Sidedness matters: Surrogate biomarkers prognosticate early colorectal cancer upon anatomic location

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Background

- Recent evidence indicates that the anatomic location of primary tumors across the colon correlates with survival in the metastatic setting, whereas left-sided tumors may exhibit superior survival compared with right-sided ones.

**Definition of primary tumor location**

- Transverse colon
- Hepatic flexure
- Ascending colon
- Cecum
- Appendix
- Sigmoid colon
- Descending colon
- Splenic flexure
- Rectum

Right-sided tumors

Left-sided tumors
Embryonal-derived disparity

Right-sided (proximal) colon cancer
- Common in women
- Microsatellite instability
- Derived from MLH1 gene
- Maltase, BAPR, MMR mutations
- MAPK signaling, serrated pathway
- Mutagenic CYP52a metabolites
- HNPCC

Left-sided (distal) colon cancer
- Common in men
- Chromosomal instability
- Derived from hamartoma
- APC, k-ras, DCC, p53 mutations
- EGFR signaling, Wnt signaling
- HER1, HER2 amplification
- FAP

Adapted from Kim et al., World J Gastroenterology 201

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Presented by: Irit Ben-Aharon MD, PhD
Stage II colon cancer – the challenge

- At presentation 25% of colon cancer cases are diagnosed as stage II, for which surgical resection remains the mainstay of treatment.
- The role of adjuvant chemotherapy remains controversial.
- Several prognostic tools have been developed and validated; their role in clinical practice remains to be elucidated.
The 12-Gene Oncotype DX® Colon Cancer

- The colon cancer Recurrence Score result is based upon the quantitative expression of seven cancer genes identified as consistently and significantly associated with recurrence-free interval (RFI) in 1,851 patients.
- These seven cancer genes include the cell cycle group (Ki-67, MYBL2, C-MYC), the stromal group (FAP, INHBA, BGN) and GADD45B.
The 12-Gene Oncotype DX® Colon Cancer

- The pre-specified Recurrence Score gene panel result was validated in 1,436 Stage II colon cancer patients with tissue from the QUASAR trial, and in two other clinical studies.
CDX2

- Human colon cancer stem cells are characterized by high expression levels of ALCAM on the cell surface.

- Tumors enriched in cells with an undifferentiated, stem-like phenotype might exhibit more aggressive clinical behavior.

- Tumors lacking CDX2 expression were characterized by high levels of ALCAM expression, high pathological grade, as well as a more aggressive natural history.

Dalerba et al, NEJM 2016
CDX2

- In independent discovery and validation datasets, patients with CDX2-negative tumors had a significantly higher risk of recurrence than patients with CDX2-positive tumors.
- Patients with CDX2-negative tumors seem to benefit from adjuvant chemotherapy, which was noted not only in stage III, but also in stage II disease.

Dalerba et al, NEJM 2016
Study objectives

- To evaluate whether the 12-Gene Oncotype DX score and/or CDX2 status correlate with primary tumor location, and whether tumor location may reflect differential prognosis in stage II CRC.

- To evaluate whether the pattern of potential correlation applies also for stage III CRC.
Methods

- The 12-gene assay has been commercially available since 2010 and has been reimbursed by Clalit Health Services (CHS) in Israel since 1/2011 for T3 mismatch repair proficient (MMR-P) stage II average risk CRC.

- Within the frame of a clinical trial, the assay was also prospectively performed in a small cohort of stage III CRC patients.

- Retrospective analysis of a cohort of patients with T3 MMR-P stage II CRC for whom the 12-gene assay was prospectively performed (1/2011-8/2016) and a subgroup of stage III CRC patients.

- The histopathological report was reviewed for precise primary tumor location.
# Results

<table>
<thead>
<tr>
<th></th>
<th>Right Sided</th>
<th>Left Sided</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Gender</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>276 (50%)</td>
<td>333 (56%)</td>
</tr>
<tr>
<td>Female</td>
<td>275 (50%)</td>
<td>263 (44%)</td>
</tr>
<tr>
<td><strong>Age</strong> Median</td>
<td>72 (40-90)</td>
<td>68 (31-86)</td>
</tr>
<tr>
<td><strong>Stage</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>II</td>
<td>551 (48%)</td>
<td>596 (52%)</td>
</tr>
<tr>
<td>III</td>
<td>60</td>
<td>72</td>
</tr>
<tr>
<td>II - Rectal</td>
<td></td>
<td>78</td>
</tr>
<tr>
<td><strong>Anatomic location</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cecum</td>
<td>95 (17.2%)</td>
<td></td>
</tr>
<tr>
<td>Hepatic Flexure</td>
<td>38 (6.8%)</td>
<td></td>
</tr>
<tr>
<td>Right side (Not specified)</td>
<td>335 (60.7%)</td>
<td>73 (11.6%)</td>
</tr>
<tr>
<td>Transverse</td>
<td>83 (15.3%)</td>
<td></td>
</tr>
<tr>
<td><strong>Anatomic location</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Splenic Flexure</td>
<td></td>
<td>53 (8.8%)</td>
</tr>
<tr>
<td>Sigmoid</td>
<td></td>
<td>306 (51.3%)</td>
</tr>
<tr>
<td>Rectosigmoid</td>
<td></td>
<td>103 (17.5%)</td>
</tr>
<tr>
<td>Left side (Not specified)</td>
<td>134 (22.4%)</td>
<td>78 (12.5%)</td>
</tr>
<tr>
<td>Rectum</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Presented at: 2017 Gastrointestinal Cancers Symposium | #GI17
Presented by: Irit Ben-Aharon MD, PhD*
Methods – Cont.

