

Genomic Instability and the Cellular Response to Ionizing Radiation

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Cancer Genomic Instability

Chromosomal number: aneuploidy

treatment strategies needed

Large scale deletions and rearrangements

HRD: platinum/RT, PARP inhibitors

APOBEC-type mutational phenotype

Increased C-T mutations: therapy opportunities?

Microsatellite Instability

high mutational burden: better immunological responses

DNA Replication Errors

high mutational burden: immunotherapy

None of the above: key transformations

classic targeted therapy: Gleevec - CML

Homologous Recombination (HR) in Cancer

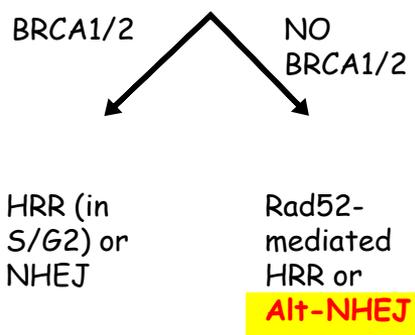
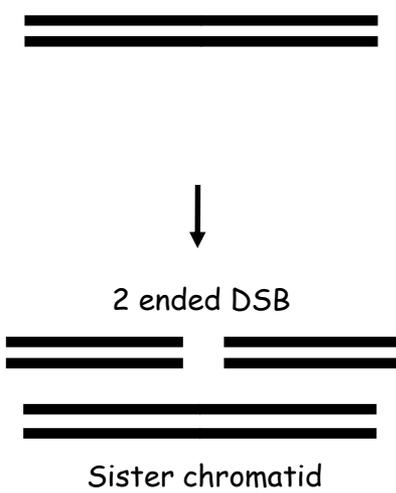
- **Many cancer cells have a greater dependence on HR for repairing double-strand DNA damage**
 - Cell cycle distribution, p53 status, stem cell program
- **A subset of cancers have defective HR**
 - HR defective: target the back-up pathway: "Synthetic Lethality"
 - Rad52; TLS polymerases; POLQ (AltEJ); Base-excision repair?
- **Clinical testing of tumor samples for defects in HR suggests that:**
 - significantly more prevalent than just BRCA1/2 mutation carriers
 - mechanism?

Pathways for repairing DSB:

- Non-homologous end-joining: canonical and alternative pathways
- Homologous recombination (gene conversion): mechanisms and proteins
- Other homology-directed pathways (single-strand annealing)
- Choice of repair partner: sister chromatid, homologous chromosome, ectopic sequences (e.g., Alu and other repeats)
- Collaboration and competition of DSB repair pathways

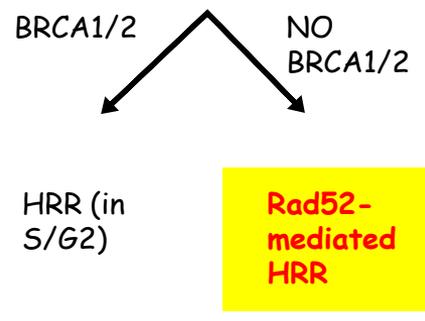
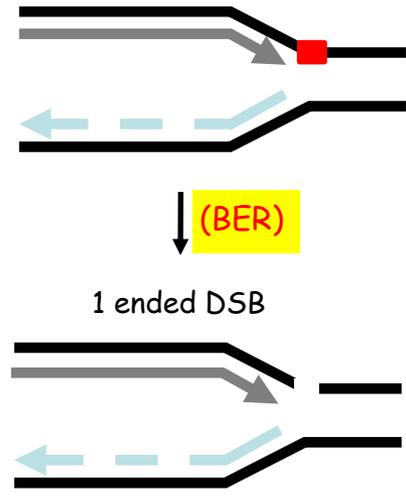
Repair of a DSB

Exposure to DSB agents



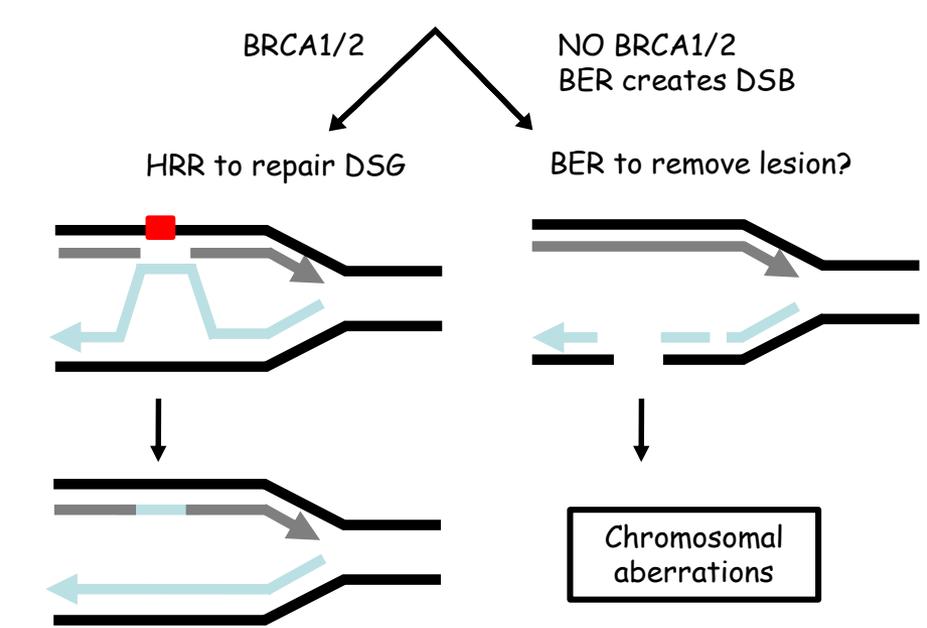
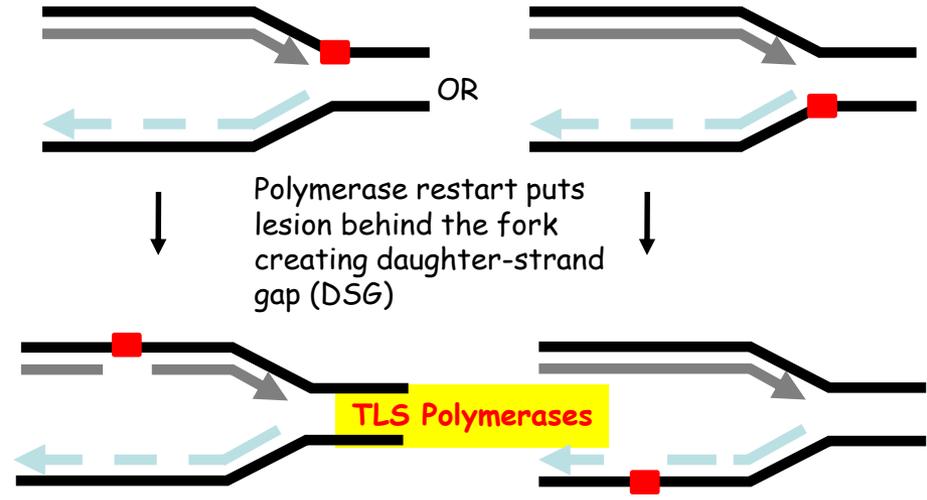
Repair at a fork

Stalled replication fork

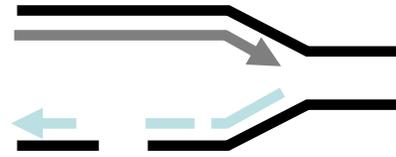


Repair behind a fork

Blocking lesion at replication fork



BRCA1/2 "deficient" cancers produce DNA breaks



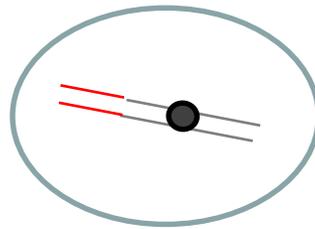
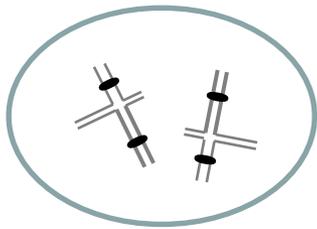
Accumulation of chromatid breaks (CTB)

