Targeting DNA damage repair beyond PARP – further drugs or targets in development

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Disclosure

- I have no conflicts of interest to disclose in relation to the targets discussed in this talk.

- Newcastle University receives research funding for ongoing or previous in projects in this area

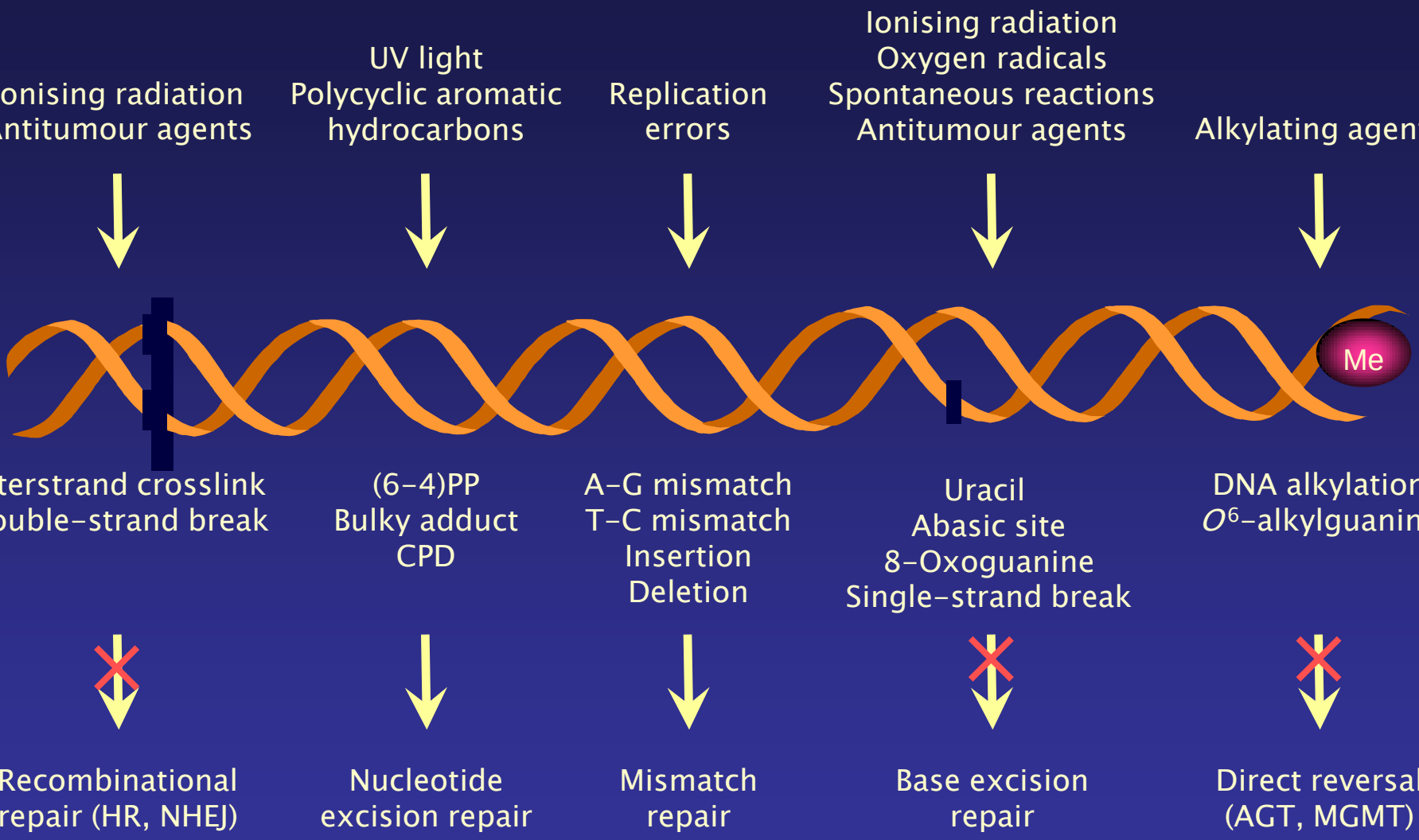
Brief

- Overview of DNA damage response (DDR) pathways
- Lessons from history of DNA repair inhibition and chemotherapy
- Brief description of DDR targets currently in development or late pre-clinical
- Thoughts on trial design
- Opportunities for combinations

DDR Inhibitors in Cancer Treatment

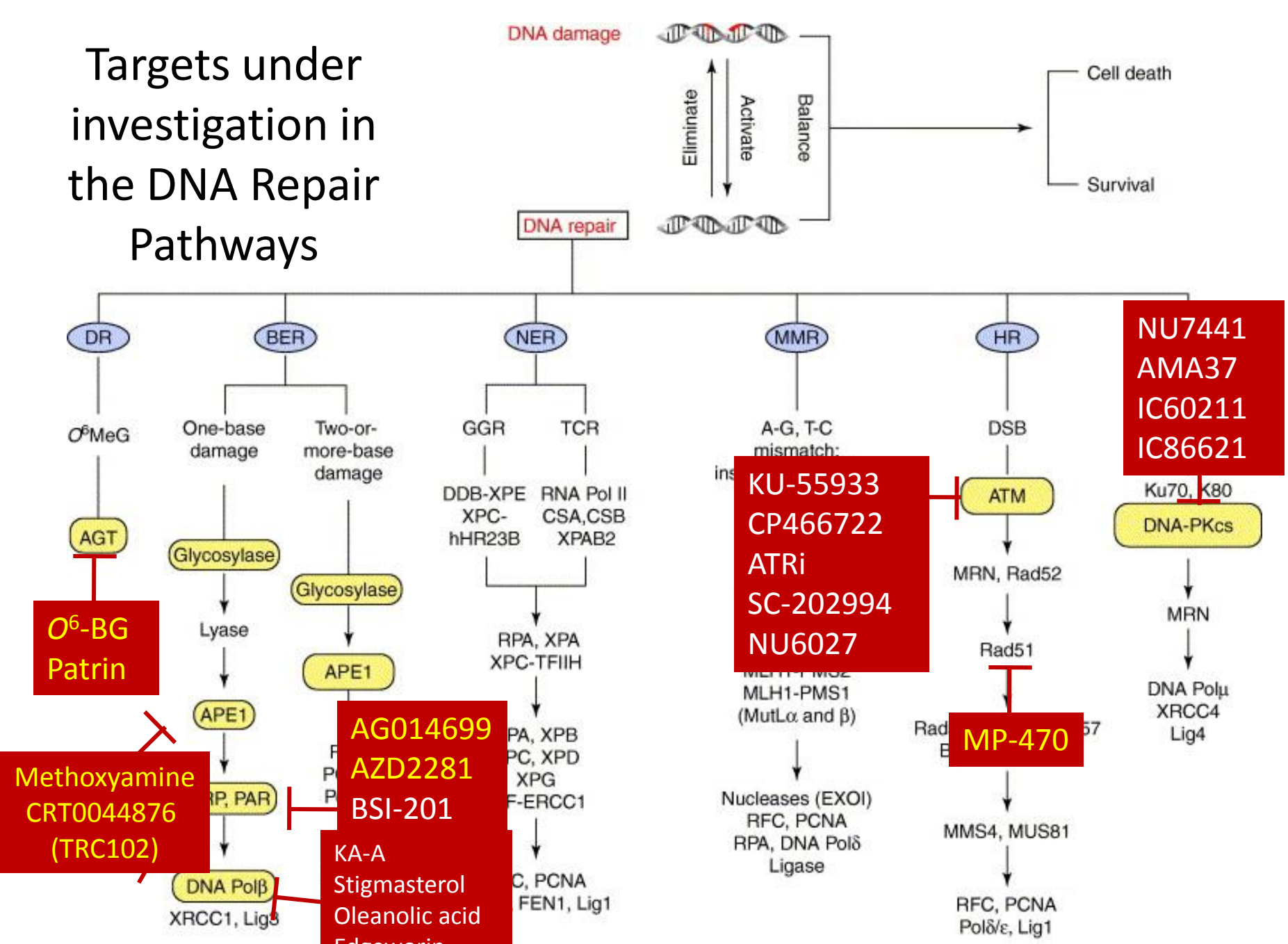
- Radiotherapy and many anticancer drugs act by damaging DNA
  - DNA repair in the tumour may be a cause of resistance
    - DNA repair inhibitors may be chemo- or radiosensitizers
- Some tumours may be more effective at DNA repair than the target normal tissue
  - Inhibiting the DNA repair pathways will level the playing field
- Some tumours lack specific DNA repair pathways (eg, BCRA1/BRCA2, HNPCC, ATM, DNA-PK, Fanconi)
  - Inhibiting alternate repair pathways may be a mechanism for antitumour selectivity *("synthetic lethality")*
  - Sensitivity to specific DNA-reactive drugs (eg carboplatin) may occur

