<table>
<thead>
<tr>
<th>Prognostic Factor</th>
<th>Hazard Ratio (95% CI)</th>
<th>Univariable P Value</th>
<th>Multivariable P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>( \text{KRAS} )</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Wild type</td>
<td>1 [Reference]</td>
<td>1 [Reference]</td>
<td></td>
</tr>
<tr>
<td>All codon 12 mutants</td>
<td>1.54 (1.05-2.27)</td>
<td>.03</td>
<td>1.7 (1.13-2.55)</td>
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<tr>
<td>All codon 13 mutants</td>
<td>1.47 (0.83-2.62)</td>
<td>.19</td>
<td>1.61 (0.87-2.97)</td>
</tr>
<tr>
<td>Age</td>
<td>1.01 (0.99-1.02)</td>
<td>.38</td>
<td>...</td>
</tr>
<tr>
<td>Female</td>
<td>1.11 (0.80-1.54)</td>
<td>.52</td>
<td>...</td>
</tr>
<tr>
<td>AJCC 7th edition T stage</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>T1/T2</td>
<td>1 [Reference]</td>
<td>1 [Reference]</td>
<td></td>
</tr>
<tr>
<td>T3/T4</td>
<td>1 (0.62-1.60)</td>
<td>.99</td>
<td>...</td>
</tr>
<tr>
<td>Location of tumor</td>
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<td></td>
<td></td>
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<tr>
<td>Colon</td>
<td>1 [Reference]</td>
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<td></td>
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<tr>
<td>Rectal</td>
<td>1.08 (0.76-1.52)</td>
<td>.67</td>
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<tr>
<td>Disease-free interval &lt;12 mo</td>
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<td>Regional lymph node status</td>
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<td>Negative</td>
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<tr>
<td>Positive</td>
<td>1.8 (1.24-2.62)</td>
<td>&lt;.001</td>
<td>2.06 (1.39-3.04)</td>
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<td>Tumor size</td>
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<tr>
<td>No. of lesions</td>
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<td>.09</td>
<td>...</td>
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<td>Bilobar lesions</td>
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<td>CEA ≥ 30 ng/mL</td>
<td>1.00 (1.00-1.00)</td>
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<tr>
<td>Chemotherapy</td>
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<tr>
<td>Preoperative</td>
<td>1.19 (0.82-1.74)</td>
<td>.36</td>
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<tr>
<td>Adjuvant</td>
<td>0.84 (0.61-1.17)</td>
<td>.30</td>
<td>...</td>
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<tr>
<td>Ablation</td>
<td>1.54 (1.08-2.20)</td>
<td>.02</td>
<td>1.81 (1.25-2.62)</td>
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<td>1 [Reference]</td>
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<tr>
<td>R1</td>
<td>1.55 (1.09-2.21)</td>
<td>.02</td>
<td>1.73 (1.17-2.57)</td>
</tr>
</tbody>
</table>

Abbreviations: AJCC, American Joint Committee on Cancer; CEA, carcinoembryonic antigen; NA, not applicable; ellipses, not included in the multivariable analysis.
Similar to previous studies reporting an incidence of \textit{KRAS} mutations ranging from 35% to 45%,\textsuperscript{5,28-30} we found \textit{KRAS} mutations in 27.5% of tumor specimens from patients undergoing hepatic resection for CRLM. \textit{KRAS} codon 12 and 13 mutations were detected in about one-third of patients undergoing resection of CRLM, with \textit{KRAS} codon 12 mutations being more common (20.2%) than \textit{KRAS} codon 13 mutations (7.3%). Among patients with \textit{KRAS} codon 12 mutations, G12V and G12D were noted in about one-quarter of specimens, while mutations in G12C, G12S, and G12A were much less common, occurring in less than 10% of patients. Among those patients who did have a \textit{KRAS} codon 13 mutation, G13D was the most commonly observed mutation, occurring in about 30% of patients. Interestingly, the incidence of different \textit{KRAS} codon mutations noted in the current study was consistent with previous data reported by Thierry and colleagues.\textsuperscript{31} Specifically, Thierry et al\textsuperscript{31} reported on specific \textit{KRAS} point mutations among patients with metastatic colorectal cancer and noted G12V (20.5%), G12D (25.6%), and G13D (28.2%) as the most common point mutations in metastatic colorectal cancer, whereas G12A (7.7%), G12C (5.1%), and G12S (12.8%) were less common. Similar frequencies of \textit{KRAS} mutations have been reported in the Sanger COSMIC database: G12V (21.9%-24.4%), G12D (33.5%-34.4%), G13D (18.9%-19.2%), G12A (6.2%-6.6%), G12C (7.9%), and G12S (4.9%-5.7%).\textsuperscript{32}

