TOPRA MSc Module 2

Selection of a Candidate Compound: Studies to Identify Likely Candidates

A presentation by Sally Old, PhD, Exec Director, Early Development, Covance Laboratories Ltd

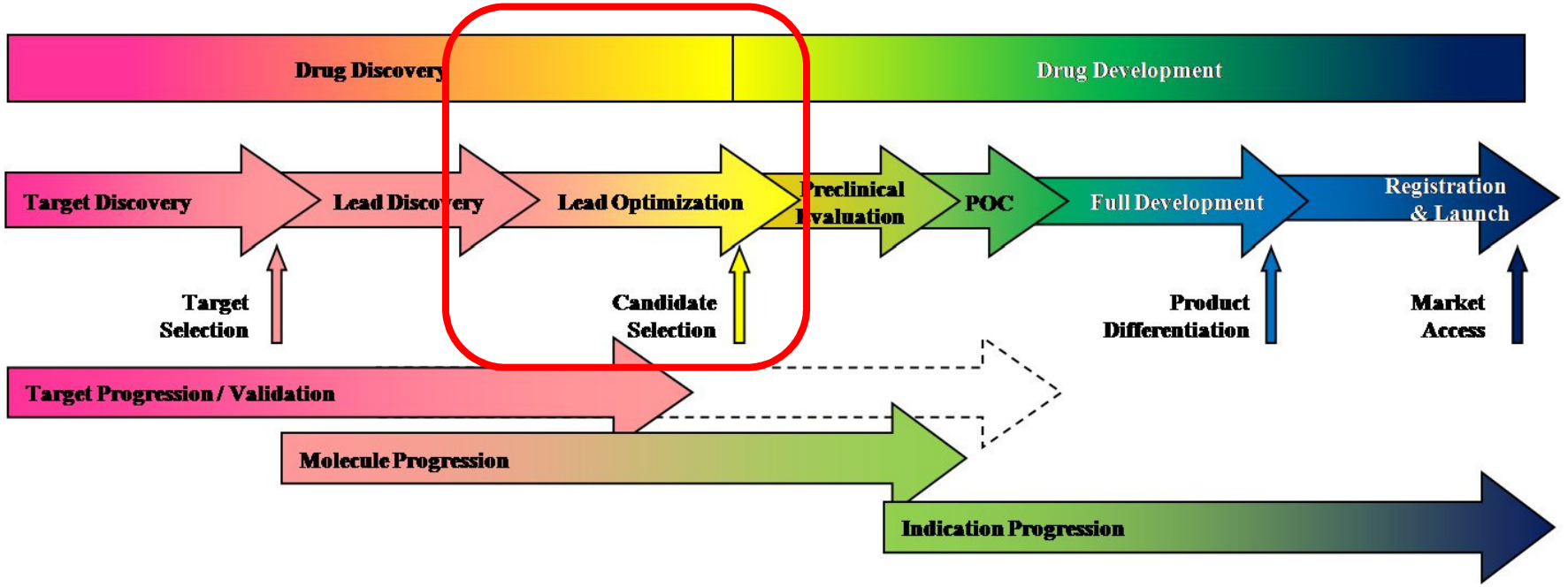
Learning Outcomes

- **Awareness of Drug Discovery process**
  - And how this links into Non-clinical Development
- **Candidate Selection**
  - Key considerations leading to selection of a candidate compound
  - Understanding of types of studies conducted
- **Relevance & impact for future development of compound**

In This presentation we will cover

- **Overview of Drug Development "continuum"**
- **Drug Discovery**
  - Key considerations, role of Lead Optimisation and Candidate Selection
  - "Typical" process and methodologies
  - Regulatory requirements
  - Interpretation and use of data
- **Concluding comments**

The Discovery-Development continuum



Reference: portfoliomanagementsolutions.com; © JamesSamanenConsulting.com

Why is Drug Discovery Important?

Choosing the right compound to develop is critical to the long term success

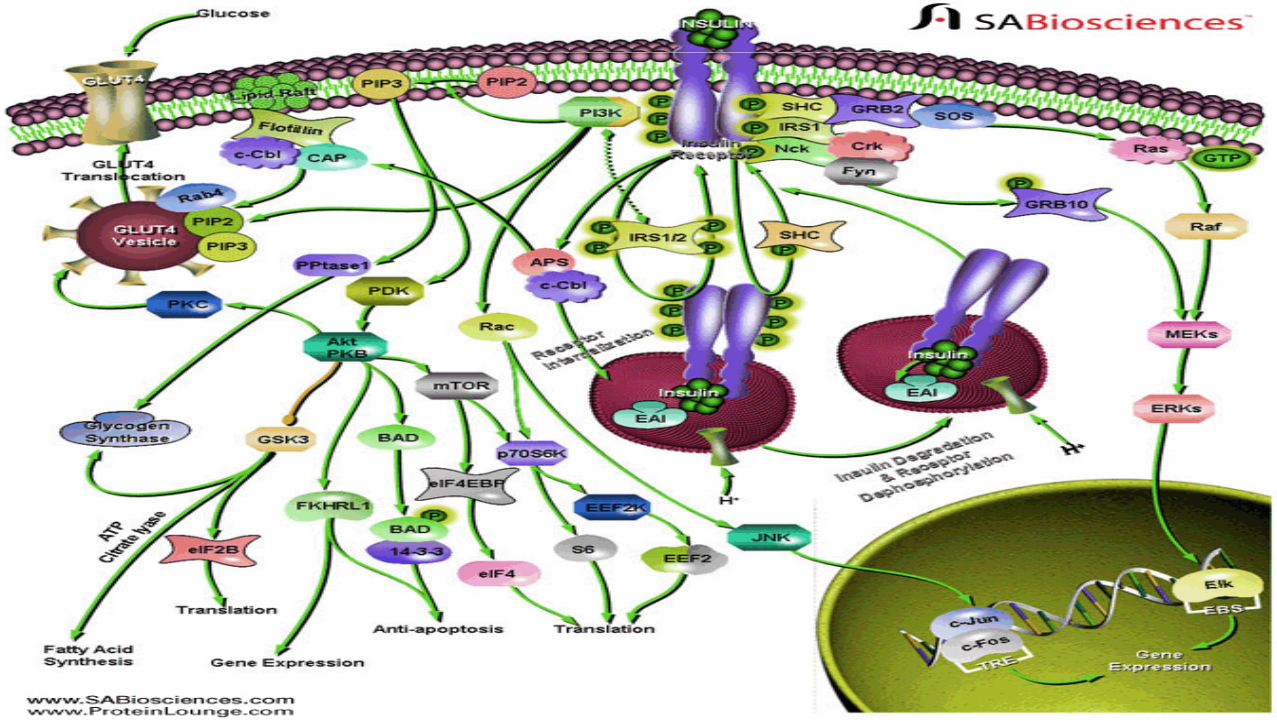
The drug needs to be

- clinically effective
- safe
- cost effective
- reproducibly manufactured in high quality
- commercially viable

Integration of efficacy and safety evaluations in early drug discovery improves the probability of technical success

How are Drugs Discovered?

