Results Summary
- 1260 subjects with a Lynch-associated cancer referred for clinical Lynch testing
- 9% with Lynch mutation
- 12.3% with ≥1 mutation on 25-gene panel
- 3.4% with a non-Lynch mutation
- 34% with ≥1 VUS in a non-Lynch gene
- 28% of mutation carriers had mutations in non-Lynch cancer susceptibility genes
- 54% of non-Lynch mutations in high-penetrance genes
- 10% of mutation carriers had BRCA1/2 mutations
- Clinically appear more “Lynch-like” than “BRCA-like”

Strengths/Limitations
- Strengths
  - Large cohort of consecutive individuals
  - Representative sample of patients referred for clinical Lynch testing
- Limitations
  - Clinical data obtained via clinician report
  - Unable to verify accuracy or completeness
  - No data on other non-Lynch genetic testing done clinically
  - No data on tumor testing (MSI, mismatch repair IHC)
  - Do the identified mutations explain clinical phenotype?

Conclusions: Multi-Gene Panel Testing in Suspected Lynch Patients
- Identification of unexpected actionable mutations in high-penetrance non-Lynch genes
  - BRCA1/2 mutations in “Lynch-like” patients who do not fulfill clinical criteria for HBOC
- Increased yield comes at the cost of VUS identification and discovery of mutations in moderate-penetrance genes

Background – Genetic Testing for Hereditary Cancer Syndromes
- Traditional model
  - Analyze specific genes for patients who fulfill clinical criteria for a specific syndrome
  - Per NCCN guidelines, Lynch syndrome testing recommended for patients whose histories fulfill Bethesda guidelines or Amsterdam criteria
- Emerging model – Multi-gene panel testing
  - Next generation sequencing of numerous cancer susceptibility genes in parallel
- Advantages:
  - Analyze multiple genes simultaneously
  - Cost is dropping
- Concerns:
  - Identification of uninformative variants of uncertain significance (VUS)
  - Identification of mutations in moderate-penetrance cancer susceptibility genes
  - Does panel testing offer meaningful advantages over targeted, criteria-based testing strategies?

Background – Lynch syndrome (HNPCC)
- Most common hereditary GI cancer syndrome
  - Germline mutations in DNA mismatch repair genes: MLH1, MSH2, MSH6, PMS2, and EPCAM
  - Up to 30% of families fulfilling clinical criteria for Lynch do not have germline mutations
- 3% of colorectal cancers, 2% of endometrial cancers
  - Increased lifetime risk of other associated neoplasms:
    - GI: Gastric, small intestine, pancreatic, hepatobiliary
    - Gyn: Ovarian
    - Urinary tract: Bladder, ureter/renal pelvis, adrenocortical, kidney
    - Cutaneous: Sebaceous adenomas/carcinomas, keratoacanthomas
    - Brain tumors
    - Slightly increased risk of breast and prostate cancers

Study Aims
- Using 25-gene panel:
  - Determine prevalence of non-Lynch mutations in patients undergoing testing for Lynch syndrome
  - Describe clinical phenotype of mutation carriers
Methods: Study Population
- 3057 consecutive subjects
  - Personal history of Lynch-associated cancer and/or polyps
  - DNA submitted in 2012-13 for clinical Lynch testing
- Subjects undergoing testing for <5 Lynch syndrome genes were not included
- After completion of clinical Lynch testing, samples anonymized for research-based testing
- 1797 subjects excluded
  - Testing originated from one of 10 states that mandate destruction of samples after clinical genetic testing (N=1615)
  - Technical factors (insufficient remaining DNA, non-blood sample) N=182
- Final study population:
  - 1260 subjects
  - All with personal history of Lynch-associated cancer and/or polyps

Methods: 25-Gene Hereditary Cancer Panel

Methods: Clinical Characteristics
- As part of routine clinical testing, clinicians completed standard test request forms
  - Ancestry
  - Personal history of cancer and/or polyps
  - Age at diagnosis
  - Family history of cancer
- Personal/family history data broadly categorized to protect anonymization
- “Lynch-associated” cancers included
  - Colorectal, endometrial, ovarian, gastric, pancreatic, small bowel, urinary tract, hepatobiliary, and brain cancers, and sebaceous adenomas/carcinomas
  - Breast cancer not considered Lynch-associated, but data on personal/family histories of breast cancer were tracked
- Fulfillment of NCCN criteria for Lynch testing and hereditary breast ovarian cancer (HBOC) testing
  - Determined based on reported personal/family history data

Results: Subject Characteristics (N=1260)
- 73% female
- 41% Western/Northern European ancestry
- Median age 1st cancer diagnosis: 47 years (IQR 39-55.5)
- 63% with history of colorectal cancer
  - 34% with colorectal cancer age <50
  - 23% with endometrial cancer
  - 7% with ovarian cancer
  - 5% with breast cancer
  - 14% with multiple primary cancers
  - 74% with family history of any Lynch-associated cancer
  - 23% with family history of breast cancer
  - 88% met NCCN criteria for Lynch testing
- 25% met NCCN criteria for hereditary breast/ovarian cancer (HBOC) testing

