Penn Medicine

Hereditary Breast Cancers: An Update

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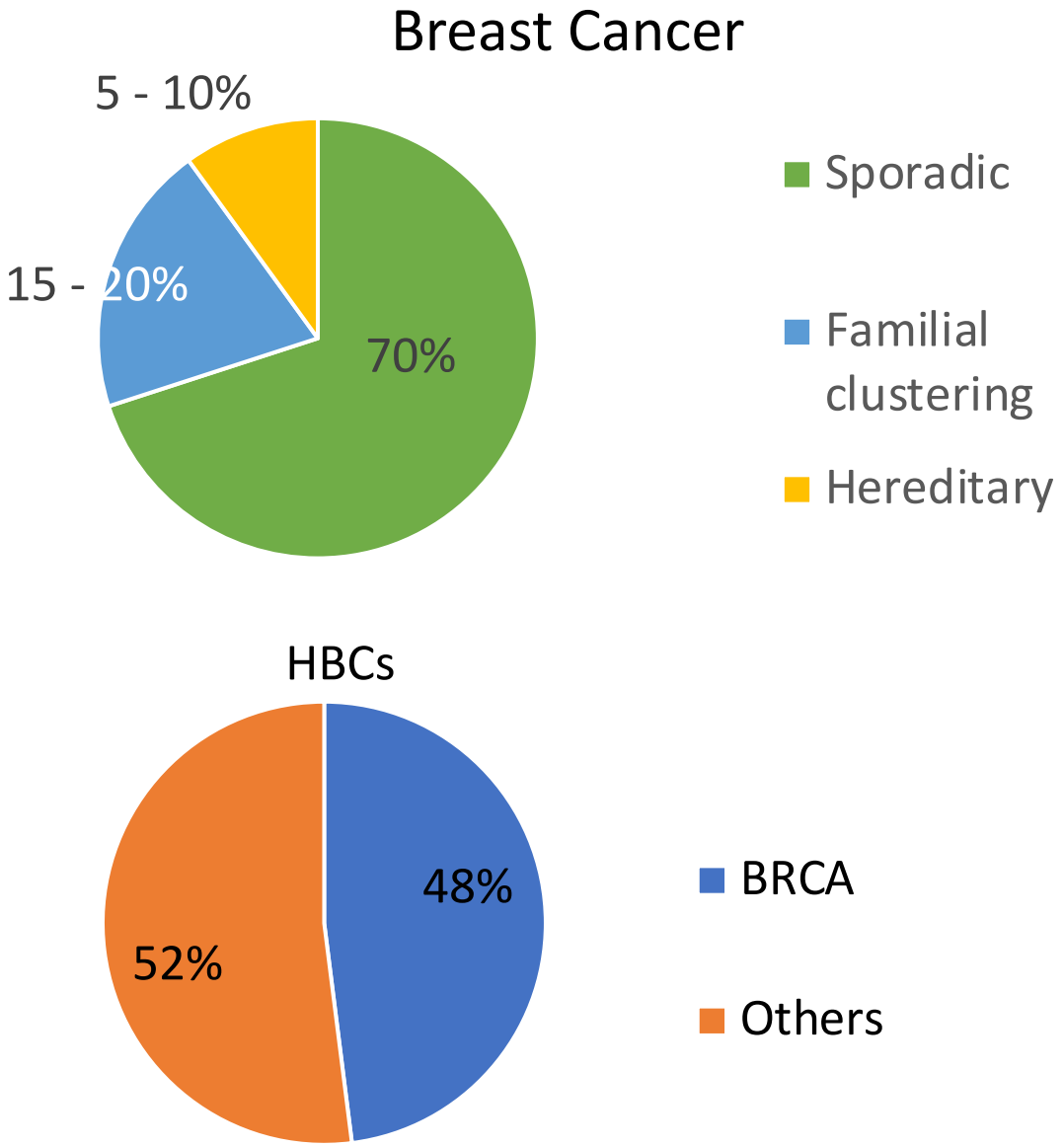
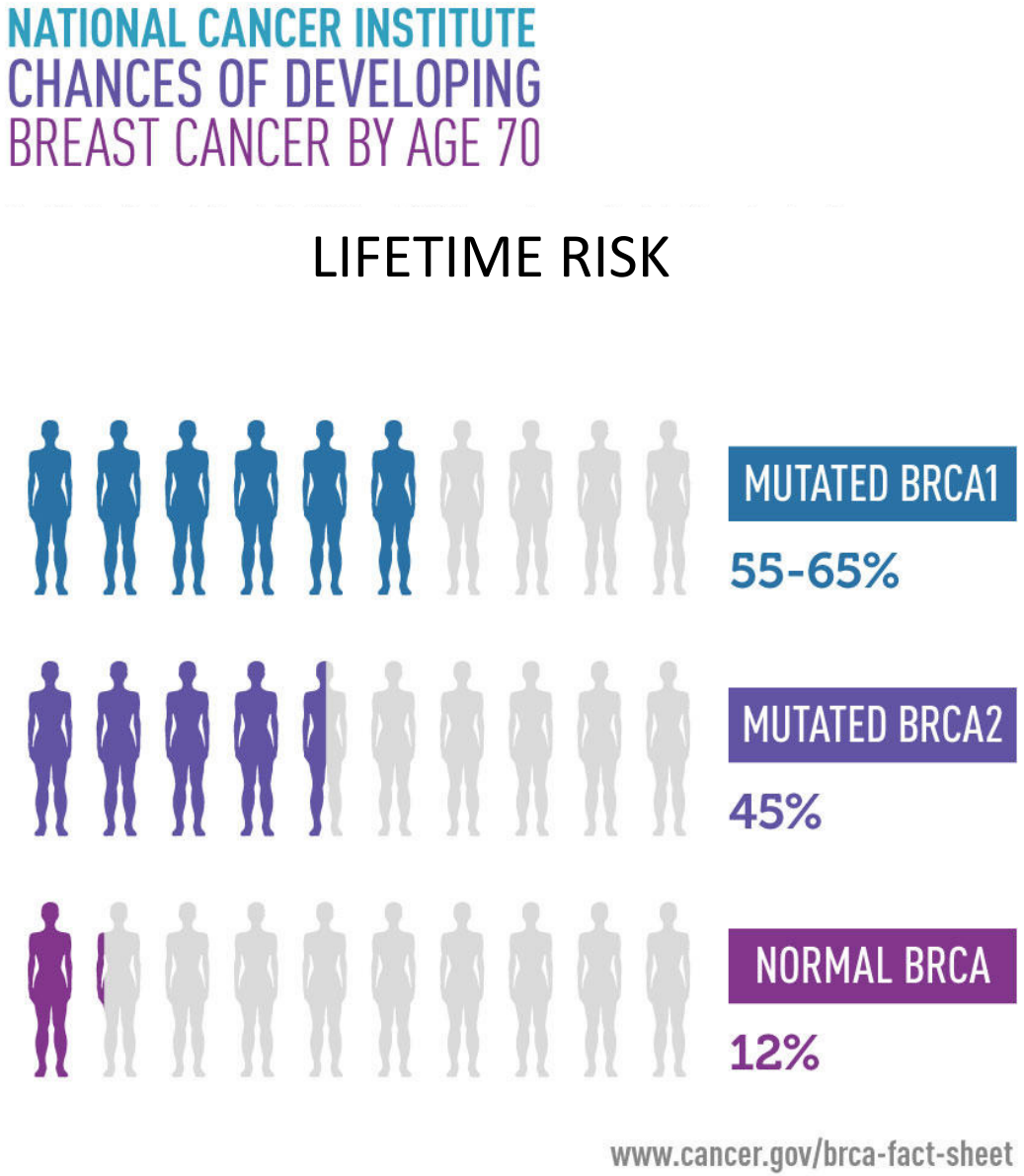
Content

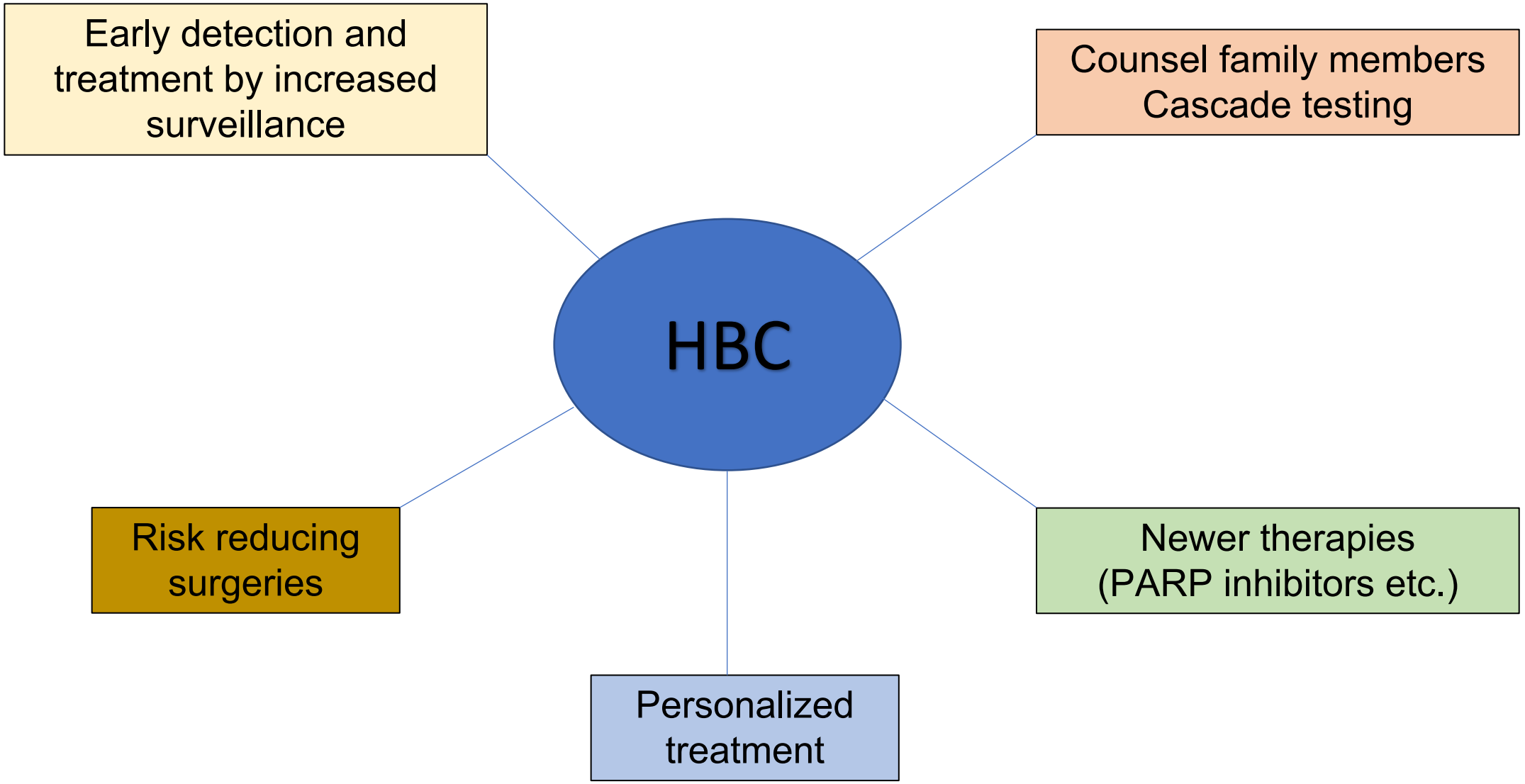
- Newly recognized non-*BRCA* genes in familial predisposition to breast cancer
- Current NCCN guidelines for genetic testing in patients with breast cancer
- Challenges with germline testing
- Polygenic risk score
- Emerging therapies and predictive tests in the treatment of hereditary breast cancer

Hereditary Breast Cancer (HBC)



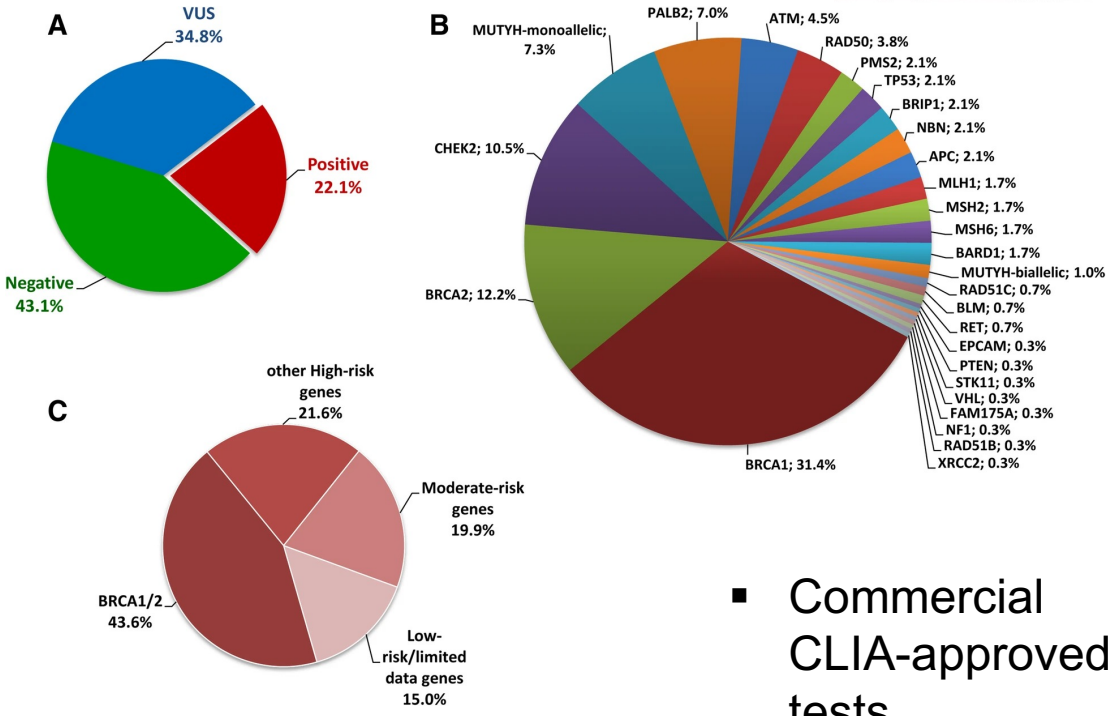


WHY is it important to recognize HBCs by germline testing?



Loss of ability to patent genes in 2013

Next Gen Sequencing



- Commercial CLIA-approved tests
- Direct-to-consumer tests

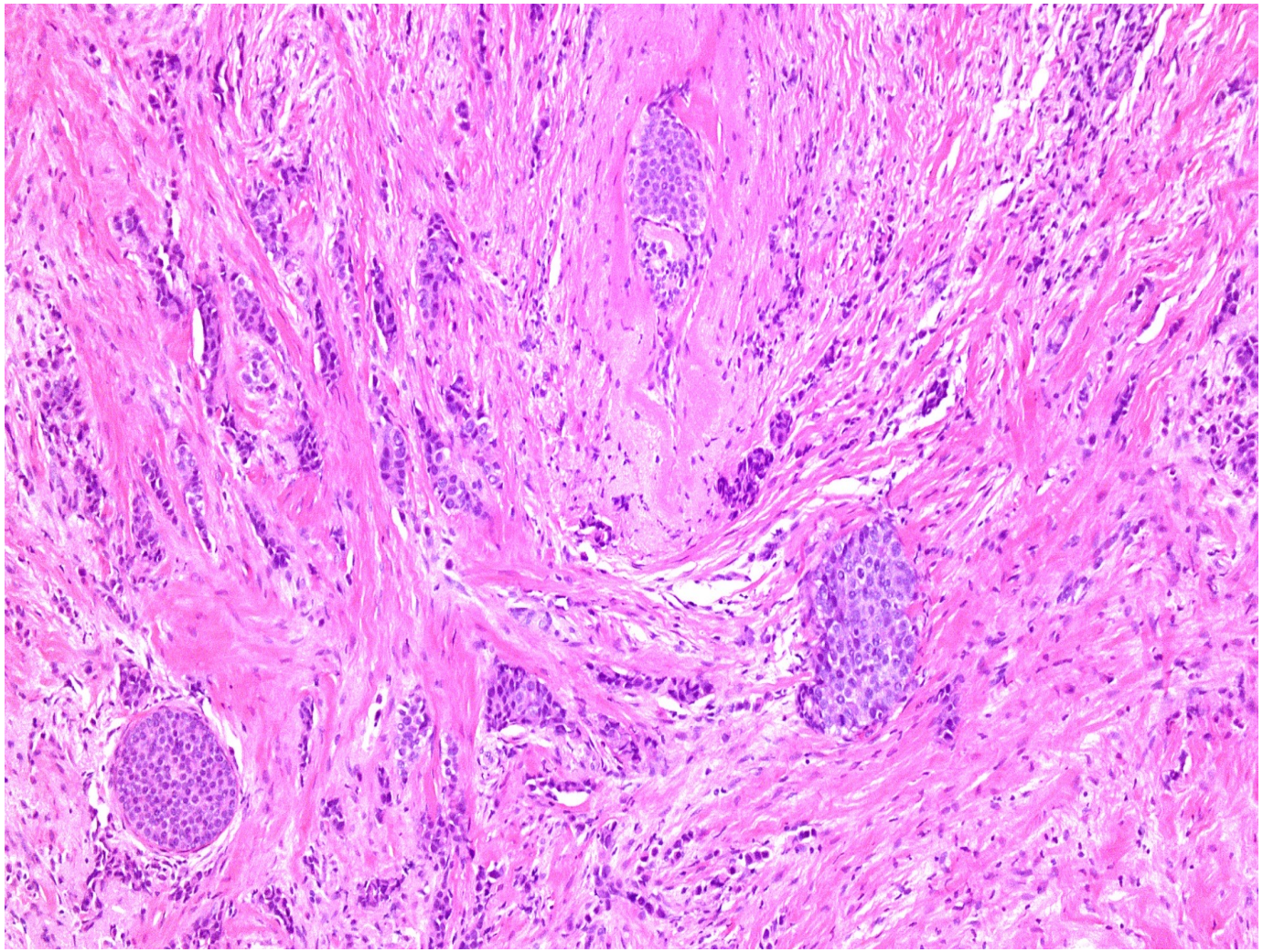
Multigene Panel Tests: A New Paradigm in HBC Testing

Germline Testing in breast Cancer

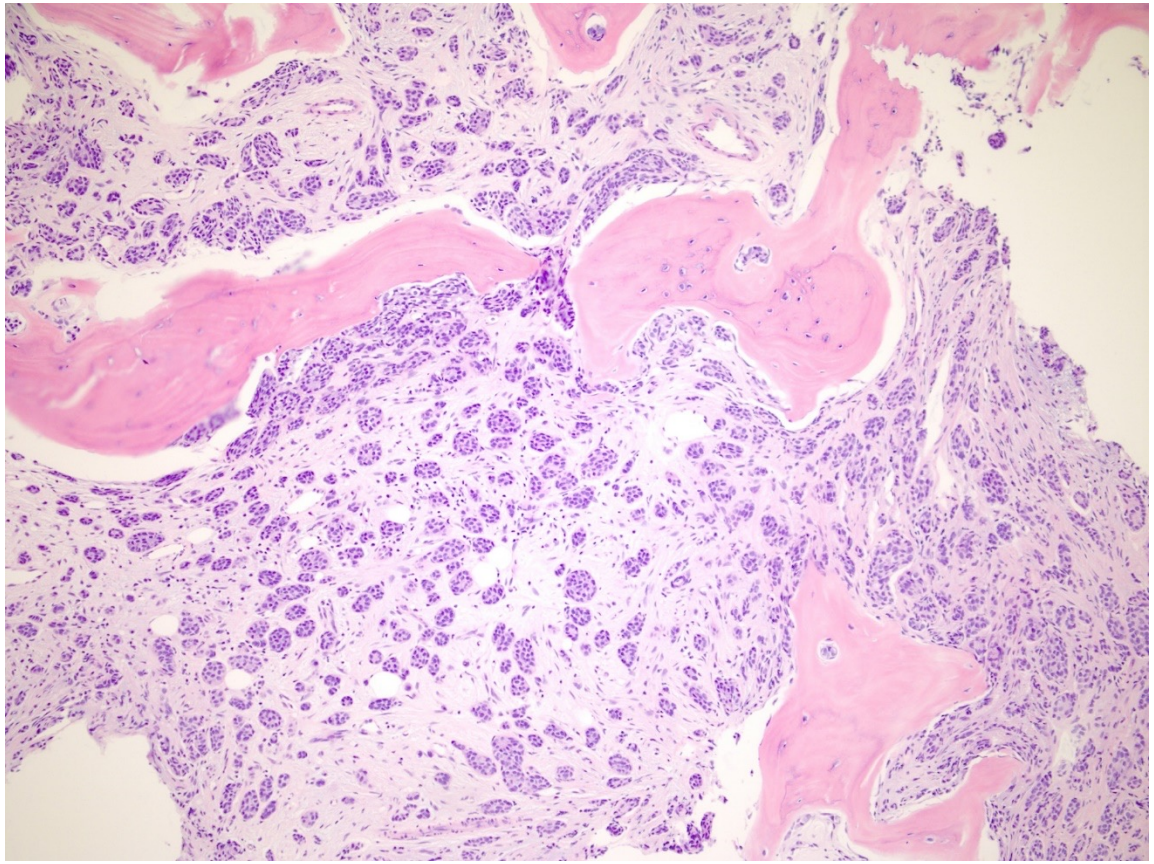
- WHO is eligible for testing?
- WHAT panel to use for germline testing?
- WHICH genes to include in the testing panel?
- WHAT is the magnitude of risk?
- HOW to utilize the test results clinically?

