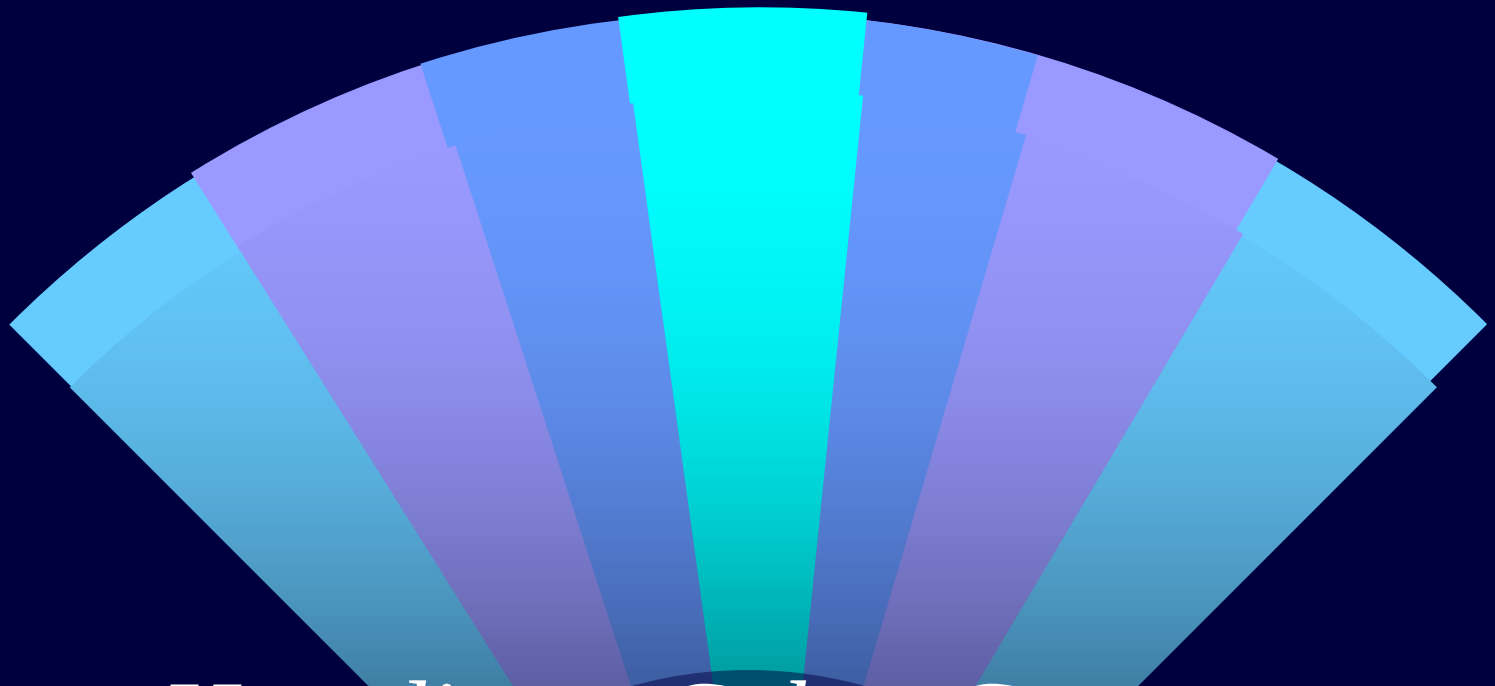


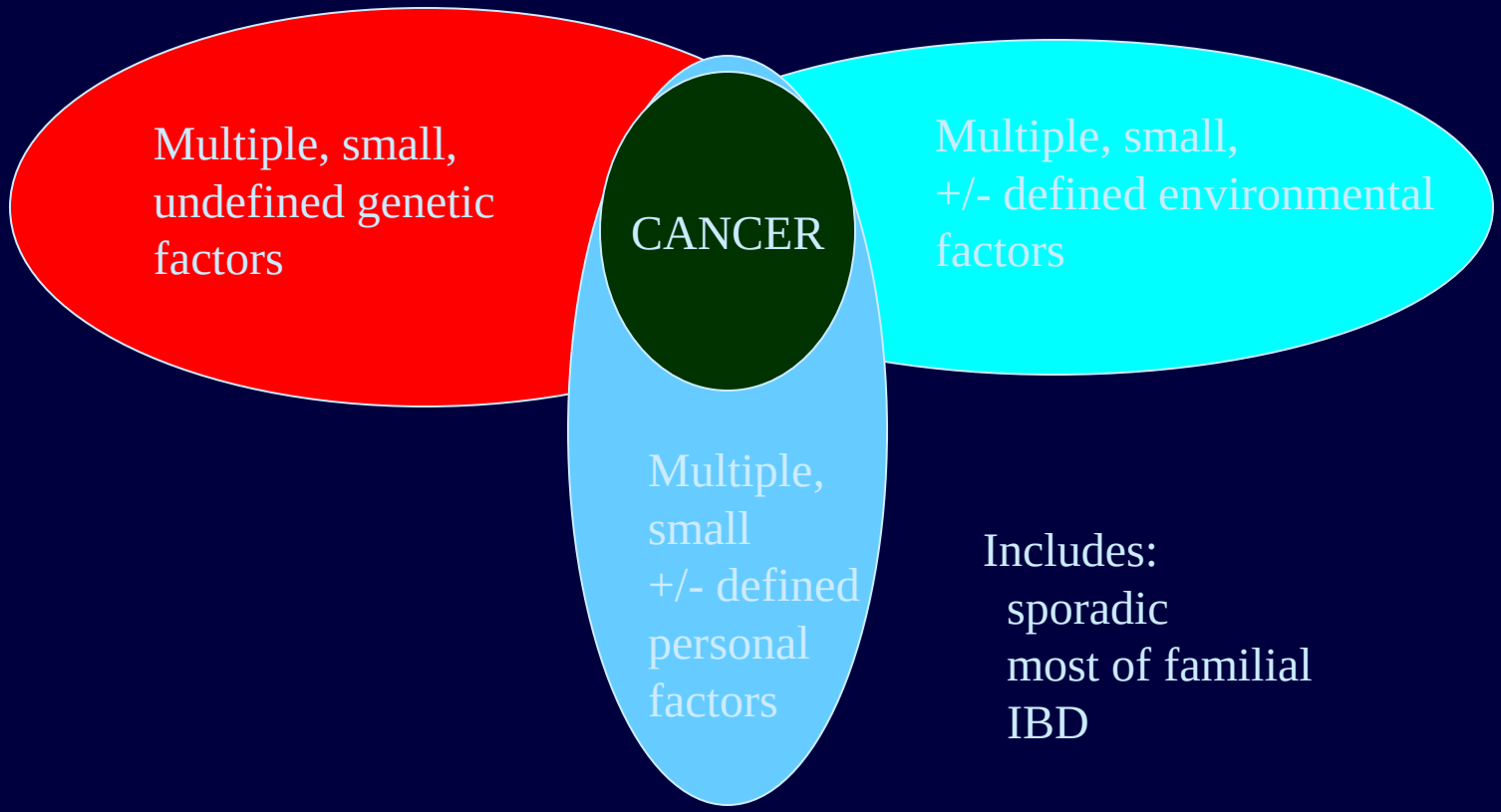
Hereditary Colon Cancer – Determination of Risk

Dawna Gilchrist, MD, FRCPC FCCMG

Objectives

- Review of Colon Cancer from a genetic perspective
- Diagnosis of Lynch Syndrome
  - Clinical
  - Genetic
  - Pathological
- Doing this all in real time

