Imperial College London

Drug Discovery at Imperial: not just an academic exercise

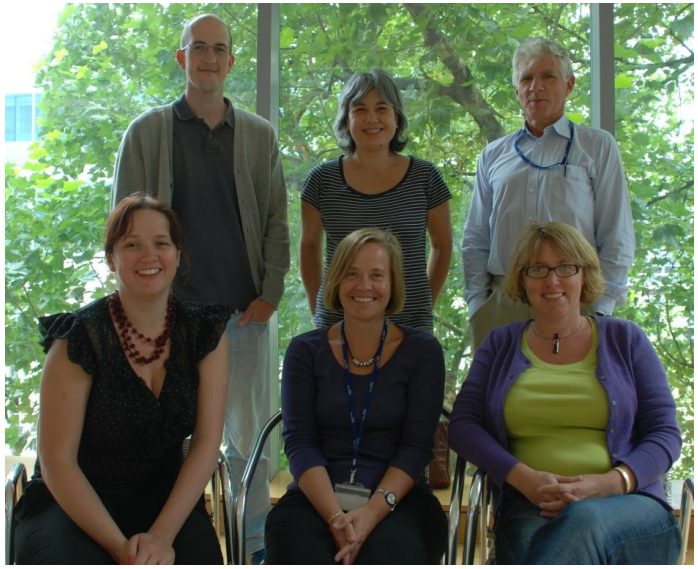
Cathy Tralau-Stewart PhD FRSC
Head of Drug Discovery
Drug Discovery Centre
Imperial College

www.imperial.ac.uk/medicine/drugdiscoverycentre

Imperial College London

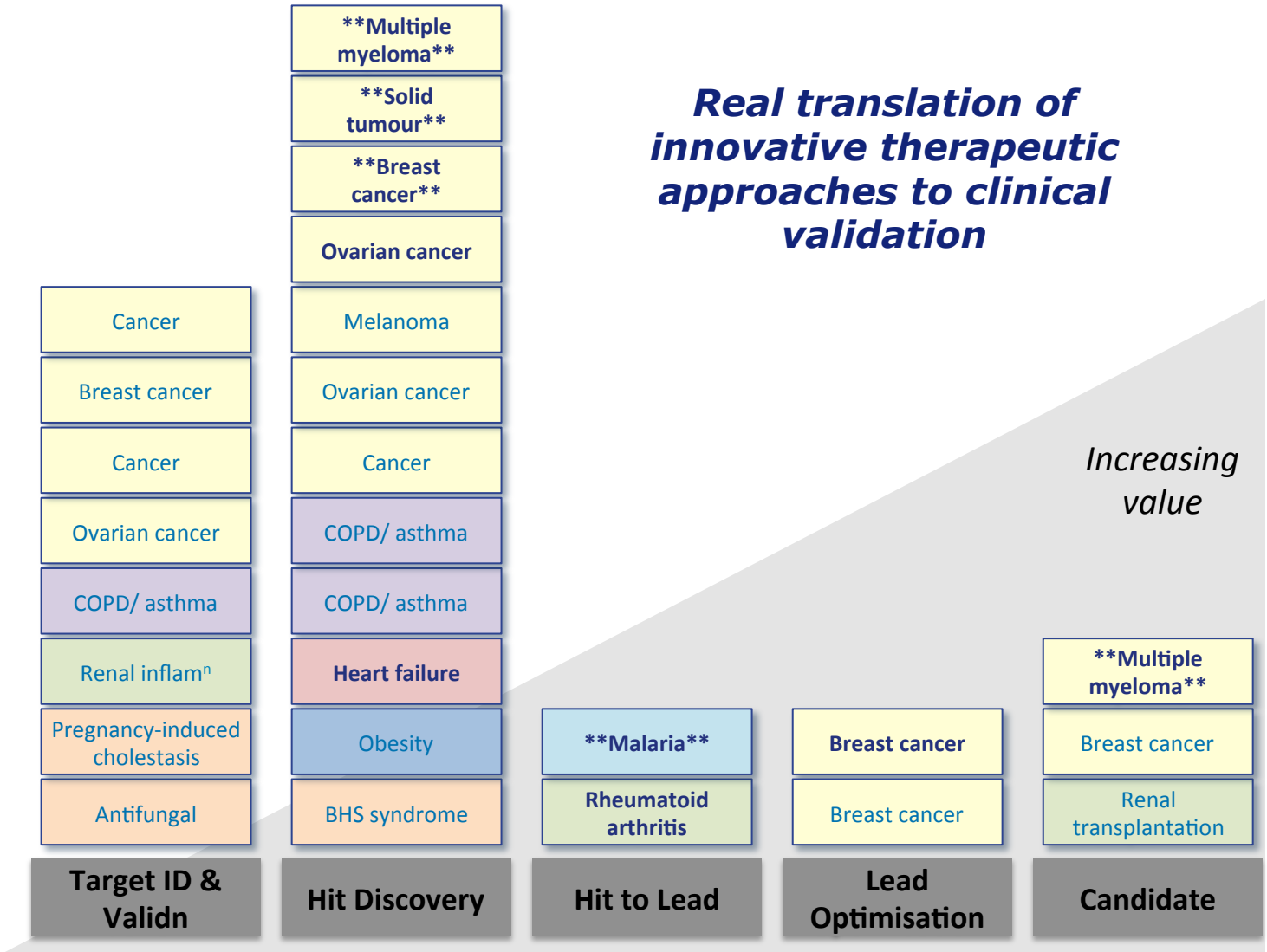
The Drug Discovery Centre (DDC)

- **Industry expertise working directly with basic & clinical researchers to translate projects to clinical validation**
- Projects sourced from Imperial's 3400 + researchers
- Multidisciplinary industry expertise in-house (4.6 FTEs)
- Limited Infrastructure
- Outsources specific expertise, skills and capabilities
- Project and outsourcing management
- ***Challenging how we do drug discovery***



Imperial College London

Current DDC portfolio by stage, target, therapeutic area & DDC rank importance



But..

Despite;

- Most Pharma saying that 50-70% of future projects will come from outside (academia/ Biotech)

- Very significant DDC cross-therapeutic area project success;
  - >25 early projects assessed
  - 7 projects in hit – candidate phases
  - 1 spinout + 1 other under consideration

The DDC will be closed in its current configuration

- Lack of core funding

Imperial College London

Funding needs to fully support translation

Funding:

- ***Research Councils***
- ***Charities***

Limited

- ***Wellcome***
- ***MRC***
- ***CRUK***
- ***Disease specific Charities***

Very limited

NIHR/ NOCRI
- **BRC/BRUs**
- **Translational Research partnerships**

INDUSTRY

Limited

Limited funding for:

Translation from basic research to Clinical validation

- MRC/TSB Biomedical Catalyst Fund ? ?