MAJOR MECHANISMS OF DNA DAMAGE AND REPAIR



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

Targets under investigation in the DNA Repair Pathways

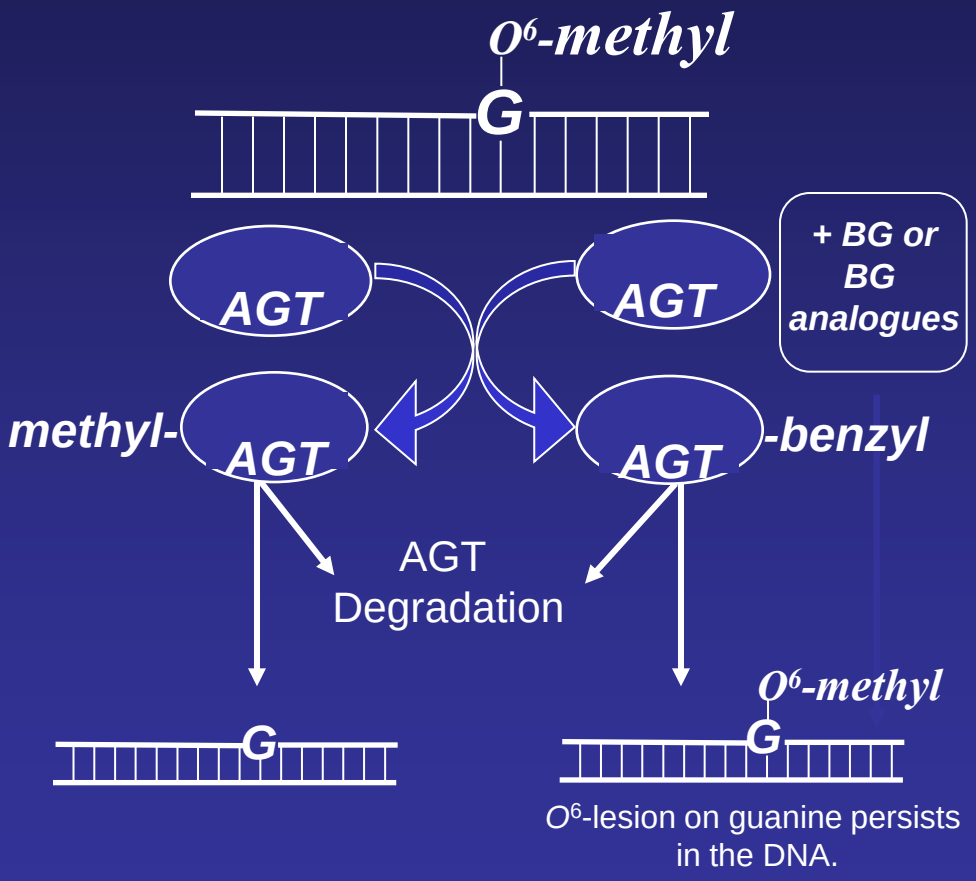


Modified from Ding et al. Trends in Pharm. Sciences, 2006

With thanks to Mark Kelly

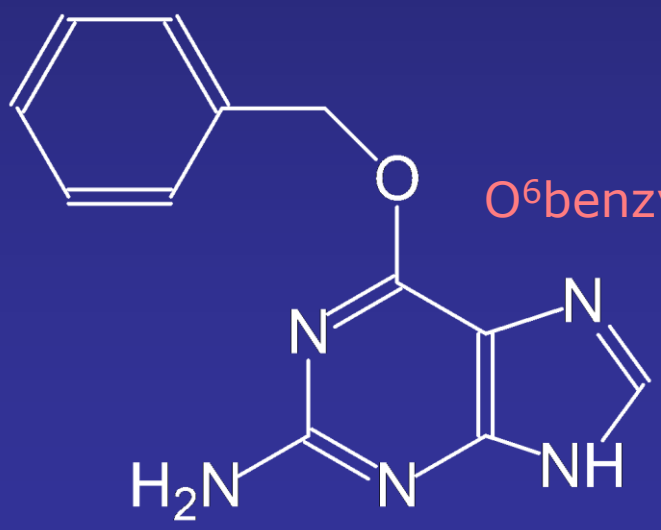
LESSONS FROM CHEMOPOTENTIATION TRIALS WITH DDR MEDIATORS

AGT or MGMT (direct reversal repair)



Carmustine Combined with O^6-Benzyl guanine

Carmustine

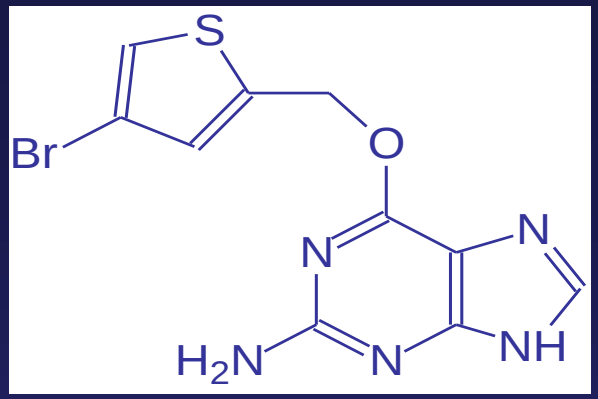


O^6benzylguanine

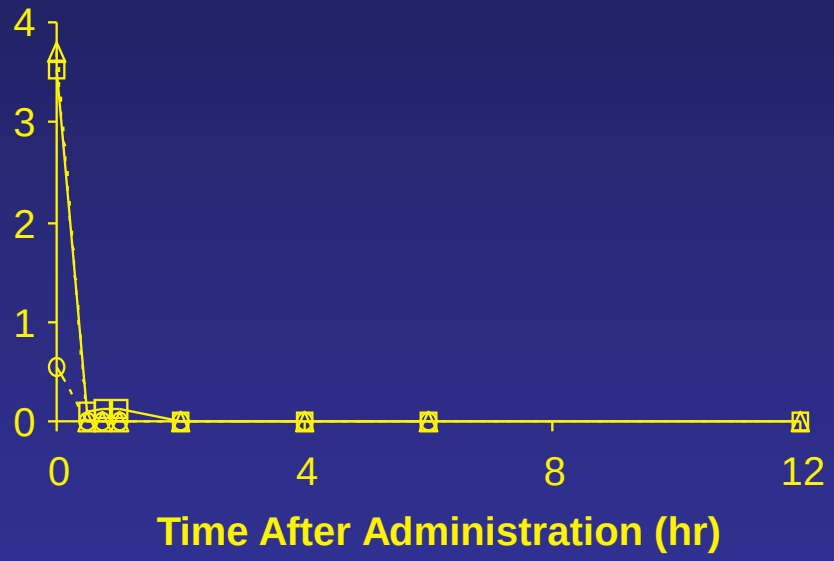
- Recommended dose for combination 40 mg/m^2 carmustine + 120 mg/m^2 O^6-benzylguanine
- Carmustine dose has to be reduced to 20-25% of single agent dose

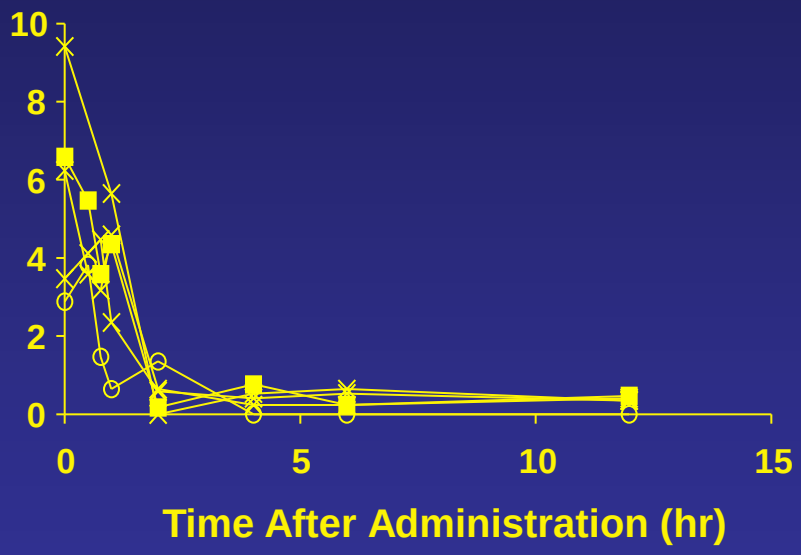
Schilsky et al, Clin Cancer Res 6:3025, 2000