Radials

Deletions + Insertions

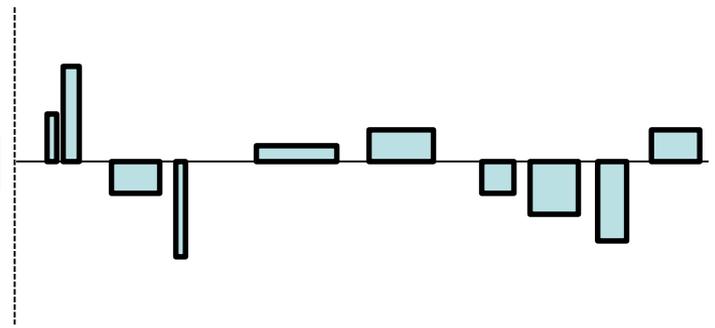
Dicentrics
(cell death)

Translocations
(cell survival)

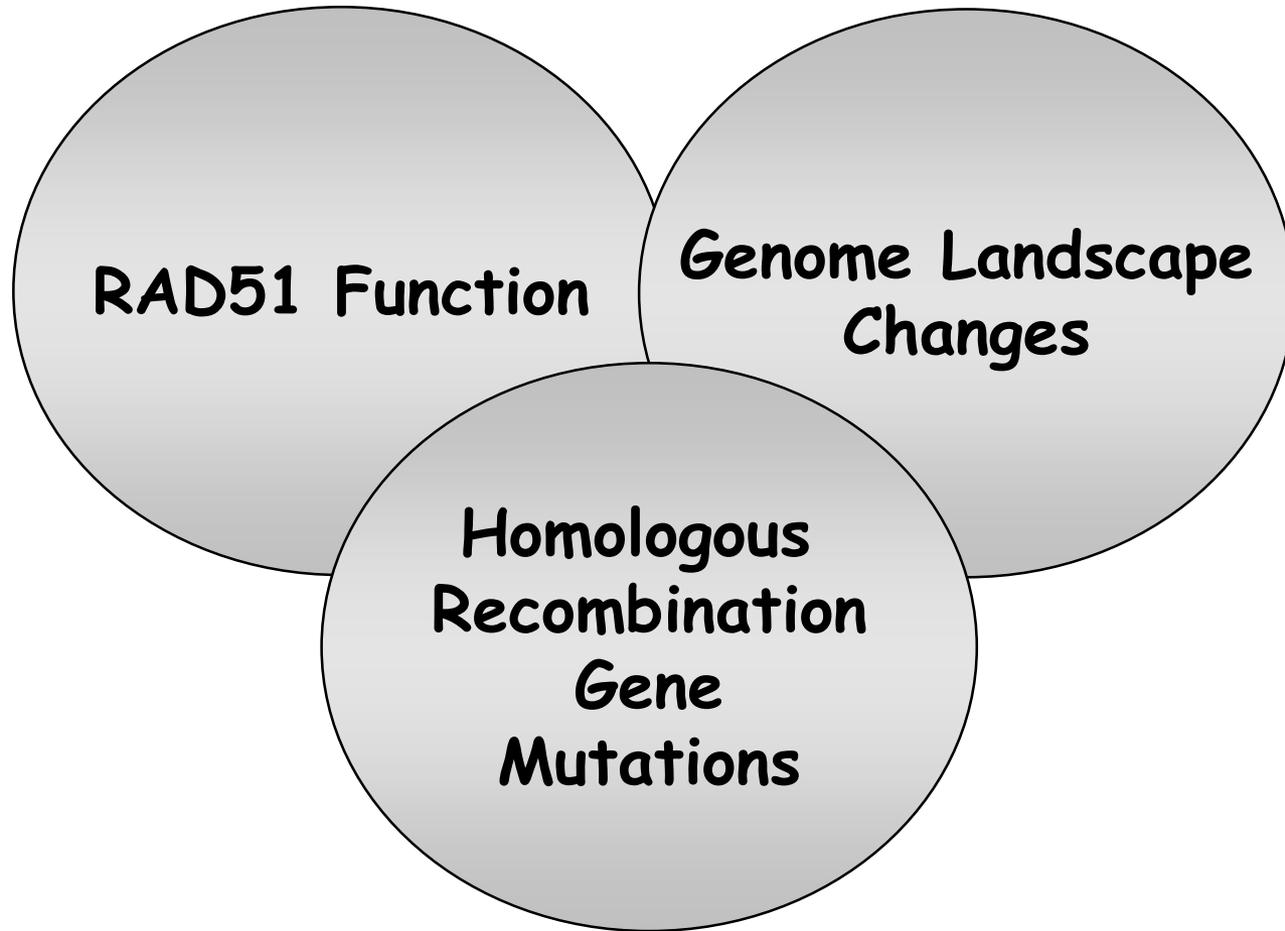


Translocation produced by
exchange and cell division

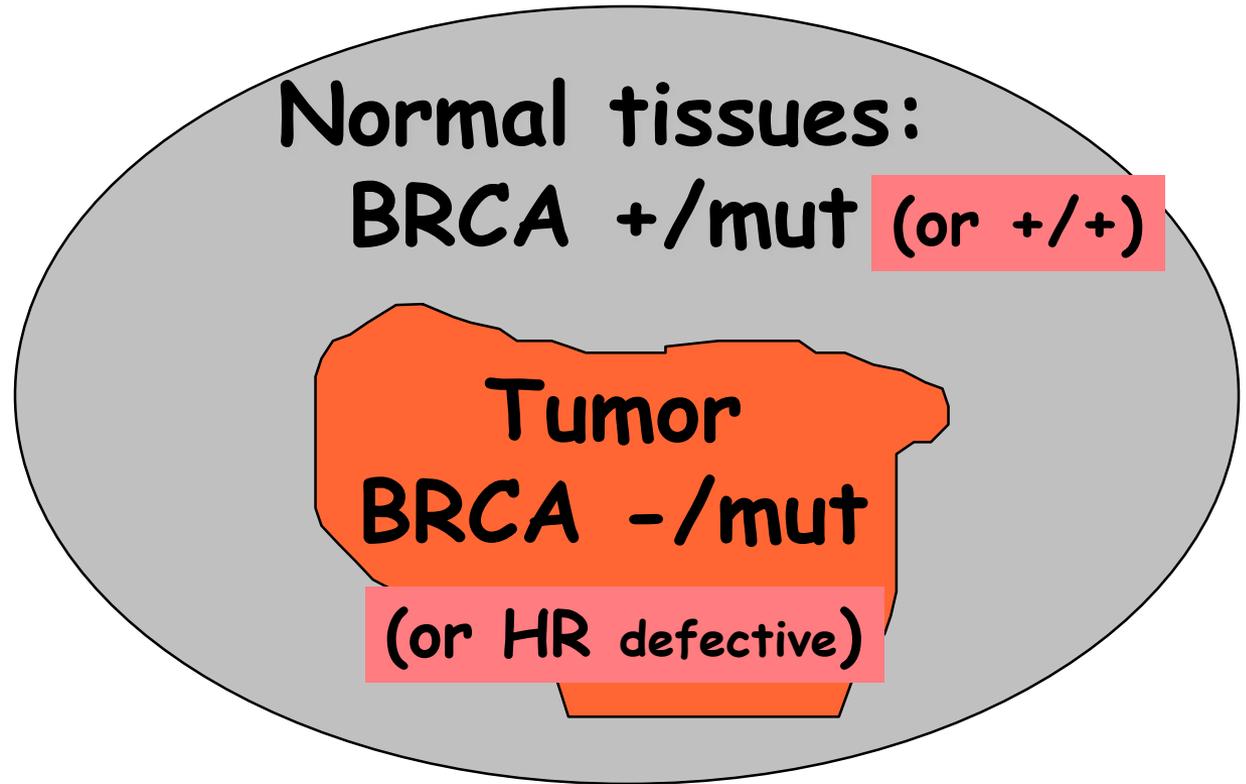
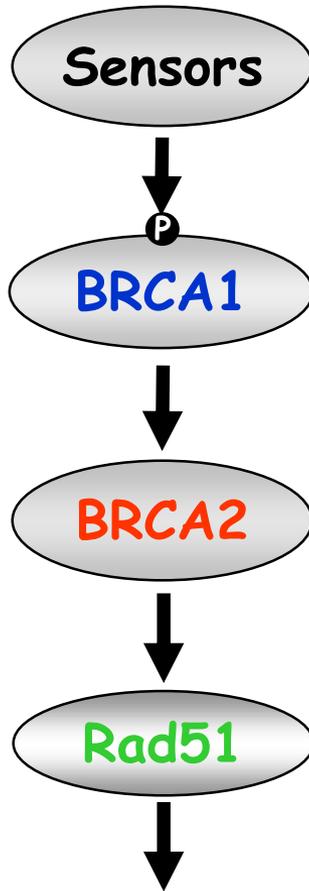
aCGH