Most of cancer, including most of colon cancer



Includes:
- sporadic
- most of familial
- IBD

Bad Luck

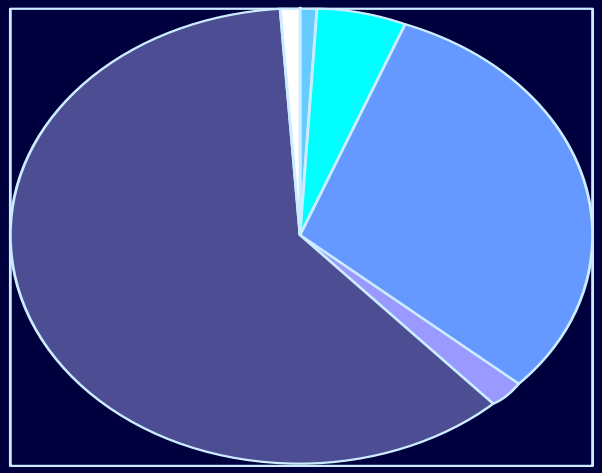
- "bad luck" is 2/3 of all of Cancer

  Science Vol. 347 pp78-81 Jan 2015
- Can occur at ANY age
- A truly singleton case of any type of cancer, regardless of age, is more likely to be multifactorial, even mostly bad luck, than genetic in nature

Somatic versus Germline mutations

- A somatic mutation causes cancer
- A germline mutation causes a genetic syndrome

Colon Cancer



FAP
Lynch
Familial
IBD
Sporadic
Funny Polyps

Familial Adenomatous Polyposis

- Dominant, related to APC gene
  - Profuse, classic or attenuated
  - Polyps throughout gi tract (remember the stomach)
  - Extracolonic features
- Recessive, related to MYH gene

"Funny" Polyps

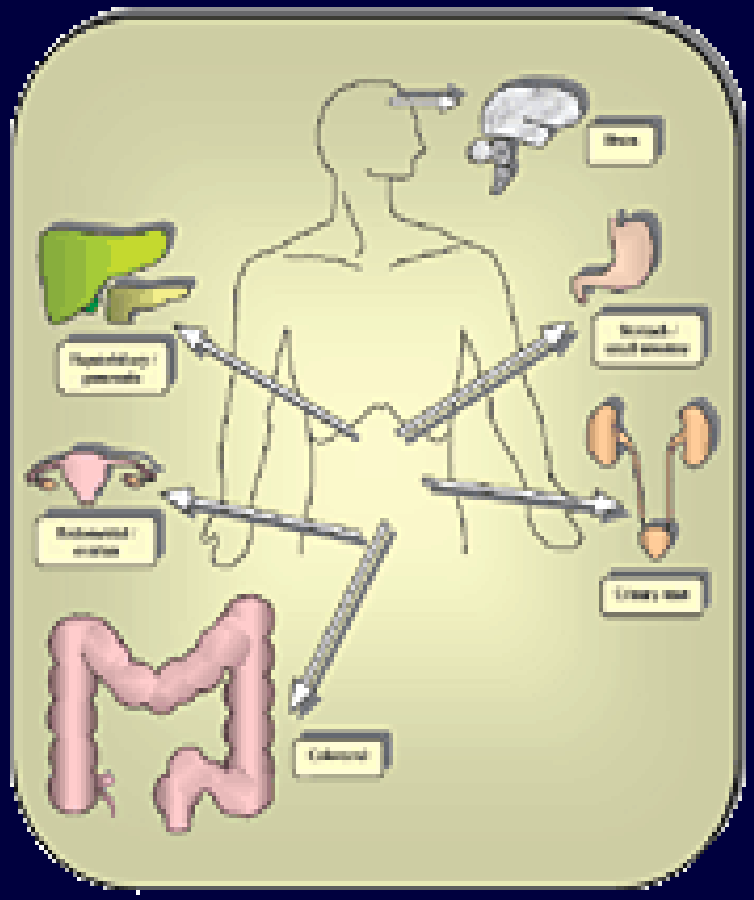
- Peutz Jeghers
- Juvenile Polyposis
- Mixed Polyposis
- Polyps associated with other genetic syndromes eg. Cowden, Hereditary Hemorrhagic Telangiectasia