Phase I Trial of 4-BTG & Temozolomide



Depletion of PBMC ATase after IV and PO 4-BTG





ADD of 4-BTG is 10mg/m^2

Phase II dose 4-BTG 40mg/day with temozolomide 125mg/m^2/day

Ransom, Middleton et al, CCR (2006) 12, 1577-84

PARP Inhibitors in Clinical Trials

Agent	Company	Single/Combination therapy	Route of administration	Disease	Clinical status
AG014699 (PF0367338) 2003	Pfizer (New York, NY)	Combination and single agent	I.v. And oral	Solid tumors, melanoma	Phase I + II MM complete TMZ), phase II in BRCA pts open, phase I ongoing
KU59436 (AZD2281) (olaparib) 2005	AstraZeneca/ KuDOS (London, United Kingdom)	Single/ Combination ++	Oral	Various	Phase I complete. Numerous phase II studies
ABT-888 (veliparib) 2006	Abbott Laboratories (North Chicago, IL)	Single/ Combination ++	Oral	Solid tumors and lymphoid malignancies	Phase 0/I completed Numerous phase II studies RT trials
BSI-201 (SAR 240550) 2006	BiPar (Brisbane, CA) (SanofiAventis)	Combination with gem carbo, tmz, RT	I.v.	Triple negative breast cancer	Phase II completed Phase III completed
INO-1001 2005/6	Inotek/ Genentech (Beverly, MA)	Combination with temozolomide, single	I.v.	Melanoma, glioblastoma multiforme	Small phase II in melanoma Reformulation
MK4827 2008	Merck	Single	Oral	Solid, BRCA ovarian	Phase I completed
E7016 (GPI 21016) 2010	Eisai Inc (MGI Pharma)	Combination with temozolomide	Oral	Solid tumors	Phase I ongoing
CEP-9722 2009	Cephalon	Combination with temozolomide	Oral	Solid tumours	Phase I ongoing
LT673 2011	Lead Therapeutics/Biom arin (Novato, USA)	Single agent and combination	oral	Solid tumours	Phase I planned

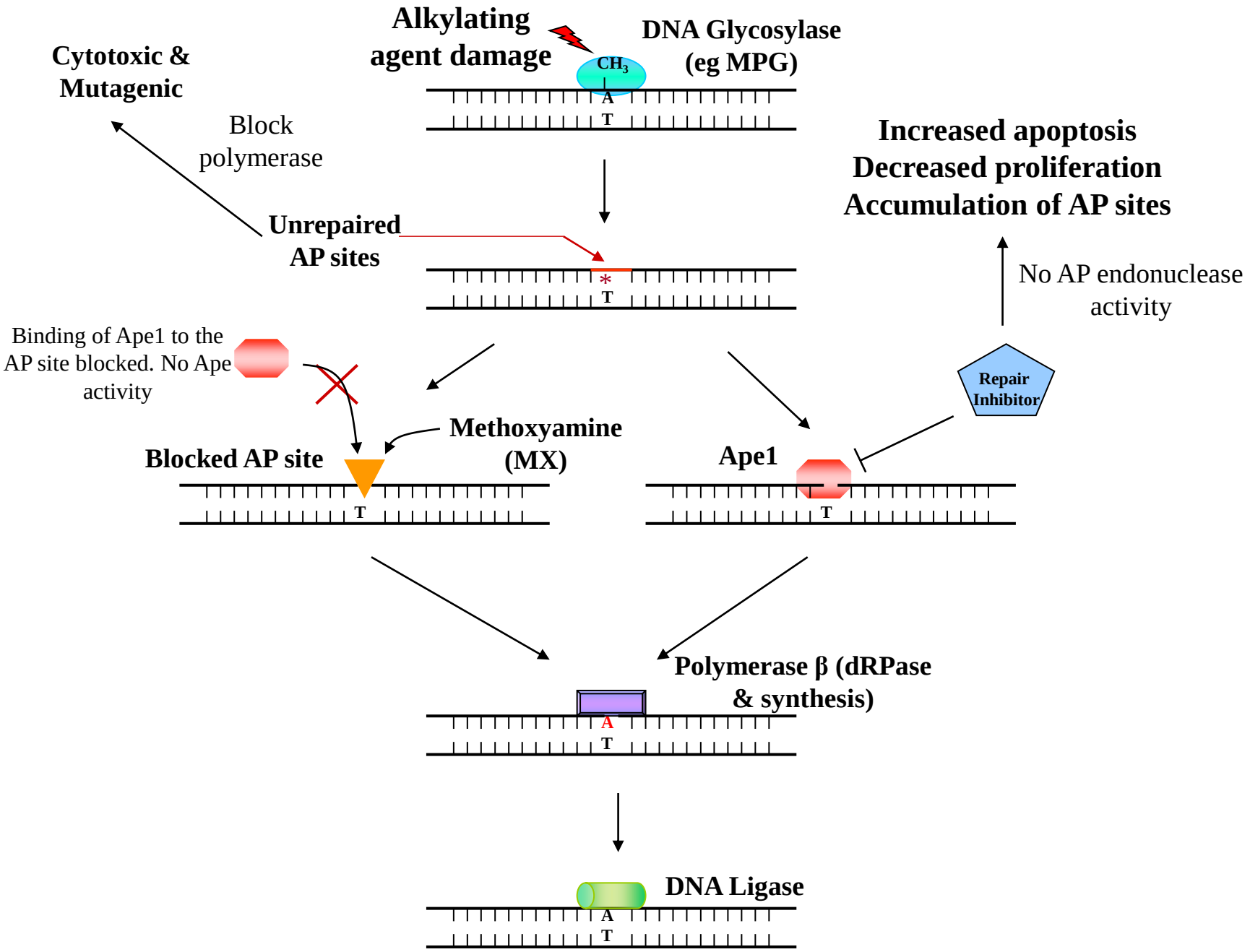
PARP inhibitor chemopotentiation studies

- AG014699 + TMZ – enhanced myelo-suppression (↓ TMZ 25%)
- AZD2281 + gem/cis (PID 400mg bd)
  - Ola 100 mg days 1-4 cis 50 day 3 gem 400 days 3 and 10
  - Ola 100 mg day 1 only cis 50 day 1 gem 500 day 1 and 8
- ABT888 + topotecan
  - PID in combination with topo 0.6 mg/m^2 tolerable (d1-5)

- Outlier = BSI-201 gem/carbo or TMZ – no enhancement of myelosuppression

NOVEL TARGETS IN DEVELOPMENT - BER

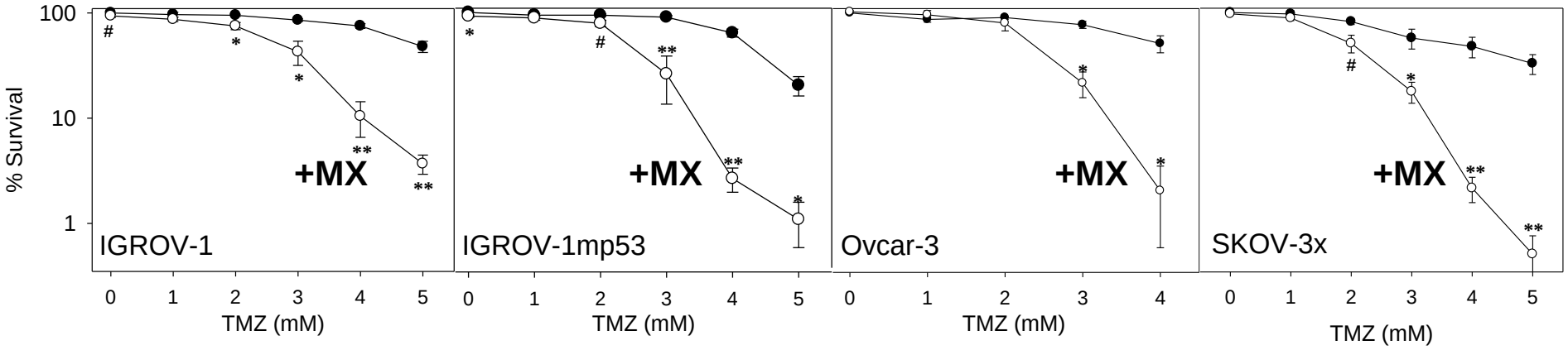
Consequences of inhibiting Ape1 and BER



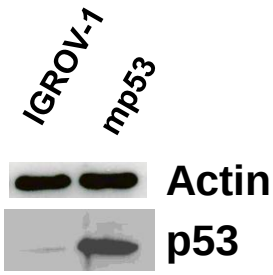
Treatment of four ovarian cancer cell lines with TMZ and MX

(MTS assay)

A.