Case 1: 26/F, African-American, palpable mass, breast, left (2012)

Invasive mammary ca. with mixed ductal and lobular features (ER+PR+HER2+)
Lymph nodes and bone metastases at presentation (stage IV)



Breast biopsy



Bone biopsy

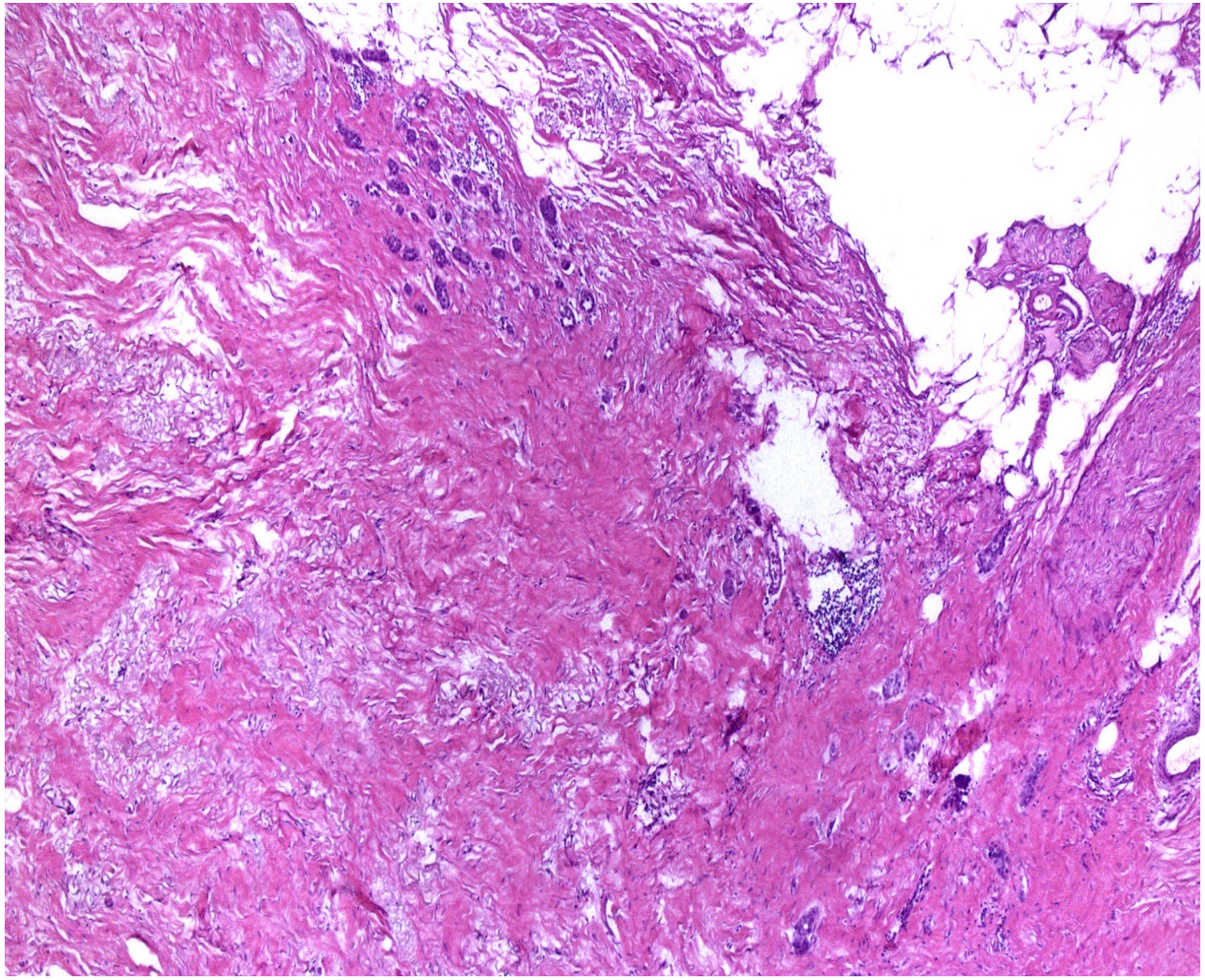
Case 1: 26/F, African-American, palpable mass, breast, left (2012)

- **FAMILY HISTORY**

- Maternal grandmother: died of breast cancer (age unknown)
- Maternal great aunt: breast cancer (age unknown)
- First cousin (maternal): breast cancer (age unknown)

2012
Germline testing for *BRCA1/2:*
NEGATIVE RESULTS

Case 1: Neoadjuvant chemotherapy (6 cycles) near complete response

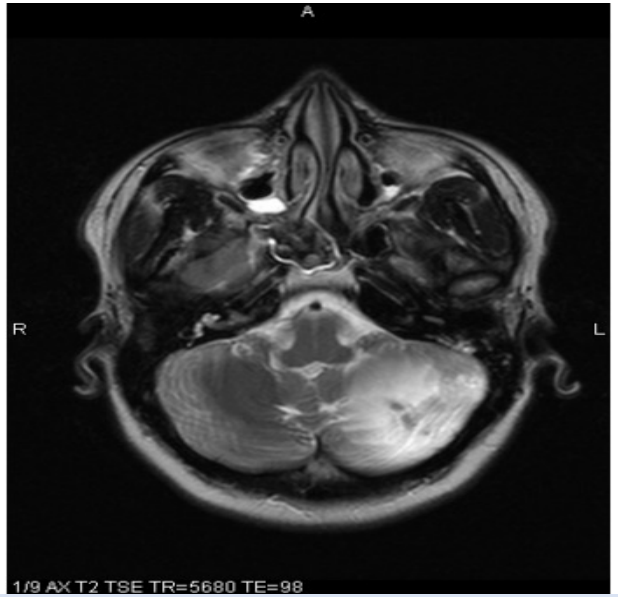


- 2013: Bilateral mastectomy with ALND
  - residual small foci of disease in left breast
  - 1/17 LN+
- 7/2013 PET scan: No FDG avid disease

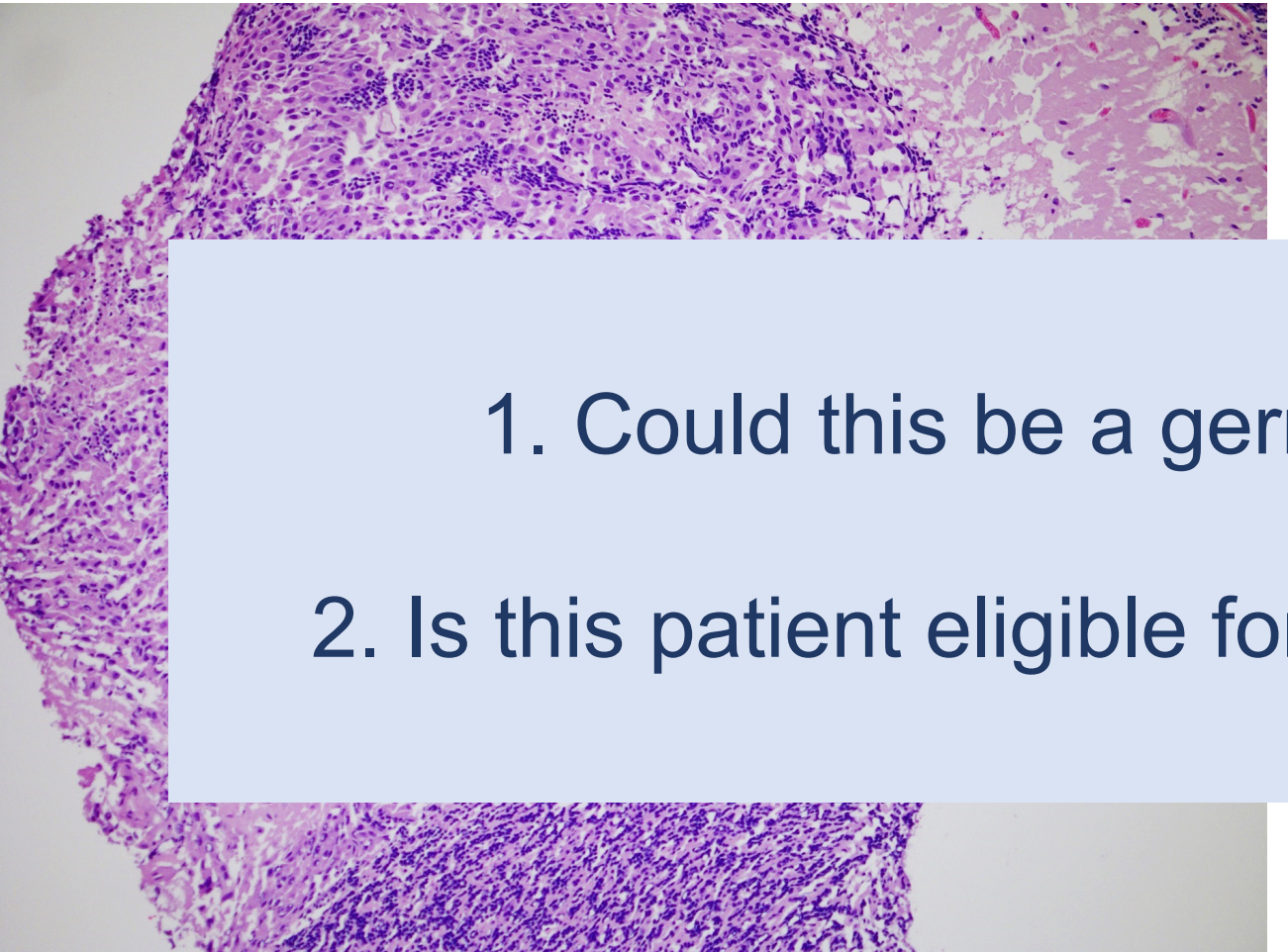
Case 1

- 10/2013: multiple brain metastases, treated with WBRT

01/2016



Anaplastic astrocytoma WHO Grade III



1. Could this be a germline *TP53* mutation?

2. Is this patient eligible for repeat germline testing?

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WHO is eligible for germline testing?

- Differing recommendations for germline testing?
  - **USPSTF (The United States Preventive Services Task Force):**
    - women with a personal or family history of breast, ovarian, tubal, or peritoneal cancer OR who have an ancestry associated with *BRCA1/2* gene mutations should be assessed with a **familial risk-assessment tool (ONTARIO; IBIS; BRCAPRO etc.)**
      - Genetic counseling and testing **ONLY IF** increased lifetime risk based on above risk assessment tools
  - **ASBS (American Society of Breast Surgeons):** testing **ALL** patients with breast cancer
  - **NCCN:** cautions **AGAINST** genetic testing in breast cancer patients diagnosed >60 years of age without a family history of breast, ovarian, pancreatic, or prostate cancer
    - very low probability (<2.5 percent) of yielding results with clinical utility

NCCN guidelines for genetic testing in breast cancer patients

Blood relative with a mutation in a cancer susceptibility gene

AGE
≤50 yrs

GENDER
Male breast cancer

RACE
Ashkenazi Jewish

H/O Other Cancers: Pancreas, Ovarian, or Prostate

PATHOLOGY

- **TNBC**
- **Multiple primary breast cancers (synchronous or metachronous)**
- **Lobular breast cancer with personal or family H/O diffuse gastric cancer**

LFS or Cowden testing criteria +

If it can aid in systemic therapy (PARP-inhibitors in metastatic setting; Olaparib in high risk HER2 – breast ca)

FAMILY HISTORY

- ≥1 close blood relative with ANY:
  - breast cancer at age ≤50
  - male breast cancer
  - ovarian cancer
  - pancreatic cancer
  - prostate cancer with metastatic, or high- or very-high-risk group
- ≥3 total diagnoses of breast cancer in patient and/or close blood relatives
- ≥2 close blood relatives with either breast or prostate cancer (any grade)

Mutation detected on solid tumor sequencing with germline implications

Individuals who meet these criteria but tested negative with a prior limited genetic test

>5% probability of a BRCA1/2 pathogenic variant based on prior probability models (eg, Tyrer-Cuzick, BRCAPro, CanRisk)

Case 1: Repeat Germline Testing Using Multigene Panel Test (2016)

- **FAMILY HISTORY**

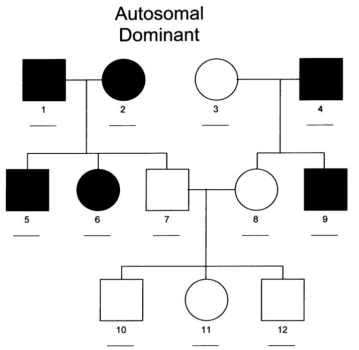
- Maternal grandmother: died of breast cancer (age unknown)
- Maternal great aunt: breast cancer (age unknown)
- First cousin (maternal): breast cancer (age unknown)
- Son: dx with adrenal cortical ca. at 18 months of age few months ago
- Sister: breast cancer dx. at 37y few months ago

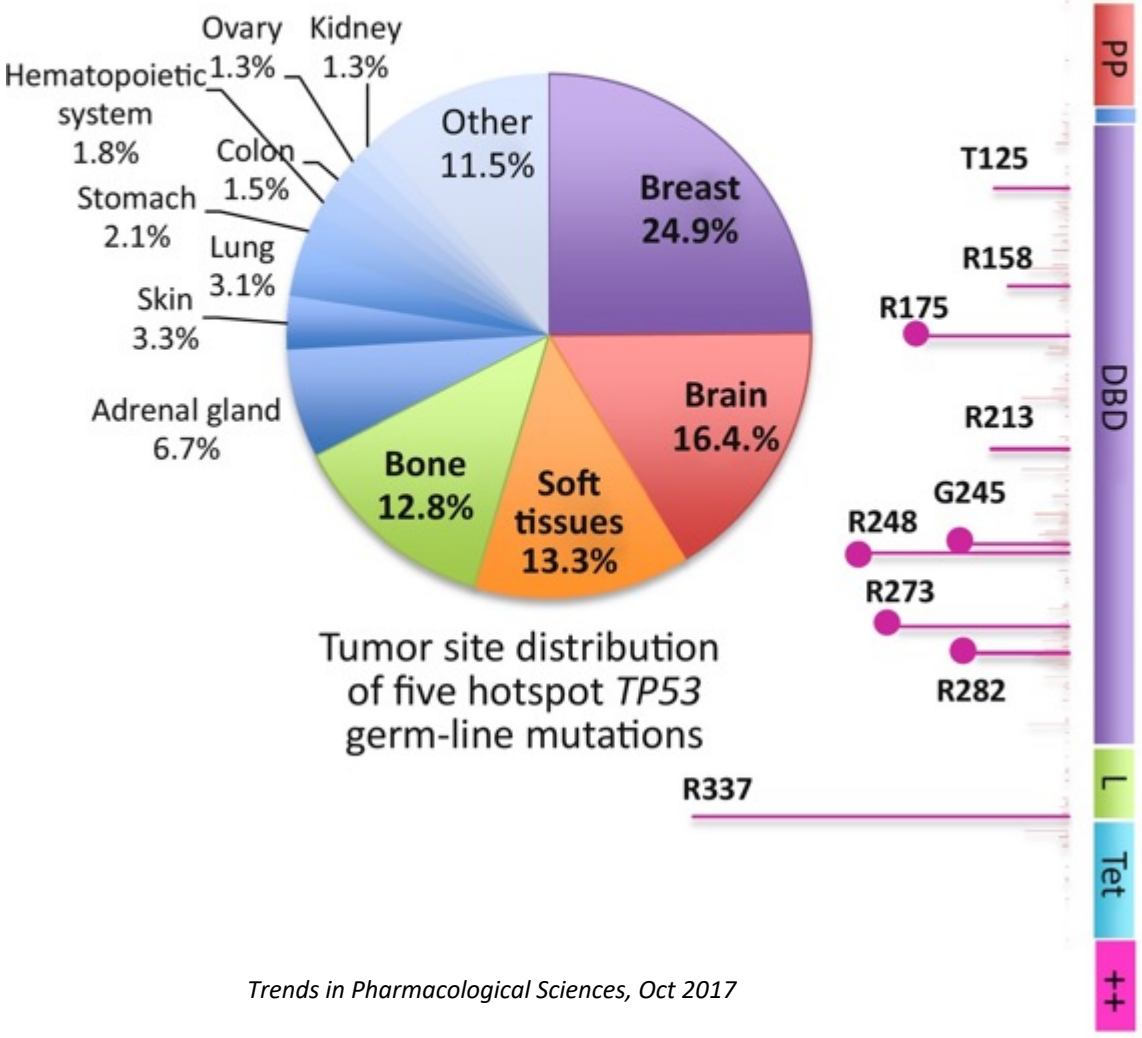
2012
Germline testing for *BRCA1/2:*
NEGATIVE RESULTS

Diagnosis
Li-Fraumeni Syndrome (LFS)

2016
Repeat Germline testing
Positive for mutation *TP53*
p.R196* C.586C>T
(same as solid tumor panel result)

Cancers associated with LFS



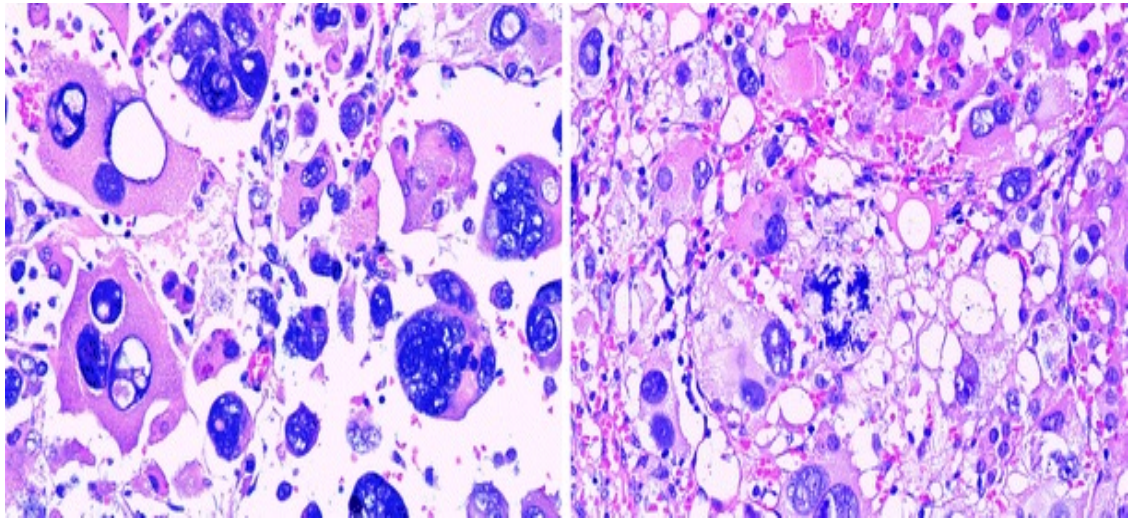


- Rare (1 in 5000-20,000 person years)
- <1% of all HBCs
- 50% risk by age 40 years
- 90% risk by age 60
- Types of cancer and ages of onset can vary from
  - Family to family
  - Person to person within the same family
- Number of cancers may vary
  - MOST individuals develop <u>more than one</u> cancer during his/her lifetime
  - Rarely only one cancer
- Risk of developing second cancer/sarcoma post radiation