Familial Colon Cancer

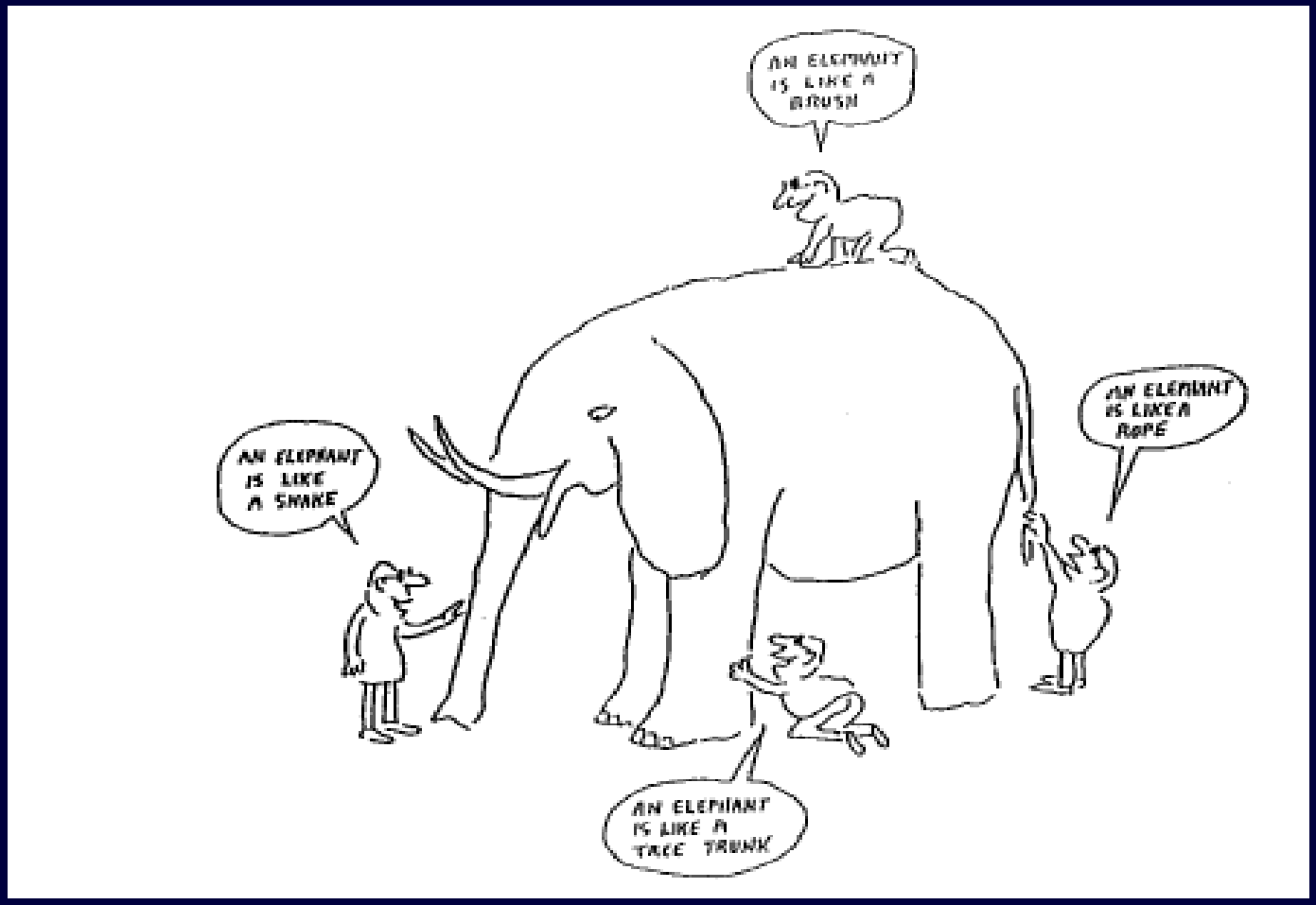
- Two or more cases of colon cancer in the family
- Does not meet criteria for genetic syndrome
- A “grab bag” of multifactorical cases and syndromic cases NYD

