Pathogenic somatic mutation (SM) of mismatch repair (MMR) genes and associations with microsatellite instability (MSI), tumor mutational burden (TMB) and SM in other DNA repair pathways in 24,223 tumor genomic profiles

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*Caris Life Sciences, Phoenix, AZ*
Mismatch repair (MMR) and Lynch syndrome (LS)

**TAKEAWAYS**

MLH1, MSH2, MSH6, PMS2 are the clinically relevant MMR genes

MLH1/PMS2 & MSH2/MSH6 function as heterodimers

Deficient MMR (dMMR) leads to microsatellite instability

LS is the autosomal dominant inheritance of a MMR gene mutation
Interactions between DNA repair pathways

1. MMR
2. HR
3. BER
4. NER
5. NHEJ
6. POL

DNA DAMAGE SIGNALING (cell cycle arrest or cell death)

CANCER

Adapted from:
- Arora et. al., Cancer Biol. & Therp., 2017
- Nicolas et. al., Gene, 2016

Presented by: Bodor, JN

Presented at: 2018 ASCO Annual Meeting
Mono- (mSM) and bi-allelic (bSM) somatic mutations causing deficient MMR (dMMR)

- bSM in MLH1, MSH2, MSH6 or PMS2 is a clinically relevant cause of dMMR
- Primary cause of tumor dMMR (1.9%) after germline MMR mutation (2.8%) and MLH1 promoter methylation (11.9%)

**dMMR is found in histologically diverse tumors**

<table>
<thead>
<tr>
<th>Tumor type</th>
<th>MSI-H rate (%)</th>
<th>Sample size</th>
</tr>
</thead>
<tbody>
<tr>
<td>Uterine</td>
<td>14.1</td>
<td>39/277</td>
</tr>
<tr>
<td>Small Bowel</td>
<td>8.6</td>
<td>6/70</td>
</tr>
<tr>
<td>Prostate</td>
<td>6.2</td>
<td>11/178</td>
</tr>
<tr>
<td>Colorectal</td>
<td>3.5</td>
<td>42/1185</td>
</tr>
<tr>
<td>Cancer of Unknown Primary</td>
<td>2.7</td>
<td>22/815</td>
</tr>
<tr>
<td>Hepatobiliary</td>
<td>2.3</td>
<td>9/389</td>
</tr>
<tr>
<td>Gastroesophageal</td>
<td>1.5</td>
<td>6/400</td>
</tr>
<tr>
<td>Neuroendocrine</td>
<td>0.2</td>
<td>1/431</td>
</tr>
<tr>
<td>Pancreas</td>
<td>0.2</td>
<td>1/459</td>
</tr>
<tr>
<td>NSCLC</td>
<td>0.2</td>
<td>5/2112</td>
</tr>
<tr>
<td>Breast</td>
<td>0.1</td>
<td>2/1459</td>
</tr>
</tbody>
</table>

- MSI-H was identified in nearly every tumor histology, many non-Lynch syndrome spectrum tumors.
- Unclear how MSI related to high tumor mutational burden, but mutational patterns seen.
- Could MSI and/or MMR mutations result from mutations in non-MMR DNA repair pathways?

Hall MJ et al. JCO 34, no. 15_suppl (May 20 2016) 1523-1523.
Study Objectives

- Determine the frequency of mSM and bSM in the MMR genes (MLH1, MSH2, MSH6, PMS2) in a large sample of histologically diverse tumors undergoing commercial tumor genomic testing.

- Associate these with the MSI-H phenotype, TMB, and SM in other DNA repair pathways.

- Associate these with tumor histology and CRC sidedness (L vs R).
Methods

Sample for analyses

- The Caris Life Sciences database was queried for tumors with ≥ 1 SM of an MMR gene (N= 24,223). Convenience sample of tumors undergoing tumor genomic testing.
- Tumors tested with a NGS multi-gene panel (Illumina NextSeq 592 gene panel).

Variables of interest and statistical approach

- Likely pathogenic/pathogenic mSM or bSM in an MMR gene (MLH1, MSH2, MSH6, PMS2).
- MSI, TMB (high ≥17 mut/Mb), tumor histology/location, non-MMR DNA repair pathway mutation.
- Statistics: Associations tested by Fisher’s exact test and logistic regression.
Prevalence and frequency of bSM and mSM

24,223 tumor genomic profiles

470 tumors with ≥ 1 SM

1.94%

bSM in a MMR gene
n = 80

0.33%

bSM in MLH1
n = 7

bSM in MSH2
n = 22

bSM in MSH6
n = 49

bSM in PMS2
n = 2

Mean age
mSM 61.8 yrs
bSM 58.1 yrs

p < 0.01

mSM in a MMR gene
n = 390

1.61%

MSH6 F1088fs
Frequency of MMR mSM by tumor histology

Percent Distribution

- Breast (14), 4%
- CRC (90), 23%
- Endocrine (6), 2%
- Upper GI (19), 5%
- GU (19), 5%
- Lung (45), 12%
- Ovarian (23), 6%
- Pancreabil (14), 4%
- Prostate (8), 2%
- Other (38), 10%
- Sarcoma (4), 1%
- Skin (12), 3%
- Small bowel (14), 4%
- Uterine (84), 22%

