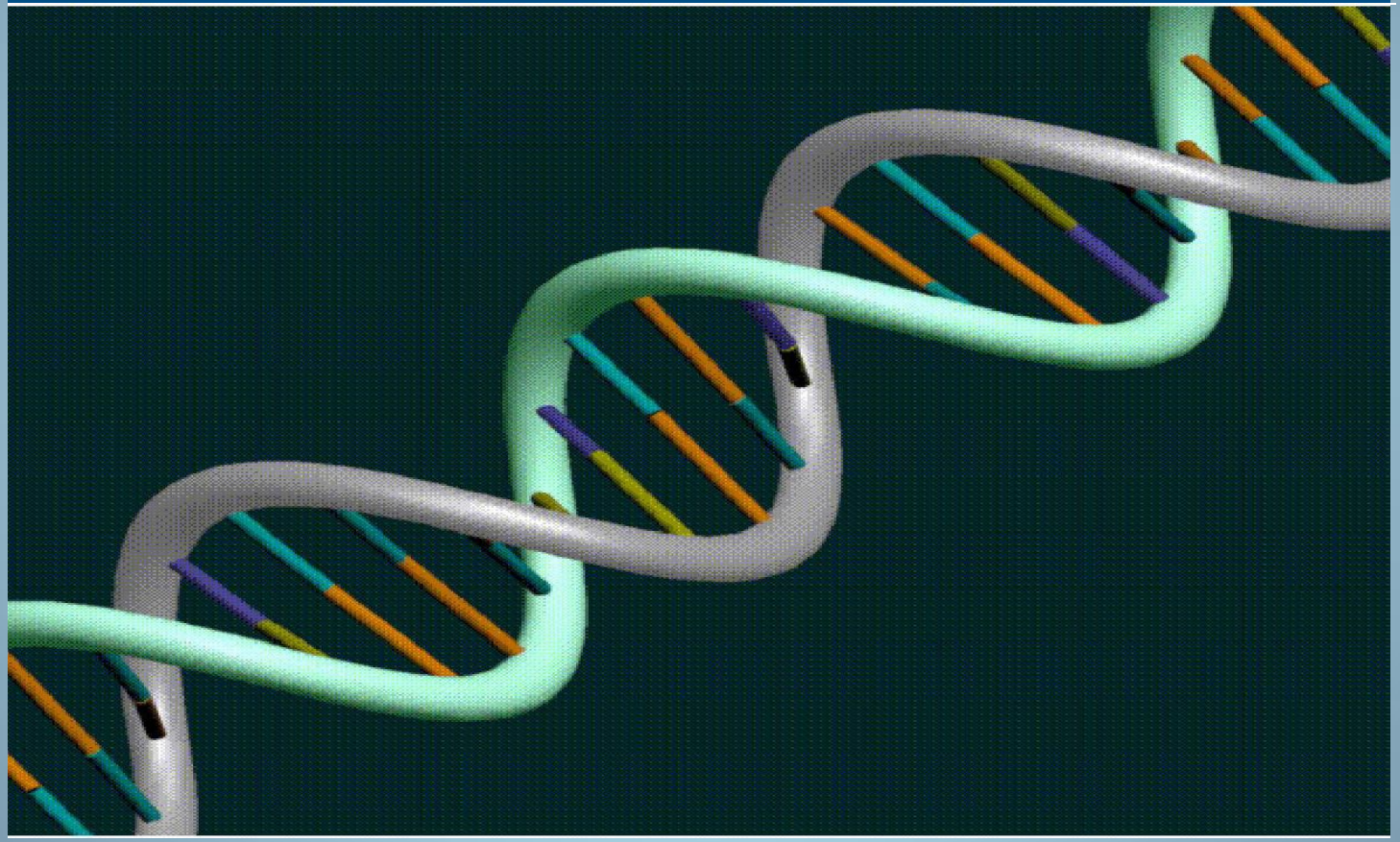
Boca Raton Regional Hospital



58 year old with T1cN0 infiltrating ductal carcinoma right breast

2013 Lumpectomy ER PR positive, Her 2 negative breast cancer

2013 Oncotype 21 Gene recurrence score: Adjuvant aromatase inhibitor

2013 BRCA 1 and BRCA 2 negative

2014 "Genova lab Estrogenomic gene snp: Dimpro

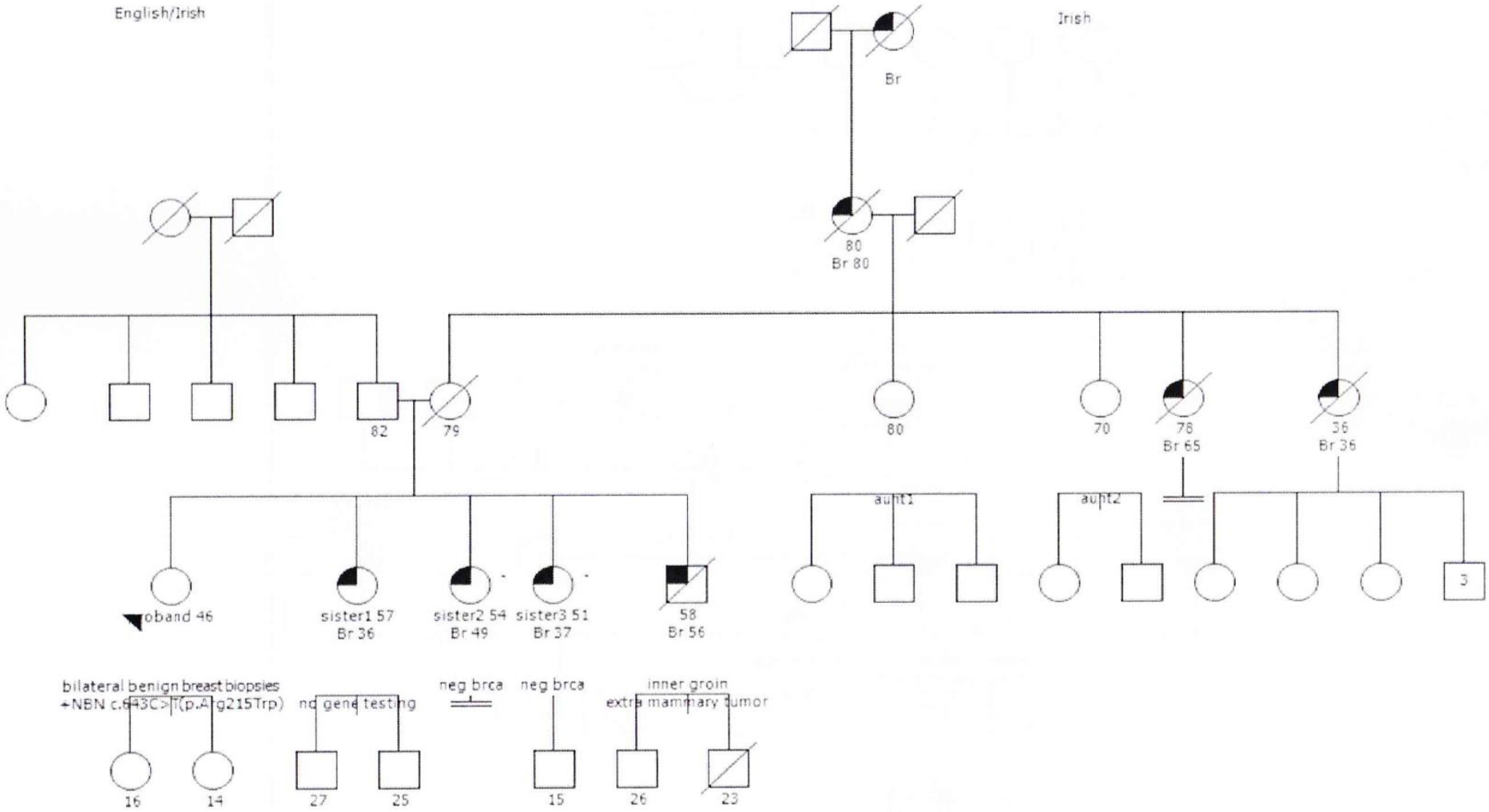
2015:Mediastinal nodal relapse: Foundation One genetic tumor test

2015: Ambry germline testing

2016: affinitor based on genetic biomarkers

Outline

- Current role of germline cancer testing
- Major Cancer Genetic syndromes: what test, who to test, interpretation of positive, negative and variant results
- Actionable results: current guidelines by site
- Discuss challenges of current testing: including VUS , Lack of reliable data for intermediate risk genes
- Other issues: direct to consumer testing


English/Irish
Irish
Br
80
Br 80
82
79
80
70
78
Br 65
56
Br 36
aunt1
aunt2
3
oband 46
sister1 57
Br 36
sister2 54
Br 49
sister3 51
Br 37
58
Br 56
bilateral benign breast biopsies
+NBN c.643C>T(p.Arg215Trp)
no gene testing
neg brca
neg brca
inner groin
extra mammary tumor
16
14
27
25
15
26
23

Hereditary Susceptibility to Cancer

- Who to test
- What test to select, and when to update testing on previously tested families
- What are the cancer risks associated with the mutation and how accurate are the estimates
- What interventions are indicated
- Is there evidence supporting improved clinical outcomes

Genetic testing for High Risk Cancer Syndromes

- Mutations are associated with cancer risk syndromes: more than one type of cancer clustered in a family or individual due to the same genetic mutation
- There are a small number of mutations that confer a significantly elevated lifetime risk of cancer in individual carriers
- Not all inherited cancer syndromes have an identifiable high risk gene, this does not mean the family is without risk. Family history and other risk factors must be used to assess risk

Genetic testing for Cancer Risk

Who Should be tested

- What we know

Who Could be tested

- What we don’t know

Genetic testing for Cancer Risk

Who Should be tested

What we know

- What is the risk of cancers associated with the mutation
- What actions can be taken to prevent or reduce risk
- What is the proven utility of the intervention

Who Could be tested

What we don't know

- What is the risk of cancers associated with the mutation
- What is the proven utility of interventions to reduce risk or prevent cancer

Genetics in Cancer

- 5-10% of all malignancies are due to highly penetrant hereditary cancer predisposition syndromes [Ballinger, 2012]

- Over 400 cancer-related genes have been identified
  - May account for many familial cancers
  - *Caution! Current clinical testing may include some of these genes of lower-risk*

http://www.sanger.ac.uk/genetics/GCP/Census

SYNDROME	Risk cancer	Recommendations
HBOC: **Hereditary Breast and Ovarian Cancer** BRCA 1 BRCA2	Breast 85% Ovary 40%	MRI and Mammogram age 25 Prophylactic oophorectomy
Hereditary Colon Uterine LYNCH	Colon 80% Uterine 40%	Colonoscopy yearly age 25 Gyn prophylactic surgery
Hereditary Pancreatic Cancer PALB2, BRCA2	60% Pancreas breast 40%	Research screening
Prostate Cancer	Younger, more aggressive forms	Increased screening

Updates and Key features of the following Cancer types:

- Colorectal Cancer
- Breast Cancer
- Ovarian Cancer
- Pancreatic Cancer
- Prostate Cancer

Colon Cancer key considerations

- 5-10% of colorectal cancers associated with genetic syndrome
- Primarily associated with young age, multiple generations, uterine cancers, other cancers
- Individual carriers are at increased risk earlier
- Universal Screening is available to capture 80% of carriers
- Intervention improves mortality