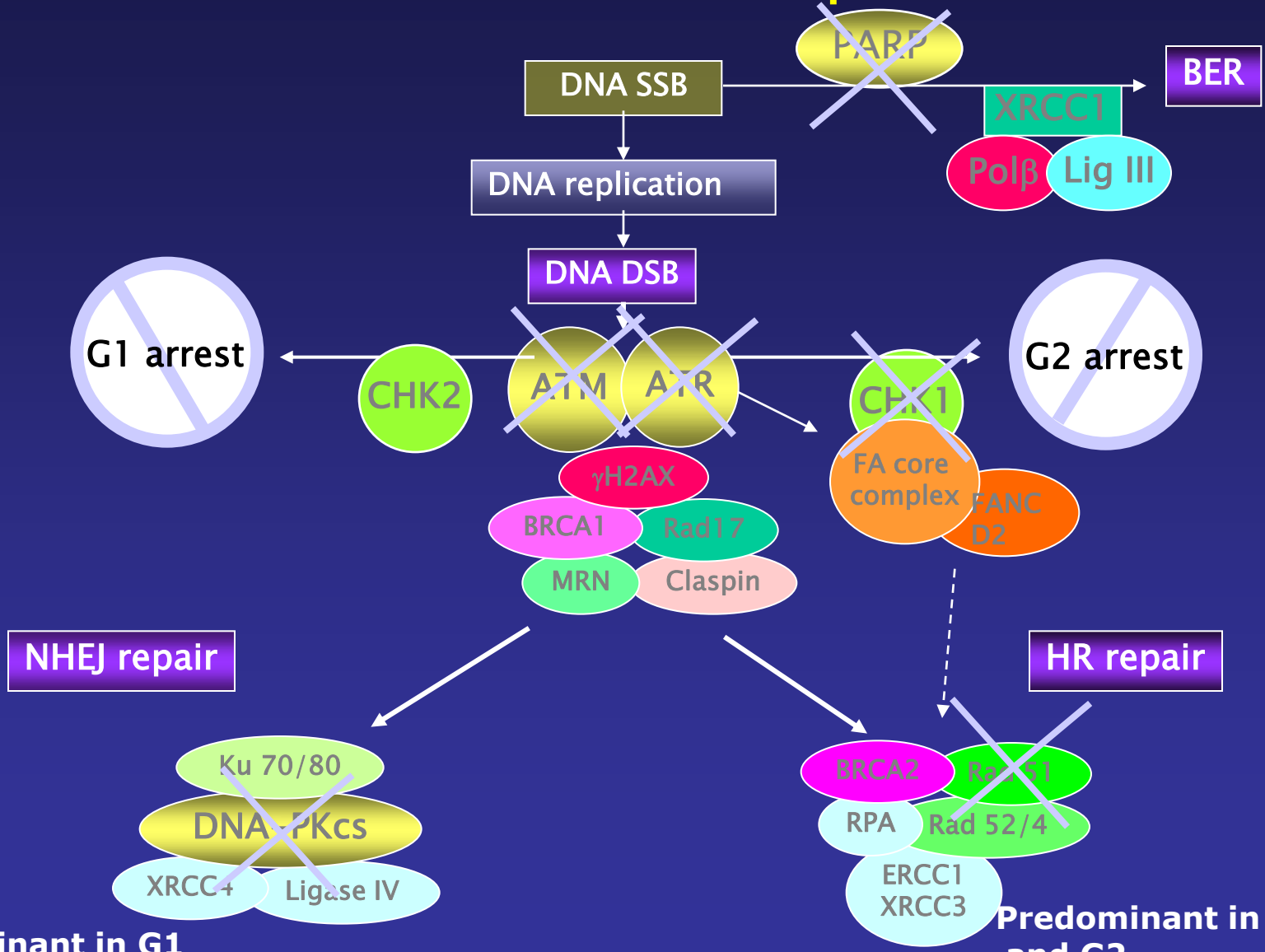


B.

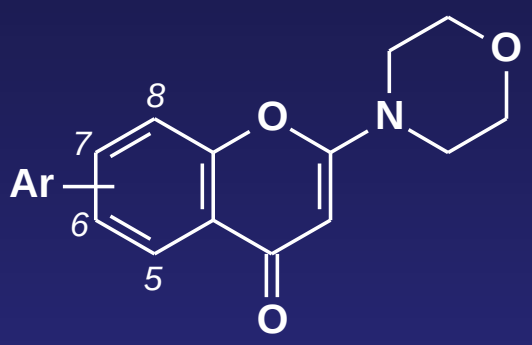


NOVEL TARGETS IN DEVELOPMENT – DSB REPAIR

Multiple targets of DNA Double and single strand break repair

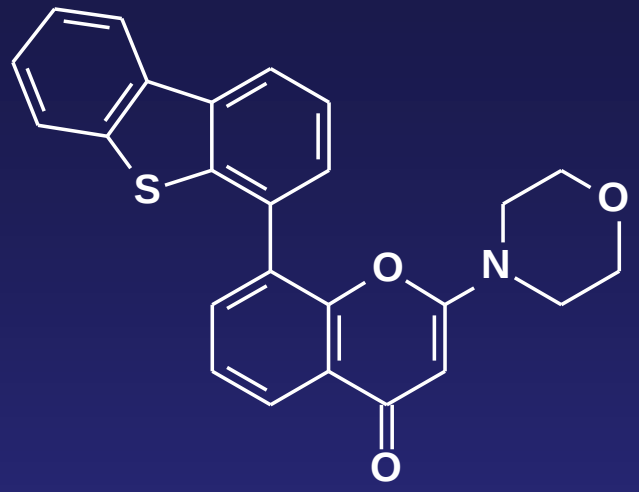


DISCOVERY OF THE DNA-PK INHIBITOR NU7441



Promising leads

Library Synthesis



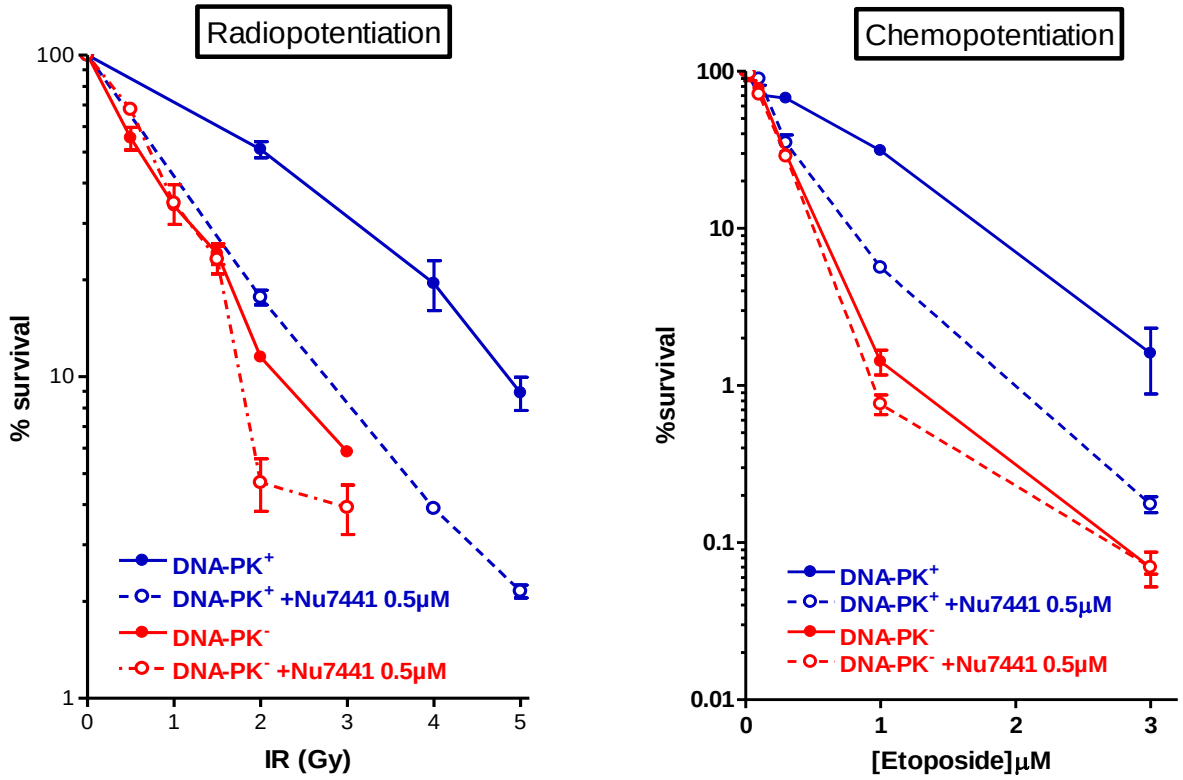
NU7441

DNA-PK IC_{50} (μM)	ATR IC_{50} (μM)	ATM IC_{50} (μM)	PI 3-K IC_{50} (μM)	PI 4-Kβ IC_{50} (μM)	mTOR IC_{50} (μM)
0.012	>100	>100	5	40	1.7

No activity seen at 10 mM in a screen against 60 diverse kinases

Leahy *et al* (2004) *Bioorg. Med. Chem. Lett.* **14**, 6083-6087.
Hardcastle *et al* (2005) *J. Med. Chem.* **48**, 7829-46 .