Tumor Type (N), %

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- Other (38), 10%
- Sarcoma (4), 1%
- Skin (12), 3%
- Small bowel (14), 4%
- Uterine (84), 22%
Frequency of MMR bSM by tumor histology

**Percent Distribution**

- Uterine (34), 43%
- CRC (21), 26%
- Upper GI (2), 3%
- Ovarian (14), 18%
- Pancreobil (3), 4%
- Other (4), 5%
- Small bowel (2), 3%

**Tumor Type (N), %**

- CRC (21), 26%
- Upper GI (2), 3%
- Ovarian (14), 18%
- Pancreobil (3), 4%
- Other (4), 5%
- Small bowel (2), 3%
- Uterine (34), 43%

**Percent bSM by CRC Site**

- Right: 12.5%
- Left: 30.3%
- Unspecified: 20.0%

**P = 0.102**
Association of MMR mSM with high TMB

Percent with high TMB

<table>
<thead>
<tr>
<th>Gene</th>
<th>Wild Type</th>
<th>Mutated</th>
</tr>
</thead>
<tbody>
<tr>
<td>MLH1</td>
<td>68.2%</td>
<td>70.5%</td>
</tr>
<tr>
<td>MSH2</td>
<td>65.0%</td>
<td>84.4%</td>
</tr>
<tr>
<td>MSH6</td>
<td>62.0%</td>
<td>74.5%</td>
</tr>
<tr>
<td>PMS2</td>
<td>72.9%</td>
<td>36.0%</td>
</tr>
</tbody>
</table>

P < 0.01  P < 0.05  P < 0.001
Association of MMR mSM with MSI-H

Percent with MSI-H

<table>
<thead>
<tr>
<th>Gene</th>
<th>Wild Type</th>
<th>Mutated</th>
</tr>
</thead>
<tbody>
<tr>
<td>MLH1</td>
<td>58.1%</td>
<td>68.5%</td>
</tr>
<tr>
<td>MSH2</td>
<td>59.7%</td>
<td>67.7%</td>
</tr>
<tr>
<td>MSH6</td>
<td>55.5%</td>
<td>65.6%</td>
</tr>
<tr>
<td>PMS2</td>
<td>63.6%</td>
<td>40.0%</td>
</tr>
</tbody>
</table>

P < 0.001
P < 0.01

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Association of MMR bSM with TMB and MSI

TMB

Percent bSM

P < 0.001

MSI

Percent bSM

P < 0.001

bSM

Low Intermediate High

TMB

% 0% 3.2% 8.3% 22.5%

0% 5% 10% 15% 20% 25% 30%

MSI

Stable Equivocal High

% 0% 6.6% 15.4% 23.4%

0% 5% 10% 15% 20% 25% 30%
Association of MMR bSM with SM in HR and NER DNA Repair Pathways

SM in HR and NER DNA Repair

<table>
<thead>
<tr>
<th></th>
<th>HR</th>
<th>NER</th>
</tr>
</thead>
<tbody>
<tr>
<td>SM Yes</td>
<td>24.4%</td>
<td>24.5%</td>
</tr>
<tr>
<td>SM No</td>
<td>14.0%</td>
<td>13.2%</td>
</tr>
</tbody>
</table>

P < 0.01

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### Associations between MMR bSM and SM in HR and NER DNA Repair

<table>
<thead>
<tr>
<th>MMR genes</th>
<th>OR</th>
<th>(95 % CI)</th>
<th>OR</th>
<th>(95 % CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td><strong>HR</strong></td>
<td></td>
<td><strong>NER</strong></td>
<td></td>
</tr>
<tr>
<td><strong>bSM vs. no mutations</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MLH1</td>
<td>4.21</td>
<td>(1.49 – 11.84)**</td>
<td>2.06</td>
<td>(0.77 – 5.53)</td>
</tr>
<tr>
<td>MSH2</td>
<td>10.30</td>
<td>(4.13 – 25.65)**</td>
<td>2.24</td>
<td>(0.96 – 5.23)</td>
</tr>
<tr>
<td>MSH6</td>
<td>3.82</td>
<td>(1.75 – 8.33)**</td>
<td>2.97</td>
<td>(1.40 – 6.26)**</td>
</tr>
<tr>
<td>PMS2</td>
<td>0.55</td>
<td>(0.10 – 3.14)</td>
<td>0.49</td>
<td>(0.11 – 2.24)</td>
</tr>
</tbody>
</table>
Study Limitations

- No paired germline data therefore unknown if the somatic mutations observed are also found in germline, though a number of the mutations identified here have been reported in germline series.

- bSM were assumed to be in trans as the likelihood of two pathogenic mutations on the same MMR gene allele is very low.

- Loss of heterozygosity studies not conducted.

- No individual-level family history data or treatment data.
Summary

- MMR mSM are found in a diverse range of tumor histologies.

- MMR bSM occur more often in younger patients and primarily in Lynch syndrome spectrum tumors, suggesting:
  - Germline MMR mutations precede many bSM and/or
  - Some organs are more vulnerable to develop bSM in the setting of mSM

- MMR mSM and bSM are associated with high TMB, MSI-H, and mutations in non-MMR DNA repair pathways (i.e. HR and NER DNA repair pathways).

- Findings suggest both interactive and cascade effects of DNA repair pathways, which may elucidate tumor biology and new treatment targets.