BRCA1-BRCA2 Pathway Defects in Breast Cancer

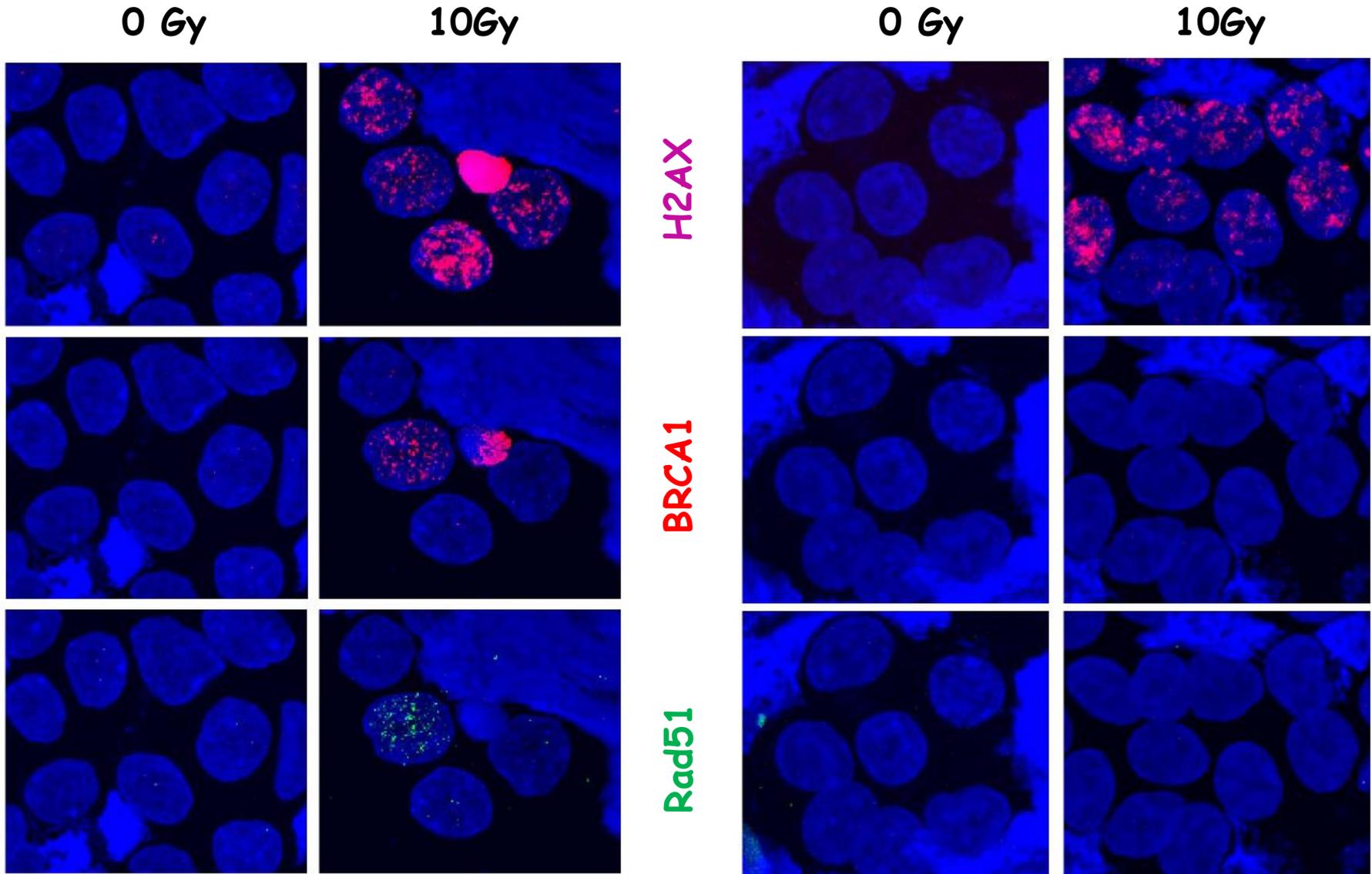


BRCA1-BRCA2 Pathway Defects in Breast Cancer

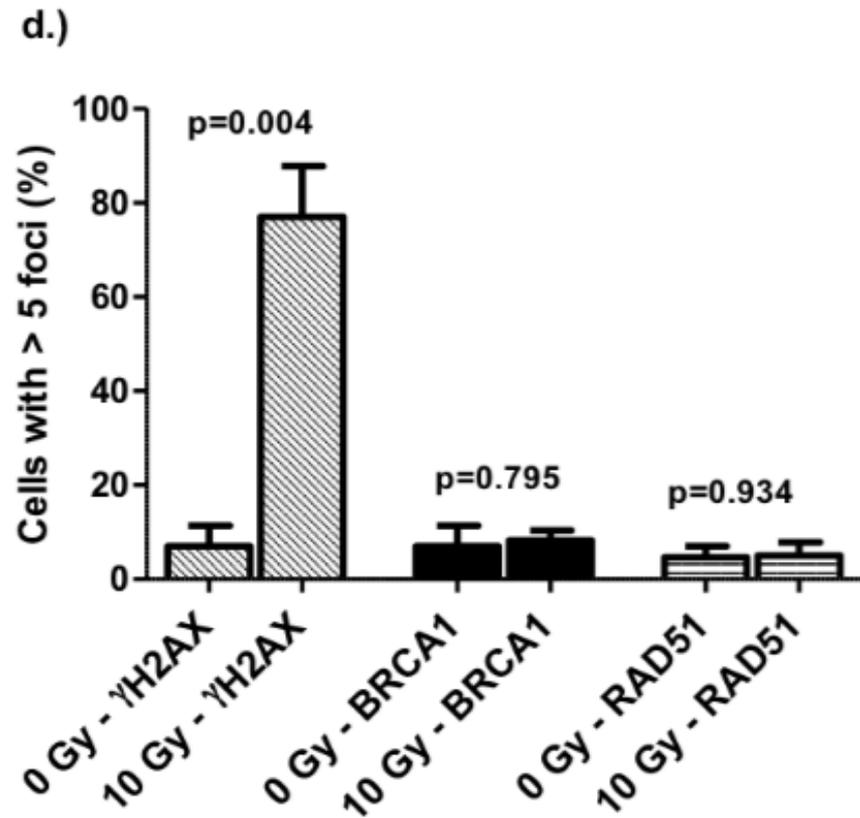
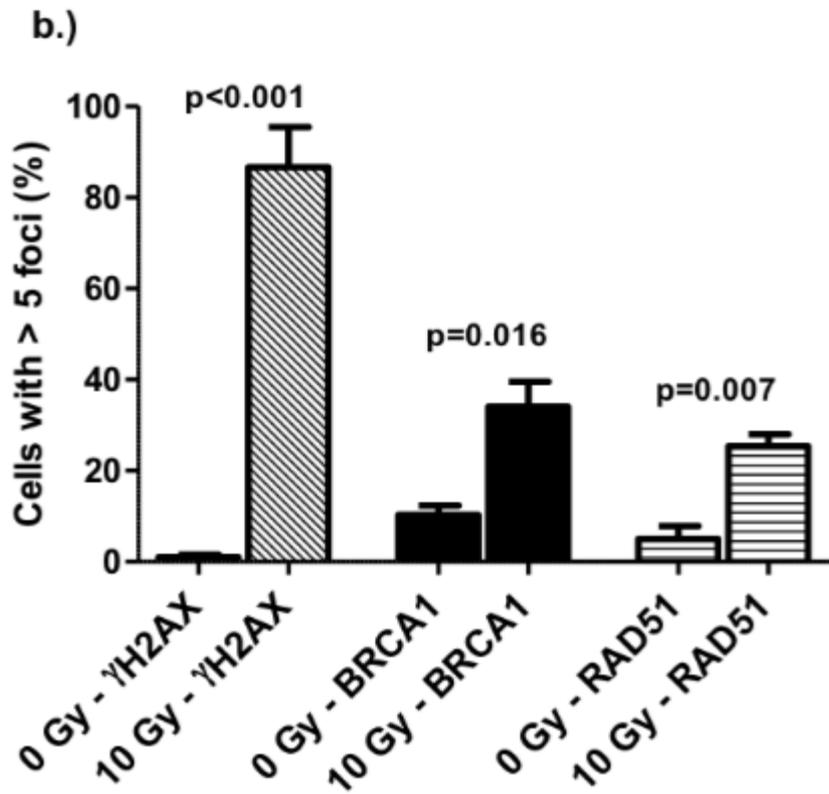


