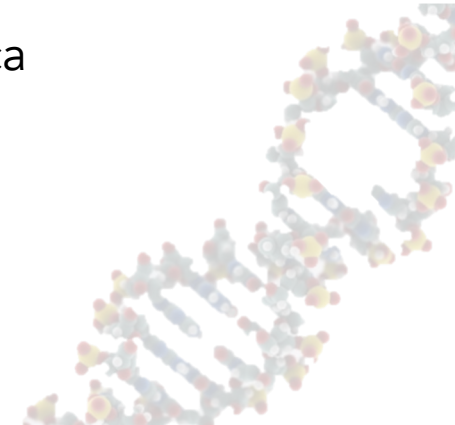
# LA CELLULA STAMINALE DEL CANCRO

# Evoluzione darwiniana del tumore: non solo frequenza di mutazione

Altri meccanismi contribuiscono alla trasformazione multi-step e complicano il modello e le tempistiche attese:

- Cambiamenti epigenetici
- Perdita oncosoppressore richiede 2 eventi mutazionali
- Mutazioni driver e mutazioni passenger

- Ambiente
- Variazione frequenza di mutazione in mutanti instabilità genetica
- Presenza di cellule diverse nel tumore



Weinberg, la Biologia del cancro

# Cellule staminali nella leucemia mieloide acuta (AML)

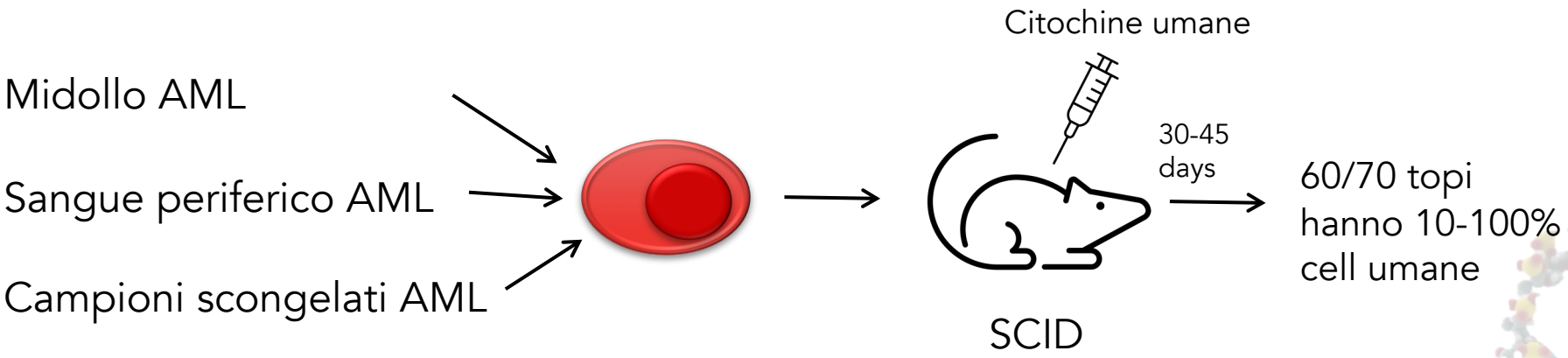
**A cell initiating human acute myeloid leukaemia after transplantation into SCID mice**

**Tsvee Lapidot, Christian Sirard, Josef Vormoor, Barbara Murdoch, Trang Hoang*, Julio Caceres-Cortes*, Mark Minden†, Bruce Paterson‡, Michael A. Caligiuri§ & John E. Dick‖**

NATURE VOL 367 · 17 FEBRUARY 1994

Cellule leucemiche umane in topo → Non sempre inducono AML → Perchè?

1Q: cellule ematopoietiche dipendono fortemente da citochine → trattamento con citochine favorisce tumorigenesi nel topo




