Towards New Medicines?

The Importance of
Biological Target Validation in Drug Discovery

David Haigh
Professor in Medicinal Chemistry & Head of Drug Discovery
Centre for Cancer Research & Cell Biology
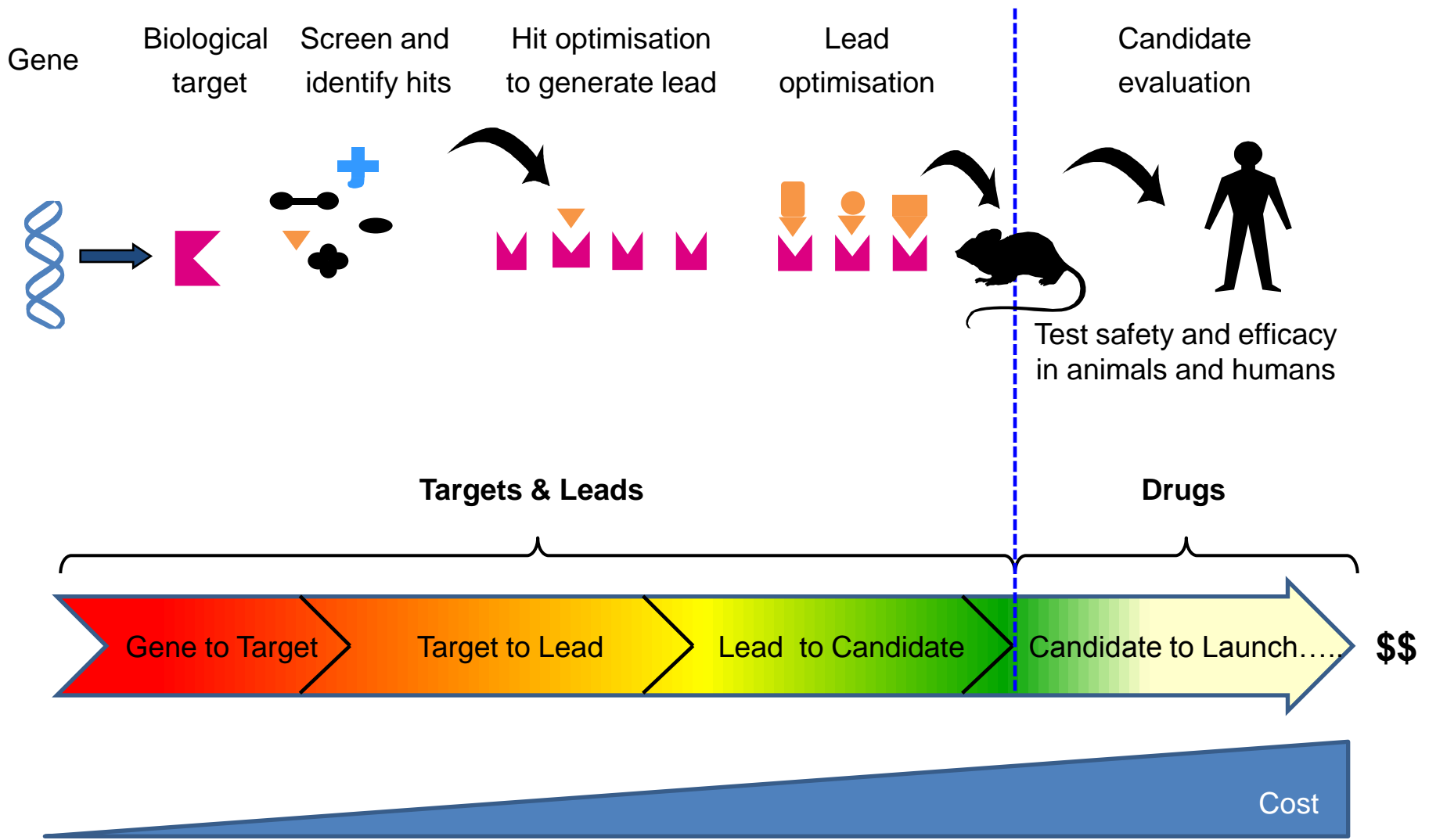**Queen's University, Belfast**

Antiparasitic and Anticancer Drugs Workshop – 9 September 2011

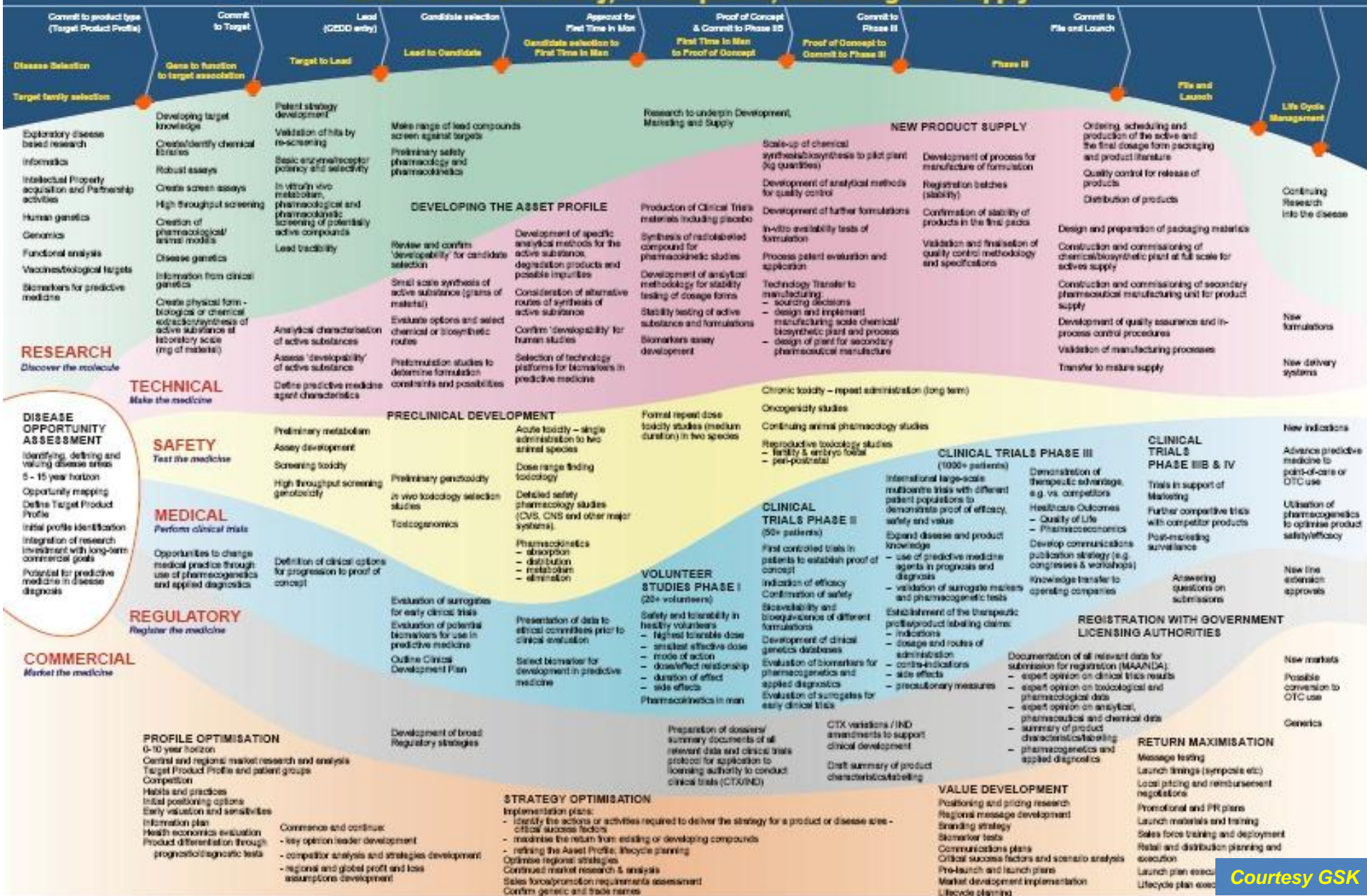# *Linking Cause and Effect......*

Why are firemen found at burning houses?