Homologous Recombination

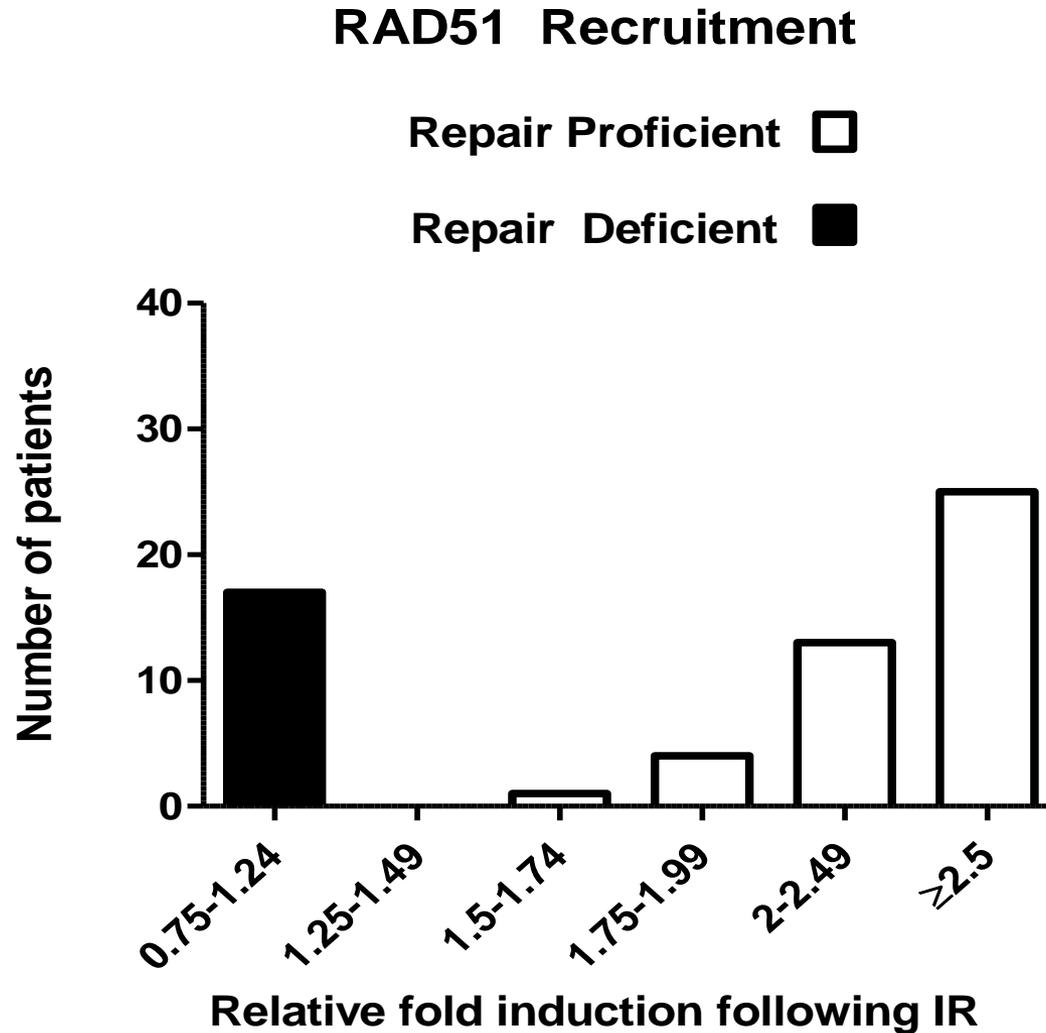
RAD51 Function in Human Tumor Cells



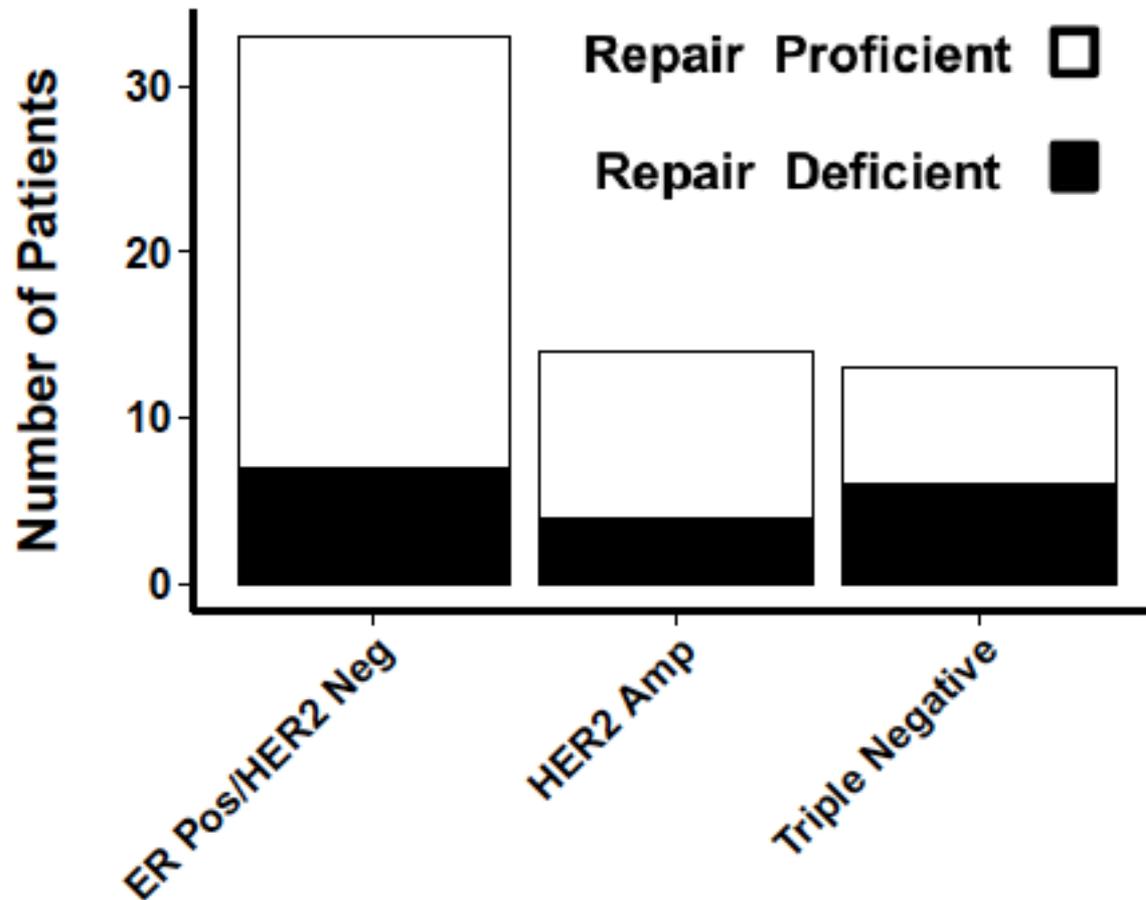
Quantification of foci data



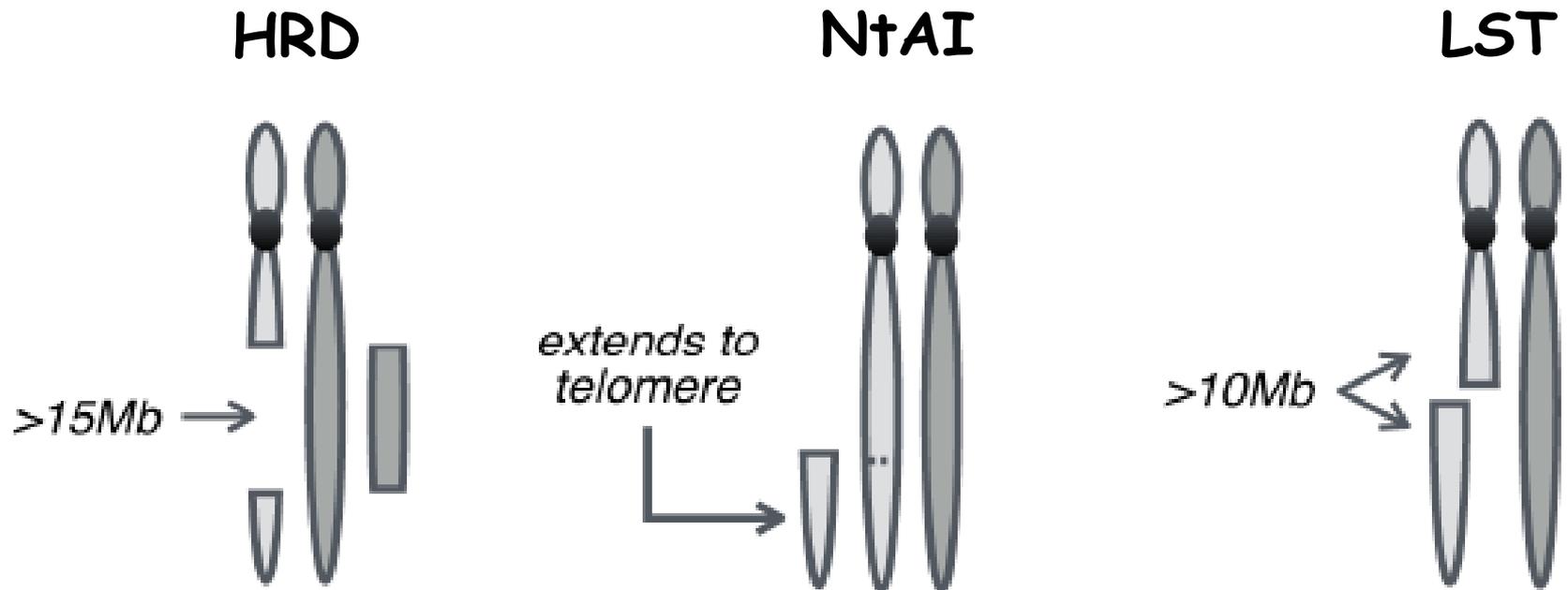
BRCA1-BRCA2 Pathway Defects as measured by RAD51 Function



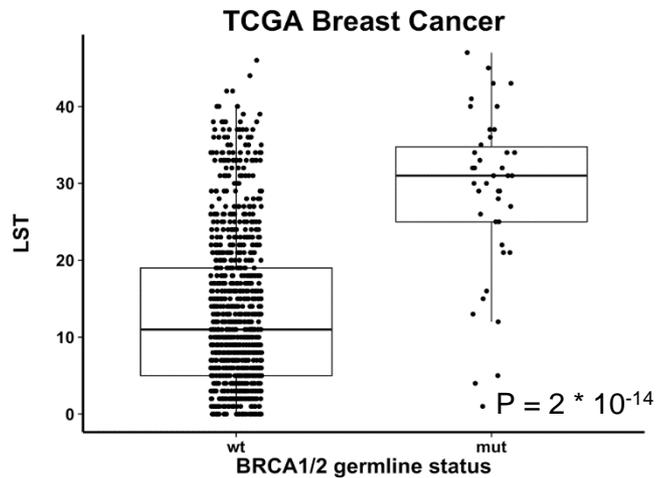
RAD51 Function by Breast Cancer Sub-types



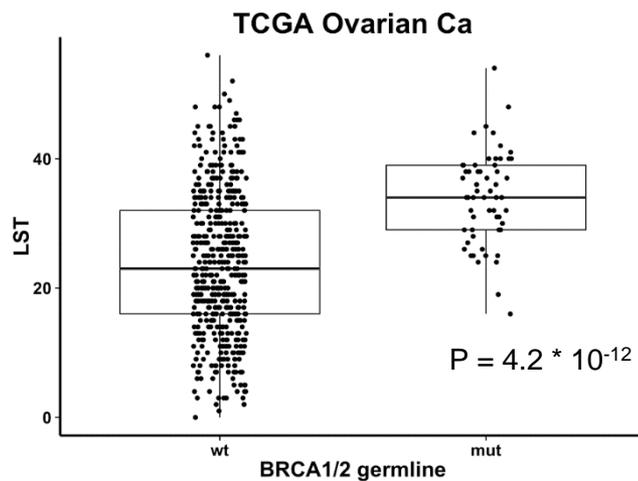
Changes in the Genomic Landscape



