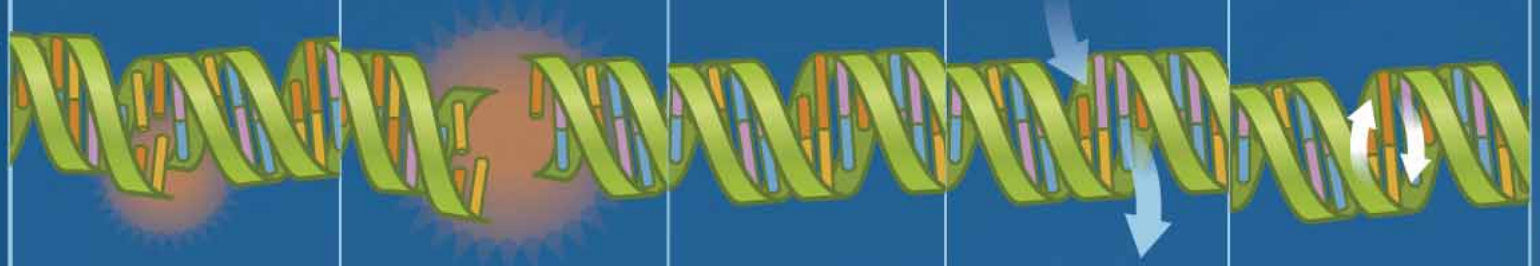
"BRCAness," PARP and the Triple-Negative Phenotype

Prof Alan Ashworth, FRS

Disclosures for Professor Alan Ashworth, FRS

Consulting Agreements	GlaxoSmithKline, Pfizer Inc
Patent	AstraZeneca Pharmaceuticals LP

Types of DNA Damage and Repair

	Single-strand breaks (SSBs)	Double-strand breaks (DSBs)	Bulky adducts	Insertions deletions	06-alkylguanine
Type of damage					
Repair pathway	Base excision repair	Recombinational repair	Nucleotide excision repair	Mismatch repair	Direct reversal
Repair enzymes	PARP	HR → ATM; NHEJ → DNA-PK	XP, polymerases	MSH2 MLH1	AGT

Rationale for Targeting DNA Repair Defects in Tumours

- Germ-line defects in DNA repair components lead to cancer predisposition (eg, BRCA, mismatch repair, etc)
- Many (most?) adult sporadic cancers show evidence of genomic/genetic instability
- Defects in different DNA repair pathways confer sensitivity to specific DNA damaging agents

Tumour Cells in BRCA1 or BRCA2 Mutation Carriers Have Lost Normal BRCA Function

Normal Tissues

1 mutant copy, 1 intact copy of BRCA gene

Tumour

1 mutant copy of BRCA gene

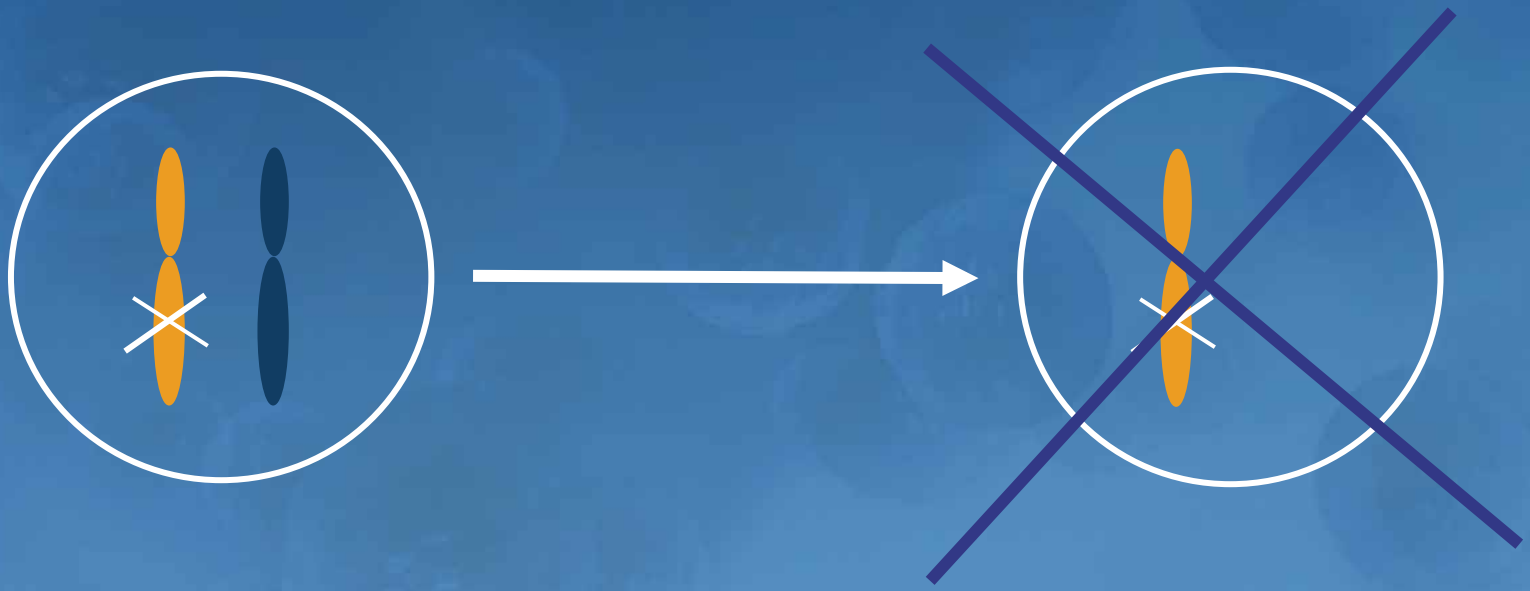
How Can BRCA1 or BRCA2 Mutant Cells Be Selectively Killed While Not Affecting Normal Cells in Mutation Carriers?