1. Identify a medical need
2. Understand the underlying mechanism
3. Identify a molecular target
4. Identify a lead compound
5. Identify a commercially viable product
6. Move into drug development



Target Considerations

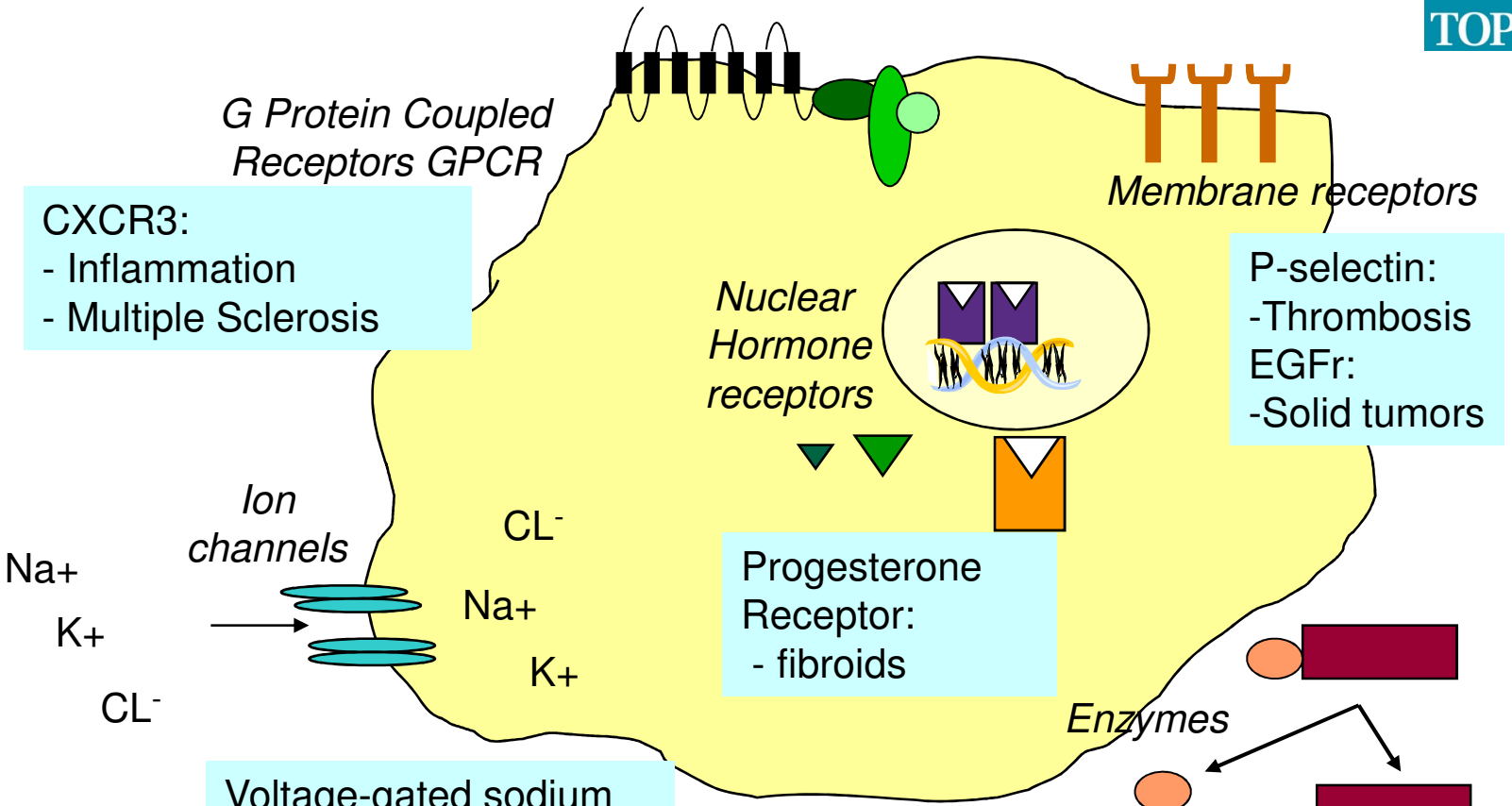
Target Characterization

- Tissue distribution and expression levels
- Pathway
- *In vitro* expression of protein
- Assay development

Target Validation – evidence that affecting the target will modify the disease

- Cell-based models
- Animal models
- Clinical evidence
- Biomarkers

Target Types - Examples



Identification of lead compounds

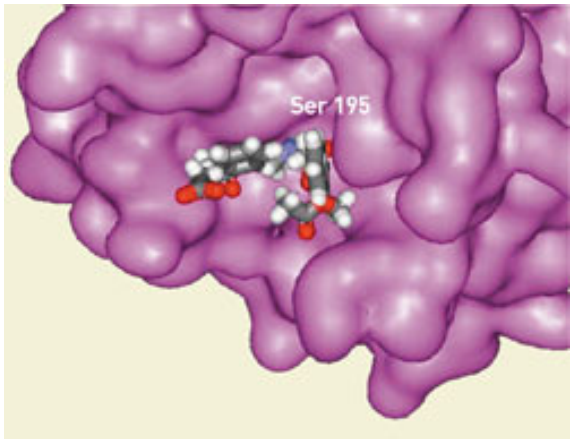
Combinatorial chemistry

- Synthesis of thousands of lead compounds

Chemical libraries

High-throughput screening

- Binding assays
- Activity assays
- Cell-based assays



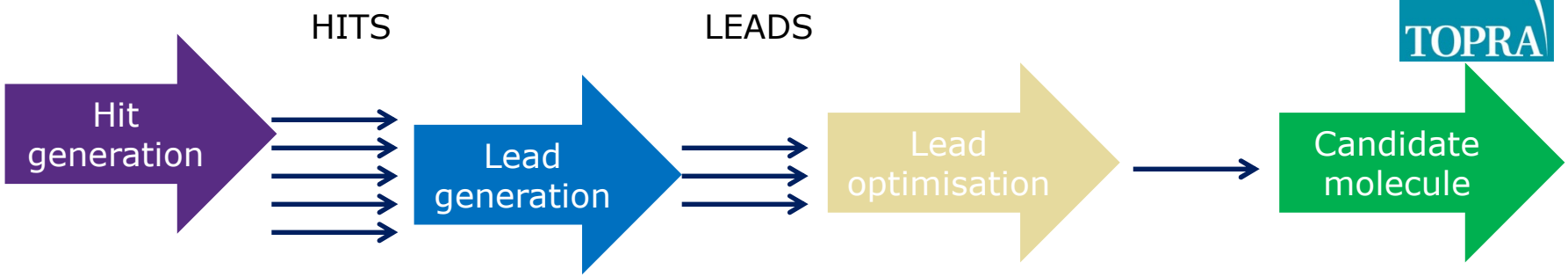
Molecular modeling

- 3-dimensional images of macromolecular binding site
- Structure-based drug design

