Genes in genome guidance, mismatch repair

Thanh-Trang Vo

Cancer Biology

1/24/2013
Progression of Research

Boland RC & Goel A. Gastroenterology 2010 138:2073
Role of Mismatch Repair

Normal Errors in Replication/Environmental Damage

- Base pairing mismatch ➔ point mutation
- Slippage of DNA ➔ microsatellite instability
Role of Mismatch Repair

Normal Errors in Replication/Environmental Damage

- Base pairing mismatch ➔ point mutation
- Slippage of DNA ➔ microsatellite instability
Role of Mismatch Repair

Key function of MMR

- Recognize mismatch or loop
- Recognize daughter strand
- Recruit excise wrong sequence EXO1
- Recruit DNA polymerase to synthesize new DNA
Role of Mismatch Repair

Key function of MMR

- Recognize mismatch or loop
- Recognize daughter strand
- Recruit excise wrong sequence EXO1
- Recruit DNA polymerase to synthesize new DNA
Clinical MMR

MMR deficiency
• Inherited – Lynch syndrome (hereditary non-polyposis colorectal carcinoma HNPCC)
• MLH1 promoter hypermethylation

Cancers Incidence
• Cancers including ovarian, endometrial, gastric, colon and other cancers
• 15-17% primary colon cancer patients MLH1 mutation
• MSH2 mutations cause cancers that are not of the colon
MMR role in progression of evolution of colon cancer

• MMR defect is associated more with β-catenin mutations instead of APC mutations; most cancers with β-catenin mutations don’t have APC mutation

• MMR with APC mutations have that are different from MMR competent cancers in that is has more alterations in repeat sequences
Colon cancers with MSI

Cancers with MSI (microsatellite instability)

• More likely to develop in the proximal colon
• More likely to appear poorly differentiated
• Less likely to be invasive
• Less likely to have mutations in KRAS and p53
• Found in younger patients than colon cancers without MSI
• Often heriditary
Detecting MSI in MMR Defective Colon Cancers

Shift in PCR product of genes with repeat sequences
N = Normal colon
T = Tumor

Boland RC & Goel A. *Gastroenterology* 2010 138:2073
Clinical MMR

MMR deficiency
- Inherited – Lynch syndrome (hereditary non-polyposis colorectal carcinoma HNPCC)
- MLH1 promoter hypermethylation

Cancers Incidence
- Cancers including ovarian, endometrial, gastric, colon and other cancers
- 15-17% primary colon cancer patients MLH1 mutation
- MSH2 mutations cause cancers that are not of the colon
Lynch Syndrome

Characteristics
• Develop cancer at an early age than other patients (20 to 30 years old)
• Germline genes found to be mutated are MSH2, MLH1, MSH6 and PMS2
• Some instead of mutations have germline promoter methylation of MMR genes
• Frequently have multiple tumors: colon, rectum, endometrium, stomach, ovary, urinary tract, small intestine, etc.
• No increase in incidence of breast, lung or prostate cancer
• Better overall survival: 5 year survival with Lynch syndrome is 65% versus 44% for patients with sporadic colon cancer older than 65 yrs
• Diagnosed with lower disease state and less metastasis
• Amsterdam Criteria for familial study
  1. 1st degree relative
  2. Colon cancer occurs in at least 2 generations
  3. Affected members are younger than 50 years of age
Clinical MMR

MMR deficiency
• Inherited – Lynch syndrome (hereditary non-polyposis colorectal carcinoma HNPCC)
  • MLH1 promoter hypermethylation

Cancers Incidence
• Cancers including ovarian, endometrial, gastric, colon and other cancers
• 15-17% primary colon cancer patients MSH1 mutation
• MSH2 mutations cause cancers that are not of the colon
Hypermethylation of MLH1

In 1997 Kane et al. showed that MLH1 is silenced by biallelic methylation of the promoter in MSI colon cancers

• Results in less MLH1 protein and associated PMS2
  – PMS2 is rapidly degraded without stabilizing MLH1 protein
• Tumors are diploid (74%)
• Have better prognosis than patients with non-MSI tumors
• Patients develop cancer at older age than patients with Lynch syndrome
• Loss of MLH1 expression increases with age (gene is lost in ~50% of colon cancer patients who are older than 90 years of age)
• Demethylation agents like 5-azacitidine restores MLH1 and PMS2 expression, restores sensitivity to 5-FU
Prevalence of MMR in Colon Cancer