Normal Tissues

1 mutant copy, 1 intact copy of BRCA gene

Tumour

1 mutant copy of BRCA gene



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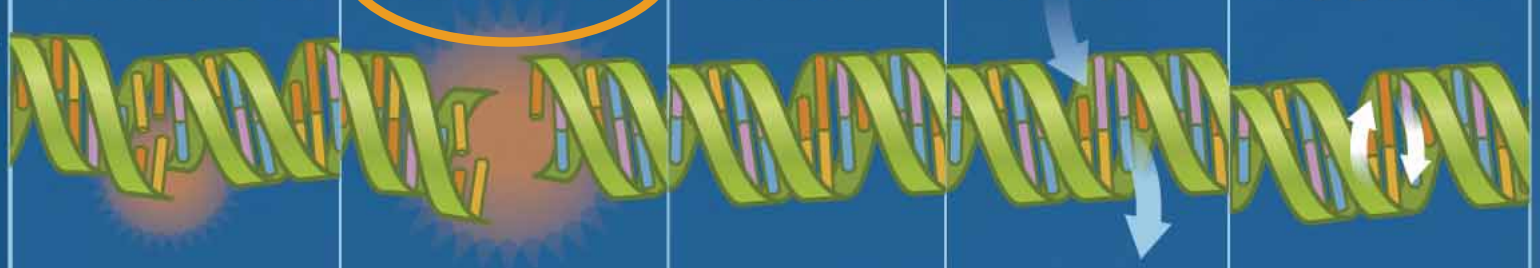
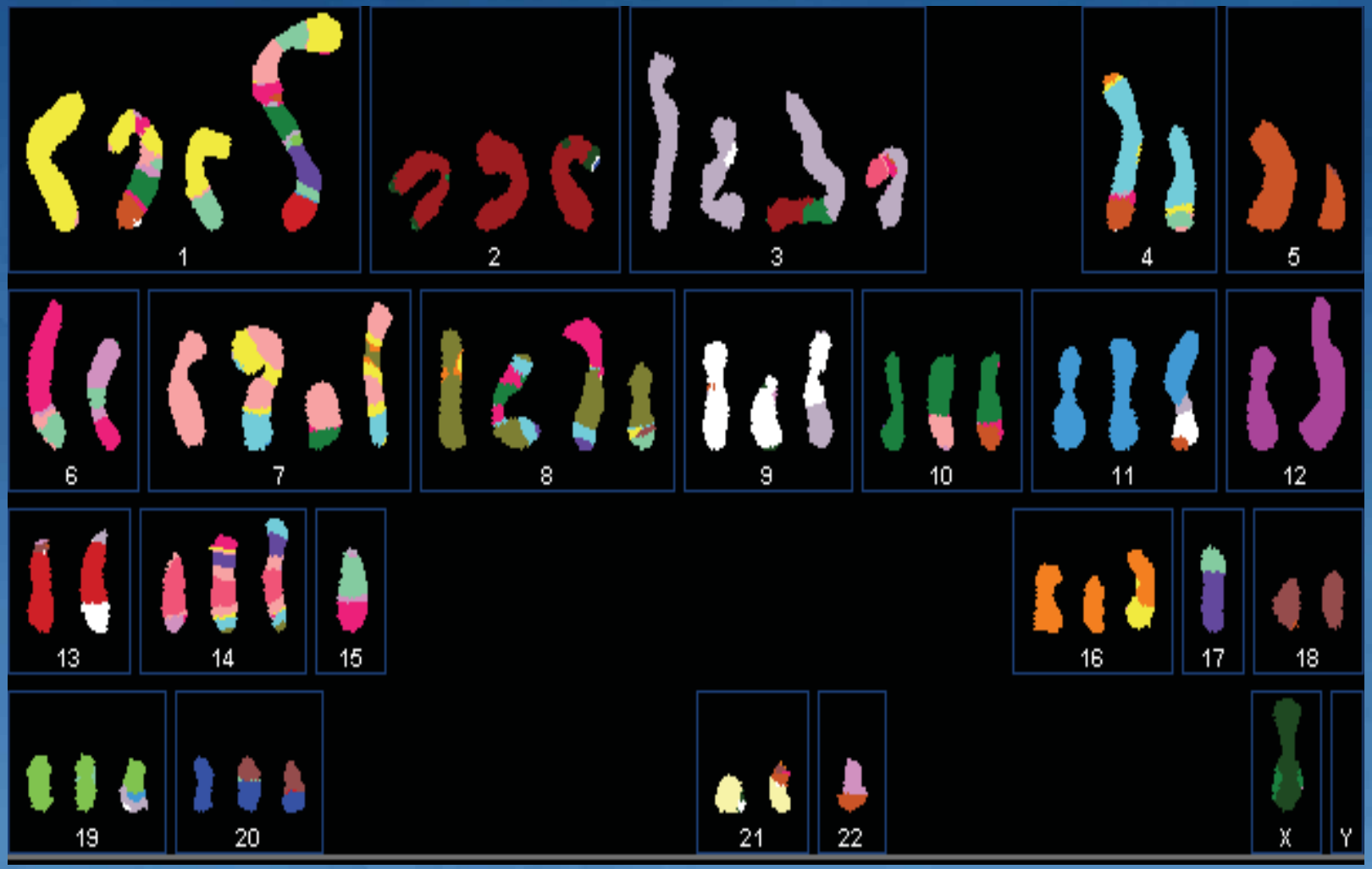
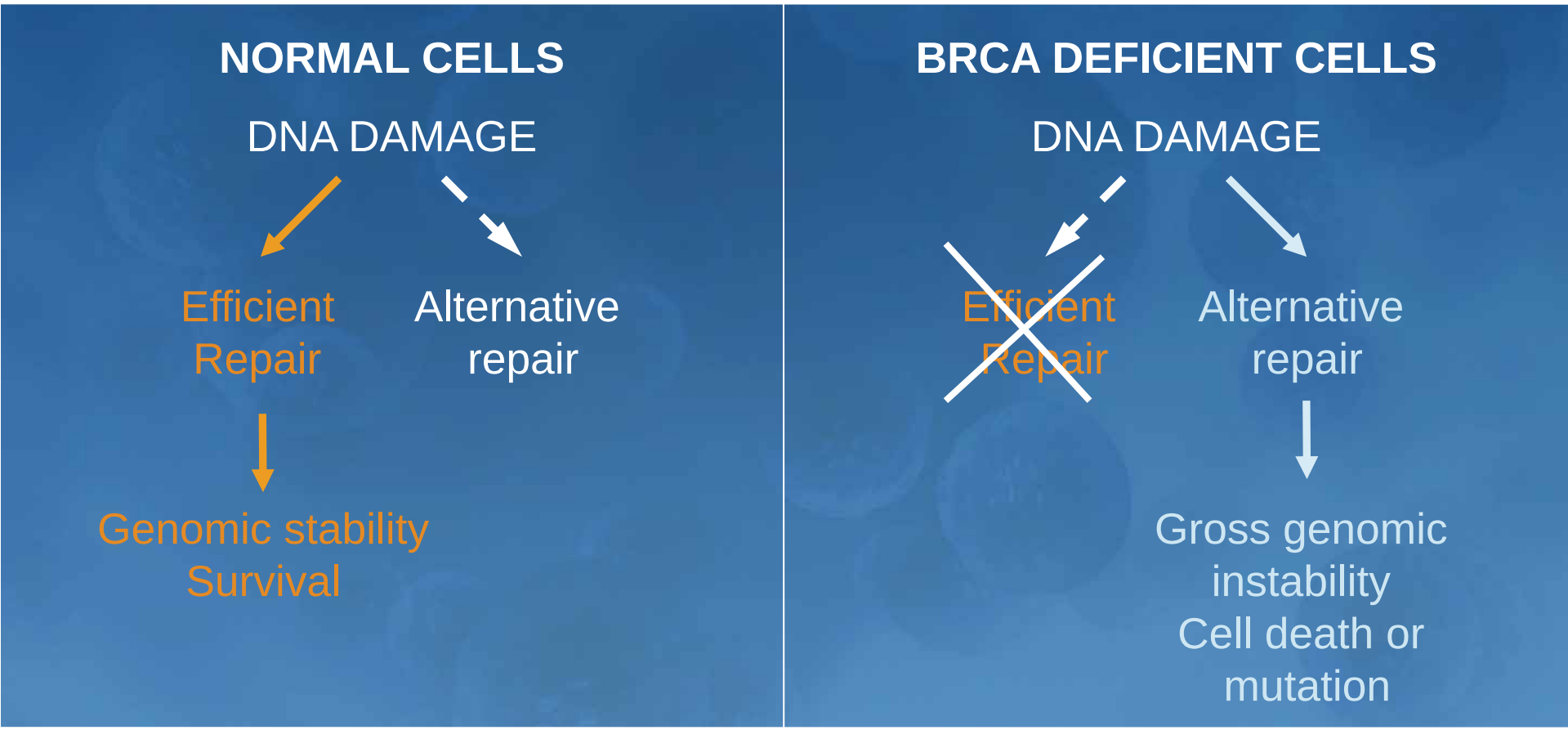