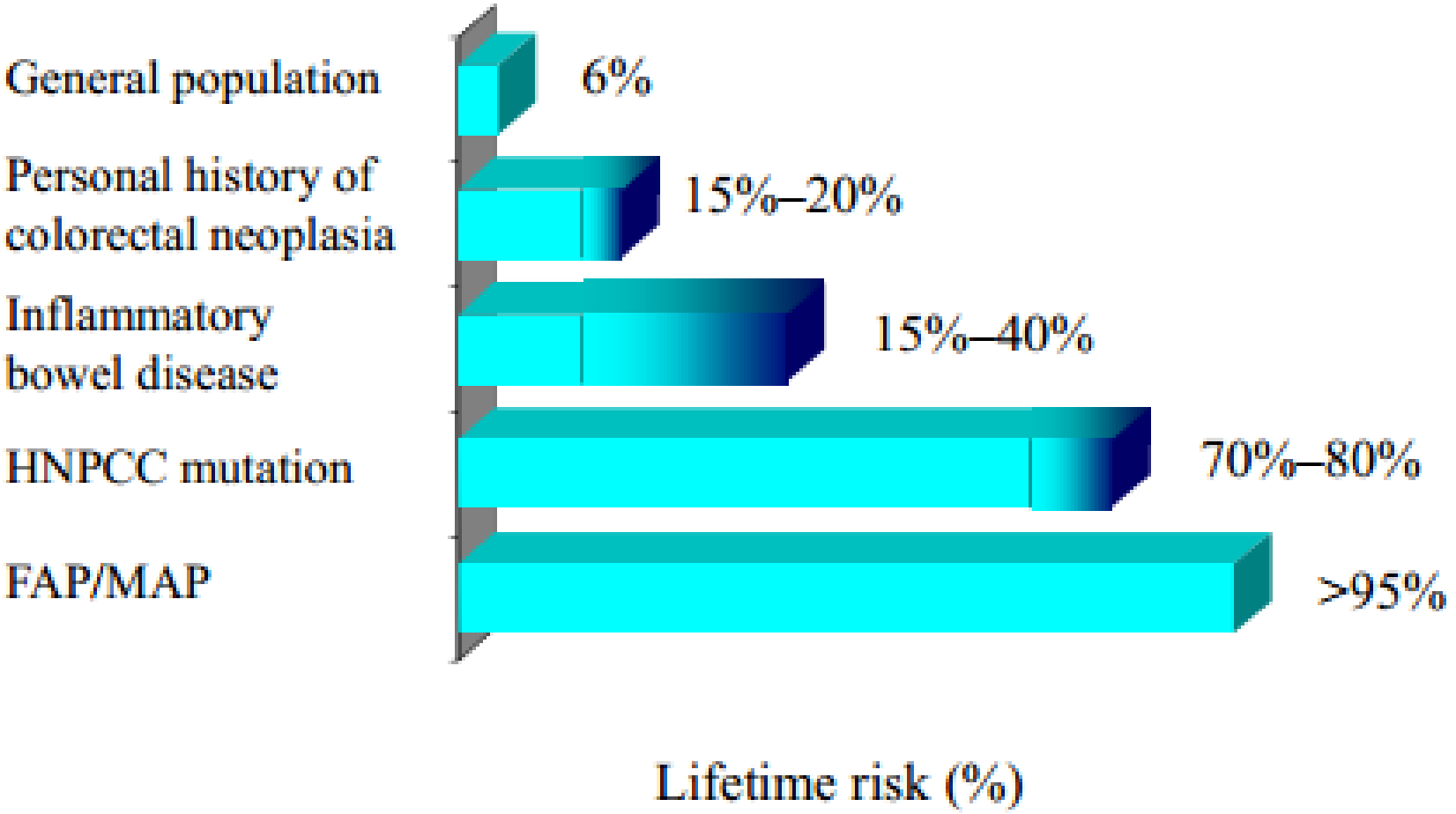
Genetic testing for Lynch Syndrome colorectal endometrial cancer families

As common as BRCA

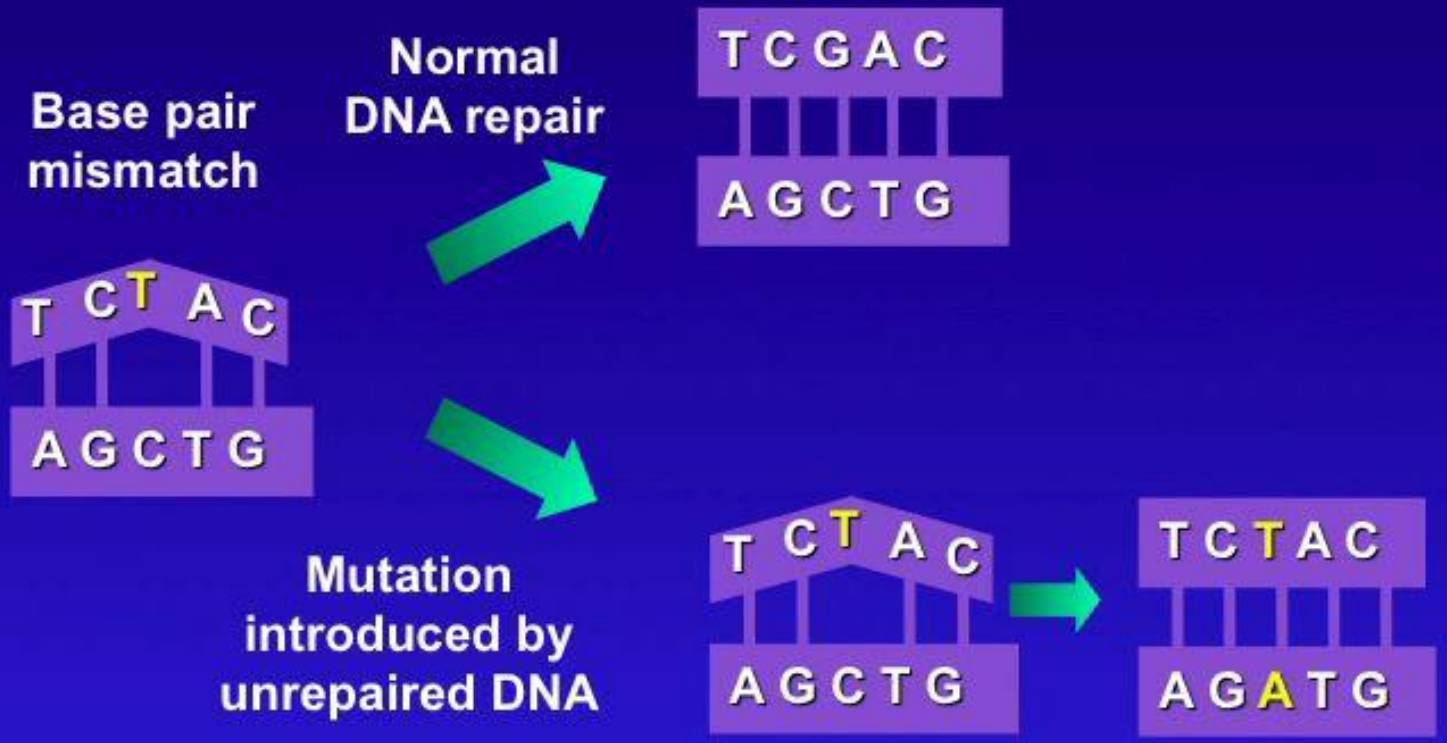
Result can save lives with as much or more impact than BRCA

Testing today in a fraction of the candidate patients

Risk of Colorectal Cancer



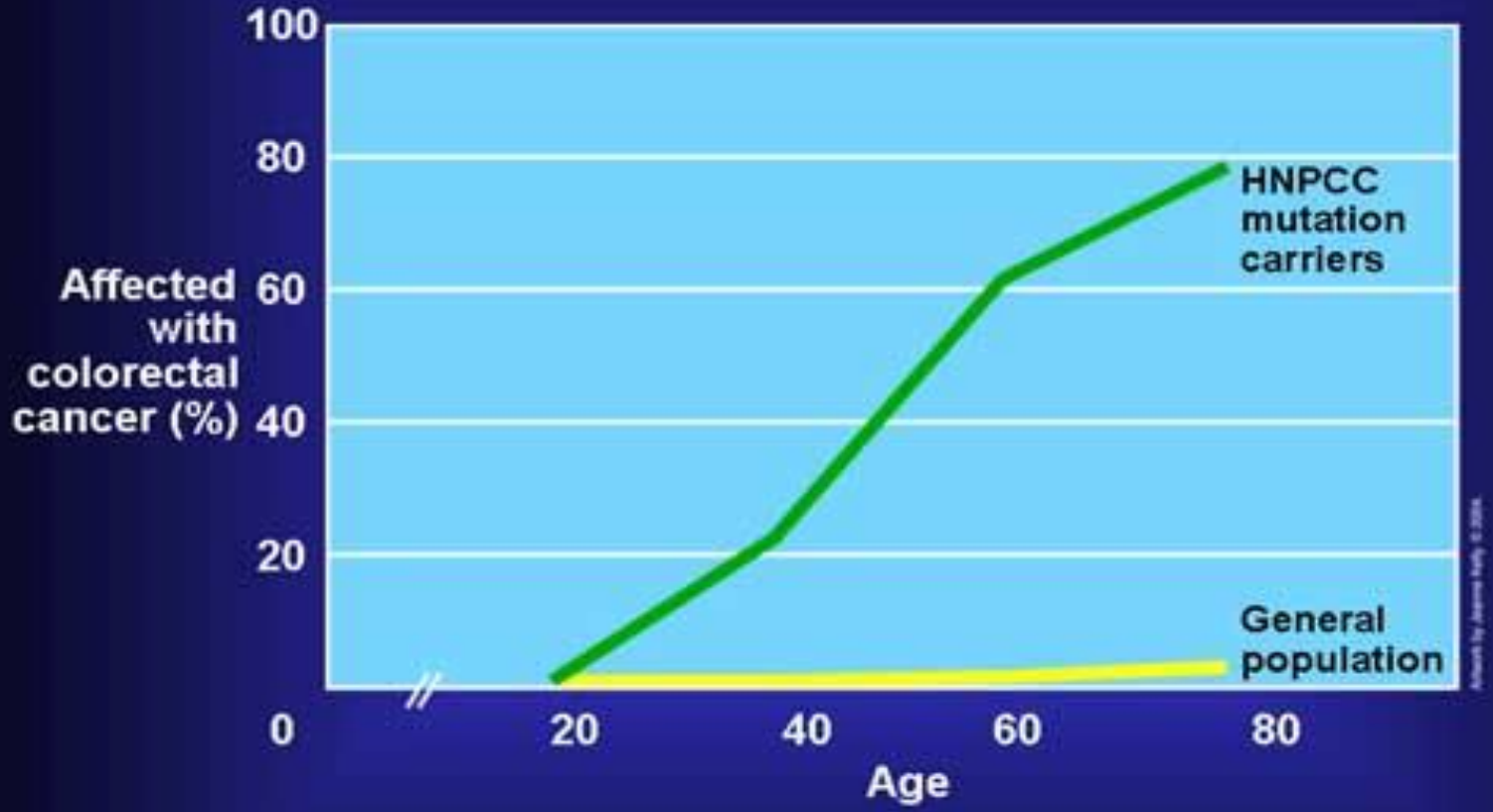
DNA Mismatch Repair



Testing Tumor for Lynch Syndrome

Tumor analysis-Screening for Lynch syndrome in the tumor (reliable in colon and uterine)
MSI-Microsatellite Instability
IHC-Immunohistochemistry