Lynch Syndrome – a syndrome of deficent mismatch repair (MMR)

- CRC R>>L, average onset 44
- Colon 50-80% (♂>♀)
- Uterine 20-60%
- Ovary, Stomach, Upper Urinary Tract – 10-20%
- Biliary Tract and Small Bowel – Rare
- 1/3 of Turcot – Lynch plus brain, usually glioblastoma
- Muir-Torre – Lynch plus sebaceous gland neoplasm, keratocanthomas



It's often a matter of perspective



The Geneticist Approach to Diagnosis of Lynch Syndrome

- Patient information
- Family History
- Pathologic Information
- Genetic Testing

Patient Information

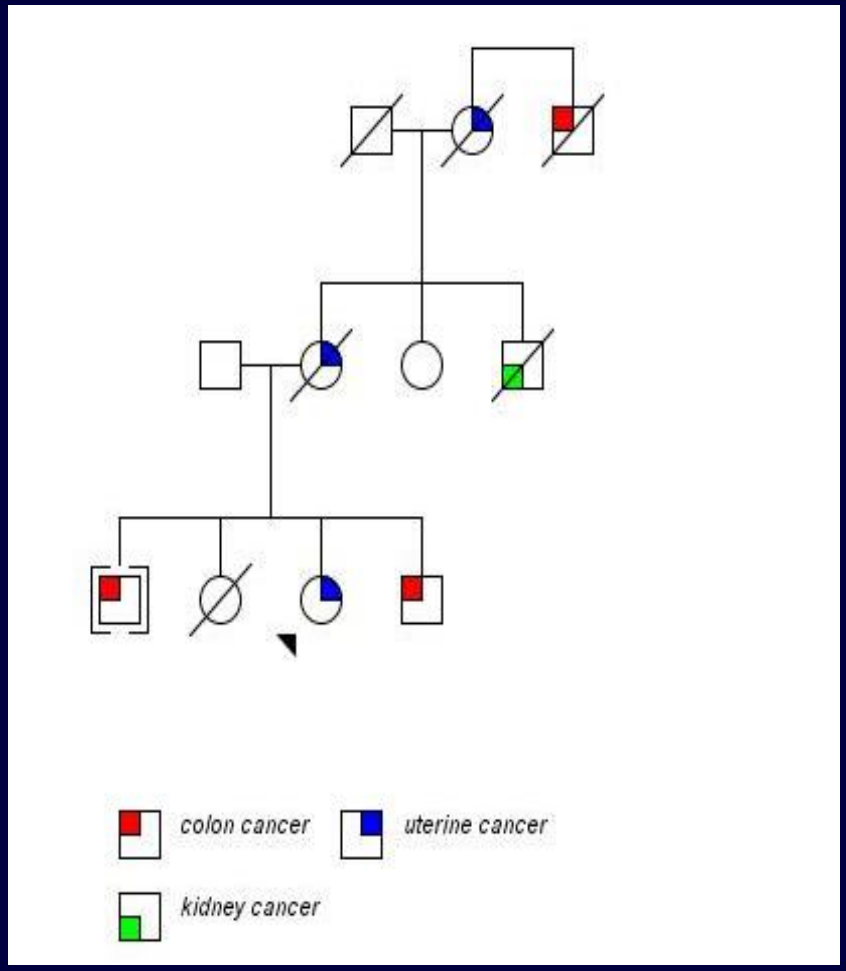
- Age of onset
- Tumour (gross and H&E)
  - If colonic, position
  - Specific cell type eg. Endometroid ovarian cancer, transitional cell tumour of urinary tract
- Special Pathologic Features
  - Crohn's like peritumour lymphocytic reaction
  - Tumour Infiltrating Lymphocytes (TIL's)

    JR Jass Disease Markers 2004 20:215-224

Patient Information

- Very Special Pathology
  - Microsatellite Instability
    - Non specific. Much of MSI associated with **somatic** mutations of MLH1
  - Immunohistochemistry (mismatch repair profile)
    - Non specific for missing MLH1 – again mostly due to **somatic** mutations of MLH1
    - Missing MSH2/MSH6/MSH2 is NOT absolutely diagnostic of a germline mutation