Cellular specificity of NU7441 for DNA-PK

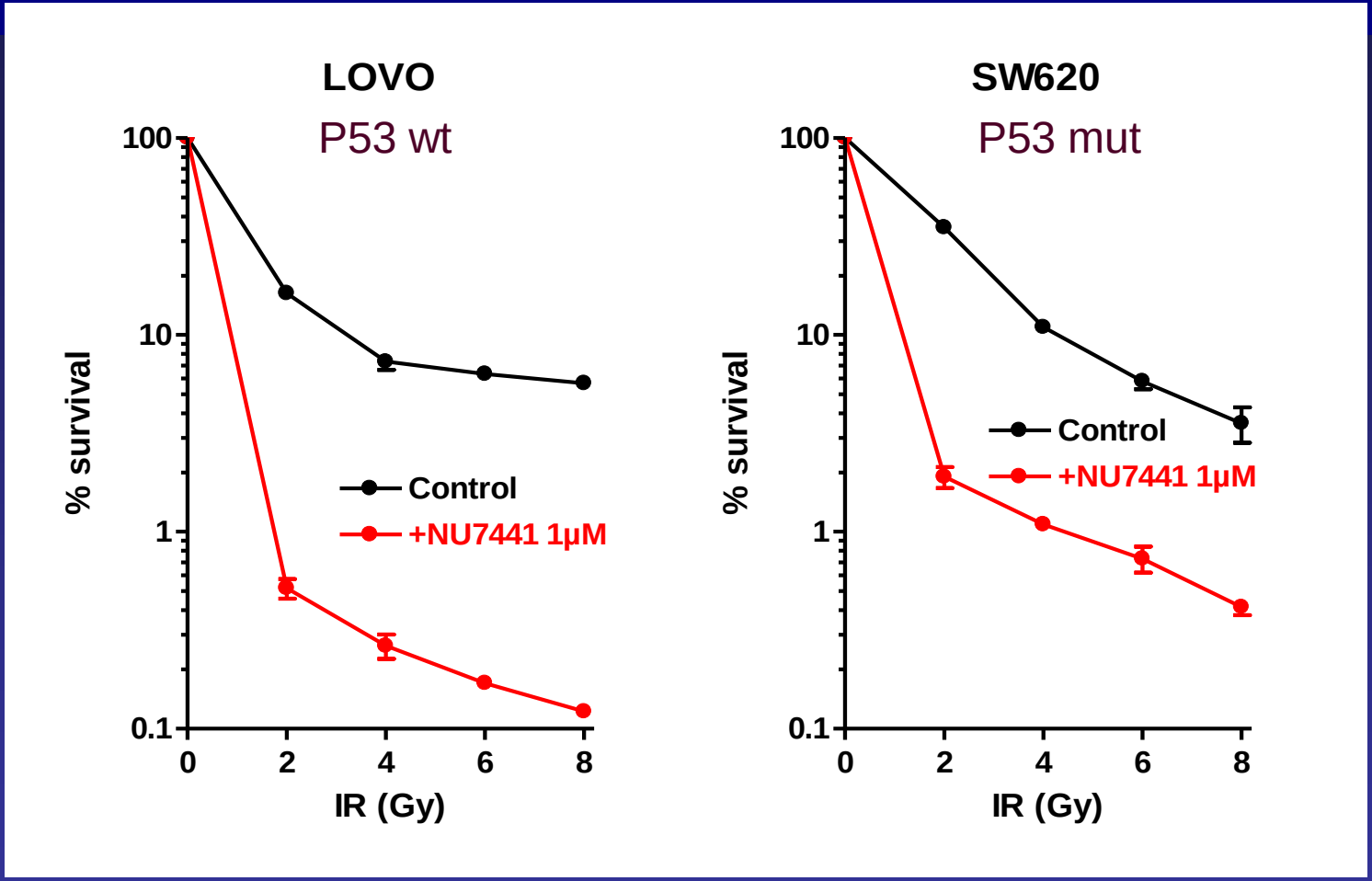


Cells were exposed to drugs for 16 hr prior to seeding for colony formation.

DNA-PK$^-$ cells (V3) are inherently more sensitive to IR and etoposide than DNA-PK$^+$ cells (V3-Yac). NU7441 potentiates IR and etoposide cytotoxicity in DNA-PK$^+$ but not DNA-PK$^-$ cells confirming that DNA-PK is the cellular target of NU7441

Nicola Curtin and Yan Zhao

Radiopotentiation of human p53 wt and mutant colon cancer cells



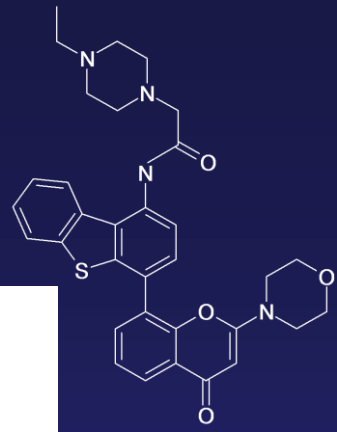
	Dose Modification Ratio (DMR)	
	LOVO	SW620
2 Gy	32	19
6 Gy	38	8.2

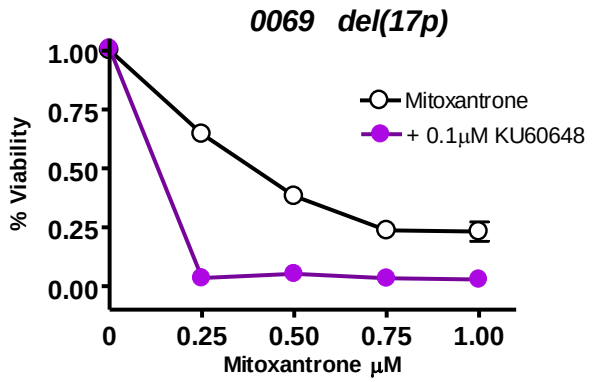
Nicola Curtin

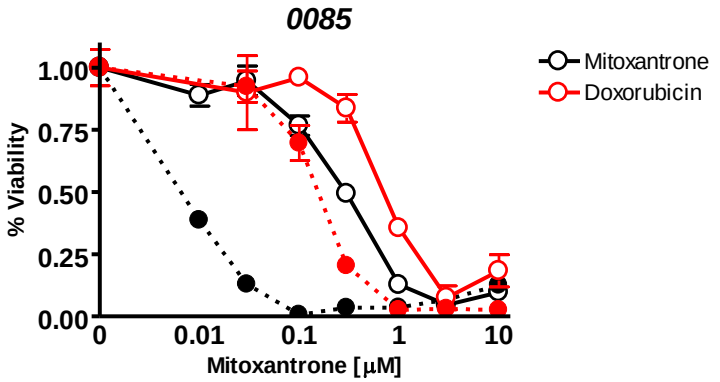
CLL data

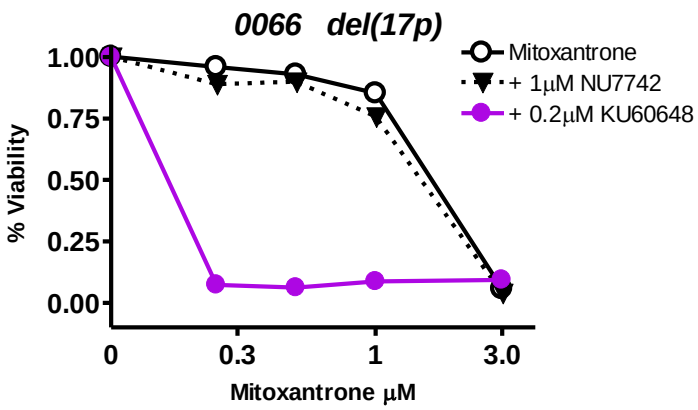
- Over-expression DNA PK in del 17q cases
- Correlates with drug resistance
- Re-sensitisation by DNA-PK inhibitor

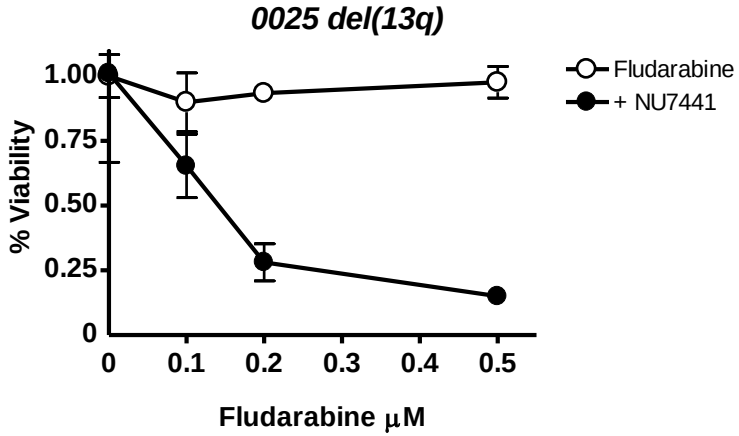
DNA-PK inhibition chemosensitises del(17p), p53 mutated cases







