TAT 2001
Targeted Anticancer Therapies

Synthetic Lethality in Cancer Treatment: Current Role of PARP Inhibitors

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UK

Funding

CANCER RESEARCH UK

Hilary Calvert
Disclosures

- Inventors reward scheme, AG014699 / PF-01367338
- Consultancy
  - Agouron / Pfizer
  - AstraZeneca
  - BiPar
  - Eli Lilly
  - Kudos
  - LEAD
- All PARP inhibitors are investigational at the present time

Problems with targeted anticancer agents

- The target may be present and have a function in normal tissues
  - Side effects limit the dose and therefore the anticancer efficacy of the drug
- Inhibiting the target may have little effect on the tumour
  - Alternative proliferative pathways

Ideal targeted agents

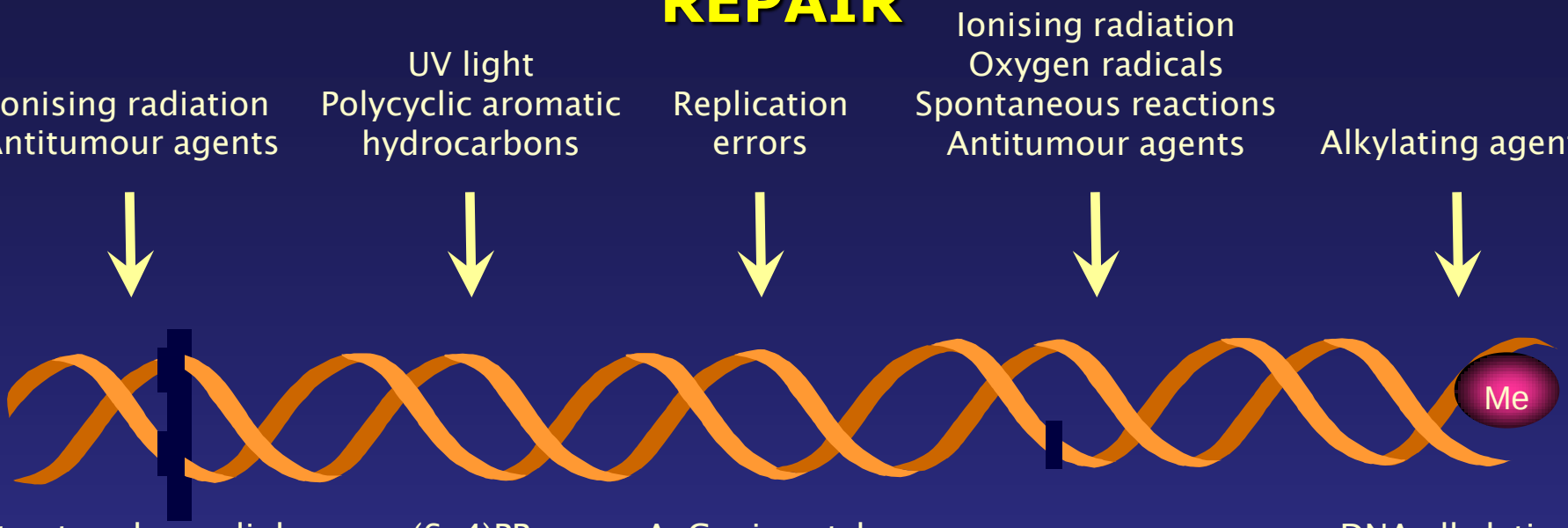
- Target a fusion protein unique to the tumour cell and responsible for its malignant transformation
  - Imatinib, CML
  - ALK inhbitors, lung cancer
- Target a mutation unique to the tumour
  - BRAF, melanoma
- Target a gene that is uniquely amplified in the tumour
  - Her2, trastuzumab
- Synthetic lethality

DNA Repair – a process essential to cell survival

How long is a piece of DNA?	
DNA length per cell	2 meters
Cells per human	2×10^{13}
DNA length per human	4×10^{13} meters
Distance from the Earth to the Sun	1.49×10^{11} meters
Number of return trips to the Sun	134

- Each cell sustains 10,000 to 30,000 episodes of DNA damage per day
- 5 Basic types of DNA damage – repair pathways
- Redundancy
  - Different pathways
  - 2 Alleles

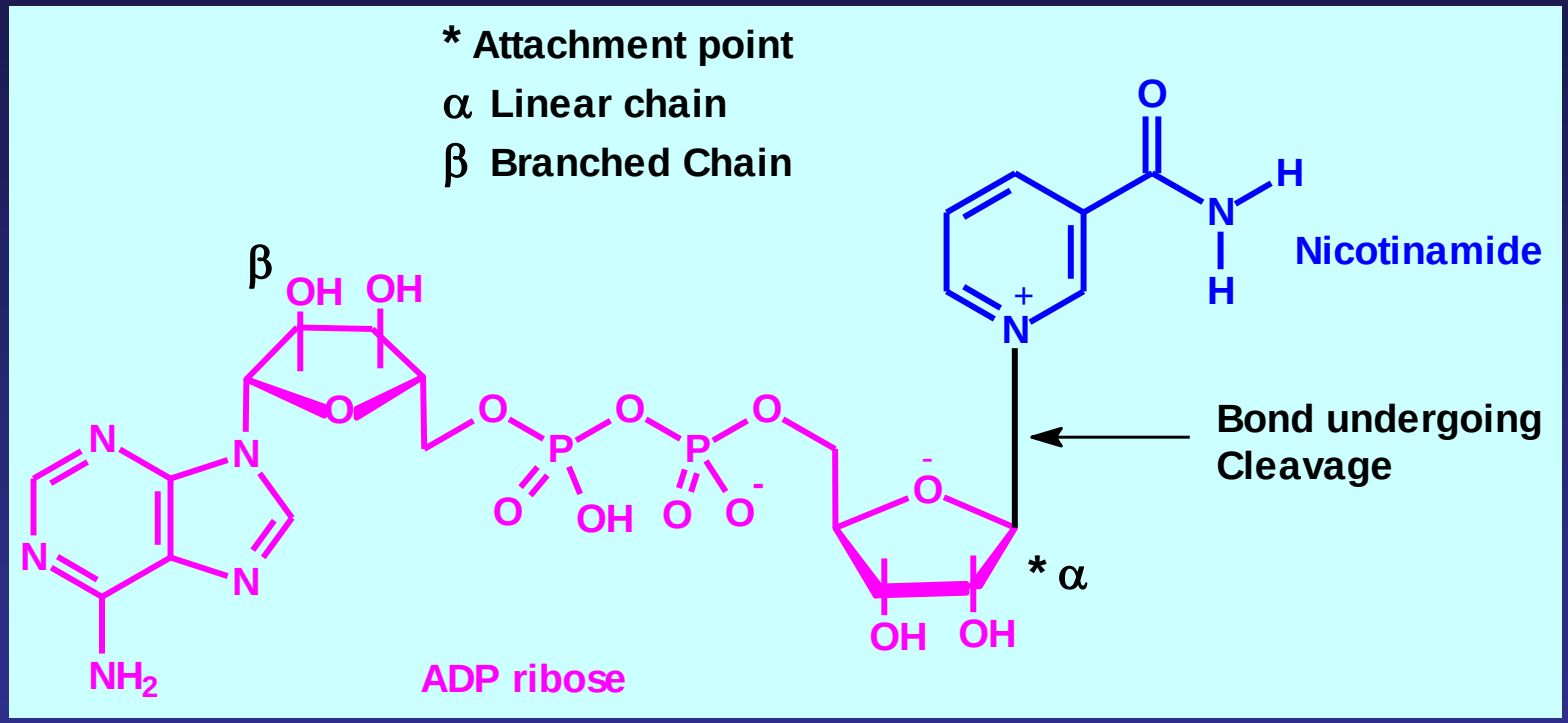
MAJOR MECHANISMS OF DNA DAMAGE AND REPAIR



Ionising radiation Antitumour agents	UV light Polycyclic aromatic hydrocarbons	Replication errors	Ionising radiation Oxygen radicals Spontaneous reactions Antitumour agents	Alkylating agents
Interstrand crosslink Double-strand break	(6-4)PP Bulky adduct CPD	A-G mismatch T-C mismatch Insertion Deletion	Uracil Abasic site 8-Oxoguanine Single-strand break	DNA alkylation O^6-alkylguanine
Recombinational repair (HR, NHEJ)	Nucleotide excision repair	Mismatch repair	Base excision repair	Direct reversal (AGT, MGMT)
DNA PKi **ATMi**			**PARPi**	**O^6BG** **PaTrin**

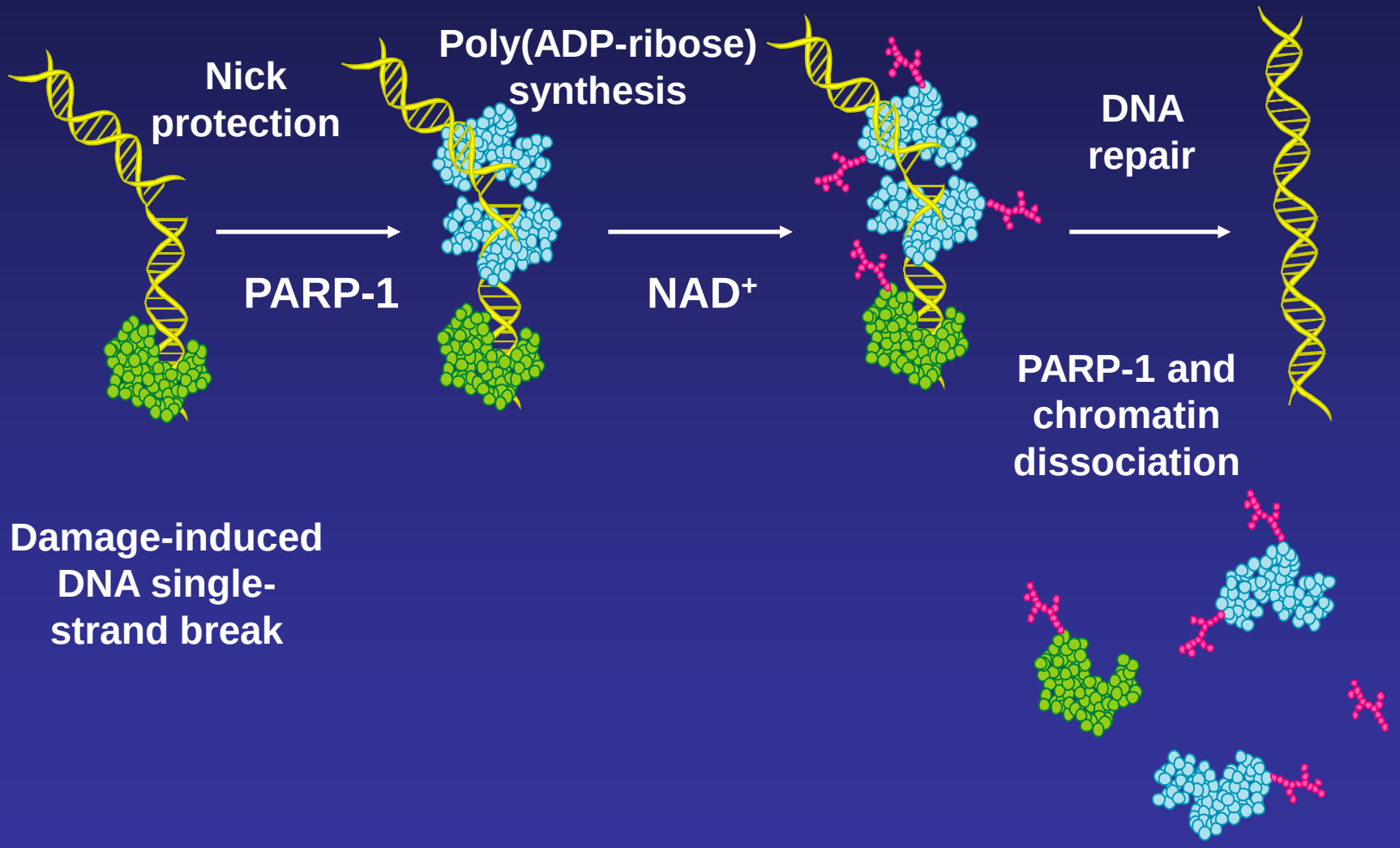
Modified from Hoeijmakers, J. H. (2001) *Nature* **114**, 366-374.