(Strategy for UK Life Sciences Dec 2011)

- Pharma ? ?

Imperial College London

The DDC: not just 'traditional' drug discovery

Examples;

1. Model peptide molecules to derive therapeutics
2. Structural approaches to understand target in combination with virtual screening & modelling
3. Find novel approaches with models of human disease systems (cell, tissue) which mimic real clinical effects

Academia, linked to patients are well placed to do drug discovery close to the clinic

Requires both academic and industrial skill sets

DDC example projects

1. Model peptide molecules to derive therapeutics
   - Peptides for Multiple Myeloma
2. Structural approaches to understand target in combination with virtual screening & modelling
   - Cell Cycle Phosphatase Inhibitors for Cancer
3. Find novel approaches with models of human disease (cell, tissue) which mimic **real** clinical effects
   - Platinum resistance in Ovarian Cancer

Imperial College London

1. Multiple Myeloma : NFkB pathway

Professor Guido Franzoso, Lorna Tornatore, Menotti Ruvo, Caroline Low, Cathy Tralau-Stewart, Albert Jaxa-Chamiec, Katie Chapman

Supported by MRC DPFS

A Peptide modelling approach

Multiple Myeloma : Background

- Current therapies typically target the NF-κB pathway and can induce temporary remission
- MM remains incurable, onset of drug resistance is common, and management of relapsed and refractory disease remains a major problem
- We have developed novel peptide leads which selectively target the interface between a **protein-protein interaction downstream of the NF-κB-dependent pathway** of survival in MM
- Enhances JNK cytotoxic signaling via inhibition of NF-κB-JNK crosstalk
- This leaves the functions of NF-κB in inflammation, immunity and development intact

Competitor approaches

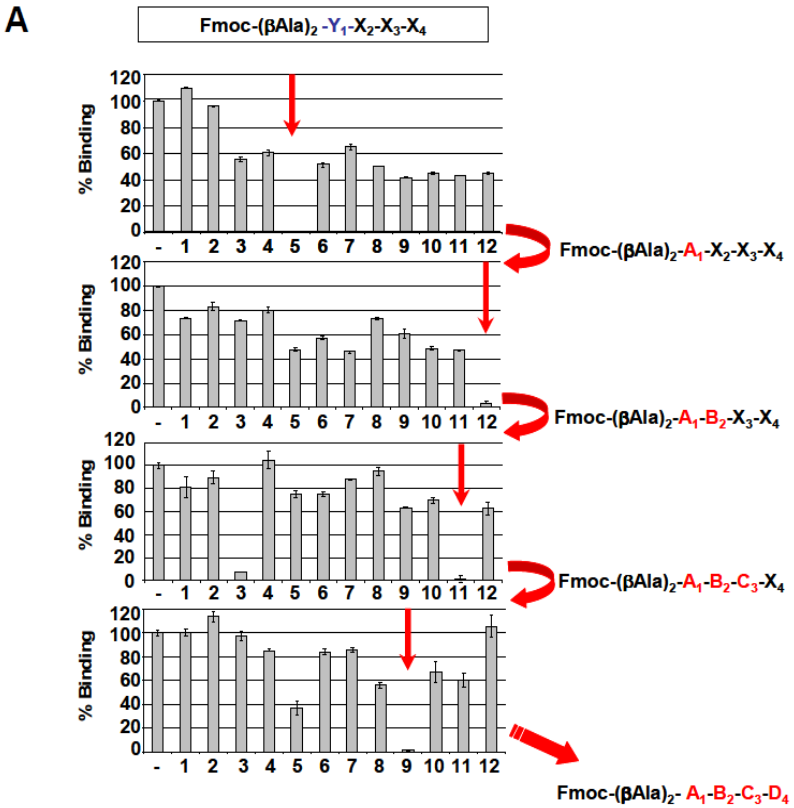
- The NF-κB pathway is a well-recognized target for drug development in MM
- Conventional NF-κB blockers, eg Proteasome Inhibitors (bortezomib), are only tolerated clinically at doses that produce ≤80% systemic proteasome inhibition, allowing some cancerous cells to escape elimination