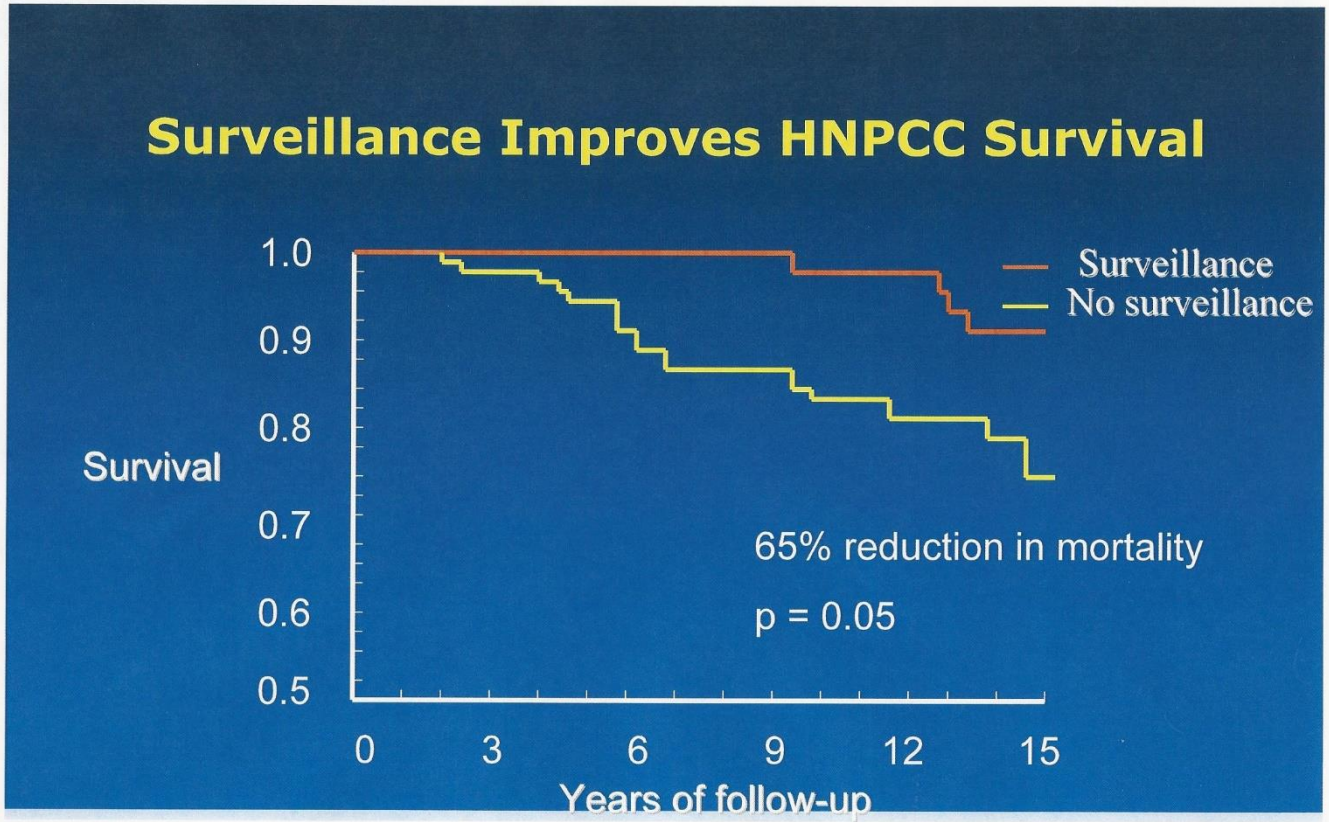
Age-Related Penetrance



Percentage of individuals with altered mismatch repair gene who develop cancer

NATIONAL CANCER INSTITUTE

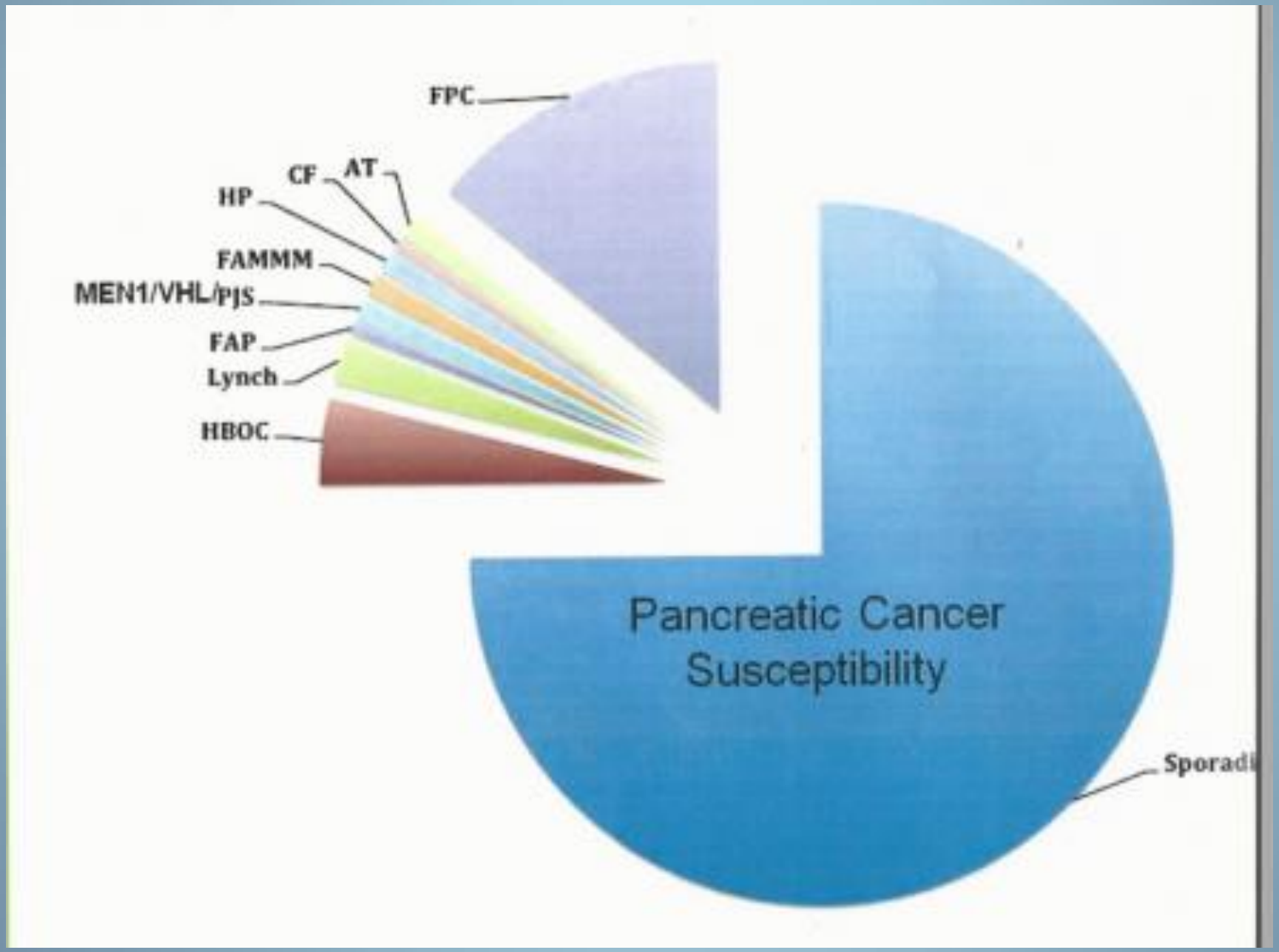
Surveillance Improves HNPCC Survival



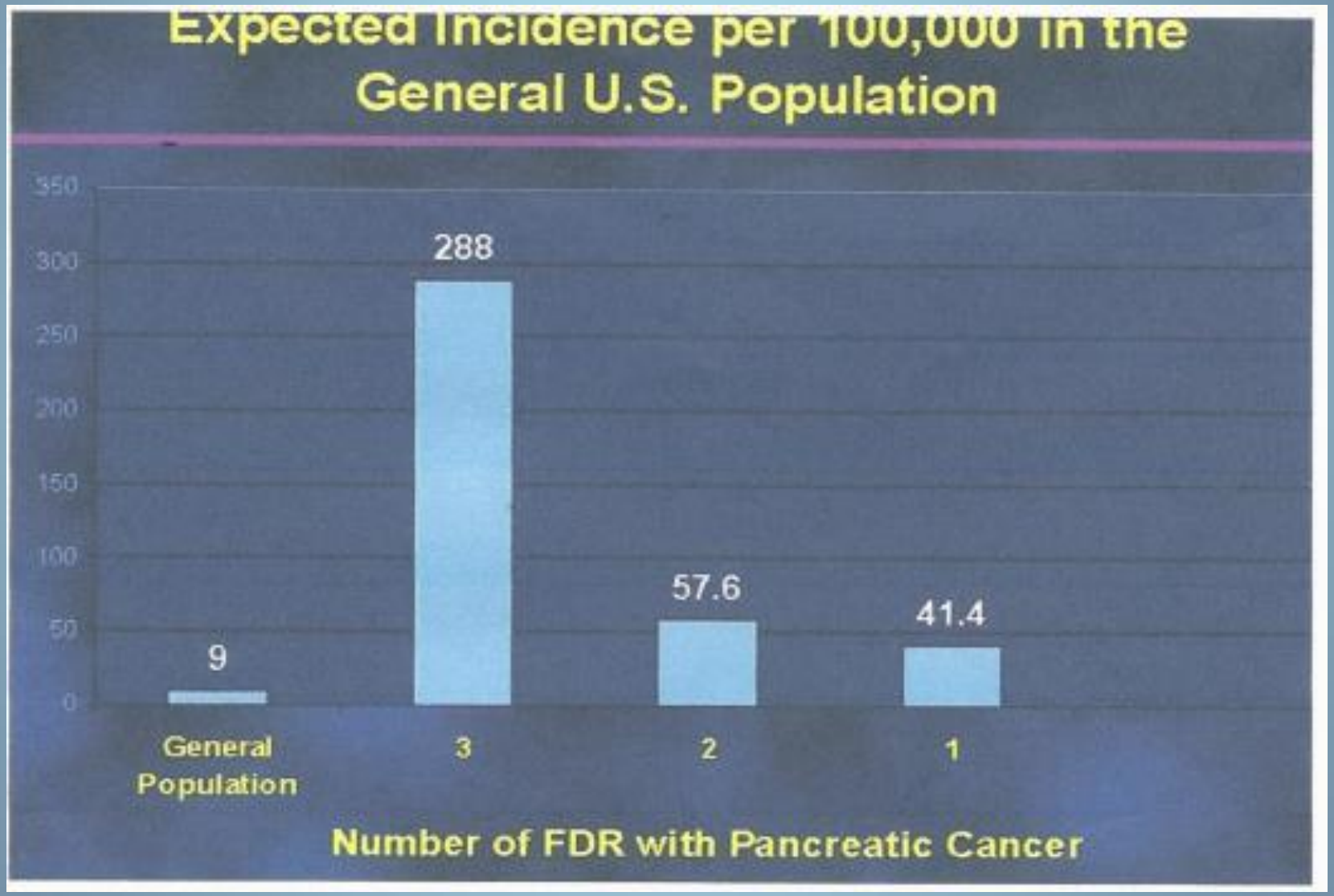
Jarvinen HJ, et al. *Gastroenterology*. 2000;118:829-834.

City of Hope

Pancreatic Cancer key considerations



Pancreatic Cancer



PANCREATIC CANCER

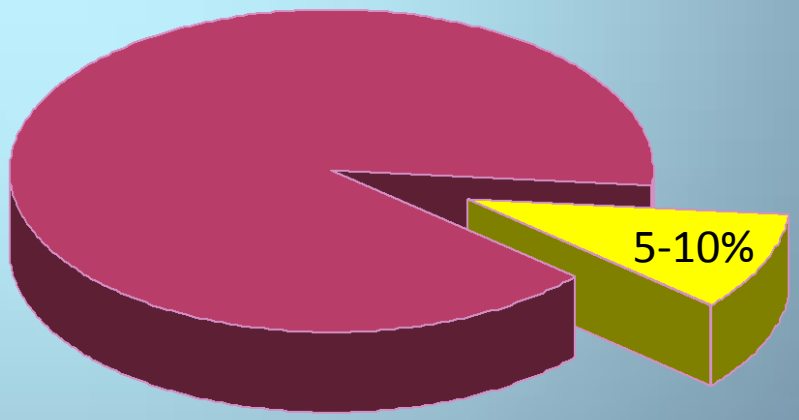
Mutation carriers to screen

- Mutation carriers
  - Peutz-Jeghers regardless of family history
  - BRCA2 with at least 1 FDR or 2 or more relatives with PC
  - PALB2 with at least 1 FDR with PC
  - P16 with at least 1 FDR with PC
  - Lynch syndrome with at least 1 FDR with PC

Prostate Cancer

- Most frequently diagnosed cancer in US men - 36% of all cancers
- Lifetime risk for men in US: 15-20%
- 200,000 new cases per year

- 5-10% is heritable
  - ~40% under 55y
  - Higher in families with breast/ovarian cancer



Prostate Cancer Germline Testing

- *ATM,*
- *BRCA1, BRCA2, CHEK2,PALB2, NBN, RAD 51*
- *HOXB13,*
- *MLH1, MSH2, MSH6, PMS2,* EPCAM
- *TP53*.