BRCA2 Tumour Cell Line CAPAN-1

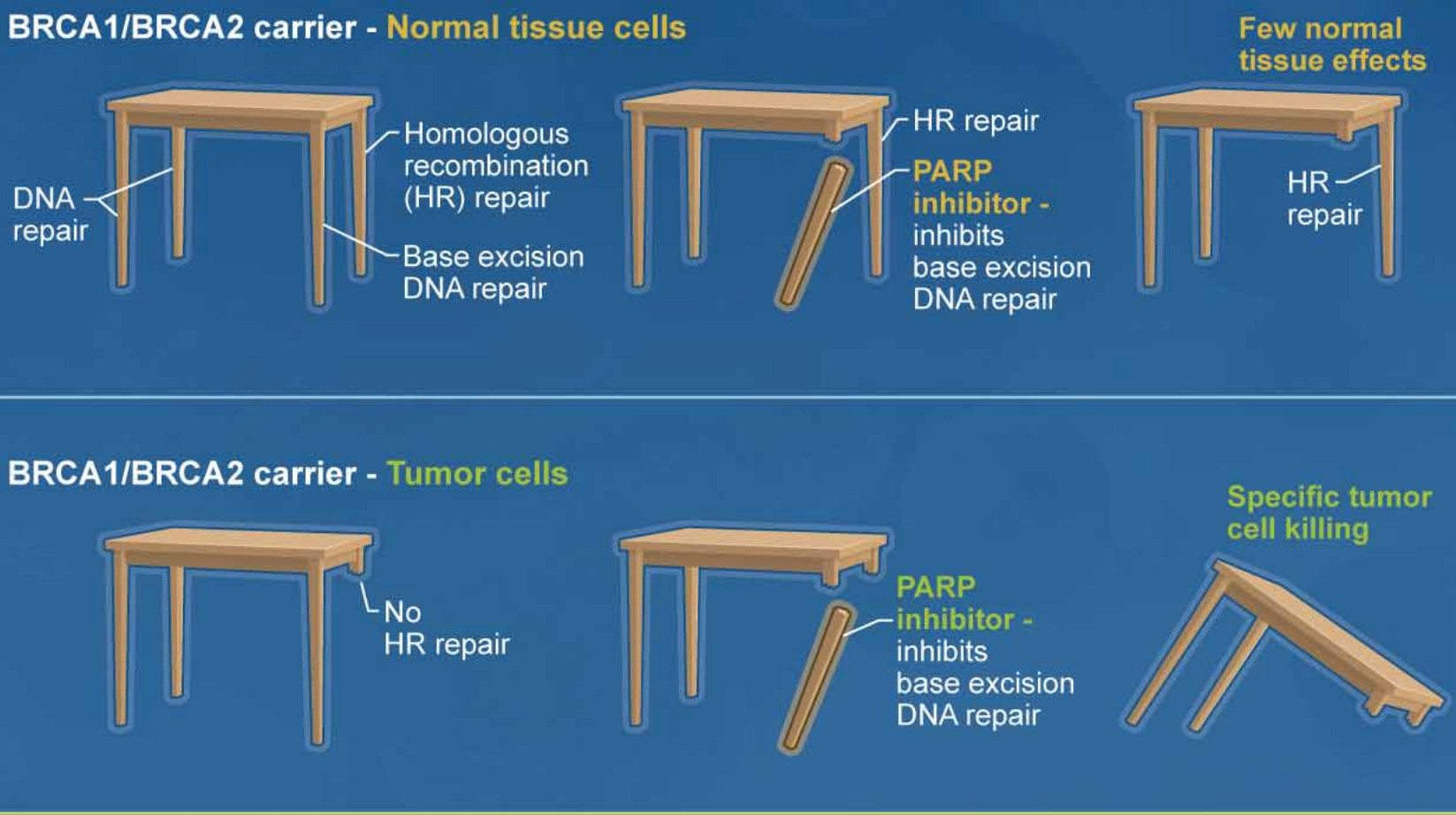


Courtesy Paul Edwards, Univ of Cambridge

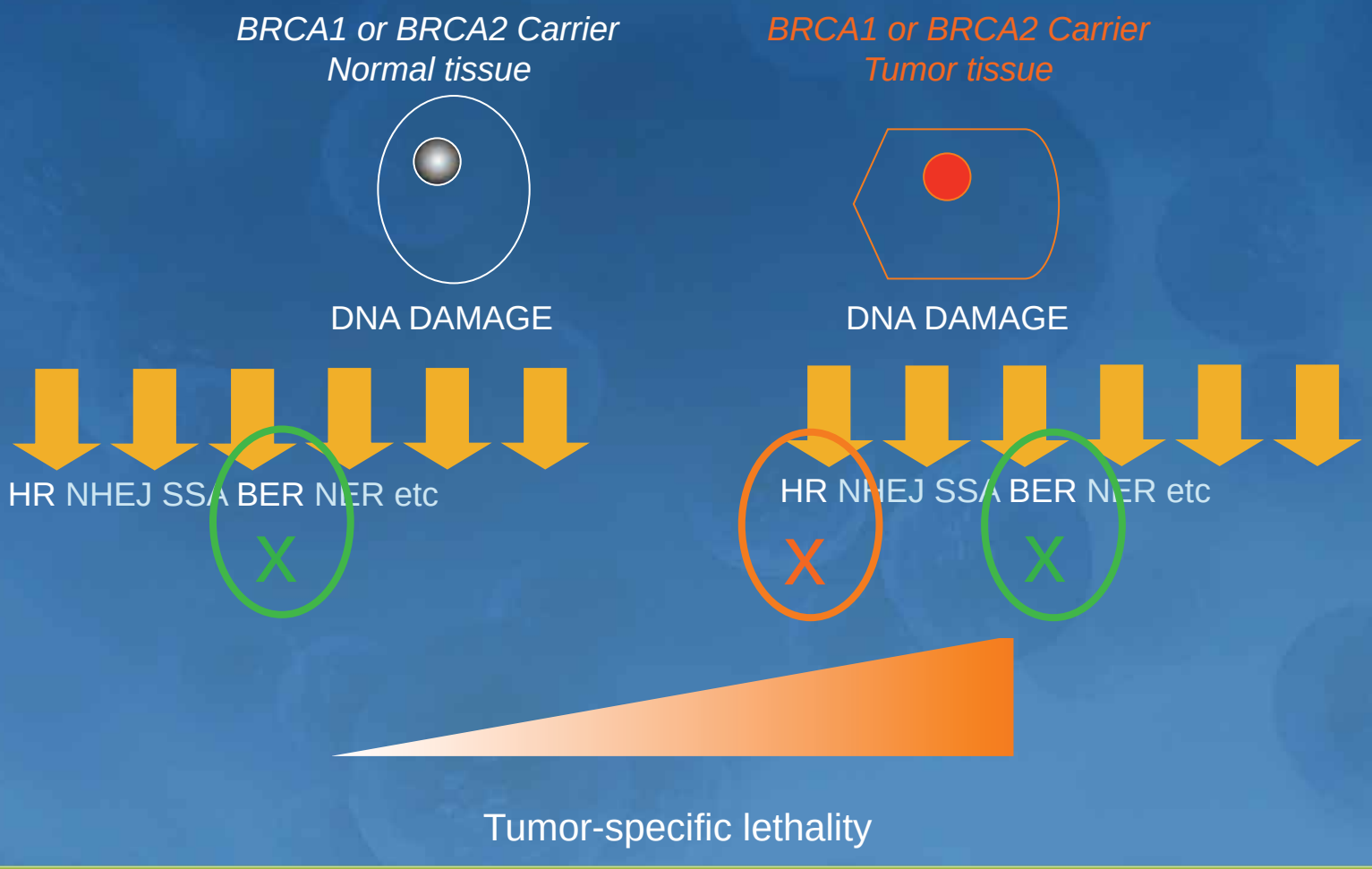
The Use of Alternative Pathways Underlies the DNA Repair Defect in BRCA Deficient Cells