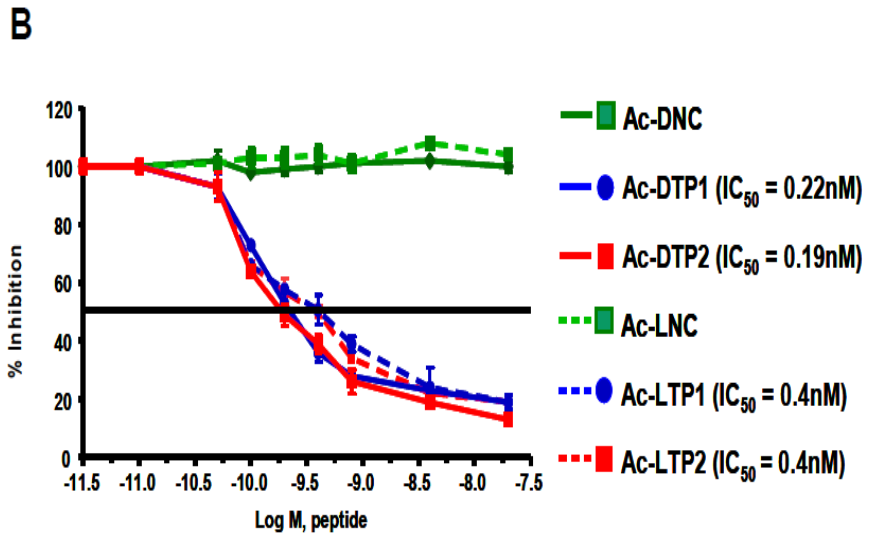
Imperial College London

Screening of peptide library

A. ELISA competition assay to screen >20,000 peptides and isolate leads

B. DTPs inhibit interaction with Sub-nM IC_{50}s

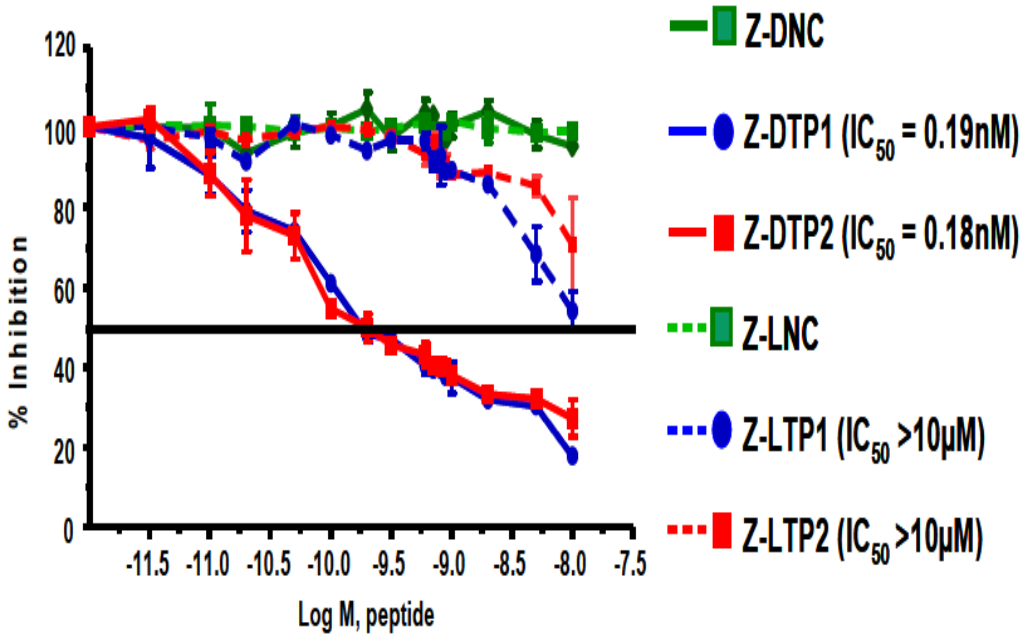




Laura Tornatore ,Menotti Ruvo & Guido Franzoso

Peptides are stable in human serum

ELISA competition assays with peptides after a 48-hr incubation with human serum at 37°C



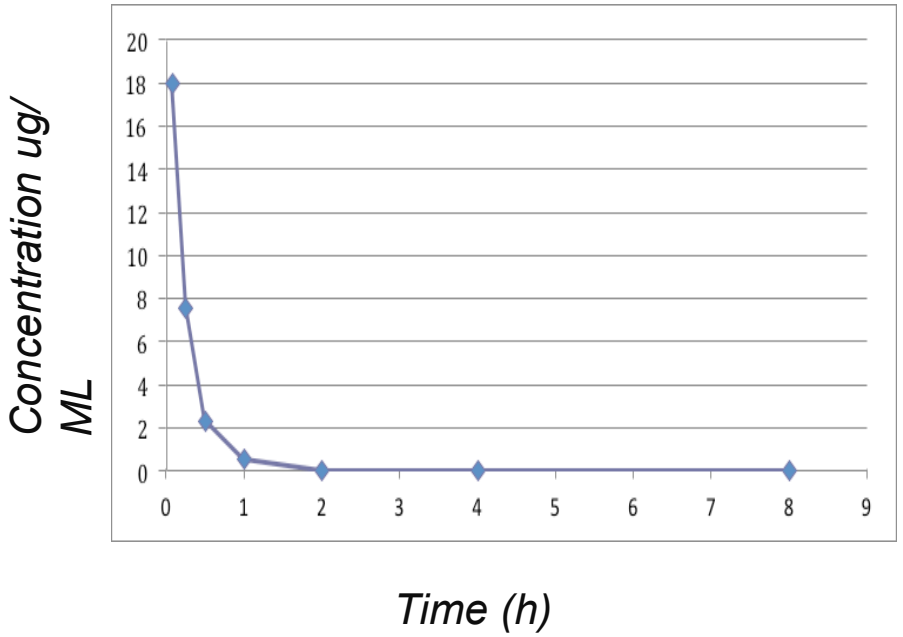
Laura Tornatore & Guido Franzoso

Pharmacokinetics

Peptide Pharmacokinetics in mouse after single iv bolus administration

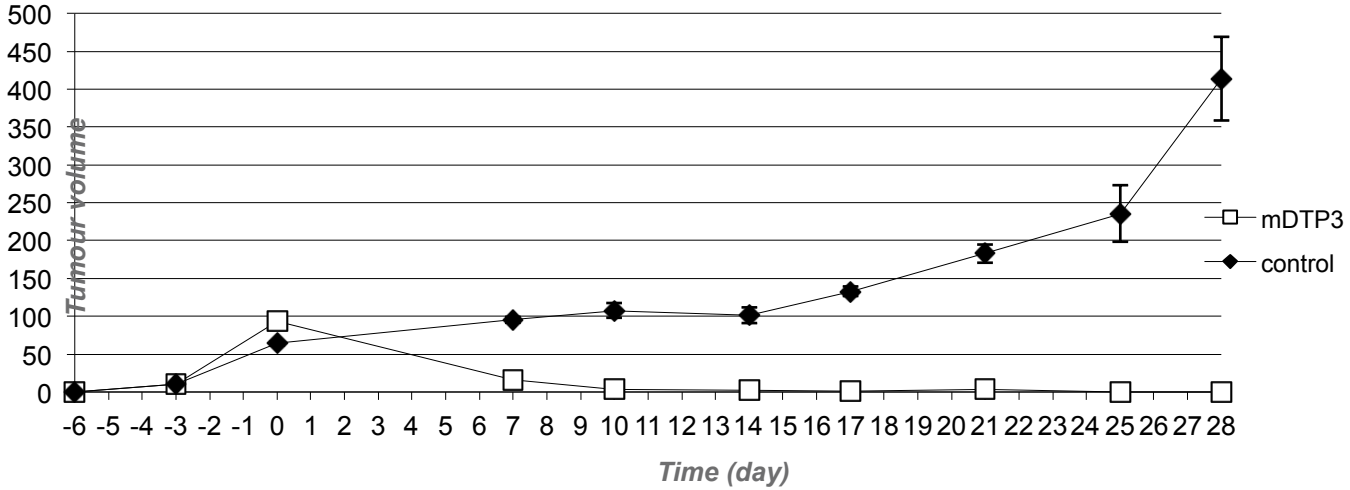
Parameter	Mean values N=3
T1/2(h)	1.26
AUC(ug.h/ml)	6.43
V (L/kg)	2.79
Cl (ml/min/kg)	27.1
Cl mL/h	44.2