Lapidot et al., 1994

# Cellule staminali nella leucemia mieloide acuta (AML)

Midollo AML

Sangue periferico AML

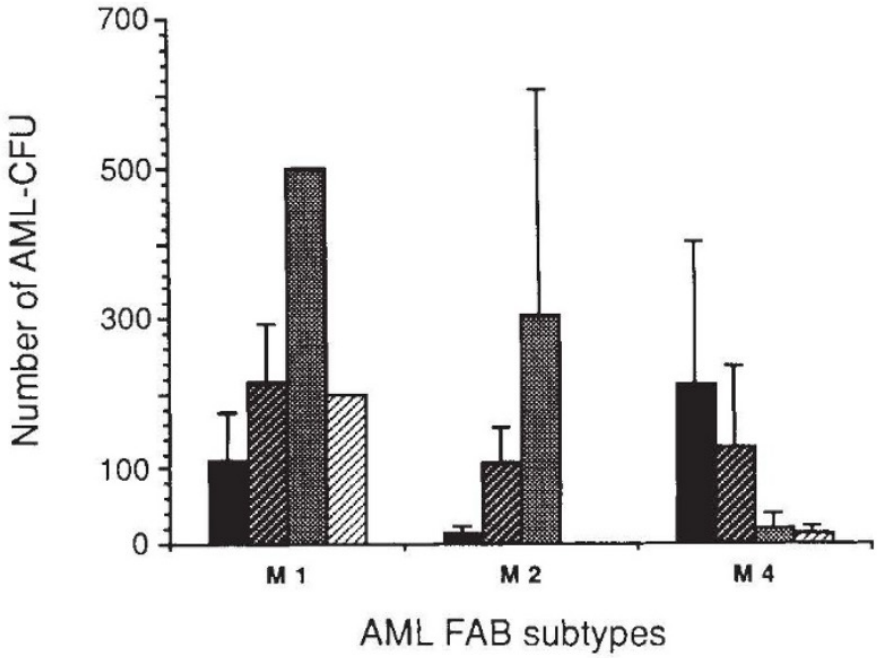
Campioni scongelati AML

Citochine umane

SCID

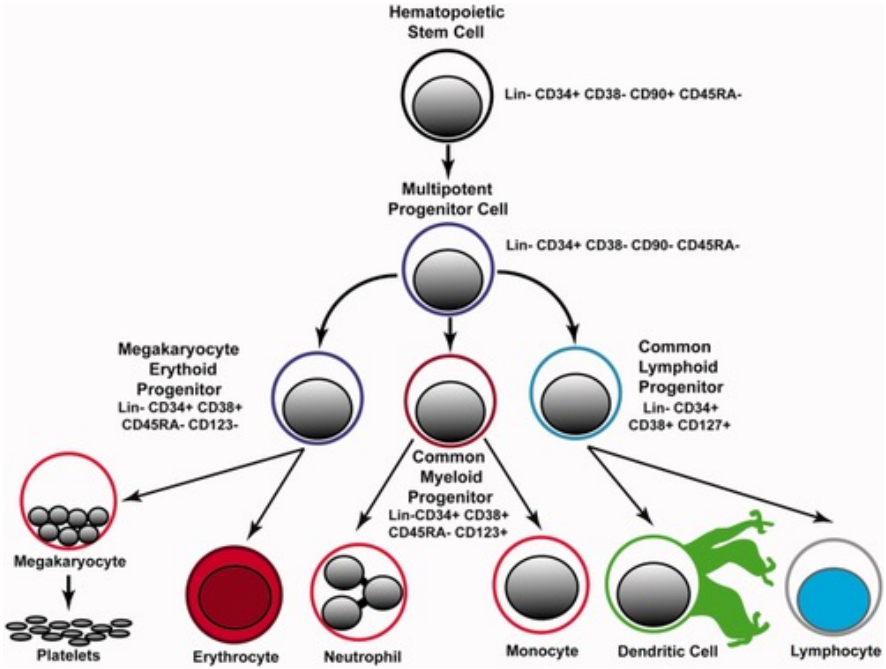
30-45 days

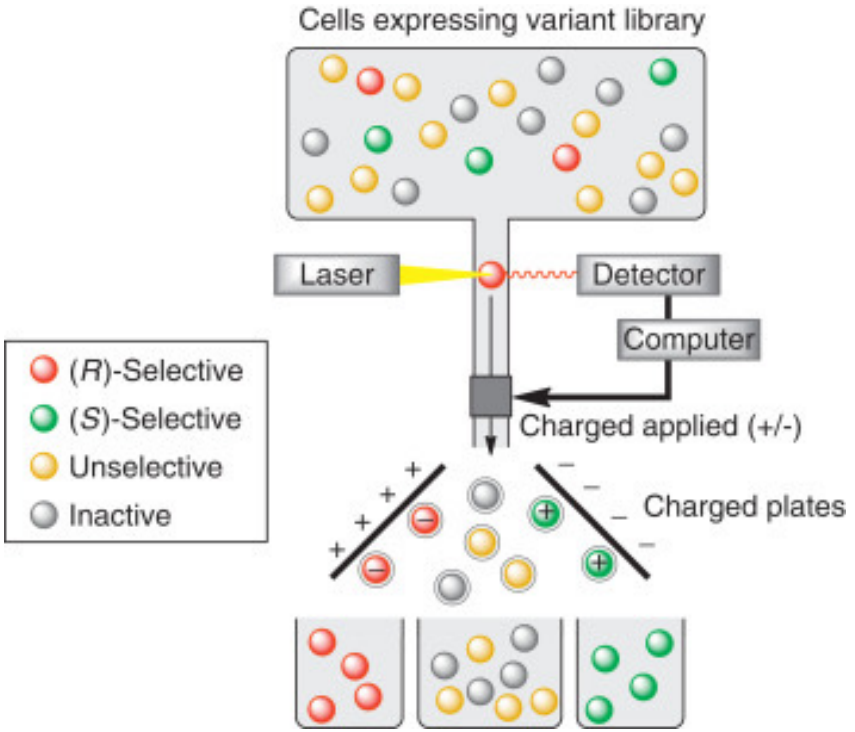
60/70 topi hanno 10-100% cell umane




Lapidot et al., 1994

# Cellule staminali nella leucemia mieloide acuta (AML)



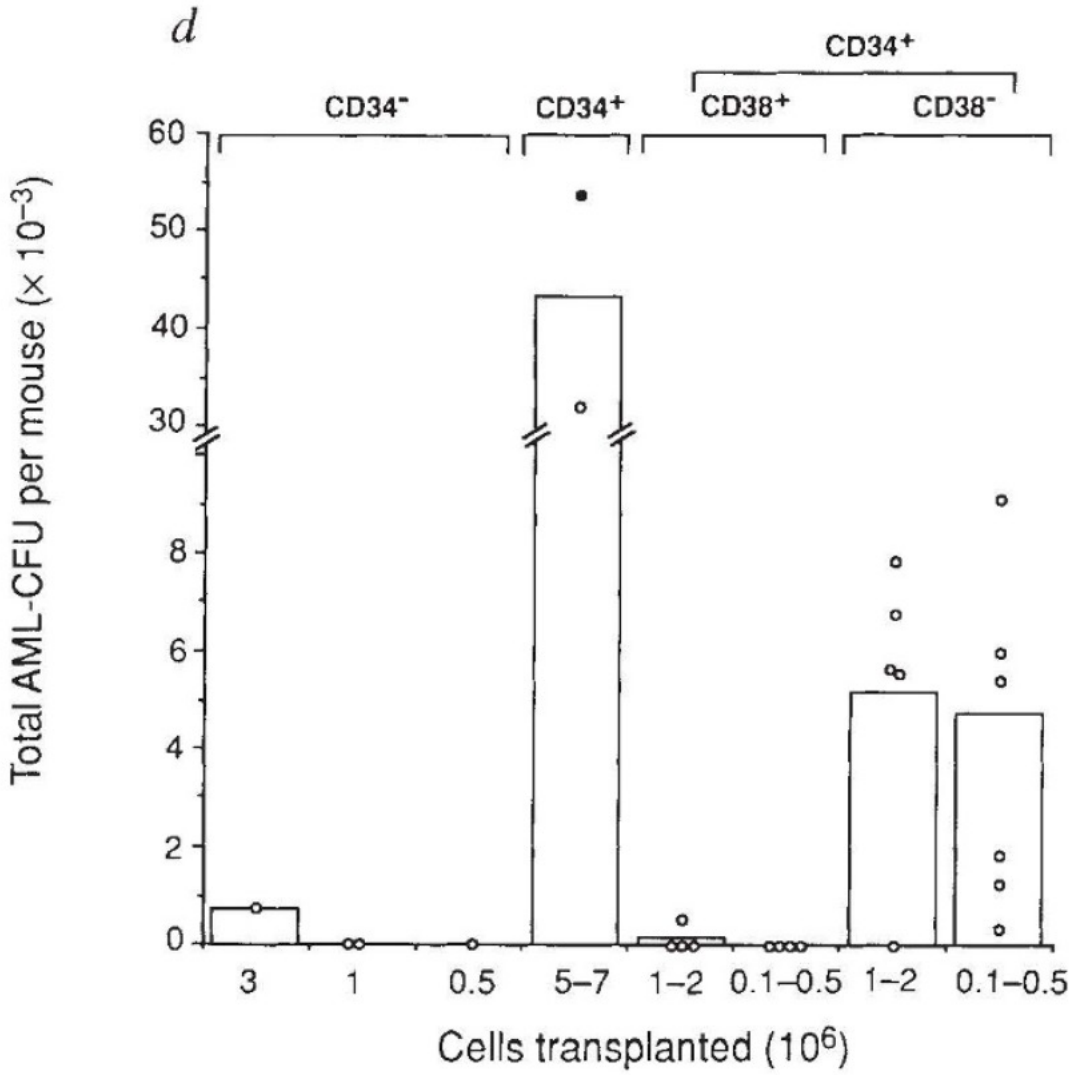




Cellule leucemiche umane in topo → quali sono responsabili del tumore?

- Separare cellule tumorali in base a marcatori di superficie
- Reinoculare cellule isolate in topi immunodeficioenti
- Anlizzare la comparsa del tumore

Wolkowicz, 2013; Lapidot et al., 1994; Cimato et al., 2015

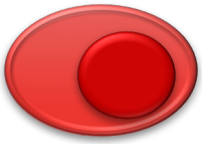
# Cellule staminali nella leucemia mieloide acuta (AML)




Lapidot et al., 1994

# Cellule staminali nella leucemia mieloide acuta (AML)

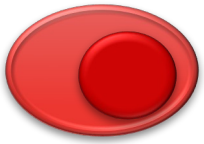
$CD34^+$ $CD38^-$



Poco abbondanti (1% cellule neoplastiche) e poco differenziate

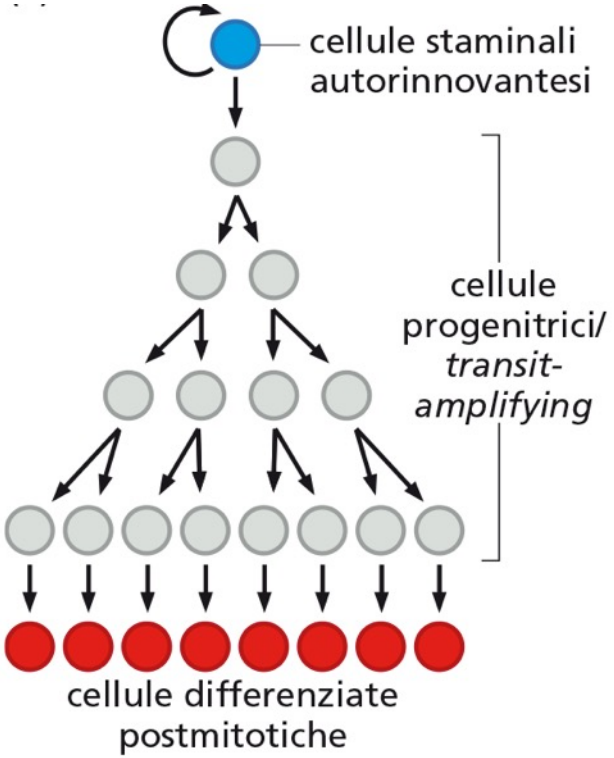
Si espandono in fretta e formano tumori in topo

$CD34^+$ $CD38^-$



Abbondanti e parzialmente differenziate

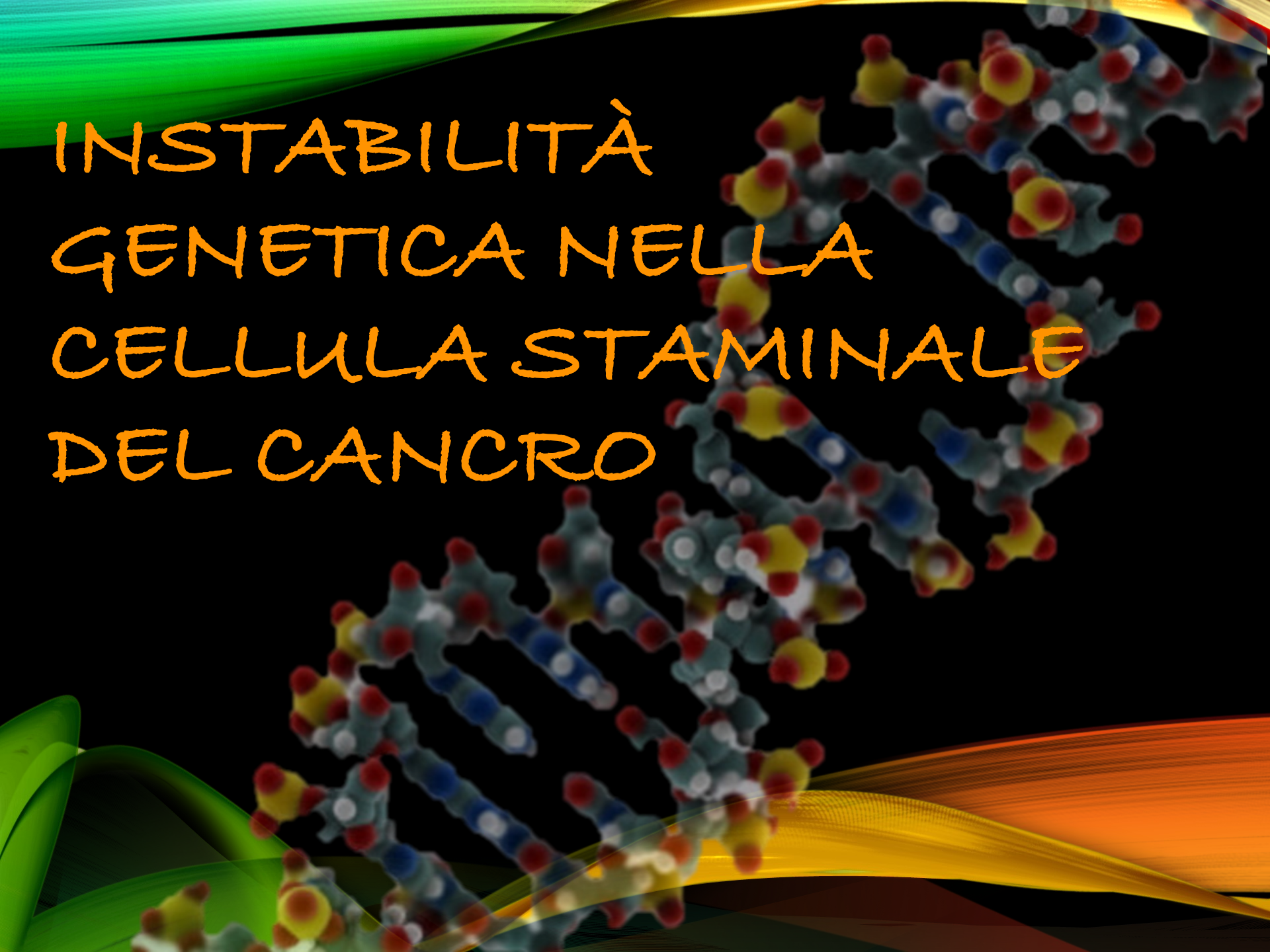
NON formano tumori in topi



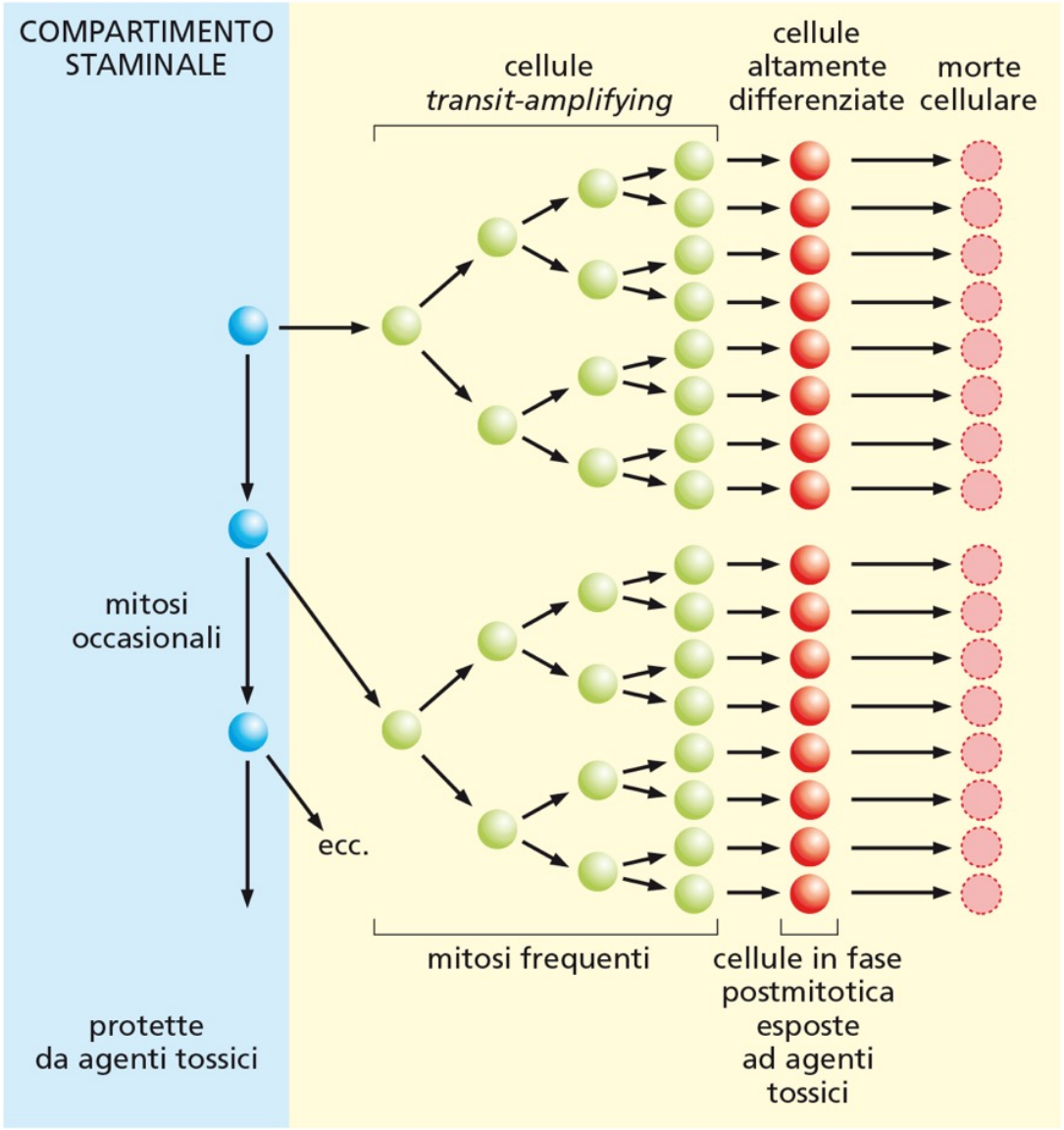

Solo poche cellule nel tumore hanno la capacità di autorinnovarsi e sono fortmente tumorigeniche → Cellula staminale del cancro (CSC)