Does marriage cause suicide?
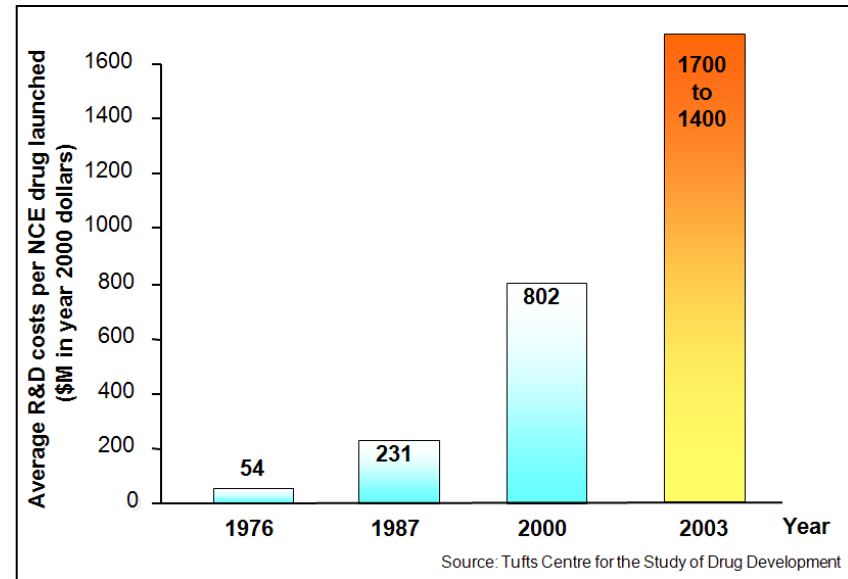
Basics of the Modern Drug Discovery Process

Gene | Biological target | Screen and identify hits | Hit optimisation to generate lead | Lead optimisation | Candidate evaluation

Test safety and efficacy in animals and humans

Targets & Leads **Drugs**

Gene to Target | Target to Lead | Lead to Candidate | Candidate to Launch….. **$$**

Cost

Timelines and costs are not to scale!

The Path to a New Medicine is Astonishingly Complex…….
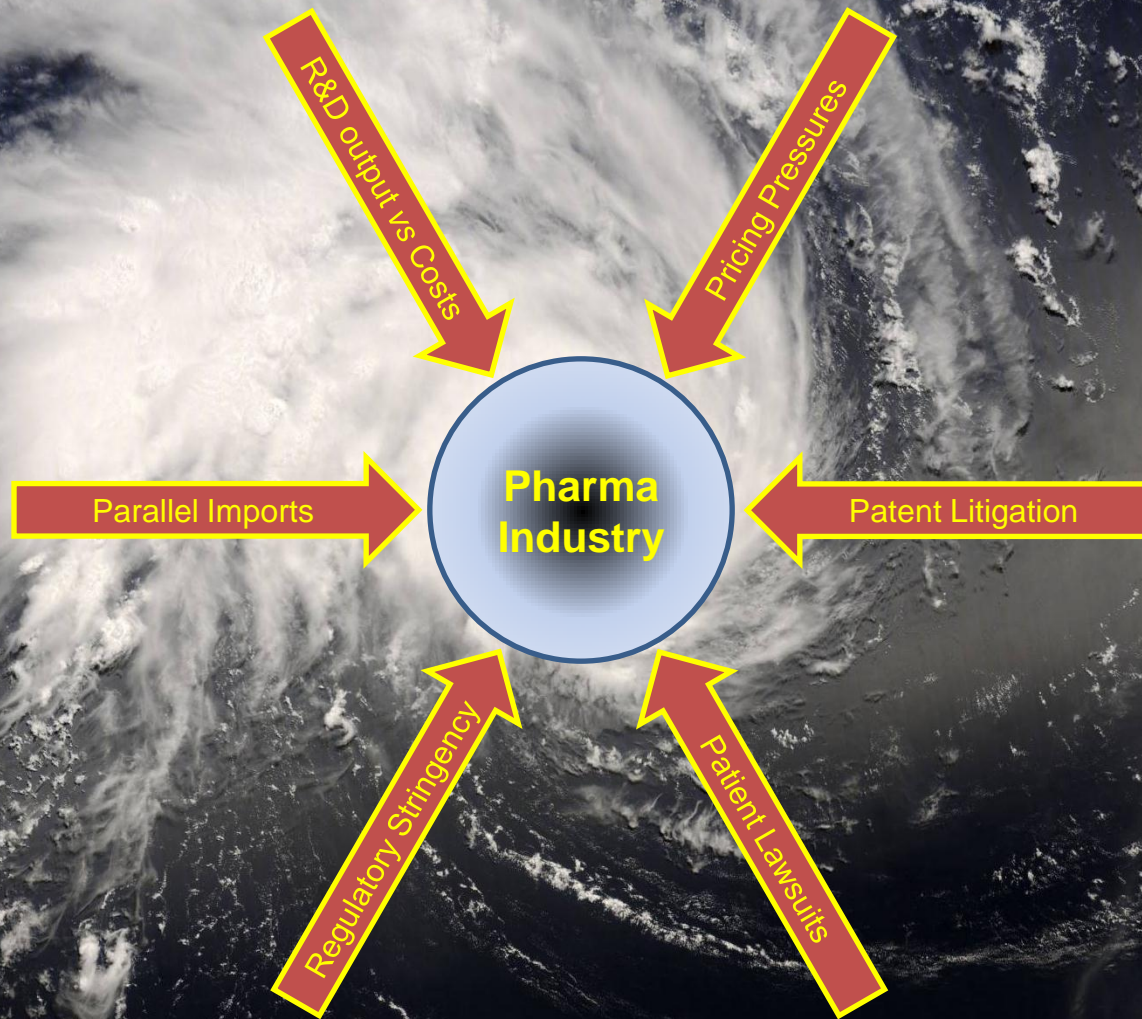

Courtesy GSK

Orthogonality of R&D Output vs Cost
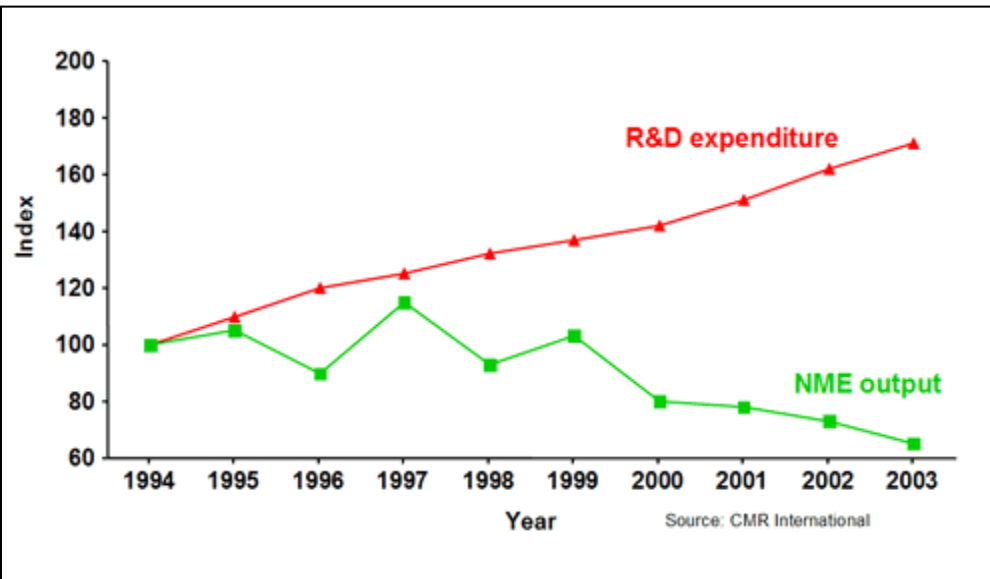
- **The Pharmaceutical industry conundrum:**
    - R&D productivity measured by New Medical Entity (NME) declined 40% (1994-2003)
    - R&D expenditure increased by 70% in same period and 600% over last 30 years
- **Estimated R&D cost per NME was *ca.* $1.4-1.7bn in 2003 (conservatively $2bn 2011)**