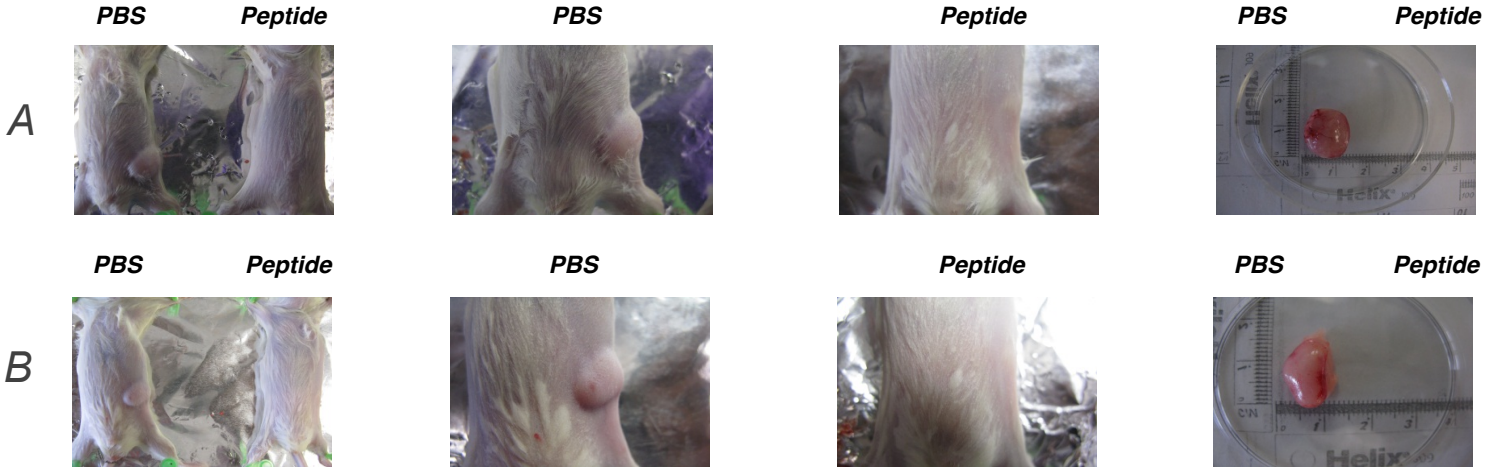
Mouse time course in plasma after iv bolus dose



Imperial College London

Tumour size decreases within 1 week of treatment with lead peptide





Laura Tornatore & Guido Franzoso

Small-peptides for Multiple Myeloma

- Low Molecular weight peptides (460-650Da)
- Good cell activity (Lead peptides kill MM cell lines with IC_{50}s in low nM to low µM range)
- High therapeutic index in vitro
- High stability in biological fluids
- Pharmacokinetics suitable for iv infusion

Imperial College London

2. Cell Cycle Phosphatase Inhibitors for Cancer

David Mann, Prof Alan Armstrong, **Caroline Low,** Cathy Tralau-Stewart, James Collins, Katie Judd, Katherine Scott, Katie Chapman

Supported by CRUK

A virtual screening approach

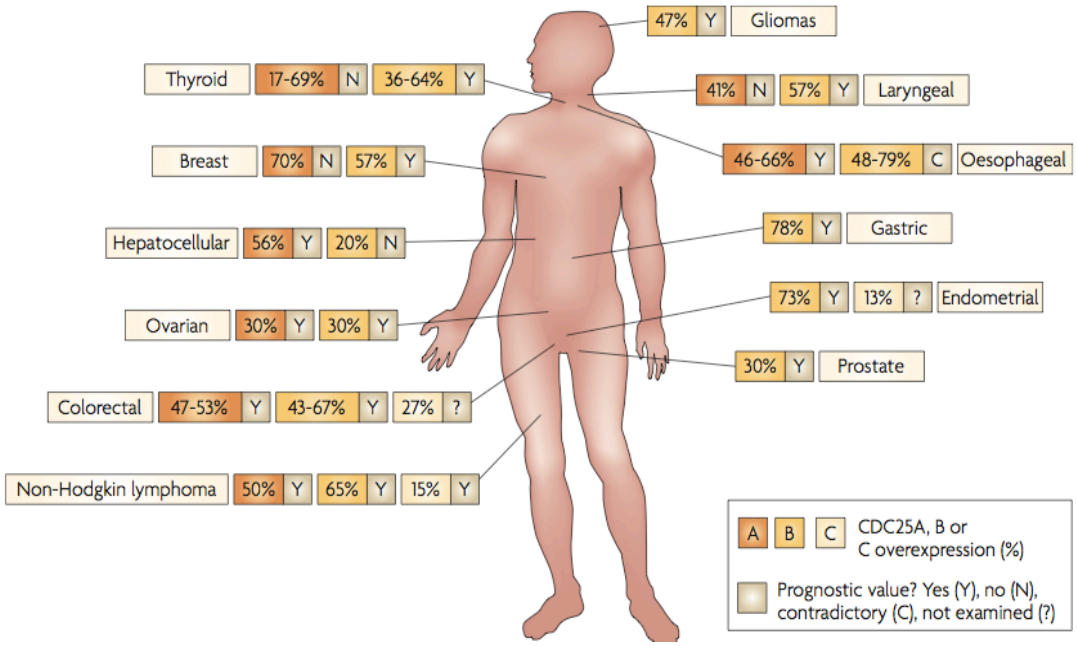
Imperial College London

Cdc25 – establishing a pharmacophore for reversible inhibitors

CDC25 phosphatases in cancer cells: key players? Good targets?

Rose Boutros, Valérie Lobjois* and Bernard Ducommun*‡*

Nature Reviews Cancer 7 (2007) 495-507

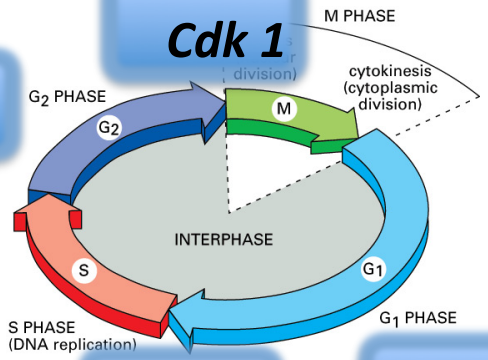


Cdc25B

Cdc25C

Cdk 1

Cdk 2



Cdk 2

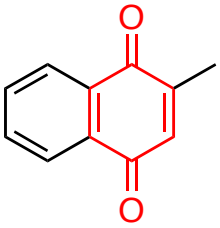
Cdk 4

Cdc25A

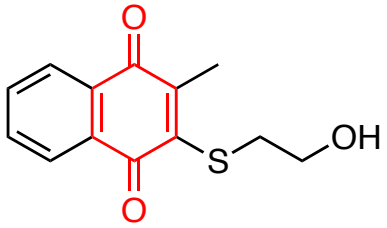
?