Catalytic Activity of PARP



PARP forms ADP-ribose polymers attached to histone proteins and to itself.
PARP uses NAD as a substrate

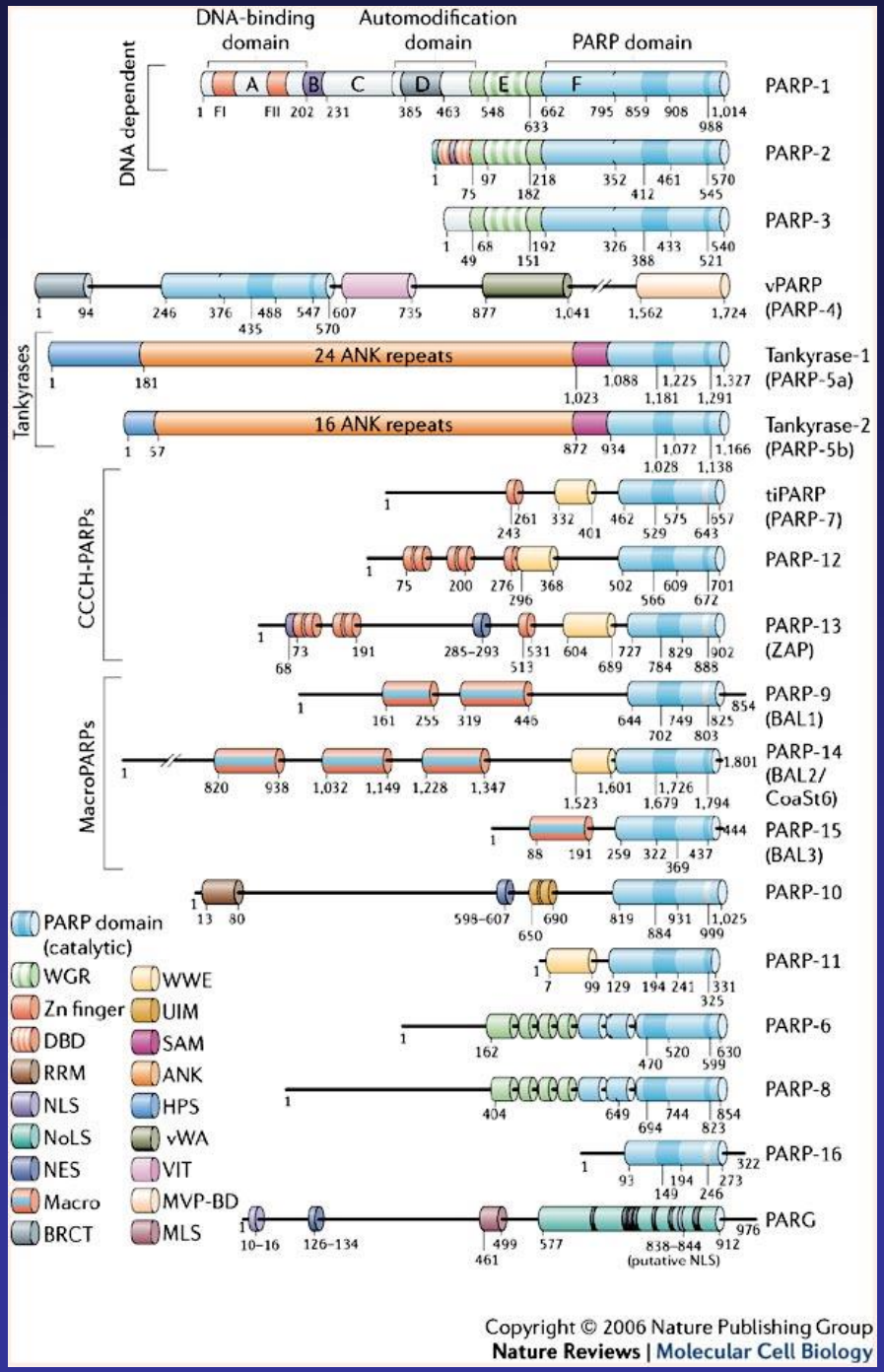
Mechanism of Action of PARP in Base Excision Repair



Poly(ADP-ribose)polymerase (PARP): A truly cancer-specific target?

- 17 isoforms – PARP-1 is the best characterised
- Present in high activity in most tissues
- Activated by DNA strand breaks and involved with single-strand break repair
  - "Housekeeping" function
- Utilises NAD as a substrate to form APD-ribose polymers on histone proteins and itself (automodification)
- Involved in numerous other processes
  - Epigenetic regulation of chromatin structure and gene expression
  - Interacts with transcription factors and co-factors (NF)-kB, PAX6, AP-2, b-Myb, TEF1
  - Interacts with kinetochore proteins
  - etc, etc

17 PARP Isoforms



Schreiber et al. Nature Reviews Molecular Cell Biology 7:517, 2006

We were fortunate in 1990 that we knew only about PARP-1

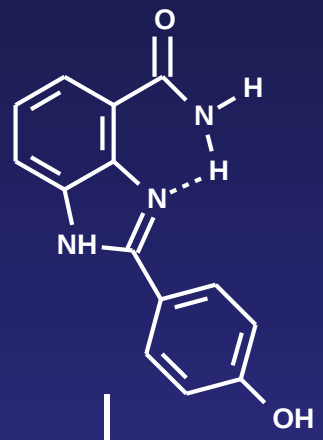
PARP Inhibitor Programme, Newcastle Anticancer Drug Development Initiative, 1990

- Rationale
  - Inhibition of PARP (PARP-1) known to potentiate in vitro
    - Monomethylating agents (temozolomide, nitrosoureas)
    - Topoisomerase 1 inhibitors (topotecan, irinotecan (SN38))
    - Radiation therapy
- Objective
  - To generate high affinity PARP inhibitors for in-vivo / clinical use
- Note
  - BRCA1 identified 1994, BRCA2 identified 1995

Development of High-Affinity PARP Inhibitors (Newcastle / Agouron)

3-aminobenzamide

Ki = 4μM



NU 1085

Ki = 10nM

Agouron collaboration - crystal structure

PD128763

Ki = 70nM

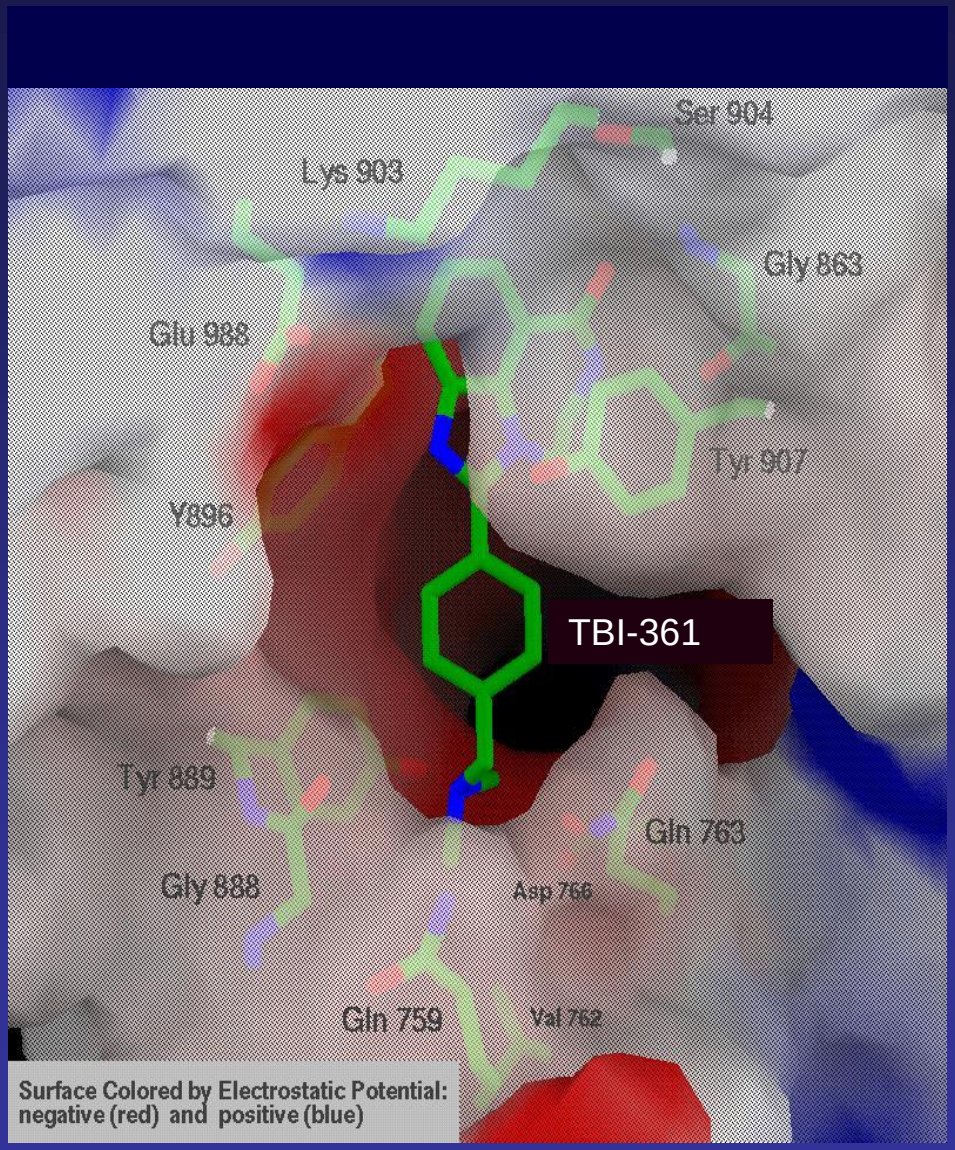
Hypothermia

AG14699

Ki = < 5 nM

Phase I 2003, In clinical development by Pfizer

TBI-361

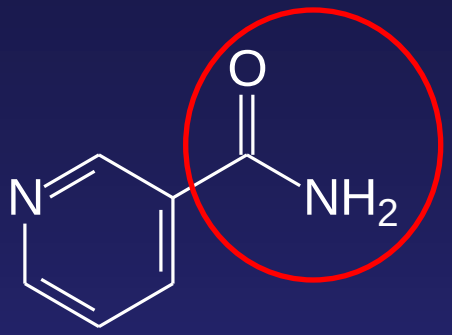


Constraining the carboxamide ring in a seven membered ring maintained the interactions with the active site.