Inherited risk of Prostate cancer germline testing

- Early-onset prostate cancer (diagnosed ≤40 years of age)
- Multiple primary cancers in one person (*e.g*. prostate and male breast cancer)
- Personal history of prostate cancer and 1 or more family members* with breast cancer (diagnosed ≤50 years of age) and/or invasive ovarian cancer
- Personal history of prostate cancer and 2 family members with breast, pancreatic, or prostate cancer
- Associated with 10% detection in metastatic prostate cancer

Prostate Cancer Germline

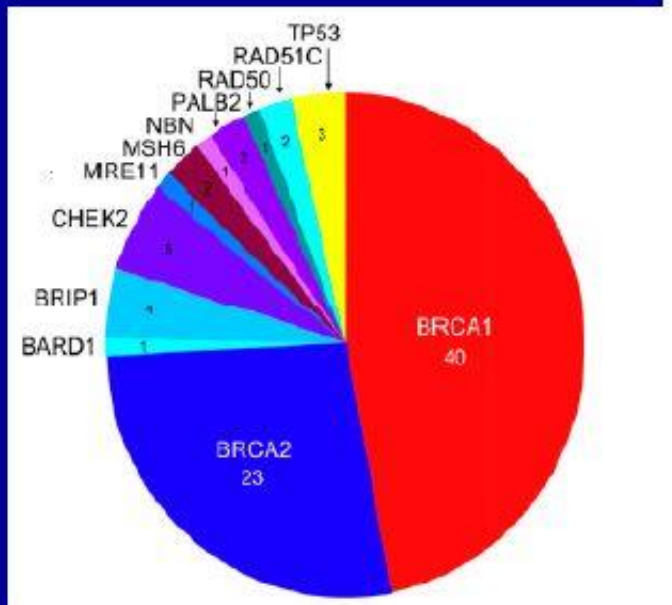
- Familial Prostate cancer risk not all explained by high penetrant genes
- Young age, high gleason score, mestatic disease along with other family history
- Actionability not standardized or proven: likey at minimum early implemenation of PSA
- Clinical Trials of PARP inhibitors suggests clinical management role
- May be Important to identify for family testing

Germline Analysis of 12 DNA Repair Genes in Women with Ovarian Cancer

- 360 women unselected for age and family history
  - 273 ovarian, 48 peritoneal, 31 FT, 8 synchronous endometrial & ovarian

- **24%** germline mutation
  - Loss of function
  - >2/3 in BRCA1 or BRCA2
  - 12 genes represented

- Of women with mutation:
  - 30% had no family history
  - 37% ≥ 60 years old at diagnosis



Walsh T et al. , PNAS, November 2011:108;10832-18037

Ovarian Cancer Testing

- Applies to epithelial ovarian cancer such as papillary serous
- Risk Reduction for at risk women results in clear demonstrated improved long term survival
- Some differences of age of onset may allow delayed prophylactic surgery
- Bilateral Salpino-oophorectomy standard
- BRCA testing for PARP inhibitor treatment in advanced disease
- First degree relative can be tested if affected member no available.

Estimated Ovarian Cancer Risk (Cumulative, assuming constant RR)

Age	Population	Average FDR Risk (RR 2.2)	BRIP1 (c-c) (RR 11.2)	BRIP1 (seg) (RR 3.41)	RAD51C (RR 5.2)	RAD51D (RR 12)
25	0.02%	0.05%	0.22%	0.11%	0.10%	0.23%
30	0.03%	0.07%	0.36%	0.17%	0.17%	0.38%
35	0.05%	0.11%	0.54%	0.25%	0.25%	0.58%
40	0.07%	0.16%	0.81%	0.40%	0.38%	0.87%
45	0.12%	0.26%	1.32%	0.65%	0.61%	1.41%
50	0.19%	0.42%	2.12%	0.99%	0.99%	2.27%
55	0.29%	0.64%	3.20%	1.40%	1.50%	3.43%
60	0.41%	0.91%	4.53%	1.91%	2.13%	4.85%
65	0.59%	1.24%	6.14%	2.54%	2.90%	6.57%
70	0.75%	1.65%	8.10%	3.27%	3.85%	8.66%
LTR (80)	**1.21%**	**2.64%**	**12.71%**	**4.06%**	**6.12%**	**13.56%**

Ovarian cancer risk management

- Cumulative risk ≥ FDR risk (2.64%) at:
  - 55 years (BRIP1 case-control RR, RAD51D)
  - 65 years (RAD51C)
  - 70 years (BRIP1 segregation RR)
- Consider RRSO around menopause (50)
- Age would be shifted younger if there is familial multiplier
- PALB2 risks are unclear as yet

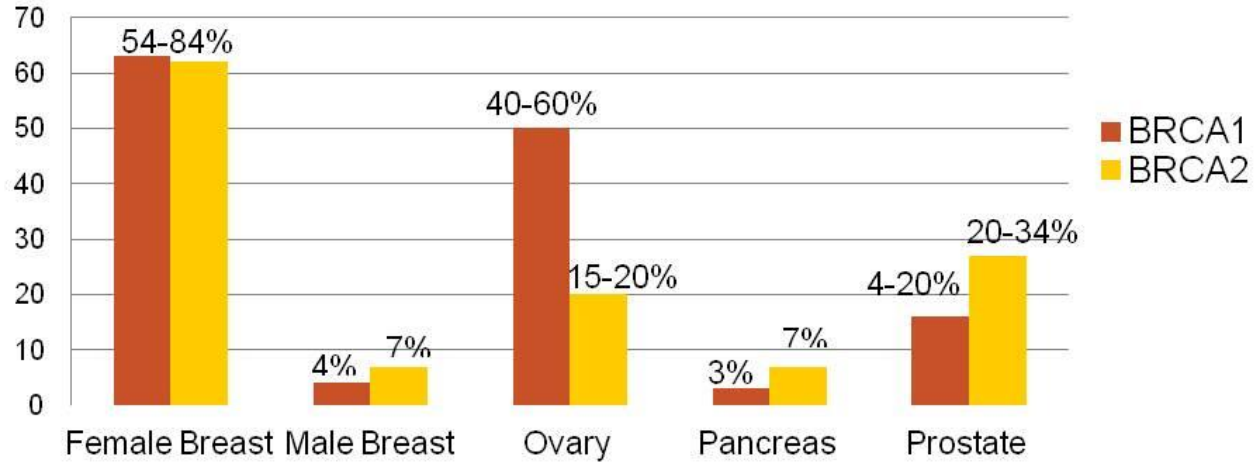
Tung, Domchek… Robson, NRCO in press 2016

Breast Cancer- Who

- Breast cancer < 45
- Triple negative breast cancer, under age 60
  - 11-28% have *BRCA1* mutations
- Two breast cancer primaries in a single individual
  - ~30% risk of second primary in 10 years for *BRCA1/2*
- Breast or ovarian cancer at any age in those of Ashkenazi Jewish ancestry
- Breast cancer at any age and…
  - ≥1 close relative* with breast cancer <45
  - ≥1 close relative* with epithelial ovarian cancer at any age
  - ≥2 close relatives* with breast cancer and/or pancreatic cancer at any age

NCCN Guidelines: Genetic/Familial High-Risk Assessment: Breast and Ovarian
*Includes third degree relatives

Cancer Risks in Carriers of Gemline Mutations in *BRCA1* and *BRCA2*



PRESENTED AT: ASCO Annual '15 Meeting

Presented By Judy Garber at 2015 ASCO Annual Meeting

Management Guidelines *BRCA1/2* Carriers

Management Option	Screening Interval/Comments
SCREENING	
• Clinical Breast Exam • Breast MRI • Mammogram	• Q6-12 mos beginning age 25 • Yearly age 25-75 (then individualize) • Yearly age 30-75 (then individualize)
• Transvaginal ultrasound* • CA-125*	• Q6 mos beginning age 30 • Q6 mos beginning age 30
PREVENTION	
• Bilateral mastectomy • Bilateral salpingo-oophorectomy	• Discuss option with patient • Recommend by age 35-40 and when childbearing complete
• Consider oral contraceptive • Consider tamoxifen	

Prophylactic mastectomy

- Decision making prevention vs bilateral as part of new diagnosis and treatment
- Age associated risk
- Accuracy of lifetime risk
- Survival outcome data
- Surgical complications
- Evidence based decision making

Breast cancer risk and surgical Prophylaxis: How the numbers work

- Risk is an annual risk number. BRCA 1 1.5-3% per year BRCA 2 1-1.5% per year
- BRCA 1, greater risk prior to age 50, BRCA 2 evenly distributed
- BRCA 1: 85% is ER negative, BRCA 2 15% is ER positive
- Nipple Sparing mastectomy, risk of breast cancer 1-4%
- Different cultures, individuals view the risks differently
- **Accurate risk estimates

