Microsatellite Instability and BRAF Mutations in Colon Cancer: Populations study

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Financial Relationships Disclosure

• I have no financial relationships, with commercial entities that produce or market products or devices related to the content of this CME activity, to disclose.
Populations Studies, Why?

- Population-based studies allow the understanding of how gene variants and gene-environment interactions contribute to disease development.
- The resulting knowledge is to be integrated into public health research programs.
- Such research might lead to more effective and targeted medical and public health interventions.
Colon Cancer

• Accounts for 13% of all cancer related deaths

• Incidence of and mortality from colon cancer in AAs are higher than in general population

• Causes for such differences are multiple: genetic, environmental and socio-economical.
Colorectal Cancer

Sporadic (average risk 65%–85%)

Family history (10%–30%)

Hereditary nonpolyposis colorectal cancer (HNPCC) (5%)

Familial adenomatous polyposis (FAP) (1%)

Rare syndromes (<0.1%)
How does CRC develop?

- Chromosomal instability (CIN) that can be studied by LOH or aCGH

- DNA Methylation and Histones modification that constitute epigenetic changes that might participate in the development of cancer

- Genes’ instability (MIN or MSI) where genes sequences are not correctly maintained (mutations)
Genes in Cancer

- **Oncogenes**: Genes of which the function participate in cell proliferation (e.g. *BRAF*).
- **Tumor Suppressor genes**: Genes of which the function is anti-proliferative (e.g. *APC*).
- The balance between the above genes functions maintain tissue **homeostasis**.
- **Caretaker genes**: Genes of which the function is not directly linked to cell division but create an environment prone to carcinogenesis if their function is altered (e.g. DNA MMR genes).
Why is it important to study the MSI phenotype?

• The DNA mismatch repair (MMR) pathway is an important post-replicative repair process. It is involved in the maintenance of genomic stability.

• Loss of MMR function prevents the correction of replicative errors leading to instability at the genome that can be detected by polymorphisms in microsatellites sequences scattered all over the genome.
Why is it important to study the MSI phenotype? (ctd.)

• Correlation between MMR repair status and clinicopathological parameters of the tumor is of value in predicting tumor behavior and response to chemotherapy.

• Chemotherapy being an integral part of multimodal therapy for locally advanced cancers and predicting response may help in tailoring regimens in patients for optimum response.
Hereditary Nonpolyposis Colorectal Cancer

Mismatch Repair

TCGAC
AGCTG

Mismatch error

TCTAC
AGCTG

TCGAC
AGCTG

MMR

MMR

TCATAC
AGCTG
Example of MSI in ABI3130
Overall survival curves of patients with MSS and MSI
Materials and Methods

- Colon cancer samples from HU-JH (AAs, n=95) and patients from Omani (African Descent, n=61) and Iranian (Caucasians, n=53) Hospitals
- MSI was checked using a panel of 5 markers: BAT25, BAT26, NR21, NR22 and NR24
- MSP was used to check the methylation status of p16 and MLH1 genes
- IHC was used to check the expression level of MLH1 and MSH2 genes
- A PCR fragment from BRAF gene was sequenced for V600E point mutation detection
Results with the African American Patients

- In a 95 patients sample, 30% were found to be MSI-H, of which 71.5% were proximal
- The mean age was 65.7 years
- Defect in expression as shown by IHC was found in 68% of cases for MLH1 and 29% for MSH2
- BRAF mutation was detected in 9.7% of cases, were predominantly proximal and 43% of these mutants express MLH1
- Only 20% of MSI-H carry the BRAF mutation
- MLH1 displayed methylation in 94% of cases

Brim et al, Molecular Cancer, 2008
Results with Iranian Patients

• In a 53 patients sample, 26% were found to be MSI-H, of which 79% were proximal
• The mean age was 59.8 years
• Defect in expression as shown by IHC was found in 77% of cases for MLH1 and 8% for MSH2
• BRAF mutation was detected in 7% of MSI-H cases and 2% total cases
• MLH1 displayed methylation in 71% of MSI-H cases.

Brim et al, Molecular Cancer, 2008
Results with the Omani Patients

- In a 61 patients of African Descent (AD), only 13% were MSI-H of which 12.5% were proximal
- The mean age of 52.7 years
- Defect in expression as shown by IHC was found in 37.5% of cases for MLH1 and 25% for MSH2
- BRAF mutation was detected in 18.6% of cases, none of which was proximal and 81% of these mutants express MLH1
- 25% of MSI-H carry the BRAF mutation
- MLH1 displayed methylation in 83.6% of cases

Ashktorab, Brim et al 2008: Dig. Dis. and Science, 2008
Conclusions

• CRC occurs at a younger age in Omani and Iranian patients where there is a lower occurrence of MSI-H tumors compared to AAs patients.
• There is an important role of MLH1 expression in these populations.
• BRAf mutations play a major role in the MSI-H tumors carcinogenesis in AA and Omani patients but not similarly in the iranian patients.
• The high MSI-H occurrence in AAs patients might have significant implications in cancer treatment design for these patients.
The way to go:

• High throughput technologies: Genetic (SNPs genome wide), Epigenetic (methylome and Histone code) and Genomic (genome structure: aCGH) are necessary to dig deeper in population specifics for CRC development.
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