- CDX2 expression was reviewed for patients of this cohort who were diagnosed in 2016 (as of when CDX2 immunostaining was routinely performed).

- Recurrence score (RS) and CDX2 expression were correlated with primary tumor location.

- Rectal tumors were analyzed separately.
Patient and sample accounting

All patients of CHS for whom Oncotype DX colon was performed  
n=1370

MMR-P Stage II  
n=1159

Stage III  
n=133

Stage II - Rectal cancer  
n=78

Eligible for Primary Analysis  
Stage II n= 1147  
Stage III n= 132

CDX2 Eligible for Primary Analysis  
Stage II n= 109

Excluded  
n=13

* Excluded since pathological report was not available

Presented by: Irit Ben-Aharon MD, PhD
Results – Stage II

- Recurrence score was higher in right-sided tumors compared with left-sided tumors.

<table>
<thead>
<tr>
<th></th>
<th>N</th>
<th>Score (Mean)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Right side</td>
<td>551 (48.03%)</td>
<td>27.72 Range (6-71)</td>
</tr>
<tr>
<td>Left side</td>
<td>596 (51.97%)</td>
<td>25.79 Range (6-54)</td>
</tr>
<tr>
<td>Total</td>
<td>1147 (100%)</td>
<td>p-0.002</td>
</tr>
</tbody>
</table>
Results – Stage II

- Recurrence score gradually decreased across the colon (cecum - highest score, sigmoid-lowest score).

<table>
<thead>
<tr>
<th></th>
<th>Score (Mean)</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>N</td>
<td>Score</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cecum</td>
<td>95</td>
<td>29.75</td>
<td>Range (8-71)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hepatic flexure</td>
<td>38</td>
<td>27.76</td>
<td>Range (7-57)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sigmoid</td>
<td>306</td>
<td>24.49</td>
<td>Range (0-52)</td>
<td></td>
<td>p-0.014</td>
</tr>
</tbody>
</table>
CDX2

- Right-sided tumors exhibited more CDX2-negative tumors compared with left-sided tumors.

<table>
<thead>
<tr>
<th></th>
<th>Right side</th>
<th>Left side</th>
</tr>
</thead>
<tbody>
<tr>
<td>CDX2 pos</td>
<td>34 (64.2%)</td>
<td>47 (83.9%)</td>
</tr>
<tr>
<td>CDX2 neg</td>
<td><strong>19 (35.8%)</strong></td>
<td>9 (16.1%)</td>
</tr>
<tr>
<td>Total (N=109)</td>
<td>53</td>
<td>56</td>
</tr>
<tr>
<td></td>
<td>p-0.029</td>
<td></td>
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</tbody>
</table>
Results – Stage III

- Recurrence score was higher in right-sided tumors compared with left-sided tumors, and higher than in stage II tumors.

<table>
<thead>
<tr>
<th></th>
<th>N</th>
<th>Score (Mean)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Right side</td>
<td>60 (45.4%)</td>
<td>31.15 Range (3-63)</td>
</tr>
<tr>
<td>Left side</td>
<td>72 (54.6%)</td>
<td>24.6 Range (7-52)</td>
</tr>
<tr>
<td>Total</td>
<td>132 (100%)</td>
<td>p-0.001</td>
</tr>
</tbody>
</table>

Presented by: Irit Ben-Aharon MD, PhD
Conclusions

- Our study indicates that right-sided colorectal tumors may display worse prognosis compared with left-sided tumors in MMR-P stage II CRC upon these two prognostic tools.

- CDX2 lack of expression is correlated with a higher Oncotype score.

- Further recurrence analysis should be performed to indicate whether primary tumor location may actually serve as a **prognostic factor**.

- Further studies are warranted to confirm the role of tumor location as a predictive factor to adjuvant treatment.
Limitations

- CDX2 negative tumors represent a larger fraction than in the NEJM cohort.
- The biological correlation between the Oncotype gene panel and CDX2 expression has not yet been established.
- No recurrence/survival data (most included cases were diagnosed in 2014-2016)

Strengths

- Large number of cases, MMR-P, average risk stage II.
- Confirms the role of CDX2 expression and its correlation to tumor location.
CDX2 and Oncotype score

- CDX2-negative tumors had a higher Oncotype DX score.

<table>
<thead>
<tr>
<th></th>
<th>Oncotype Score (Mean)</th>
<th>SD</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>CDX2 pos</td>
<td>24.42</td>
<td>10.30</td>
<td></td>
</tr>
<tr>
<td>CDX2 neg</td>
<td>32.00</td>
<td>12.686</td>
<td>0.020</td>
</tr>
</tbody>
</table>
Results – Rectal cancer

- Recurrence scores for stage II and stage III rectal cancer were higher compared with left-sided tumors.

<table>
<thead>
<tr>
<th></th>
<th>N</th>
<th>Score (Mean)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stage II Left Colon</td>
<td>596</td>
<td>25.79</td>
</tr>
<tr>
<td>Stage II Rectal</td>
<td>78</td>
<td>27.06*</td>
</tr>
<tr>
<td>Stage III Left Colon</td>
<td>72</td>
<td>24.6</td>
</tr>
<tr>
<td>Stage III Rectal</td>
<td>14</td>
<td>27.15**</td>
</tr>
</tbody>
</table>

*p-0.04, **p-0.05
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Hillel Yaffe Medical Center, Hadera
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