15% colon cancers have MSI
- 75-80% acquired methylation of MLH1
- 2-3% have germline mutation (Lynch syndrome)
## Genes Affected by MSI

### Cell proliferation genes
- GRAB1
- TCF-4
- WISP3
- Activin receptor-2
- Insulin-like growth factor-2 receptor
- Axin-2
- CDX

### Cell Cycle/Apoptosis genes
- BAX
- Caspase-5
- RIZ
- BCL-10
- PTEN
- hG4-1
- FAS

### DNA Repair genes
- MBD-4
- BLM
- CHK1
- MLH3

### MMR genes (except MLH1)
<table>
<thead>
<tr>
<th>Microsatellite length</th>
<th>Gene</th>
</tr>
</thead>
<tbody>
<tr>
<td>A10</td>
<td>AIM2</td>
</tr>
<tr>
<td></td>
<td>CASPASE-5</td>
</tr>
<tr>
<td></td>
<td>MBD-4</td>
</tr>
<tr>
<td></td>
<td>OGT</td>
</tr>
<tr>
<td></td>
<td>SEC63 (also, A9)</td>
</tr>
<tr>
<td></td>
<td>TGFβ1R2</td>
</tr>
<tr>
<td>A9</td>
<td>BLM</td>
</tr>
<tr>
<td></td>
<td>CHK1</td>
</tr>
<tr>
<td></td>
<td>GRB-14</td>
</tr>
<tr>
<td></td>
<td>MLH3</td>
</tr>
<tr>
<td></td>
<td>RAD50</td>
</tr>
<tr>
<td></td>
<td>RHAMM</td>
</tr>
<tr>
<td></td>
<td>RIZ (also, A8)</td>
</tr>
<tr>
<td></td>
<td>TCF-4</td>
</tr>
<tr>
<td></td>
<td>WISP3</td>
</tr>
<tr>
<td>T7</td>
<td>FAS</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Microsatellite length</th>
<th>Gene</th>
</tr>
</thead>
<tbody>
<tr>
<td>A8</td>
<td>ACVRII</td>
</tr>
<tr>
<td></td>
<td>APAF</td>
</tr>
<tr>
<td></td>
<td>BCL-10</td>
</tr>
<tr>
<td></td>
<td>hG4-1</td>
</tr>
<tr>
<td></td>
<td>MSH3</td>
</tr>
<tr>
<td>A6</td>
<td>PTEN (2 A6’s)</td>
</tr>
<tr>
<td>T10</td>
<td>OGT</td>
</tr>
<tr>
<td>T9</td>
<td>KIAA0971</td>
</tr>
<tr>
<td></td>
<td>NADH-UOB</td>
</tr>
<tr>
<td>G8</td>
<td>BAX</td>
</tr>
<tr>
<td></td>
<td>IGF2R</td>
</tr>
<tr>
<td>C9</td>
<td>SLC23A1</td>
</tr>
<tr>
<td>C8</td>
<td>MSH6</td>
</tr>
<tr>
<td>G7</td>
<td>AXIN-2 (A6, A6, C6)</td>
</tr>
<tr>
<td></td>
<td>CDX2</td>
</tr>
</tbody>
</table>

Data from Duval and Hamelin.
Colon Cancer
Tumorigenesis in MMR-deficient Cells

Rajagopalan et al Nat Rev 2003 3;695
Colon Tumorigenesis in MMR Defective Cancers

Normal APC expression but mutant b-catenin (cannot interact with APC) or downstream TCF-4
Colon Cancer
Tumorigenesis in MMR-deficient Cells

Rajagopalan et al Nat Rev 2003 3;695
Colon Tumorigenesis in MMR Defective Cancers

Lynch syndrome = KRAS mutation
Sporadic tumors with MSI = BRAF mutation
Targeting MMR Defective Tumors

Synthetic lethal approaches

**MSH2 deficiency**
- Methotrexate
- Psoralen
- DNA polymerase beta inhibition
- PTEN-induced putative kinase I (PINK1)

**MSH1 deficiency**
- DNA polymerase gamma
- PTEN-induced putative kinase I (PINK1)
References

Boland RC & Goel A. Gastroenterology 2010 138:2073
Martin SA et al Clin Cancer Res 2010;16:5107
Rajagopalan et al Nat Rev 2003 3;695