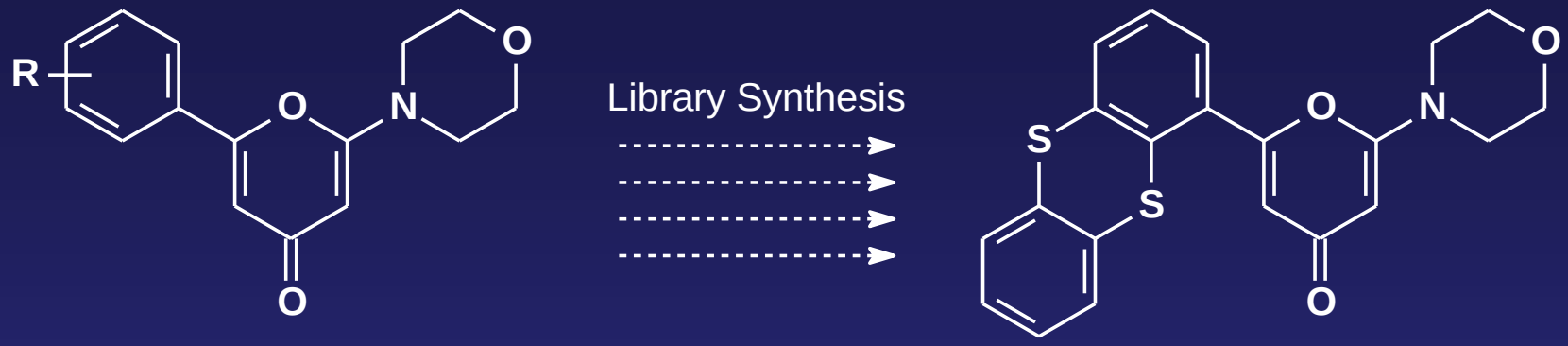




Potential Phase I trial design for DNA PKi

- Oral clinical candidate potentially to enter phase I trials in combination with doxorubicin
- MTD and "TID" (target inhibitory dose) to be defined – ex-vivo assay
- PD clinical assay to be validated in PBLs from an established pre-clinical assay
- Measurement of the ability of DNA PK to phosphorylate ser15 on n66p53

IDENTIFICATION OF THE ATM INHIBITOR KU-0055933



Weak ATM inhibition

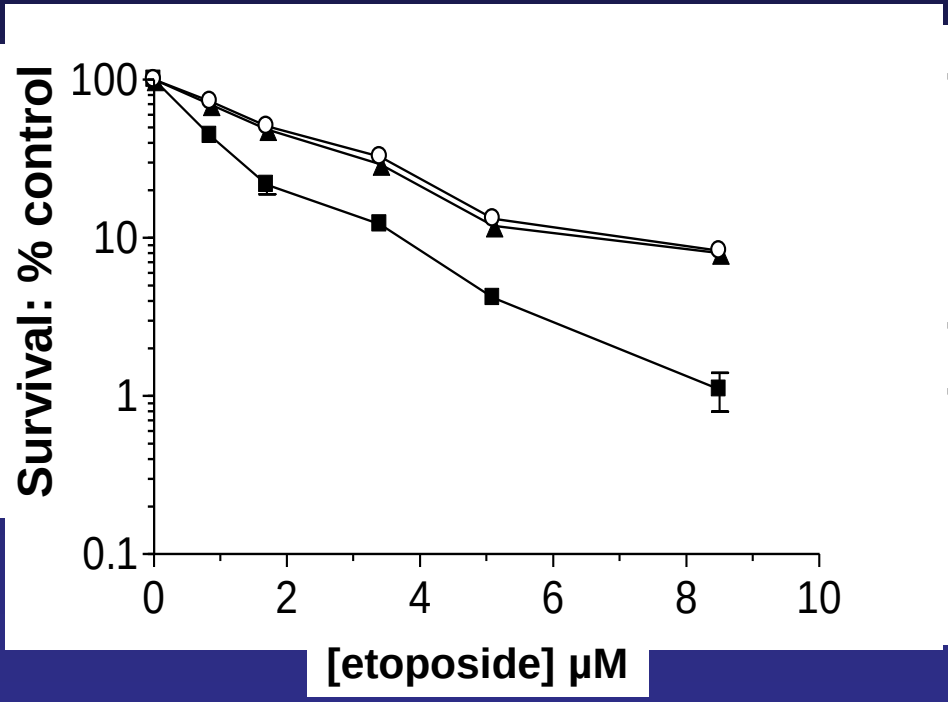
KU-0055933

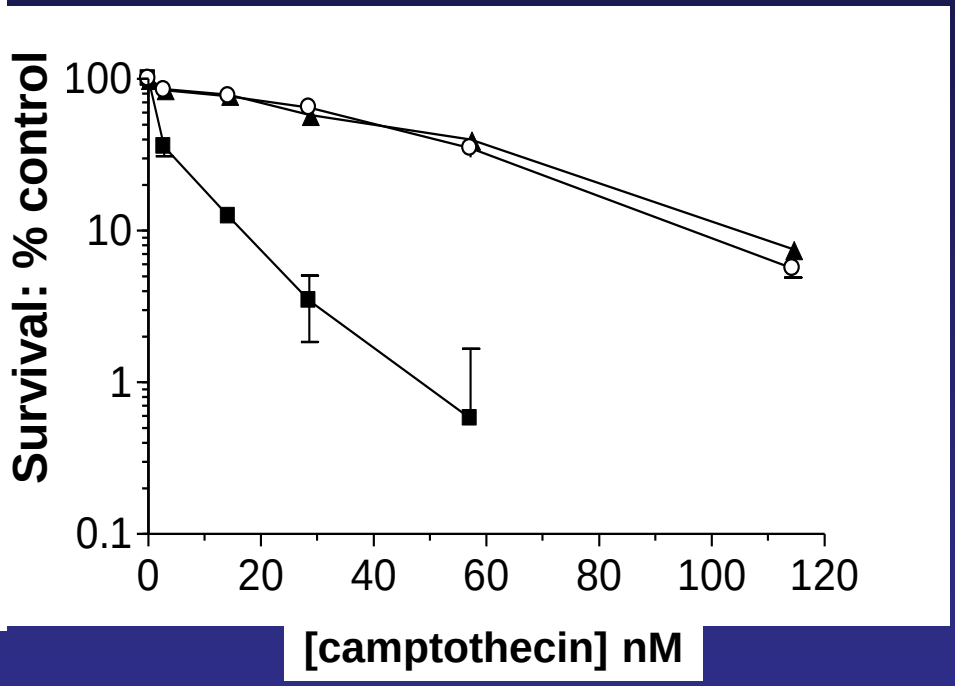
ATM IC_{50} (μM)	ATR IC_{50} (μM)	DNA-PK IC_{50} (μM)	PI 3-K IC_{50} (μM)	PI 4-Kβ IC_{50} (μM)	mTOR IC_{50} (μM)
0.013	>100	2.0	25	20	8

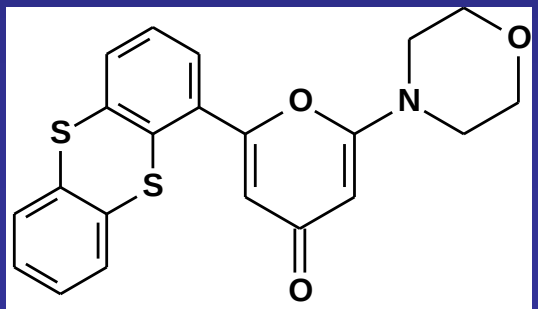
Little or no activity seen in a 60 kinase screen at 10 mM

Hickson *et al* (2004) *Cancer Res.* **64**, 9152-9159.