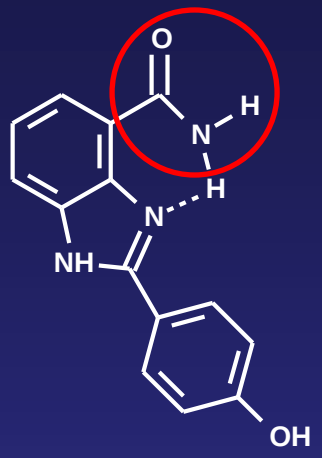
Ki < 5nM purified full length rhPARP-1

Increased

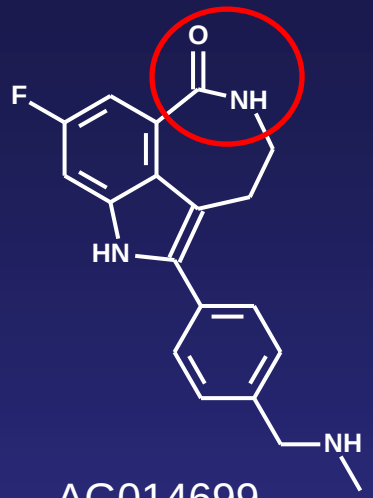
Structures of PARP Inhibitors



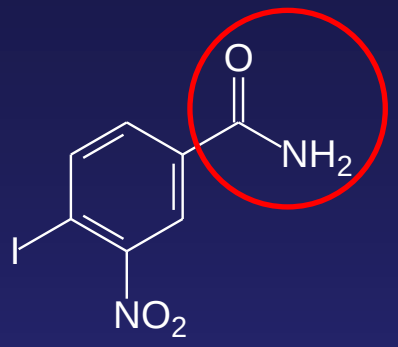
Nicotinamide
Component of NAD



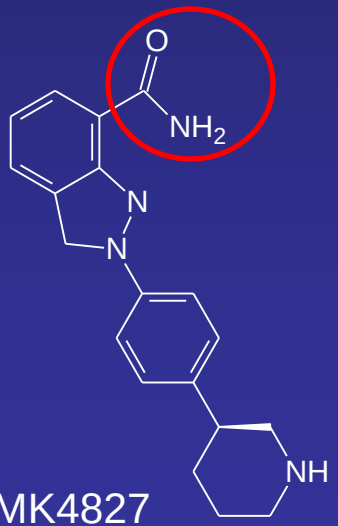
NU 1085
(Newcastle University)



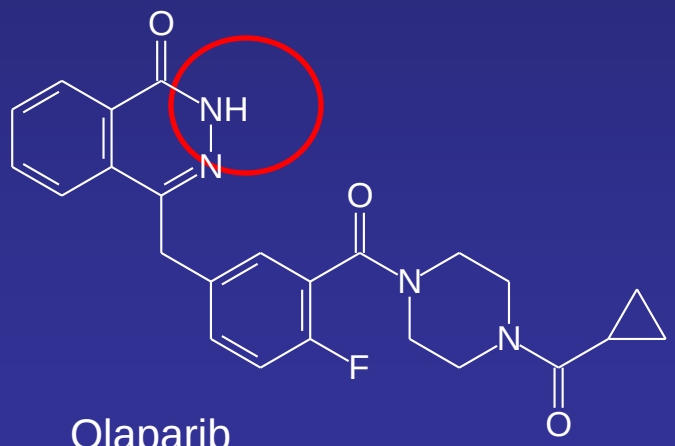
AG014699
Agouron/Pfizer



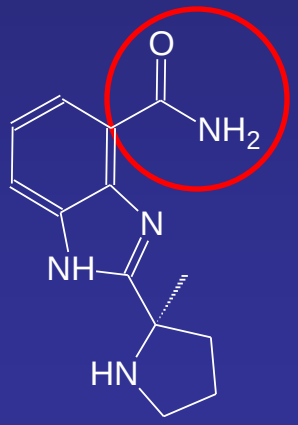
Iniparib
BSI 201
Bipar / Sanofi-Aventis



MK4827
Merck & Co



Olaparib
AstraZeneca



Veliparib (ABT888)
Abbott

PARP Inhibitors in Cancer Treatment

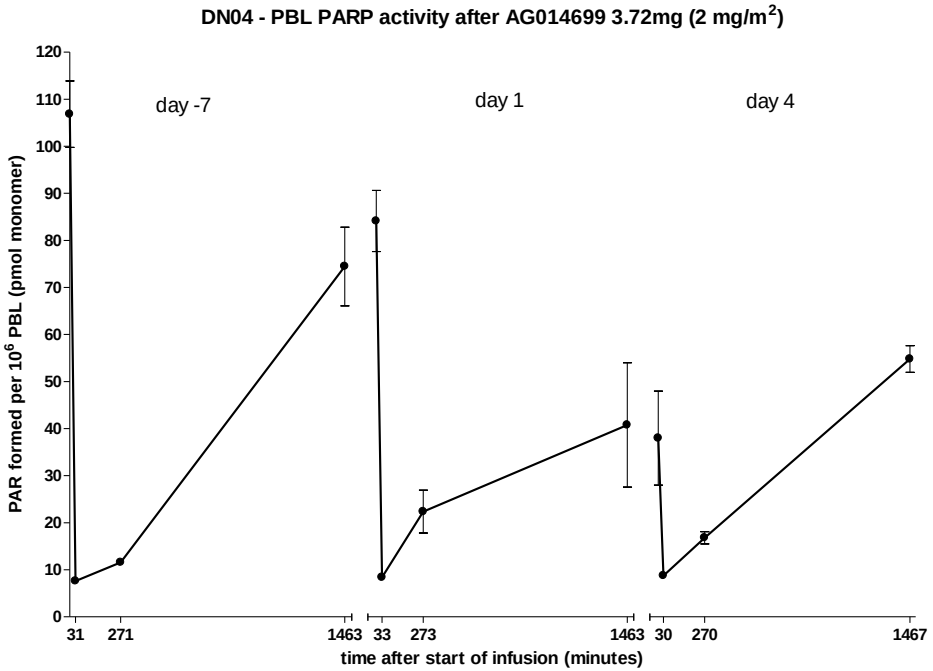
- Role 1
  - Potentiate specific cytotoxic drugs
    - Monomethylating agents (eg temozolomide)
    - Topoisomerase 1 inhibitors (eg topotecan)
- Role 2
  - Potentiate radiation therapy
- Role 3 “Synthetic lethality”
  - Single agent activity in tumours deficient in homologous recombination repair (HR) (eg BRCA1, BRCA2)
- Role 4
  - In combination with drugs known to be active in tumours which have HR defects (eg carboplatin / ovarian). Specific potentiation of these drugs not consistently seen in vitro
- (Role 5
  - Chemoprevention in BRCA carriers)

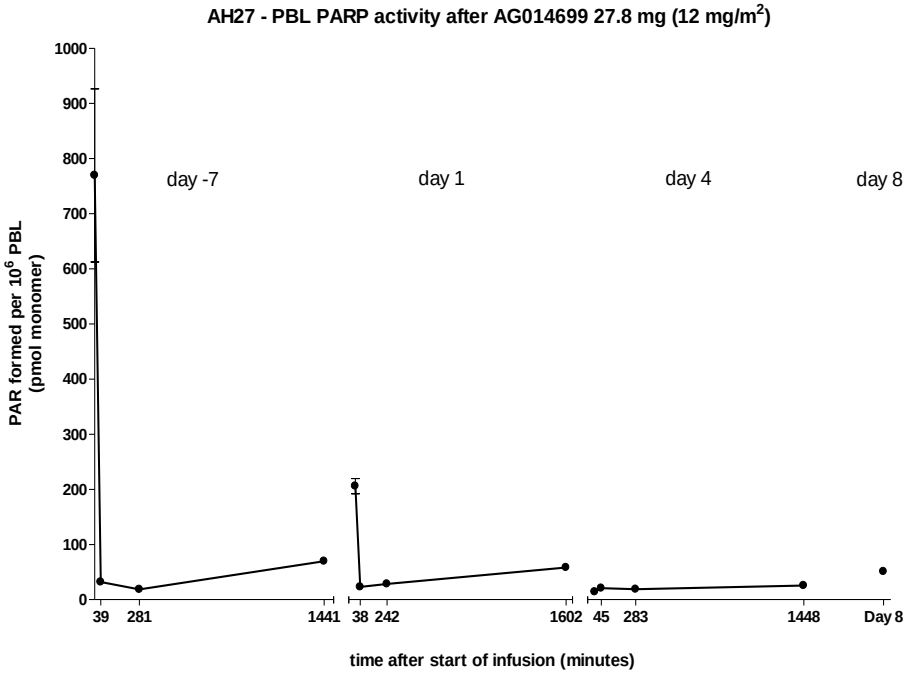
Phase 0 / 1 Trial of AG014699 Day 1-5 Schedule with temozolomide

- Substantial (≥90%) PARP inhibition seen in peripheral blood mononuclear cells and tumour biopsies
- No significant toxicities attributable to the PARP inhibitor as a single agent
- No dose-reduction for temozolomide
- Clinical activity observed

Plummer R et al, Phase I Study of the Poly (ADP-Ribose) Polymerase Inhibitor, AG014699…, Clinical Cancer Research 14(23):7917-7923, 2008

PARP Inhibitory Dose established using PD assay (immunoblot for polymer in PBLs)