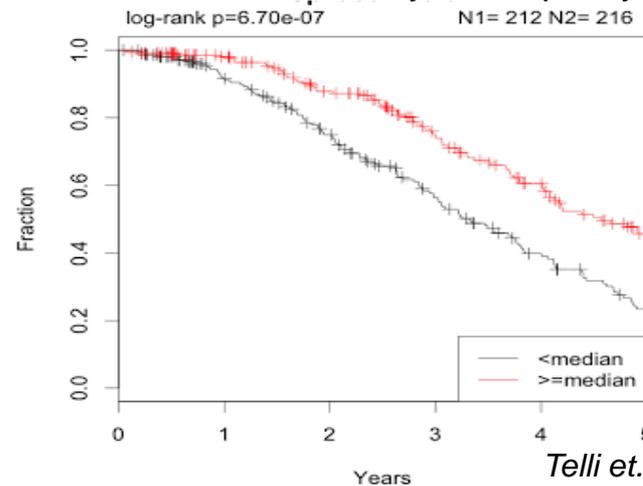
Clinical Utility of Structural Signatures



PrECOG 105: Neo-adjuvant Gem, Carbo, Iniparib.



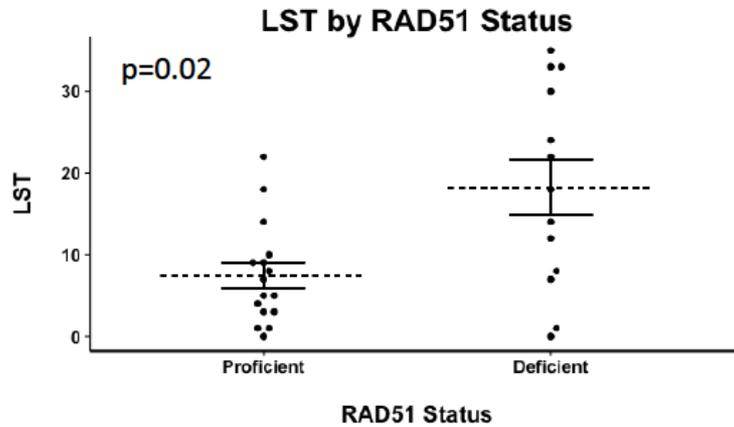
TCGA: Overall Survival by LST, split by Median



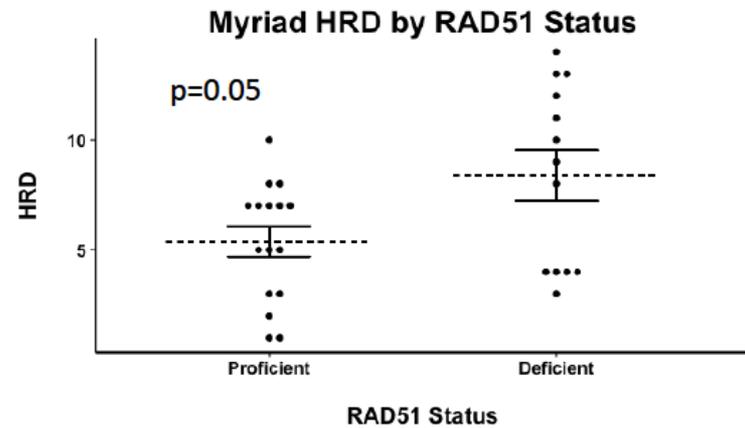
Telli et. al, JCO, 2015
Isakoff, et. al., JCO 2015

Changes in the Genomic Landscape and RAD51 Function

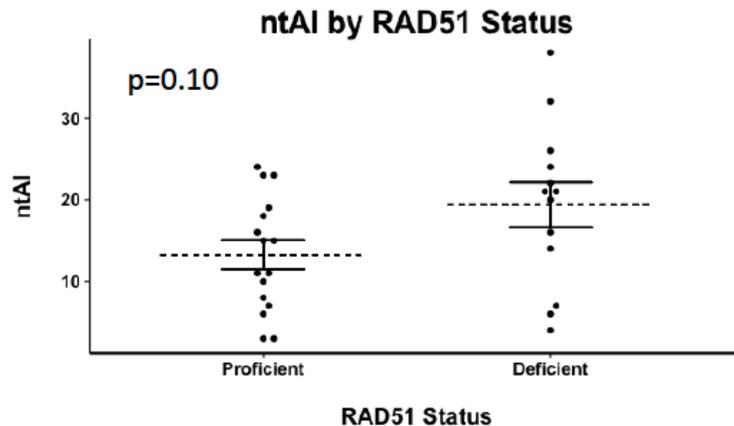
a.)