Imperial College London

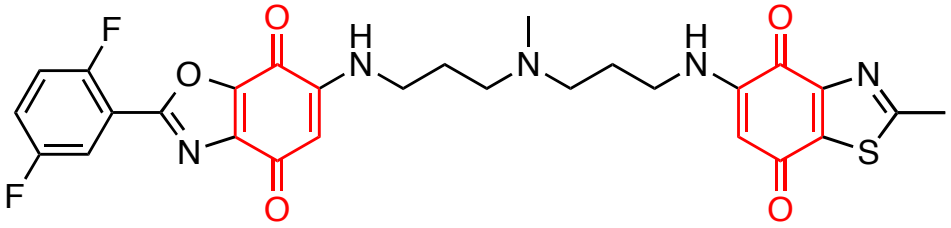
Quinones: irreversible Cdc25 inhibitors



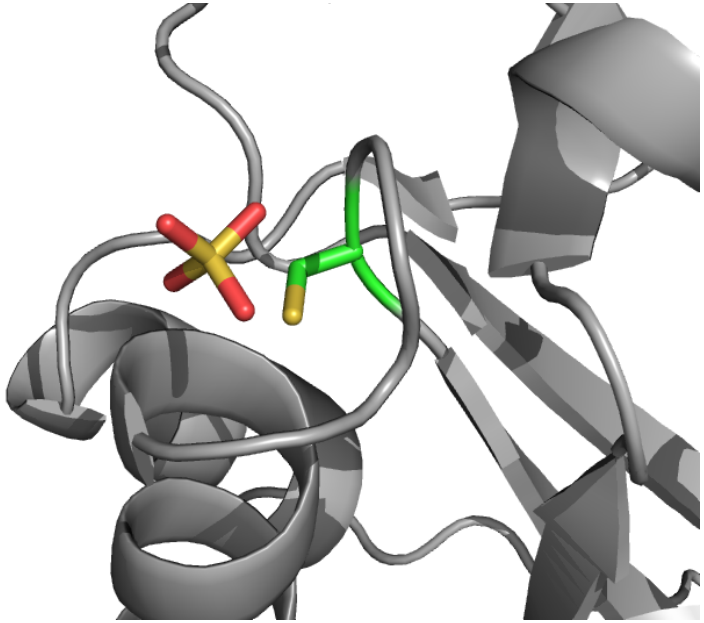
Vitamin K3



BN82685
Cdc25B IC50 3.8μM

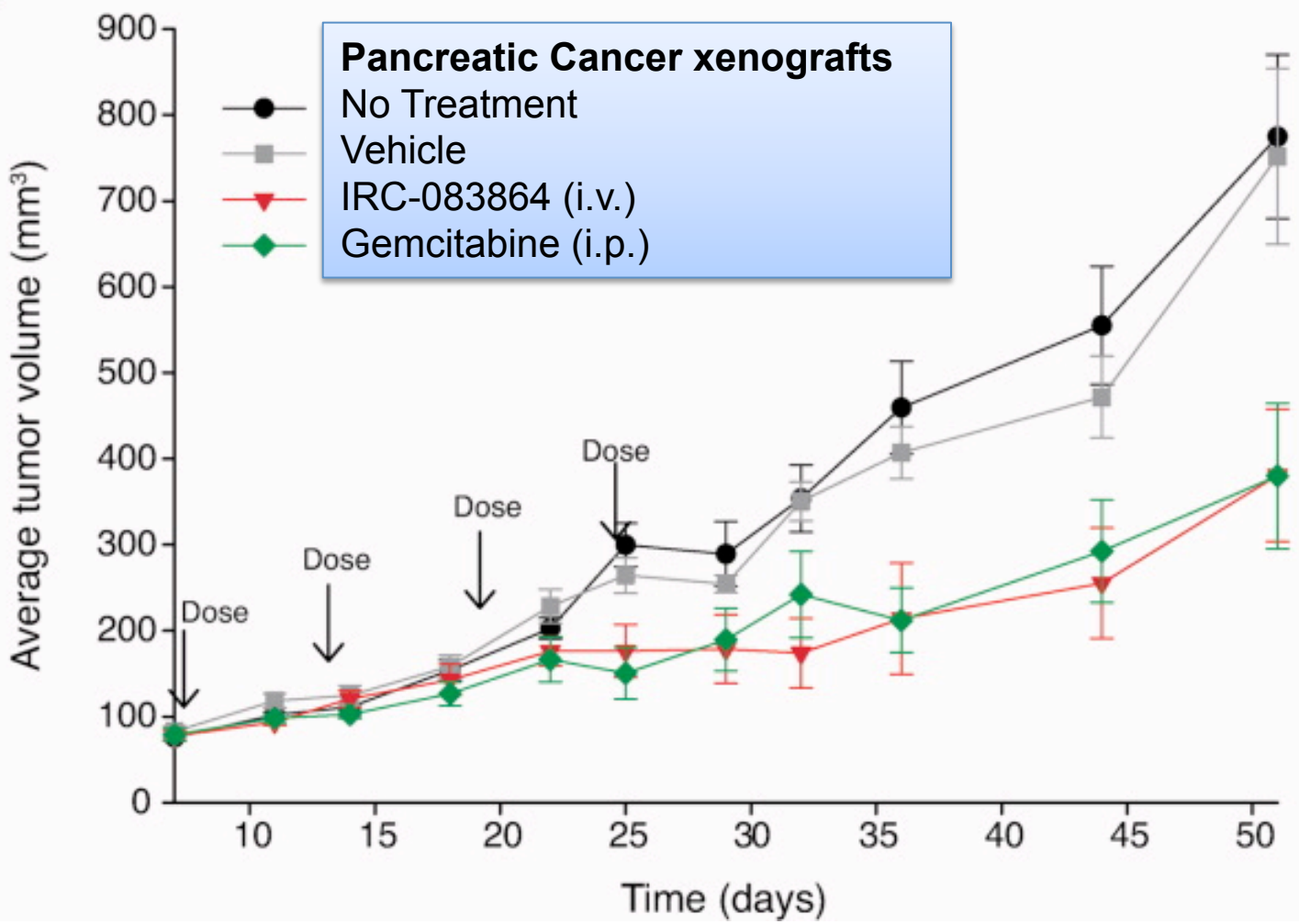


IRC-083864/Debio-0931
Cdc25A: 23 nM
Cdc25B: 26 nM
Cdc25C: 23 nM



Quinones arrest cell cycle by
- oxidation of Cys in catalytic site
- irreversible reaction with Cys

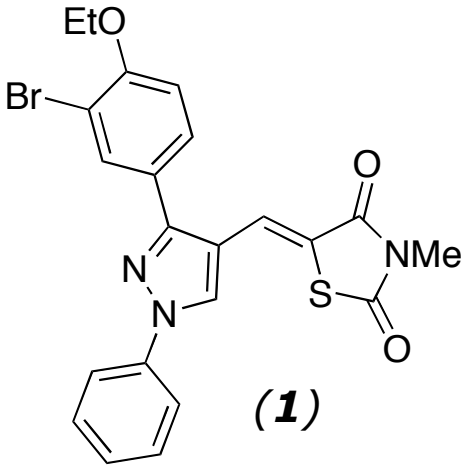
Quinone inhibitors vs standard treatment