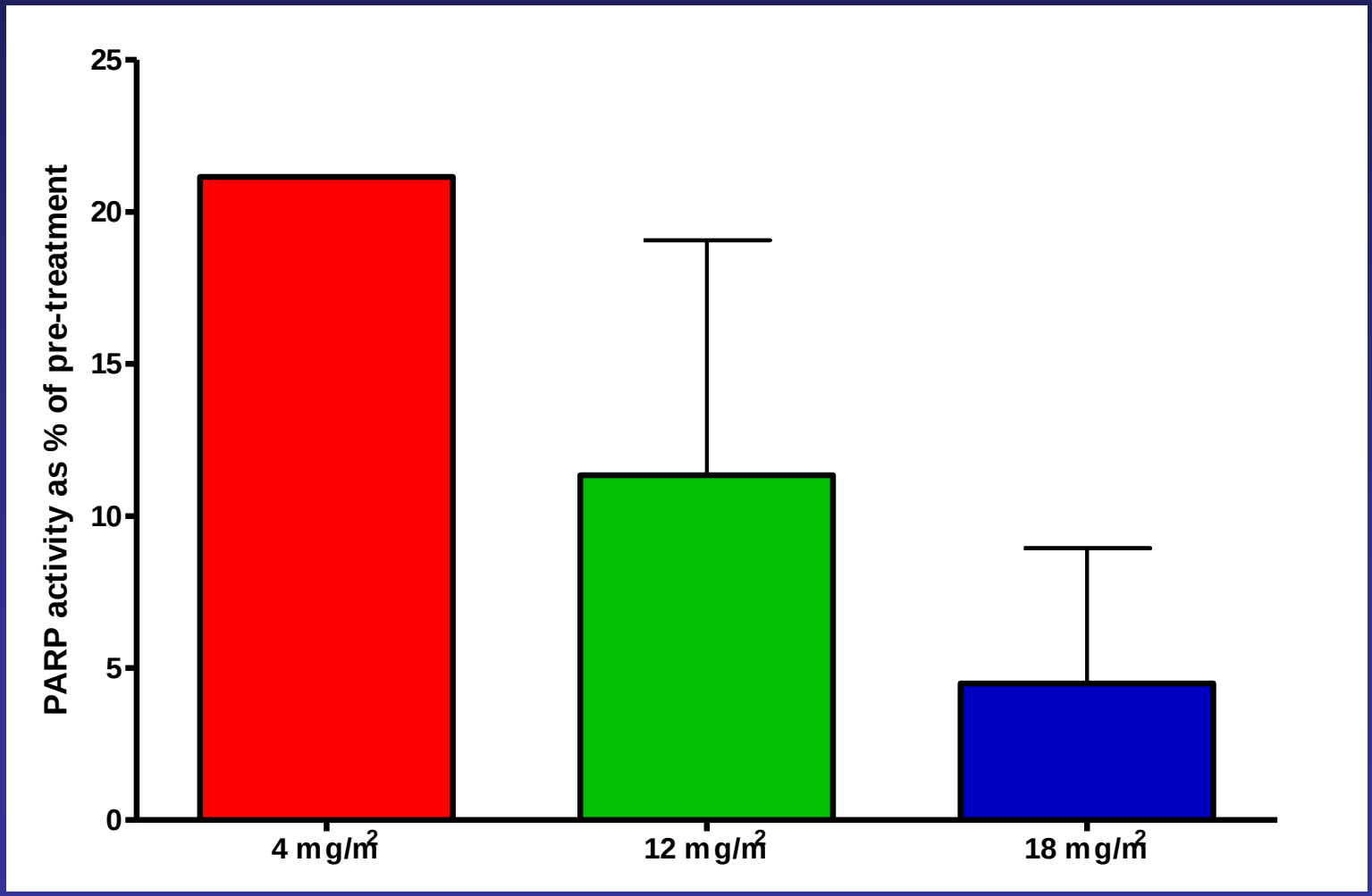




PARP inhibition in PBLs
2 mg/m^2 AG014699

PARP inhibition in PBLs
12 mg/m^2 AG014699

Mean tumour PARP activity at 6 hours after a single dose of AG014699



Comparison of response data with TMZ single agent phase III

Efficacy Endpoint	*AG014699 + Temozolomide n 46 pts	M.R. Middleton, Phase III DTIC vs. TMZ (temozolomide arm) n 156 pts
CR	--	2.6 % (95% CI N/A)
PR	17.4% (95% CI 7.9%-31.6%)	10.9 % (95% CI N/A)
SD $\geq$ 24 wks	17.4% (95% CI 7.9%-31.6%)	Not Available
PFS	3.5 months (95% CI 2.0-6.2)	1.9 months (95% CI N/A)
OS	9.9 months (95% CI 6.2-14.7)	7.7 months (95% CI N/A)

•6 UK sites: Newcastle, Belfast, Oxford, Glasgow, Manchester, Birmingham (Plummer, Wilson, Middleton, Evans, Lorigan, Steven)

Plummer R, Lorigan P, Evans J, et al. J Clin Oncol 2006;24:456S.

PARP Inhibitors in Cancer Treatment

- Role 1
  - Potentiate specific cytotoxic drugs
    - Monomethylating agents (eg temozolomide)
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- Role 2
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- (Role 5
  - Chemoprevention in BRCA carriers)

PARP Inhibitors in Development

Agent	Company	Route	Clinical Status
AG014699 PF-01367338	Pfizer	Iv / oral	Phase I/II Combos
KU59436 AZD2281 Olaparib	AstraZeneca/ Kudos	oral	Phase II/III Combos
Veliparib ABT888	Abbott	oral	Phase I/II Combos
Iniparib BSI-201	BiPar/ Sanofi-Aventis	Iv Prodrug?	Phase II/III Combos
INO-1001	Inotek	iv	Phase Ib complete
GPI21016	MGI Pharma/ Eisai	oral	Phase I
CEP-9722	Cephalon	oral	Phase I
MK4827	Merck & Co	oral	Phase I
BMN-673	Biomarin / LEAD Pharmaceuticals		Preclinical

Specific killing of BRCA2-deficient tumours with inhibitors of poly(ADP-ribose) polymerase

Helen E. Bryant[1], Niklas Schultz[2], Huw D. Thomas[3], Kayan M. Parker[1], Dan Flower[1], Elena Lopez[1], Suzanne Kyle[3], Mark Meuth[1], Nicola J. Curtin[3] & Thomas Helleday[1,2]

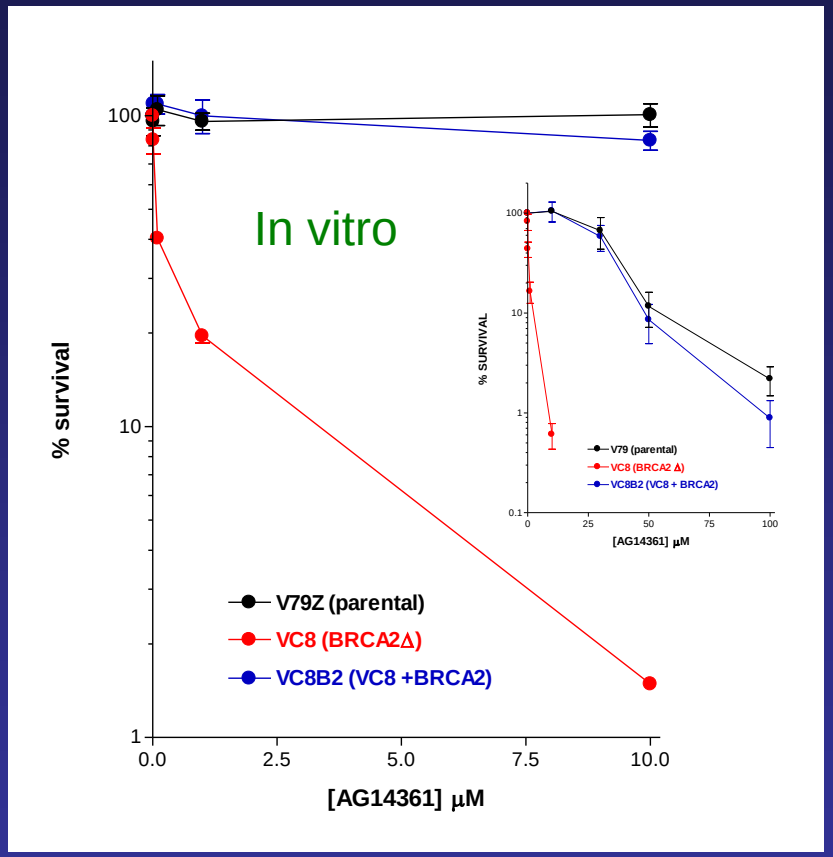
Nature 2005; 434:913-917 (Newcastle / Sheffield)

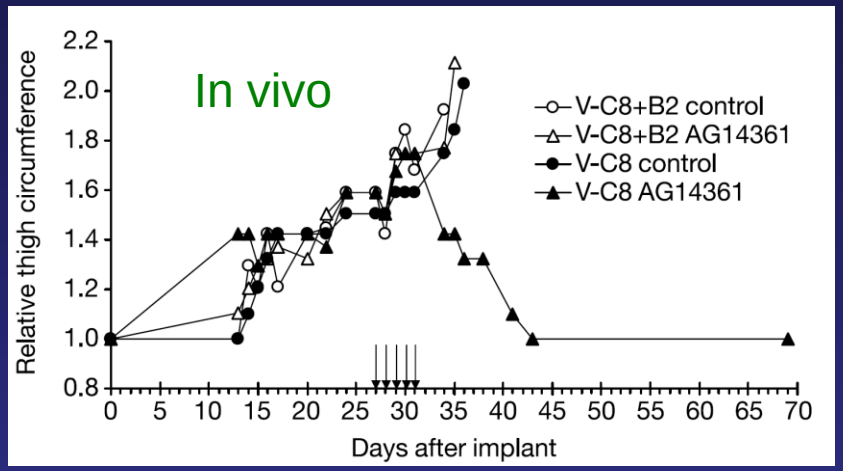
Targeting the DNA repair defect in *BRCA* mutant cells as a therapeutic strategy

Hannah Farmer[1,2*], Nuala McCabe[1,2*], Christopher J. Lord[2*], Andrew N. J. Tutt[2,3], Damian A. Johnson[2], Tobias B. Richardson[2], Manuela Santarosa[2†], Krystyna J. Dillon[4], Ian Hickson[4], Charlotte Knights[4], Niall M. B. Martin[4], Stephen P. Jackson[4,5], Graeme C. M. Smith[4] & Alan Ashworth[1,2]

Nature 2005: 434:917-921 (Institute of Cancer Research, London)

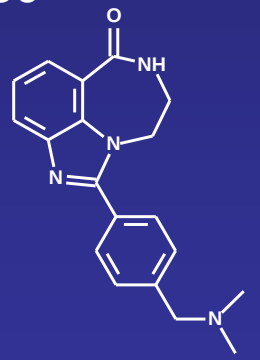
BRCA2-deficient cell lines are hypersensitive to PARP inhibitors (Newcastle / Pfizer Compounds)