The Family History



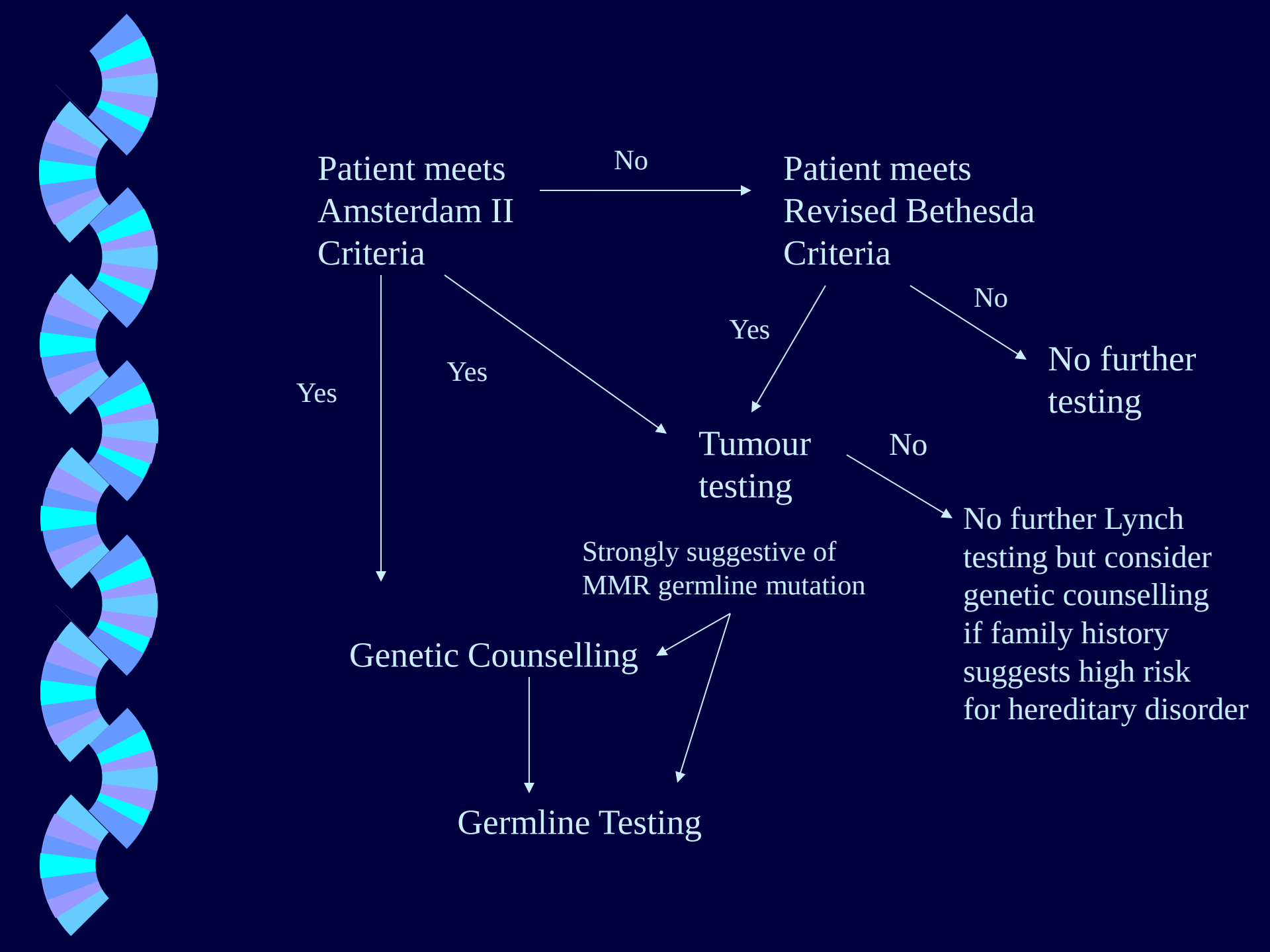
- Dominant Inheritance
- Incomplete Penetrance
- Variable Expressivity
  - Age of onset
  - Target organ(s)
  - Individual natural history
- Accuracy of reports critical