- **Overall probability of success (POS) in a new drug reaching market is very low (2%)**
    - 20% Overall POS Target to Candidate Selection (*ca.* 3-6 yrs)
    - 10% Overall POS Preclinical & Clinical Development (*ca.* 4-9 yrs)
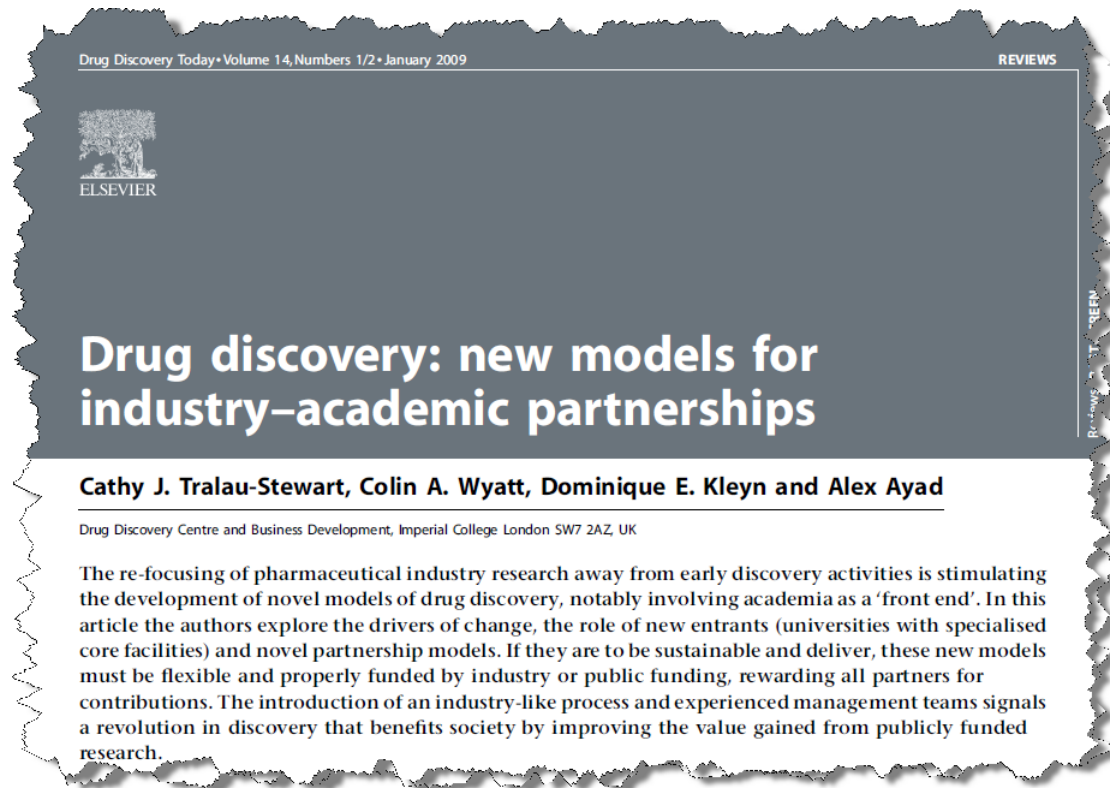   - 33% Overall POS FTIH to market for precedented mechanism (8% unprecedented)

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The Changing Pharmaceutical Landscape: "A Perfect Storm"

R&D output vs Costs

Pricing Pressures

Parallel Imports

Pharma Industry

Patent Litigation

Regulatory Stringency

Patient Lawsuits

"In this new world order, the only viable way to create true value for our patients as well as for our shareholders is through innovation"

Tachi Yamada, former Chairman of R&D, GSK

# *Academia – A Unique Niche in The New Pharma World Order*

Drug Discovery Today • Volume 14, Numbers 1/2 • January 2009 — REVIEWS

ELSEVIER

Drug discovery: new models for industry–academic partnerships

Cathy J. Tralau-Stewart, Colin A. Wyatt, Dominique E. Kleyn and Alex Ayad

Drug Discovery Centre and Business Development, Imperial College London SW7 2AZ, UK

The re-focusing of pharmaceutical industry research away from early discovery activities is stimulating the development of novel models of drug discovery, notably involving academia as a 'front end'. In this article the authors explore the drivers of change, the role of new entrants (universities with specialised core facilities) and novel partnership models. If they are to be sustainable and deliver, these new models must be flexible and properly funded by industry or public funding, rewarding all partners for contributions. The introduction of an industry-like process and experienced management teams signals a revolution in discovery that benefits society by improving the value gained from publicly funded research.

Drug Discovery Today, **2009**, *14*, 95

- **The pharmaceutical landscape is driving an inexorable paradigm change**
- **Big Pharma is suffering from a so-called "Innovation Gap"**
- **Balancing Pharma's internal "R"** *vs* **external innovation – "Portfolio de-risking"**
- **Big Pharma challenge – Can scale leverage niche "First in Class"** *vs* **"Best in Class" ?**
- **Unprecedented opportunity for academic drug discovery, especially in niche markets**

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*Finding an Innovative Drug
in the Biological Target Haystack*

Targets from the Human Genome: A Surplus of Wealth or an Embarrassment of Neglected Opportunity?
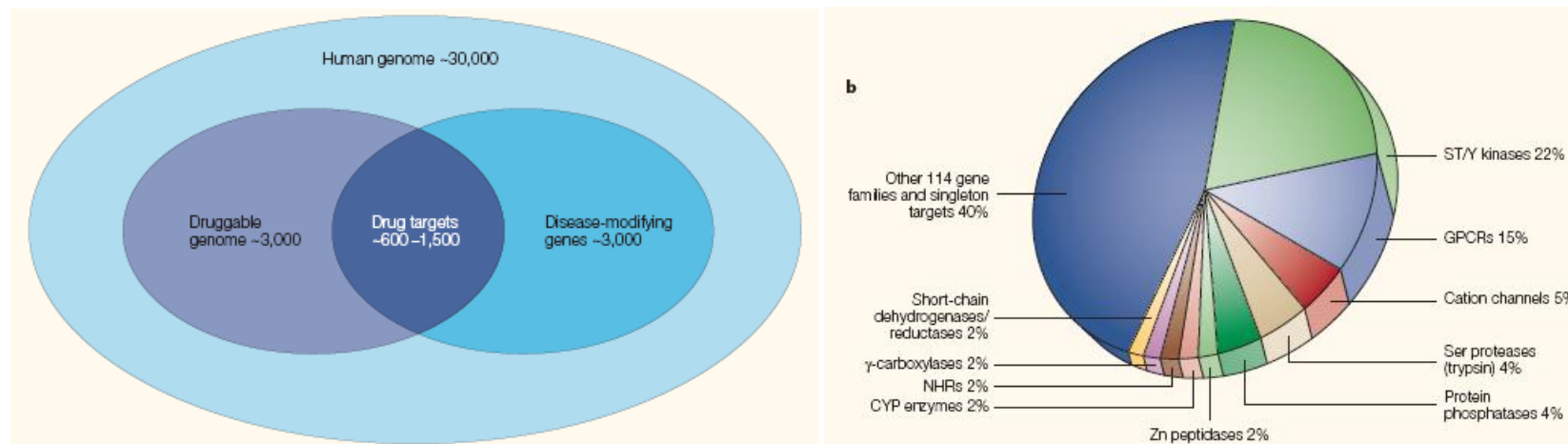