Estimating Risk of Second Primary

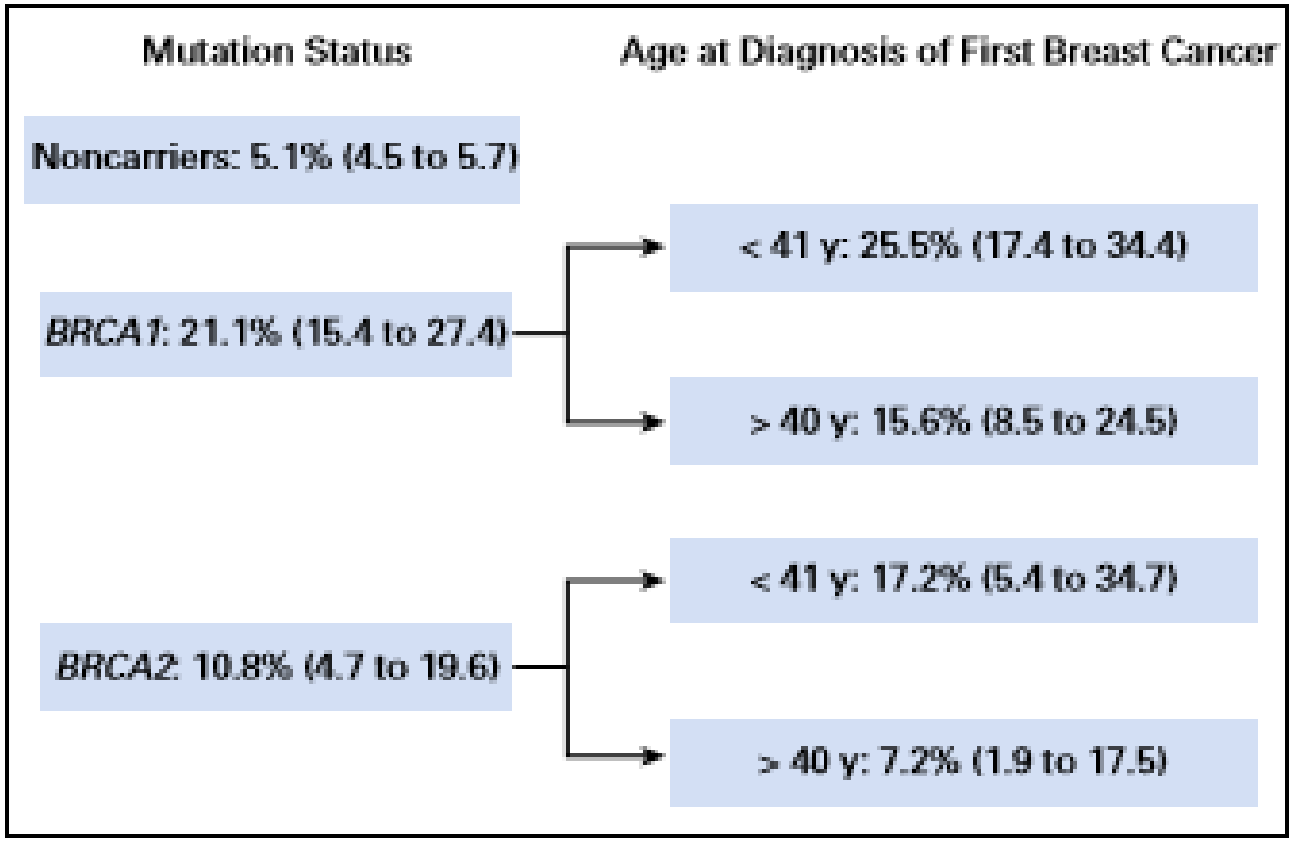
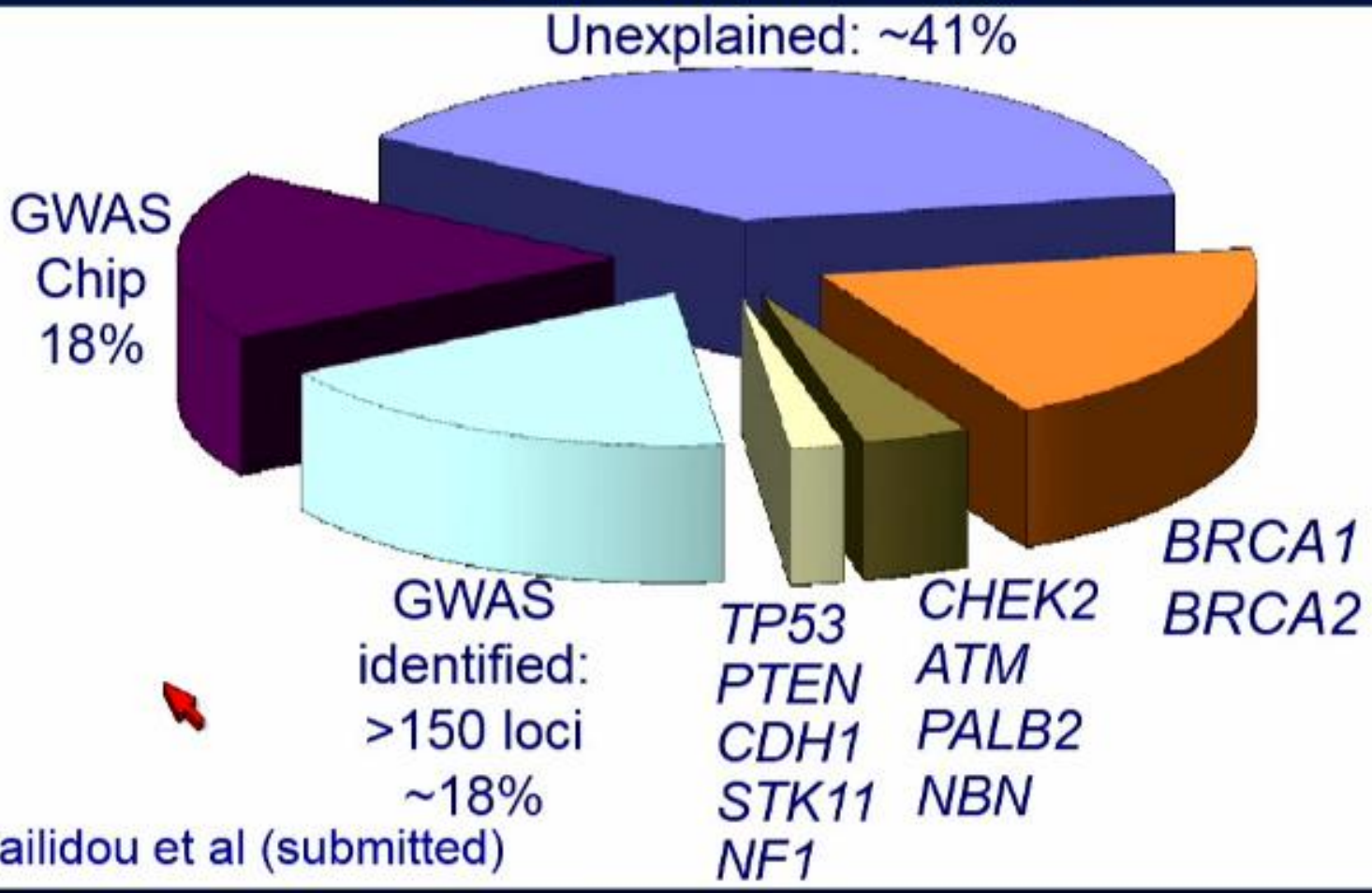


Fig 3. Summary of the 10-year cumulative contralateral breast cancer risks for noncarriers and *BRCA1/2* mutation carriers, stratified on the risk predictor (age) of this study in patients younger than 50 years of age. Numbers in brackets are 95% confidence intervals.

PALB-2

- Partner and Localizer of BRCA 2
- Homologous DNA repair
- Risk of Breast Cancer overall is 33% by age 70
- Risk is increased to 58% if family history greater than two first degree relatives
- accounts for 2.4% of familial aggregates of breast cancer
- Associated with increased risk of pancreatic cancer

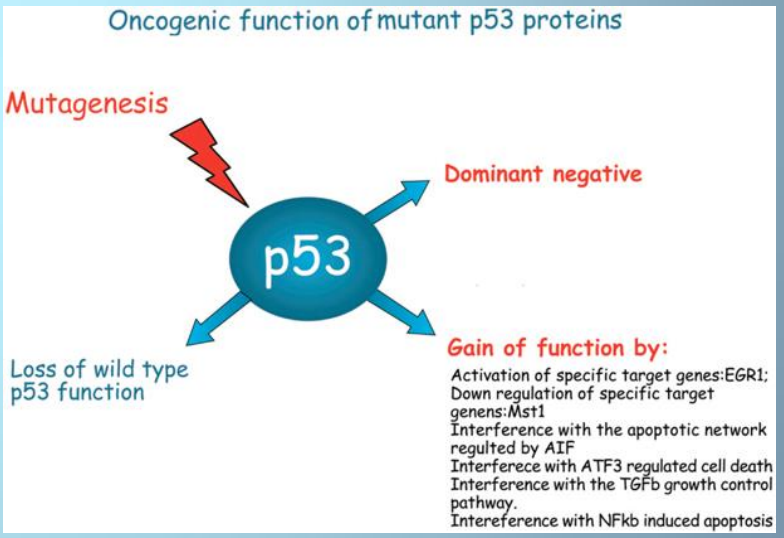
Familial aggregation explained



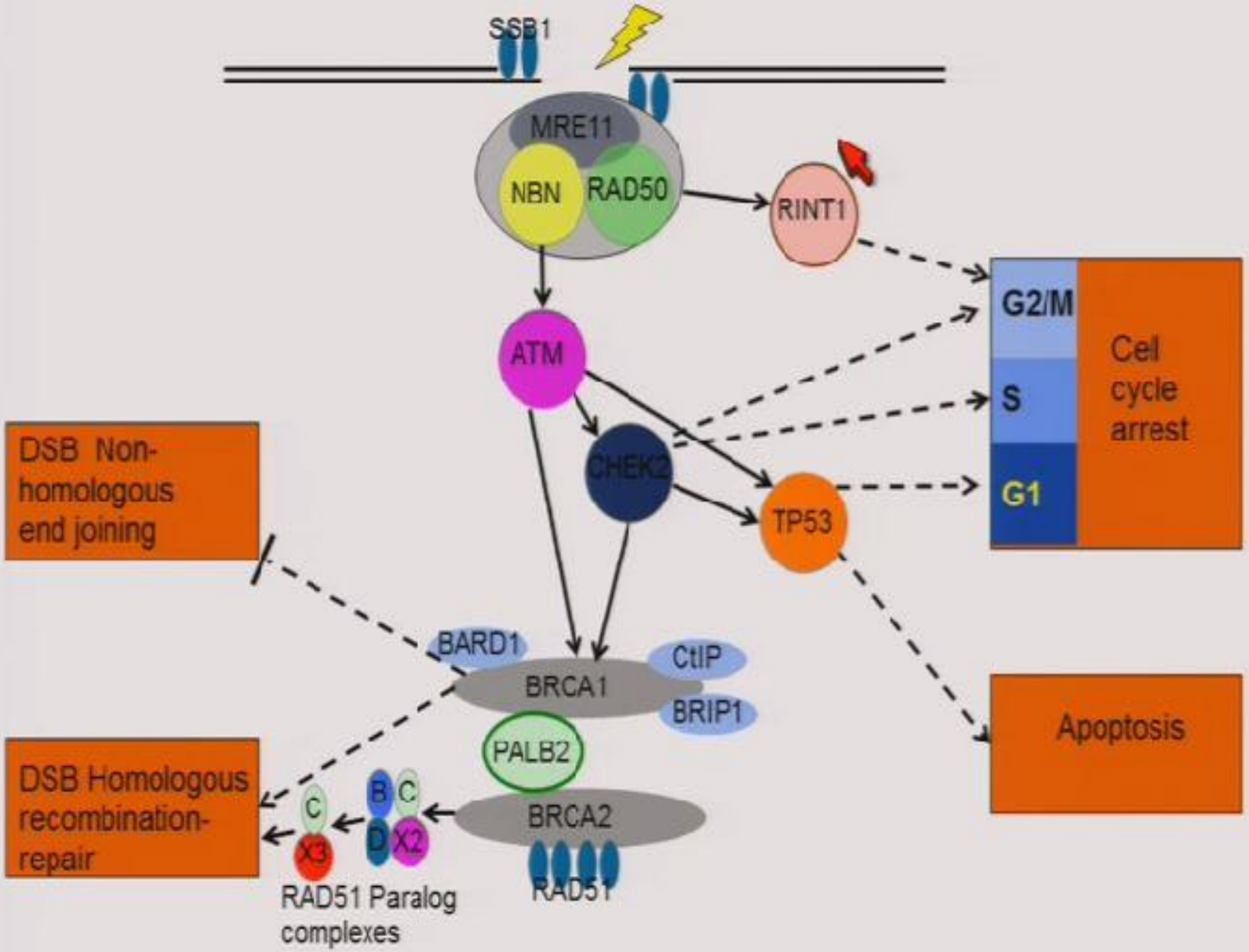
Michailidou et al (submitted)

Li-Fraumeni Syndrome

- Prevalence: Up to 1 in 20,000
- Inheritance: Autosomal dominant
- Gene: *TP53*
- Lifetime risk of cancer:
  - 50% by age 30-35y
  - 90% by 60y
  - Female lifetime risk is 90%
  - Male lifetime risk is 70%
  - 57% risk of a second primary



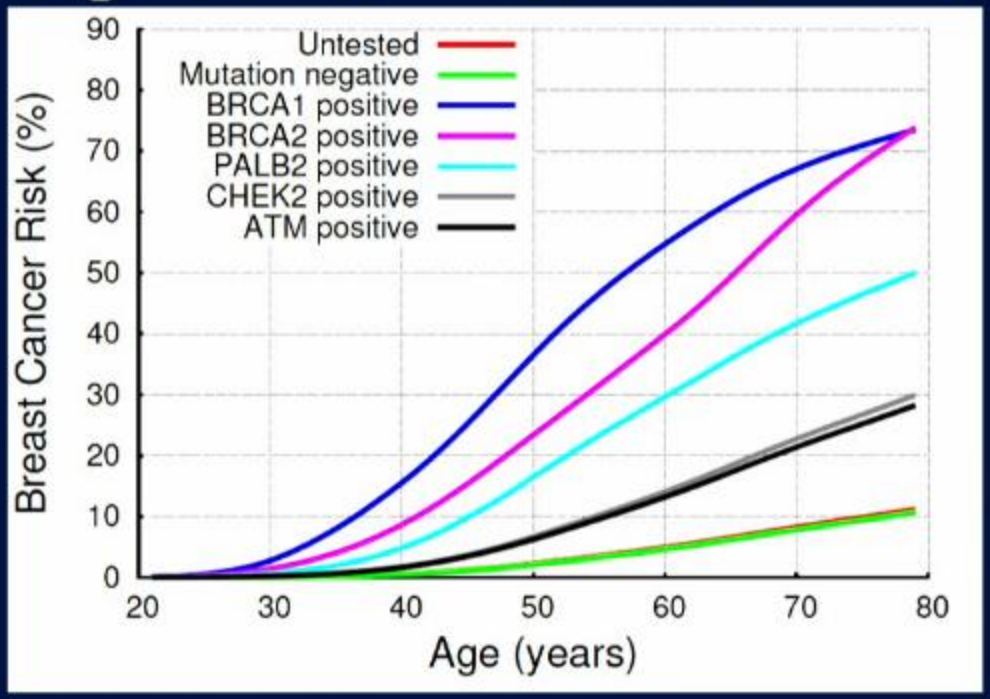
Core Genes in the Homologous Recombination-Repair Pathway