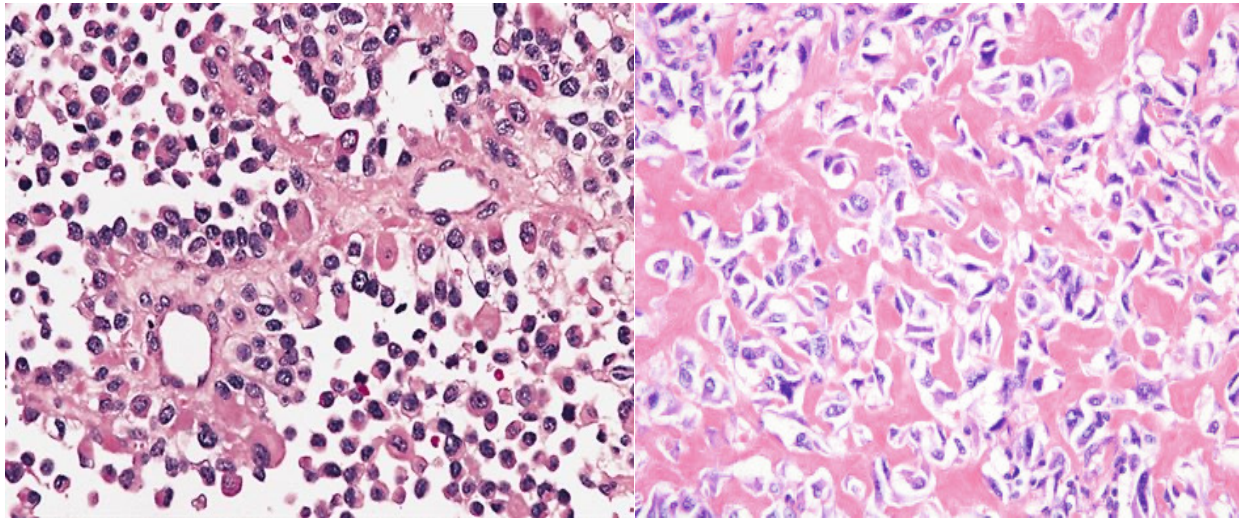
Trends in Pharmacological Sciences, Oct 2017

Strongest predictors of Li-Fraumeni syndrome

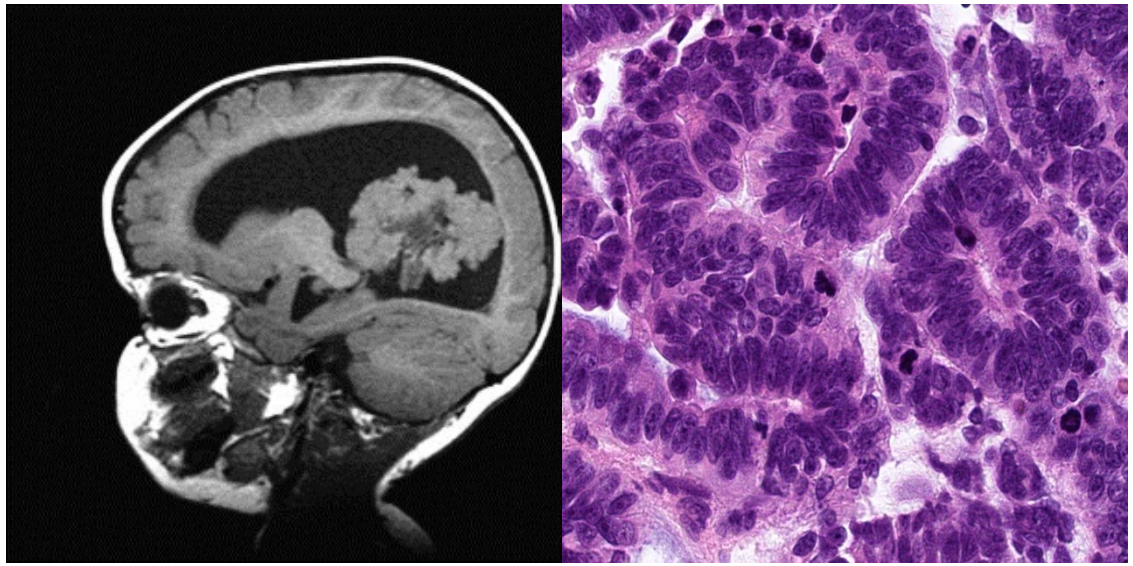
Childhood adrenocortical carcinoma



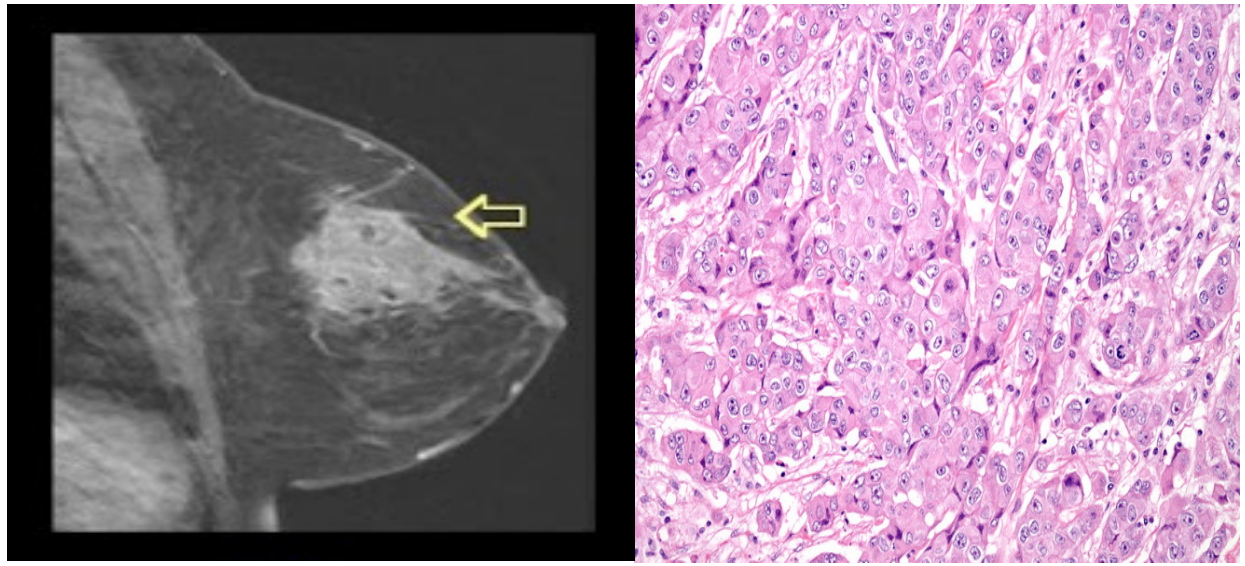
Childhood sarcoma (RMS, Osteosarcoma etc.)



Childhood choroid plexus carcinoma



Women with breast cancer under age 30



Source: pathologyoutlines.com

Clinical Diagnostic Criteria for LFS or germline testing for *TP53* mutations

- A member of a kindred with a known *TP53* pathogenic or likely pathogenic variant

- CLASSIC

A combination of ALL of the following:

- A proband with a sarcoma diagnosed before age 45
- A first degree relative with any cancer dx. before age 45
- An additional first- or second-degree relative in the same lineage with cancer diagnosed at age <45 years, or a sarcoma at any age

High positive predictive value (56%)
Low sensitivity (40%)
Li et al 1988

- REVISED CHOMPRET CRITERIA (2015)

- Criterion 1 : Family +
  - Tumor belonging to LFS spectrum before the age of 46
  - At least 1 first-degree or second-degree family member with a LFS-tumor (except breast) before the age of 56
  - Our pt. <26y + son with ACC
- Criterion 2 : Multiple cancers
  - Person with multiple tumors (2 belonging to LFS spectrum) and the first occurred before age 46
  - Our pt. <26y + Astrocytoma
- Criterion 3 : Specific type of cancer
  - Person with adrenal cortical carcinoma, tumor of choroid plexus, RMS embryonic anaplastic type, regardless of family history OR breast cancer before age 31
  - Our pt. <26y

- Pediatric hypodiploid acute lymphoblastic leukemia

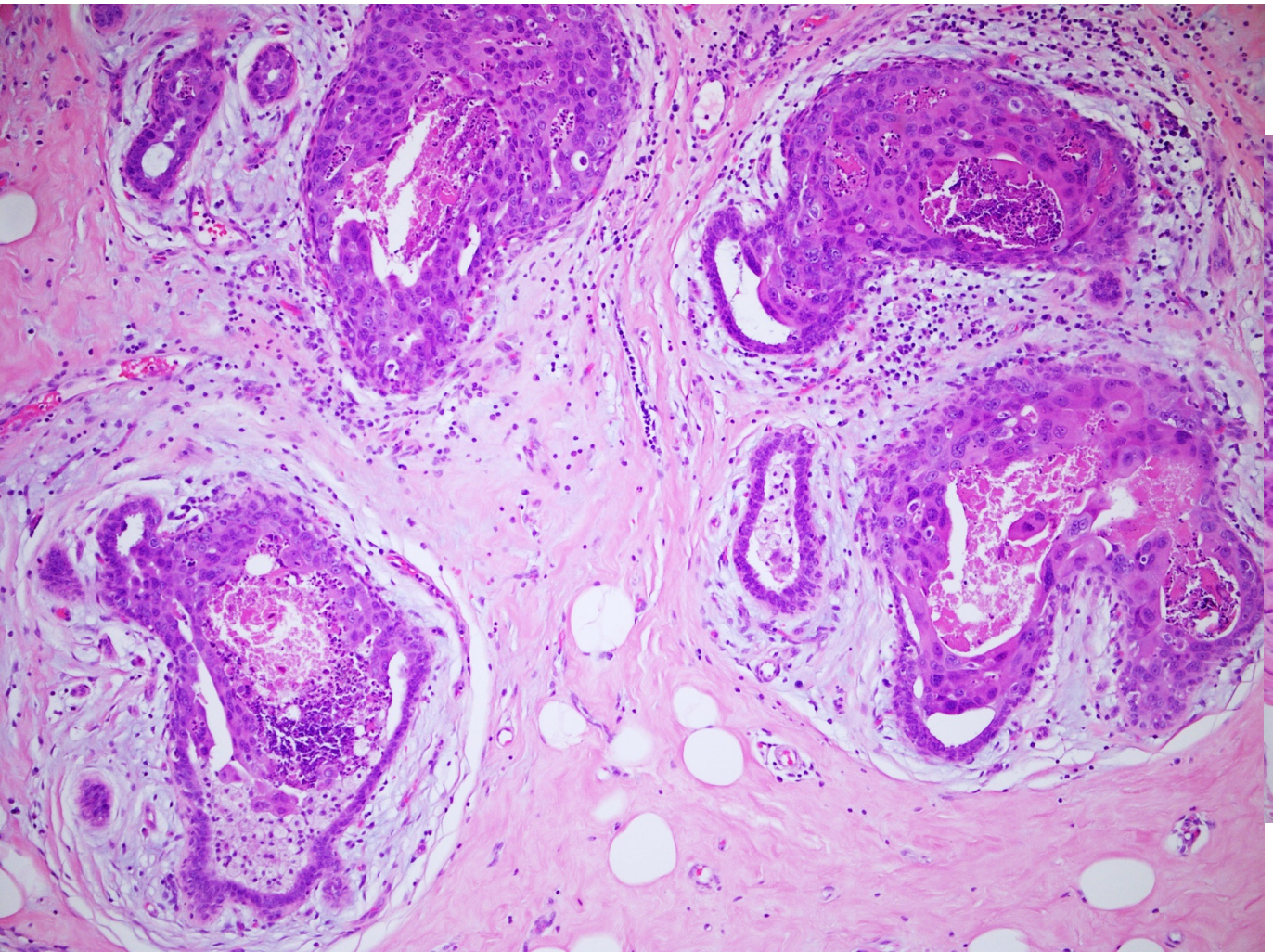
What do we learn from Case #1?

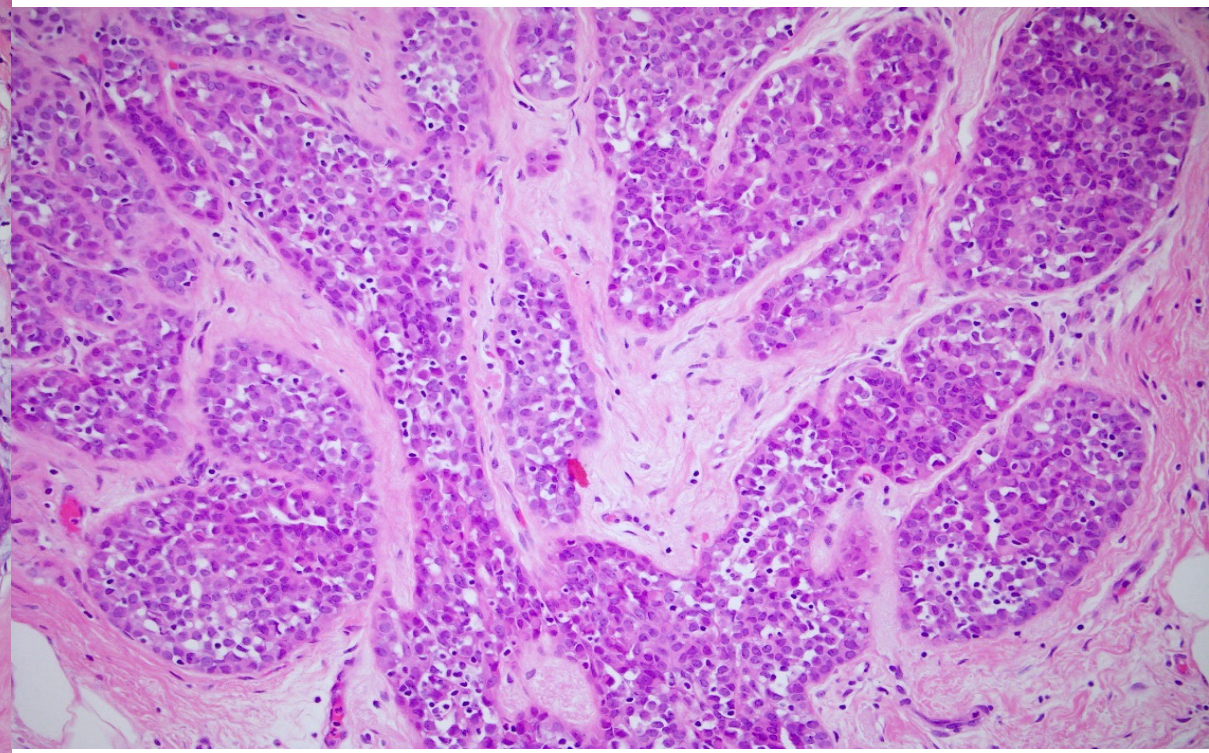
- Multigene panel NGS testing on solid tumors may help diagnose germline mutations in patients
- Patients that test negative with single gene panel (BRCA1/BRCA2) germline test may test positive for other pathogenic germline mutations with newer multigene panel tests
- Clinical diagnostic criteria are not sensitive enough to diagnose hereditary breast cancer syndromes (LFS, etc.)

Case 2

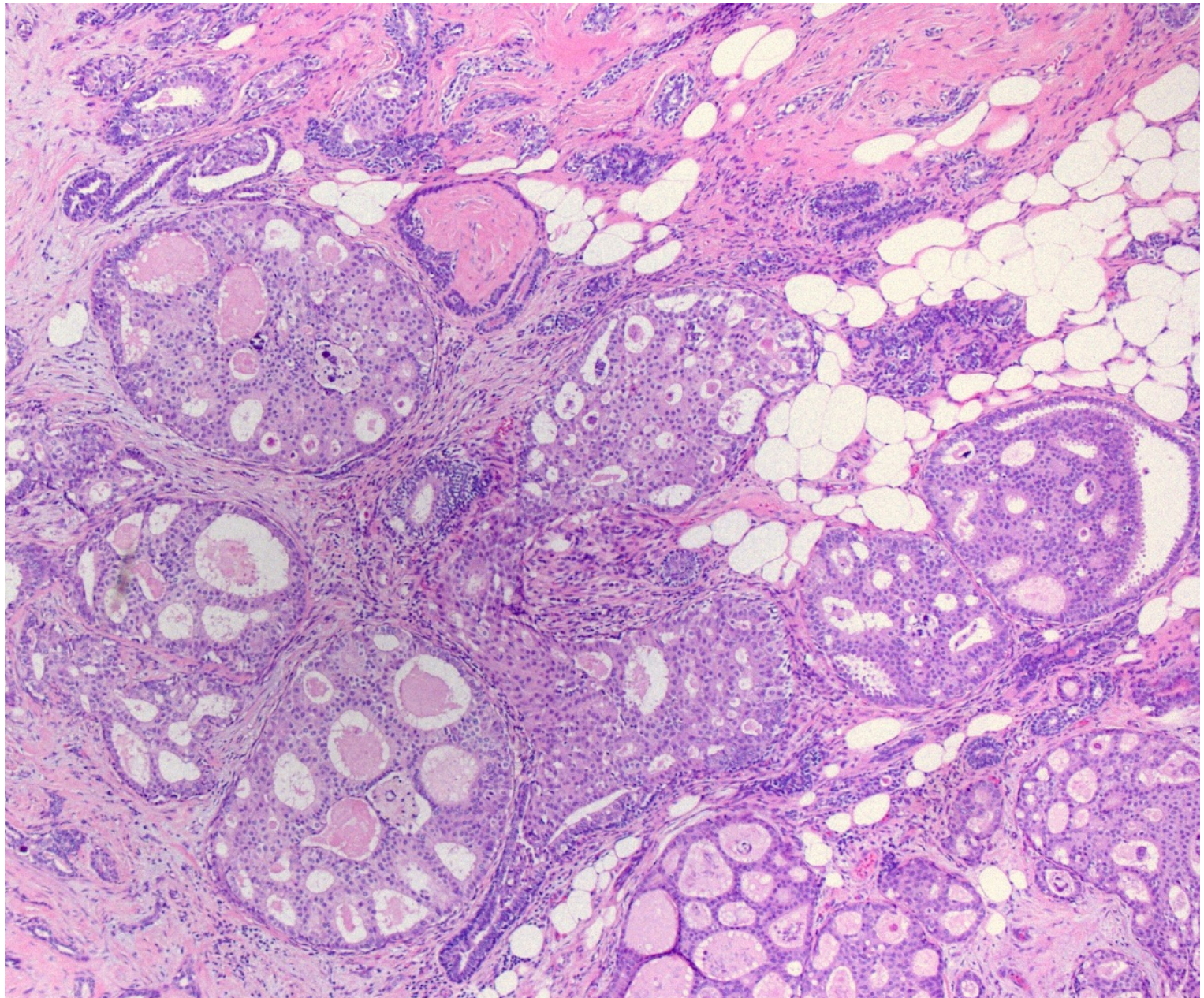
62y/F Caucasian, with abnormal calcifications on annual mammography screening in bilateral breasts