Nature Reviews Drug Discovery, **2002**, *1*, 727

- **Estimate 3,000 drugable genes, perhaps ~600-1,500 real targets implicated in disease**
- **HOWEVER, a 2006 Analysis indicated...**
  - **21,000 Drug products, arising from...**
    - **1357 Unique drugs, of which 1204 are considered "small molecules"**
      - **803 Small molecules are orally active**
        - **These oral drugs act *via* 186 human genome targets**
          - **Only 12-31% of the anticipated "real" targets currently exploited**
            - ➢ **Substantial scope for novel, drugable targets**

Nature Reviews Drug Discovery, **2006**, *5*, 821
Nature Reviews Drug Discovery, **2006**, *5*, 993
Drug Discovery Today, **2007**, *12*, 998

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Gene-Family Distribution of Current Drugs
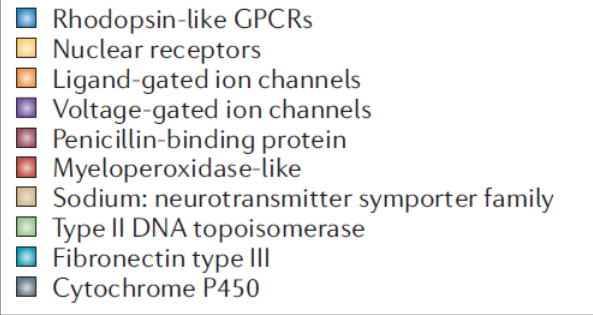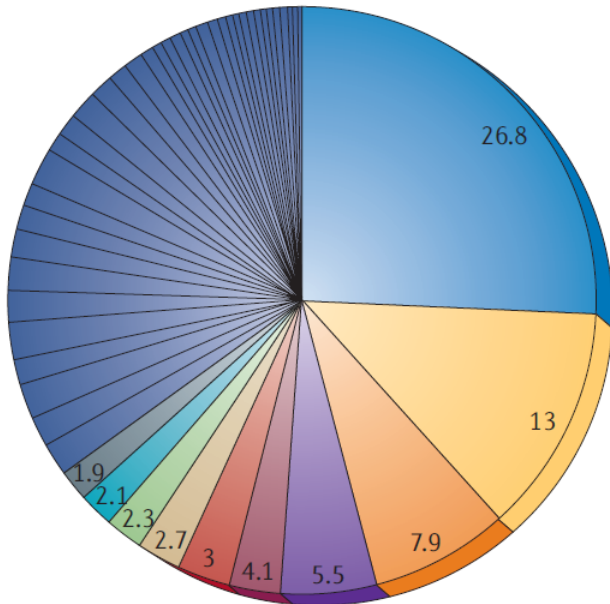
- Family share as % of all FDA-approved drugs is displayed for the top 10 drug families (2005).

- Additionally, there are a further 120 domain families or singletons for which only a few drugs have been successfully launched.

- Based on 1,357 dosed components from >20,000 approved products.

FDA, December 2005.

Nature Reviews Drug Discovery, **2006**, *5*, 993

Nature Reviews Drug Discovery, **2006**, *5*, 821

Legend:
- Rhodopsin-like GPCRs
- Nuclear receptors
- Ligand-gated ion channels
- Voltage-gated ion channels
- Penicillin-binding protein
- Myeloperoxidase-like
- Sodium: neurotransmitter symporter family
- Type II DNA topoisomerase
- Fibronectin type III
- Cytochrome P450

- **GPCRs, Ion Channels & Nuclear Receptors represent >50% of drugged targets**
- **Enzymes represent a large family of targets (not apparent in this classification)**
   - 47% of human genome vs 30% for GPCRs
- **Is the Pharmaceutical industry risk-averse in respect of novel targets?**

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# The Rate of Molecular Target Innovation
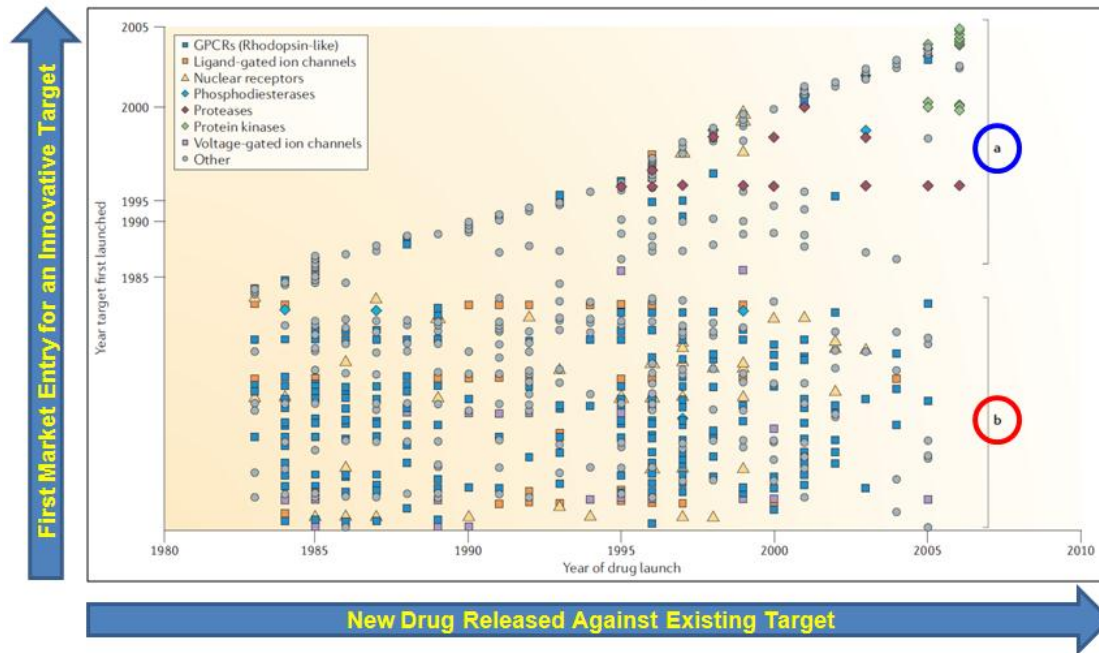
- **Plot shows the rate of innovation *vs* the 'Me-Too' syndrome**

  - More recent targets have higher Y ordinate

  - **Region a** reflects periods of high target innovation (after 1982)

  - **Region b** is predominantly the re-use of established mechanisms

- **Corporate & strategic factors also influence choice of precedented *vs* innovative targets**

  - Efficacy (novel mechanism) *vs* Efficiency (precedented, 'Me-too'; - better potency, PK, IP etc)

- **The average rate of new 'drugged' targets is relatively constant at ~5 per year**

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Biologically Relevant Drug-Target Interactions