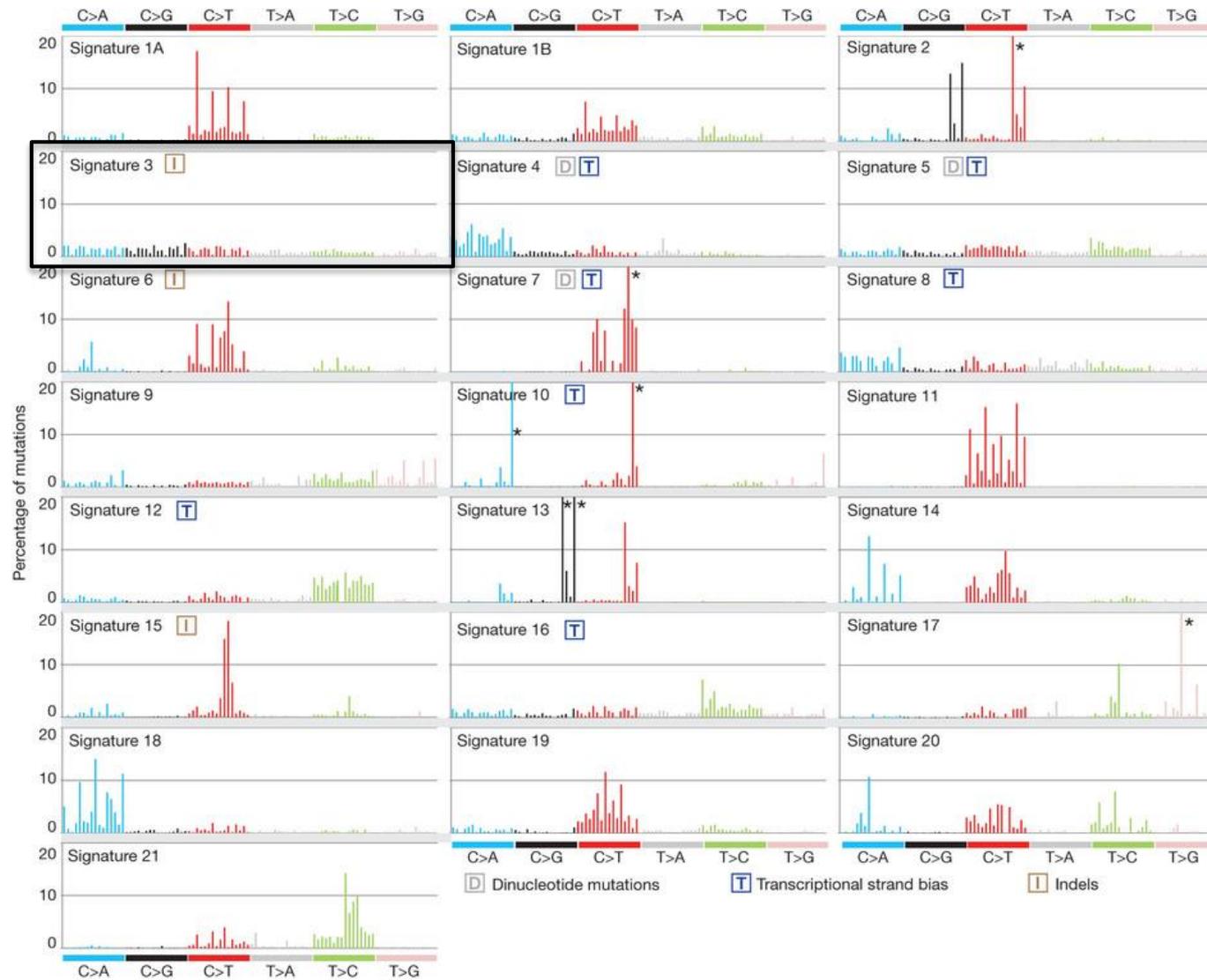
b.)



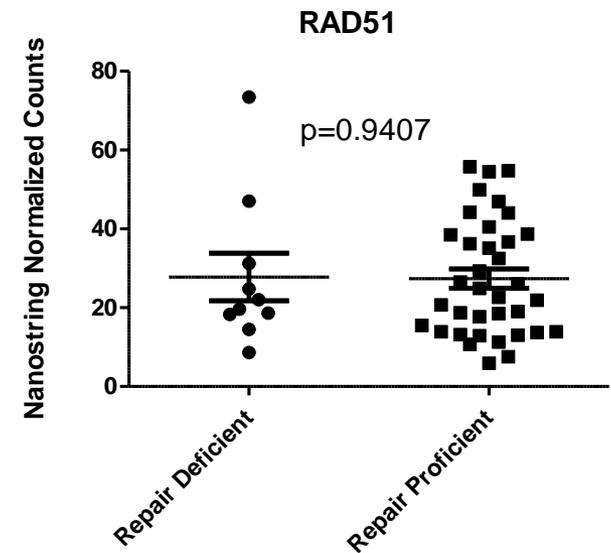
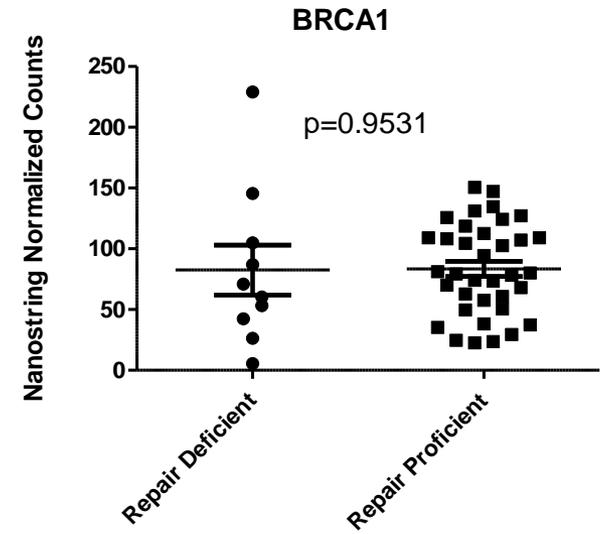
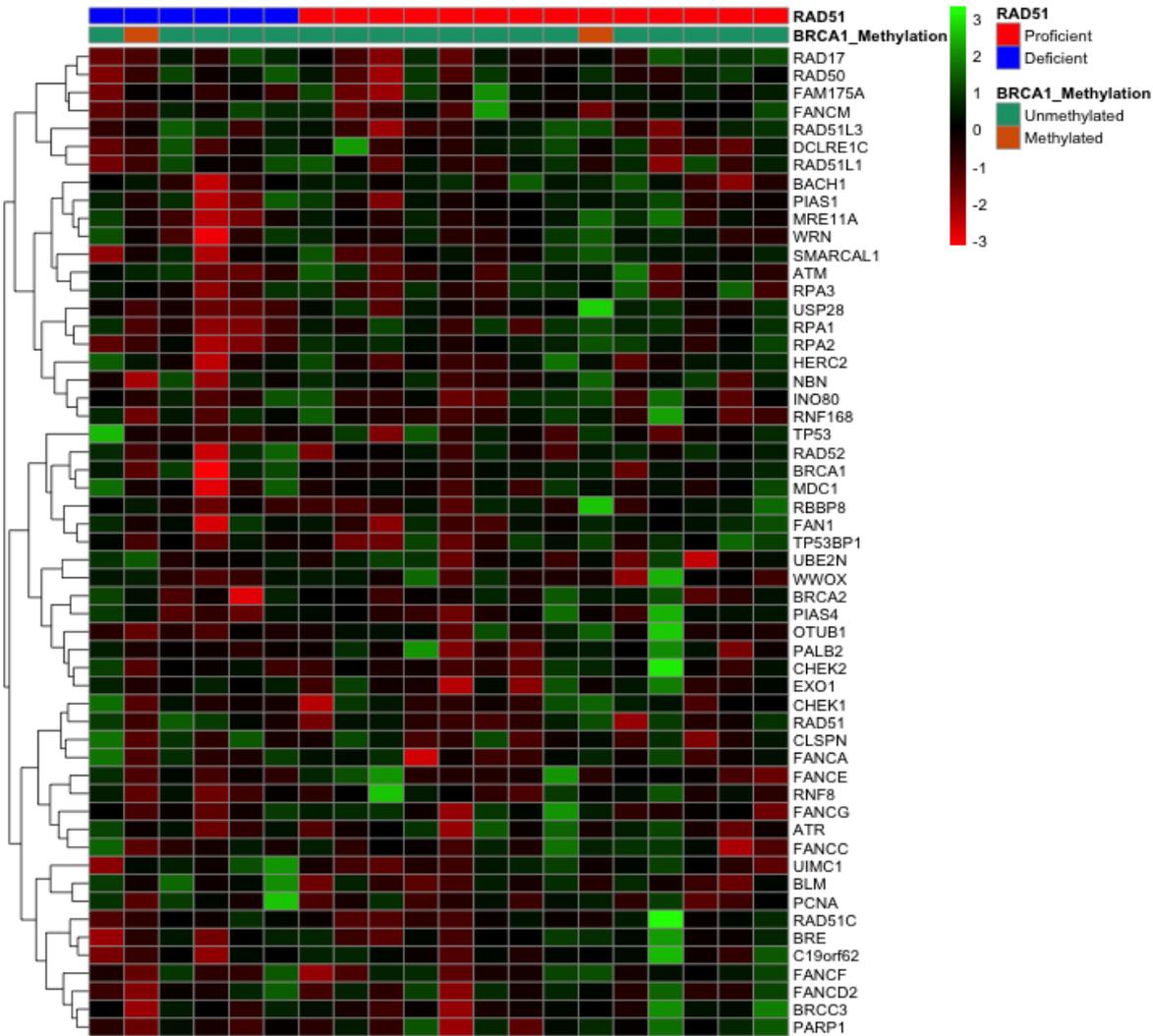
c.)