Core biopsy, right breast: DCIS, high grade and LCIS



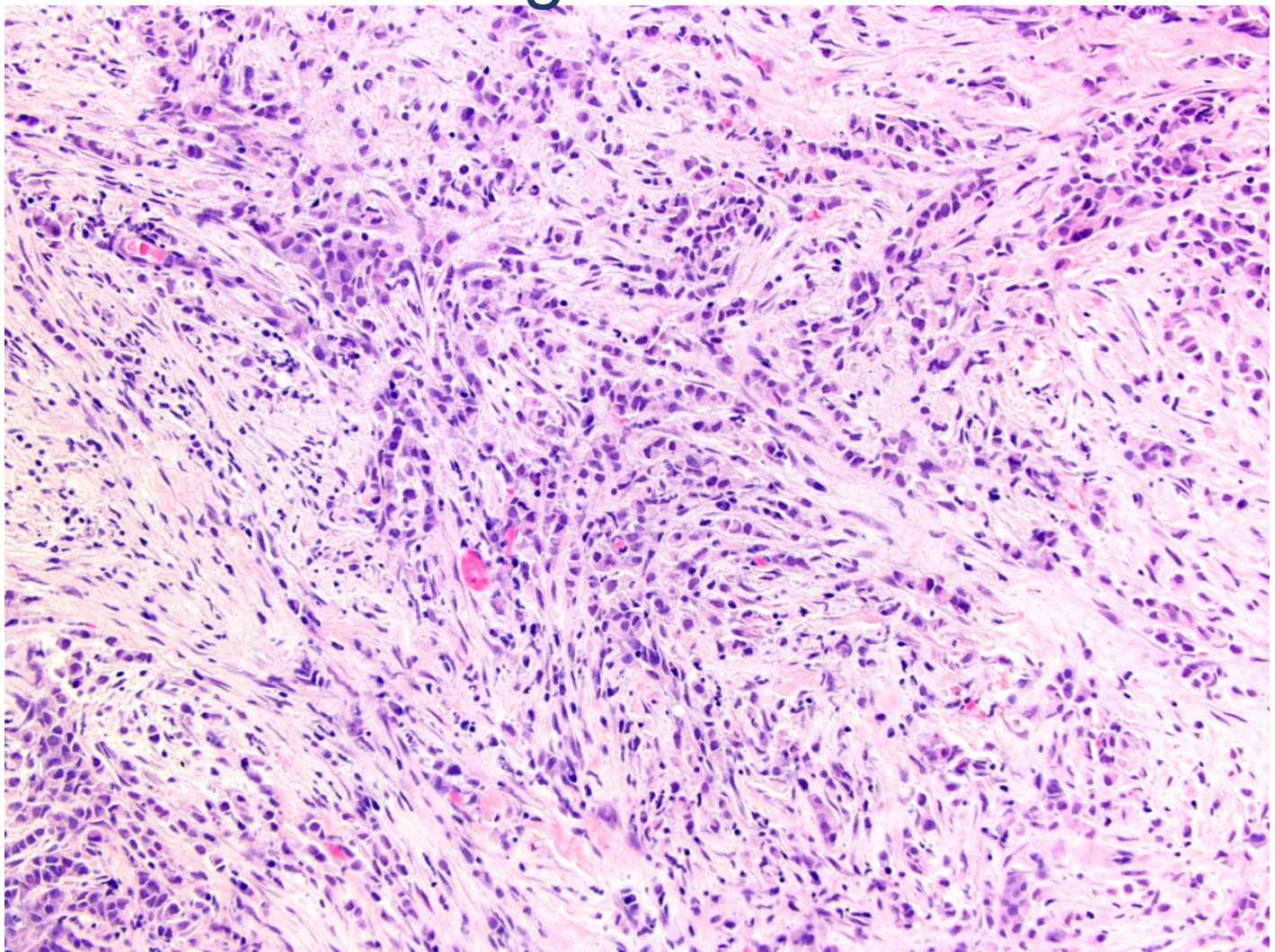


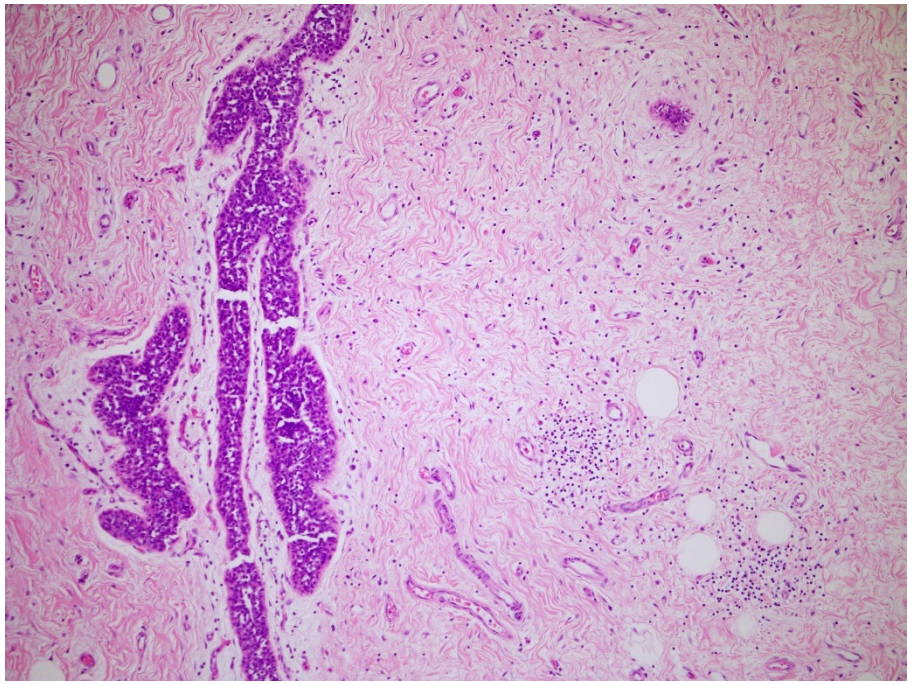
Core biopsy, left breast: DCIS, low to intermediate grade



Case 2: Bilateral needle-loc excisions

ILC, left breast, ER+/PR+/HER2-, stage T1a N0





Residual LCIS, right breast

Case 2: Family History

- Detailed family history:
  - Breast Cancer in her sister @ 37y
  - Breast Cancer in her maternal grandmother @ 58y
  - Colon Cancer in her paternal grandmother @ 74y
  - Ovarian Cancer in her paternal relative @ 45y
  - Both parents alive in their 80's and cancer free

Sister

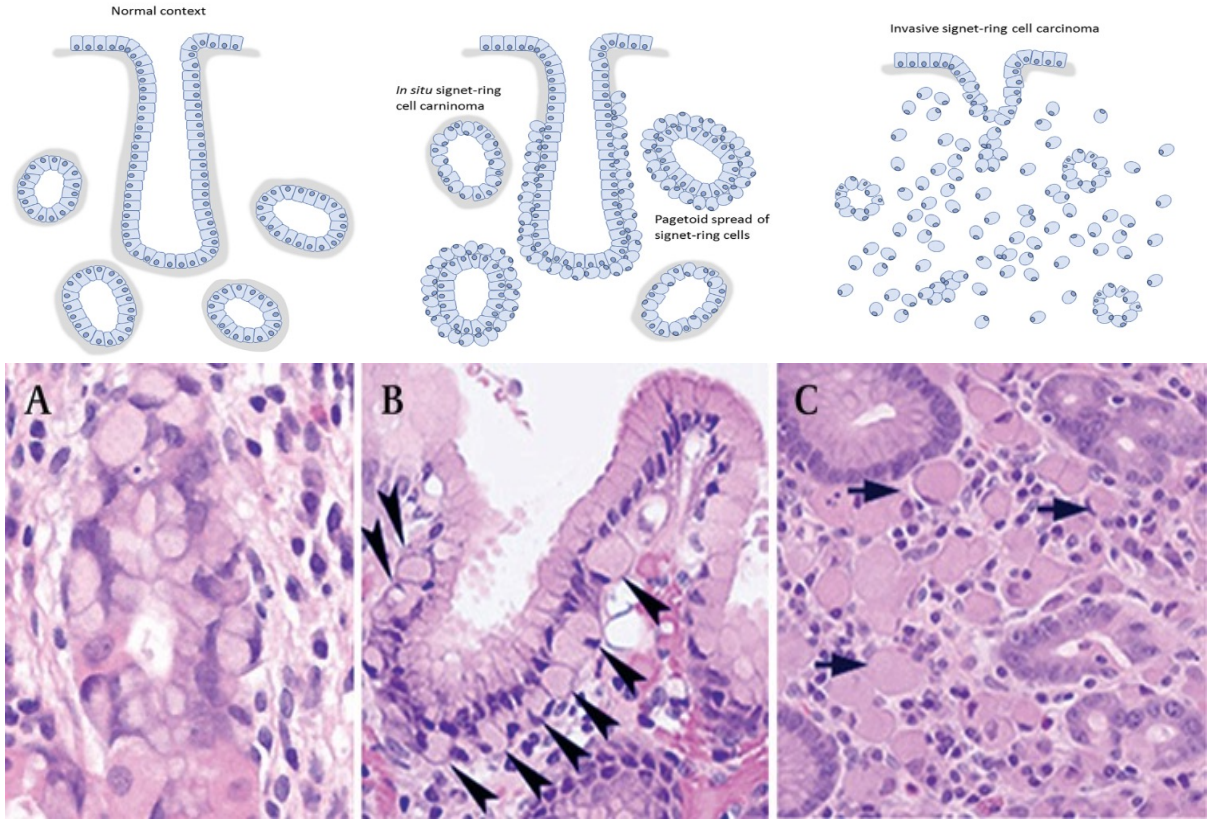
- **recently tested positive for *CDH1* germline mutation on multigene panel testing**
  - **deleterious mutation *CDH1* c.1711+1G>A**

CDH1 germline mutations and cancer

- First described in 1999
  - Familial clustering of diffuse gastric cancer and lobular breast cancer in New Zealand Maōri families and its linkage with germline *CDH1* variants *(Guilford et al., 1998)*
  - Hereditary diffuse gastric cancer (HDGC)
- Increased lifetime risk
  - Diffuse-type gastric adenocarcinoma (Original estimates: 70%; Recent estimates: 37% - 42% for men and 25% - 33% for women)
  - Lobular breast cancer in women (42 – 55%)
  - Cleft lip and palate (14%)
  - Blepharocheilodontic syndrome

CDH1 germline mutations and gastric cancer

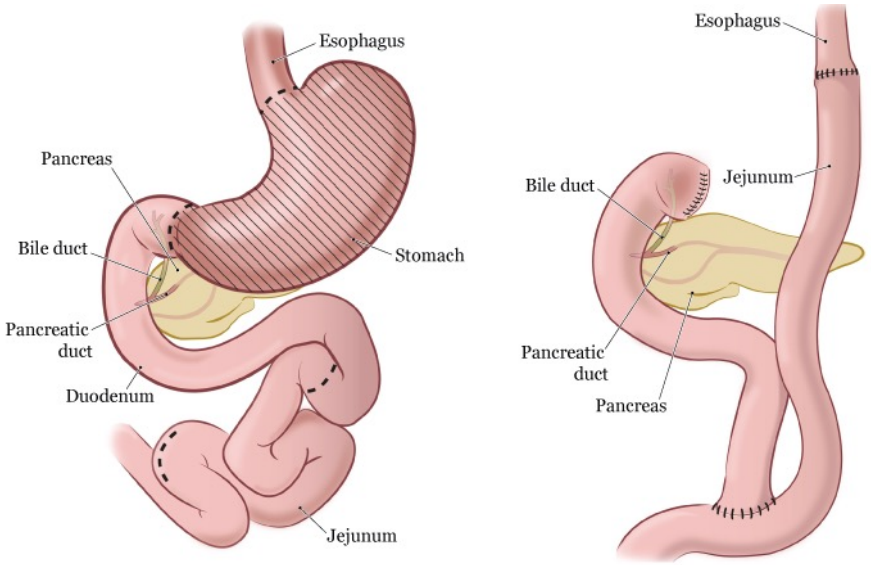
Multiple small foci of occult signet ring cell cancer (in-situ, Pagetoid spread and invasive)

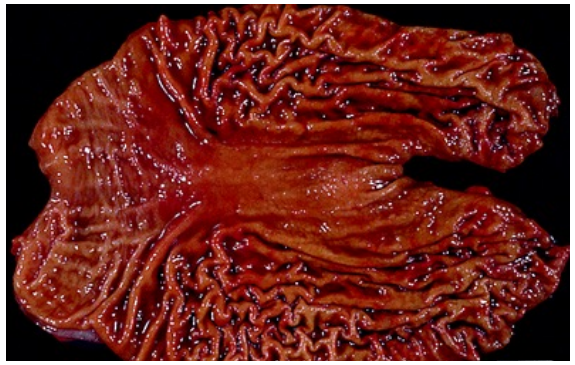


Gamble LA. JAMA Surg. 2021;156(4):387-392. doi:10.1001/jamasurg.2020.6155

Corso G et al. Cancer Metastasis Rev (2014) 33:1081–1094

CDH1 and gastric cancer managemement/surveillance

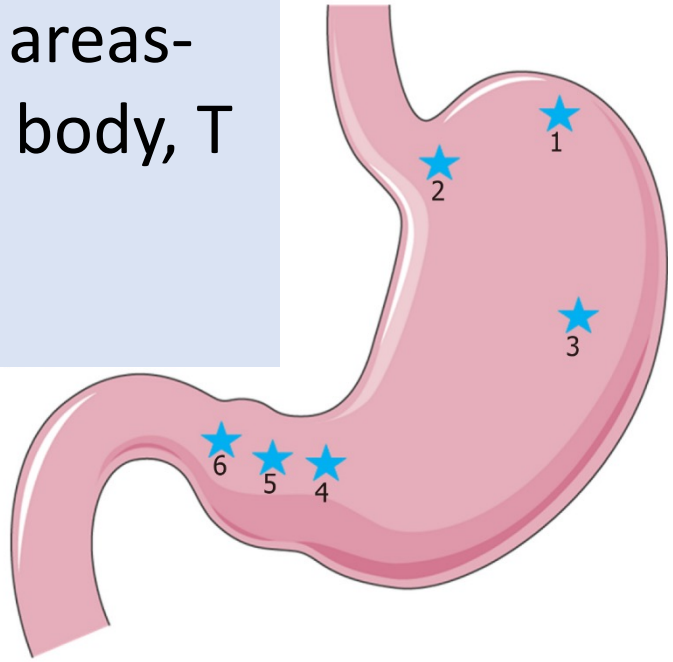




Prophylactic gastrectomy
(complete sampling for SRC foci)
Occult SRC detection rate 80-100%

Upper GI Endoscopy

Random 5 biopsies from each of 6 areas- fundus, cardia, body, T zone, antrum, prepyloric



Cambridge protocol
Occult SRC detection rate 20-63%

Lynch HT. J Med Genet July 2010 Vol 47 No 7

Genetic testing for *CDH1* germline mutations

International Gastric Cancer Linkage Consortium Guidelines (IGCLC)

- ≥2 Cases of gastric cancer in family (any age), with at least 1 confirmed DGC
- Isolated individual diagnosed with DGC at age <40 from a low incidence population
- Personal or family h/o both DGC and LBC, with 1 case <50y at time of diagnosis
- Personal history of DGC and personal or family history of cleft/lip-palate
- In-situ SRC or pagetoid spread of SRC on gastric biopsy

New criterions added

- Bilateral ILC in a pt. <50y with or without family h/o ILC
- Unilateral ILC in a pt. <45y with a family h/o ILC

Back to Case 2….

- Detailed family history:
  - Breast Cancer in her sister @ 37y
  - Breast Cancer in her maternal grandmother @ 58y
  - Colon Cancer in her paternal grandmother @ 74y
  - Ovarian Cancer in her paternal relative @ 45y
  - Both parents alive in their 80's and cancer free

NO FAMILY H/O GASTRIC CA

Case 2

Tested *CDH1* positive: deleterious mutation c.1711+1G>A in the CDH1 gene (similar to that found in her sister)

???

- Is she a candidate for prophylactic gastrectomy?
- Should she opt for B/L risk reducing mastectomy OR opt for follow-up by MRI surveillance of breast?
- Genetic testing and cancer screening of family members?

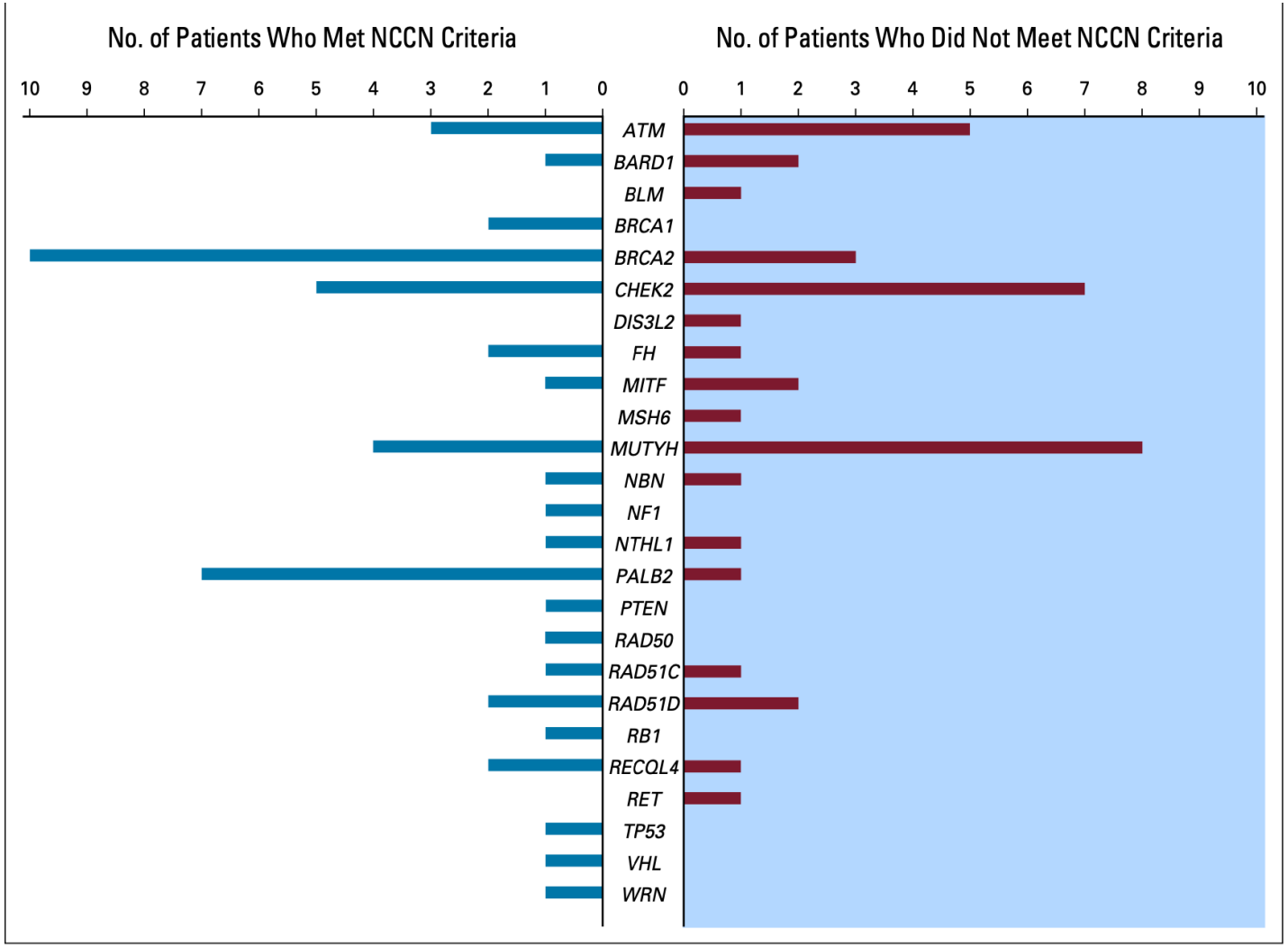
Case 2 follow up…

- Patient opted for B/L mastectomy with adjuvant hormonal therapy
- Follow up EGD biopsies- signet ring cell carcinoma in stomach cardia bx
- Total gastrectomy
  - Pathology: 16 microscopic foci of invasive poorly differentiated signet ring cell adenocarcinoma (T1a N0)
- Both son and daughter positive for *CDH1* and diagnosed with signet ring cell gastric ca. on EGD biopsies- underwent gastrectomy
  - Daughter- High risk MRI screening for breast ca.
- Sister diagnosed with B/L ovarian Krukenberg tumor from metastatic gastric carcinoma- currently receiving chemotherapy
- Father (asymptomatic) tested positive for *CDH1* (she inherited the gene from father)

Germline Testing in Breast Cancer

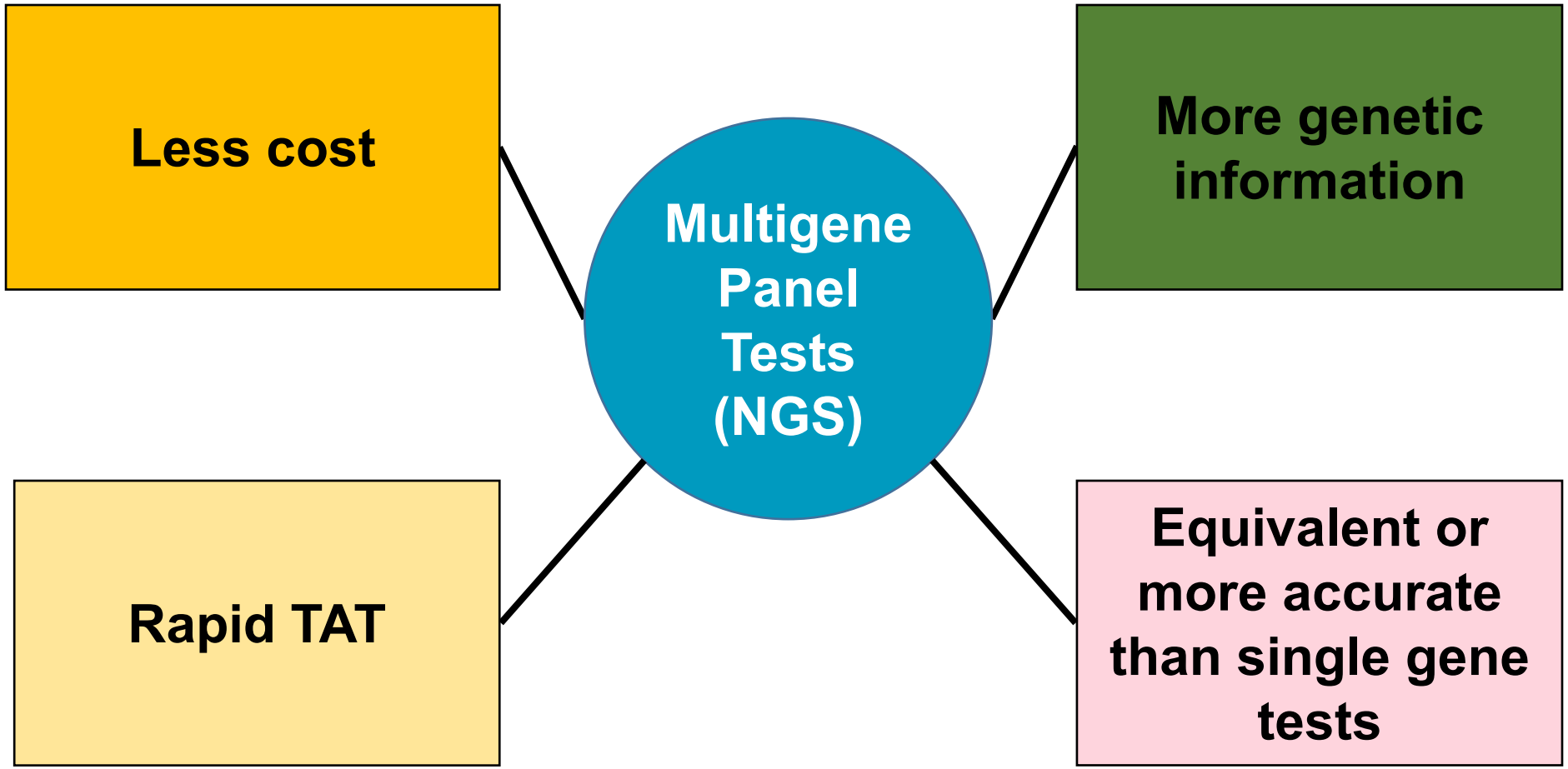
- WHO is eligible for testing?
- WHAT panel to use for germline testing?
- WHICH genes to include in the testing panel?
- WHAT is the magnitude of risk?
- HOW to utilize the test results clinically?