- **Four main types of interaction of drug molecules with their biological targets**
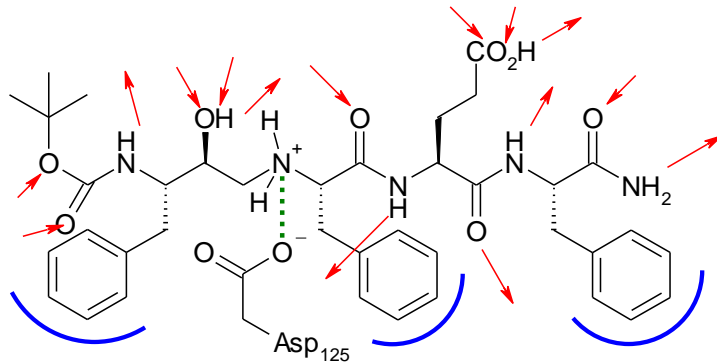
HIV protease inhibitor
Acta Cryst. Sec D, **2006**, *D62*, 489

- **Charge Interactions & Hydrogen Bonds**
  - Strong, reversible binding forces fundamental in biology and drug-target interactions
  - "Structural" water molecules often implicated in H-bonding interactions
- **Hydrophobic interactions**
  - Weak interaction, but drives much of the small molecule affinity for its target
  - Desolvation enhances binding of hydrophobic groups to lipophilic pockets *vs* aq solvent
- **Covalent Interactions**
  - Not generally preferred since irreversible (or very slow off-rate) modification of protein(s)
  - Can lead to failure of body to recognise self, hence causing immunogenicity

The Druggable Genome – What Makes a Good Drug Target?
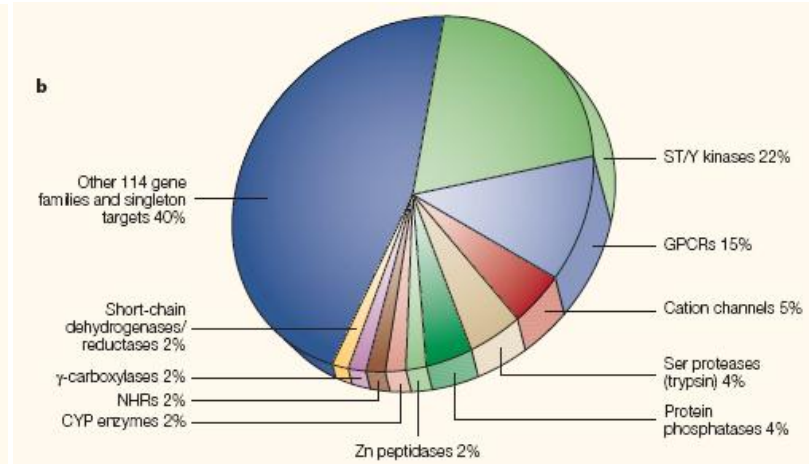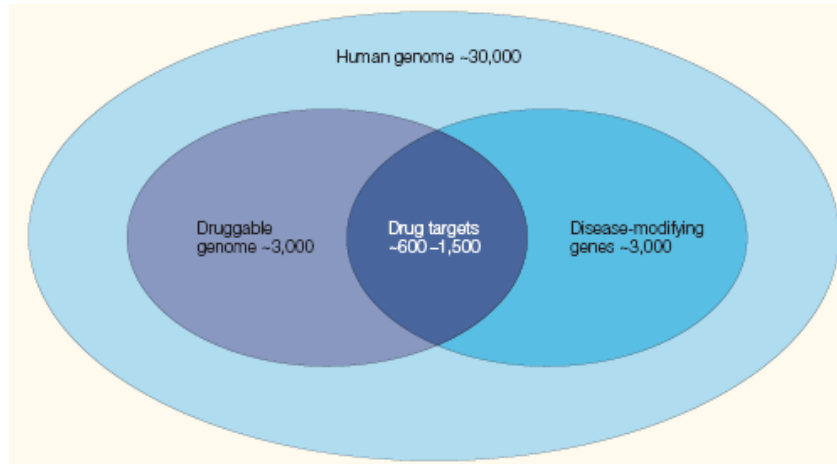
Nature Reviews Drug Discovery, **2002**, *1*, 727

- **Estimate 3,000 druggable genes, perhaps ~600-1,500 real targets implicated in disease**

- **Usually need protein with cleft to allow small molecule to bind and inhibit biological process**
  - Eg Staurosporine bound to GSK3β

- **Target Classes & Systems Biology**
  - GPCRs, Enzymes, Nuclear Receptors, Ion Channels,
  - Integrins,

- **Others?**
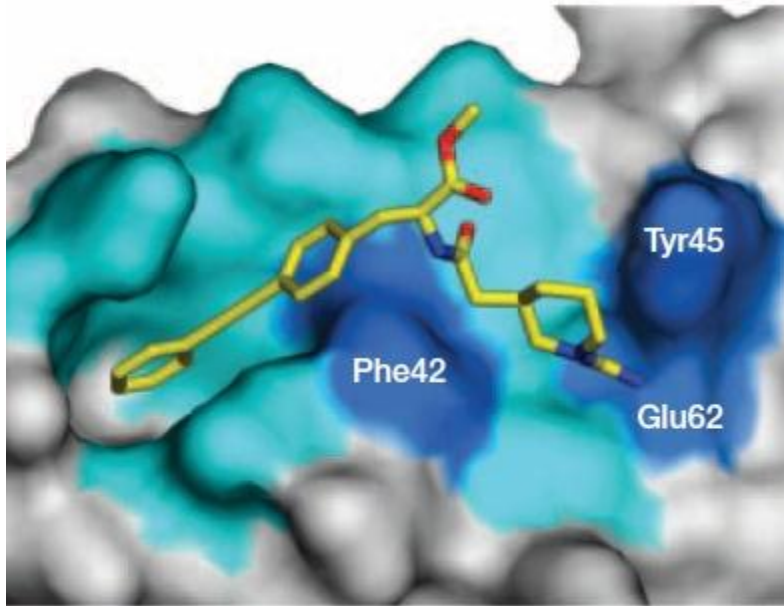 - Protein-Protein, Nucleic Acids, and.........?
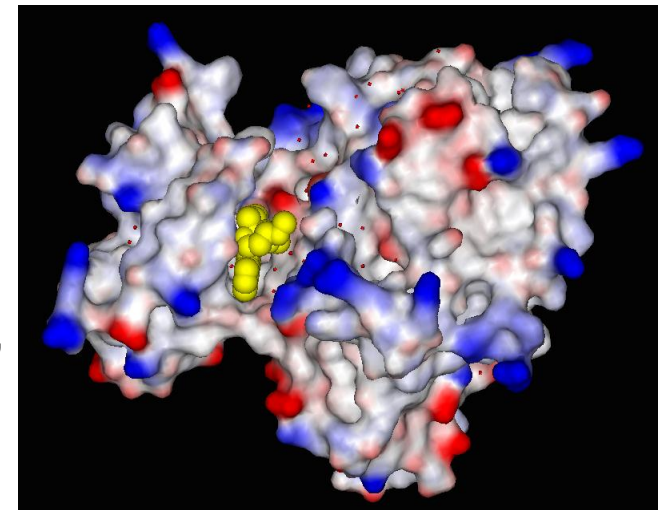
J. Mol. Biol., **2003**, *333,* 393-407

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What About the Hard Targets?