Synthetic Lethality — The Principle

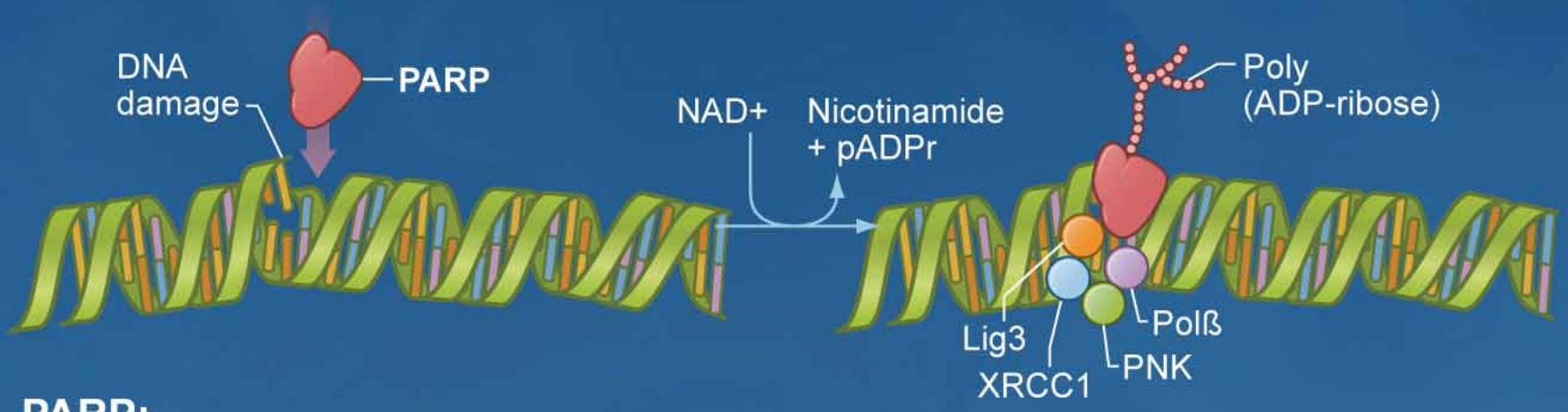


Synthetic Lethality in DNA Repair Pathways



Tutt A et al. *Cold Spring Harb Symp Quant Biol* 2005;70:139-48;
Ashworth A. *J Clin Oncol* 2008;26(22):3785-90.

Poly(ADP-ribose) Polymerase (PARP)

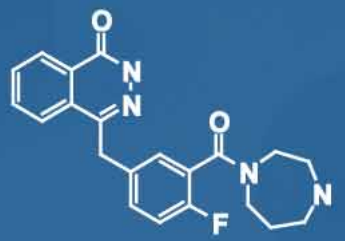


PARP:

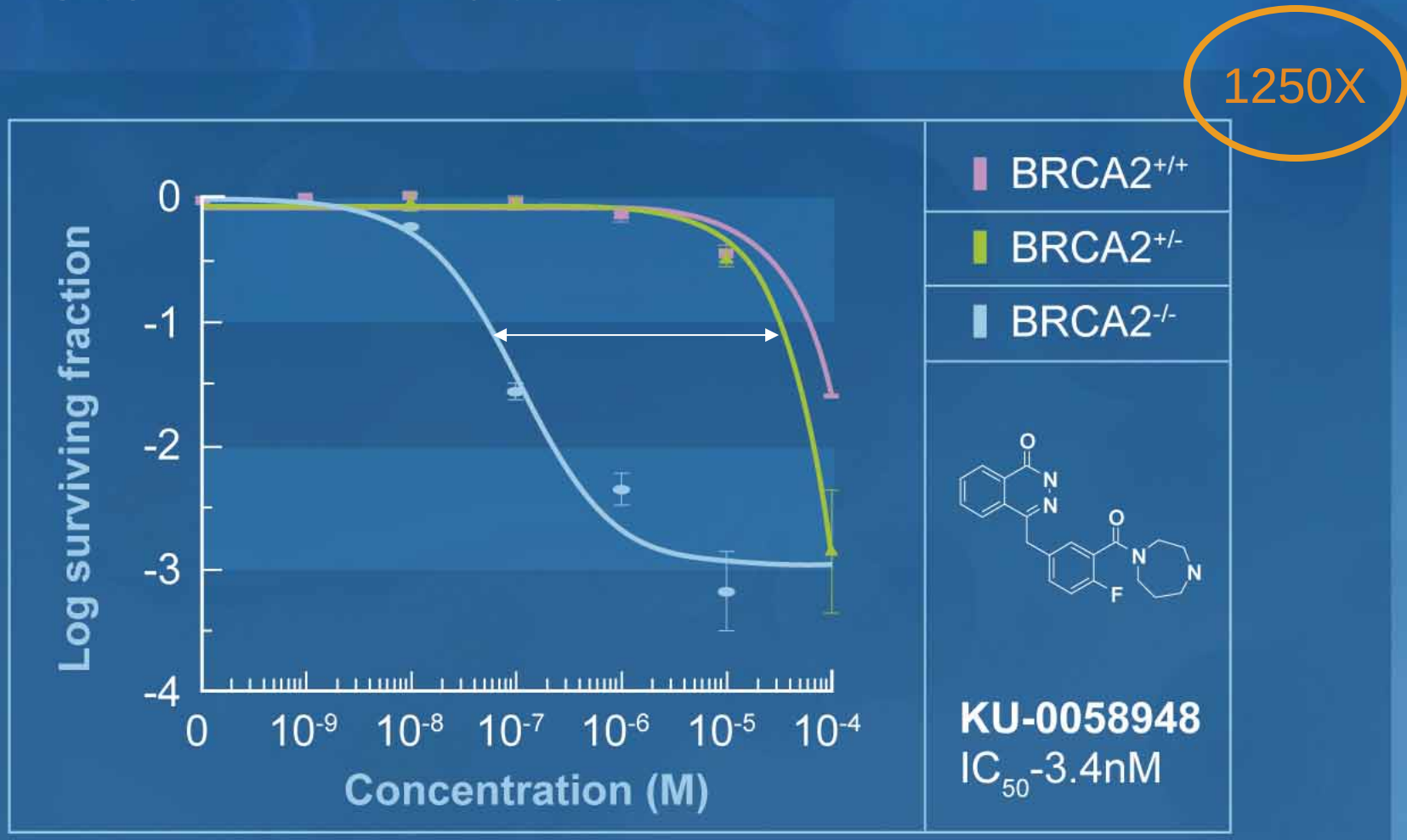
- Involved in DNA base-excision repair
- Binds directly to DNA damage
- Produces large branched chains of poly(ADP-ribose)