Nearly half of patients with breast cancer with a clinically actionable mutation and/or management guidelines in development are missed by current testing guidelines



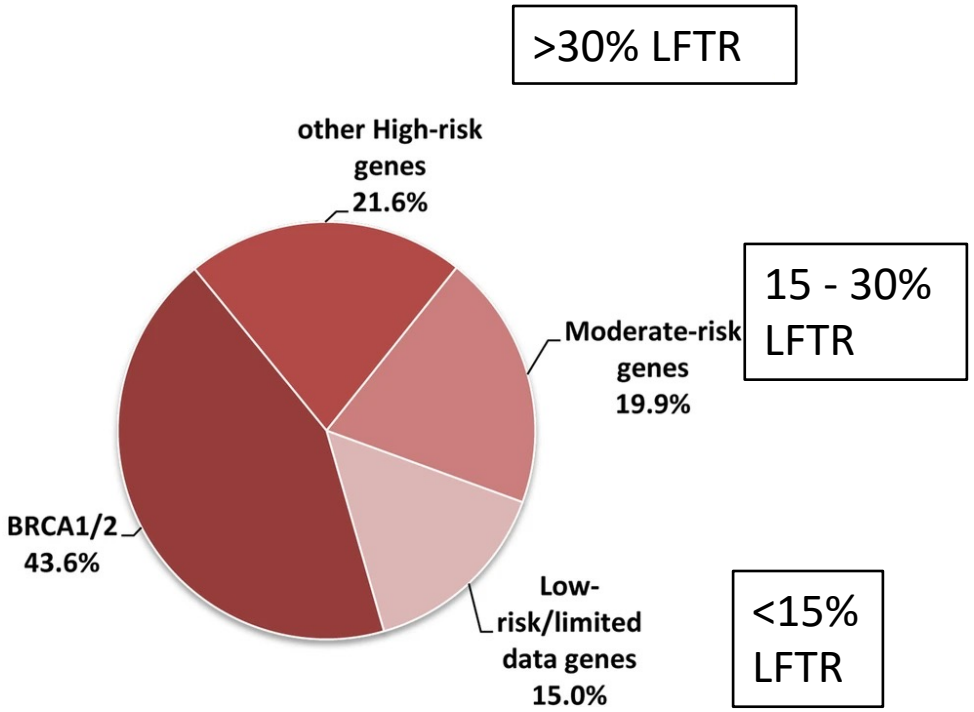
Beltsch P et al. J Clin Oncol 37:453-460. © 2018 by American Society of Clinical Oncology

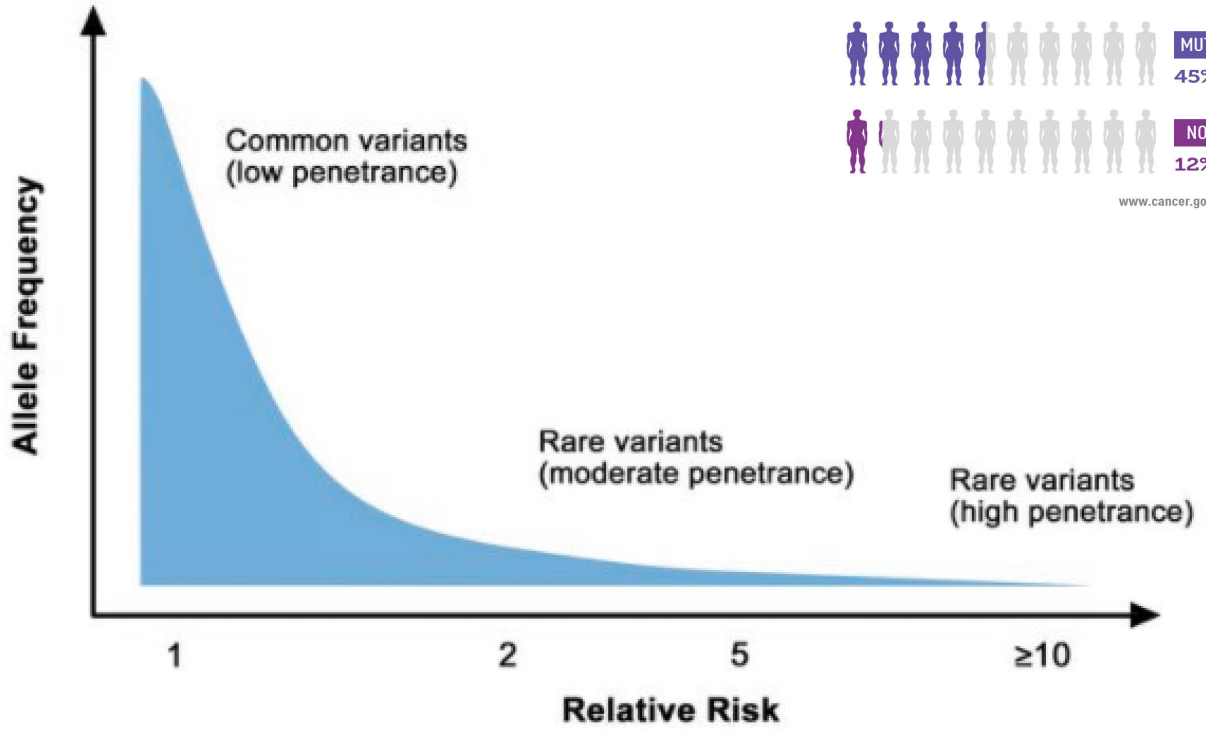
Advantages of Germline Testing with Multigene Panels

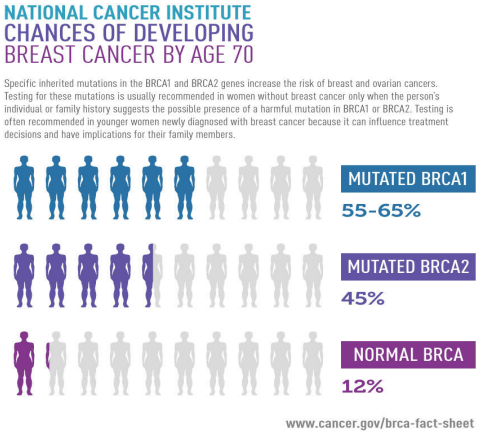


Multigene Panel Testing-Challenges

Uncertain Risk of Variant Penetrance



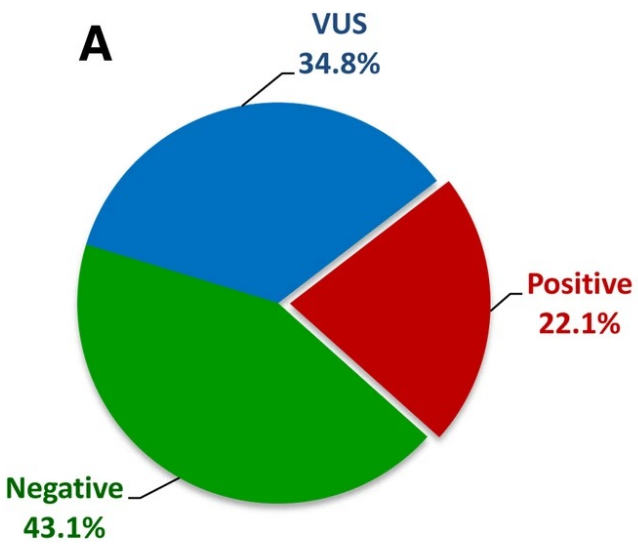




- Which genes fall under low risk for disease penetrance vs moderate risk vs high risk?

Multigene Panel Testing-Challenges

Variants of Uncertain Significance (VUS)



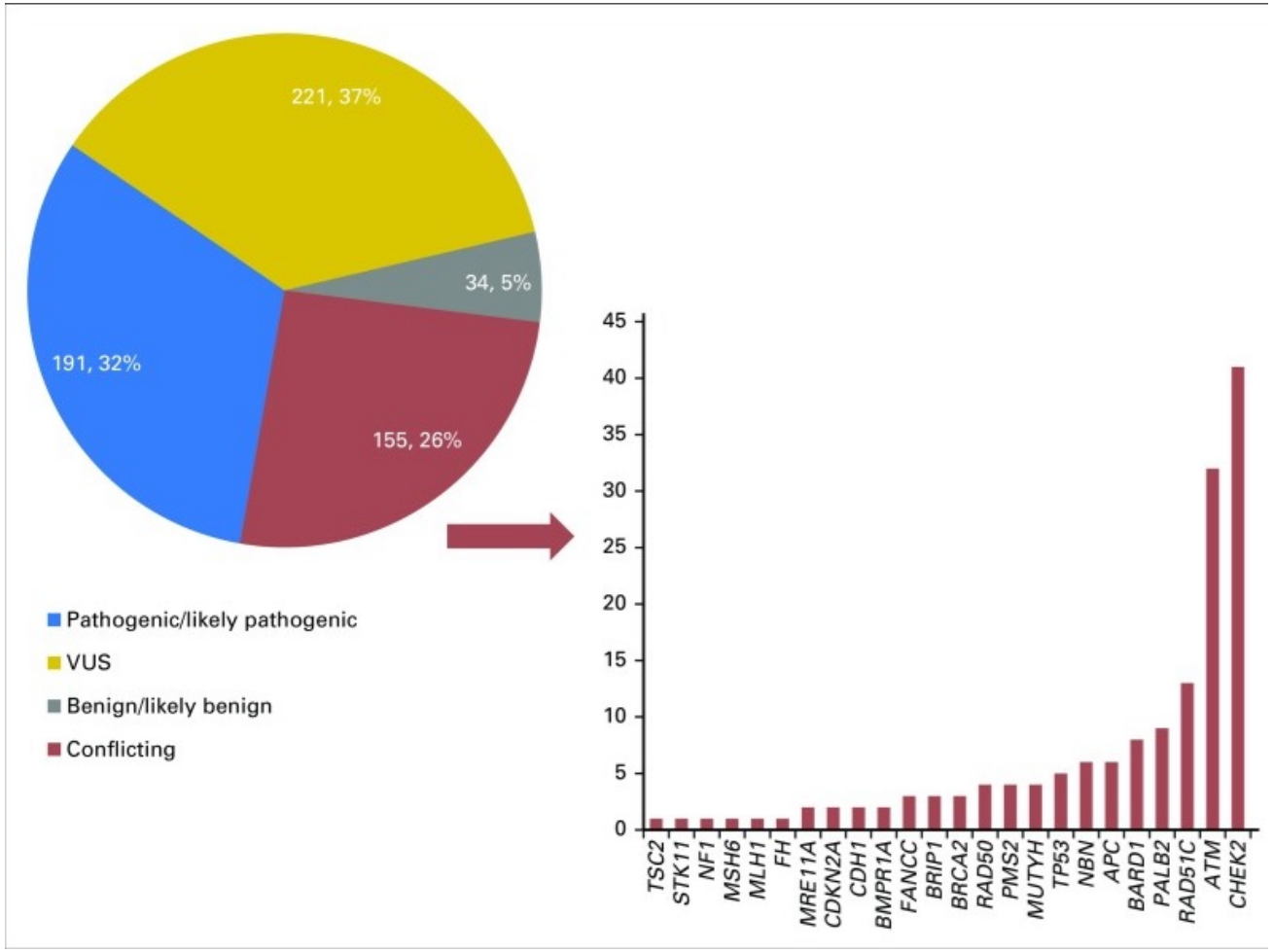
Pathogenic (P)	Sufficient evidence to classify as capable of causing disease. Targeted testing of at-risk family members and appropriate changes in management (e.g., high risk surveillance, chemoprevention or risk-reducing surgery).
Variant, Likely Pathogenic (LP)	Strong evidence in favor of pathogenicity. Targeted testing of at-risk family members and appropriate changes in management (e.g., high risk surveillance, chemoprevention or risk-reducing surgery).
Variant, Unknown Significance (VUS)	Limited and/or conflicting evidence regarding pathogenicity. Targeted testing of informative family members to collect segregation data recommended. Medical management based on personal and family histories, not VUS carrier status.
Variant, Likely Benign (VLB)	Strong evidence against pathogenicity Targeted testing of at-risk family members not recommended. Medical management based on personal and family histories.
Benign	Very strong evidence against pathogenicity. Targeted testing of at-risk family members not recommended. Medical management based on personal and family histories.

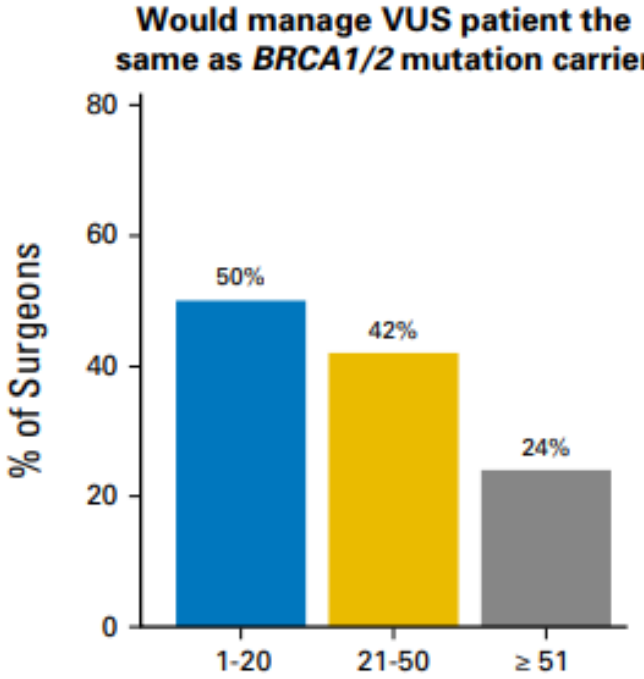
Multigene Panel Testing- Databases

- Many online tools have become available to assist in variant interpretation:
  - ClinVar (www.ncbi.nlm.nih.gov/clinvar)
  - ClinGen (www.clinicalgenome.org)
  - ENIGMA (https://enigmaconsortium.org)
  - PROMPT (https://promptstudy.info)
  - ExAC (http://exac.broadinstitute.org)
  - ASK2me (https://ask2me.org/index.php), includes curated management guidelines
  - IARC TP53 database

Multigene Panel Testing-Challenges

Inter-laboratory discrepancy
in variant classification





Lack of understanding for VUS amongst surgeons

Multigene Panel Testing-Challenges

- Increased identification of low-moderate penetrance PVs without established cancer risk reduction guidelines
- Increased VUS
- Interlaboratory discrepancy
- Challenges in genetic counseling
  - Shortage
  - Cost, racial and socioeconomic barriers

NCCN recommends carefully selected panels performed at a CAP or CLIA Certified Laboratory

J Clin Oncol. 2018 Feb 1;36(4):432

Germline Testing in Breast Cancer

- WHO is eligible for testing?
- WHAT panel to use for germline testing?
- WHICH genes to include in the testing panel?
- WHAT is the magnitude of risk?
- HOW to utilize the test results clinically?

Two recent large case control studies (NEJM, Feb 2021)

The NEW ENGLAND JOURNAL of MEDICINE

ORIGINAL ARTICLE

Breast Cancer Risk Genes — Association Analysis in More than 113,000 Women

Breast Cancer Association Consortium*

ABSTRACT

BACKGROUND

Genetic testing for breast cancer susceptibility is widely used, but for many genes, evidence of an association with breast cancer is weak, underlying risk estimates are imprecise, and reliable subtype-specific risk estimates are lacking.

Dorling et al.