Results: Germline Testing (N=1260)
- 155 (12.3%) subjects with ≥1 pathogenic mutation on the 25-gene panel
  - 114 (9.0%) subjects with a Lynch mutation
  - 43 (3.4%) with a non-Lynch mutation
  - Including 2 subjects with both Lynch and non-Lynch mutations
    - One with MSH6 and STK11 mutations
    - One with MSH2 and ATM mutations

Methods: Clinical Characteristics

Pathogenic mutations identified by multi-gene panel testing

BRCA1/2 carriers (N=15) 10% of all mutations identified

Other high-penetration mutation carriers (N=8)
- APC (N=5) and biallelic MUTYH (N=2)
  - 5 (71%) with colorectal cancer
  - 3 (43%) with history of colorectal polyps
  - 1 (14%) with history of breast cancer
  - 100% with family history colorectal cancer
  - 100% met NCCN Lynch criteria
- STRK1 (N=1); same patient also carried pathogenic MSH6 mutation
  - Personal history of 3 primary cancers
    - Colorectal, endometrial, and breast cancers
    - Met NCCN Lynch criteria
- Note: 28 subjects (2% of study cohort) with monoallelic MUTYH mutations
  - Significance unclear
  - 23/28 were G196D or Y179C

Other high-penetrance genes
- Lynch syndrome
  - BRCA1/2
  - MSH2
  - MSH6
  - MSH2
  - MSH6
  - EPCAM
  - APC
  - CDH1
  - PTEN
  - TP53
  - APC
  - BARD1
  - CHEK2
  - BRIP1
  - RAD51C
  - PALB2
  - CDK12
  - NBN

Moderate-penetrance
- ATM
- BARD1
- BRCA1
- BRCA2
- CDX2
- CHEK2
- EPCAM
- MLH1
- MSH2
- MSH6
- MUTYH
- PMS2
- SMAD4
- STK11
- BRCA1
- BRCA2
- CHEK2
- MLH1
- MLH2
- MSH2
- MSH6
- TP53

All sequence variations and large rearrangements classified for pathogenicity

Variants of Uncertain Significance (VUS)
- Of the 20 non-Lynch genes, 594 VUS were seen in 433 (34%) subjects
- Most common genes to have a VUS
  - ATM (N=114 subjects)
  - APC (N=50)
  - MLH2 (N=50)
  - BRCA2 (N=50)
  - CDX2 (N=32)
  - CHEK2 (N=25)

Pathogenic mutations identified by multi-gene panel testing

BRCA1/2 carriers (N=15) 10% of all mutations identified

Non-Lynch mutations identified by multi-gene panel testing

BRCA1/2 carriers (N=15) 10% of all mutations identified

N=1260

33% fulfilled NCCN HBOC testing criteria versus 16% Lynch carriers

93% fulfilled NCCN Lynch testing criteria versus 95% of Lynch carriers

Lynch syndrome mutations identified by multi-gene panel testing

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Non-Lynch mutations identified by multi-gene panel testing

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33% fulfilled NCCN HBOC testing criteria versus 16% Lynch carriers

93% fulfilled NCCN Lynch testing criteria versus 95% of Lynch carriers
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- 3057 consecutive subjects
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- 1797 subjects excluded
  - Testing originated from one of 10 states that mandate destruction of samples after clinical genetic testing (N=1615)
  - Technical factors (insufficient remaining DNA, non-blood sample) N=182
  - Final study population: 1260 subjects
  - All with personal history of Lynch-associated cancer and/or polyps

Results: Subject Characteristics
- 1260 subjects
- At least 1 personal/family history of breast cancer
- At least 1 personal/family history of colorectal cancer

Methods: Clinical Characteristics
- As part of routine clinical testing, clinicians completed standard test request forms
  - Ancestry
  - Personal history of cancer and/or polyps
  - Age at diagnosis
  - Family history of cancer
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  - “Lynch-associated” cancers included:
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Methods: 25-Gene Hereditary Cancer Panel

High-penetrance genes
- Lynch syndrome
  - BRCA1/2
  - MLH1
  - MSH2
  - PMS2
  - EPCAM

Moderate-penetrance genes
- CDKN2A
- ATM
- PMS2
- STK11
- CDH1
- PTEN
- TP53
- MSH6
- BARD1
- CHEK2
- BRIP1
- NBN
- BRCA1/2

Other high-penetrance genes
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- CDX2
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- APC
- EPCAM
- BRCA1

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- 60% colorectal cancer
- 33% colorectal cancer age <50
- 7% ovarian cancer
- 0 with breast cancer
- 0 with pancreatic cancer

33% fulfilled NCCN HBOC testing criteria

Other high-penetrance mutation carriers (N=8)
- APC (N=5) and biallelic MUTYH (N=2)
  - S (71%) with colorectal cancer
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  - 100% with family history colorectal cancer
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Non-Lynch mutations identified by multi-gene panel testing
Multi-Gene Panel Testing in Patients Suspected to Have Lynch Syndrome*

Matthew B. Yurgelun, Brian Allen, Rajesh Kaldate, Karla Bowles, Benjamin Roa, Richard J. Wenstrup, Anne-Renee Hartman, Sapna Syngal

*Poster Presented at ASCO - June 2014

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