PARP Inhibitors

KU-0058948
IC_{50}-3.4nM

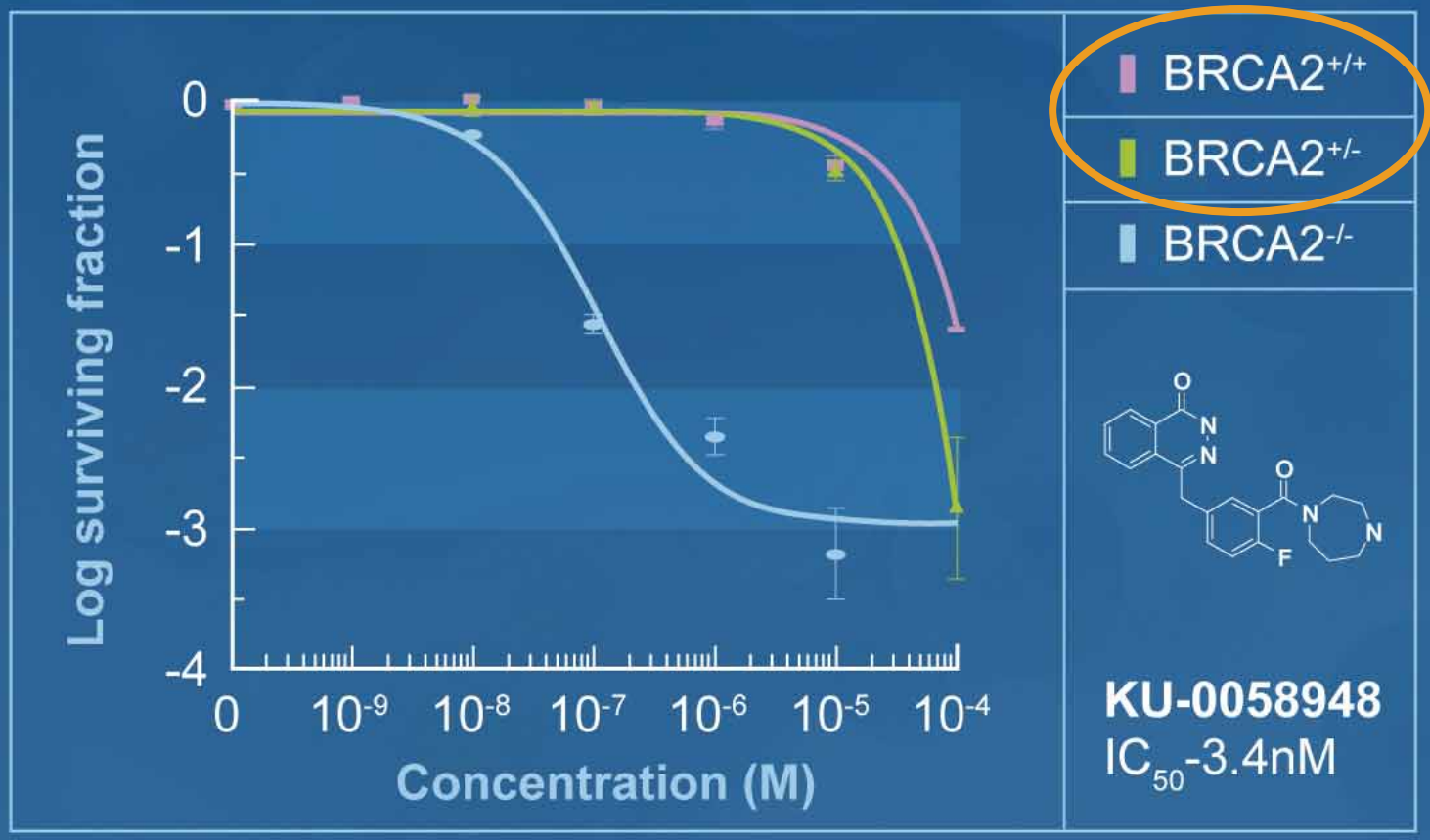


Extreme Sensitivity of BRCA2-Deficient Cells to PARP Inhibition



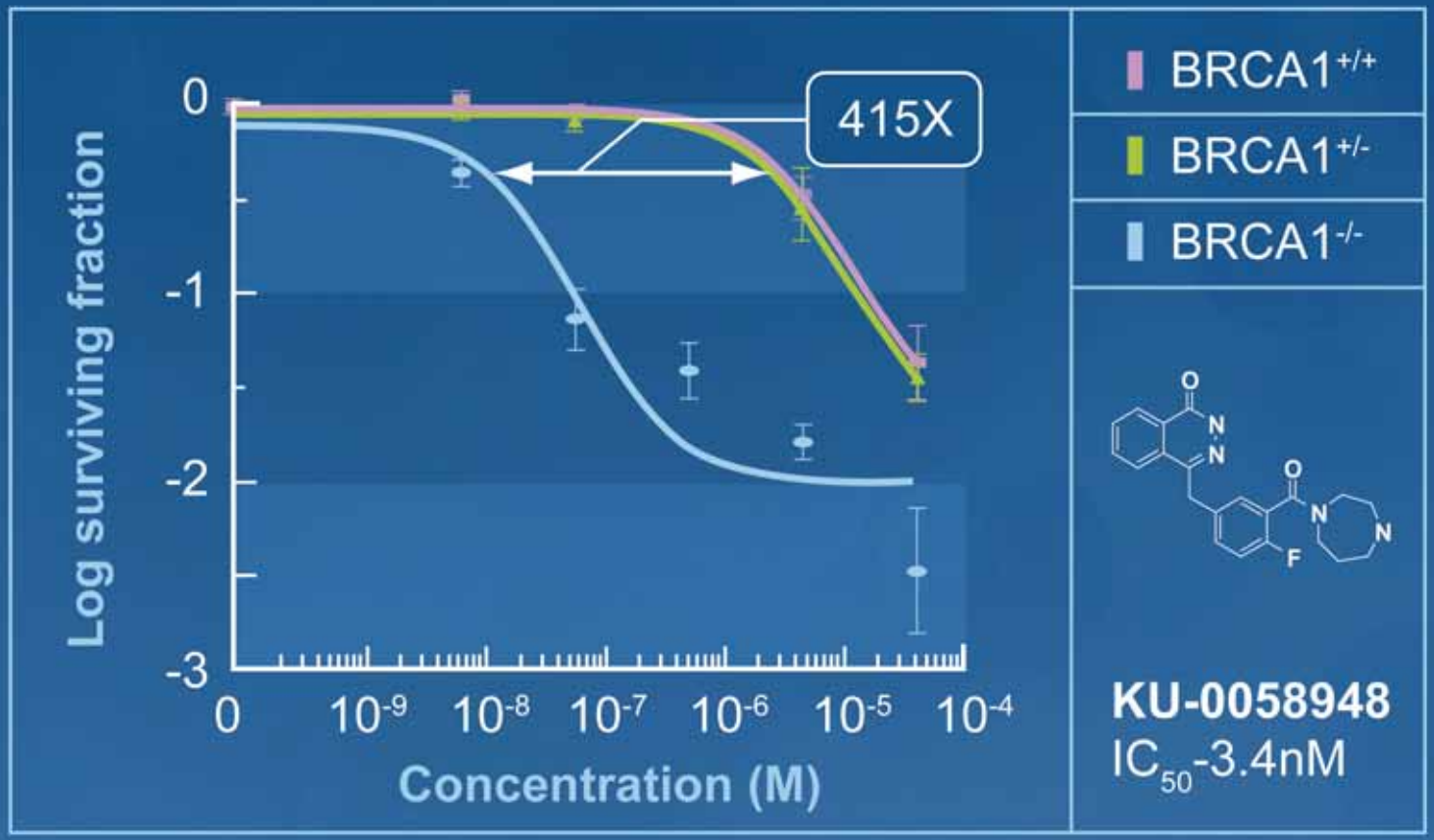
Farmer H et al. *Nature* 2005;434(7035):917-21.