- 34 genes
- 113,000 women (60,000 with breast cancer and 53,000 unaffected controls)
- 25 countries

The NEW ENGLAND JOURNAL of MEDICINE

ORIGINAL ARTICLE

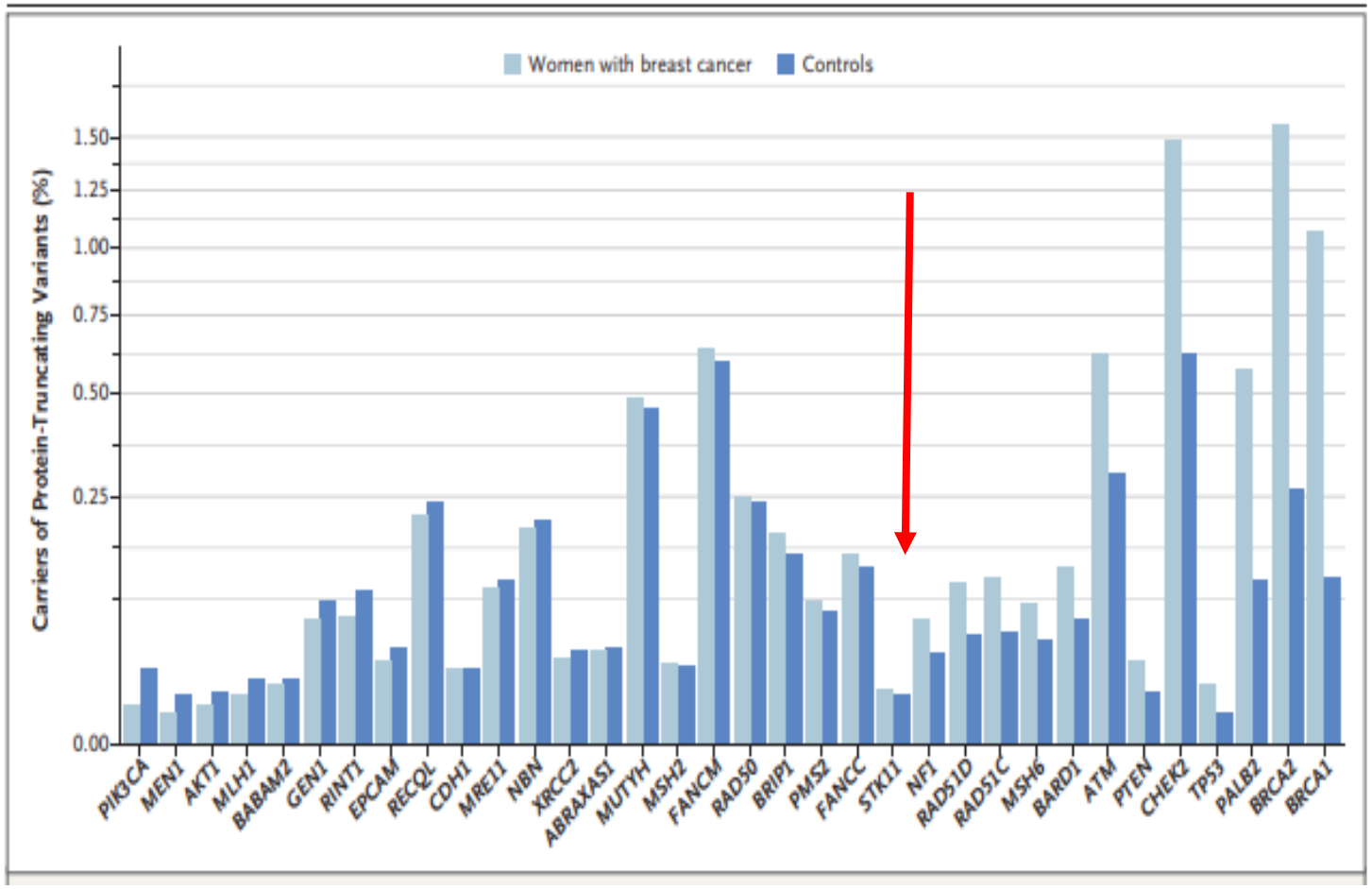
A Population-Based Study of Genes Previously Implicated in Breast Cancer

C. Hu, S.N. Hart, R. Gnanaolivu, H. Huang, K.Y. Lee, J. Na, C. Gao, J. Lilyquist, S. Yadav, N.J. Boddicker, R. Samara, J. Klebba, C.B. Ambrosone, H. Anton-Culver, P. Auer, E.V. Bandera, L. Bernstein, K.A. Bertrand, E.S. Burnside, B.D. Carter, H. Eliassen, S.M. Gapstur, M. Gaudet, C. Haiman, J.M. Hodge, D.J. Hunter, E.J. Jacobs, E.M. John, C. Kooperberg, A.W. Kurian, L. Le Marchand, S. Lindstroem, T. Lindstrom, H. Ma, S. Neuhausen, P.A. Newcomb, K.M. O'Brien, J.E. Olson, I.M. Ong, T. Pal, J.R. Palmer, A.V. Patel, S. Reid, L. Rosenberg, D.P. Sandler, C. Scott, R. Tamimi, J.A. Taylor, A. Trentham-Dietz, C.M. Vachon, C. Weinberg, S. Yao, A. Ziogas, J.N. Weitzel, D.E. Goldgar, S.M. Domchek, K.L. Nathanson, P. Kraft, E.C. Polley, and F.J. Couch

Hu et al.

- 28 genes
- 64,000 women (32,347 with breast cancer and 32,544 unaffected controls)
- United States

Association with breast cancer risk



(Dorling et al. NEJM Feb 2021)

Significant Risk (Odds ratio 5 – 10.6)	Modest Risk (Odds ratio 1.8 – 6.0)
BRCA1	BARD1
BRCA2	RAD51C
PALB2	RAD51D
ATM	PTEN
CHEK2	NF1
	TP53
	MSH6 (Dorling et al.)
	CDH1 (Hu et al.)

Two recent large case control studies (NEJM, Feb 2021)

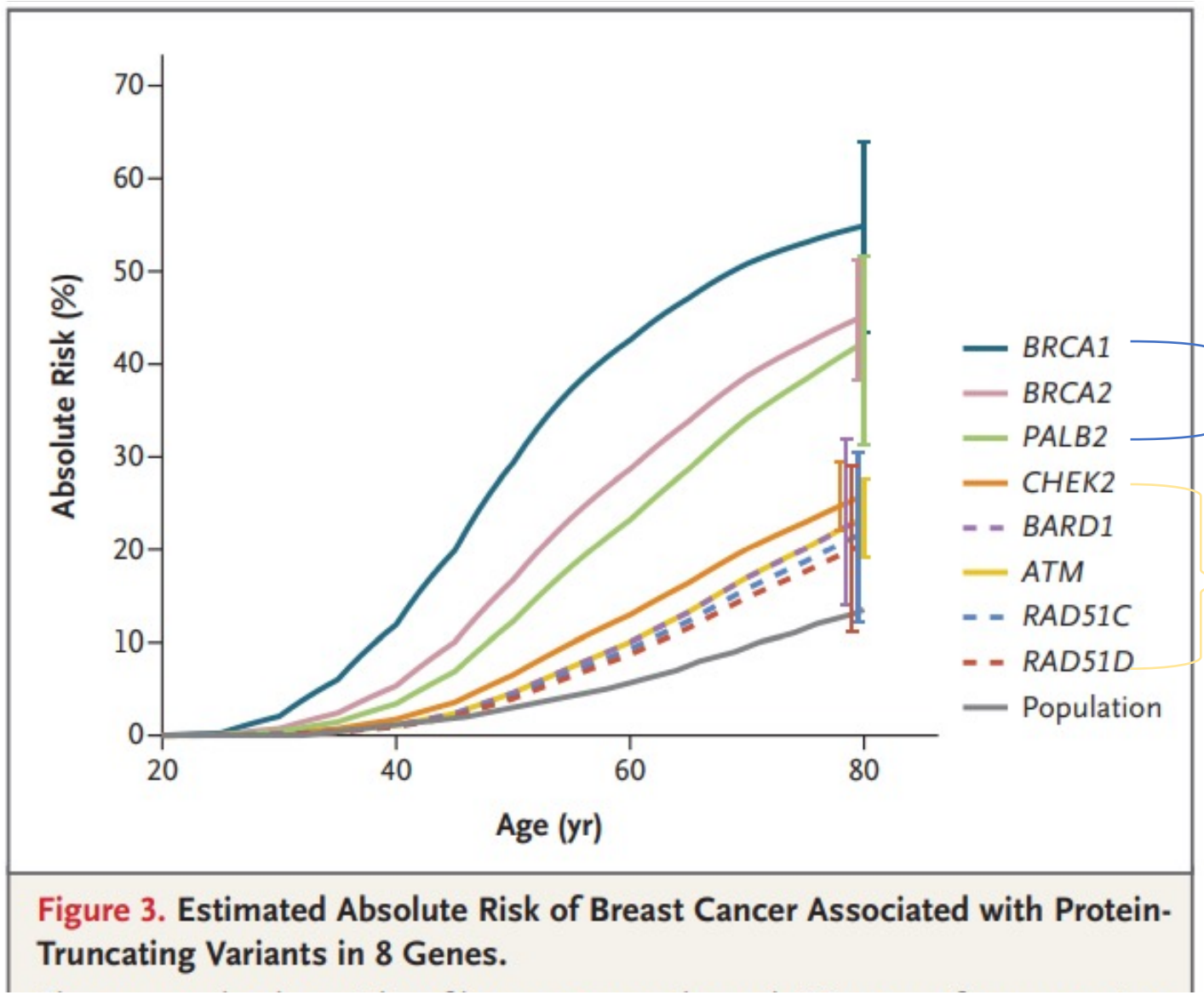


Figure 3. Estimated Absolute Risk of Breast Cancer Associated with Protein-Truncating Variants in 8 Genes.

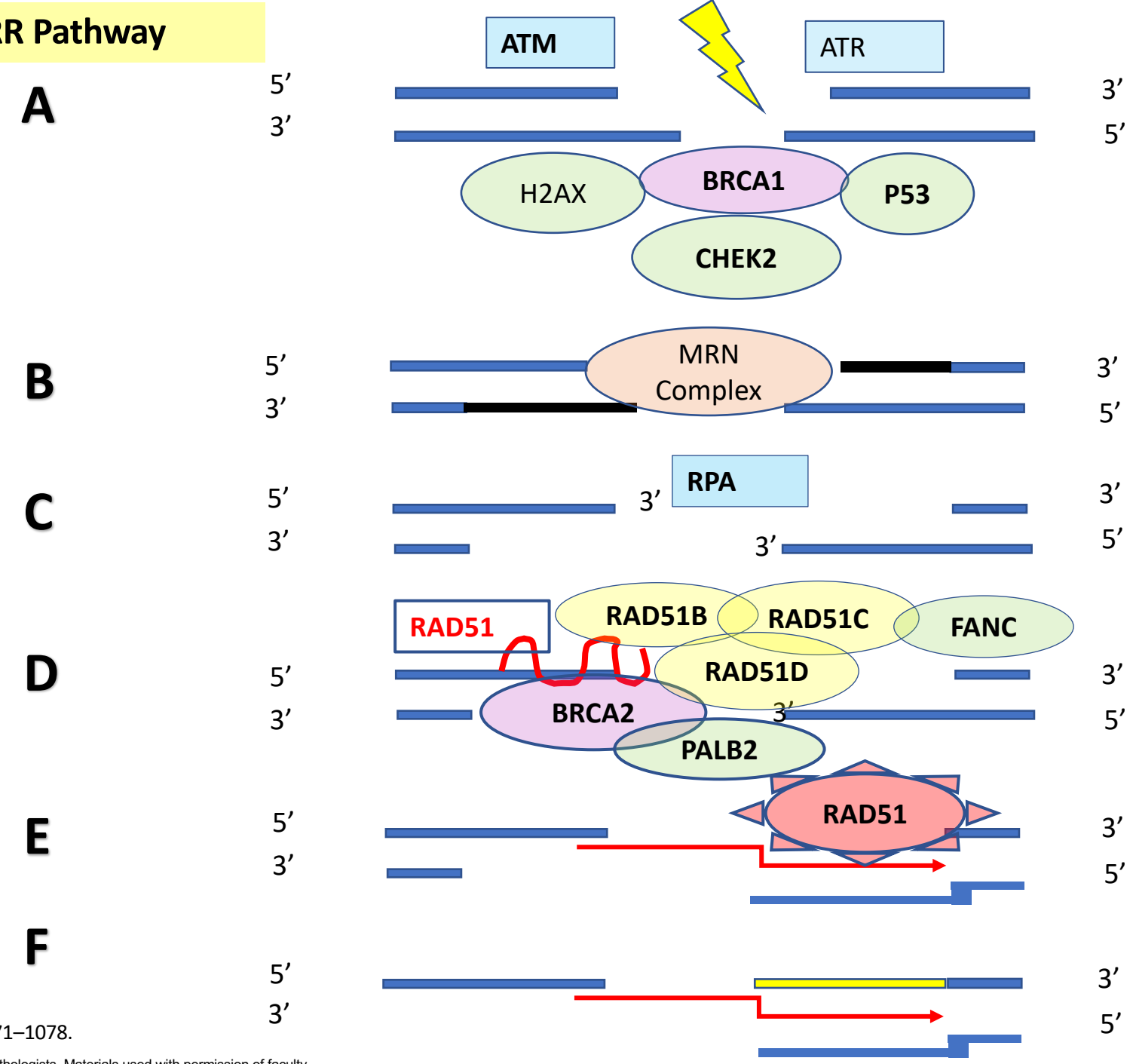
(Dorling et al. NEJM Feb 2021)

HIGH RISK GENES FOR HBC
(>30% LIFETIME RISK BY 80Y)

MODERATE RISK GENES FOR HBC
(17 - 30% LIFETIME RISK BY 80Y)

- ER POSITIVE TUMORS: ATM AND CHEK2
- ER NEGATIVE TUMORS: BRCA1, BRCA2, BARD1, PALB2, RAD51C, RAD51D

HRR Pathway



Recognition and Assembly

DNA End Resection

RPA binds to 3’ overhangs

RAD51 Loading

Homologous strand invasion

DNA Synthesis and repair

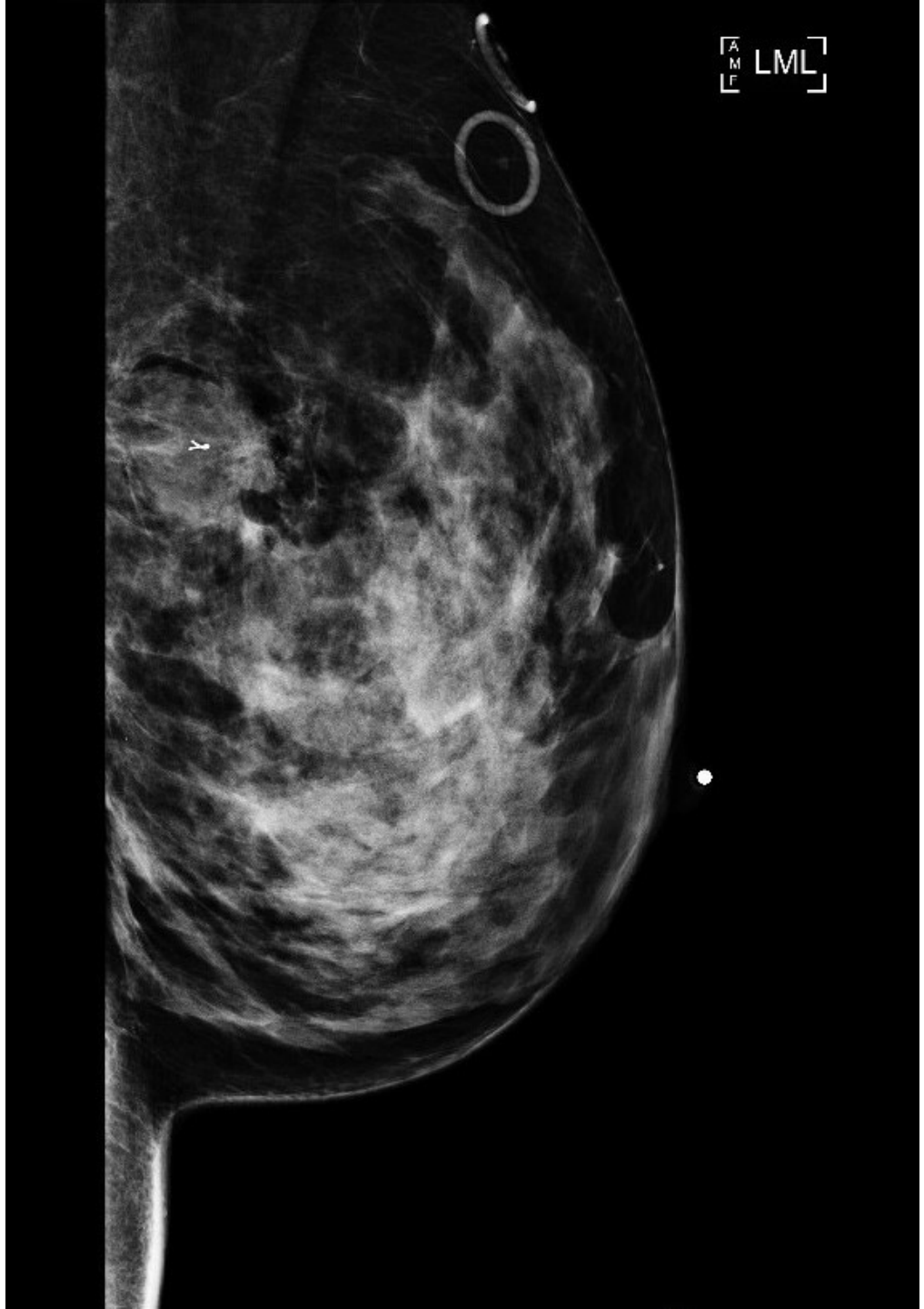
Courtesy: Dr. Rania Bakkar

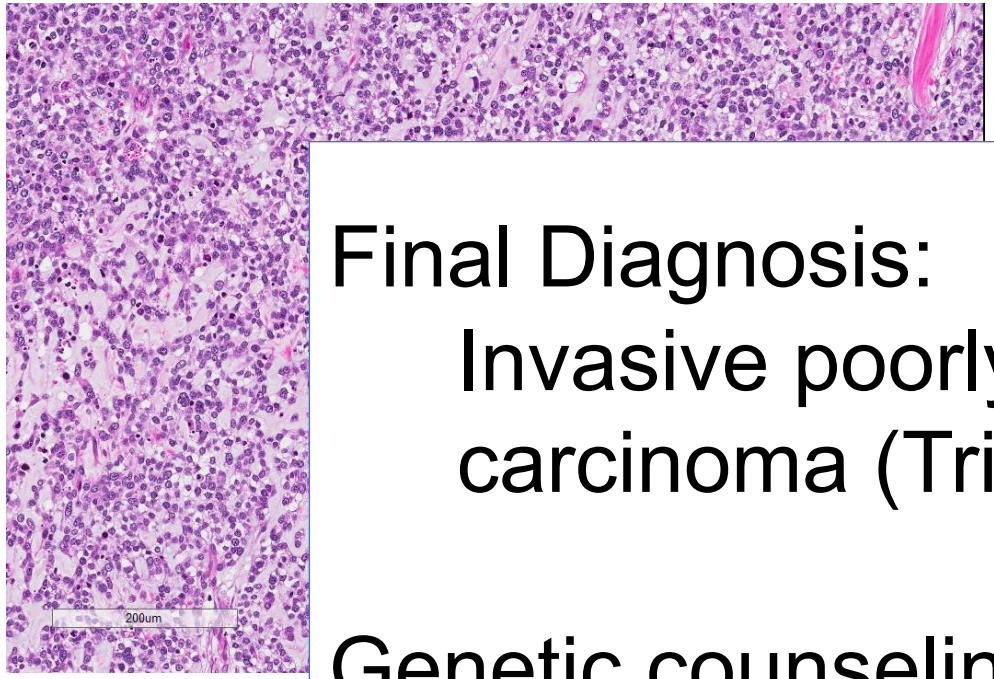
Nature 2009, *461*, 1071–1078.