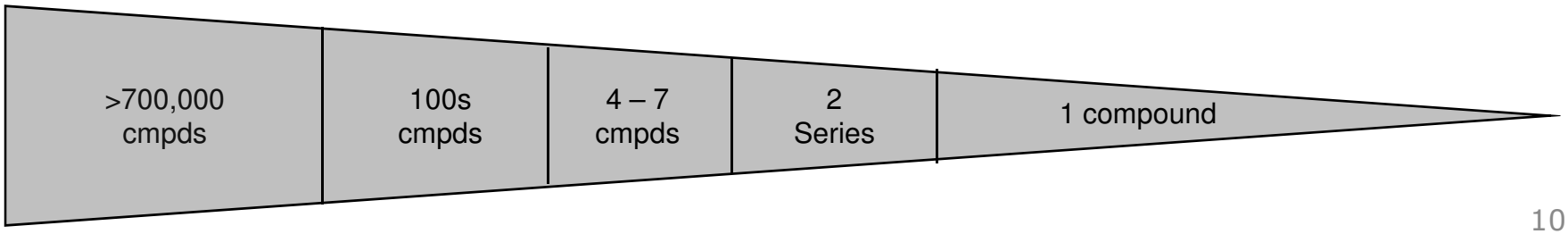
Enhancement of existing compounds

- Improve efficacy, eg statins
- Reduce drug interactions e.g. terfenidine -> fexofenidine
- Improve bioavailability or duration of action

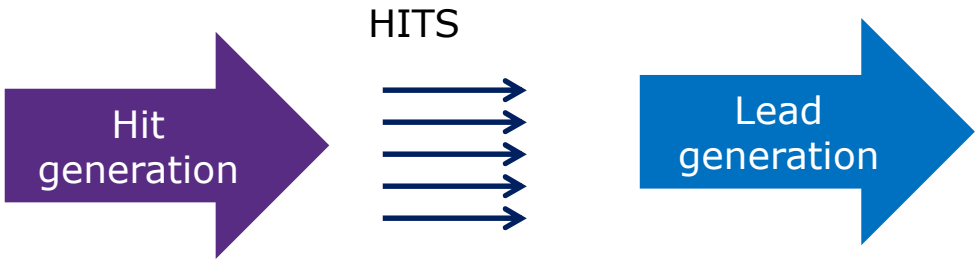
Selection of a candidate compound



- Drug development: starting with the end in mind enables the right foundation to be laid at the start
- Lead optimisation to candidate selection can be time consuming and expensive
- Important to prospectively define how will you select your candidate molecule
- Decide what attributes are important for the molecule
  - For example what is the desired pharmacological profile?
  - Affinity for target, safety, pharmacokinetics and ADME (absorption, distribution, metabolism, elimination, toxicology)
- Design the studies to generate the data to enable the decisions to be made



Drug discovery process and methodologies



- *in vitro* assays
  - Provide ADME properties
  - Physicochemical and pharmacokinetic (PK) measurements
- Examples include
  - Aqueous solubility: Important for *in vitro* assays and *in vivo* delivery of drug
  - Log $D_{7.4}$: Measure of lipophilicity; movement across membranes (e.g. BBB)
  - Hep G2 hepatotoxicity: Surrogate for effects of toxicity on human liver
  - Cytotoxicity in suitable cell line: Likelihood of cellular toxicity *in vivo*
  - *ADME – see following slide*

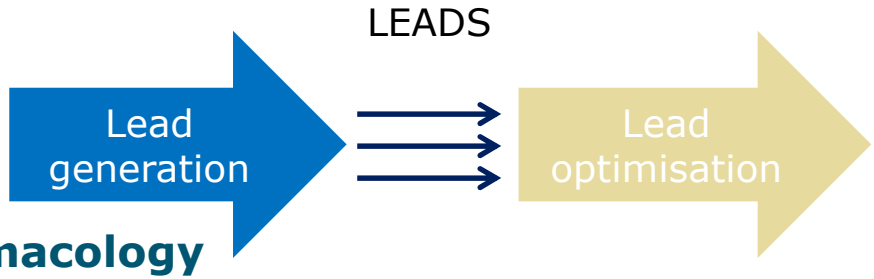
Assessment of DMPK Properties

Determine the "drugability" of the molecule

A	Caco-2 IV/PO PK screening	▪ Moderate to high absorption ▪ Mechanism of absorption ▪ Bioavailability ▪ Animal pharmacokinetics
A/D	P-gp via Caco-2	• Efflux transporter substrate for absorption & impact on CNS uptake
D	RBC distribution* Plasma protein binding Brain to plasma ratio*	• Interspecies comparison to better interpret biological and tox results • Brain uptake for centrally acting compounds or potential for CNS based adverse events
M	Microsomal incubation Hepatocyte incubation CYP IC_{50} PXR activation Reaction phenotyping	• Projection of human clearance • Cross-species metabolite comparison (coverage of human metabolites) • Potential for drug-drug interactions • Potential for CYP or P-gp induction • Role of polymorphic enzymes
E	PK Screening w/ urine analysis	• Animal pharmacokinetics and mechanism of clearance

*Optional based on compound need

Drug discovery process and methodologies



- ***in vitro* Pharmacology**
  - Biochemical Assay
  - Selectivity Assay
  - Cellular Assay
- **PK/PD**
  - *in vivo* PK
  - Multiple doses
  - Different species (support species selection)
- ***in vivo* efficacy**
  - Single and repeat dose to confirm *in vivo* pharmacologic effects
- **Preliminary safety (non-GLP)**
  - MTD studies – to help design pivotal tox studies

In Vitro Assays

Provide a readout in a test tube, plate or cell culture system to identify molecules that "hit" a target

- Activation of epidermal growth factor receptor [EGFR] tyrosine kinase, up-regulated in cancer
  - Measure level of tyrosine kinase autophosphorylation
- Inhibition of tumor necrosis factor [TNF], an inflammatory cytokine with a monoclonal antibody
  - Binding assay to measure affinity
  - Cell-based assay to measure neutralization of cytotoxic effect of TNF