Moderate Risk Germline mutations: CHEK2

- CHEK 2 DS DNA break repair
- Most well know is 1100delC mutation: 2-3 fold RR
- 1100dlC and family history RR is 5 or 37% by age 70
- Homozygous does occur, increases risk multiple cancers
- More common in Europe

BRCA1, BRCA2, PALB2, ATM and *CHEK2* average breast cancer risks in BOADICEA



Lee et al, Genet Med (2016)

Risks from:
Antoniou et al, NEJM (2014)
Easton et al, NEJM (2015)
Weischer et al, JCO (2008)

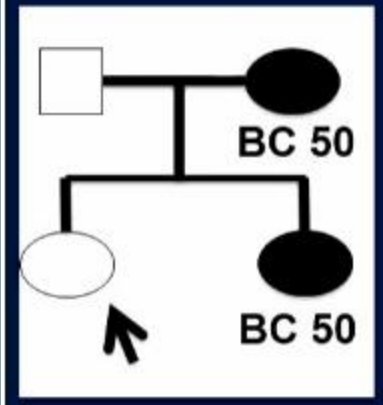
Estimated average 5-year risks (constant RR)

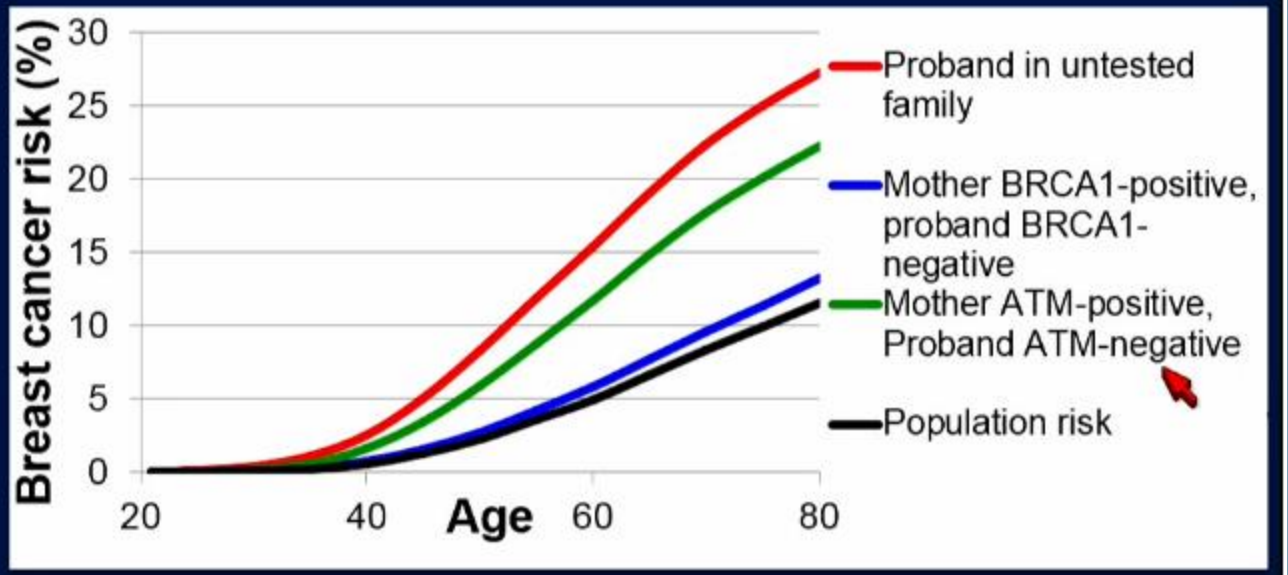
	Population	ATM/NBN (RR 2.7-2.8)*	CHEK2 (1100delC) (RR 3.0)‡	CHEK2 (I157T) (RR 1.58)	PALB2
Age	5 year incidence	5 year incidence	5 year incidence	5 year incidence	5 year incidence
25-29	0.04%	0.12%	0.13%	0.07%	0.35%
30-34	0.14%	0.38%	0.41%	0.21%	1.05%
35-39	0.30%	0.84%	0.90%	0.48%	2.5%
40-44	0.61%	1.70%	1.83%	0.96%	4.25%
45-49	0.94%	2.64%	2.83%	1.49%	6.35%
50-54	1.12%	3.14%	3.36%	1.77%	8.00%
55-59	1.33%	3.71%	3.98%	2.09%	7.25%
60-64	1.72%	4.81%	5.15%	2.71%	7.35%
65-69	2.11%	5.92%	6.34%	3.34%	5.95%
70-75	2.20%	6.17%	6.61%	3.48%	6.70%

Other risk genes

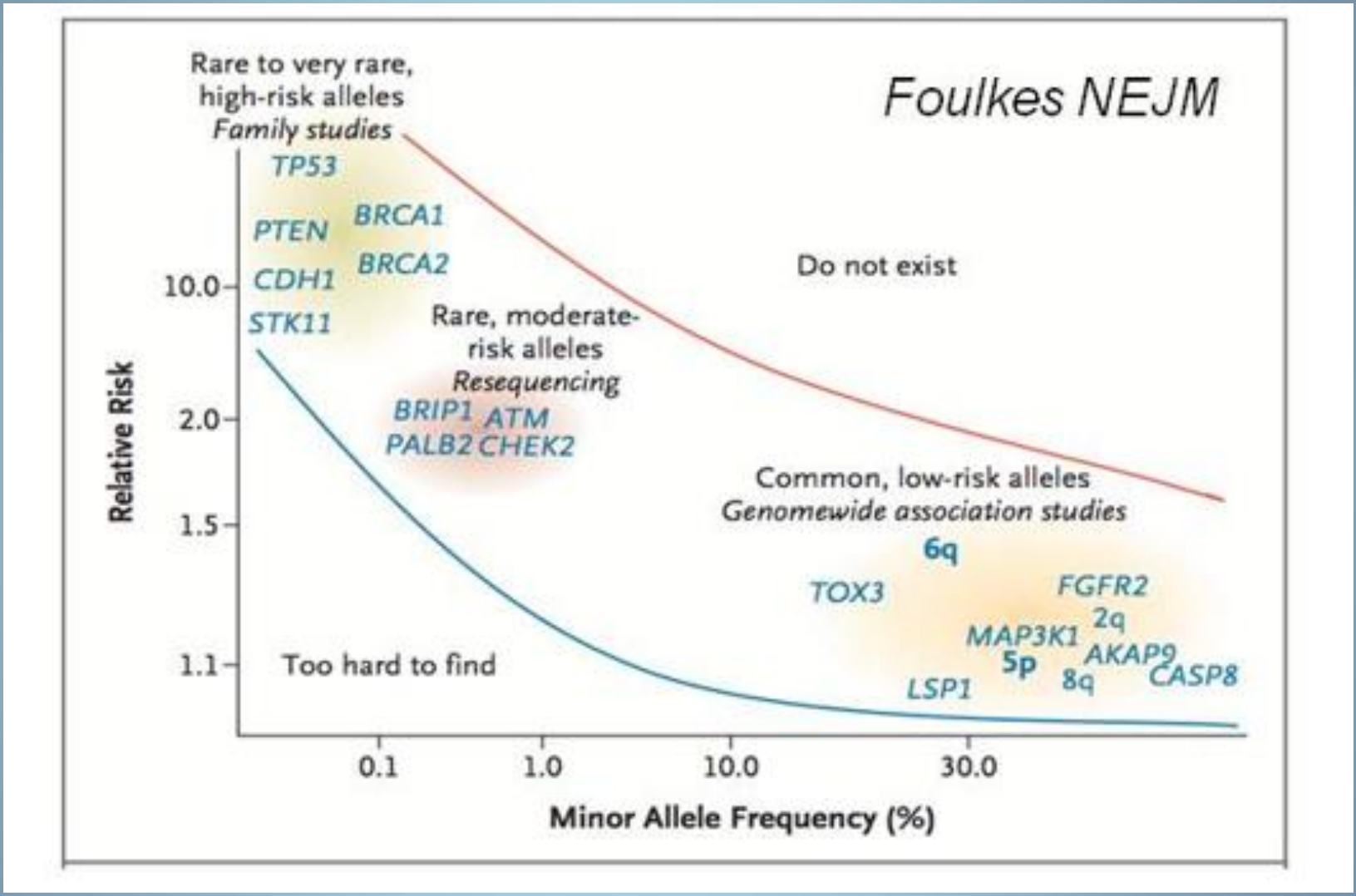
- MRE11A minimal data
- NBN: Slavic heterozygous may have 2-3 fold increase risk breast
- RAD50: very little data
- BARD1 very little data
- BRIP1 increase ovarian cancer in Finland
- RAD 51 mainly ovarian cancer risk
- MUTYH: homozygous know, but heterozygous very little data

Negative predictive testing





Lee et al, Genet Med (2016)


Foulkes NEJM
Rare to very rare, high-risk alleles
Family studies
TP53
PTEN
BRCA1
BRCA2
CDH1
STK11
Do not exist
Rare, moderate-risk alleles
Resequencing
BRIP1
ATM
PALB2
CHEK2
Common, low-risk alleles
Genomewide association studies
6q
TOX3
FGFR2
2q
MAP3K1
5p
AKAP9
8q
CASP8
LSP1
Too hard to find
Relative Risk
10.0
2.0
1.5
1.1
0.1
1.0
10.0
30.0
Minor Allele Frequency (%)

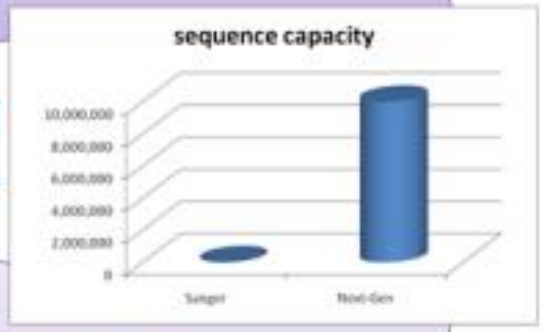
Breast Cancer and Ovarian Cancer and Panel Testing

- Testing has become clinically available on a widespread basis for only about 1-2 years
- Most labs offer wide variety of panels
- First data are recently published on results of panel testing
- Issues of VUS and estimates of cancer and incidental findings are common to all cancers
- Which panels, and what are the main issues
- What are the results in Breast, ovarian cancer in early use of testing

Sanger Sequencing

Hundreds of bases / read
Hundreds of reads per run

Thousands of bases per run



Multiple Sequence Reactions

Next-Generation Sequencing

"Massively Parallel Sequencing"

