New Polyposis Genes: Beyond FAP and MAP

Robert Gryfe MD, PhD, FRCSC
No disclosures
Polyposis Syndromes

Polyposis

Attenuated Polyposis

Adenoma, Hamartoma, Sessile Serrated
<table>
<thead>
<tr>
<th>Yr</th>
<th>Synd</th>
<th>Gene</th>
<th>CR</th>
<th>Pheno</th>
<th>CRC</th>
<th>GI</th>
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<td>1991</td>
<td>FAP</td>
<td>APC</td>
<td>Ad</td>
<td>&gt;100</td>
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<td>DuoAd GFGP GA</td>
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<td>10-99</td>
<td>&lt;70%</td>
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<td>2002</td>
<td>MAP</td>
<td>MUTYH</td>
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<td>40-70%</td>
<td>Gastr SB</td>
<td>Stomach Pancreas</td>
<td>AVM's</td>
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<td>1998</td>
<td>PJS</td>
<td>STK11</td>
<td>PJ</td>
<td>≥1-2</td>
<td>10-40%</td>
<td>Gastr SB</td>
<td>Breast Pancreas SB Stomach Gonadal Lung</td>
<td>Mucocutaneous pigmentation</td>
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DNA Repair Colorectal Cancer Caretakers

Mismatch Repair Deficiency
Lynch syndrome

Base Excision Repair Deficiency
MUTYH Associated Polyposis

Others?
New Polyposis Genes: Beyond FAP & MAP

- 2002-2012: very little new in inherited CRC genetics
- Recent advances likely due to more powerful ‘omics’ sequencing
- Polymerase proofreading associated polyposis (PPAP)
- NTHL1 associated polyposis (NAP)
- Hereditary mixed polyposis syndrome (HMPS)

All:
- Described in past 5 years
- DNA repair deficiencies
- Rare
- Variable phenotype

- Don’t blame me for the acronyms!
Polymerase Proofreading Associated Polyposis

Germline mutations affecting the proofreading domains of POLE and POLD1 predispose to colorectal adenomas and carcinomas

Claire Palles1, Jean-Baptiste Cazier2,18, Kimberley M Howarth1, Enric Domingo1, Angela M Jones1, Peter Broderick3, Zoe Kemp1, Sarah L Spain1, Estrella Guarino4, Israel Salguero4, Amy Sherborne3, Daniel Chubb3, Luis G Carvajal-Carmona1, Yusanne Ma3, Kulvinder Kaur5, Sara Dobbins3, Ella Barclay1, Maggie Gorman1, Lynn Martin1, Michal B Kovac1,6, Sean Humphray7, The CORGI Consortium8, The WGS500 Consortium8, Anneke Lucassen9, Christopher C Holmes10, David Bentley7, Peter Donnelly2,10, Jenny Taylor5, Christos Petridis11, Rebecca Roylance12, Elinor J Sawyer11, David J Kerr13, Susan Clark14, Jonathan Grimes15,16, Stephen E Kearsleyy, Huw J W Thomas17, Gilean McVean2, Richard S Houlston3, Ian Tomlinson1,5

Palles et al, 2013 (UK)

• 15 probands + 5 relatives with ≥10 adenomas, <60 yrs
  - 8 had CRC, 12 had affected FDR (≥5 adenomas)
  - No APC, MUTYH or MMR mutation
  - 1x APC, 1x MSH6

• whole genome sequencing
POLE p.Leu424Val (exonuclease/proofreading domain)
• 3 affected individuals in 1/13 selected CRC/pedigrees

POLE L424V Validation:
• 12/3,805 (0.3%) European CRC; 0/6,721 controls; 0/10,755 controls
• All 12 POLE CRC carriers were in highly selected case groups (vs. clinical trials)

Phenotype:
• Variable tendency to multiple or large adenomas
• All pedigrees consistent with AD & high penetrance of multiple adenomas & young MSS CRC
• Adenomas = 10 (1-68); CRC = 33 yrs (28-53 yrs)
• No obvious: accelerated adenoma > carcinoma; adenoma, carcinoma or extracolonic phenotype (i.e. sidedness, histology, etc)

Somatic:
• APC substitution mutations (but not G:C > T:A transversions)
PPAP cont'd

POLD1 p.Ser478Asn (exonuclease/proofreading domain)
- In 2/13 selected CRC/pedigrees (2 probands + 1 relative)
- Likely a common ancestor by shared microsatellite testing

POLD1 S478N Validation:
- Identified in a 28 yo CRC, whose father had CRC at 44 yrs