Wolkowicz, 2013; Lapidot et al., 1994

# INSTABILITÀ GENETICA NELLA CELLULA STAMINALE DEL CANCRO

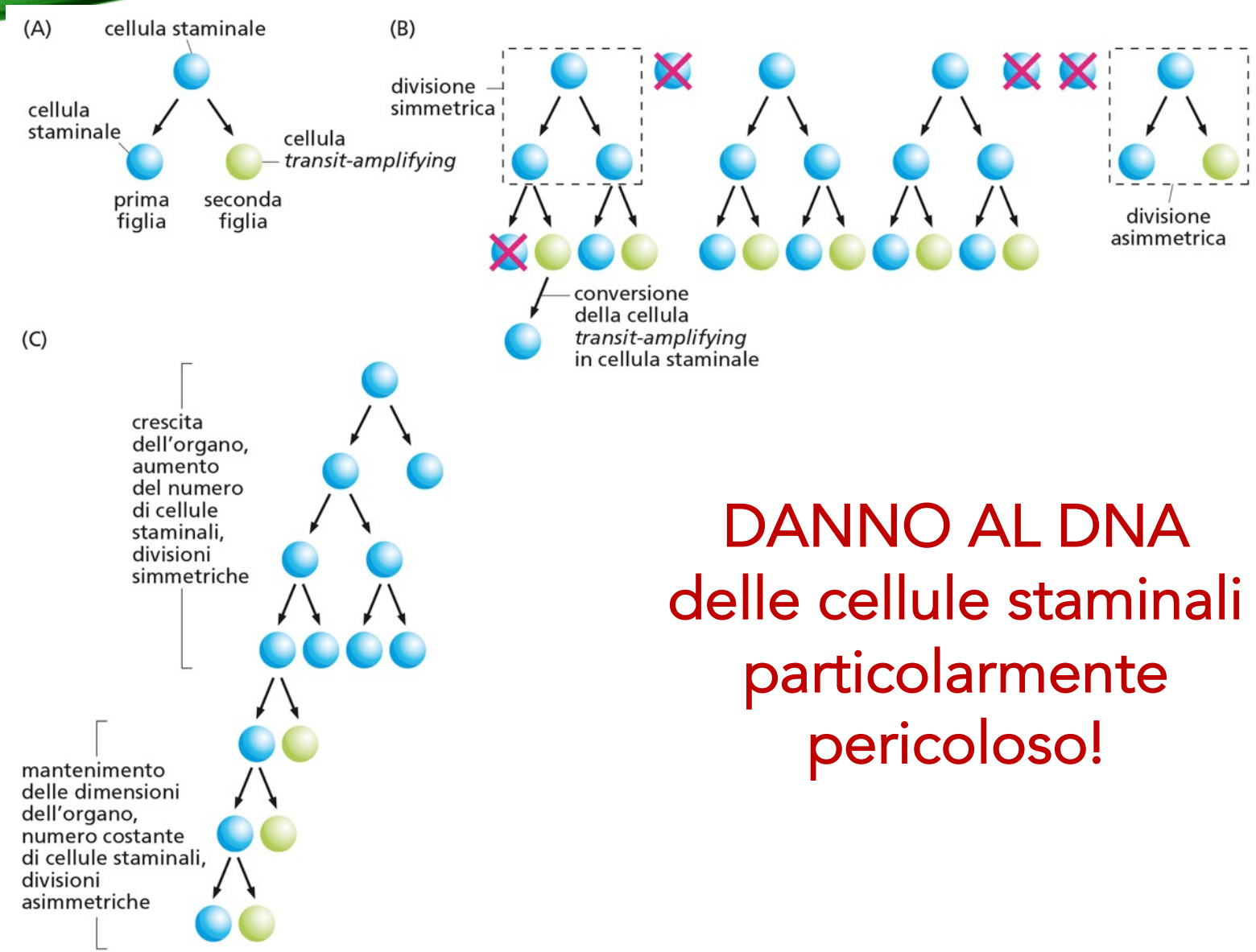


# Cellule staminali nei tessuti




Weinberg, La biologia del cancro

# Cellule staminali nei tessuti




DANNO AL DNA
delle cellule staminali
particolarmente
pericoloso!

Weinberg, La biologia del cancro

# LETALITÀ E CITOTOSSICITÀ SINTETICHE NEL CANCRO

# Interazioni genetiche

Combinazioni di mutazioni multiple che generano un fenotipo inatteso

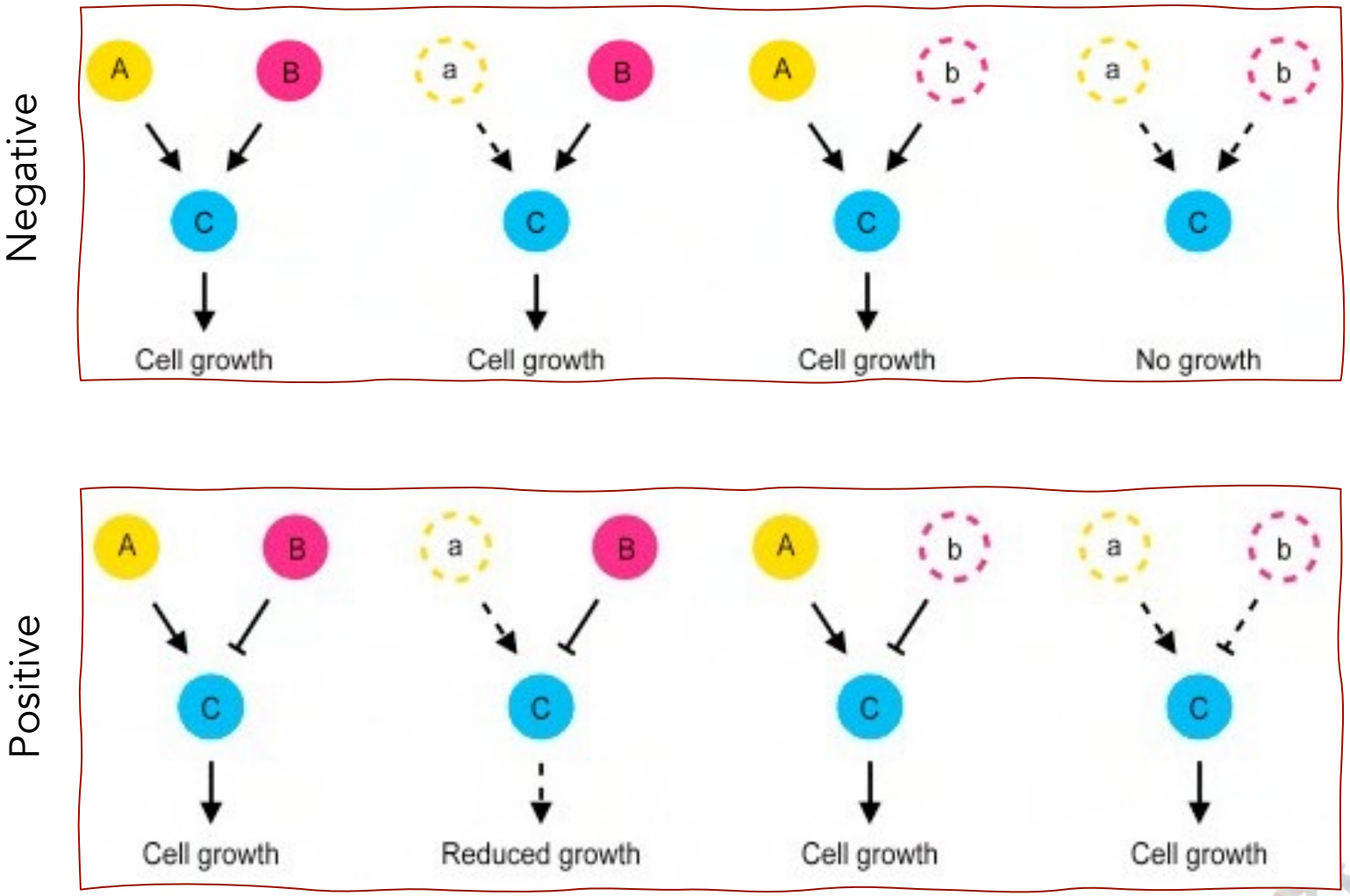
**SOPPRESSIONE:** recuperare l'effetto di una mutazione con una seconda mutazione

**PEGGIORAMENTO SINTETICO:** esacerbare l'effetto di una mutazione con una seconda mutazione

Interazione genetica **POSITIVA**

Interazione genetica **NEGATIVA**

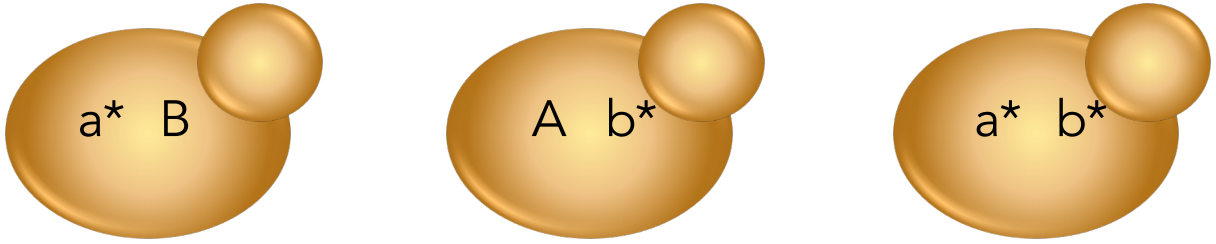
# Interazioni genetiche positive e negative




Van Leeuwen et al., 2017

# Letalità sintetica

Mutazioni simultanee in due geni (A e B) causano morte cellulare ma non hanno effetto singolarmente




viable viable dead

La letalità sintetica è stata inizialmente descritta in *Drosophila* e in lievito

# Letalità sintetica nei tumori

## Integrating Genetic Approaches into the Discovery of Anticancer Drugs

Leland H. Hartwell, Philippe Szankasi, Christopher J. Roberts, Andrew W. Murray, Stephen H. Friend*

The discovery of anticancer drugs is now driven by the numerous molecular alterations identified in tumor cells over the past decade. To exploit these alterations, it is necessary to understand how they define a molecular context that allows increased sensitivity to particular compounds. Traditional genetic approaches together with the new wealth of genomic information for both human and model organisms open up strategies by which drugs can be profiled for their ability to selectively kill cells in a molecular context that matches those found in tumors. Similarly, it may be possible to identify and validate new targets for drugs that would selectively kill tumor cells with a particular molecular context. This article outlines some of the ways that yeast genetics can be used to streamline anticancer drug discovery.