Brezak et al, (2009), Int. J. Cancer, ***124****, 1449*

Imperial College London

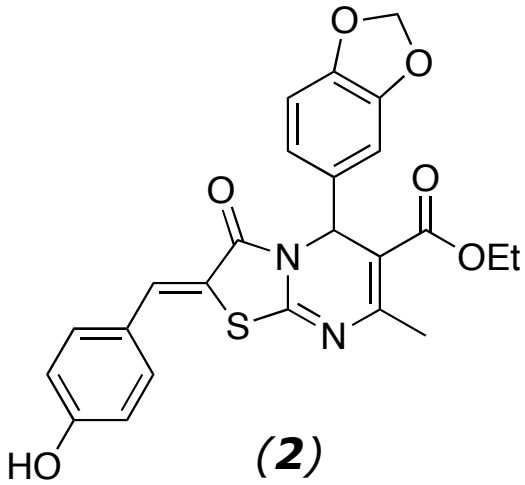
Small set of reversible inhibitors known



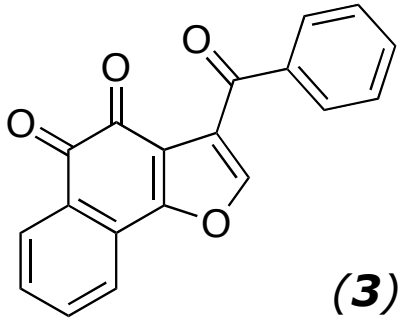
(1)

Assay	IC50 (μM)
Cdc25B	2.0

Kim et al WO2006/101307



(2)



(3)

PITT-9131

Assay	IC50 (μM)
Cdc25A, B, C	5-10

Brisson et al (2004), Mol. Pharm., 824

Assay	IC50 (μM)
MBP-Cdc25B3	13.0 ± 0.5

Montes et al (2008), J. Chem. Inf. Model ,157

Imperial College London

Create new class of reversible Cdc25 inhibitor using field points

Model

- Create single model from 3 different ligands
- Dissect out field point pattern for one compound

Virtual Screen

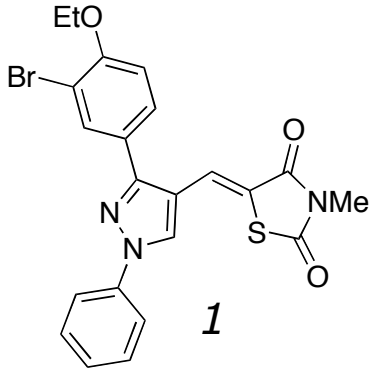
- Use as pharmacophore probe for virtual screen
- Hunt for compounds with similar field point patterns

Test

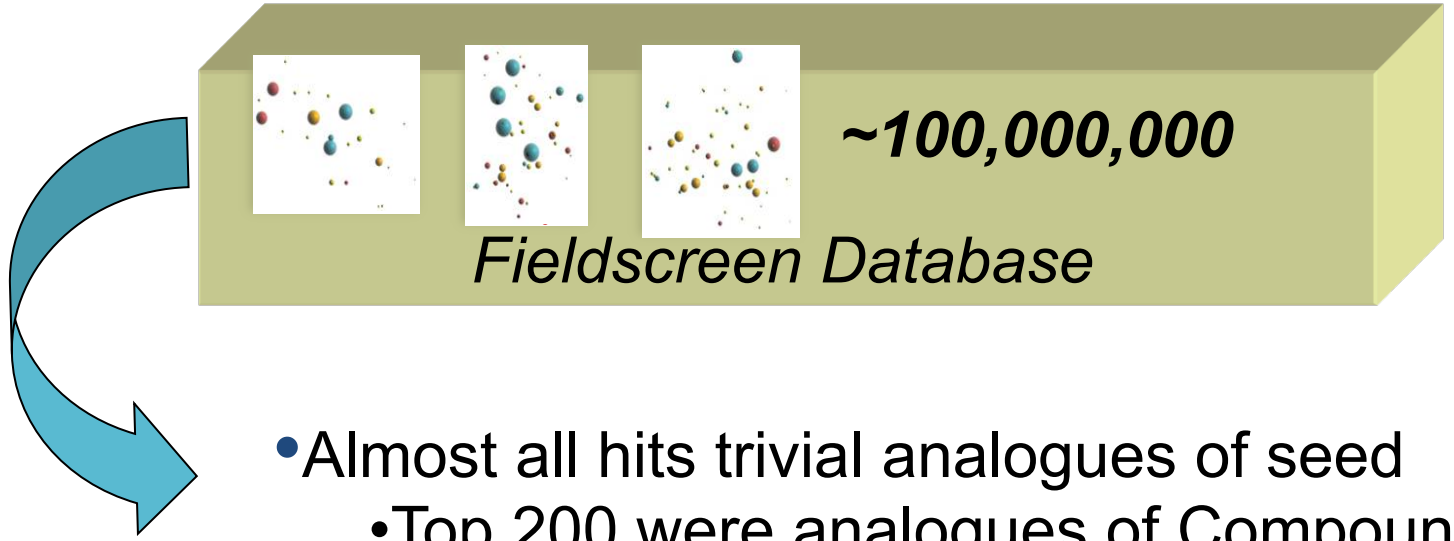
- Purchase commercial compounds suggested
- Test compounds in enzyme bioassay