Loss of One Copy of the *BRCA2* Gene Does NOT Cause Sensitivity to PARP Inhibitors



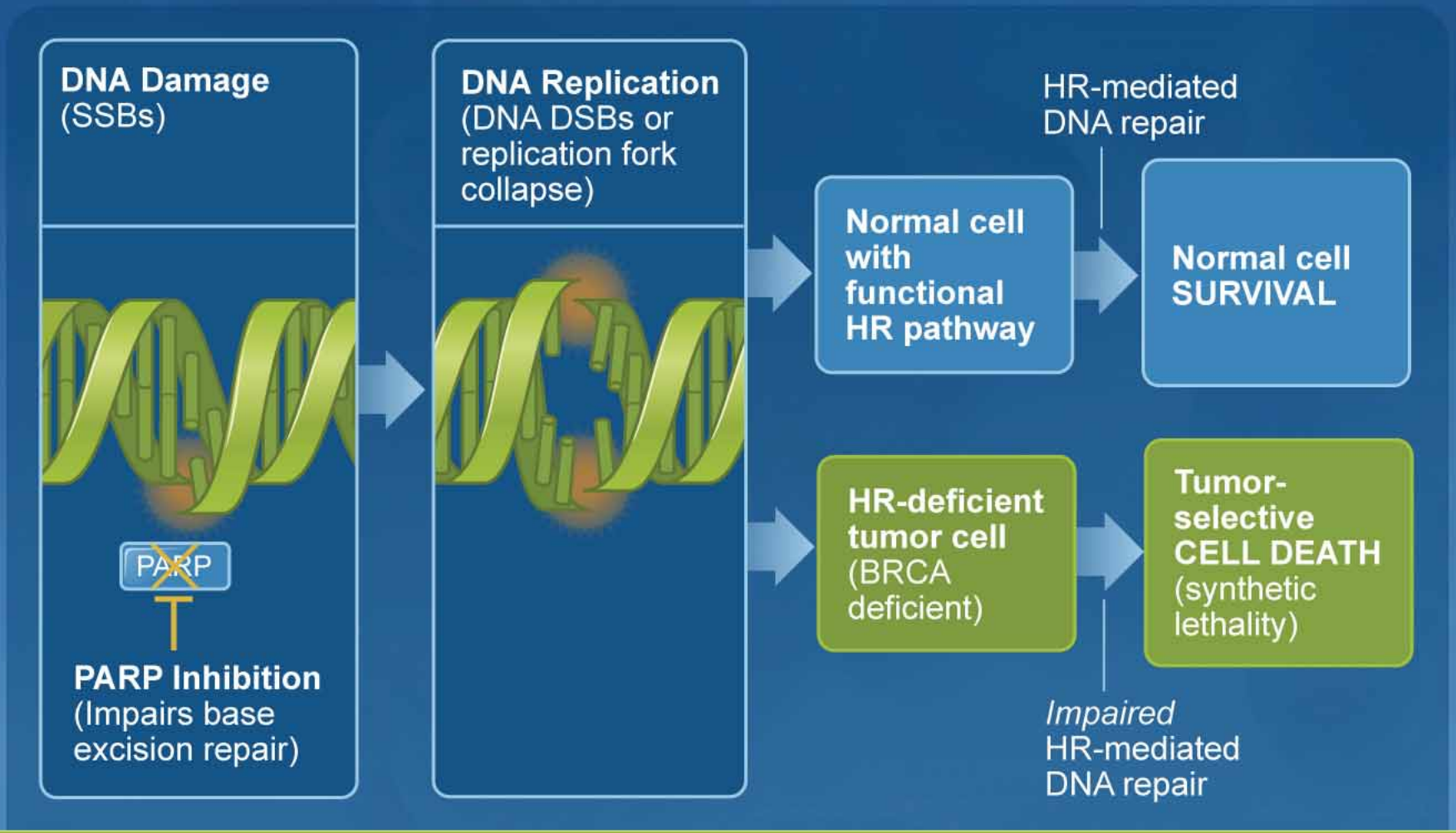
Farmer H et al. *Nature* 2005;434(7035):917-21.

BRCA1-Deficient Cells Are Also Extremely Sensitive to PARP Inhibition



Farmer H et al. *Nature* 2005;434(7035):917-21.

Synthetic Lethality between PARP Inhibition and BRCA1/2 Mutation

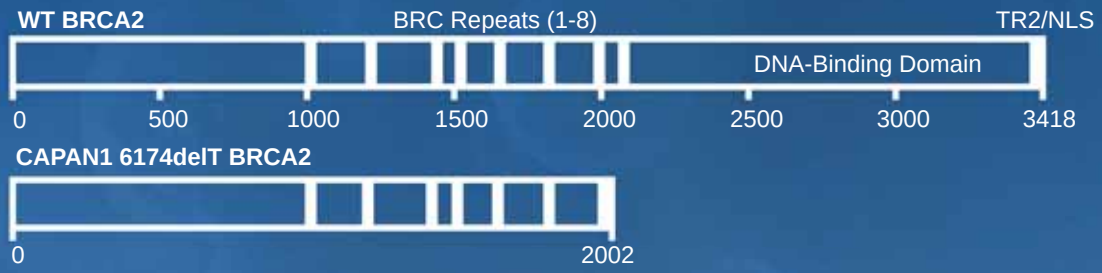


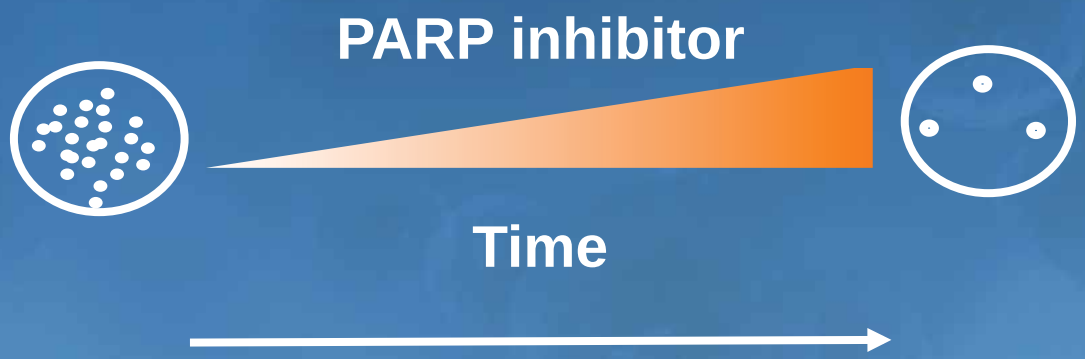
Synthetic Lethal Resistance

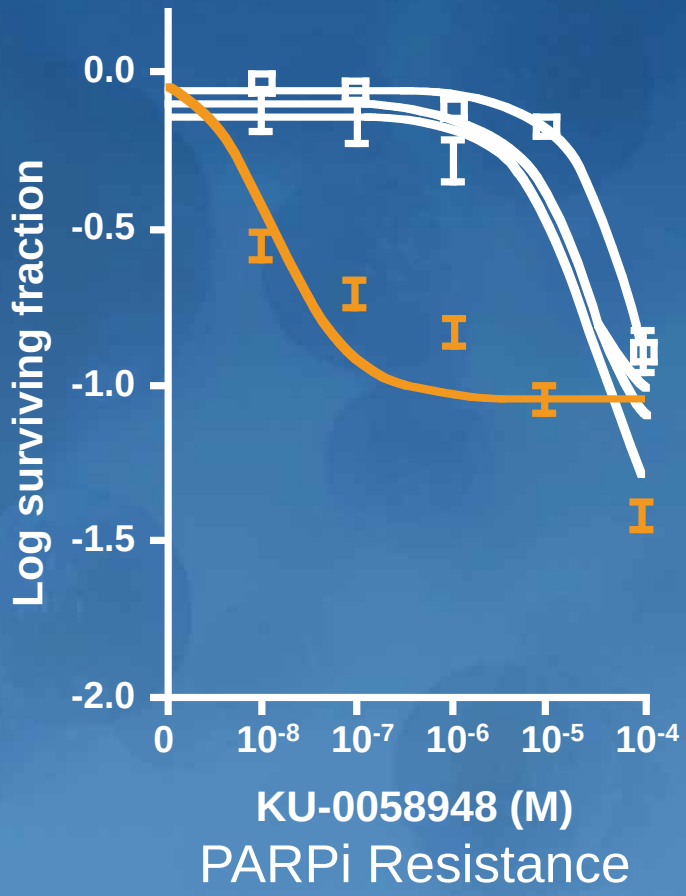
- Resistance arises to many targeted therapies
- Frequently due to mutation of “target” (eg, imatinib/cAbl)
- How does resistance to a synthetic lethality arise?