L'instabilità genetica del tumore lo rende vulnerabile

Combinare una mutazione tumore-specifica necessaria per lo sviluppo del tumore con l'inibizione farmacologica di un pathway compensativo

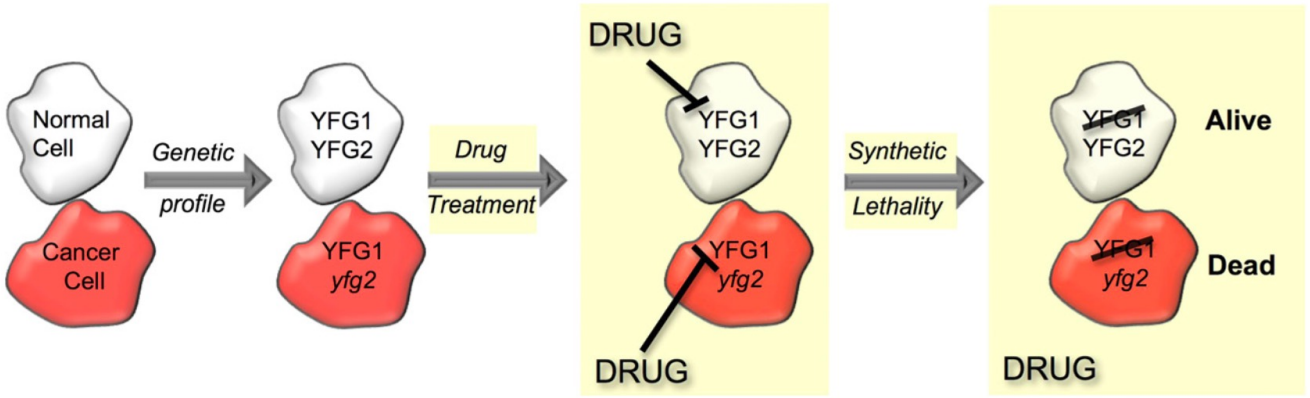
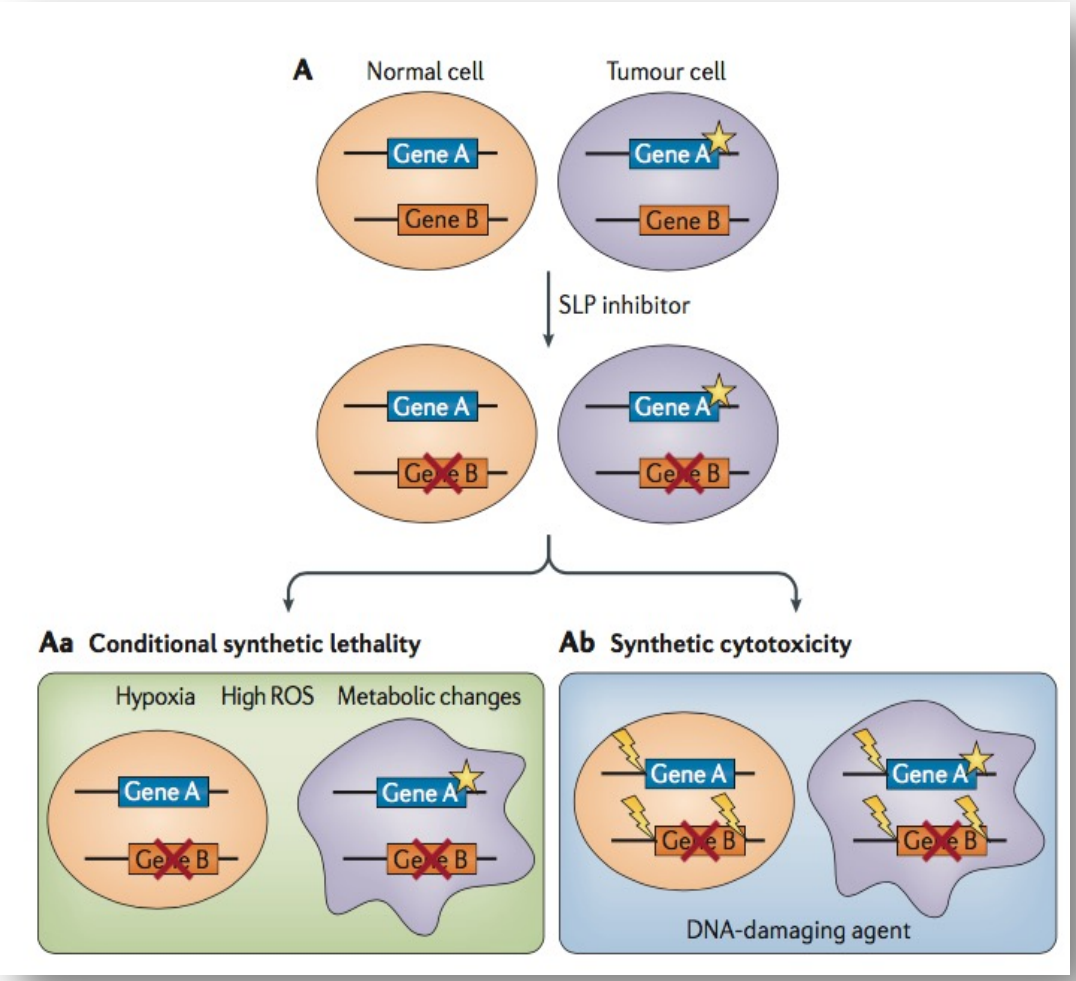



**Fig. 2 – Synthetic lethality in chemotherapy. Differently from healthy cells, cancer cells are characterised by mutations; in the figure, *yfg2* represents the cancer mutation (YFG: your favourite gene). If YFG1 and YFG2 represent a SL-pair, a drug that inhibits YFG1 can selectively damage the tumour cells, without affecting the normal cells.**

# Letalità e citotossicità sintetiche



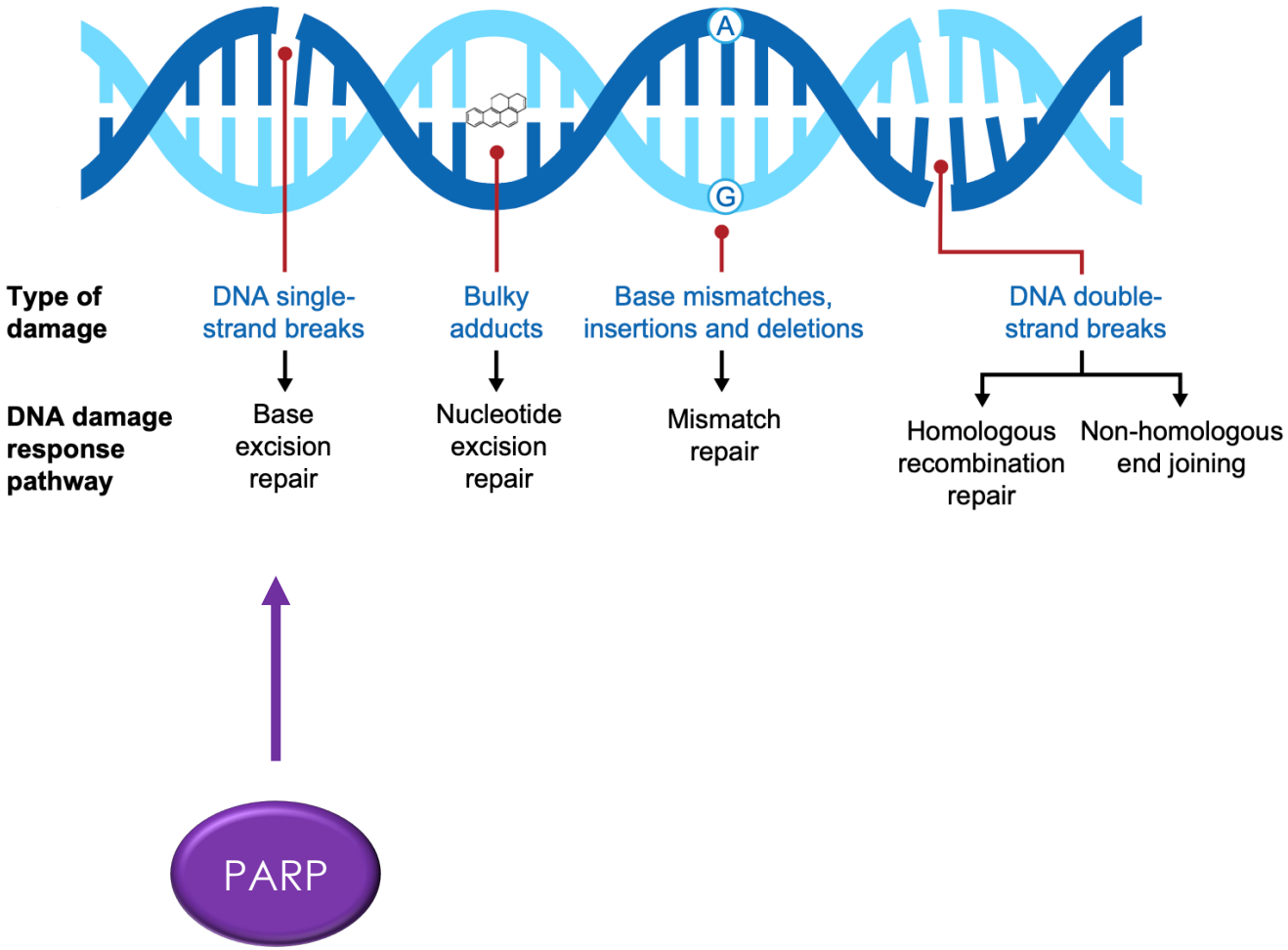

La combinazione di una mutazione tumore-specifica e un inibitore farmacologico aumenta la sensibilità delle cellule tumorali a un agente che danneggia il DNA