Criteria

- Amsterdam Criteria *(specific, not very sensitive)*
  - 3 or more relatives with CRC cancer; one should be 1° relation to other two; 2 or more generations affected; at least one CRC before age of 50
  - FAP ruled out; pathologic verifications
- Modified Amsterdam
  - Includes other Lynch associated tumours
- Bethesda *(sensitive, not very specific)*

Purpose of Criteria

- Amsterdam and Modified Amsterdam
  - For purposes of molecular testing
  - DNA extracted from blood
- Bethesda
  - For testing for MSI and IHC
  - Tumour material (not validated on polyps)


Patient meets Amsterdam II Criteria
No
Patient meets Revised Bethesda Criteria
Yes
No
No further testing
Yes
Yes
Tumour testing
No
No further Lynch testing but consider genetic counselling if family history suggests high risk for hereditary disorder
Strongly suggestive of MMR germline mutation
Genetic Counselling
Germline Testing

Genetic Testing

- MLH1, MSH2 (majority)
- MSH6, PMS2
- Epcam (promoter for MSH2)
- Sequencing and deletion/duplication

Everything in agreement

- 37 yo male patient with colon cancer (ascending colon)
- Mother + 2 maternal uncles with colon cancer
- Tumour shows moderate peritumour lymphocytic response and 3 TIL's per HPF
- MSI-H
- IHC shows missing MSH2 and MSH6
- Pathologic mutation in MSH2

Mostly in agreement

- 53 year old male with colon cancer x 2 (38; 53) with H&E suggestive of possible Lynch syndrome
- MSI-H and abnormal MMR for MSH6
- Significant family history of abdominal-pelvic cancers
- Variant of unknown significance found in MSH6
- 3 other family members tested (total 8 meioses) and have VUS. Their tumours are also MMR abnormal for MSH6
- We have found a previously unreported mutation.

Unusual Family History

- 2 sisters with cancer – colon at 43; uterine at 38
- MSI-H, abnormal MMR
- No family history of abdominal-pelvic cancer
- Lynch mutation found in both sisters
- Mutation not found in either parent
- ?? How to explain this

Possible Explanations

- Sample mix-up somewhere
- Non-paternity
- What about … A New Mutation.

And, the answer is

- Re-examination of father's molecular test results shows a small spike consistent with 30% mosaicism for same mutation at daughters

Frustrating

- Patient with Lynch type tumour
- Family history consistent with Lynch syndrome by Amsterdam II i.e. more than just colon cancer
- MSS, MMR profile normal
- Normal genetic testing for Lynch genes

- Lynch-like syndrome

Due to ???

- Always assess accuracy of all information
- Confluence of bad luck
- Other genes

- “The biggest problem is the unknown unknowns” Donald Rumsfeld

Pulling out of hair

- Patient with Lynch type tumour
- Familial history variable but often paucity of other cases
- MSI-H, abnormal MMR suggestive of germline mutation
- No mutation found on genetic testing of Lynch genes
- ?? How to explain

If the problem is with MLH1 – do the hypermethylation tests

- Hypermethylation of MLH1 is a common somatic event, particularly in the elderly
- Causes MSI-H and missing MLH1/PMS2 on IHC
- Hypermethylation studies on tumour
- BRAF V600 in approximately 2/3

What about MLH1 promotor germline hypermethylation

- Prevalence ?? but rare
- To evaluate:
  - make sure you have clearly normal tissue to evaluate (not just immediately peri-tumour)
  - Test for hypermethylation of MLH1 on patient's blood

Counselling Issue

- This cannot be tested for by sequencing and MLPA of MLH1 gene
- Does germline hypermethylation have implications for at-risk individuals?
  - If due to mutation of imprinting centre, could be inherited
  - If sporadic event, likely erased in meiosis