"Therapeutic ratio" ~ 250

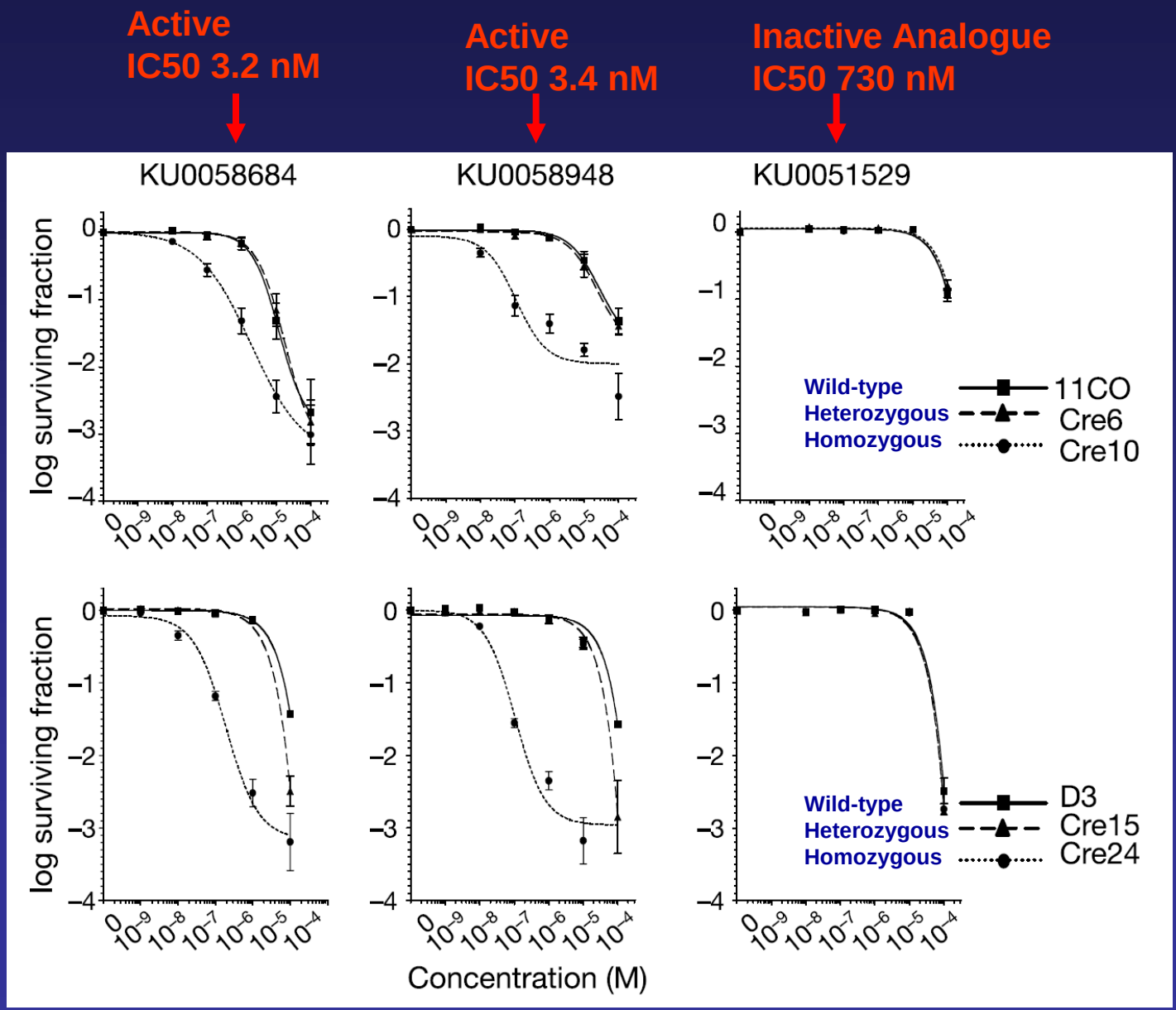
AG014699 (clinical development)



AG014361 (this expt)

Bryant HE, Schultz N, Thomas HD, Parker KM, Flower D, Lopez E, Kyle S, Meuth M, Curtin NJ, Helleday T. Specific killing of BRCA-2 deficient tumours with inhibitors of poly(ADP-ribose) polymerase. Nature 2005; 434:913-917

Mutation in BRCA1 or BRCA2 Results in Extreme Sensitivity to PARP Inhibition (Kudos/AZ Compounds)

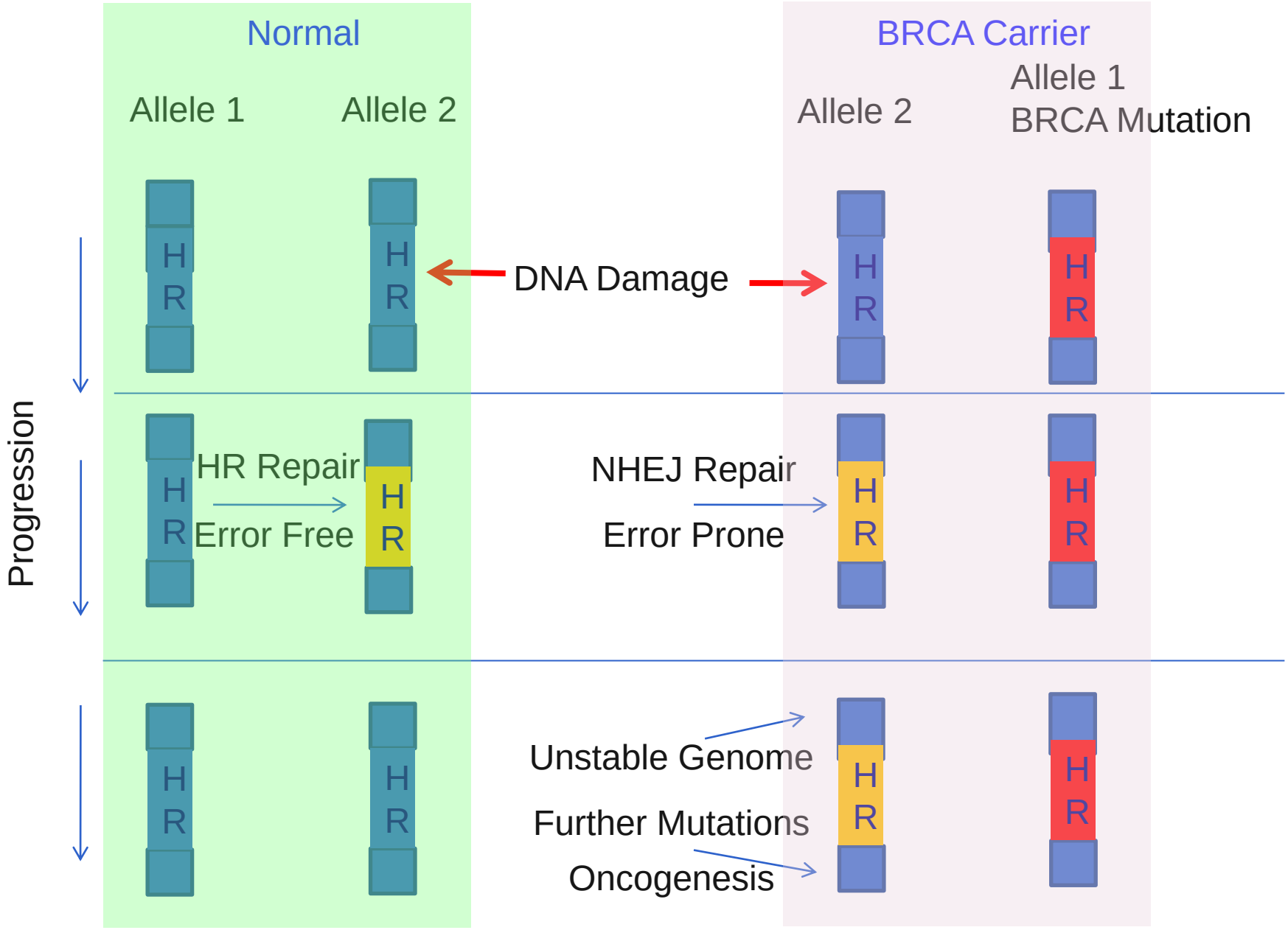


Adapted from:
Farmer et al. Nature 434, 917-921, 2005

BRCA1 and BRCA2 Cancer Predisposition Genes

- Mutation carriers are predisposed to breast, ovarian, prostate, pancreatic and other cancers
- BRCA1 and BRCA2 are involved in homologous recombination repair – error-free repair of double strand breaks
- Carriers have one allele carrying a mutant, non-functioning gene. Damage to the functioning copy results in error-prone DNA repair and is oncogenic

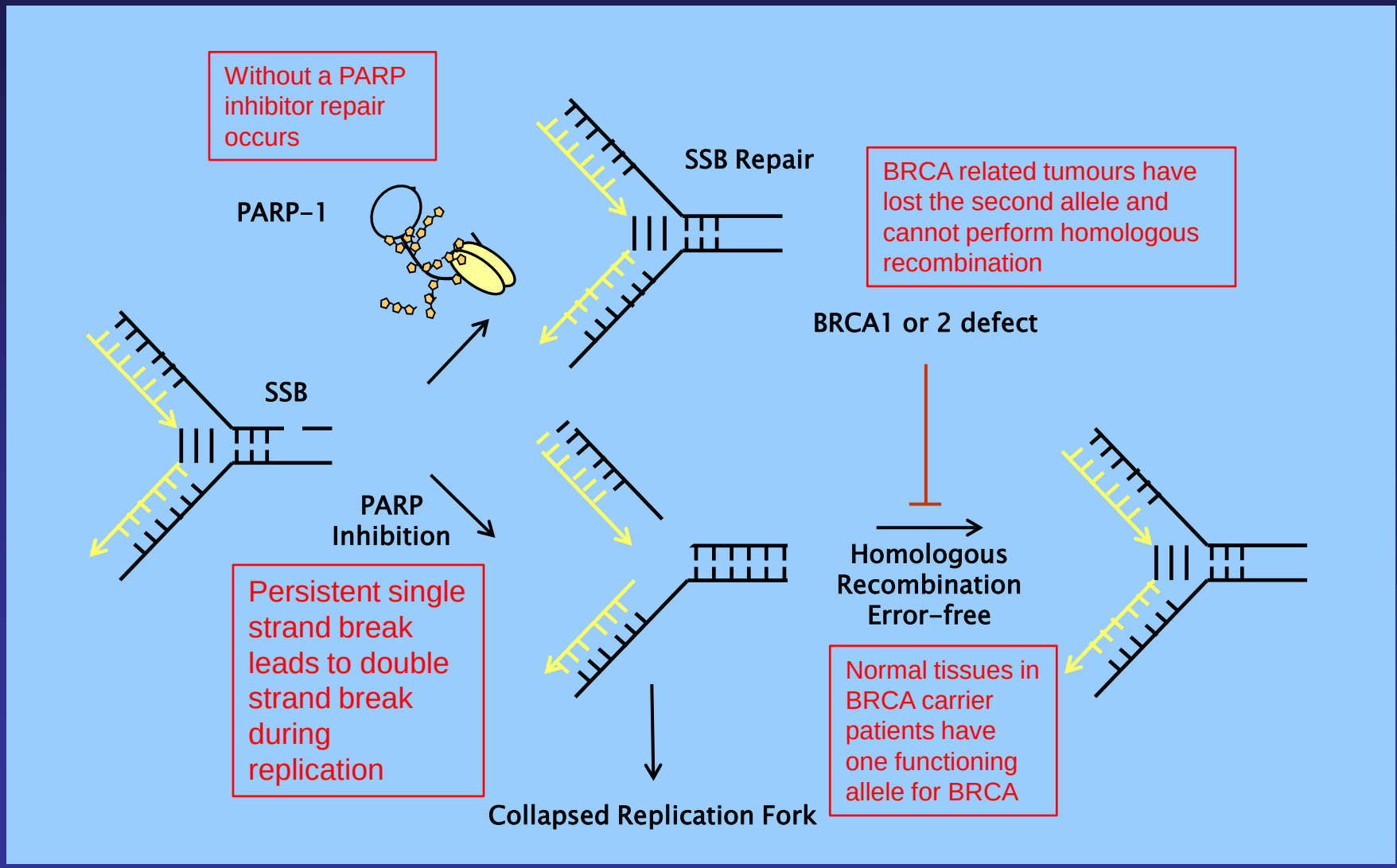
BRCA Carriers and Cancer Susceptibility



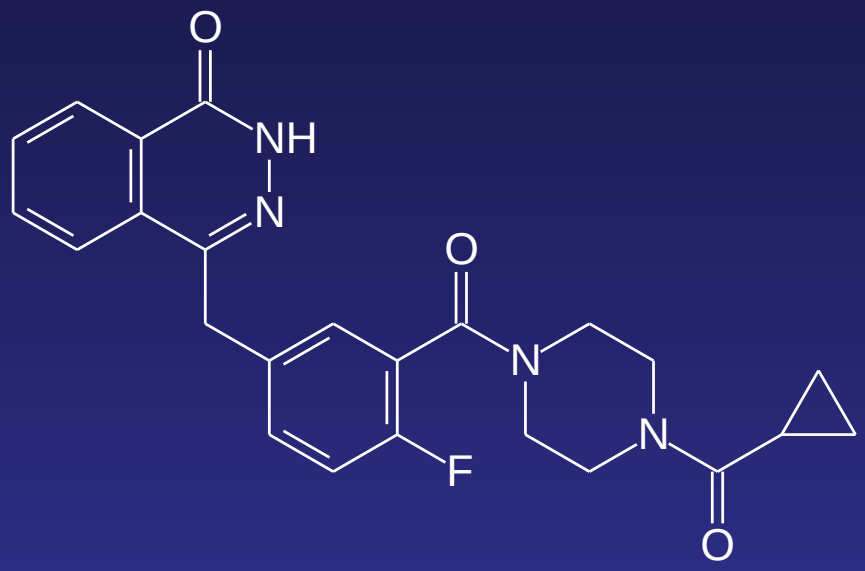
Properties of Cancers Arising in BRCA1/2 Carriers

- The cancer has lost the ability to carry out HR (homologous recombination repair)
- Ovarian cancers are typically more sensitive to platinum treatment than sporadic (non-BRCA) cases.