Letalità sintetica

Citotossicità sintetica

O'Neil et al., 2017; Lavin and Yeo, 2020

# Inibitori di PARP e tumori al seno




Cortesi et al., 2001

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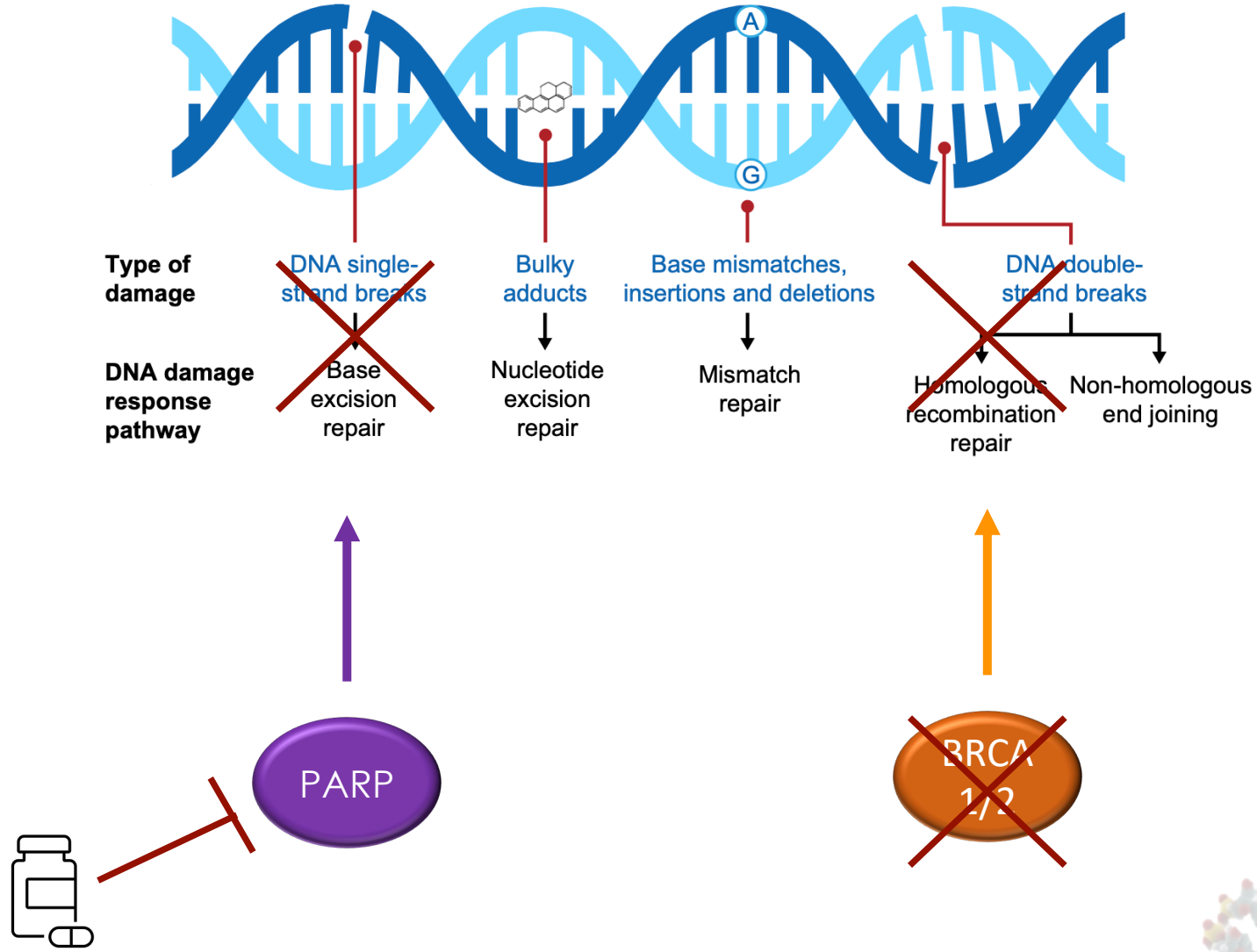


Cortesi et al., 2001

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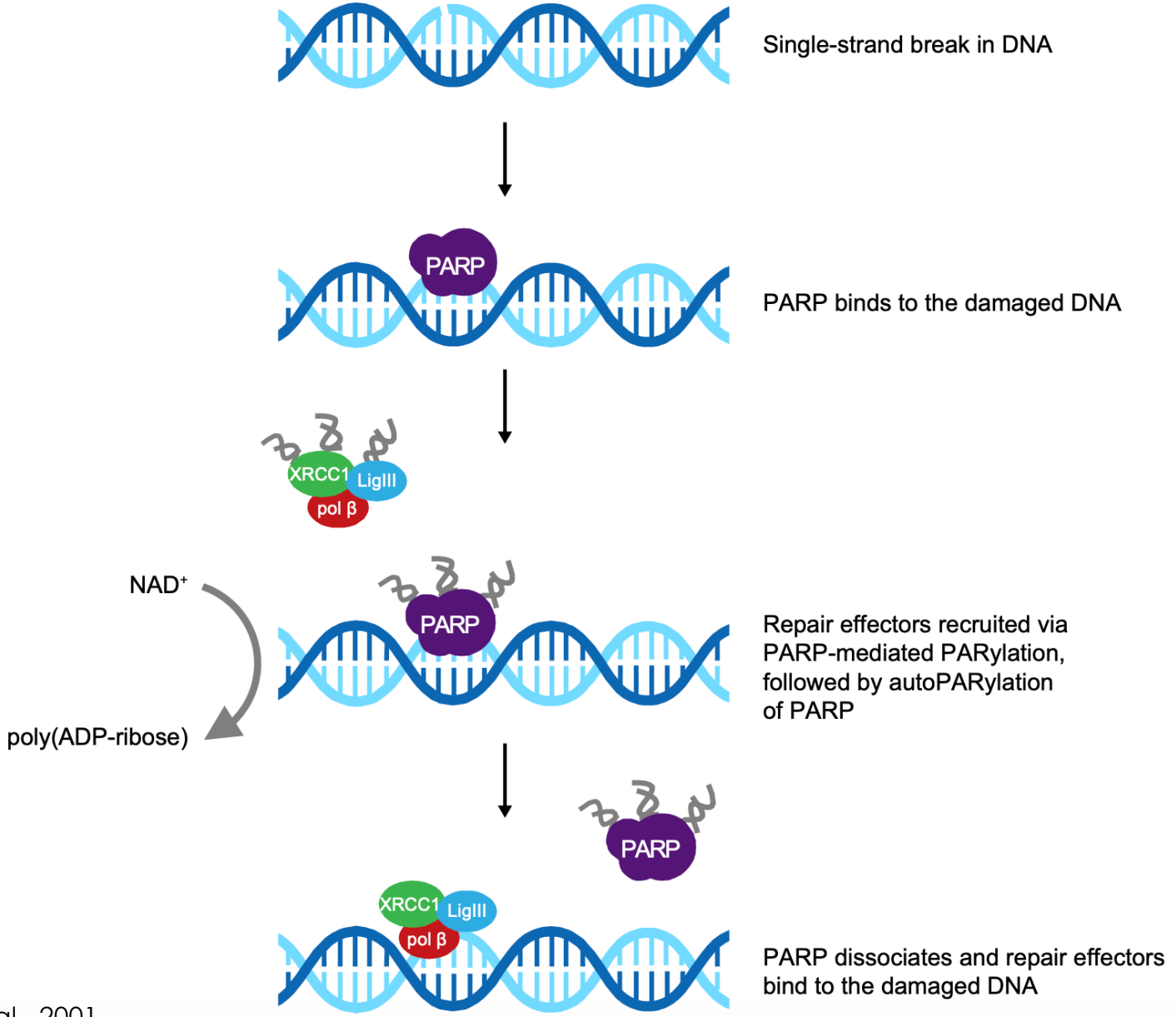


Cortesi et al., 2001

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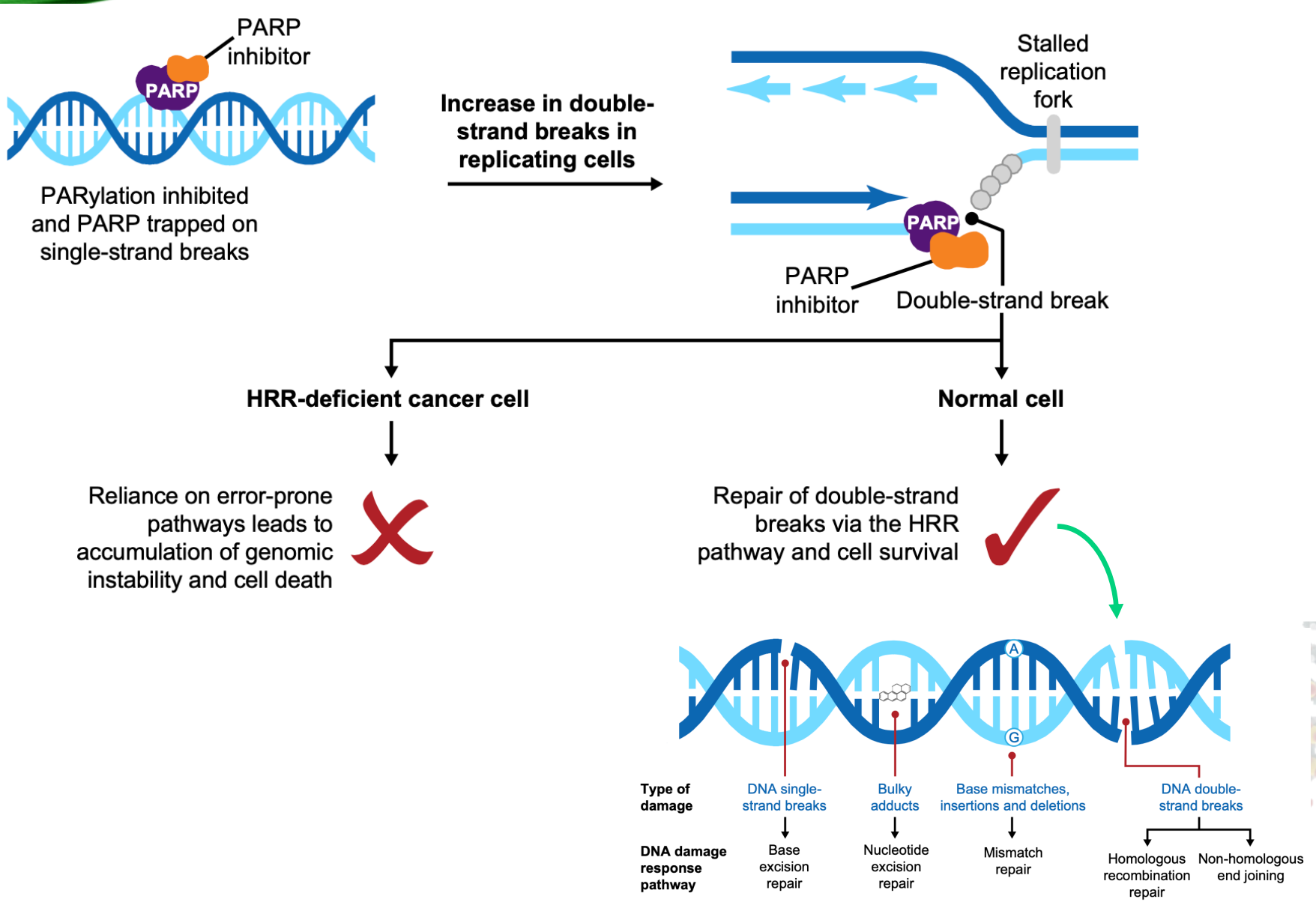



Cortesi et al., 2001

# DDR come bersaglio terapeutico nella terapia dei tumori

nature reviews cancer

https://doi.org/10.1038/s41568-022-00535-5

Review article

Check for updates

## Targeting DNA damage response pathways in cancer

Florian J. Groelly[1], Matthew Fawkes[2], Rebecca A. Dagg[1], Andrew N. Blackford[2] & Madalena Tarsounas[1]