- **The industry is familiar with GPCRs, enzymes, Nuclear receptors, integrins etc**
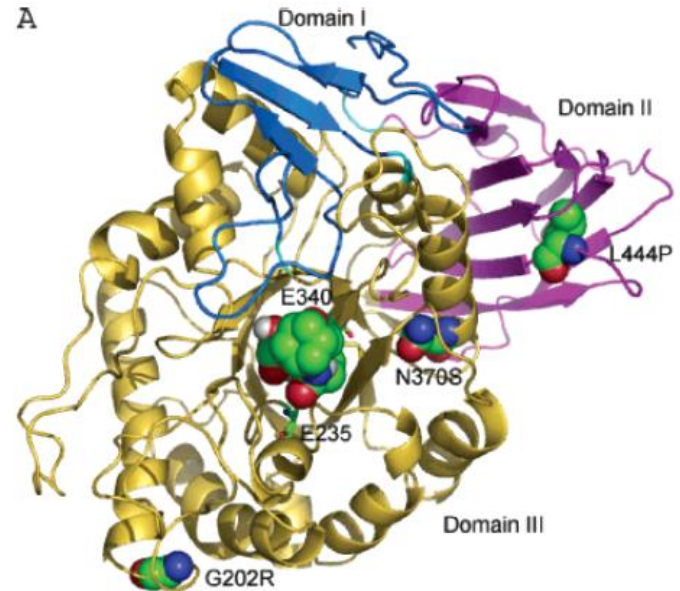- **What about the "undruggable" targets?**

Protein- Protein

- **Ro-26-4550 binds cytokine IL-2 and prevents binding to it's receptor**

    cf Nat. Rev. Drug. Disc., **2004**, *3*, 301
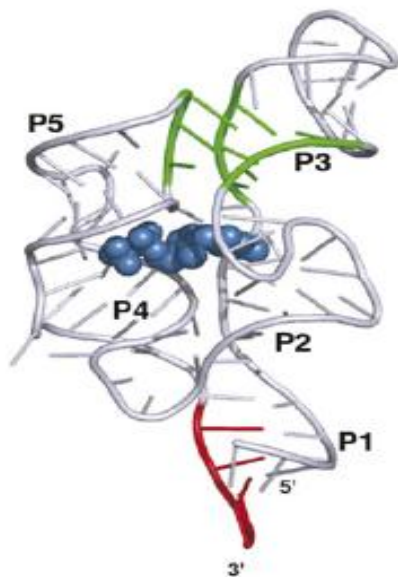

Protein Misfolding

- **Pharmacological chaperones bind & correct folding of glucocerebrosidase**

    J. Med. Chem., **2007**, *50*, 94

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What About the Hard Targets? (ctd)

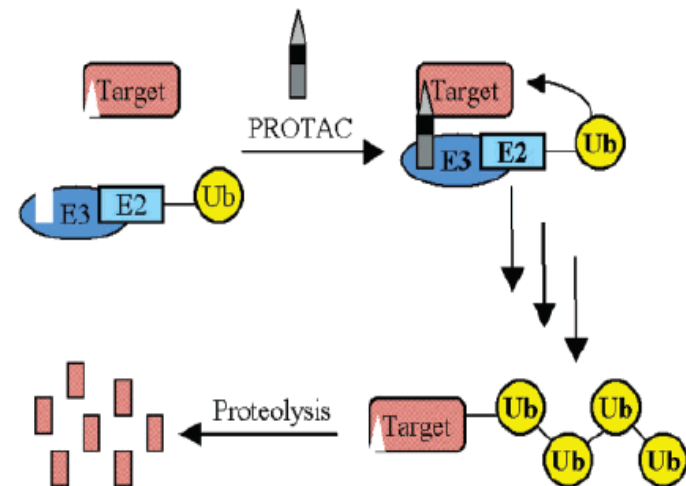## Aptamer Riboswitches

- **Thiamine pyrophosphate metabolite binding to aptamer regulates gene transcription**

  Curr Op Struc Biol, **2007**, *17, 273*

Targeting for UPS Degradation

- **Proteolysis-Targeting Chimeric Molecule targets Androgen Receptor to UPS**
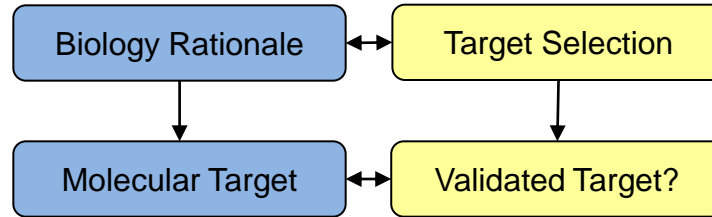
  ChemBioChem, **2005**, *6, 40*

Why Do (Cancer) Drugs Fail?

- **Overall success with Oncology drug development in recent years has been mixed**
- **>30 new cancer treatments were approved by the FDA between 2001-2006**
- **Many of these are antibodies or non-novel, non first-in-class small molecule agents**
    - 2001: Gleevec (Imatinib) – CML (BCR-ABL)
    - 2003: Iressa (Gefitinib) – Metastatic NSCLC (EGFR)
    - 2004: Avastin (Bevacizumab) – Metastatic Colorectal cancer (VEGF)
    - 2006: Zolinza (Vorinostat) – Percutaneous T-cell lymphoma (HDAC)
    - 2006: Herceptin (Trastuzimab) – Breast cancer (ERBB2)

- **Figures for 1990-2000 show only a 5% success rate for Oncology drugs in the clinic**
    - Prior to 1991, poor PK or bioavailability was chief reason for failure (40%)
    - By 2000, this had fallen to 10% by introduction of PK, metabolism & permeability assays
- **Failure now often occurs late in development**
    - Chiefly due to **insufficient therapeutic activity** (30%) and toxicity (30%)

- **Better predictive/molecularly defined animal models & in vitro toxicity models and increasing use of clinical biomarkers to define appropriate patients are helping**

- **BUT, still need better defined targets**

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Target Selection & Validation

```
┌──────────────────┐      ┌──────────────────┐
│ Biology Rationale │◄────►│ Target Selection │
└──────────────────┘      └──────────────────┘
         │                         │
         ▼                         ▼
┌──────────────────┐      ┌──────────────────┐
│ Molecular Target │◄────►│ Validated Target?│
└──────────────────┘      └──────────────────┘
```

- **THE most important aspect of any drug discovery programme is target selection**
- **Easy to assume a target is appropriate for intervention, but much harder to prove....**
- **Better to fail early (and cheaply) than to fail later in the R&D process...!**

Target Validation for Drug Discovery: "Omics" Correlation Does Not Establish Causation

- **Does protein (over)expression cause disease?**

- **Maybe protein (over)expression is a defensive (good) mechanism......**

  Firemen are found at burning houses

  Firemen are not found at normal houses

  Therefore, firemen cause house fire

  Therefore, eliminate firemen to prevent fires

- **Or maybe it is completely coincidental......**

  More weddings occur in June than in any other month

  More suicides occur in June than in any other month

  Therefore weddings cause suicides

  Therefore eliminate weddings to prevent suicides

- **Thorough Target Validation is essential**

See Rydzewski, RM in *"Real World Drug Discovery"*, Elsevier, Oxford, **2008**, p184

Target Validation

- **Link gene to disease**
    - Population-based genetic studies

- **Determine expression pattern in normal vs. disease tissues**
    - Detect target mRNA and/or protein expression pattern using TaqMan, immunohistochemistry, and in situ hybridization

- **Manipulate target**
    - Gene manipulation in mice, e.g. transgenic and knock-out/knock-in mouse
    - Target blocking, e.g. gene interference, antibody, peptides, and tool compounds

- **Elucidate disease pathways or mechanisms of action with tool compounds in in vitro & in vivo assays**
    - How does the tool compound affect the target?