Pharmacokinetic Modeling

In Vivo PK

In Vitro Scaling

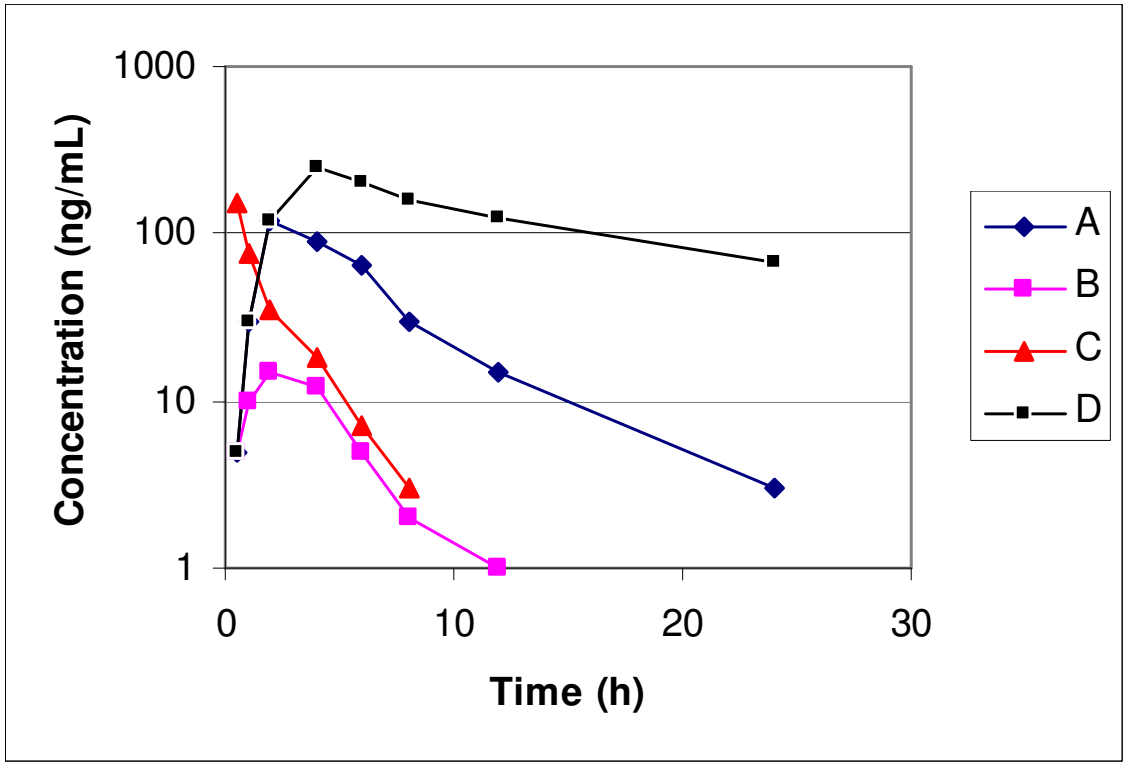
Pharmacology

Clinical Projections

Drive Towards an Appropriate Target Clinical Profile

Pharmacokinetic Screening

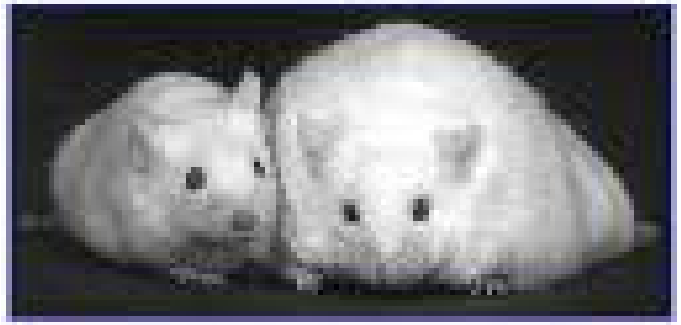
- Pharmacokinetics of drug candidates in animals are often predictive of PK in human
- Test compounds are dosed to rodents, dogs, or primates
- Drug levels measured by high throughput methods
- PK parameters may be optimized based on therapeutic target
  - Onset of action
  - Duration of action
  - Bioavailability



In Vivo Assays

Animal models of disease to show that a molecule not only hits the target but has the desired effect

- Tumors known to express over-activated EGFR could be transplanted into animals to assess the effects of potential inhibitors
  - Effects on tumor growth
  - Effects on EGFR autophosphorylation – Western blot
- For anti-TNF antibodies, measure reduction in inflammatory response in various animal models of inflammation
  - Collagen-induced arthritis
  - Mouse autograft model
- Antidiabetic compounds
  - db/db mouse model
  - Feed high fat diets



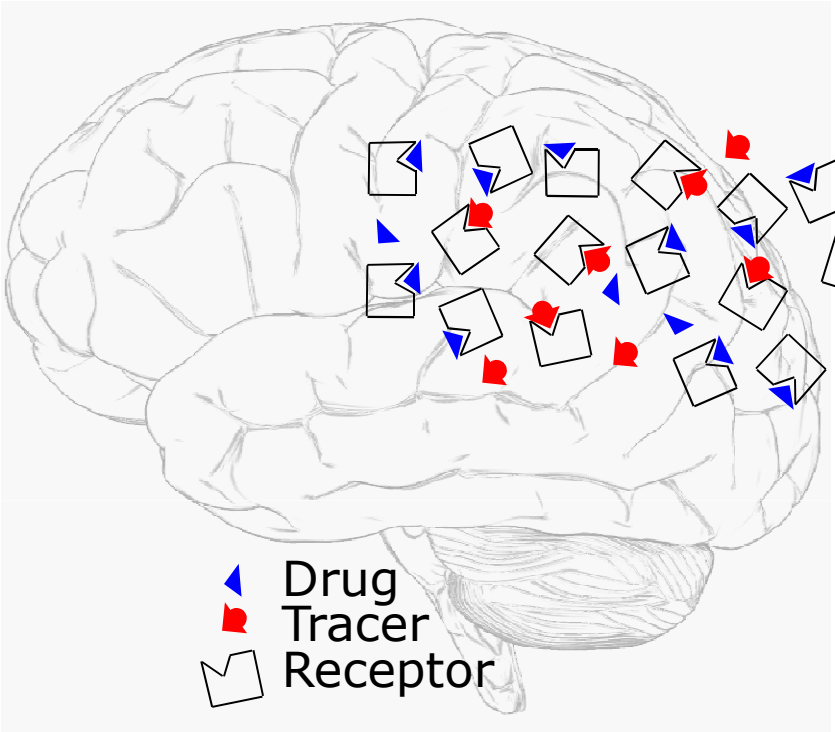
The same assay may be applied to address either safety or efficacy issues; data interpretation is different

- Example: Seizure threshold study
  - Safety Application - Identify adverse effects of compound on convulsive liability
  - Efficacy Application – Identify compound that might be effective in treating epilepsy

Receptor Occupancy [*in vivo*]

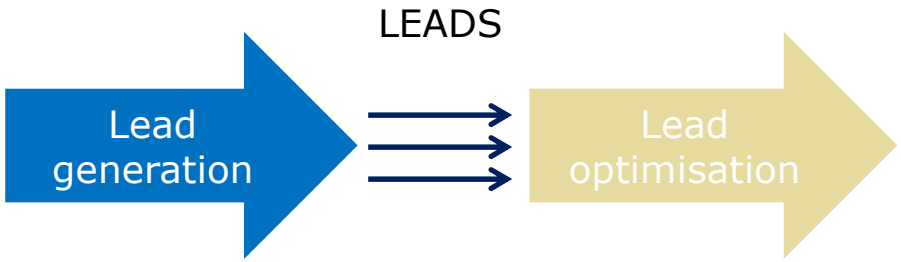
Does the drug reach the target receptor?