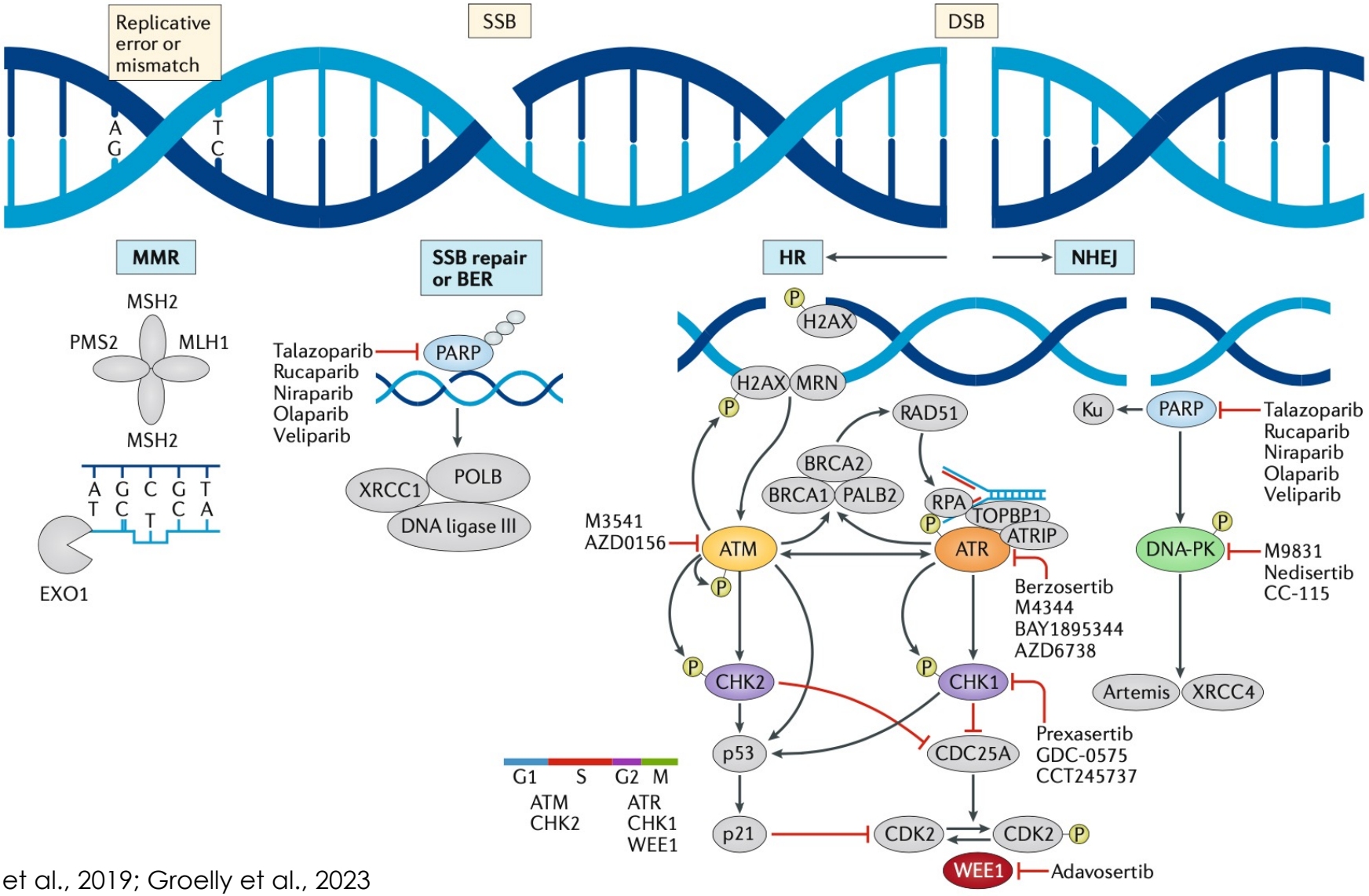
REVIEWS

## State-of-the-art strategies for targeting the DNA damage response in cancer

*Patrick G. Pilié[1,8], Chad Tang[2,3,8], Gordon B. Mills[4,5] and Timothy A. Yap[3,5,6,7]**

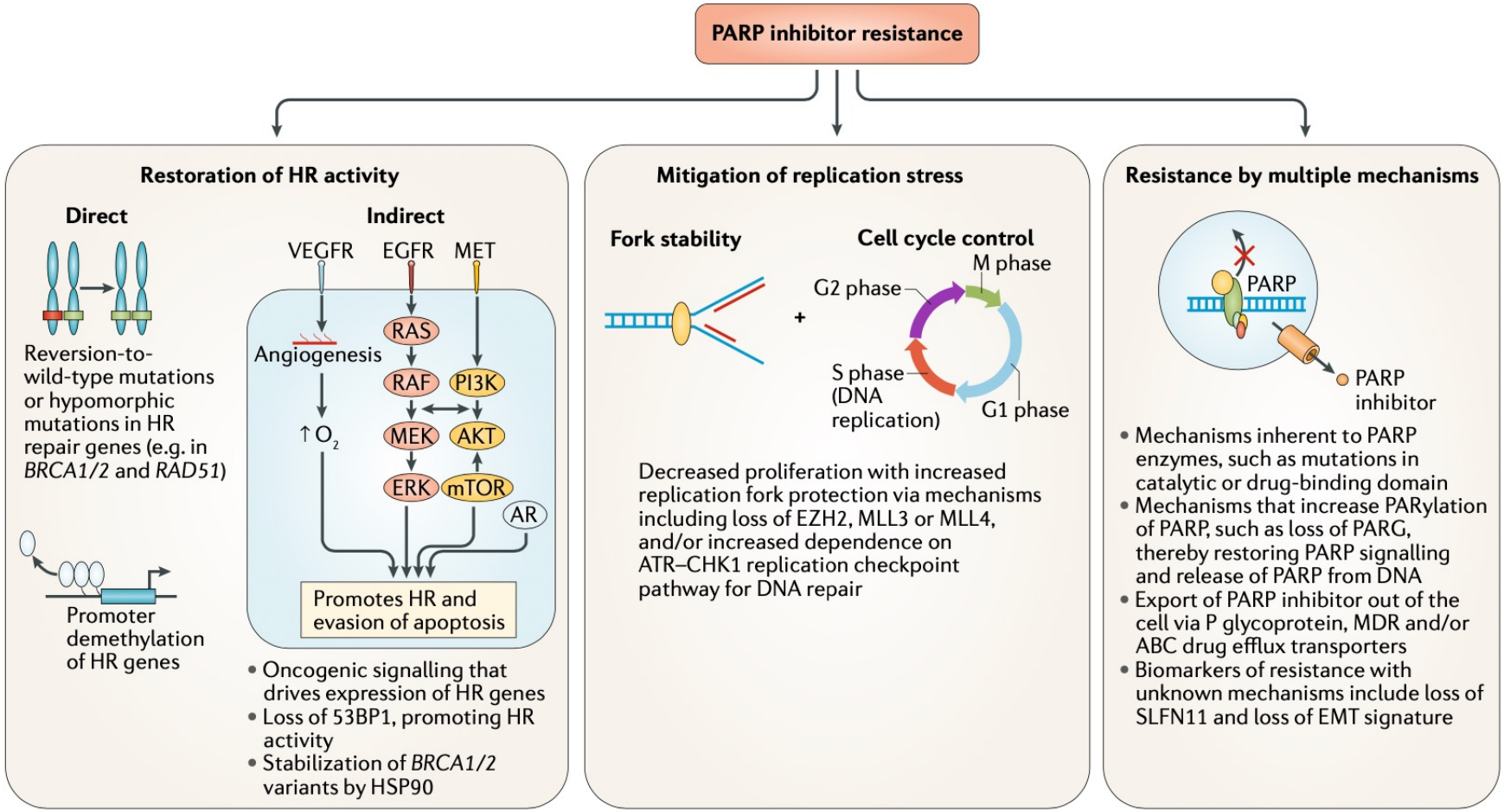
Pilié et al., 2019; Groelly et al., 2023