- **A target is never fully validated until the drug is tested in humans**
    - But, reliability is gained by exploiting target classes with proven track records

Nature Reviews Drug Discovery, **2007**, *6*, 902
Current Opinion Chemical Biology, **2004**, *8*, 371

ChemBioChem, **2005**, *6*, 468
Current Opinion in Pharmacology, **2003**, *3*, 571

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# *Target* vs *Phenotype-Based Drug Discovery?*
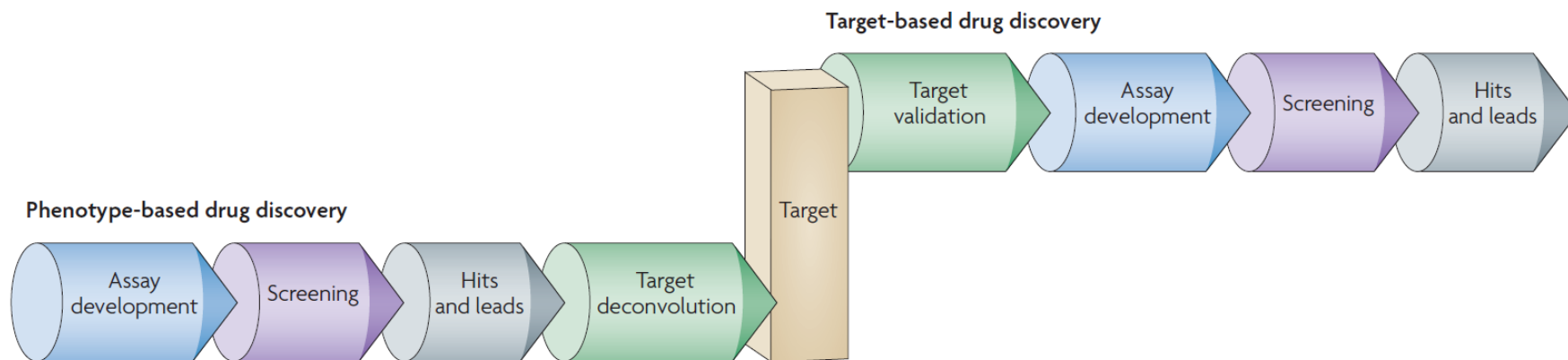
- **Target driven discovery is well exemplified but poorly correlated with clinical efficacy**
  - **-** Is over-reliance on "omics" the root cause of declining pharma innovation & productivity?

- **Phenotypic ("High Content Screening") drug discovery is emerging (*Re-emerging?*):-**
  - Lead molecules obtained first and used to deconvolute & identify target
  - Correlates target with phenotype, hence greater likelihood of clinical efficacy

- **Deconvolution strategies facilitate modern target validation**
  - Affinity chromatography; Yeast-3-Hybrid; Phage Display
  - Gene & Protein Microarray profiling; Biochemical suppression RNAi technology
  - Knockout vs Knockin approaches

- **Key message:-   Patient ➡ Pathophysiology ➡ Target(s) ➡ Screen ➡ Drug(s)**

Nature Reviews Drug Discovery, **2007**, *6*, 902
Current Opinion Chemical Biology, **2004**, *8*, 371

ChemBioChem, **2005**, *6*, 468
Current Opinion in Pharmacology, **2003**, *3*, 571

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Choice of Target Type
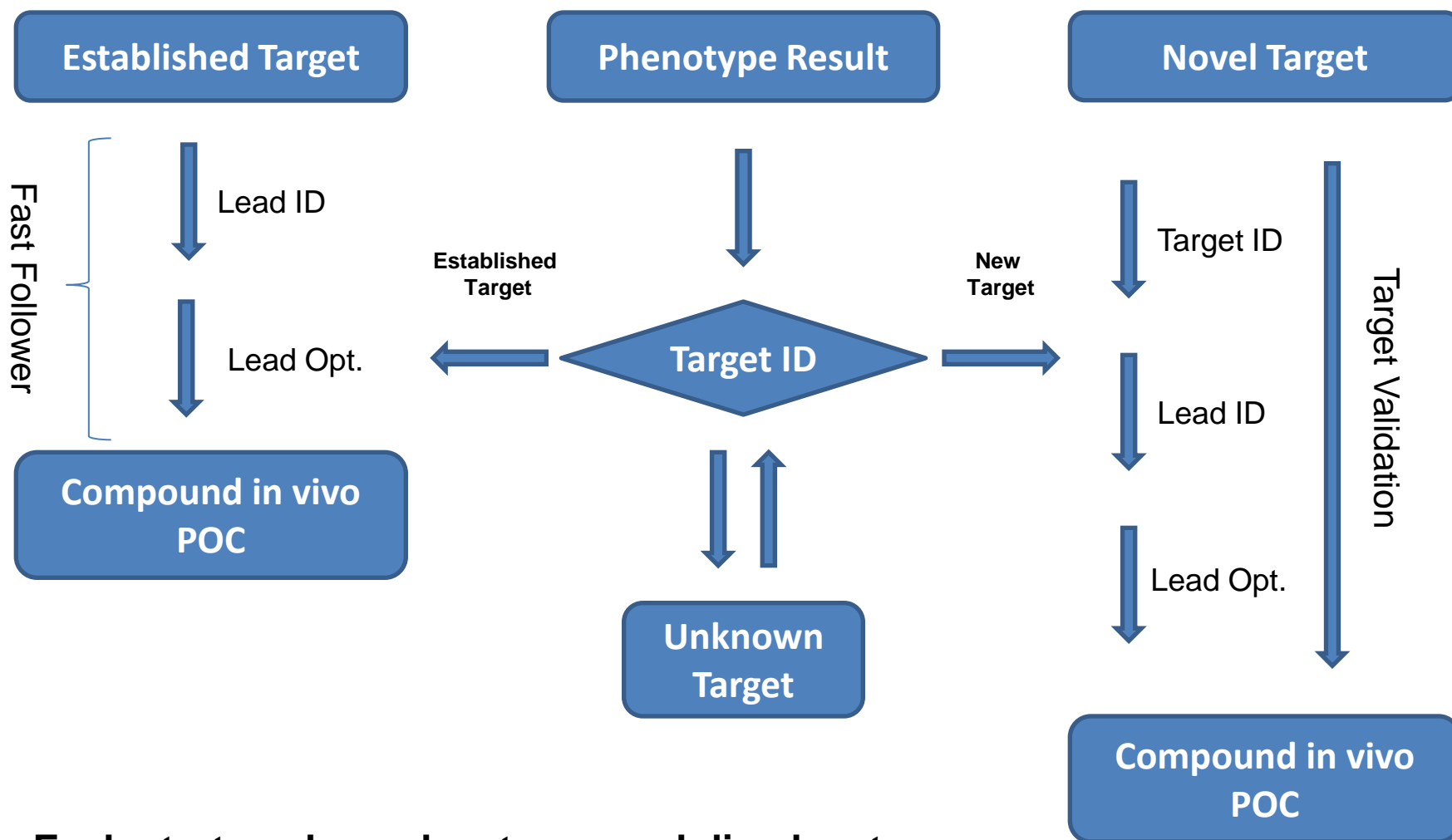
- **Each strategy has advantages and disadvantages**

See Rydzewski, RM in *"Real World Drug Discovery"*, Elsevier, Oxford, **2008**, p176

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Cancer Target Validation – A Special Case?