- 5-HT1a, 2a, 2c Receptors
- Cannabinoid-1 Receptor
- Kappa Opiate Receptor
- Mu Opiate Receptor
- Delta Opiate Receptor
- Dopamine-1, -2 Receptors
- Serotonin Transporter
- Dopamine Transporter
- Norepinephrine Transporter
- mGlu5 Receptor
- mGlu1 Receptor
- Nicotinic α4β2 Receptor
- Histamine-1 Receptor



1. Dose animal with test compound
2. Dose animal with tracer
3. Analyze tracer bound to receptor

Drug discovery process and methodologies

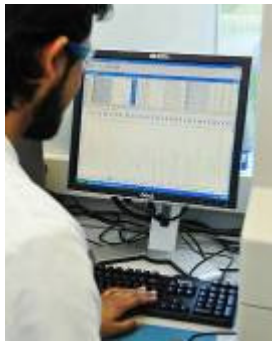


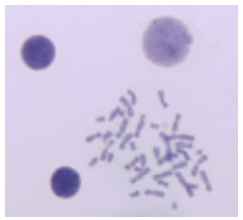
- **Preliminary safety (non-GLP)**
  - MTD studies – to help design pivotal tox studies
- **Depending on the molecule and risk may also conduct**
- **Genotoxicity**
  - *See next slide*
- **Safety Pharmacology**
  - Evaluation can be incorporated into preliminary toxicology/efficacy studies
  - Cardiovascular
  - CNS
  - Respiratory
  - Gastrointestinal

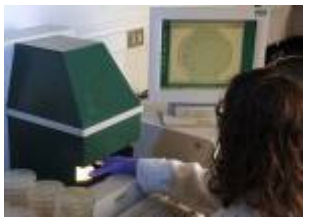
Screening – Genetic Toxicology

Genetic Toxicology

- Philosophy
  - Flexible approach
  - Scaled down Regulatory assays often used (best prediction for later in development)
- Options
  - *in silico* analysis can be useful as first step (e.g. Derek Nexus/ Leadscope)
  - Include *in vitro* cell-based assays for gene mutation & chromosomal damage





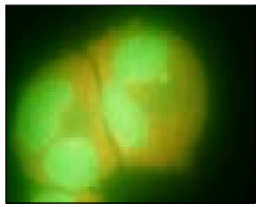


Screening – Genetic Toxicology

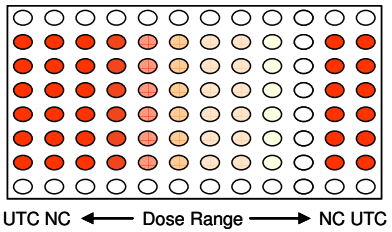
Typical screening assays

- Ames assay (bacterial gene mutation)
  - Normally 2 of the 5 standard strains used (TA98 and TA100)
    OR
  - Mini-Ames test (modified bacterial mutation test)

- *In vitro* micronucleus in mammalian cells (detection of chromosomal damage/aneuploidy)

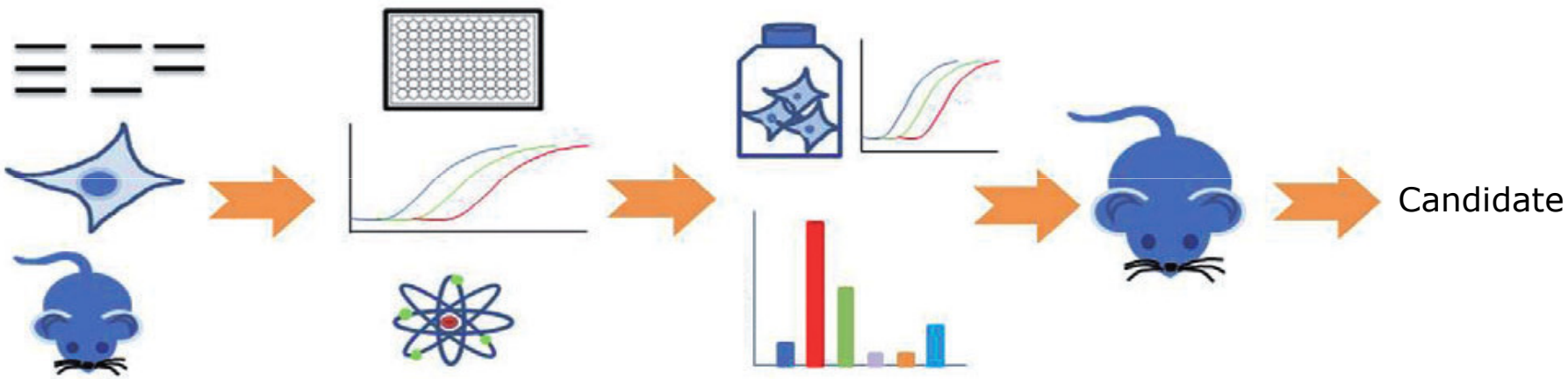


  - Conducted in human lymphocytes
    OR
  - Conducted in cell lines such as CHO / L5178Y / TK6



- Screens for other Genetic Toxicology endpoints and also Cytotoxicity may also be used

Drug discovery process and methodologies - summary



Target validation	Compound screening	Secondary assays	*In vivo* analysis	
•Genetic, cellular and *in vivo* experimental models to identify and validate target	•HTS & selective library screens; structure based design •Reiterative directed compound synthesis to improve compound properties	•*in vitro and ex vivo* secondary assays (mechanistic) •Selectivity & liability assays	•Compound pharmacology •Disease efficacy models •Early safety & toxicity studies	•Preclinical safety & toxicity package

Reference: Hughes JP *et al* (2011) Brit J. Pharmacol . **162** 1239–1249

Regulatory Requirements

GUIDANCE DOCUMENTS: PHARMACOLOGY

- **ICH M3(R2): Non-clinical safety studies for the conduct of human clinical trials and marketing authorisation for pharmaceuticals**
  - Primary PD studies (*in vivo* and/or *in vitro*) intended to investigate the mode of action and/or effects of a substance in relation to its desired therapeutic target
    - Generally conducted during the discovery phase of pharmaceutical development
    - Not conducted in accordance with Good Laboratory Practices (GLP)
    - Can contribute to dose selection for both non-clinical and clinical studies
  - Safety pharmacology (S7A)

Regulatory Requirements

GUIDANCE DOCUMENTS: PHARMACOLOGY

- **S7A Safety Pharmacology Studies for Human Pharmaceuticals**
  - Studies that investigate the potential undesirable pharmacodynamic effects on physiological functions in relation to exposure in the therapeutic range and above
  - Primary: Studies on the mode of action and or effects in relation to the desired therapeutic target
  - Secondary: Studies on the mode of action and/or effects not related to the desired therapeutic target
  - Rational Approach in Design and Conduct
  - Based on Pharmaceutical's Properties and Uses Scientifically Valid Methods
    - Use of New Technologies and Methodologies is encouraged
    - Potential to Incorporate SP Endpoints into Toxicology, Kinetics, Clinical studies etc.
- **ICH S7B: The Non-clinical Evaluation of the Potential for delayed Ventricular Repolarization (QT Interval Prolongation) by Human Pharmaceuticals**
- ICH S7A and S7B are discussed in detail in another presentation