Mutational Signatures

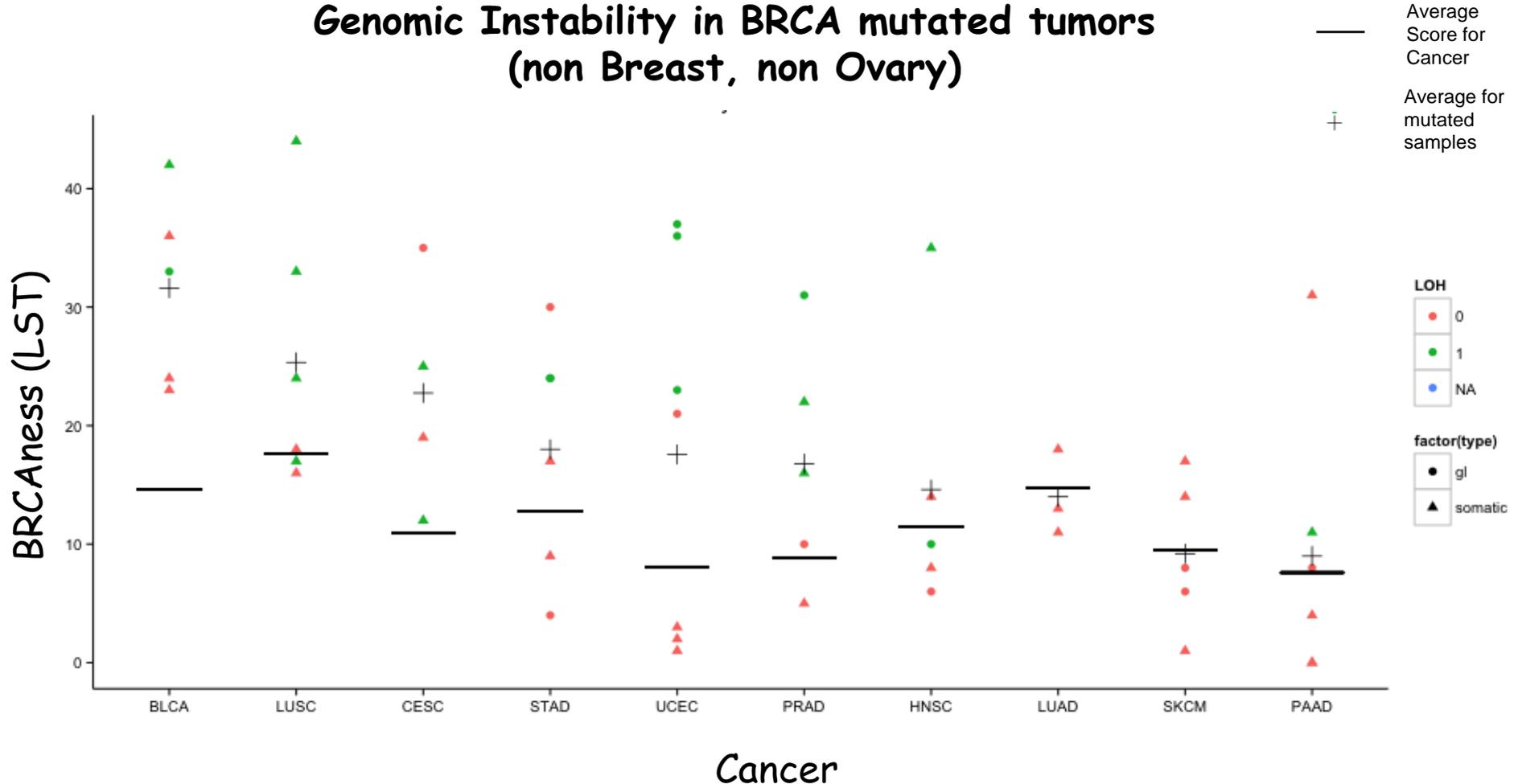


RAD51 Function and Gene Expression

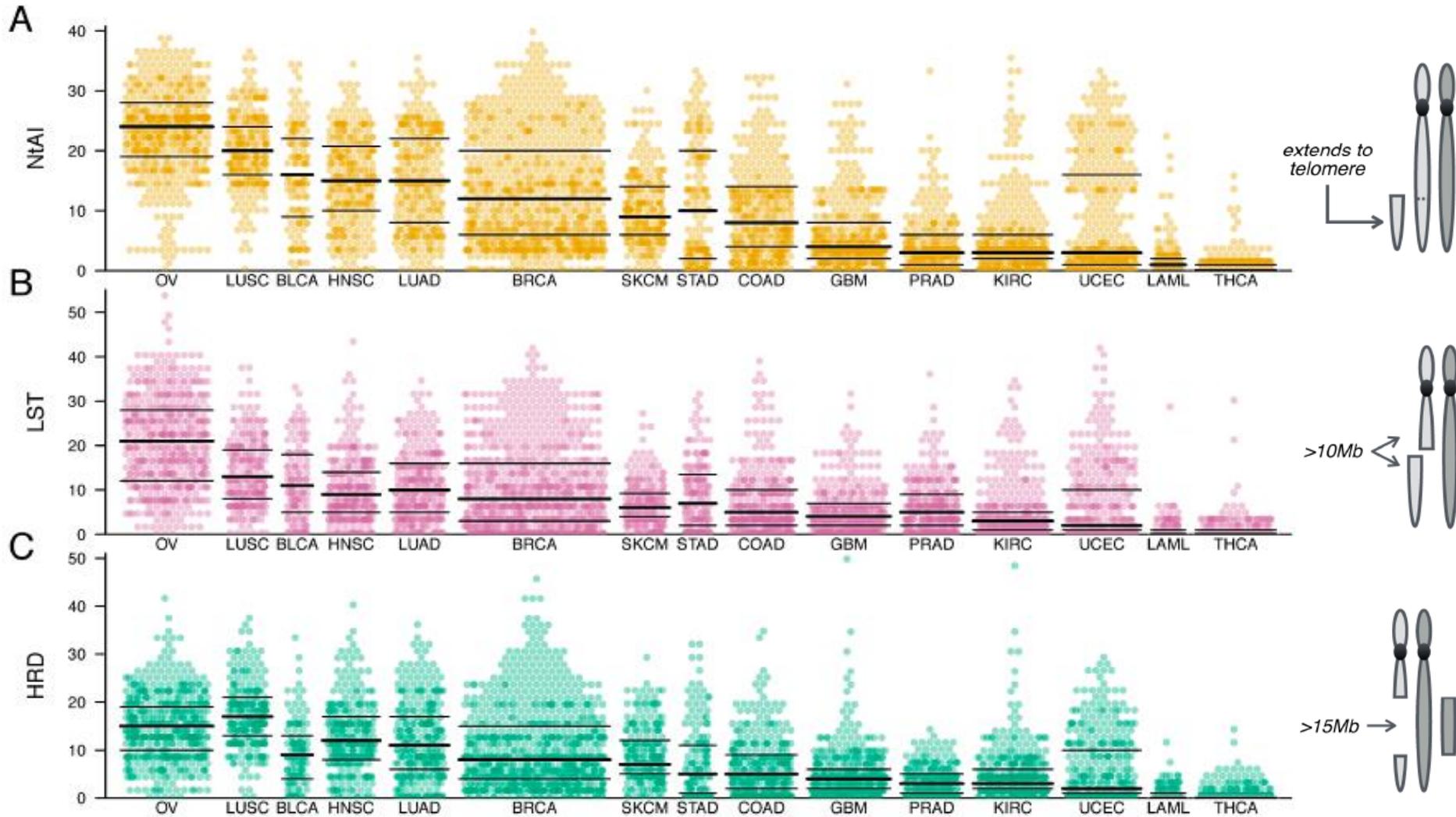


Mutations in BRCA1/BRCA2 in non-Breast & Ovarian Cancer in TCGA

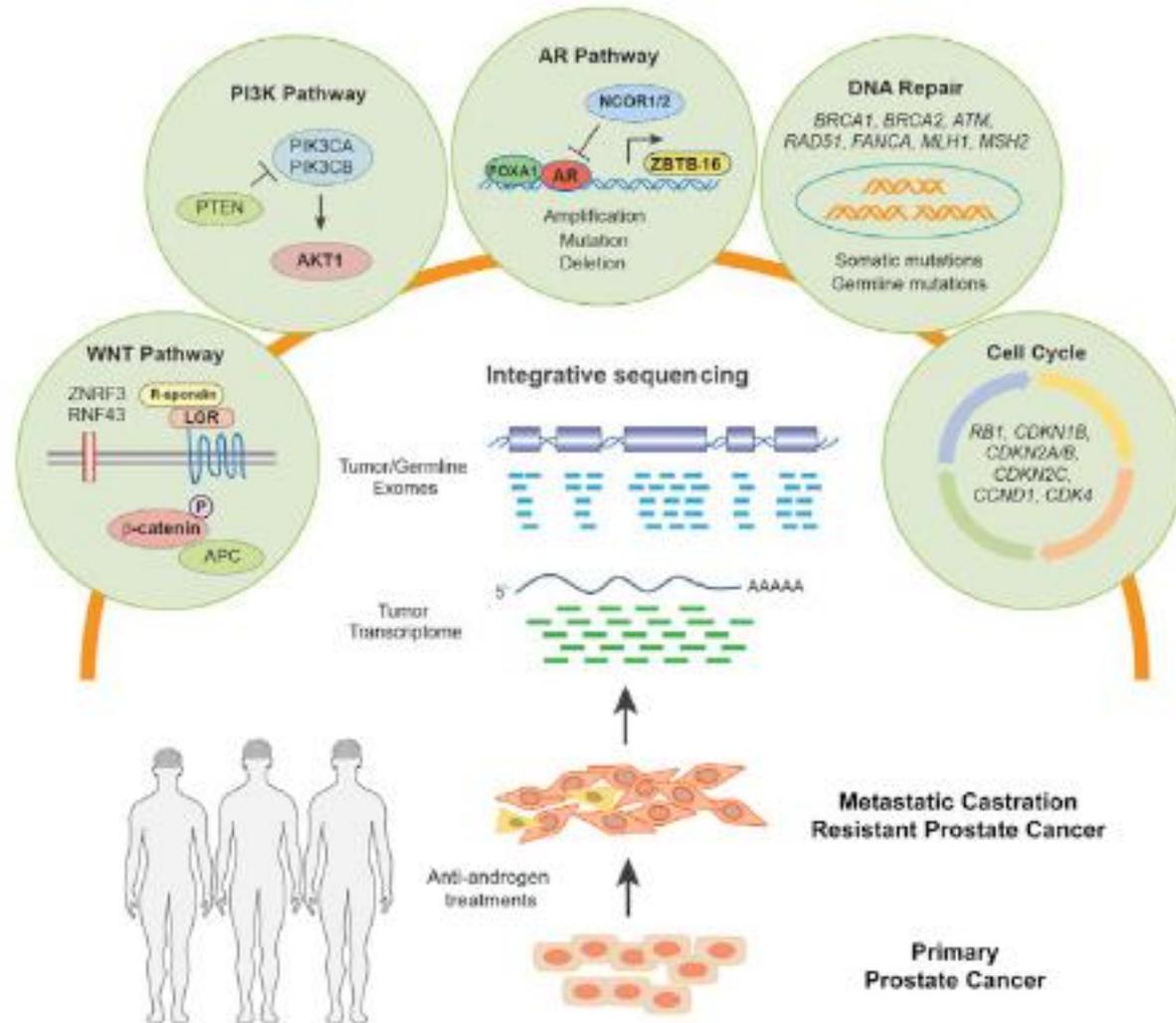
Genomic Instability in BRCA mutated tumors
(non Breast, non Ovary)



BRCA1-BRCA2 Pathway Defects across broad tumor types



Cancer Genomics



Sporadic BRCAness/HRD

- RAD51 function is a direct measure of the BRCA1-BRCA2 pathway
- Genome Landscape changes are now easy to measure with the availability of tumor DNA
- The BRCA1-BRCA2 pathway can be disrupted by inactivating mutations anywhere in the pathway
- All 3 measures correlate well, but for individual patient prediction of response to therapy, we need an assay that accurately determines response to “HR-directed therapies”
- There are lots of new potential opportunities for synthetic lethality to exploit this Achilles heel of HRD cancers

Cancer Genetics and DNA Repair

- DNA Repair abnormalities in cancer cells
- Mutational signatures observed in TCGA
- Methods to detect HR-Deficiency
 - Functional Assessment