	Genetics Track	Synergy Track	Lineage Track	Host Track
Concept	• Mutations • Translocation • Expression	• Phenotypically invisible genetic changes	• Differentiation Status • Lineage Markers • Lineage specific signalling	• Angiogenesis • Hypoxia • Stromal Environment
Key Question	Dependence or oncogene addiction	Cancer cell specificity	Appropriate tumour response	Therapeutic Impact
Experimental Approach	RNAi Knockout in cell lines	RNAi Knockout in cell lines Drug sensitivity	RNAi Known modulators Antibodies	Therapeutic Impact
Example	BCR-ABL Gleevec	PARP-BRCA	ER Tamoxifen	VEGF Sutent

- **Cancer cell mechanistic dependencies allow definition of 4 subtypes of targets**
  - Each with its own key question & experimental TV approach

Nature, **2006**, *441*, 451

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Target Validation for Cancer Drug Discovery – Checklist (1)

Specific for Oncology; but general principles apply

- **About the Target**

  - Protein Name & MW; Gene Name

  - Novelty & biological function of target

  - Which pathway is target involved in & function of that pathway

  - Which animal system is being studied (human, yeast etc)

- **Target Validation**

  - Evidence for target mutation/disregulation in human cancer

  - Does disregulation lead to disease phenotype & correlate with clinical findings

  - Is there evidence that disease phenotype is influence by target modulation

  - What is the basis for selectivity for tumour vs normal cells

  - Are there clinically relevant groups of patients and how would they be identified

  - What are the potential adverse consequences of modulating this target

- **Target Drugability**

  - Is the target active catalytically

  - Does the target have a known drugable interaction site & are there known inhibitors

  - Is the target predicted to be drugable based on target class (enzyme, NHR, PPI etc)

  - Availability of crystal structure or homology model

Target Validation for Cancer Drug Discovery – Checklist (2)

- **The Target Screen and Downstream Assays -The Test Cascade**
    - Is there an assay format suitable for HTS & availability of reagents
    - Is there a second, independent assay, for HTS hit confirmation
    - Is there a cellular assay & availability of biomarkers
    - Is the cellular readout compatible with *in vivo* target monitoring

- **Other Target Tools**
    - Availability of antibodies against target protein
    - Is there a matched (Isogenic) pair of +/-cell lines for the target
    - Is there an assay to measure target expression
    - Is there a knockout mouse

- **Chemical Intervention Strategies and Competitive Landscape**
    - External competitive interest, IP, literature coverage
    - Current/anticipated basic science collaborations
    - Availability of tool compounds to aid biology & assay development
    - Chemistry strategies: Libraries; Fast-Follower; SBD approaches & virtual screening

- **Hit and Lead Developability Criteria**

Early Drug Discovery (Simplified!)
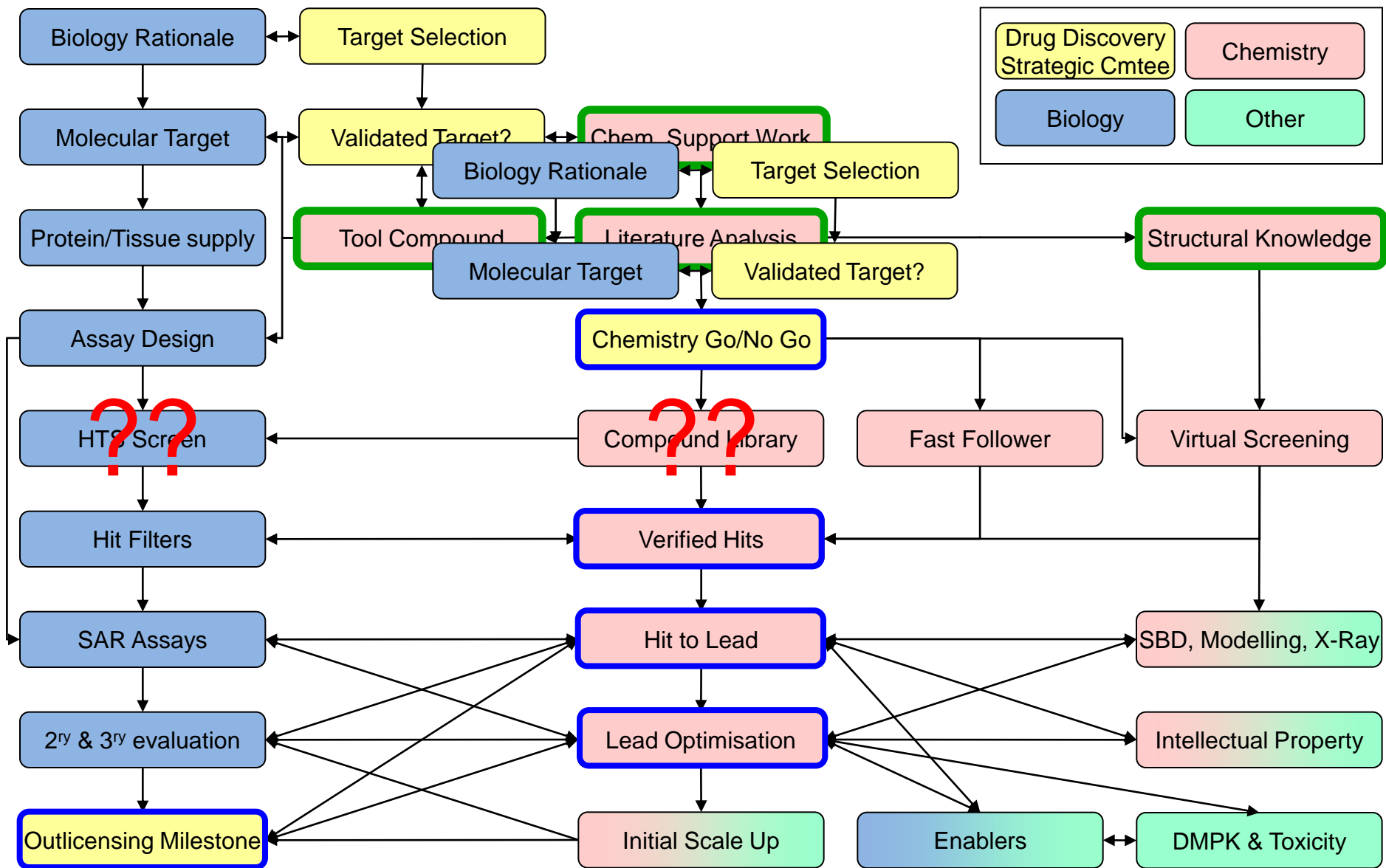
- **Programmes driven by key milestones at each stage**

Milestones for HV, H2L and LO Activities

- **Milestone driven Hit Validation, Hit-to-Lead and Lead Optimisation continuum**
- **Specific parameters likely to vary with each programme, typically including:-**

- **Hit Validation**
  - Potency (%I or %A) confirmed *via* IC_{50} or EC_{50} etc; anticipate <5-20 uM
  - Appropriate profile in hit-filter assays
  - Hit structure validated by NMR, MS and subsequent resynthesis
  - No undesirable functionality
  - Potential for chemical diversity, ease of synthesis
  - IP landscape

- **Hit-to-Lead**
  - Anticipate <1 uM Potency in chemical series
  - Appropriate & intelligible SAR (no flagpole cpds)
  - Acceptable early biological & (non-specific) toxicity profile
  - Evidence of acceptable enabler profile (P450, solubility, permeability etc) and PK
  - Viable synthetic routes to chemistry diversity
  - Scope for IP generation

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Milestones for HV, H2L and LO Activities (ctd)

- **Lead Optimisation**
  - <100 nM Potency with robust SAR
  - Correlated biological profile in 2^{ry} (and 3^{ry}) assays; appropriate assays in place
  - Evidence of PK appropriate to anticipated target profile
  - No series-specific biological profile, toxicity or enabler related issues
  - Established chemistry; scope for scaffold optimisation & additional IP
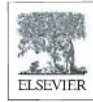
- **Out licensing**
  - Will seek input from potential partners at all stages, where appropriate, in order to ensure acceptable asset profile is being generated

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Work From My Own Laboratory – Watch This Space

With D. Longley

So is it Really That Easy?