Regulatory Requirements

GUIDANCE DOCUMENTS: PHARMACOLOGY

- **ICH S9: Non-clinical evaluation for anticancer pharmaceuticals**
- **Pharmacology studies**
  - Prior to Phase I studies
  - Preliminary characterization of the mechanism(s) of action and schedule dependencies
  - Anti-tumor activity of the pharmaceutical should have been made
  - Appropriate models should be selected based on target and mechanism of action
  - These studies can:
    - provide nonclinical proof of principle;
    - guide schedules and dose-escalation schemes;
    - provide information for selection of test species;
    - aid in start dose selection and selection of investigational biomarkers, where appropriate; and,
    - if relevant, justify pharmaceutical combinations
  - Understanding the secondary pharmacodynamic properties of a pharmaceutical could contribute to the assessment of safety for humans, and those properties might be investigated as appropriate

Regulatory Requirements

GUIDANCE DOCUMENTS: PHARMACOKINETICS

- **ICH S3A: Toxicokinetics - A Guidance for Assessing Systemic Exposure in Toxicology Studies**
  - The primary objective of toxicokinetics is to describe the systemic exposure achieved in animals and its relationship to dose level and the time course of the toxicity study.
  - Secondary objectives are:
    - to relate the exposure achieved in toxicity studies to toxicological findings and
    - contribute to the assessment of the relevance of these findings to clinical safety.
    - to support the choice of species and treatment regimen in non-clinical toxicity studies.
    - to provide information which, in conjunction with the toxicity findings, contributes to the design of subsequent non-clinical toxicity studies
  - If the tox study is to GLP the TK assessment needs to be to GLP

Regulatory Requirements

GUIDANCE DOCUMENTS: PHARMACOKINETICS

- **Guideline on the non-clinical development of fixed combinations of medicinal products (EMEA/CHMP/SWP/258498/2005)**
- Potential additive, potentiation or antagonistic effects of the compounds when used together with respect to the pharmacology, pharmacokinetics or toxicology of the combination under development
- What you do (which studies and their design) to support them from a non-clinical perspective depends on what they are:
  - Fixed combination of compounds already approved as free combination therapy
  - Fixed combination of approved compounds NOT approved as free combination therapy
  - Fixed combination containing one or more New Active Substances