# DDR come bersaglio terapeutico nella terapia dei tumori



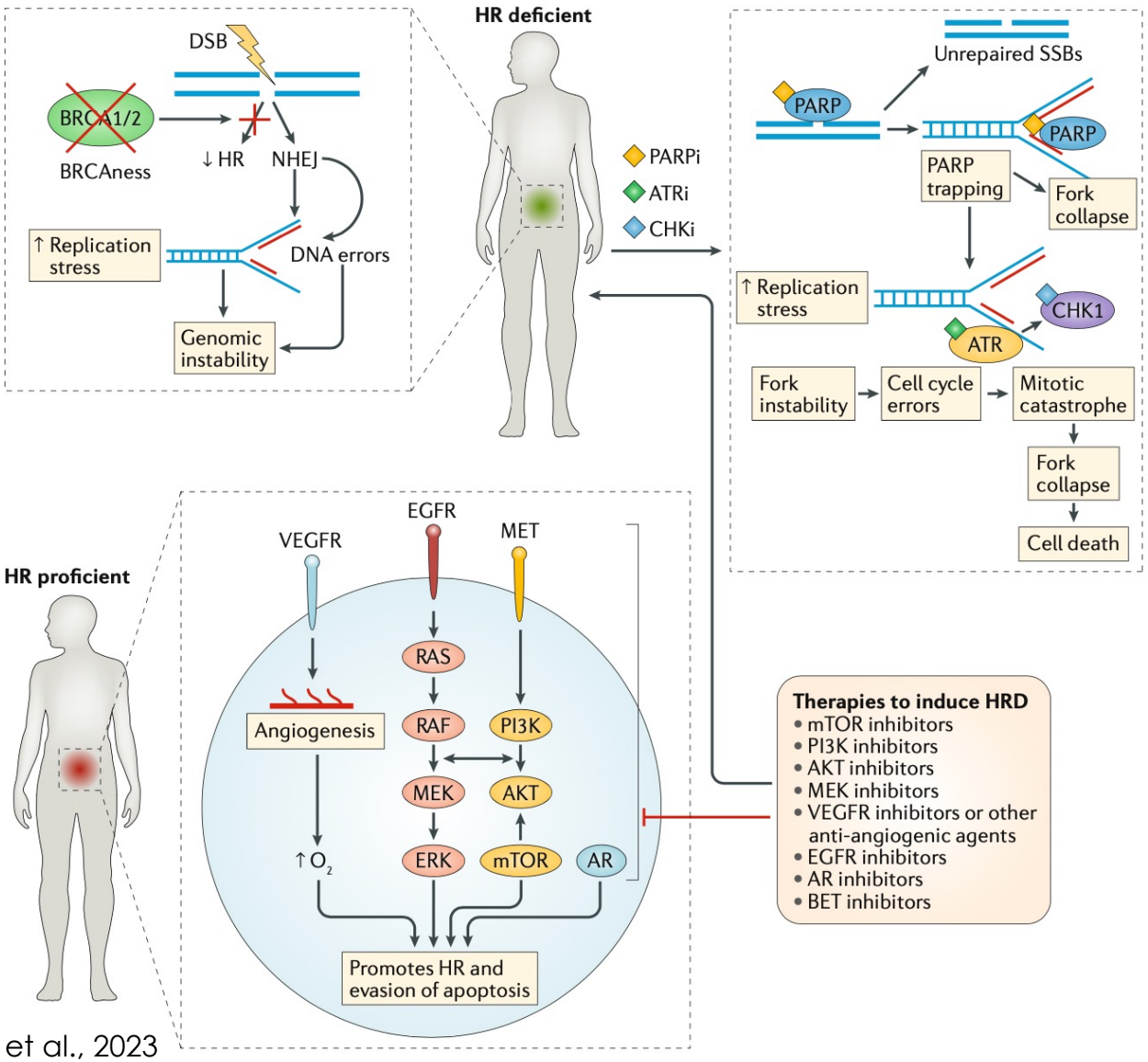

Pilié et al., 2019; Groelly et al., 2023

# Meccanismi di resistenza a inibitori di PARP




Pilié et al., 2019; Groelly et al., 2023

# Efficacia degli inibitori di PARP in diversi background genetici




Pilié et al., 2019; Groelly et al., 2023

# Inibitori DDR

| Target | Agent[a] | Combination | Phase | Cancer types | Biomarkers | Clinical trial ID[b] or reference |
|---|---|---|---|---|---|---|
| ATM | AZD0156 | Monotherapy, olaparib, irinotecan or FOLFIRI | I | ASTs | – | NCT02588105 |
| | AZD1390 | Radiotherapy | I | Grade IV glioma | – | NCT05182905 |
| | | | I | Brain | – | NCT03423628 |
| | | | I | Soft tissue sarcoma | – | NCT05116254 |
| | | | I | Lung | – | NCT04550104 |
| | M4076 | – | I | ASTs | – | NCT04882917 |
| ATM and DNA-PKcs | XRD-0394 | Radiotherapy (palliative) | I | ASTs, MSTs, RSTs | – | NCT05002140 |
| ATR | ART0380 | Monotherapy, gemcitabine or irinotecan | I/II | ASTs, MSTs | ATM deficiency | NCT04657068 |
| | ATRN-119 | – | I/II | ASTs | DDR gene mutations | NCT04905914 |
| | BAY1895344 | – | I | ASTs, lymphomas | – | NCT03188965 (ref.[196]) |
| | | Niraparib | I | ASTs, ovarian | – | NCT04267939 |
| | | Pembrolizumab | I | Solid tumours | DDR gene mutations | NCT04095273 |
| | Berzosertib (VX-970, M6620, VE-822) | Veliparib or cisplatin | I | Solid tumours | – | NCT02723864 |
| | | Various chemotherapies | I | ASTs | – | NCT02157792 |
| | | | | | | Yap et al.[196] |
| | | Gemcitabine±berzosertib | II | Ovarian | – | NCT02595892 (ref.[197]) |
| | | Topotecan | II | Lung | – | NCT02487095 (ref.[134]) |
| | | Avelumab | I/II | Solid tumours | DDR gene mutations | NCT04266912 |
| | Ceralasertib (AZD6738) | Olaparib | II | Gynaecological | – | NCT04065269 |
| | | Durvalumab | I | Head and neck, lung | – | NCT02264678 |
| | IMP9064 | – | I | ASTs | – | NCT05269316 |
| | M4344 | Monotherapy or carboplatin | I | ASTs | *ARID1A*, *ATRX*, *DAXX* or *ATM* mutations | NCT02278250 |
| | RP-3500 | Monotherapy or talazoparib+gemcitabine | I/II | ASTs | – | NCT04497116 |

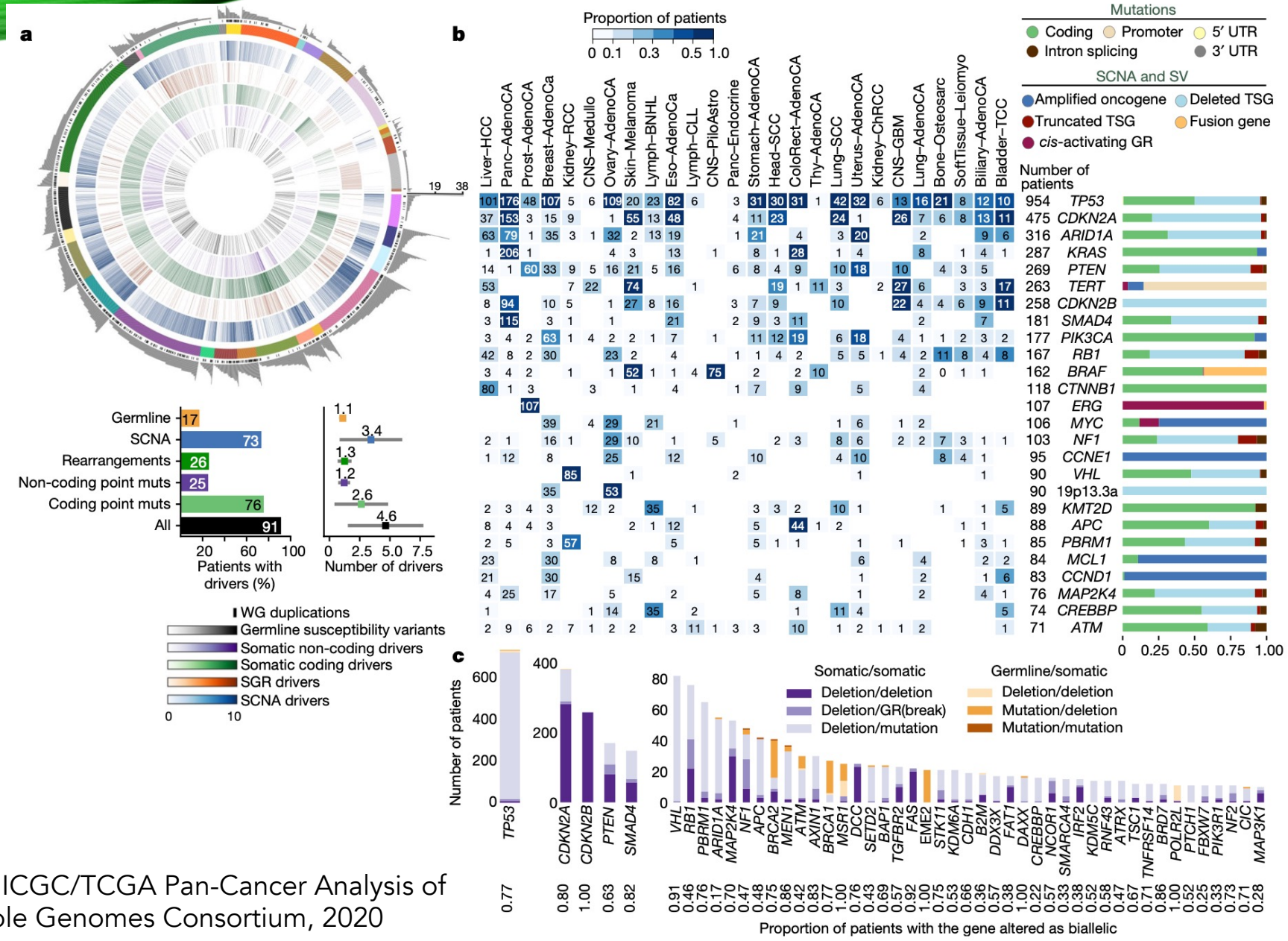
Pilié et al., 2019; Groelly et al., 2023

# Inibitori DDR

| | | | | | | |
|---|---|---|---|---|---|---|
| CHK1 | Prexasertib (LY2606368) | Irinotecan | I/II | DSRCT, rhabdomyosarcoma | – | NCT04095221 |
| | | – | II | Lung | – | NCT02735980 |
| | | – | II | Solid tumours | *MYC* or *CCNE1* amplification, *RB* loss or *FBXW7*, *BRCA1*/*BRCA2*, *PALB2*, *RAD51C*, *RAD51D*, *ATR*, *ATM*, *CHK2* or Fanconi anaemia gene mutations | NCT02873975 |
| | | – | II | Ovarian | *BRCA1*/*BRCA2* mutations | NCT03414047 |
| | | – | II | Breast, ovarian, mCRPC | *BRCA1*/*BRCA2* mutations | NCT02203513 |
| | SRA737 | Gemcitabine±cisplatin | I/II | ASTs | Predicted sensitivity to CHK1 inhibition[c] | NCT02797977 |
| | | – | I/II | ASTs, NHL | Predicted sensitivity to CHK1 inhibition[c] | NCT02797964 |
| | LY2880070 | Gemcitabine | I/II | ASTs, MSTs | – | NCT02632448 |
| | | | II | Ewing sarcoma | – | NCT05275426 |

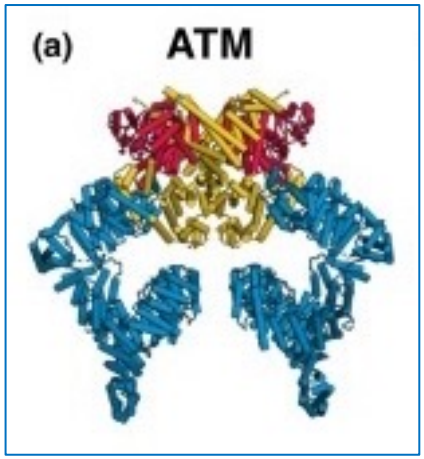
Pilié et al., 2019; Groelly et al., 2023