Phenotype:
- Pedigrees consistent with AD & high penetrance of multiple adenomas & young MSS CRC
- Variable tendency to multiple or large adenomas
- 7 POLD1 carriers developed endometrial cancer
  - 0/386 early onset endometrial cancers without a FHx of CRC
- 1 POLD1 carrier developed 2 primary astrocytomas

Somatic:
- APC substitution mutations (but not G:C > T:A transversions)
PPAP cont’d

Elsayed et al, 2014 (Netherlands)
• 1,188 familial CRC/polyposis
• 3 (0.25%) POLE L424V (0 POLD1 S478N)
• Somatic MSI & MSH2/MSH6-deficiency in 2/3 POLE L424V CRC's

Valle et al, 2014 (Spain)
• 858 polyposis & familial/young CRC
• 1 (0.1%) POLE L424V (de novo), in 28 yo CRC with polyposis
• 1 (0.1%) POLD1 L474P (novel), MMR-proficient, Amsterdam II (CRC & EC)

Chubb et al, 2015 (UK)
• 646 CRC ≤55 yrs & ≥1 affected FDR
• 2 (0.3%) POLE L424V; 1 (0.15%) POLD1 S478N

PPAP can be MSS/MSI, Am+/familial/de novo & is rare, even in selected families
**PPAP cont’d**

**Bellido et al, 2015 (Spain)**

- 544 polyposis & familial/young CRC (from Valle et al, 2014)
- Phenotype review of previous studies
- 4 likely pathologic POLE (non-L424V) mutations:
  - D316H, D316G, R409W, L474P

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<th>Gene</th>
<th>Carriers/Families</th>
<th>CRC Rate</th>
<th>Mean Age (yrs)</th>
<th>Adenomas</th>
<th>&gt;2 Adenomas</th>
<th>&gt;5 Adenomas</th>
<th>Duodenal Adenomas</th>
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<td>47/20</td>
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<td>40.7</td>
<td>19</td>
<td>82%</td>
<td>74%</td>
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<td>POLD1</td>
<td>22/8</td>
<td>59%</td>
<td>35.9</td>
<td>12</td>
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Polymerase Proofreading Associated Polyposis

POLE
- AFAP (20-100 adenomas)
- Amsterdam I
- CRC & 5-20 adenomas, <50 yrs
- CRC or 5-20 adenomas & FDR CRC, both <50yrs
- CRC or 5-20 adenomas & ≥2 FDR/SDR with CRC, regardless of age

POLD1
- AFAP (20-100 adenomas)
- Amsterdam II (CRC & EC)
- CRC or EC & 5-20 adenomas, CR <50 yrs & EC <60yrs
- CRC or EC or 5-20 adenomas & FDR CRC, CR <50yrs & EC <60yrs
- CRC or EC or 5-20 adenomas & ≥2 FDR/SDR with CRC or EC, regardless of age

Screening
- Colonoscopy q1-2 yrs, at 20-25 yrs
- EGD q3-5 yrs, at 20-25 yrs
- POLD1 EC: pelvic exam & US, q1-2 yrs, at 30-40 yrs
Weren et al, 2015 (Netherlands)

- 48 probands + 3 relatives with 5-250 polyps
  - 21 had CRC & 27 affected FDR
  - No APC & MUTYH mutation
- Whole exome sequencing
  - 1x POLD1 p.Gly321Ser & 1x POLE p.Lys284Glu
Homozygous NTHL1 pGln90* mutations

- Exonuclease/proofreading domain (like POLE, POLD1)
- Base excision repair gene, autosomal recessive (like MUTYH)
- 4 individuals from 3 families
  - Consanguinity in 1/3 families
  - 3 additional affected individuals from these families
  - 1 additional heterozygous carrier not affected
  - 0/149 in additional cohort of 149 with polyposis
- Heterozygous carriers = 0.36%, Homozygous ≈ 1/75,000
- 7 CRC’s in 4 individuals at 53 yrs (40-63 yrs)
- Adenomas = 28 (8-50)
- Endometrial ca or complex hyperplasia in 3/3 females
- Duodenal ca & multiple duodenal adenomas in 1+1 of 7
- NTHL1 C:G > T:A transitions: 11/12 APC, p53, KRAS & PIK3CA mutations
- MUTYH C:G > A:T transversions
Rivera et al, 2015 (Canada)
- German decent woman: Colon ca (41 yrs), subsequent bladder & breast ca’s, H&N SCC, CR adenomas (<30) & ovary cystadenoma
- Germline NTHL1 pGln90* & c.709+1G>A (splice-site) mutation