Candidate Selection: Interpretation and use of data

Selecting the right molecule to advance

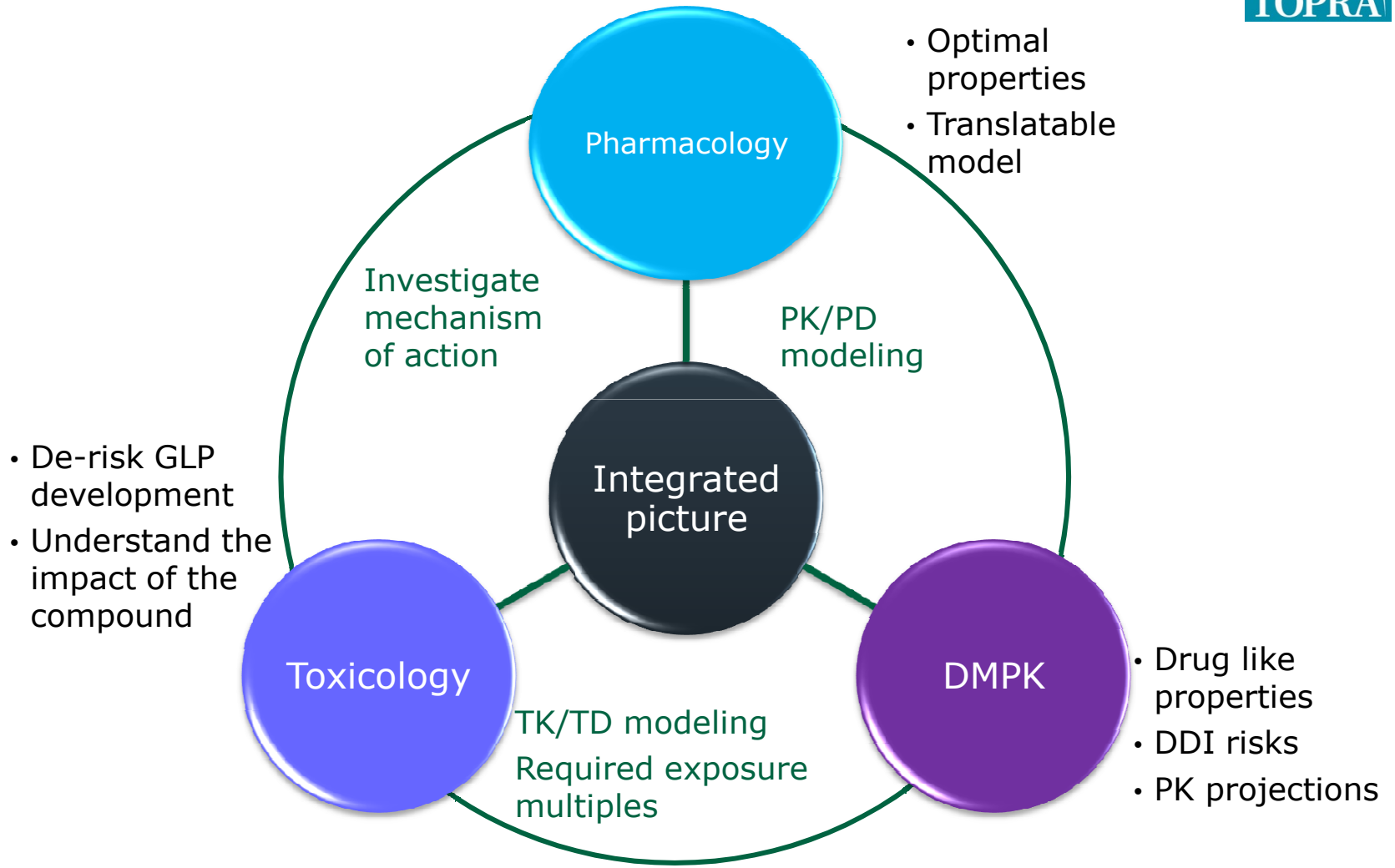
Product Target Profile

- Indication
- Dosing
- Potency
- Market landscape

Defined Scientific Endpoints

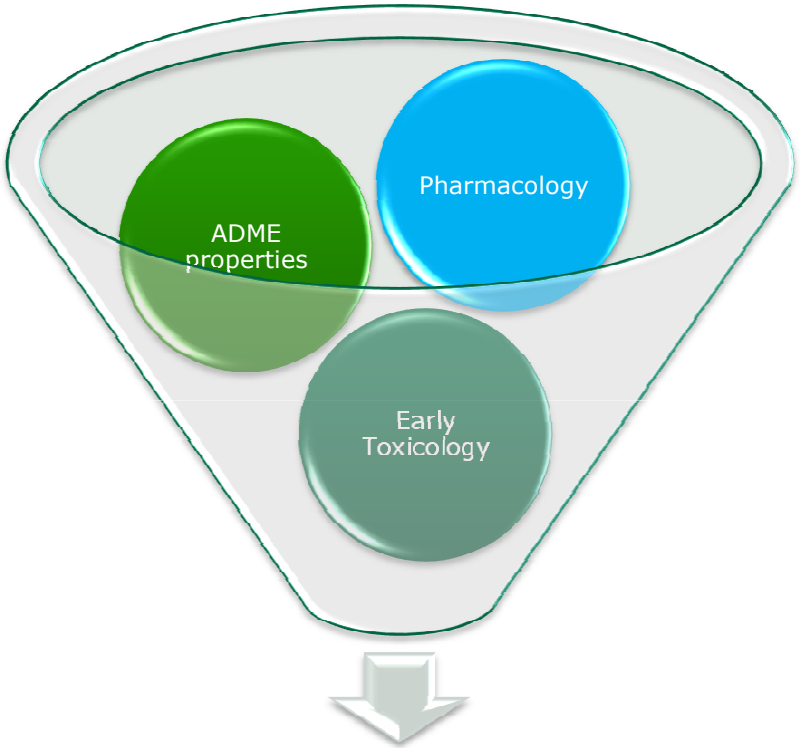
- Receptor interaction
- Performance against pharmacological model
- Active comparators
- Predicted clinical properties
- Potential toxicology outcome

Candidate Selection: Interpretation and use of data



Advancing the Molecule

Bringing the data & expertise together for decision making



Candidate Molecule

- Collation of individual scientific results
- Assess the experimental results against the Product Target Profile
- Choose the best molecule to advance

Medical Need vs. Commercial Viability

Risk/Benefit Ratio

- What is the feasibility of developing a drug with a favorable risk/benefit ratio?

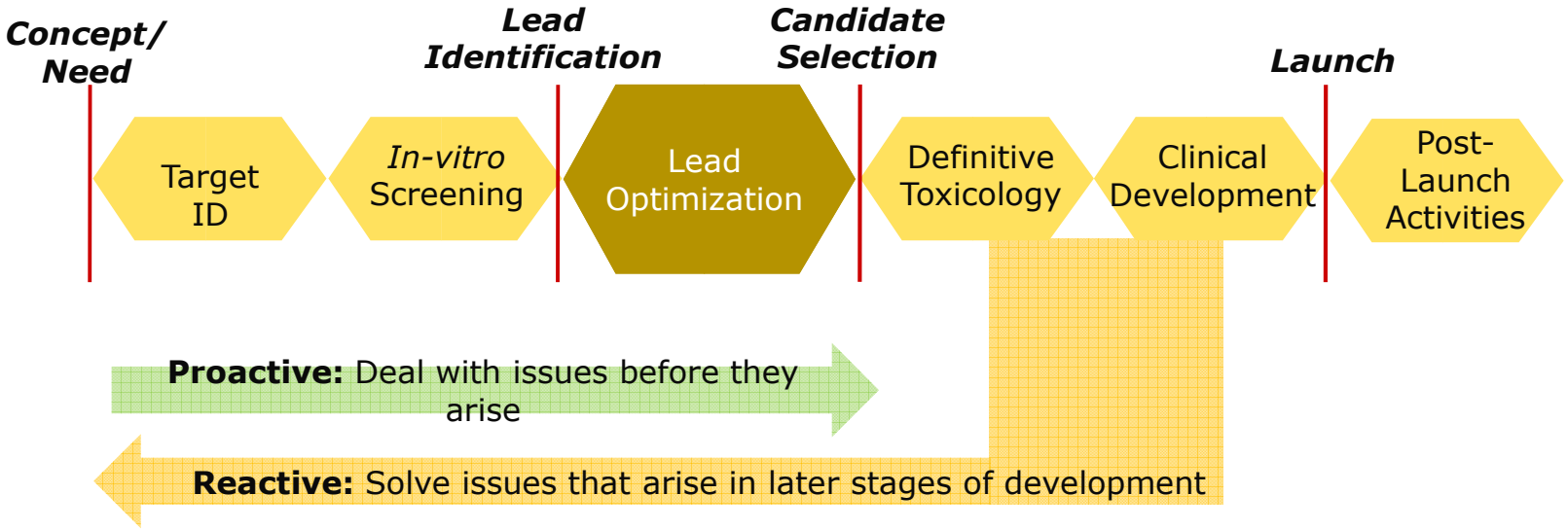
"Drugability"

- What is the feasibility of delivering a therapeutic to the target

Commercial Viability

- What is the likelihood that the developed drug will recover development costs and return a profit?
  - Commercial discussions are happening earlier and earlier
  - Includes regulatory environment and cost/benefit discussions

The Drug Development [re]Cycle



Integrating Safety and Efficacy improves the probability of technical success

In this presentation we covered

- **Overview of Drug Development "continuum"**
- **Drug Discovery**
  - Key considerations, role of Lead Optimisation and Candidate Selection
  - "Typical" process and methodologies
  - Regulatory requirements
  - Interpretation and use of data
- **Concluding comments**

Recommended references

- **Hughes J.P., Rees S., Kalindjian, S.B., Philpott, K.L. Principles of early drug discovery. Br J Pharmacol, 2011; 162(6): 1239–1249**
- **Pritchard J.F., Jurima-Romet M., Reimer M.L., Mortimer, E, Rolfe B, Caven M.N. Making better drugs: Decision gates in non-clinical drug development. Nat Rev Drug Discov. 2003** Jul;2(7):542-53.
- **Hefti, F. Franz. Requirement for a lead compound to become a clinical candidate. BMC Neurosic 2008; 9(Suppl 3): S7.**
- **ICH series of guidelines**

QUESTIONS?

Acknowledgements

William Hanlon, Shawn Heidel, Mark Holbrook, Michelle Scott, Marina Seme Nelson, Margaret Urwin [Covance colleagues]

