Defective DNA Mismatch Repair and Endometrial Carcinogenesis

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Objectives

• Review current state of knowledge regarding causes and consequence of defective DNA mismatch repair in endometrial cancer

• Identify biologically and clinically important questions that should be a priority for future studies

• Stimulate cross disciplinary discussion

• Avoid redundancy with presentations made by Drs. Levine, Goodman, Brinton, Hunter, Lu and others
DNA Mismatch Repair in Endometrial Cancers

• High prevalence (20-30% of tumors) lose MMR
  – cellular recessive (loss of function defects)
  – accumulation of strand slippage and distinct tumor phenotype
    referred to as microsatellite instability (MSI)

• Most common in endometrioid carcinomas (~33%)

• Causally associated with endometrial tumorigenesis
  – germline mutations in MLH1, MSH2, MSH6 and PMS2 confer greatly
    increased risk for endometrial cancer
Questions

• Why are mismatch repair defects less common in nonendometrioid histologies?

• Why does MSH6 mutations confer high risk for endometrial cancer and low risk for colon cancer
DNA Mismatch Repair in Endometrial Cancers

• Somatic inactivation of mismatch repair accounts for most cases
  – epigenetic silencing of MLH1

• MLH1 methylation is associated with clinicopathologic features
  – tumor grade
  – older age at diagnosis
  – lower BMI

• Methylation is associated with cis variation (rs1800734)
Questions

- Why are the key genetic and environmental factors leading to MLH1 methylation, loss of MMR and the mutator phenotype?
- What are the mechanisms?

Meyer, Broaddus and Lu, *Cancer Control* 2009
Clinical Significance of MMR Abnormalities

• Tumor defects, clinical and family history data can point to inherited disease  
  – cancer prevention

• Whereas MSI is a predictive and prognostic marker in colorectal cancers, studies in endometrial cancer have shown association with improved outcomes, poor outcomes or that there are no differences in outcomes for women whose tumors have MSI or have normal mismatch repair
Selection for Loss of DNA Mismatch Repair

• Confers a mutator phenotype that drives genotypic and phenotypic progression (Loeb 2001)

• Non-mutator effects: disruption of processes essential to normal cell growth

Fishel, Cancer Res 2001
Mutator Phenotype

• ~100X somatic mutation rate
  – model organisms
  – reporter systems
  – large scale genomic efforts

Heinen, *Current Drug Targets* 2014
Endometrial Cancer Mutator Phenotype

- low number of copy number changes
- distinct pattern of gene mutations (37 significantly mutate genes)
- distinctive gene expression profile

Integrated genomic characterization of endometrial carcinoma

We performed an integrated genomic, transcriptomic and proteomic characterization of 373 endometrial carcinomas using array- and sequencing-based technologies. Uterine serous tumours and 25% of high-grade endometrioid tumours had extensive copy number alterations, few DNA methylation changes, low oestrogen receptor/progesterone receptor levels, and frequent TP53 mutations. Most endometrioid tumours had few copy number alterations or TP53 mutations, but frequent mutations in PTEN, CIN8B1, PIK3CA, ARID1A and KRAS and novel mutations in the SWI/SNF chromatin remodelling complex gene ARID5B. A subset of endometrioid tumours that we identified had a markedly increased transversion mutation frequency and newly identified hotspot mutations in POLE. Our results classified endometrial cancers into four categories: POLE ultramutated, microsatellite instability hypermutated, copy-number low, and copy-number high. Uterine serous carcinomas share genomic features with ovarian serous and basal-like breast carcinomas. We demonstrated that the genomic features of endometrial carcinomas permit a reclassification that may affect post-surgical adjuvant treatment for women with aggressive tumours.
MSI Target Genes
(where is the meat?)

• MMR defects in colorectal cancer are associated with mutations in “driver” genes
  – strand slippage mutation in TGFRB2 are common in MSI+ colorectal cancers
  – TGFRB2 mutations seen in tumors with normal MMR (MSS)
  – germline mutation associated with risk for colorectal cancer (Markowitz et al, 1995; Lu et al, 1998)

• MMR “target genes” in endometrial cancer remain elusive
Unbiased Large Scaled Genomic Approaches

• TCGA data mined to detect mutations in MMR defects in endometrial and colorectal cancers (30 and 27 MSI+ cases respectively) and compared to MSS cases

- tumor type specificity
- elevated ratio of frameshift-to-inframe mutations
- lower transcript levels for genes with mutations
- mutations more common euchromatic and intronic regions
- depletion of MSI at nucleosome-occupied sequences
Candidate Endometrial MSI Target Genes

- Endometrial cancer genes identified
  - JAK1
  - TFAM (EC exclusive)
  - PDS5B
  - others
CTCF is an Endometrial Cancer MSI Target

- strand slippage mutations present in 25% of MSI+ tumors
  - first identified in a small cohort of tumor subjected to whole exome sequencing and validated by Sanger methods
• other CTCF mutations seen at high frequency and the strand slippage mutations are likely deleterious

– CTCF mutations seen in tumors with normal MMR (MSS)

– frameshift mutations are associated with nonsense mediated decay
Current Approaches Have Limitations
(endometrial cancer deserves more attention)

- False positives and false negatives

- Where is CTCF?
- AIM2 is mutated at high frequency in endometrial cancers (47%)
• discovery of an MSI target gene does not mean it is a driver...and not all drivers will be clinically relevant

• discovery efforts may not be sufficiently powered to address key questions; phenotypes not fully appreciated; environmental factors not adequately considered
Wrap Up Questions
(the un-summary)

• What are the genetic and environmental factors that contribute to somatic inactivation of mismatch repair? How do they interact?
• Can MMR deficiencies be prevented or the molecular evolution to cancer be avoided?
• What are the genes/pathways in MSI+ endometrial cancers that matter (treatment/prevention/biology)
• Do DNA mismatch repair defect matter clinically other than in the context of Lynch syndrome?