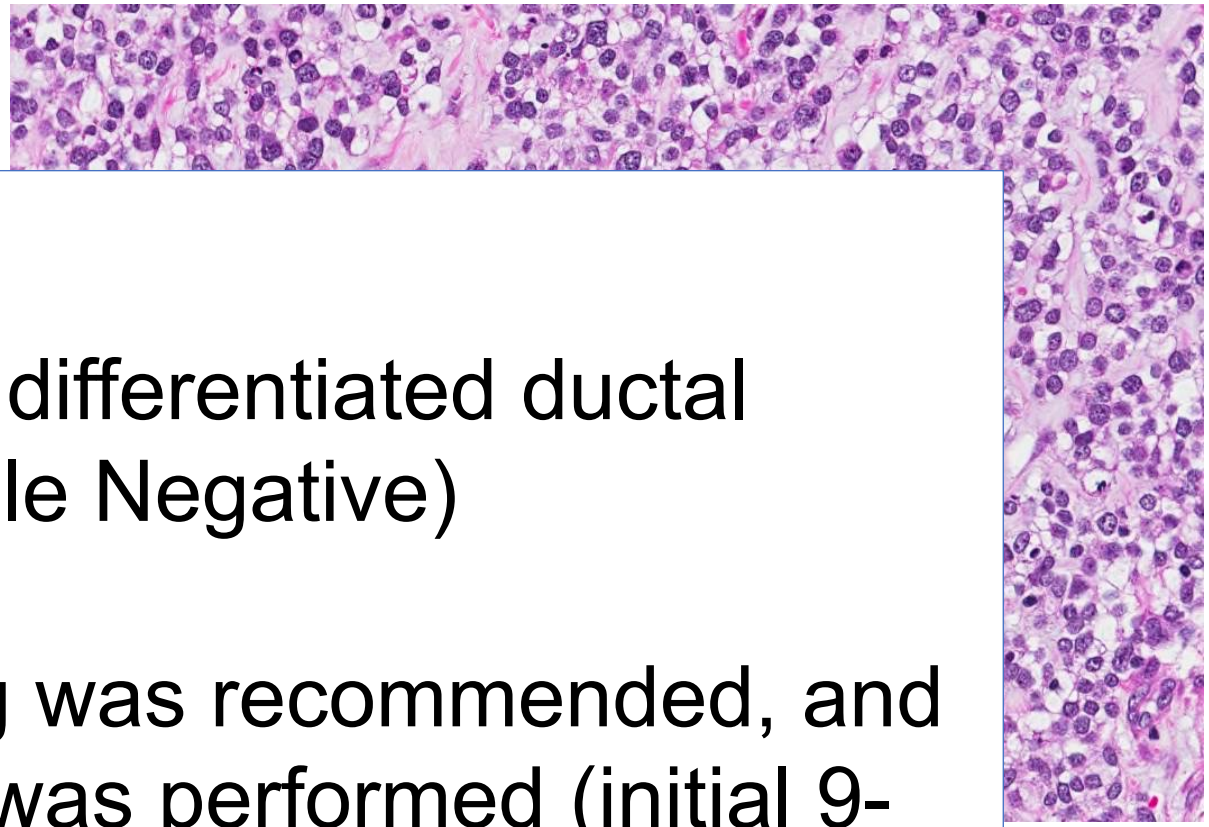
Gene	Absolute Lifetime Risk	Breast Cancer Risk and Management (Current NCCN recommendations)
BRCA1/2	>60%	Screening: Annual breast MRI with contrast starting at age 25y and annual breast MRI screening with contrast and mammogram for age 30–75 y Risk reduction: Discuss option of risk-reducing mastectomy (RRM)
TP53	>60%	Screening: Annual breast MRI with contrast starting at age 20y and annual breast MRI screening with contrast and mammogram for age 30–75 y Risk reduction: Discuss option of RRM
PTEN (Cowden Syndrome)	>60%	Screening: Annual mammography and breast MRI with contrast starting at age 35y or 10y before the earliest known breast cancer in the family Risk reduction: Discuss option of RRM
STK11	32 – 54%	Screening: Annual mammogram and breast MRI with contrast starting at age 30y Risk reduction: Discuss option of RRM
CDH1 PALB2	41 - 60%	Screening: Annual mammogram and consider breast MRI with contrast starting at age 30y Risk reduction: Discuss option of RRM
ATM CHEK2 (Frameshift P/LP mutations)	20 – 40%	Screening: Annual mammogram at age 40 y and consider breast MRI with contrast starting at age 30–35y Risk reduction: Evidence insufficient for RRM, manage based on family history
BARD1	20 – 40%	Screening: Annual mammogram at age 40 y and consider breast MRI with contrast starting at age 40y Risk reduction: Evidence insufficient for RRM, manage based on family history
NF1	20 – 40%	Screening: Annual mammogram starting at age 30 y and consider breast MRI with contrast from ages 30–50y Risk reduction: Evidence insufficient for RRM, manage based on family history
RAD51C, RAD51D	20 – 40%	Screening: Annual mammogram and consider breast MRI with contrast starting at age 40y Risk reduction: Evidence insufficient for RRM, manage based on family history

Case 3

- A 35y/F
- Ill-defined 5 cm mass in right breast
- No significant family history (FH)
- MRI-guided core biopsy



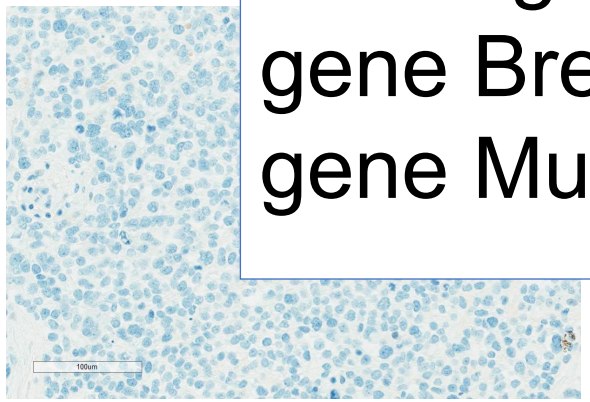




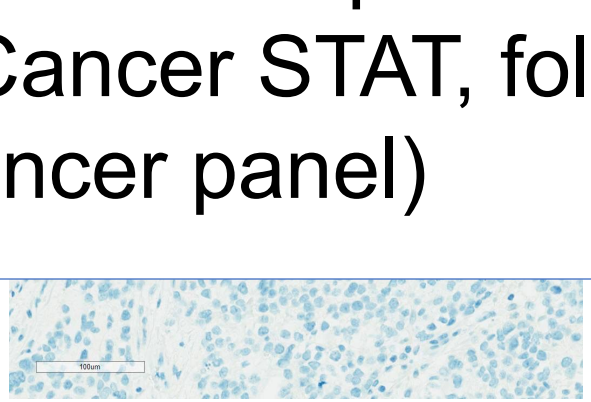
Final Diagnosis:

Invasive poorly differentiated ductal carcinoma (Triple Negative)

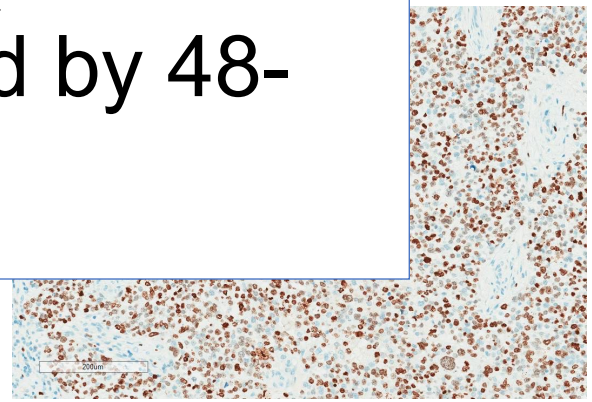
Genetic counseling was recommended, and a multigene panel was performed (initial 9-gene Breast Cancer STAT, followed by 48-gene Multi Cancer panel)



ER/PR



Her-2



Ki-67

RESULT: NEGATIVE

About this Test: This test evaluates 9 genes for variants associated with BC. Benign and likely benign variants are not included but available upon request. Diagnostic genetic testing, when combined with family history and other clinical test results/ findings, can assist in supporting clinical diagnosis, individual risk assessment and personalized management plan development.

GENE	TRANSCRIPT	GENE	TRANSCRIPT	GENE	TRANSCRIPT
ATM	NM_000051.3	CDH1	NM_004360.3	PTEN	NM_000314.4
BRCA					4
BRCA					5

NCCN recommends to test or at least ask that affected family members be tested (if applicable) in cases when an affected individual gets a negative test result

Summary of Recommendations:

No reportable genetic variants were identified by this analysis; however, this individual may still be at risk for certain medical conditions based on other factors such as family history, genetic causes not evaluated by this test or other environmental influences. This result should be discussed with a health care provider, such as a genetic counselor, clinical follow up of this individual and surveillance of family members may still be indicated. This result should be interpreted within the context of additional laboratory results, family history and clinical findings.

Hereditary Breast Cancer Workflow

Personal or family history of breast/ gynecological cancers/ancestry/known *BRCA* mutations

Oncologist/ geneticist

NCCN Guidelines

Familial risk assessment tools
-Ontario family history assessment tool
-Manchester scoring system
- Tyrer-Cuzick
-BRCAPRO
-BCSC

Multigene panel testing

Positive for PV/ LPV

-Genetic counselling
-Risk reducing strategies (intensified screening strategies, risk reducing surgery, chemoprevention)
- Targeted therapy

Negative for PV/ LPV

True Negative

Uninformative

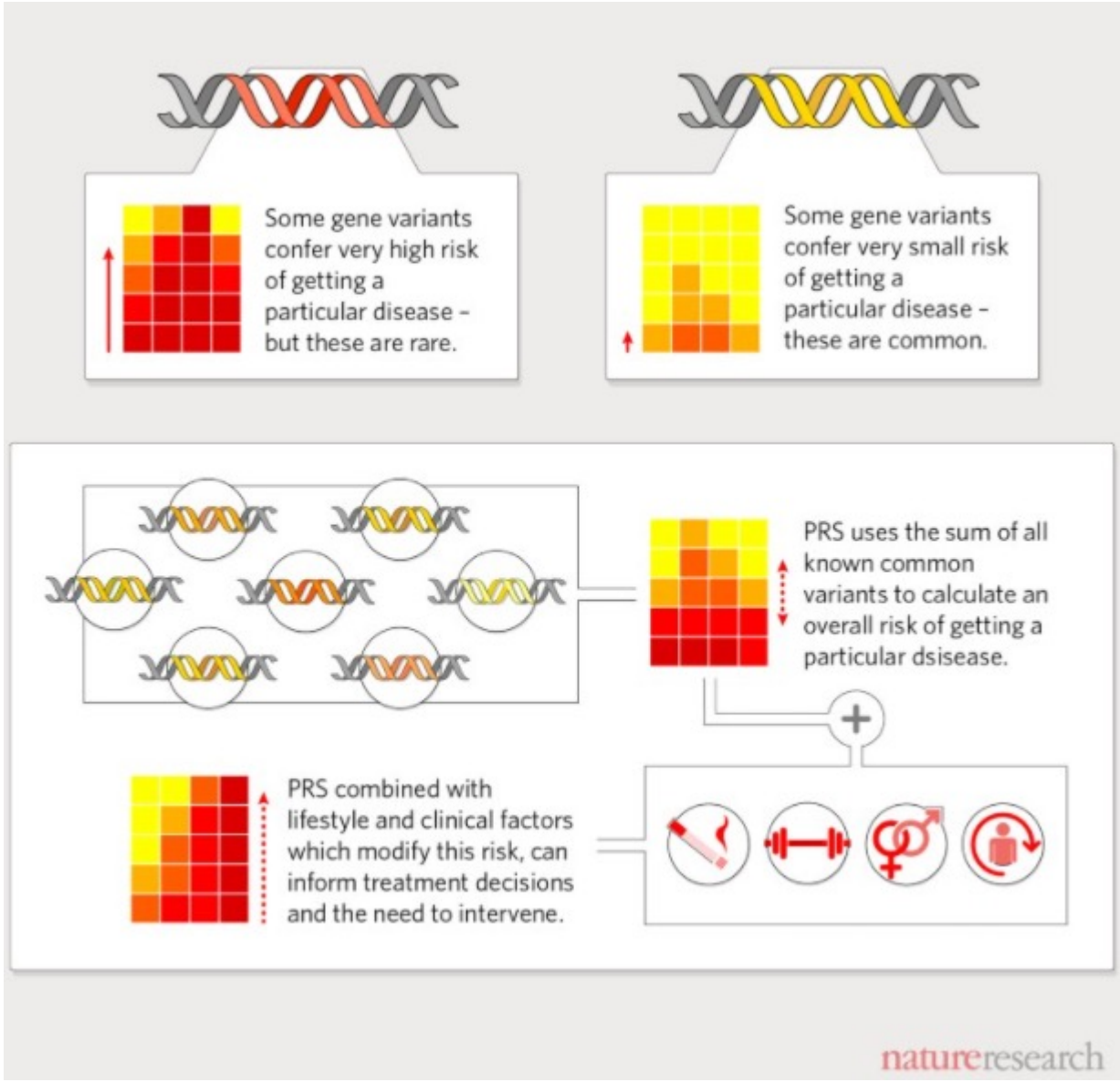
Polygenic Risk Score

VUS

-Management according to family history and other risk factors
-Continued follow up in case of re-classification

Polygenic Risk Score for Breast Cancer

- Additional low penetrance common genetic variants
  - Single Nucleotide Polymorphism (SNP)
  - >182 SNPs identified by over 100 genome wide associations studies (GWAS)
- Minimal risk associated with each allele, when combined substantial risk
- PRS= sum of the log odds ratios for each common risk associated variant
- Additional 18% of HBC risk
- HR+ BC, ductal histotype



Michallidou K et al. Nature 2017

Association of a Polygenic Risk Score With Breast Cancer Among Women Carriers of High- and Moderate-Risk Breast Cancer Genes

The 86 SNV score is associated with modified risk for carriers of *BRCA1*, *BRCA2*, *CHEK2*, *ATM*, and *PALB2* PVs

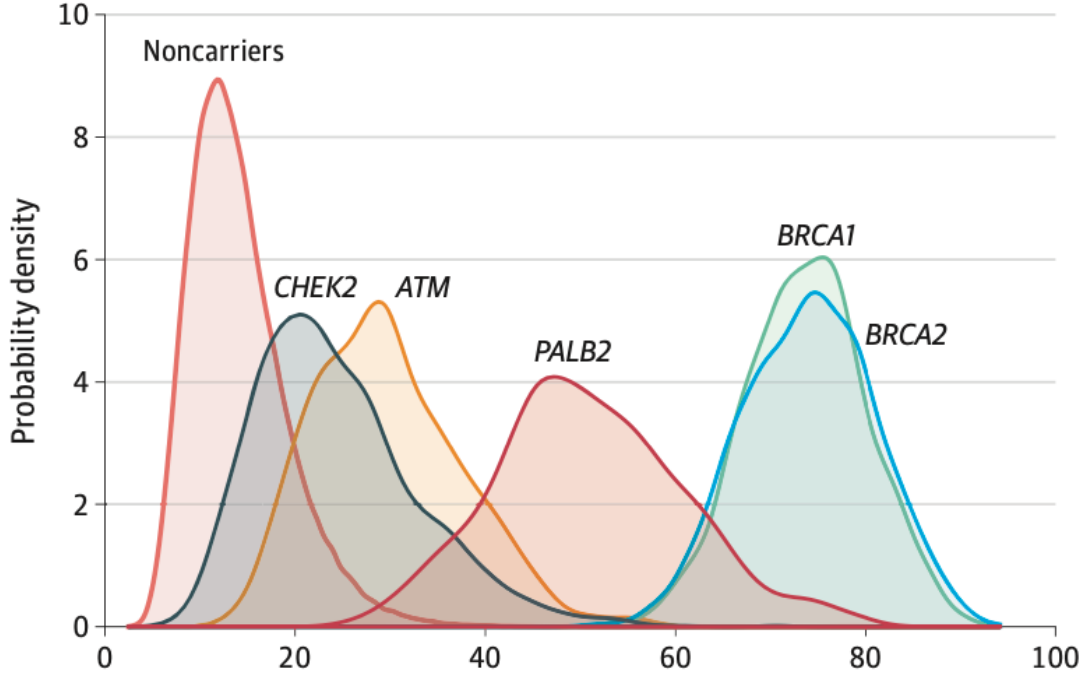


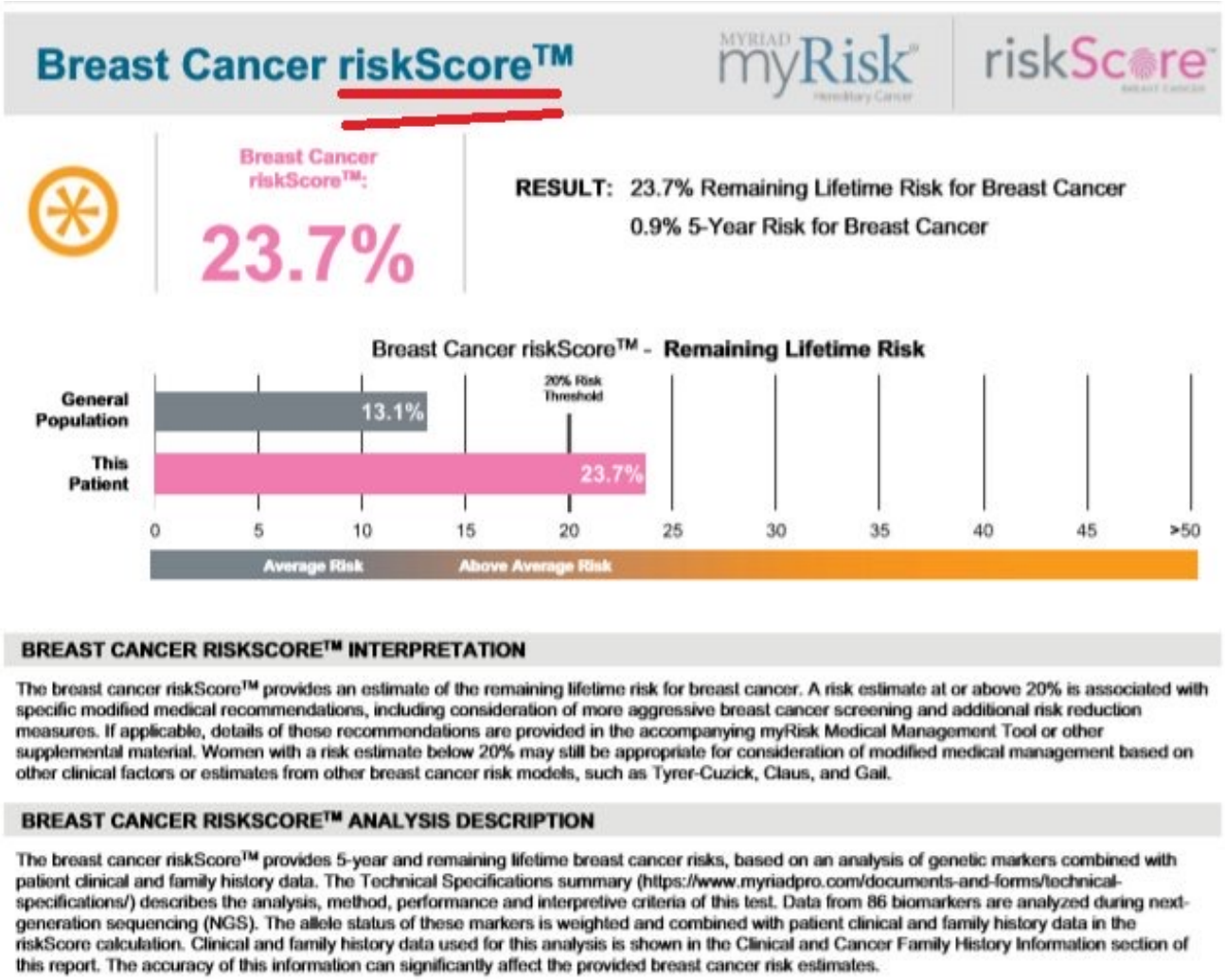
Table 4. Estimated Lifetime Breast Cancer Risk to Age 80 Years and Modification by an 86-SNV Score

Gene[a]	Gene-based risk, %	Adjusted lifetime risk, %				
		Minimum	Quintile 1	Median	Quintile 3	Maximum
ATM[31]	28.2	12.9	23.9	29.0	34.7	58.3
BRCA1[31]	73.5	53.1	69.4	73.8	77.9	91.5
BRCA2[31]	73.8	50.8	69.0	74.2	78.9	94.2
CHEK2[17]	22.1	6.6	18.1	23.0	29.1	70.6
PALB2[31]	50.1	26.2	44.4	50.3	57.3	79.2
Noncarriers[32,33]	12.7	2.5	10.4	13.2	16.9	62.4

JAMA Network Open. 2020;3(7):e206501.
doi:10.1001/jamanetworkopen.2020.8501

Polygenic Risk Score: Benefits and Limitations

- To assess whether risk reducing intervention should be considered even in the absence of high-risk pathogenic variant
- To further stratify risk in carriers of high-risk pathogenic variants
- Personalization of population-based screening (20% BC risk improvement)
- Limitation
  - Limited evidence and consensus to support implementation
  - Lack of enough studies in non-European ancestry

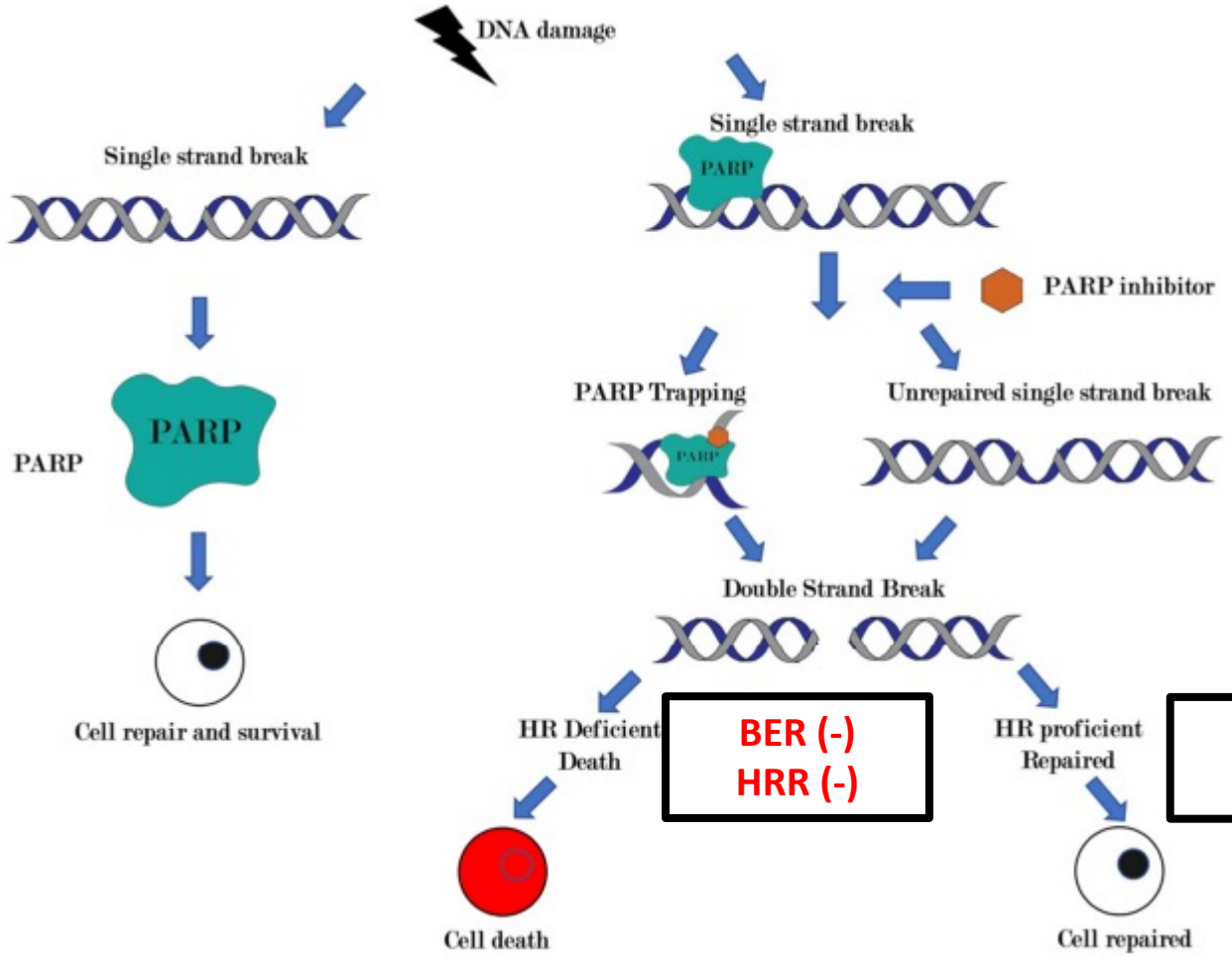


Yanes et al. Breast Cancer Research (2020) 22:21

Germline Testing in Breast Cancer

- WHO is eligible for testing?
- WHAT panel to use for germline testing?
- WHICH genes to include in the testing panel?
- WHAT is the magnitude of risk?
- HOW to utilize the test results clinically?