Sensitization of HeLa cells to etoposide and camptothecin by ATM inhibition

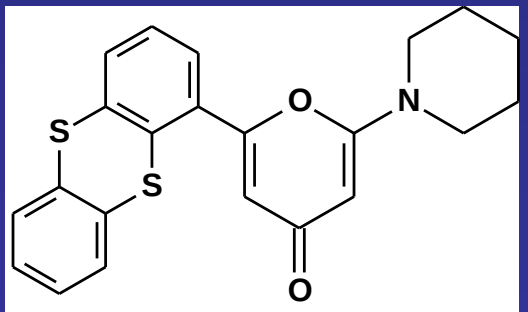






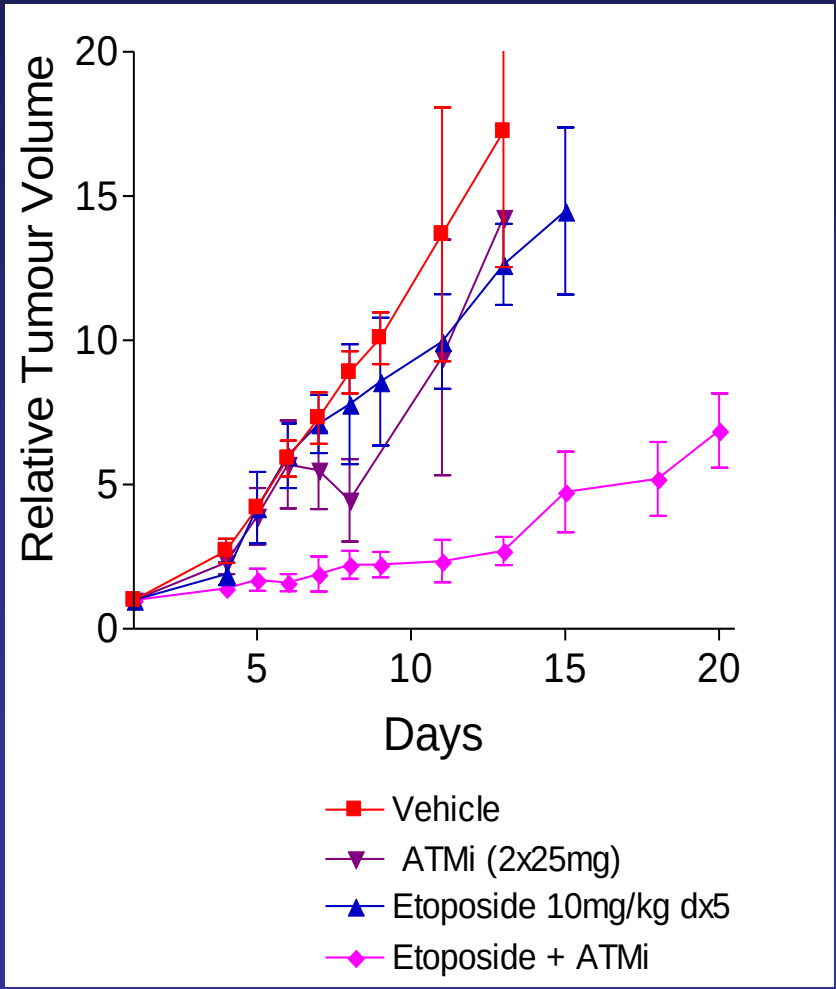
KU-0055933
IC_{50} = 13 nM

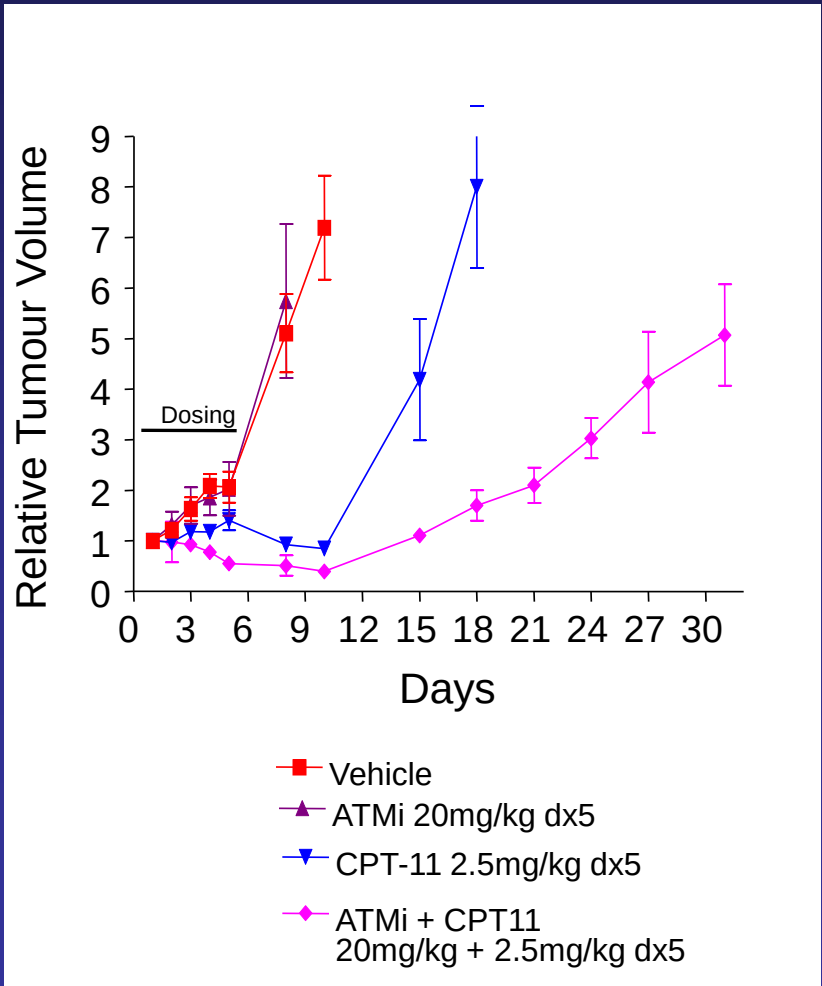
control
10 μM KU-55933
10 μM KU-58050



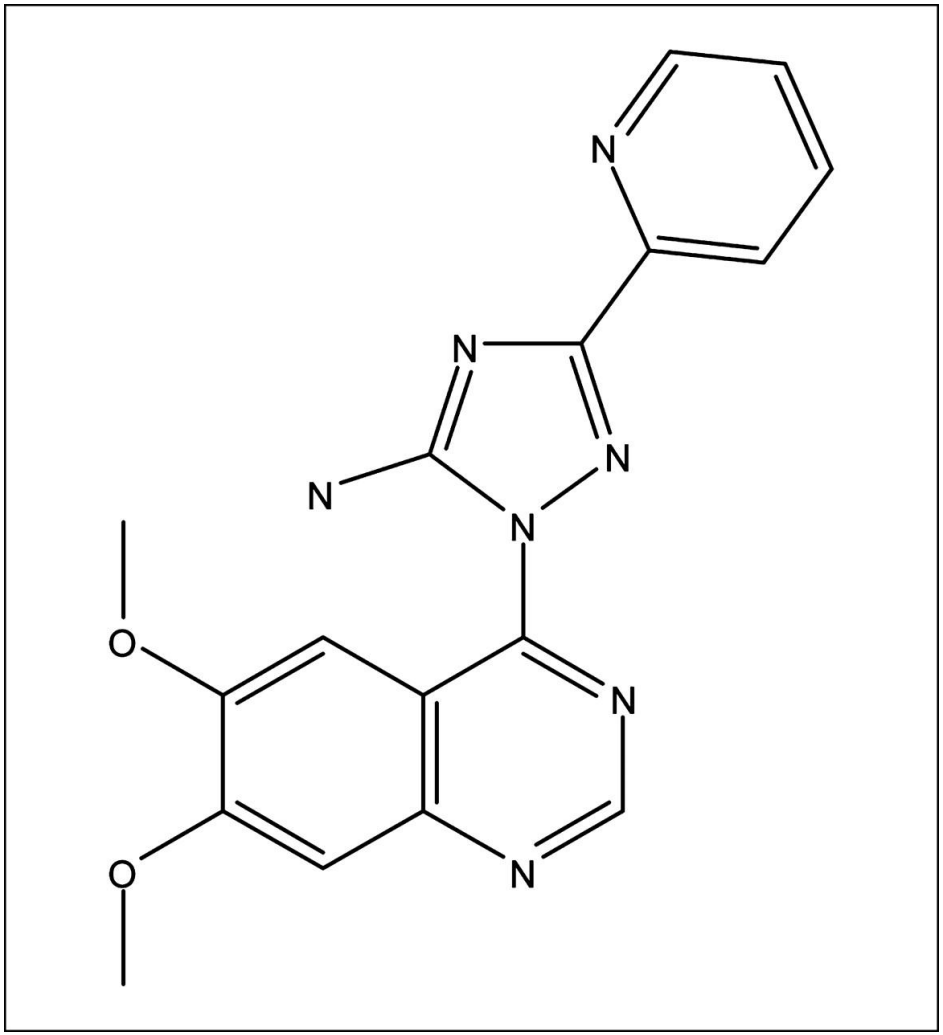
KU-0058050
IC_{50} = 3.0 μM

Effect of ATMi in combination with etoposide or irinotecan in the SW620 xenograft model