Belhadj et al, 2017 (Spain)
- Homozygous NTHL1 pGln90* in:
  - 2/88 (2.3%) polyposis & 0/523 MSS familial CRC
  - Male with CRC & 24 adenomas at 48 yrs
  - Female with breast ca x2 (47, 50 yrs), bladder ca (66 yrs), CRC x3 (67 yrs) & >15 adenomas
- Heterozygous in 3/1348 (0.2%) CRC’s & 12/2743 (0.4%) controls
- Spanish: likely recurrent (vs. founder) mutation

Phenotype:
- Attenuated (<50) polyposis, CRC (7/10), breast ca (3/5), endometrial ca (2/5); multiple malignancies (7/10) diagnosed 40-74 yrs
- Similar phenotype/screening to MAP?
Hereditary Mixed Polyposis Syndrome

- Autosomal dominant
- Varying histology polyps, confined to the colon & rectum
- Atypical juvenile polyps (mixed hamartoma/adenoma), adenomas, serrated/hyperplastic polyps
- BM change, bleeding, pain, obstruction, anemia at 40 yrs (23-65 yrs)
- ↑ risk for CRC, average 47 yrs (32-74 yrs)
Hereditary mixed polyposis syndrome is caused by a 40-kb upstream duplication that leads to increased and ectopic expression of the BMP antagonist GREM1.

Jaeger et al, 2012 (UK)

- **CRAC1** locus (15q15.3-q22.1)
  - **SCG5, GREM1 & FMN1 genes**
- 40 kb duplication spanning 3' **SCG5** to 5' of **GREM1**
  - Contains multiple gene regulatory regions
  - 190 bp specific amplification product
- Perfect concordance with 40 affected & 50 unaffected in 6 Ashkenazi Jewish HMPS families
- 0/935 AJ controls, 1/718 AJ CRC → HMPS phenotype
- ↑ ectopic epithelial GREM1 expression; normal SCG5 expression
- ↑ GREM1 predicted to ↓ BMP pathway expression (like JPS)
Venkatachalam et al, 2011 (Netherlands)
• Duplication of entire GREM1 gene & upstream SCG5 exons 3-6 in a CRC at 35 yrs

Rohlin et al, 2016 (Sweden)
• GREM1 16 kb regulatory region duplication co-segregated with 4/4 affected polyposis & 0/2 unaffected in an atypical polyposis family.

Lieberman et al, 2017 (Israel)
• GREM1 40 kb duplication: 16 carriers from 4 Ashkenazi Jewish families
• Polyposis in 3/4 families
• Amsterdam I in 1/4 families

**GREM1 40 kb duplication in Ashkenazi Jews**

**Other duplications in other ethnicities**

**Most look like HMPS, but Lynch syndrome phenotype possible**
HMPS cont’d

HMPS vs JPS:
- Similar genetic defect
- JPS extracolonic:
  - Gastric, SB, HHT
- HMPS serrated/HP polyps, older presentation

Recommendations:
- No established guidelines, very rare disease
- Colonoscopy q2 yrs at 18 yrs
- No apparent extracolonic phenotype
New Polyposis Genes: Beyond FAP and MAP

Polymerase proofreading associated polyposis (PPAP)
- POLE L424V, others?
- POLD1 S478N, (L474P)
- Autosomal dominant
- Am+/familial/de novo
  - POLD1 & endometrial ca

NTHL1 associated polyposis (NAP)
- NTHL1 Gln90*, (c.709+1G>A)
- C:G >T:A transitions
- Autosomal recessive
- Attenuated polyposis
- Extracolonic cancers appear common

Hereditary mixed polyposis syndrome (HMPS)
- GREM1 40 kb duplication in Ashkenazi Jews
- Other duplications in other ethnicities
- Most look like HMPS, but Lynch syndrome phenotype possible

Rare syndromes, even in selected populations
Variable phenotypes

Challenges:
- Testing strategies
- Clinical recommendations
New Polyposis Genes: Beyond FAP and MAP

- BMPR1A
- BMP
- GREM1
- HMPS
- FZ
- Wnt
- LKB1
- PIS
- PTEN
- PI3K/AKT
- AMPK
- TSC1/2
- Rheb
- mTORC1
- mTOR
- β-catenin
- APC
- AXIN
- DSH
- GSK3β
- FAP
- POLE
- PPAP
- MMR defect
- Proofreading defect
- NTHL1
- MSH2
- MLH1
- MSH6
- PMS2
- LS

Transcriptional effects, for example:
- mTOR: growth and proliferation, translation, increased VEGF-A expression
- β-catenin complexes with TCF/LEF, activating c-Myc and cyclinD1, leading to cell proliferation
- SMAD4 complex regulates cell growth and proliferation

1991-2002
≥2012