PARP-inhibitors and the Concept of Synthetic Lethality



Base-excision repair (BER)

BER (-)
HRR (-)

BER (-)
HRR (+)

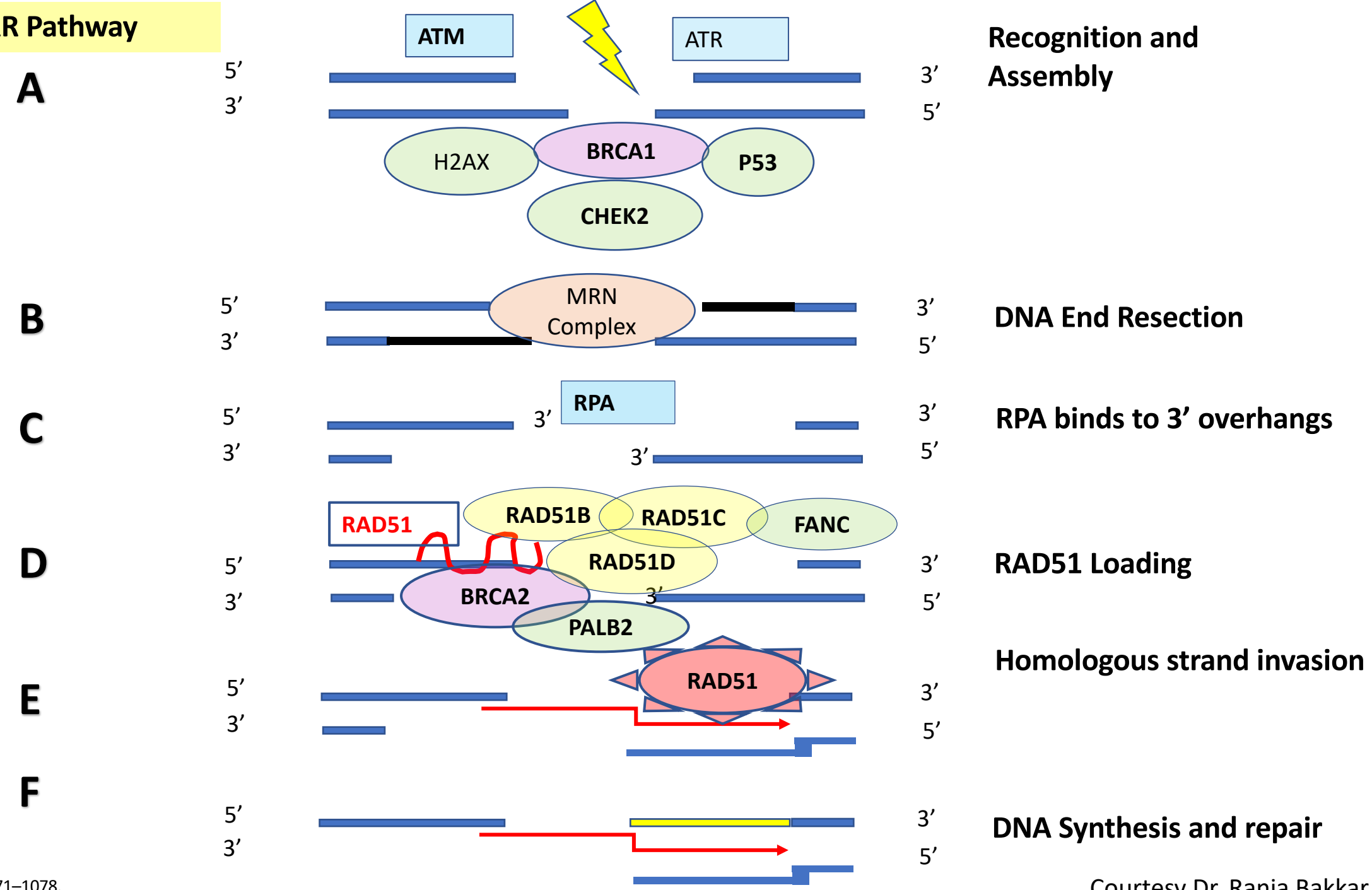
Synthetic lethality

Cell repair

PARP-inhibitors as promising targeted treatment options for all cancers with HRD, including HBCs

Chan WY et al. Cancers 2021, 13, 4520

HRR Pathway



Nature 2009, *461*, 1071–1078.

Courtesy Dr. Rania Bakkar

PARP-inhibitors in Treatment of HBCs

***Olaparib**	FDA approved (OlympiAD)	germline *BRCA* mutations and HER2-negative breast cancer who have previously been treated with chemotherapy in the neoadjuvant, adjuvant, or metastatic disease setting
***Talazoparib**	FDA approved (EMBRACA)	patients with germline *BRCA* mutations and HER2-negative, locally advanced, or metastatic breast cancer
Niraparib	BRAVO (phase III ongoing)	previously treated, Her2- negative, gBRCA mutated, metastatic BC, ≤2 previous therapies for metastatic disease
Olaparib	Clinical trials (TBCRC 048 phase II)	patients with MBC and germline PALB2 or somatic BRCA1/2 mutation
Veliparib	BROCADE-3 (phase III ongoing	HER2 negative germline BRCA mutated breast cancer
Olaparib + Trastuzumab	OPHELIA (phase II ongoing)	Metastatic HER2-positive BRCA-mutated breast cancer

*Proven to be superior to conventional chemotherapy for progression-free survival (PFS), response and toxicity; however, no change in overall survival (OS)

PARP-inhibitors in Combination with Immunotherapy in Treatment of HBCs

Niraparib + Pembrolizumab	TOPACIO (phase I/II active)	Advanced or metastatic triple negative breast cancer or recurrent ovarian cancer
Olaparib + Durvalumab	MEDIOLA (phase I/II active)	Advanced solid tumors (NSCLC, gBRCA metastatic TNBC, gBRCA metastatic ovarian cancer, gastric cancer)
Talazoparib + Avelumab	JAVELIN BRCA/ATM (phase II active)	Locally advanced or metastatic solid tumors with BRCA or ATM defect

Genomic instability due to HRD may result in increased immunogenicity and response to immunotherapy

Need for developing biomarkers for predicting response to PARPi

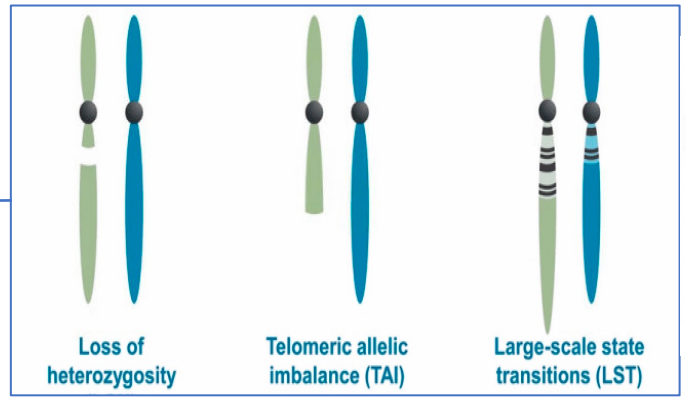
- Acquired resistance to PARPi is common

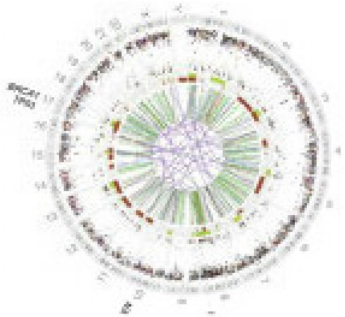
- Mechanisms for resistance to PARPi
  - PARPi efflux
  - PARP mutations
  - Restoration of HR/ BRCA1/2 functions
  - Replication fork stalling

Lee EK *Cancers (Basel)*. 2020;12(8):2054.

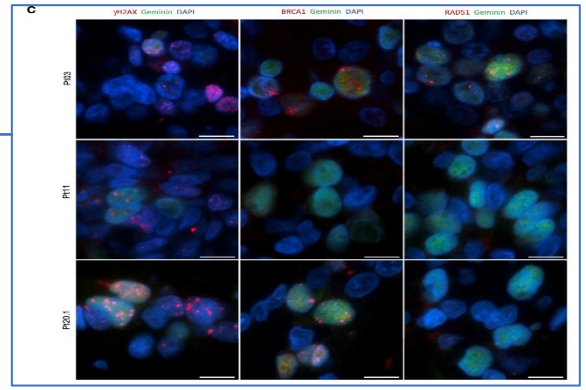
PREDICTIVE BIOMARKERS

- CLINICAL
  - Platinum sensitivity
- HRR GENE MUTATIONS
  - Germline BRCA mutations
  - Germline non-BRCA HRR mutations
  - Somatic BRCA mutations
  - Tumor BRCA mutations
- GENOMIC SIGNATURES AND SCARS (HRD SCORE)





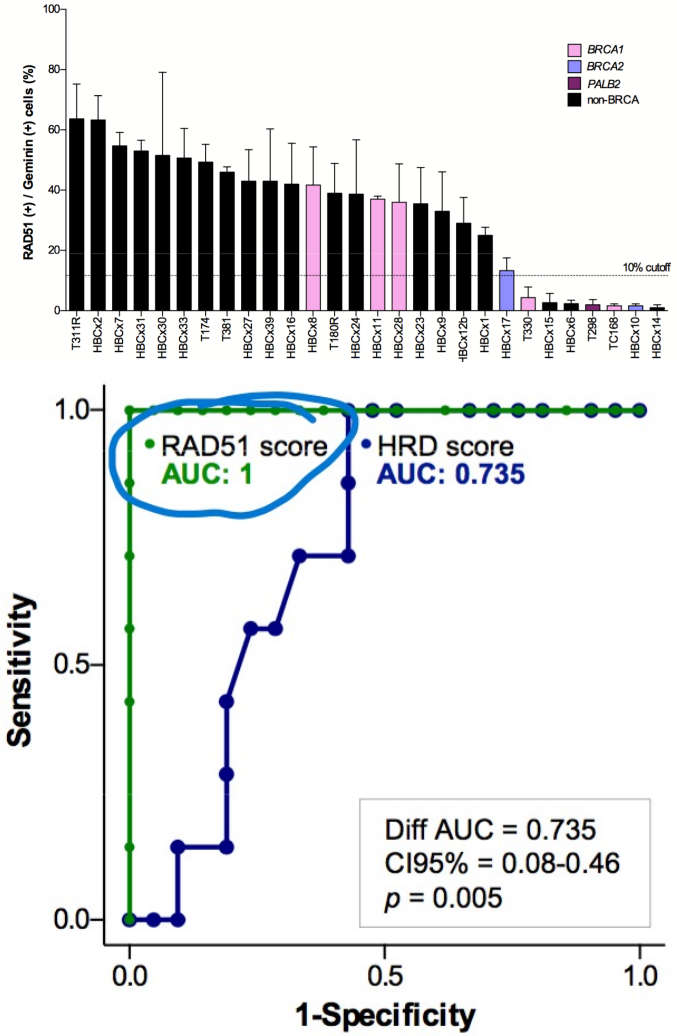
- FUNCTIONAL RAD51 ASSAY



- FDA approved for ovarian cancers
- Clinical trials and research use for breast cancers

A RAD51 assay feasible in routine tumor samples calls PARP inhibitor response beyond BRCA mutation

- Untreated g*BRCA* tumors and an independent TNBC cohort
- Correlated with PARPi resistance regardless of the underlying mechanism of HRR function restoration
- Lack of RAD51 nuclear foci associated with PARPi response
- Identifies HRR-deficient tumors among patients with hereditary breast and ovarian cancer syndrome, including *PALB2*-related tumors, *RAD51C* or *RAD51D*
- A RAD51 score cutoff of 10% predicted the response to PARPi
    - with high specificity and sensitivity, outperforming the HRD score



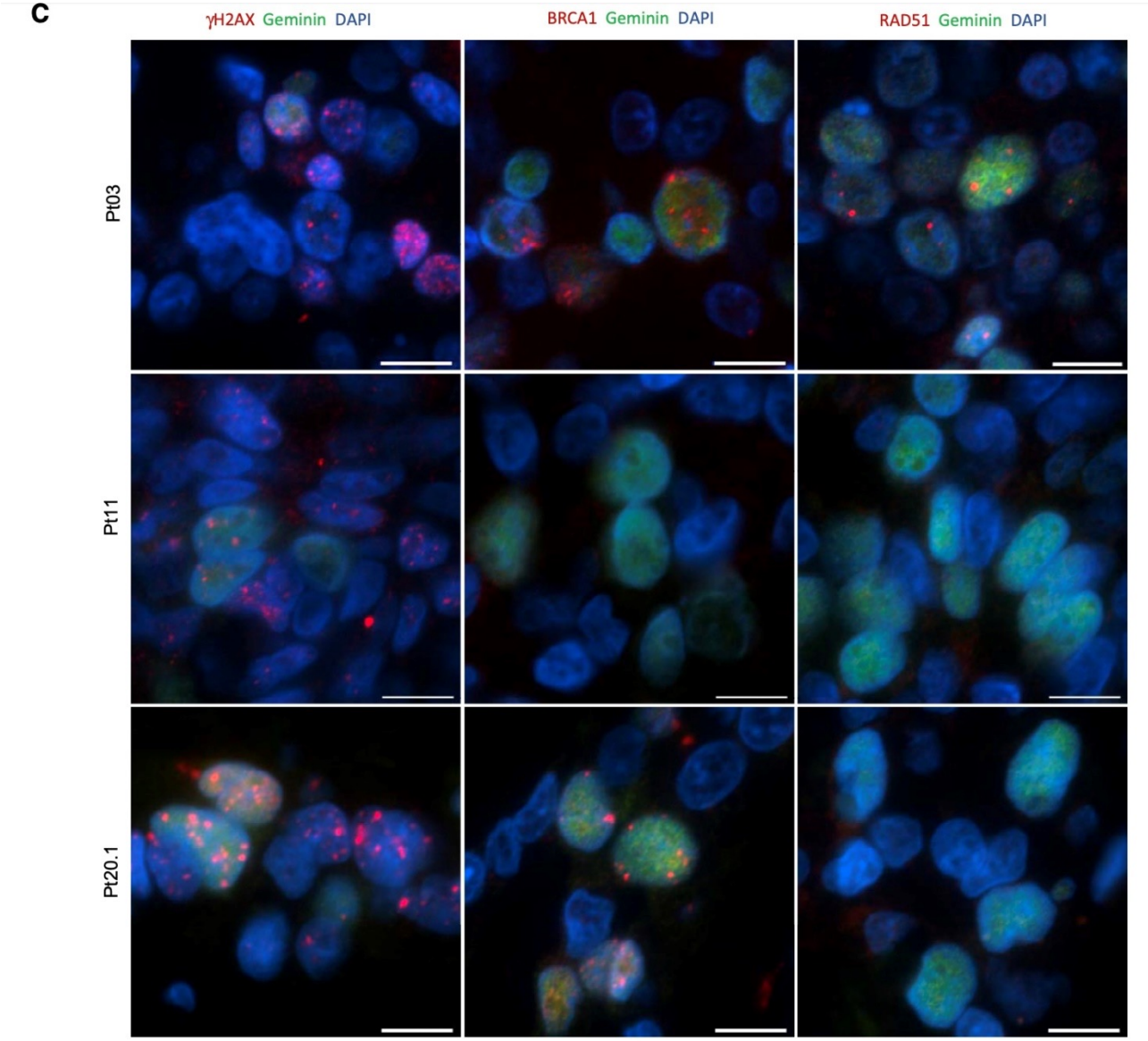
Castroviejo-Bermejo et al. EMBO Mol Med (2018) 10: e9172

RAD51 Functional Assay

HRR Proficient

HRD

HRD



Immunofluorescence based assay on FFPE tumor tissue

- Lack of RAD51 foci denotes HRD and potential response to PARPi

Castroviejo-Bermejo et al. EMBO Mol Med (2018) 10: e9172

ESMO 2021

Detection of homologous recombination repair deficiency (HRD) in treatment-naive early triple negative breast cancer (TNBC) by RAD51 foci and comparison with DNA-based tests

- To evaluate RAD51 assay and correlate this with HRD score or treatment activity

- The RAD51 test is feasible in treatment-naive FFPE tumor samples from early TNBC to assess the functional status of HRR and identifies PARPi-sensitive tumors

V. Serra et al.

Association of RAD51 with Homologous Recombination Deficiency (HRD) and clinical outcomes in untreated triple-negative breast cancer (TNBC): analysis of the GeparSixto randomized clinical trial

- To compare the performance of RAD51assay with HRD tests and assess its capacity to select patients with primary TNBC sensitive to platinum-based neoadjuvant chemotherapy (NACT)

- The RAD51 test highly concordant with BRCA mutation and HRD

- RAD51 independently predicts clinical benefit from adding Carboplatin to NACT in TNBC

- Results support further development to incorporate RAD51-testing in the clinical decision making

A. Llop-Guevara et al.

Take Home

- Non-BRCA hereditary breast cancer susceptibility genes
- High risk for penetrance (>30% LFTR)
  - BRCA1, BRCA2, and PALB2 (Common)
  - TP53, PTEN, CDH1 (Rare)
- Moderate risk for penetrance (17-30% LFTR)
  - ATM, CHEK2, BARD1, RAD51C, RAD51D, MSH6 etc.
- Common histologic subtype
  - Mostly TN (BRCA1, BRCA2, PALB2, RAD51 etc.)
  - ER positive (ATM and CHEK2)
  - HER2 positive (TP53)
- Multigene panel tests have a higher diagnostic yield for HBCs
- Patients with VUS should not be treated as patients with pathogenic variants
- PARPi are FDA approved for locally advanced or metastatic TNBCs with germline BRCA mutations (clinical trials in progress for other indications including, non-BRCA germline, somatic BRCA, ER and HER2 positive breast cancers)
- Need for developing robust predictive biomarkers (RAD51 etc.)