Imperial College London

High throughput virtual screening to identify novel series



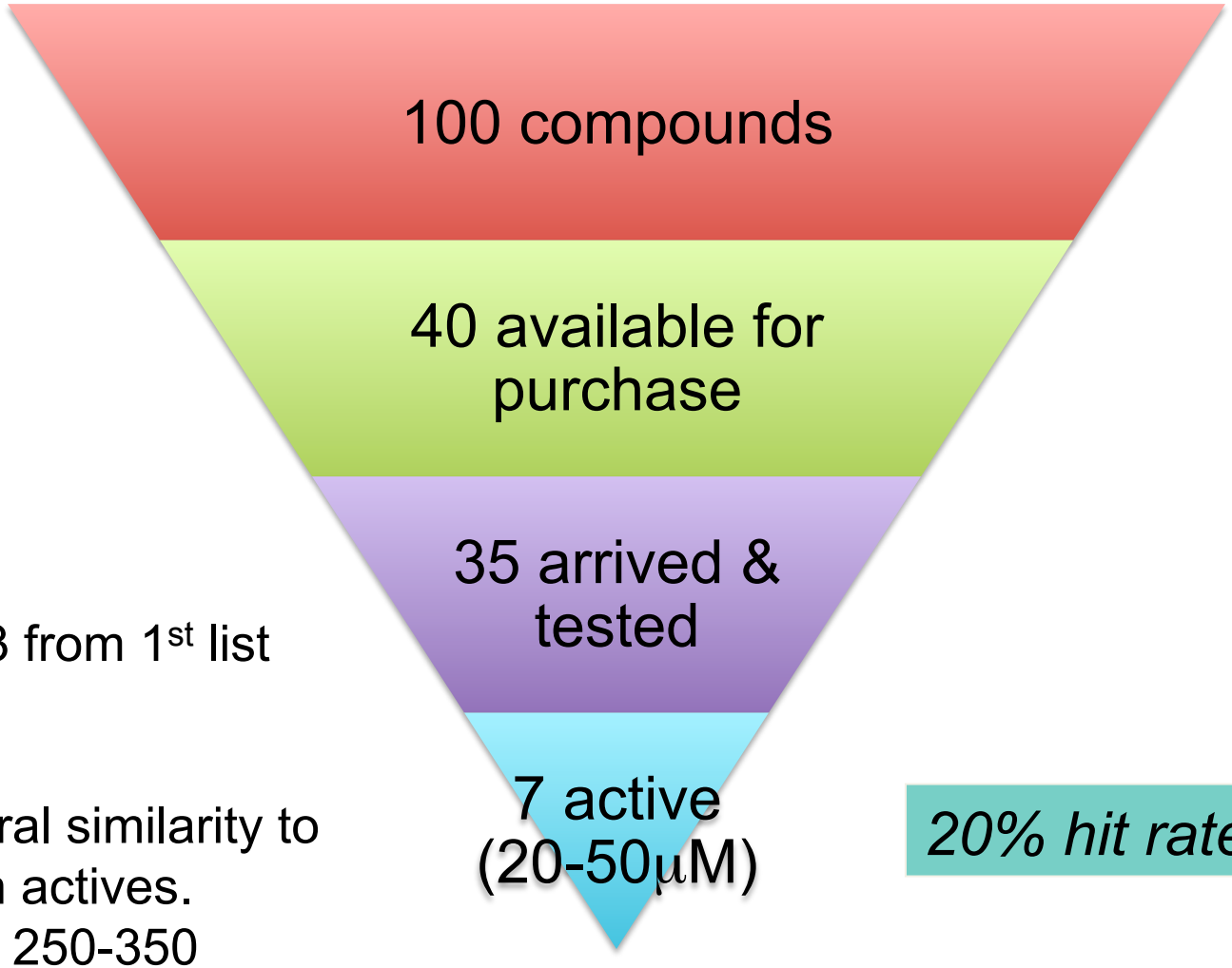
1



- Almost all hits trivial analogues of seed
  - Top 200 were analogues of Compound 1
  - 989/1000 were pyrazoles

Imperial College London

CDC25 screen re-run with pyrazoles excluded



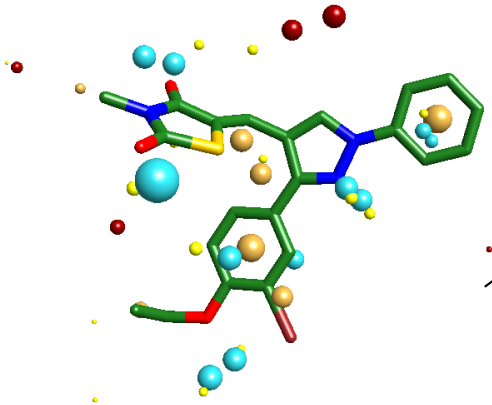
Including 3 from 1st list

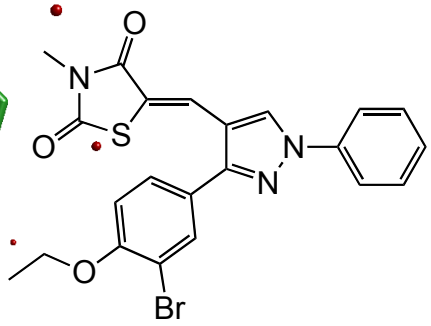
No structural similarity to any known actives.
MW range 250-350

20% hit rate

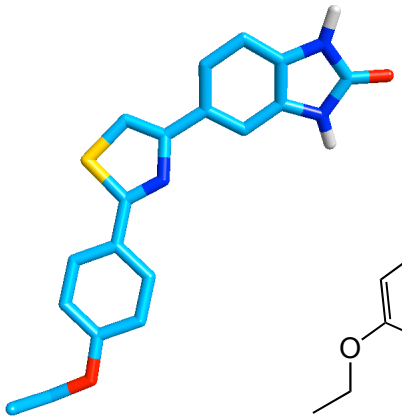
Imperial College London

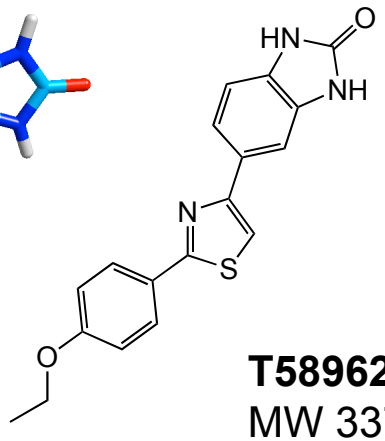
Initial thiazole hits from virtual screen





(**1**) MW 484





T5896241
MW 337

Cdc25B IC_{50} 2.3 μM

- Selective against related phosphatases
  - PTP1B, MKP-1 & 3 and alkaline phosphatases
- Cellular target confirmed (n=1)
  - predicted increase in phosphorylated CDK2
- Later compounds amongst most potent reversible Cdc25 inhibitors described

Cdc25A IC_{50} 35.5 ± 0.1 μM
Cdc25B IC_{50} 17.2 ± 0.1 μM
Cdc25C IC_{50} 47.3 ± 0.1 μM

3. Reversing platinum resistance in ovarian cancer

Prof Hani Gabra, Euan Stronach**, Albert Jaxa-Chamiec, Hayley Cordingley**, Caroline Low, Katie Chapman, Michelle Heathcote , Azadeh Cheraghchi Bashi Astaneh , Cathy Tralau-Stewart

A phenotypic approach

Platinum resistant ovarian cancer

- Many women affected by ovarian cancer:
  - ~7000 in UK annually
  - ~ 200,000 worldwide annually
- Treatment:
  - Surgery
  - Combination with platinum chemotherapy
- Platinum resistance:
  - Initial response good
  - Relapse frequent (>75%)
  - Most eventually become resistant to platinum
- 5yr survival for advanced stage ovarian cancer is less than 25%

Imperial College London

Imperial Ovarian Cancer Action team

- Created Isogenically matched cell lines*:
  - 3 cases of high grade serous ovarian cancer
  - Lines derived from samples pre & post platinum resistance development