Discordance without a hypermethylation issue

- Redo IHC – reagents, subjectivity
- Complex rearrangements of one of Lynch associated genes (inv exons 1-7 MSH2 reported)
  - Rhees et al. 2014 Familial Cancer 13:219-225
- Other genes – POLD1, POLE, ?other
- Double somatic mutations – 69% recently reported
  - Haraldsdottir et al. 2014 Gastroenterology 147:1308-1316

And, why does this all matter

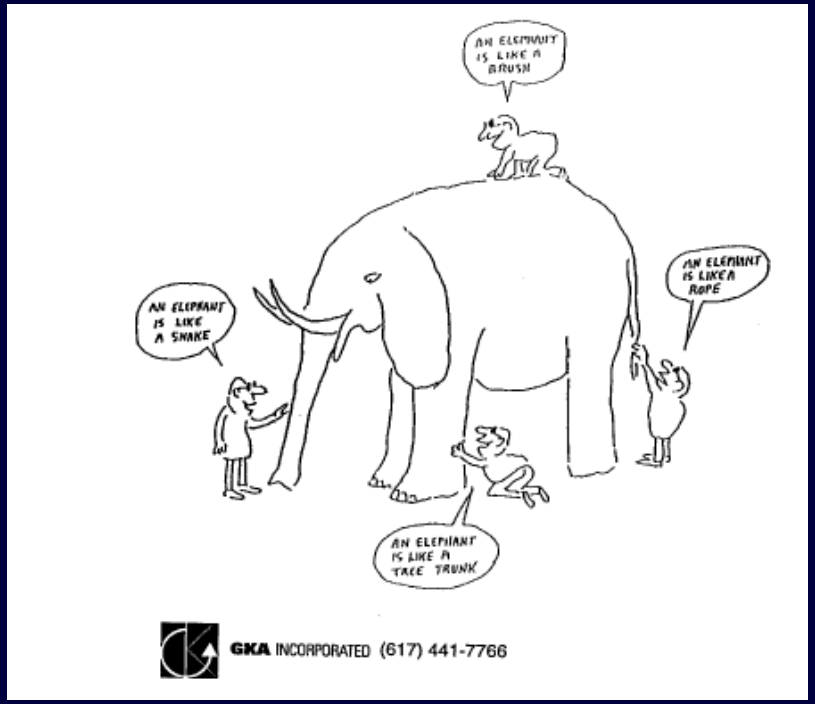
- Identification of a genetic syndrome provides more information for patient and at-risk relatives
- A known pathologic mutation in a family allows for identification of patients at risk (screening and preventive measures) and patients not at risk (routine management)

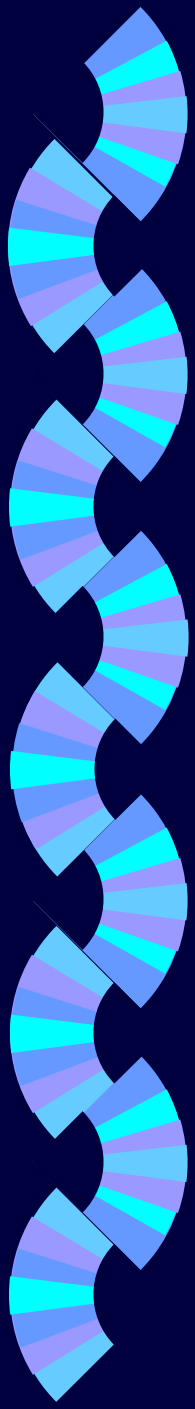
What do I think should happen

- IHC on
  - All CRC under age 60 (?70)
  - All uterine cancers under age 50
- Referral of cases suggestive of a germline mutation to Medical Genetics (rule out somatic hypermethylation first)
- Expanded panels for molecular testing

We are all members of a team

- Need the clinician to recognize individuals at risk
- Need pathology for routine and special tumour assessment
- Need genetics for identifying risks for patients and families





Thank you!

Questions & Comments