Hundreds of bases / read
Millions of reads per run

10s of BILLIONS of bases per run

Gene V Panel V Exome v Genome

	Single Gene	Gene Panel	Exome	Genome
Target Size	5000	500,000	50,000,000	3,000,000,000
Method	Sanger sequence	NGS	NGS	NGS
Estimated Variants	0-3	50	100,000	38,000,000

base pairs analyzed

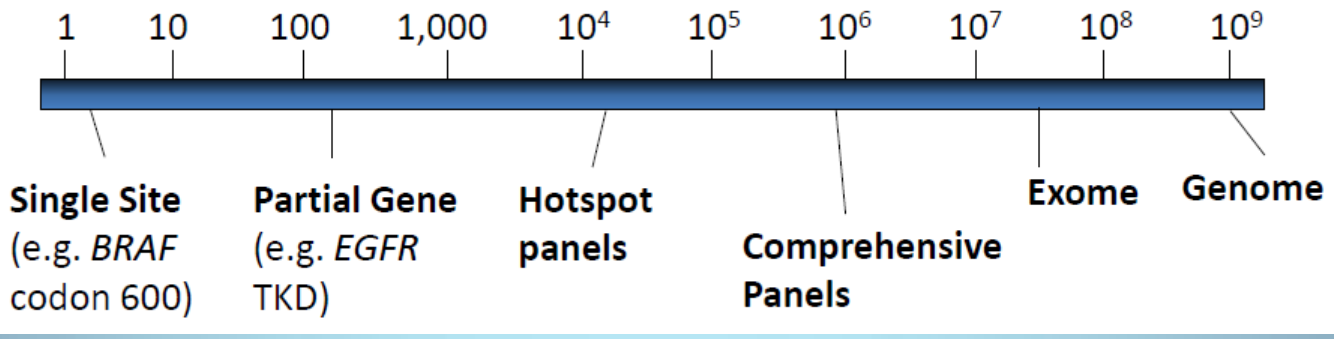


Table 1. Genes Analyzed in Commercially Available Multiplex Panels

Gene	Ambry Genetics*				University of Washington Laboratory Medicine†	
	CancerNext	BreastNext	ColoNext	OvaNext	BROCA	ColoSeq
APC	●		●		●	●
ATM	●	●		●	●	
ATR					●	
BABAM1					●	
BAP1					●	
BARD1	●	●		●	●	
BMPR1A			●		●	
BRIP1	●	●		●	●	
CDH1	●	●	●	●	●	●
CDK4					●	
CDKN2A					●	
CHEK1					●	
CHEK2	●	●	●	●	●	
FAM175A/Abraxas					●	
MLH1	●		●	●	●	●
MRE11A	●	●		●	●	
MSH2-positive *EPCAM*	●		●	●	●	●
MSH6	●		●	●	●	●
MUTYH	●	●	●	●	●	●
NBN	●	●		●	●	
PALB2	●	●		●	●	
PMS2	●		●	●	●	●
PRSS1					●	
PTEN	●	●	●	●	●	●
RAD50	●	●		●	●	
RAD51					●	
RAD51B					●	
RAD51C	●	●		●	●	
RAD51D					●	
RBBP8					●	
RET					●	
SMAD4	●		●		●	
STK11	●	●	●	●	●	●
TP53	●	●	●	●	●	●
TP53BP1					●	
UIMC1					●	
VHL					●	
XRCC2					●	
XRCC3					●	

*Aliso Viejo, CA.

Growing List of labs offering panel testing in genetics

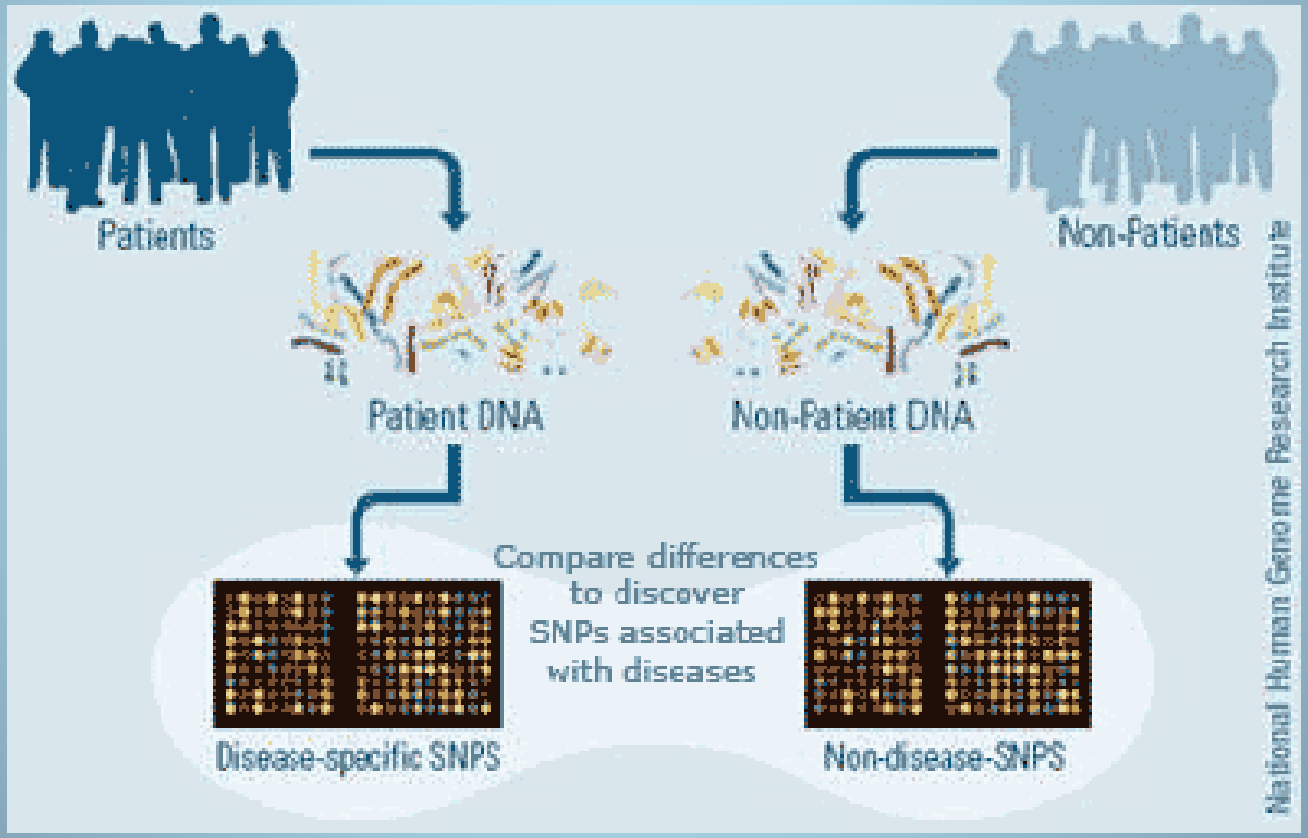
- **Ambry**
- **ApolloGen**
- **ARUP**
- **GeneDX**
- **Invitae**
- **ColorMe**
- **Myriad**
- **Pathway Genomics**
- **Prevention Genetics**
- **Quest**
- **University of Washington**
- **Fulgent Diagnostics**

Panel Testing Results for Familial Breast Cancer

Genes	N = # Patients with deleterious mutation (%)	95% Confidence interval
Any deleterious mutation	11 (10.4)	5.30, 17.81
BRCA1 or *BRCA2*	7 (6.6)	2.70, 13.13
BRCA1	4 (3.8)	1.04, 9.38
*BRCA2**	3 (2.8)	0.59, 8.05
Other genes related to breast cancer	5 (4.7)	1.55, 10.67
*ATM**	2 (1.9)	0.23, 6.65
CHEK2	1 (0.9)	0.02, 5.14
PALB2	2 (1.9)	0.23, 6.65

* One patient had a deleterious mutation in both *BRCA2* and *ATM*

GWAS: SNPs


Patients
Non-Patients
Patient DNA
Non-Patient DNA
Compare differences to discover SNPs associated with diseases
Disease-specific SNPS
Non-disease-SNPS
National Human Genome Research Institute

Understanding SNPS

- Single Nucleotide polymorphisms
- Relatively common: >1% of population
- Not known to be disease causing
- Usually but not always in “exomes” or protein mapping portion
- Investigations into possible role of combinations of SNPs impacting disease

Individual SNP associations

- Each SNP: 0, 1, 2 risk alleles
- Odds Ratio estimates per risk allele: 1.02-1.30
- Minor allele frequencies: >0.01

- Individual SNP predictive ability poor
- SNPs combine multiplicatively on risk scale

Michailidou et al, Nat Genet 2015

Types of Mutations

Normal Message:

THEBIGREDDOGRANOUT
THE BIG RED DOG RAN OUT

Deletions: Cause FRAMESHIFTS

THE BIG RED DOG RAN OUT
THE BI R EDD OGR ANO UT

Insertions:

THE BIG REB DOB GRA NOU TS

Normal Message:

THEBIGREDDOGRANOUT
THE BIG RED DOG RAN OUT

Deletions: Cause FRAMESHIFTS

THE BIG RED DOG RAN OUT
THE BI R EDD OGR ANO UT

Insertions:

THE BIG REB DOB GRA NOU TS

Variants of Unknown Significance

SUBSTITUTION

- THE BIG RED DOG RAN OUT
- THE BIG NED DOG RAN OUT

Types of Mutations

Normal Message:

THEBIGREDDOGRANOUT
THE BIG RED DOG RAN OUT

Deletions: Cause FRAMESHIFTS

THE BIG RED DOG RAN OUT
THE BI R EDD OGR ANO UT

Insertions:

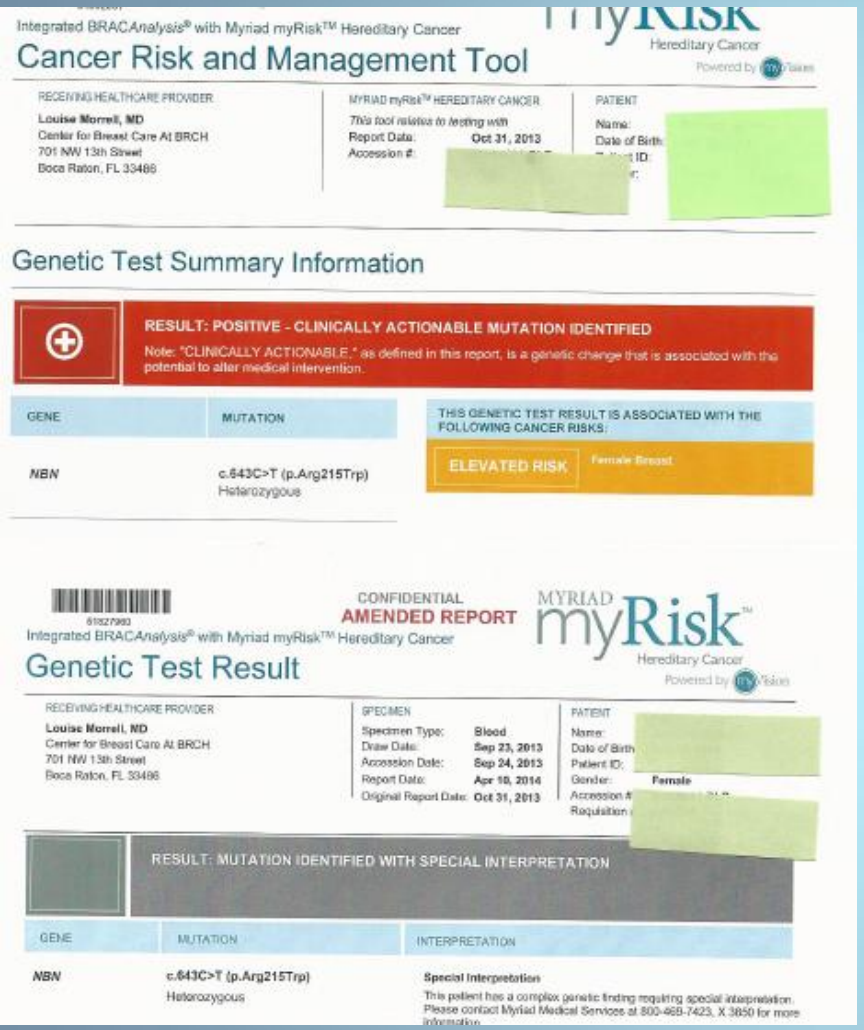
THE BIG REB DOB GRA NOU TS

Making Sense of a VUS

1. Amino acid conservation (charge, polarity, volume, hydrophobicity, etc.) and Grantham Matrix Score (GMS)
2. Prevalence in a control population (SNP?)
3. Co-segregation with disease in affected families
4. Location within gene and protein functionality
5. In silico analysis (Align-GVGD, PolyPhen, SIFT, NNsplice)
6. Evolutionary conservation
7. Concurrent with known deleterious mutations
8. LOH of wild type allele in tumors
9. Locus-specific databases (BIC, LOVD, InSiGHT, MMR Genes Variant Database, IARC TP53 Database)

VUS: Management Options

- Should not be managed as a deleterious mutation
- Estimate cancer risk based on clinical presentation and empiric risk
- No clear advantage to testing offspring
- May have different interpretations by different labs of same VUS
- Counsel patient that interpretation may change
- Participate in registry or PROMPT national data base



Integrated BRACAnalysis® with Myriad myRisk™ Hereditary Cancer

Cancer Risk and Management Tool

myRisk Hereditary Cancer — Powered by myVision™

RECEIVING HEALTHCARE PROVIDER
Louise Morrell, MD
Center for Breast Care At BRCH
701 NW 13th Street
Boca Raton, FL 33486

MYRIAD myRisk™ HEREDITARY CANCER
This test relates to testing with
Report Date: **Oct 31, 2013**
Accession #:

PATIENT
Name:
Date of Birth:
Patient ID:

Genetic Test Summary Information

RESULT: POSITIVE - CLINICALLY ACTIONABLE MUTATION IDENTIFIED

Note: "CLINICALLY ACTIONABLE," as defined in this report, is a genetic change that is associated with the potential to alter medical intervention.