In the present study, we found that patients with \textit{KRAS} codon 12 mutations had a roughly 70% increased risk for long-term death compared with patients with wt\textit{KRAS} (HR, 1.70; 95% CI, 1.13-2.55; \(P = .01\)). Specifically, among the 6 most common \textit{KRAS} codon 12 and 13 mutations, G12V (n = 22) and G12S (n = 7) mutations were particularly associated with higher mortality (G12V: HR, 1.78; 95% CI, 1.00-3.17; \(P = .05\) and G12S: HR, 3.33; 95% CI, 1.22-9.10; \(P = .02\)). In addition, in the subgroup of patients who recurred after curative intent liver resection, the effect of \textit{KRAS} mutation was even more pronounced among patients with G12V (HR, 2.96; 95% CI, 1.32-6.61; \(P = .01\)), G12C (HR, 6.74; 95% CI, 2.05-22.2; \(P = .002\)), and G12S (HR, 4.91; 95% CI, 1.52-15.8; \(P = .01\)) mutations. These findings are consistent with those by Imamura et al,\textsuperscript{33} who showed that \textit{KRAS} codon 12 mutations and, especially, G12V are associated with a worse OS compared with wt\textit{KRAS}. In fact, the authors suggested that G12V was associated with a more aggressive tumor phenotype in metastatic colorectal cancer in general. The potential differential effects of specific \textit{KRAS} mutations on clinical outcomes have also been demonstrated in a large multicenter study of 2,721 patients with metastatic colorectal cancer.\textsuperscript{5} Specifically, Andreyev et al\textsuperscript{5} reported that only the G12V point mutation was a predictive factor of worse OS among patients with advanced colorectal cancer, suggesting a more aggressive behavior of tumors with G12V mutation.\textsuperscript{5} The aggressive behavior of the G12V mutation has been explained by Al-Mulla et al,\textsuperscript{33} who showed that the G12V mutation produces proteins that behave differently than other mutated \textit{KRAS} proteins. The G12V mutation leads to a reduction of GTPase activity and affinity for GTPase-activating proteins, which in turn prevents their activation.\textsuperscript{33,34} Moreover, Guerrero et al\textsuperscript{35}...
reported that KRAS codon 12 mutations confer a more aggressive tumor phenotype than codon 13 mutations by an alteration in the threshold for apoptosis induction. Further experimental data suggest that, among the different KRAS codon 12 mutations, G12V mutation is characterized by more potent transforming ability than the others and is associated with a more aggressive biologic phenotype.36

The current study had several limitations that should be considered when interpreting the findings. As with all retrospective studies, undoubtedly there was some selection bias. Although fairly representative as evidenced by the specific mutation incidences, the sample size was relatively small for some groups. Although patients who received perioperative anti–epidermal growth factor receptor agents were excluded from the study, a remote use of those agents cannot be definitely excluded. Finally, we performed KRAS testing for somatic mutations involving only codons 12 and 13 and we did not investigate for less common mutations involving codons 61 and 146.15,37

Conclusions

KRAS codon 12 mutations, and specifically G12V and G12S mutations, were associated with worse prognosis after resection of CRLM, especially among those patients who experienced a recurrence following surgery. Specific mutations in codon 12, such as G12V and G12S, may be helpful in improving clinical decision-making and prognostic models based on both well-known clinicopathological characteristics and molecular features. In addition, our data demonstrate that different mutations, even in a single gene, can shape distinctive biologic behaviors, further supporting the unique tumor principle.38,39

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Author Contributions: Dr Pawlik had full access to all of the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis. Drs Margonis and Kim contributed equally to this article. Study concept and design: Margonis, Kim, Spolverato, Gupta, Cosgrove, Anders, Choti, Pawlik. Acquisition, analysis, or interpretation of data: Margonis, Kim, Spolverato, Ejaz, Gupta, Karagkounis, Choti.

DRAFTING of the manuscript: Margonis, Spolverato, Gupta.

Critical revision of the manuscript for important intellectual content: Margonis, Kim, Spolverato, Ejaz, Cosgrove, Anders, Karagkounis, Choti, Pawlik. Statistical analysis: Kim, Spolverato, Ejaz. Administrative, technical, or material support: Spolverato, Gupta, Cosgrove, Karagkounis, Pawlik. Study supervision: Anders, Choti, Pawlik.

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REFERENCES


Mutations in KRAS Codon 12 and Colorectal Liver Metastasis

Des Winter, MD

Invited Commentary

Colorectal Liver Metastasis Resection Outcomes Defined by Molecular Biology

Determination of tumor molecular biology is going to advance cancer care within a decade. Margonis and colleagues¹ have shown how. As found in primary cancer, the prognosis of patients undergoing resection of colorectal liver metastases is associated with specific gene mutations of the tumors. The genes in question are in the Kirsten rat sarcoma viral oncogene homologue family (ie, KRAS on the short arm of chromosome 12) that code for cell division regulatory pathways via guanosine triphosphate and mitogen-activated protein kinase. As its name suggests, the pathway was originally mapped in another tumor type but somatic (rather than inherited) mutations in the KRAS genes are found in many human tumors.

We know a lot more about these genes since their characterization in the early 1980s. Just under half of colorectal cancers express altered KRAS, subclassification of which includes exon mutations 1 (codons 12 and 13) examined here.¹ There are interactions with other genetically coded cellular factors including glucose metabolism (glucose transporter 1), microribonucleic acids (eg, let-7), protein kinases (protein kinase c), and apoptosis regulators (Bcl-xl) that result in the epigenetic and physiological phenomena causing heterogeneous tumor phenotypes. For example, KRAS mutated colorectal cancer cells are less likely to respond to anti-epidermal growth factor antibody (eg, cetuximab and panitumumab) treatments. It is a limitation of the current study that patients who were known to have received anti-epidermal growth factor therapy were excluded. It would have been even more useful to know their fate vis-à-vis KRAS mutation analysis.

Despite a greater understanding of molecular biological processes, our therapeutic options in metastatic colorectal cancer remain somewhat limited. The primacy of solid tumor excision remains the keystone of colon and rectal cancer care but approximately half of patients will have hepatic secondaries. Although never submitted to randomized trial, a criticism often levelled at liver resection, it is hard to argue against the improved survival observed following successful surgery. A trial would be useful, sure. But tax payers’ dollars are better spent on molecular developments that define the processes by which patient outcomes are determined. Optimal adjuvant care can be better only through enquiry, evidence, and evolution. As shown here, surgeons continue to champion the art of precision at the interface between scalpel and science.

REFERENCE

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