- Gene expression profiling§:
  - 10K cDNA microarrays *(Sanger H ver 1.2.1)*
    - 91 up-regulated genes; 126 genes down-regulated linked to acquired resistance
    - 13 upregulated genes were selected for siRNA functional studies
      - Reversal of platinum resistance
    - 4 genes significantly resensitised to platinum
      - **PIK3R1**
      - FOLR2
      - HDAC4
      - STAT1

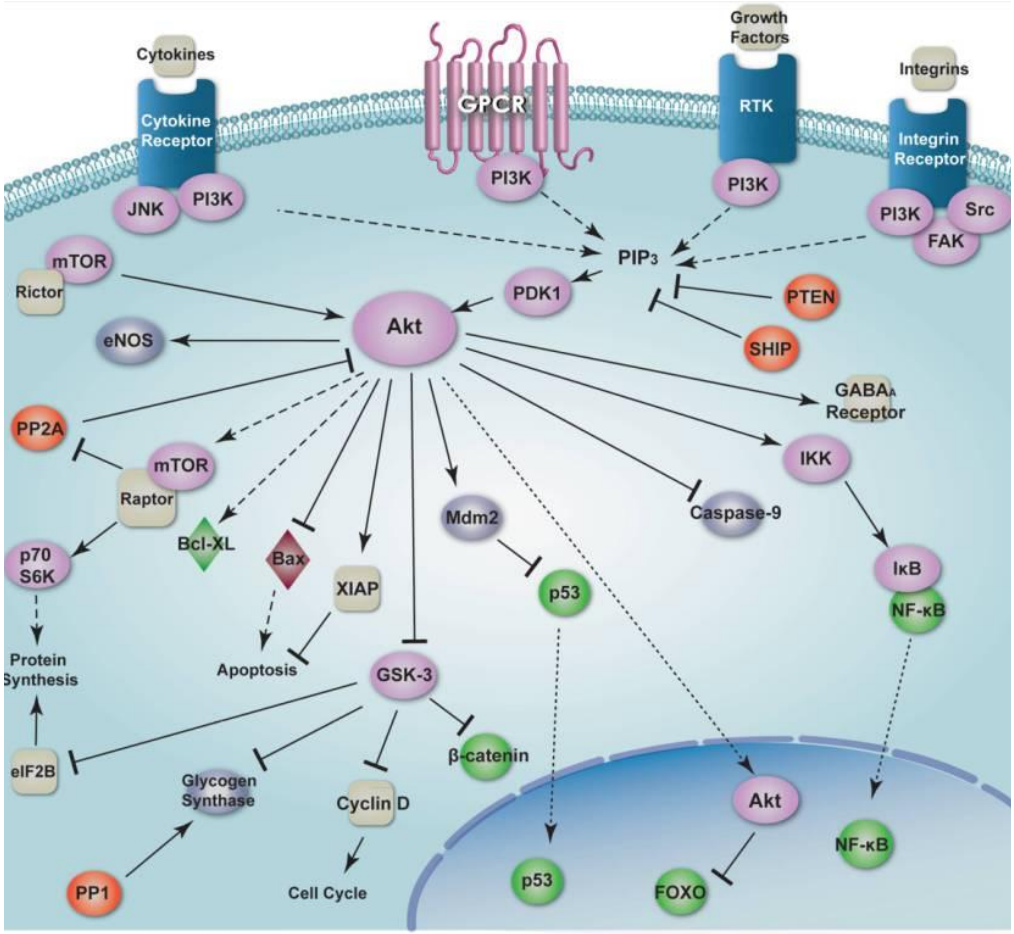
*Langdon et al (1998) Cancer Research 48
§Stronach et al (2011) Cancer Research 71

Imperial College London

Ovarian Cancer Action team

- PIK3R1
  - p85α regulatory subunit of Phospholnositide 3 Kinase
- Performed knock-down experiments on key members of AKT pathway:
  - AKT-3 more effective at resensitising to platinum than AKT-1 or -2
- Other AKT isoform analysis*:
  - AKT3 highly expressed 19/92 primary ovarian tumours
  - AKT3 shRNA inhibited proliferation highest

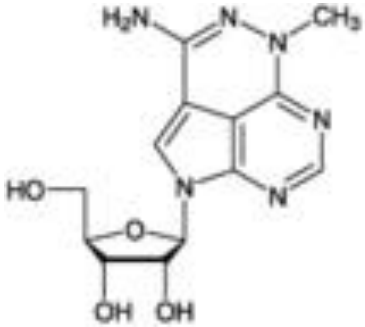
http://www.tocris.com/pathways/pdf/aktUS.pdf



*Cristiano et al (2006) Cancer Research 66

Chemical strategy

- Selected subset of AKT inhibitors in development (IDDB) to synthesise:
  - Diverse:
    - » ~ 12 series; 1-4 examples of each
  - Based on having some biological data (not just biochemistry)
  - Triciribine = “gold standard”



    - » Tricyclic nucleoside - specific inhibitor of AKT:
    - » Inhibits phosphorylaton & activation of AKT
    - » NOT kinase activity of AKT itself NOR known upstream activators (e.g. PI3K & PDK1)

Results

- Not all "AKT inhibitors" resensitised
  - Profiled compounds verses 3 AKT isoforms (pre-activated AKT)
- No isoform specific effect
- However, many had a cell phenotypic effect

Synthesised novel compounds

- "gold standard"- like activity in reversing platinum resistance in phenotypic assay
- Re characterising compounds in panel of biochemical assays : AKT isoforms +/- PH domain; PI3K; DNA-PK

Imperial College London

Successful Drug Discovery in the new environment

- **The right mix of academic (basic & clinical) and multi-disciplinary industrial drug discovery expertise**

- Open partnerships - Industry, Biotech, Academia (basic and Clinical), Research Councils, Medical Charities
  - Access to disease knowledge, compounds, expertise, models
  - Shared and equitable benefit for contributors
- Centre of Excellence model
- Funding schemes are **urgently required** which specifically address Therapeutic Discovery

Imperial College London

Drug Discovery is a team event

Drug Discovery Centre

- Albert Jaxa-Chamiec PhD
- Caroline Low PhD
- Hayley Cordingley PhD
- Katie Chapman PhD
- (Michelle Heathcote PhD)

- Richard Starkey (CRUK projects)

CDC25

- David Mann PhD
- Prof Alan Armstrong PhD
- Kathy Scott PhD
- Katie Judd PhD

Multiple Myeloma ; NFkB pathway

- Prof Guido Fransozo MD PhD
- Laura Tornatore PhD
- Prof Menotti Ruvo PhD

AKT pathway

- Prof Hani Gabra MB ChB PhD
- Euan Stronach PhD