Edwards S et al. *Nature* 2008;451(28):1111-6.

PARPi Resistant CAPAN1 (BRCA2 c.6174delT) Cells







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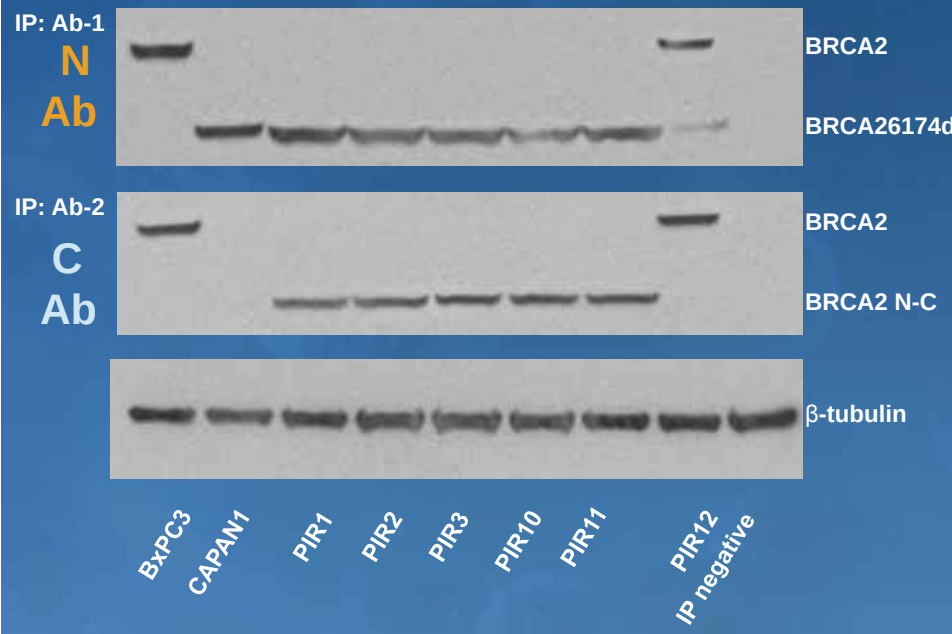
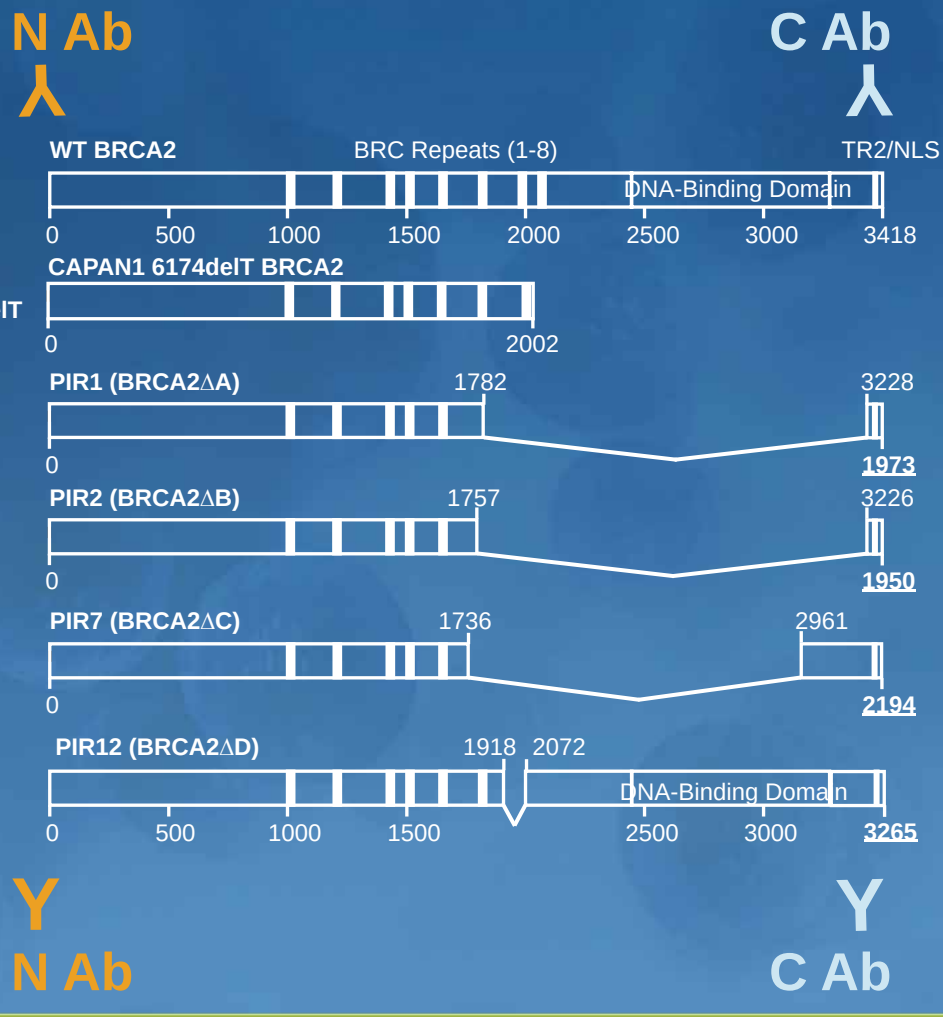
Restoration of BRCA2 Open Reading Frame in PARPi Resistant Cell Lines





Edwards S et al. *Nature* 2008;451(28):1111-6.

Lessons for Use of PARP Inhibitors

- Likely clinically relevant as similar phenomenon observed in ovarian cancer after platinum resistance
- As with other targeted therapies — mechanism based resistance can occur but **SYNTHETIC LETHAL RESISTANCE** in this case (does not preclude other mechanisms)
- Late-stage disease, resistance likely due to large target pop for resistance
- Best results likely to be achieved in early/adjuvant treatment

Extending the Approach to Sporadic Cancer

- BRCAness — Molecular features of BRCA1 or BRCA2 mutant tumours in sporadic cancers
- Suggests that therapies directed against BRCA defects might be effective in a sporadic group of tumours
- Example of BRCAness may be triple negative (ER, PR and HER2-)/basal-like and BRCA1 tumours

Turner N et al. *Nat Rev Cancer* 2004;4(10):814-9.

Similarity between BRCA1 Mutant and Basal/Triple Negative Tumours

	Basal-like and TN	BRCA1
High grade	√	√
Pushing borders	√	√
Brisk lymphocytic infiltrate	√	√
High proliferation rates	√	√
ER-	√	√
PR-	√	√
HER2-	√	√
TP53 mutations	√	√

Turner NC et al. *Oncogene* 2006;25:5846-53.