 - Structural Variation
 - Mutational Signature
- HRD found outside of Breast/Ovarian Cancer
- Expanding the ability to deduce genomic landscapes from limited sequencing data
e.g. MSKCC IMPACT data

Defective HR: Therapeutic Strategies

Cisplatin, better with ionizing radiation?

Crosslinking agents: MMC +/- radiation

PARP inhibitors, but not combined with radiation?

Depletion of nucleotide pools, MTH1 inhibition?

Topoisomerase poisons: I vs II; combination with PARP inhibitors are toxic

Reactive Oxidizing Species: produce S-phase breaks

Back-up repair pathways: RAD52 inhibitors, TLS-pols inhibitors

Acknowledgments

Memorial Sloan-Kettering

- Chris Barker
- Giana Bernheim
- Ranjit Bindra
- Elizabeth Brunn
- Beverly Cheng
- Jarin Chun
- Ram Fridlich
- Alex Goglia
- Tiffany Lai
- Ciara O'Driscoll
- Robert Delsite
- Devi Annamalai
- Alison Carley
- Christian Cleary
- Laura Eccles
- Dan Higginson
- Benjamin Lok
- Robert Mutter
- Jeffrey Park
- Rohini Roy
- Samantha Sheppard
- Adriana Vela

Collaborators:

Jorge Reis-Filho
Charlotte Ng

David Solit
Barry Taylor

Support of: NIH - NCI; BCRF; Beene Foundation; Gray Family; MSKCC