Proposed Mechanism of Synthetic Lethality of PARP Inhibitors to BRCA1 or 2-Deficient Cells



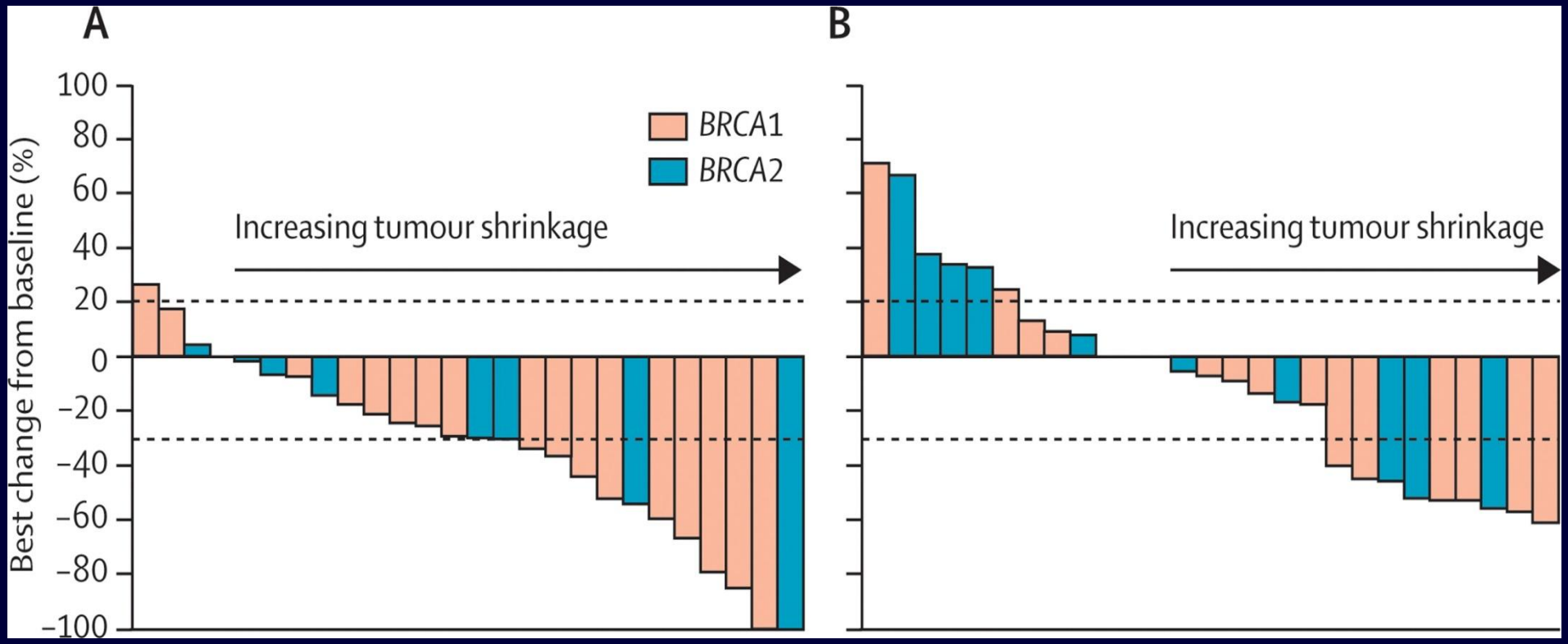
Olaparib – Kudos / AstraZeneca



- Orally available PARP inhibitor generated responses in hereditary cancers in Phase I*
- Phase II results in patients with BRCA1 or 2 related breast and ovarian cancer presented at ASCO 2009

* Peter C. Fong et al. Inhibition of Poly(ADP-Ribose) Polymerase in Tumors from BRCA Mutation Carriers. New England Journal of Medicine 361(2):123, 2009

Olaparib Phase II in Breast Cancer
Best % change from baseline in target lesions by genotype

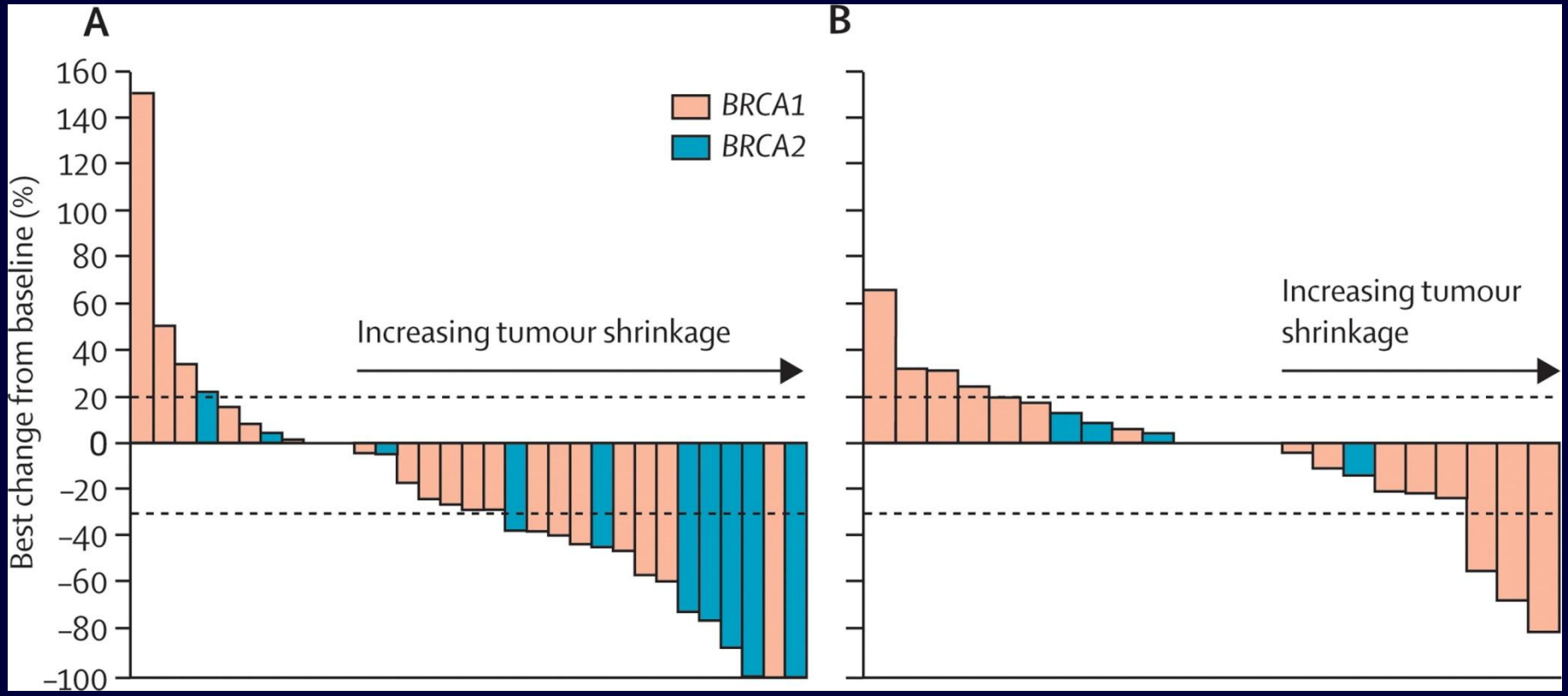


400 mg twice daily

100 mg twice daily

Tutt et al. Lancet 376:235, 2010

Olaparib Phase II in Ovarian Cancer
Best % change from baseline in target lesions