Basal-Like and TN Breast Cancers

- Account for 12-17% of all breast cancers
- More prevalent in
  - Younger women (<50 years)
  - African and Hispanic descent
  - *BRCA1* mutation carriers
- More frequently interval cancers

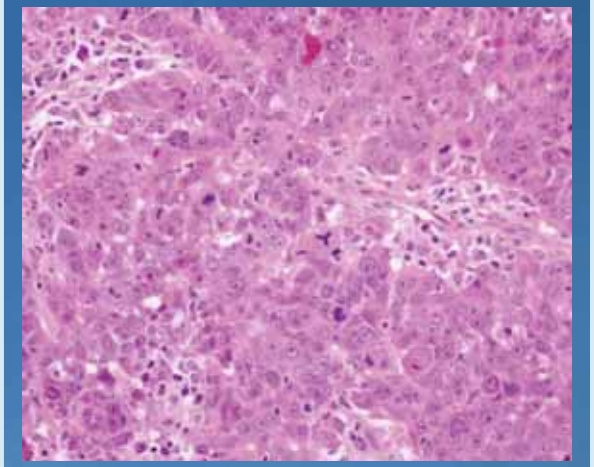
BRCA1 Downregulation

- High histological grade
- Medullary histological type
- Basal-like and TN immunophenotype
- ***BRCA1* somatic mutations are exceedingly rare**

TN and Basal-Like Carcinomas

BRCA1 downregulation

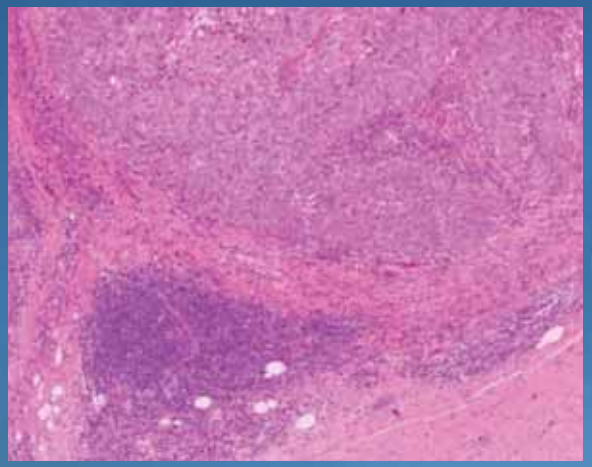
IDC
TN/Basal-like



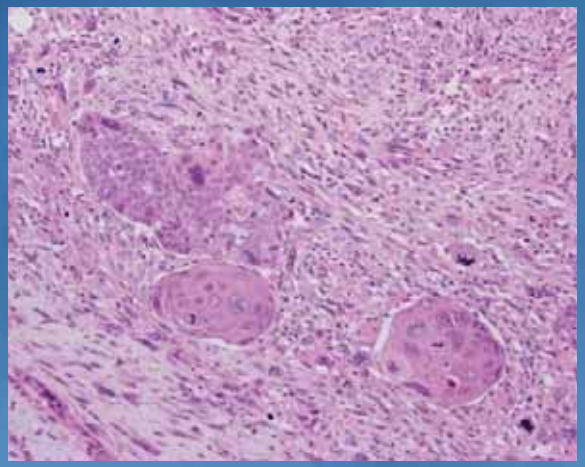
ID4 overexpression

***BRCA1* methylation**

Medullary



Metaplastic



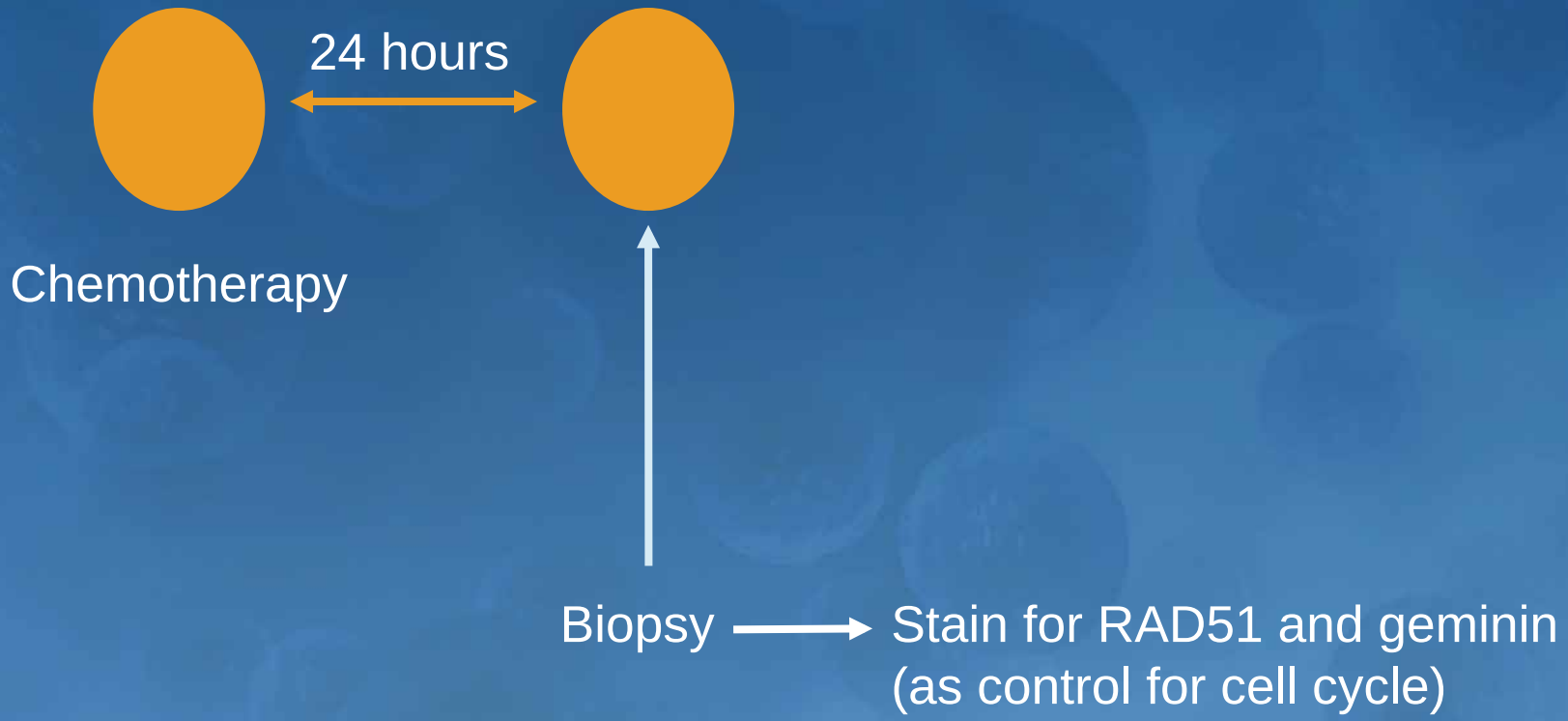
Turner NC et al. *Oncogene* 2007;26(14):2126-32; Figure adapted from Rakha EA et al. *J Clin Oncol* 2008;26(15):2568-81.

Patient Selection by Assaying DNA Repair Capacity

HR Biomarkers
RAD51 Foci Biomarker

- Lack of RAD51 focus formation after DNA damage is a robust marker of HR deficiency in cell lines
- Problematic in tumours as need to measure post damage — archival specimens can't be used

Measuring Induction of RAD51 in Breast Tumours in Response to Chemotherapy



RAD51 Scores for Tumours Treated with Neoadjuvant Chemotherapy

- Tumors that achieved a pathological complete response (pCR) with neoadjuvant chemotherapy had lower RAD51 scores.
- Of the tumors with low RAD51 score, 33% achieved pCR compared to 3% of tumors with RAD51 foci formation.
- Low RAD51 score was associated with high histological grade, ER-negative tumors and triple-negative cancers.

Graeser M et al. *Clin Cancer Res* 2010;16(24):6159-68.

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