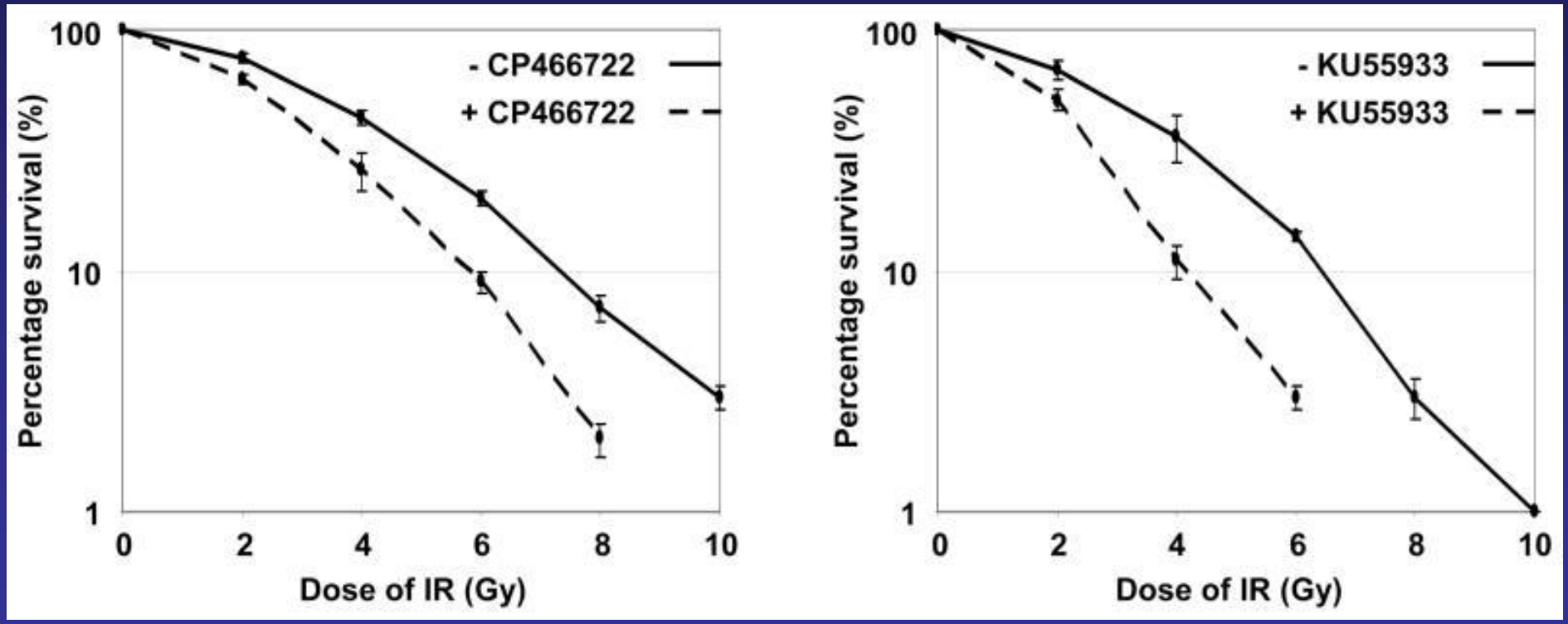
ATM inhibitors in pre-clinical development



Chemical structure of CP466722 [2-(6,7-dimethoxyquinazolin-4-yl)-5-(pyridin-2-yl)-2H-1,2,4-triazol-3-amine].

Rainey M D et al. Cancer Res 2008;68:7466-7474

ATMi and radiosensitisation



Rainey et al Cancer Research 68 (2008)

Pharmacodynamic markers for ATM

- ATM recruited to and signals DNA DSB by phosphorylation of key proteins
- 2 candidate biomarkers:
  - phosphorylation of γH2AX as marker of ATM activation
  - RAD51 foci formation as an indication of active HR
- Challenge remains HR active in cells in S phase –
  - Hair follicles, skin punch biopsies, CTCs, leukaemic cells

ATRi – NU6027

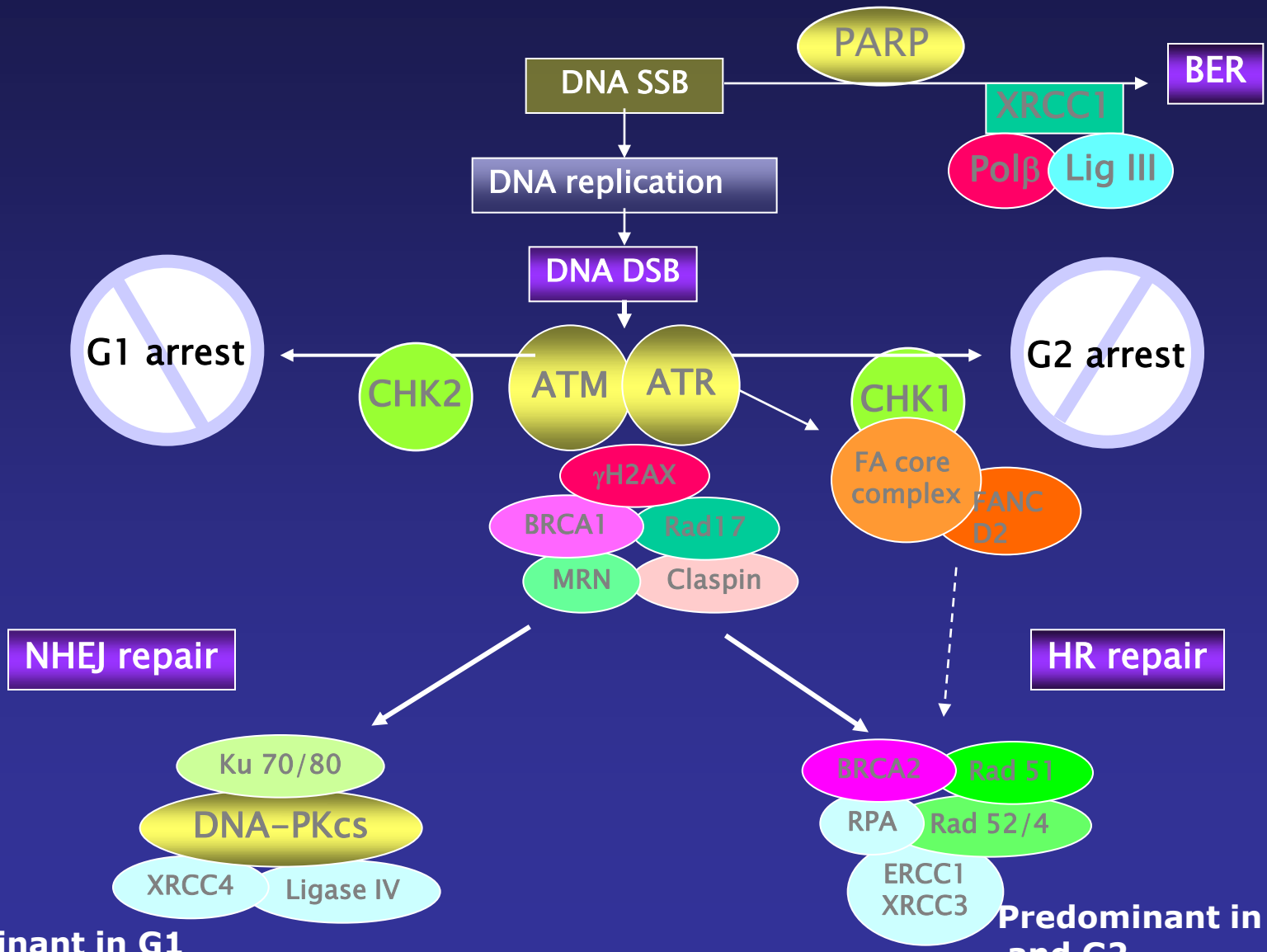
- ATR inhibitor – IC_{50} < 5μM
- Potentiates the cytotoxicity of hydroxyurea, cisplatin, temozolomide, doxorubicin and camptothecins
- CM847-KD cells
  - ATR inhibition (via induction of kinase dead or chemical inhibition) increases cytotoxicity of AG014699 – synthetic lethality by dual inhibition
- ATR inhibitors are in late preclinical development and likely to enter clinic in 2011

THOUGHTS ON TRIAL DESIGN AND POSSIBLE COMBINATIONS

Trial design with DDRi

- Combination with DNA damaging agent – as potentiator
  - Chemotherapy
  - Radiotherapy
- Synthetically lethal use in appropriate tumour type
  - Molecular biomarkers needed to ensure a therapeutic index
- Combination with other active single agents
  - Scheduling!!!!!

Are combinations of DDRi possible?



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CANCER RESEARCH UK