400 mg twice daily

100 mg twice daily

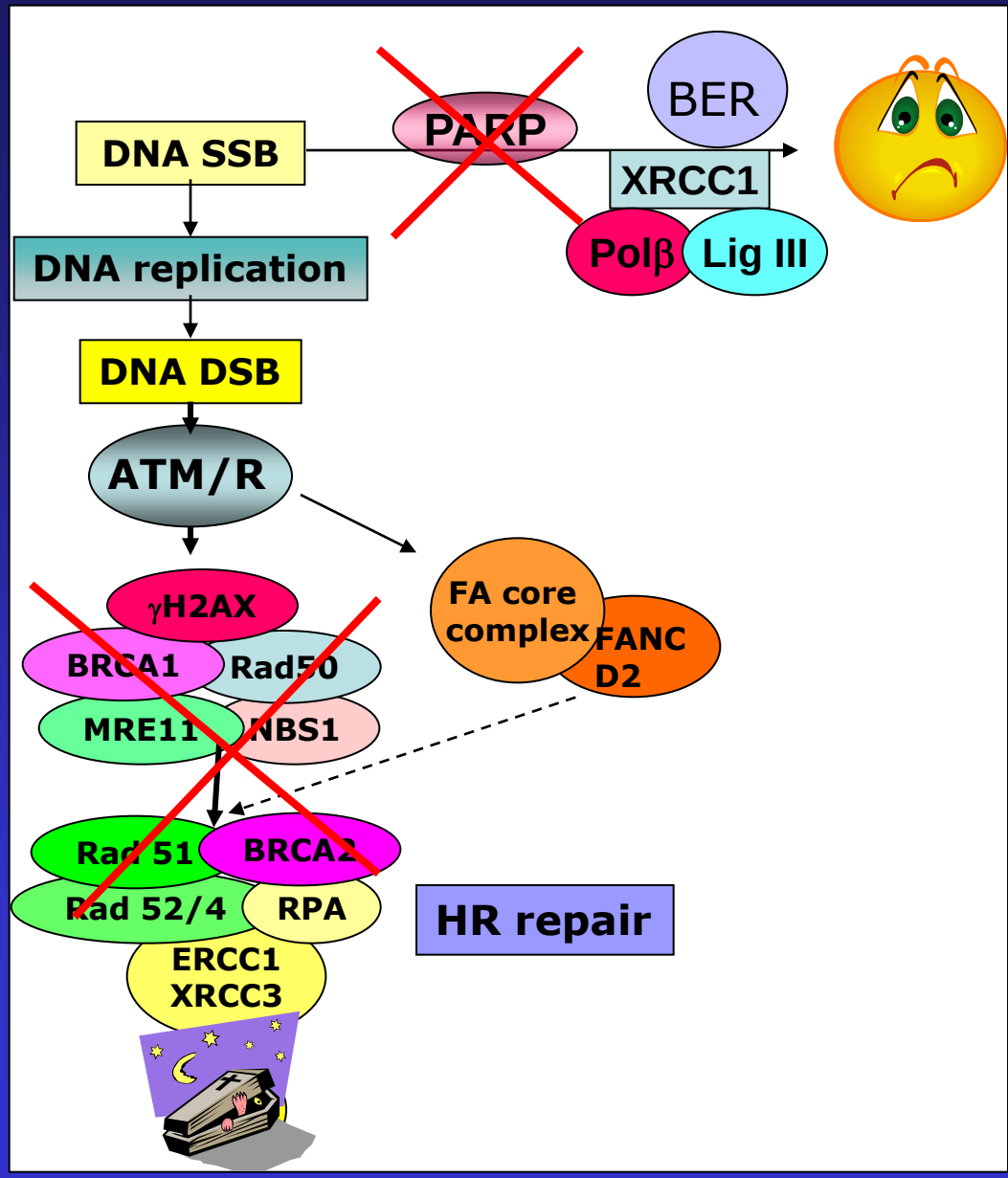
Audeh et al. Lancet 376:245, 2010

Resistance to PARP Inhibitors

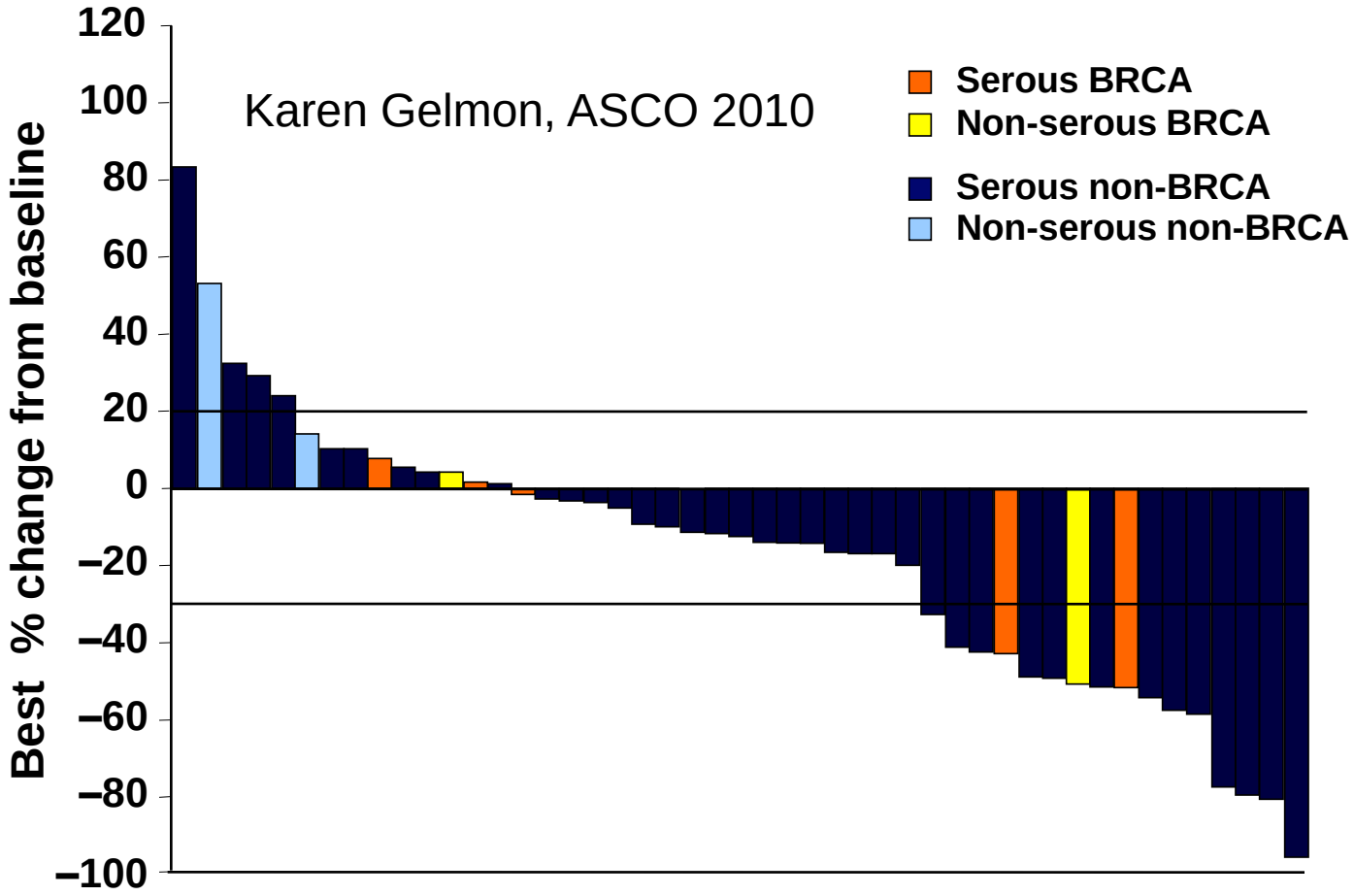
- Edwards et al. Nature 451:1111, 2008
  - BRCA2 deficient CAPAN1 cells were rendered PARP-inhibitor resistant and showed a re-activating intragenic deletion of BRCA2
  - Similar mechanism in cell lines derived from platinum-resistant patients
  - Not known whether this mechanism occurs in patients treated with PARP Inhibitors
- Swisher et al. Cancer Research 68:2581, 2008
  - 4 of 6 platinum-resistant BRCA1 tumours had acquired secondary genetic changes
  - 0 of 3 platinum-sensitive BRCA1 tumours had secondary genetic changes

Proposed mechanism and therapeutic potential

- Endogenously formed SSB are normally repaired by PARP-dependent BER.
- If PARP is inhibited SSB persist.
- SSB form DSB at replication, which are repaired by HR.
- If HR is defective the breaks are not repaired and the cell dies.
- This is the first exploitation of **synthetic lethality** in cancer therapy.
- Tumour selective.



Best % Change from Baseline in Target Lesion: High Grade Serous Ovarian/undifferentiated Tubo-ovarian; Unknown or BRCA –ve at Entry

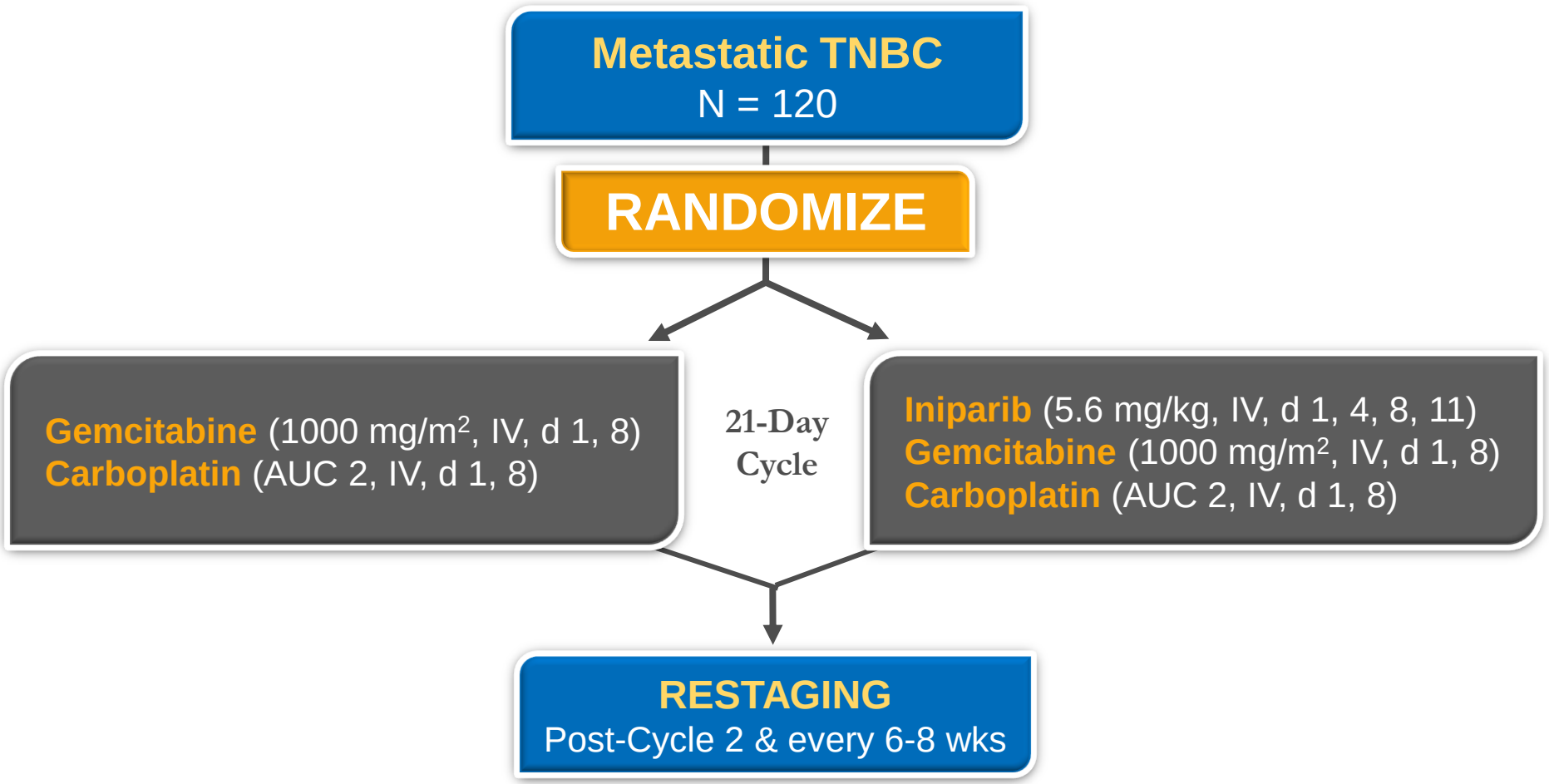


Best change in target lesion size is maximum reduction from baseline or minimum increase in absence of reduction