GENE	MUTATION
NBN	c.643C>T (p.Arg215Trp) Heterozygous

THIS GENETIC TEST RESULT IS ASSOCIATED WITH THE FOLLOWING CANCER RISKS:

ELEVATED RISK — Female Breast

CONFIDENTIAL
AMENDED REPORT

Integrated BRACAnalysis® with Myriad myRisk™ Hereditary Cancer

MYRIAD myRisk™ Hereditary Cancer — Powered by myVision™

Genetic Test Result

RECEIVING HEALTHCARE PROVIDER
Louise Morrell, ND
Center for Breast Care At BRCH
701 NW 13th Street
Boca Raton, FL 33486

SPECIMEN
Specimen Type: **Blood**
Draw Date: **Sep 23, 2013**
Accession Date: **Sep 24, 2013**
Report Date: **Apr 10, 2014**
Original Report Date: **Oct 31, 2013**

PATIENT
Name:
Date of Birth:
Patient ID:
Gender: **Female**
Accession #:
Requisition #:

RESULT: MUTATION IDENTIFIED WITH SPECIAL INTERPRETATION

GENE	MUTATION	INTERPRETATION
NBN	c.643C>T (p.Arg215Trp) Heterozygous	**Special Interpretation** This patient has a complex genetic finding requiring special interpretation. Please contact Myriad Medical Services at 800-469-7423, X 3850 for more information.

CONFIDENTIAL
AMENDED REPORT

Integrated BRACAnalysis® with Myriad myRisk™ Hereditary Cancer

MYRIAD myRisk™ Hereditary Cancer — Powered by myVision™

myRisk Genetic Result

RECEIVING HEALTHCARE PROVIDER
Louise Morrell, MD
Center for Breast Care At BRCH
701 NW 13th Street
Boca Raton, FL 33486

SPECIMEN
Specimen Type: **Blood**
Draw Date: **Sep 23, 2013**
Accession Date: **Sep 24, 2013**
Report Date: **Oct 02, 2014**
Original Report Date: **Oct 31, 2013**

PATIENT
Gender: **Female**

This amended report reflects a change in the interpretation of this individual's previous test result. The NBN variant c.643C>T (p.Arg215Trp) was originally reported as a deleterious mutation associated with elevated cancer risk (see report dated October 31, 2013). This variant was then classified as a deleterious mutation with regards to Nijmegen breakage syndrome (see report dated April 10, 2014), and is now being reclassified to a "variant of uncertain clinical significance" with regards to both Nijmegen breakage syndrome and hereditary cancer risks. While this variant has been identified in conjunction with a second deleterious truncating NBN mutation in an individual affected with Nijmegen breakage syndrome, we have recently identified the c.643C>T (p.Arg215Trp) variant on both copies of the NBN gene (homozygous) in an individual who does not have obvious signs of Nijmegen breakage syndrome. This suggests that this variant may not be associated Nijmegen breakage syndrome. Therefore, we are reclassifying this variant to a "variant of uncertain clinical significance." Please see the accompanying letter for more information.

RESULT: NEGATIVE - NO CLINICALLY SIGNIFICANT MUTATION IDENTIFIED

Note: "CLINICALLY SIGNIFICANT," as defined in this report, is a genetic change that is associated with the potential to alter medical intervention.

ADDITIONAL FINDINGS: VARIANT(S) OF UNCERTAIN SIGNIFICANCE (VUS) IDENTIFIED

GENE	VARIANT(S) OF UNCERTAIN SIGNIFICANCE	INTERPRETATION
NBN	c.643C>T (p.Arg215Trp)	**UNCERTAIN CLINICAL SIGNIFICANCE** There are currently insufficient data to determine if these variants cause increased cancer risk.

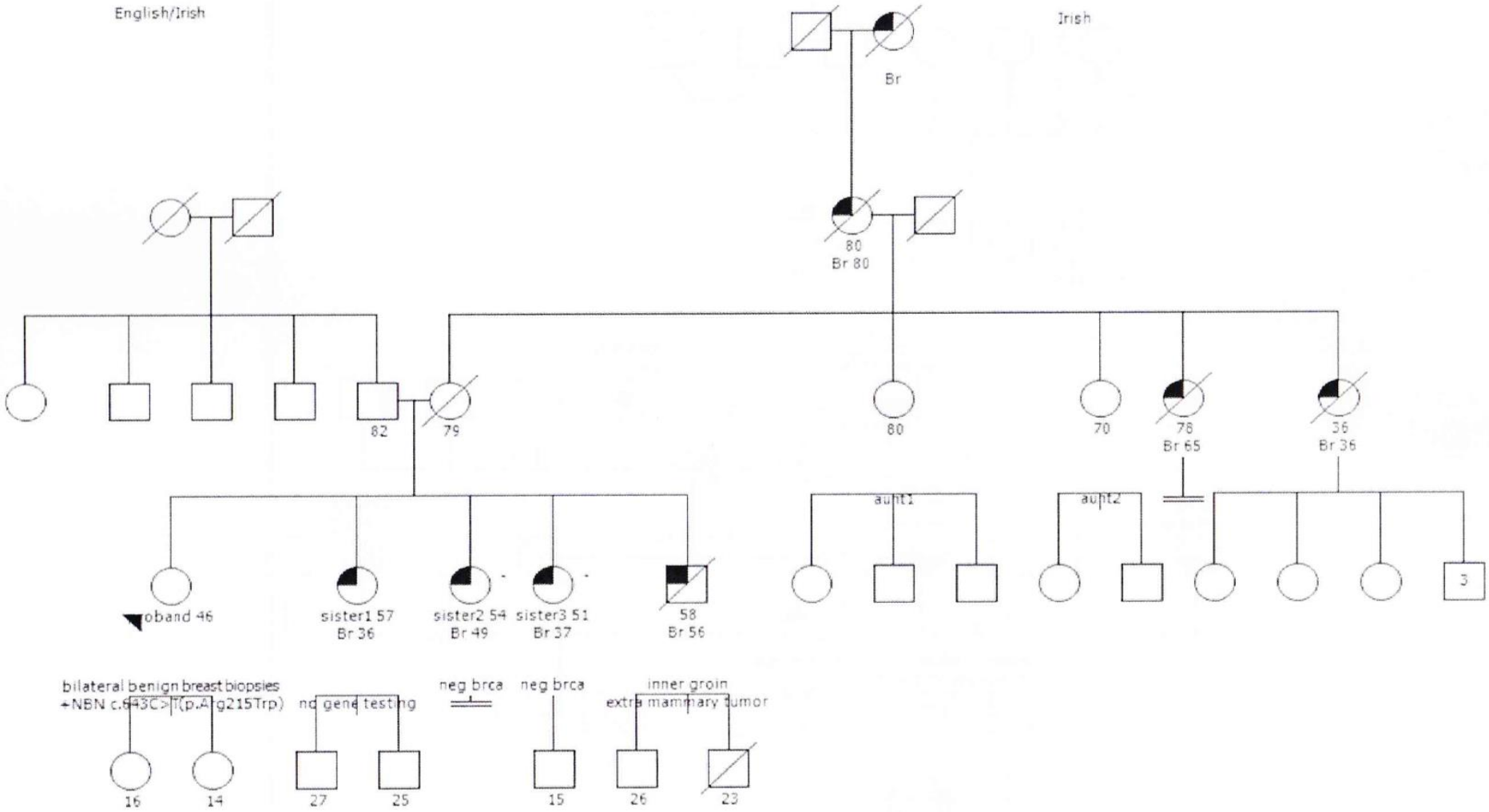
MORE DETAILS ABOUT: *NBN* c.643C>T (p.Arg215Trp): NM_002485.4

The heterozygous germline *NBN* variant c.643C>T is predicted to result in the substitution of tryptophan for arginine at amino acid position 215 of the *NBN* protein (p.Arg215Trp). Biochemical analysis indicates that this mutation impairs normal *NBN* function (di Masi A et al., Biochemical and Biophysical Research Communications 2008, 369: 835-840).

While the c.643C>T variant has been previously identified in conjunction with a deleterious truncating *NBN* mutation in an individual affected with Nijmegen breakage syndrome (a rare autosomal recessive disorder), we have also observed this variant on both copies of the *NBN* gene (homozygous) in an individual who does not have obvious signs of Nijmegen breakage syndrome (Seemanova E et al., J Natl Cancer Inst 2007, 99:1875-80; Myriad internal data). This suggests that this variant may not be associated with Nijmegen breakage syndrome. Available clinical data indicate that this mutation may not be associated with increased breast cancer risk (Seemanova E et al., J Natl Cancer Inst 2007, 99:1875-80; Desjardins S et al., BMC Cancer 2009, 9:181; Steffen J et al., Int J Cancer 2004, 111:67-71; Gao P et al., Mutagenesis 2013, 28:683-697). Therefore, patient cancer-risk management should be based upon personal and family history, as well as other clinical factors.

Details About Non-Clinically Significant Variants: All individuals carry DNA changes (i.e., variants), and most variants do not increase an individual's risk of cancer or other diseases. When identified, variants of uncertain significance (VUS) are reported. Likely benign variants (Favor Polymorphisms) and benign variants (Polymorphisms) are not reported and available data indicate that these variants most likely do not cause increased cancer risk. Present evidence does not suggest that non-clinically significant variant findings be used to modify patient medical management beyond what is indicated by the personal and family history and any other clinically significant findings.

Variant Classification: Myriad's myVision™ Variant Classification Program performs ongoing evaluations of variant classifications. In certain cases, healthcare providers may be contacted for more clinical information or to arrange family testing to aid in variant classification. When new evidence about a variant is identified and determined to result in clinical significance and management change, that information will automatically be made available to the healthcare provider through an amended report.


English/Irish
Irish
Br
80
Br 80
82
79
80
70
78
Br 65
56
Br 36
aunt1
aunt2
3
oband 46
sister1 57
Br 36
sister2 54
Br 49
sister3 51
Br 37
58
Br 56
bilateral benign breast biopsies
+NBN c.643C>T(p.Arg215Trp)
no gene testing
neg brca
neg brca
inner groin
extra mammary tumor
16
14
27
25
15
26
23

Risk Calculator Tool

www.ask2me.org

Comparison Shopping

Company	Technology	Cost
23 & Me	GW-SNP	$99-$199
Counsyl	Targeted SNP	$350
Pathway G.	Targeted SNP	$100s-varys
deCODEme	GW-SNP	$2000
Navigenics	Targeted SNP	Variable
Knome	WGS	$10-40K
Illumina	WGS	$10-40K

Direct to consumer testing

- Ancestry.com
- 23andMe
- 3 purposes: (1) identity seeking (2) disease risk information (3) curiosity or lifestyle
- Uses SNPs for identity
- Previous restriction from testing, now allowed to test for medical traits that can be passed down from both parents

Conclusion

- Germline genetic testing is standard of care for high risk individuals when test results will change patient management
- Best established in breast, ovarian cancer, and lynch syndrome
- Panel testing is readily available
- Panel testing results in identification of additional gene testing when patients undergoing testing are BRCA negative about 10% of the time.
- Risk estimates are likely to be variable based on the clinical context
- The availability of genetic testing data is being made available of the
- Cancer Risk Estimates, proven utility needs much more work

Inherited Genetics and germline testing

- Most Cancers are not due to identifiable high risk genes
- Clinical usefulness depends on accuracy, validity and utility: accurate estimates of risk plus acitionability
- Accurate estimates of risk require knowledge of family history
- Increasingly available panels results in “population Screening” which lacks utility, increased variants
- Key identification: look at one side of family at a time, look for young age, multiple cancers in one individual, bilateral disease