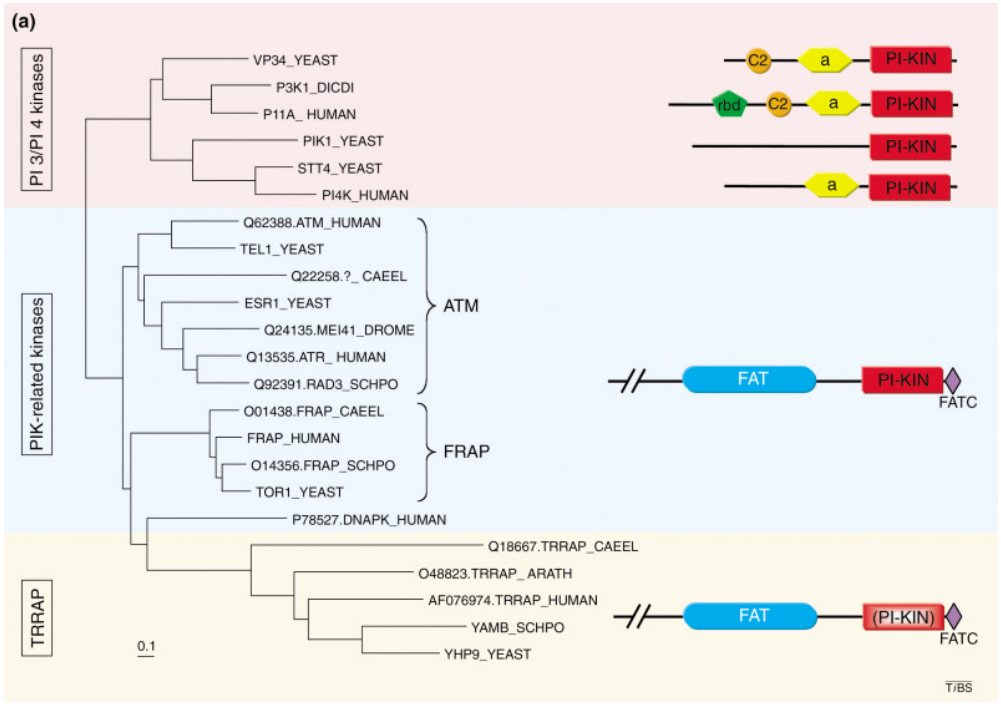
# Genomica del cancro




The ICGC/TCGA Pan-Cancer Analysis of Whole Genomes Consortium, 2020

# ATM

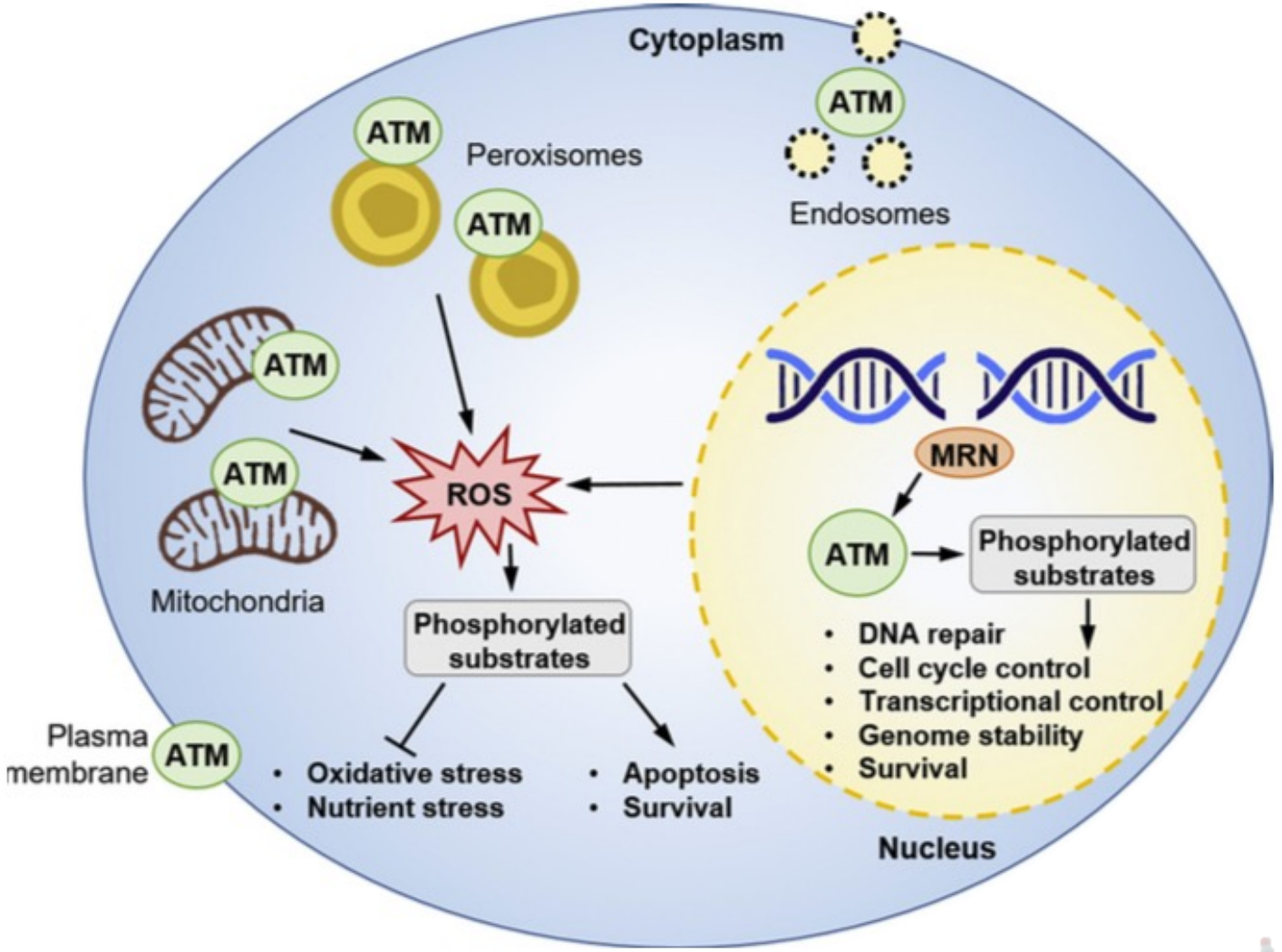




Maréchal and Zou, 2013; Hanahan and Weinberg, 2013; Williams and Zhang, 2021

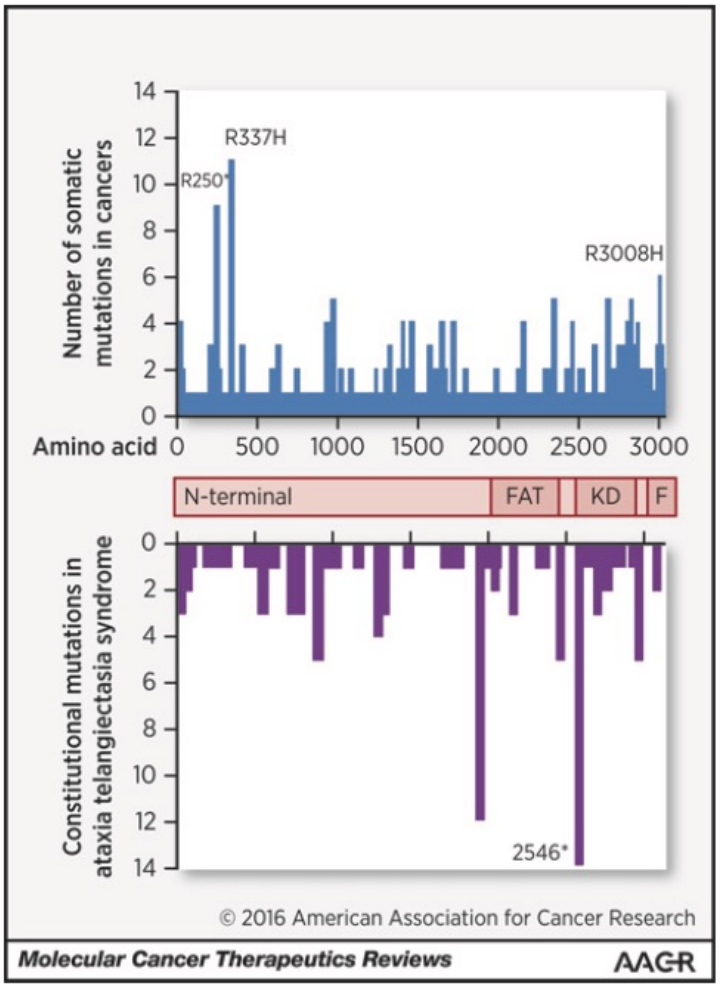
# ATM

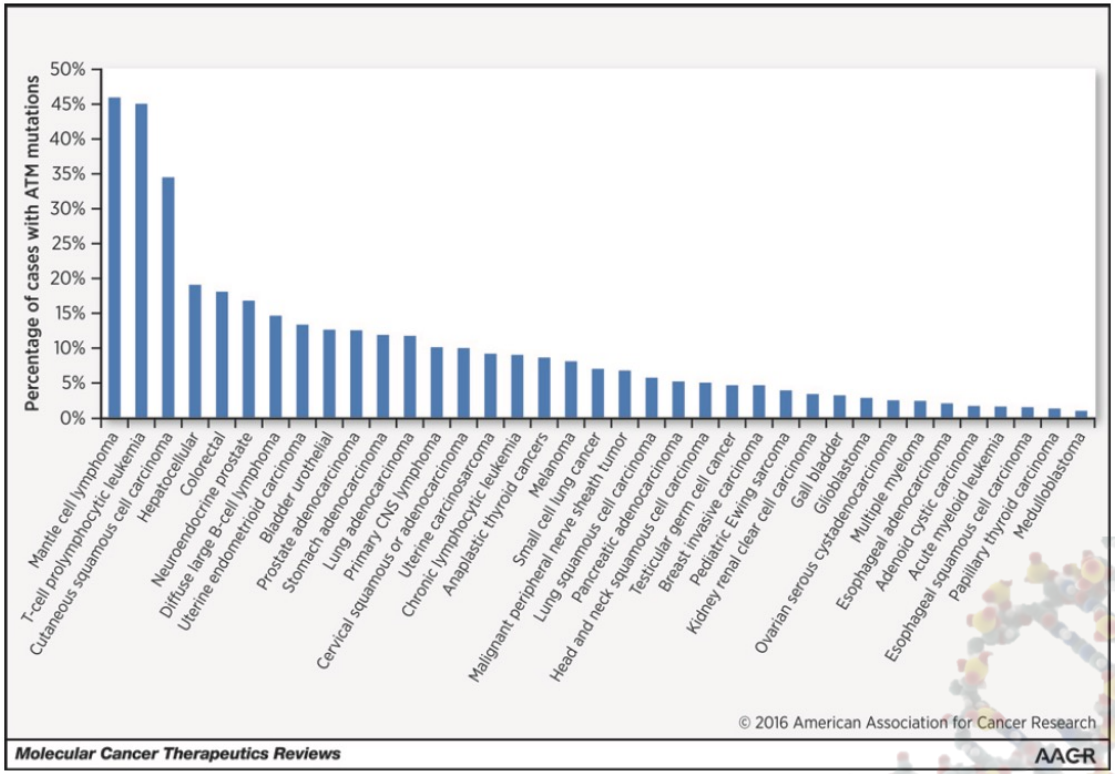

Cytoplasm
ATM
Peroxisomes
ATM
ATM
Endosomes
ATM
ATM
Mitochondria
ROS
MRN
ATM
Phosphorylated substrates
Phosphorylated substrates
• DNA repair
• Cell cycle control
• Transcriptional control
• Genome stability
• Survival
Plasma membrane
ATM
• Oxidative stress
• Nutrient stress
• Apoptosis
• Survival
Nucleus

# ATM in cancer

ATM mutations are frequent in cancer

ATM as a target in cancer therapy




Choi et al., 2016

# La mia ricerca

- ATM mutations or ATM inhibition increase sensitivity to radiotherapy
- Specific ATM inhibitors have been developed

? Cancers for which therapy benefit of ATM inhibition

? Resistence to ATM inhibition

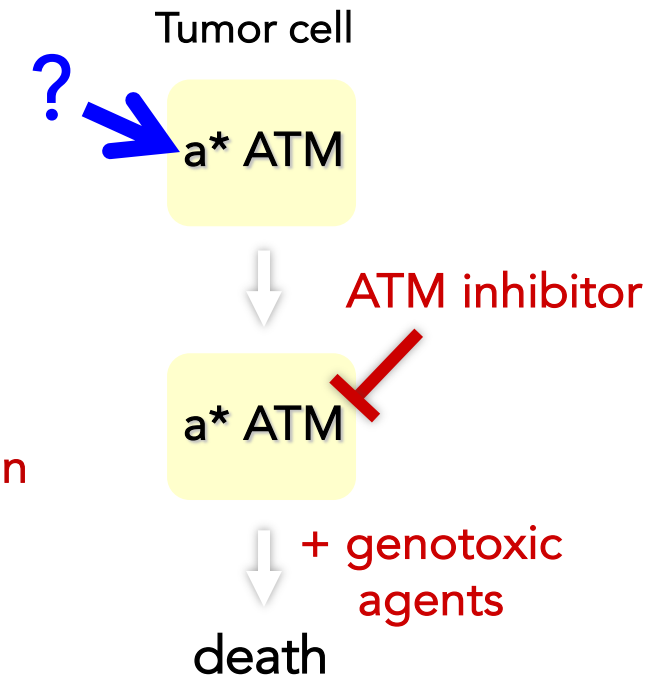



**Table 1** Phase I Clinical Trials of ATM Inhibitors in 2021

| ATMi | Combination | Tumor Types | NCT# | Recruitment Status |
|---|---|---|---|---|
| AZD0156 | Module 1: Olaparib<br>Module 2: FOLFIRI (irinotecan and 5-fluorouracil) | Module 1: Locally advanced/metastatic tumors including gastric adenocarcinoma<br>Module 2: Colorectal cancer | NCT02588105 | Active, enrolled 84 patients, but not enrolling |
| AZD1390 | Arm A: 35 Gy over 2 wk<br>Arm B: 30 Gy over 2 wk whole or partial brain RT<br>Arm C: 60 Gy over 6 wk partial brain RT | Arm A: Recurrent glioblastoma<br>Arm B: Metastatic brain tumors<br>Arm C: Primary glioblastoma | NCT03423628 | Arm A and C enrolling; Arm B enrollment completed |
| M3541 | 30 Gy over 2 wk | Advanced and metastatic solid tumors | NCT03225105 | Enrollment completed |

Lavin and Yeo, 2020