PARP Inhibitors in Cancer Treatment

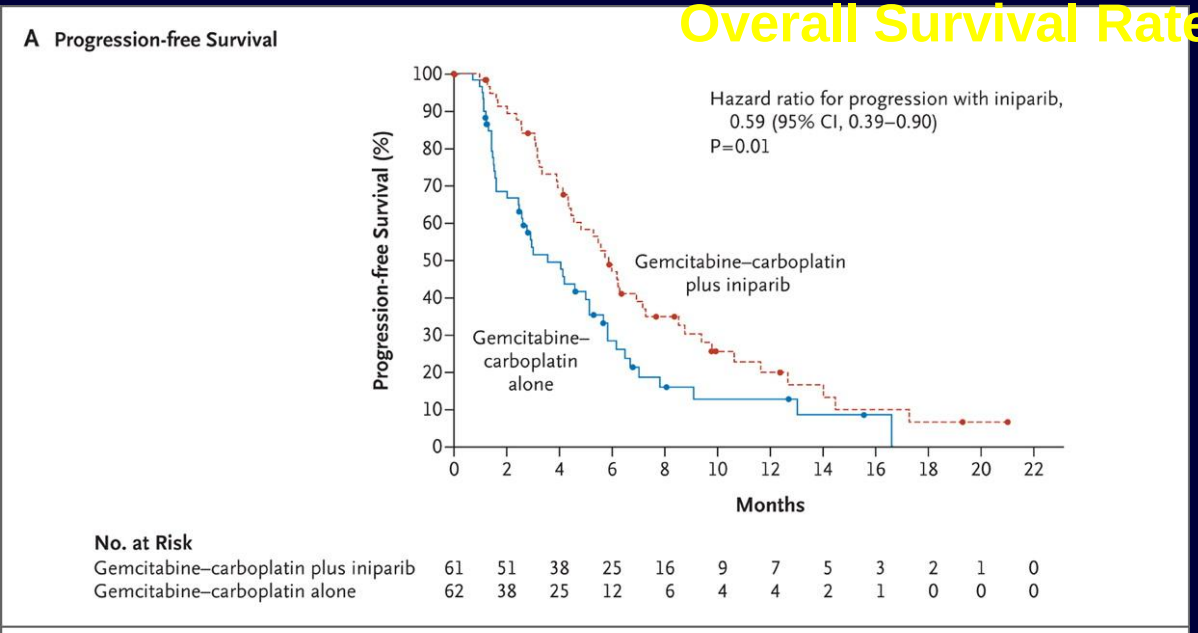
- Role 4
  - In combination with drugs known to be active in tumours which have HR defects (eg carboplatin / ovarian). Specific potentiation of these drugs not consistently seen in vitro

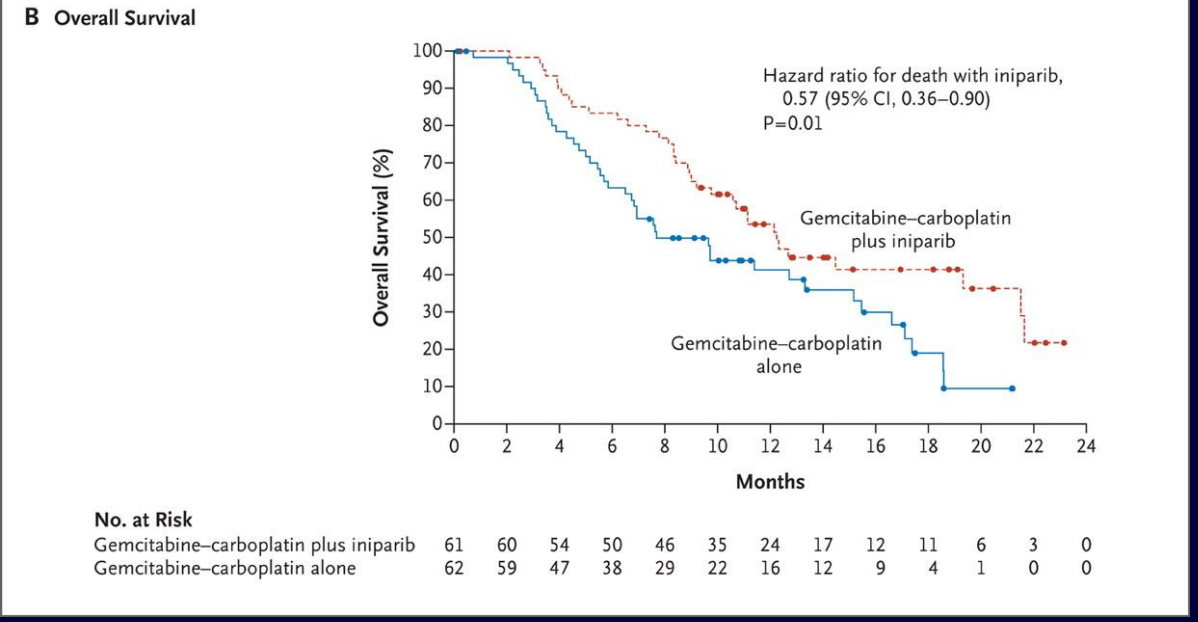
Phase 2 mTNBC Study: Treatment Schema



* Patients randomized to gem/carbo alone could crossover to receive gem/carbo + iniparib at disease progression

Iniparib Phase II in Triple Negative Breast Cancer: Progression-free and Overall Survival Rates





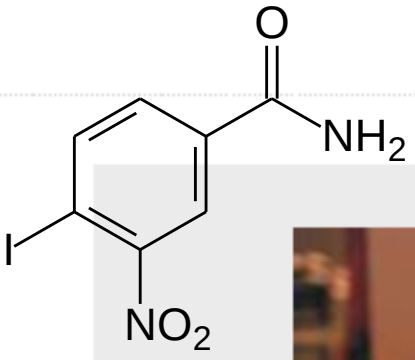
O'Shaughnessy J et al. N Engl J Med 2011;364:205-214.

The NEW ENGLAND JOURNAL of MEDICINE

BiPar breast cancer drug fails late-stage trial

San Francisco Business Times - by Ron Leuty
Date: Friday, January 28, 2011, 4:06pm PST - Last Modified: Friday, January 28, 2011, 5:11pm PST

Related: Health Care



A promising breast cancer treatment developed by BiPar Pharmaceuticals Inc. fizzled in a major late-stage study.

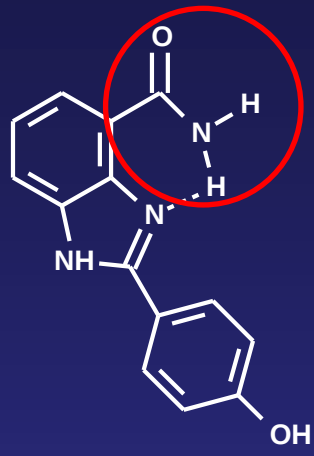
The drug, iniparib or BSI-201, did not hit either of the main goals — overall survival and progression-free survival — in a study of 519 women with metastatic triple-negative breast cancer, parent company Sanofi-Aventis said.

Iniparib has been closely watched for scientific and business reasons. For one, it belongs to a class of cancer drugs known as PARP inhibitors. Also, Sanofi made a splash when it bought tiny, South San Francisco-based BiPar in 2009 for as much as $500 million in total payouts, depending on iniparib's level of success.

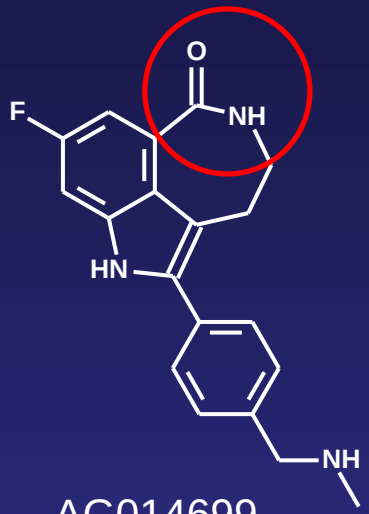
Enlarge Image

CEO Atul Dhir came to the South San Francisco-based BiPar after its acquisition by Sanofi-Aventis.

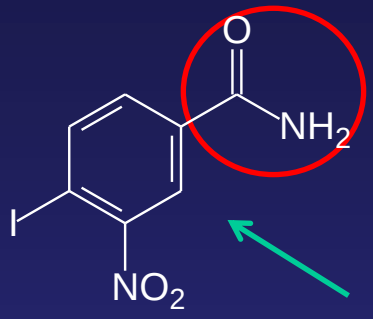
Structures of PARP Inhibitors



NU 1085
(Newcastle University)

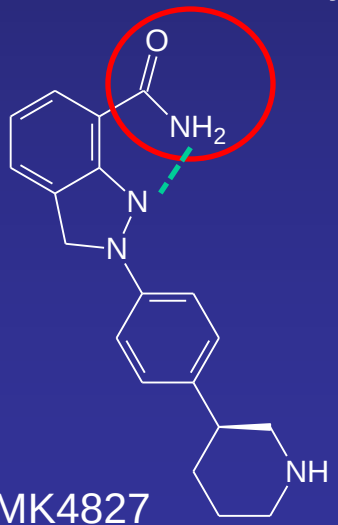


AG014699
Agouron/Pfizer

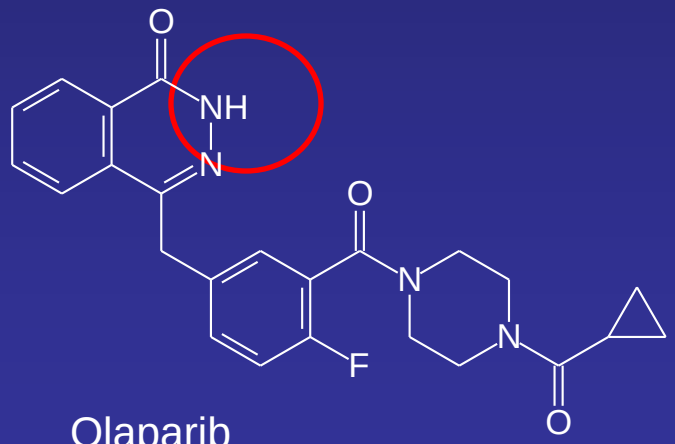


Iniparib
BSI 201
Bipar / Sanofi-Aventis

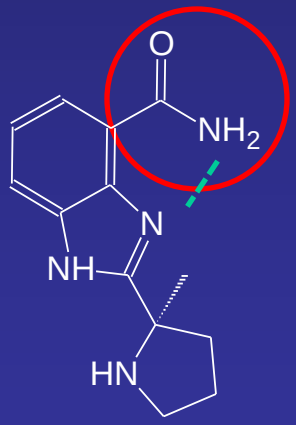
Iniparib itself is not a PARP Inhbitor – an active metabolite is proposed



MK4827
Merck & Co



Olaparib
AstraZeneca



Veliparib (ABT888)
Abbott

Identification of HR Deficient Tumours

- γH2AX, RAD51
  - Mukhopadhyay et al, Clin Cancer Res; 16(8) :2344, 2010
- PTEN
  - Mendes-Pereira et al, EMBO Mol Med 1, 315–322, 2009
- BRCA1 Expression
  - *Carser, JCO 27:15s, 2009 abstract 5527*

PARP-BRCA Summary

- PARP inhibitors provide specific therapy for tumours arising in patients who are BRCA1 or 2 mutation carriers
- PARP Inhibitors also show single agent activity in non-BRCA tumours likely to have a HR defect
- PARP inhibitors will probably potentiate chemotherapy agents – probably preferably in tumours with low homologous recombination repair capability
- Biomarkers are required to select tumours which will be sensitive to PARP inhibitors: Possible potential markers
  - RAD51 focus formation
  - PTEN deficiency
  - BRCA1 Expression
  - ……..

Newcastle Anticancer Drug Development Initiative, 1990






Barbara Durkacz

Discovered PARP1

Roger Griffin

Medicinal Chemistry

Bernard Golding

Herbie Newell

Preclinical Pharmacology

Nicola Curtin

DNA repair Biology

Ruth Plummer

Clinical Trials

The PARP Team

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