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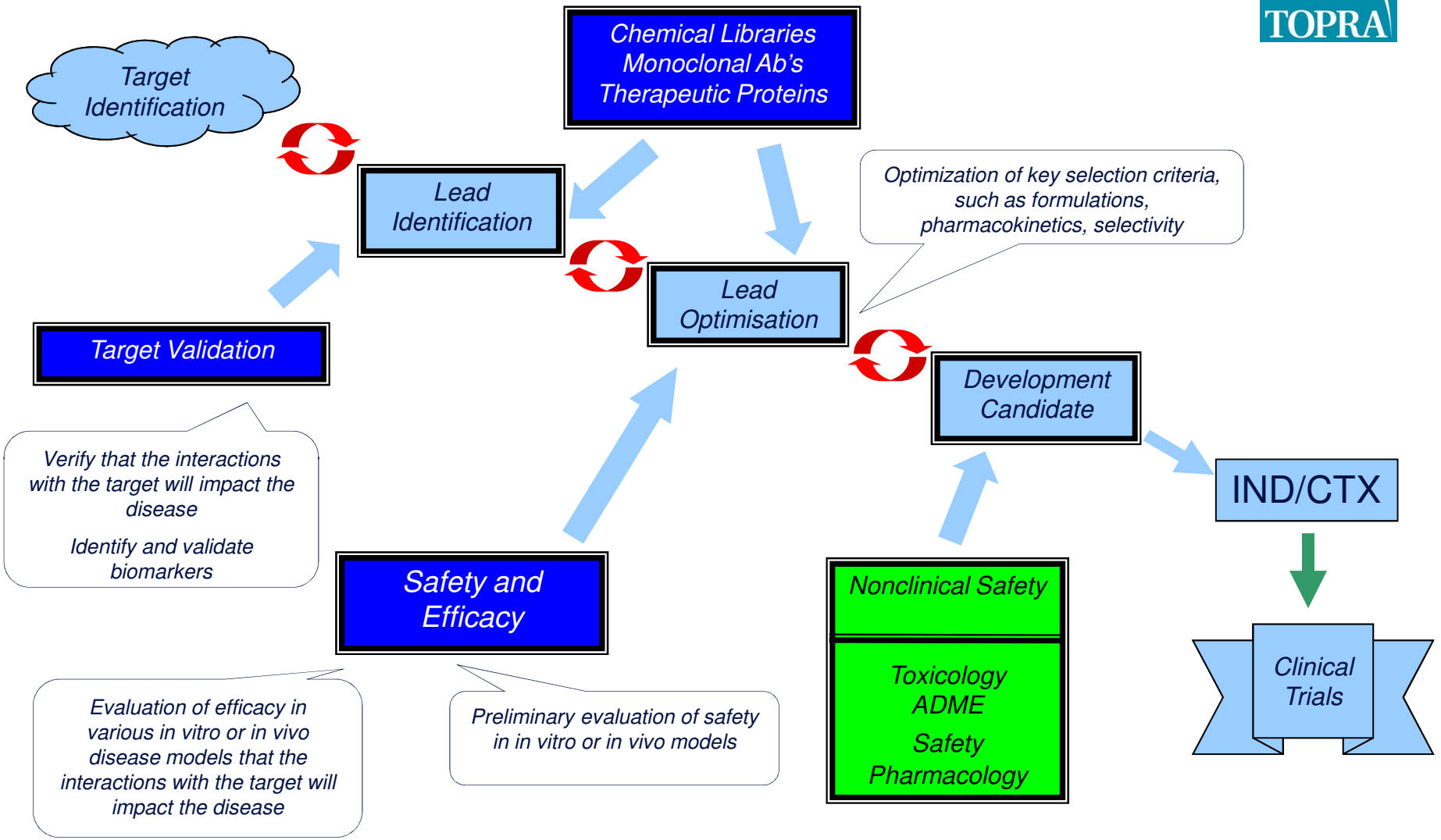
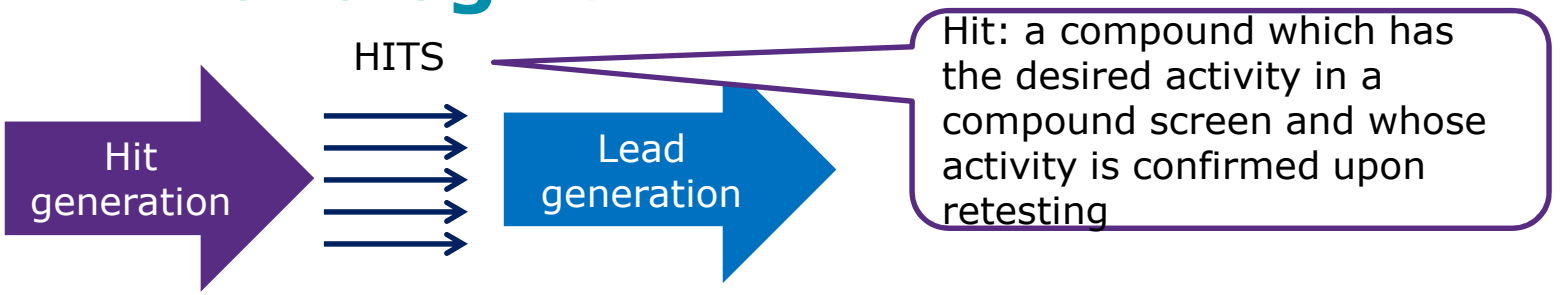
Back-up slides

Drug Discovery Process; summary

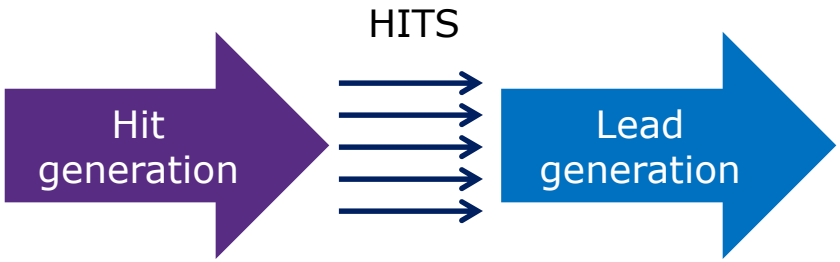


Drug discovery process and methodologies



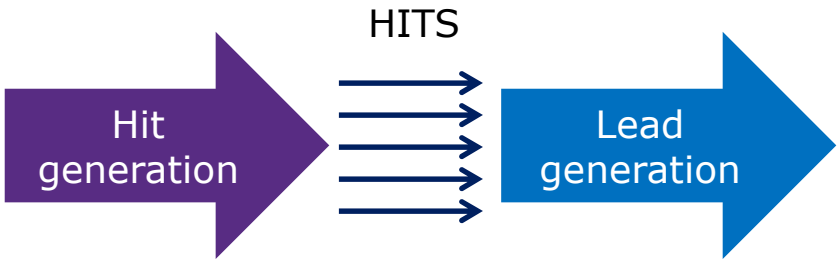
- **High throughput screening (HTS)**
  - Entire compound library directly against drug target (or may use cell-based assay)
  - Activity is dependent upon the target
  - Require secondary assays to confirm the site of action
  - Uses complex laboratory automation
  - Assumes no prior knowledge of nature of the chemotype likely to have target activity
- **Focused or knowledge-based screening**
  - Selecting smaller subsets of molecules from chemical library
  - Based on knowledge of target protein and literature (or patent precedents)
- **Fragment screening**
  - Generation of very small molecular weight compound libraries
  - Typically accompanied by generation of protein structures to enable progression
- **Physiological screening**
  - Tissue-based approach
  - Look for response more aligned with final desired *in vivo* effect

Drug discovery process and methodologies



- **Chemistry programs are conducted in parallel to screening programs**
  - Improve the potency, selectivity and physiochemical properties of the molecule
- **Further data generation to support hypothesis that intervention at the drug target will have efficacy in the disease state**
- **Output from the screen is the HIT molecule**
- **The HITS then need to be filtered/triaged:**
  - Remove frequently occurring molecules
  - Grouped according to structural similarity: broad spectrum of classes

Drug discovery process and methodologies



- **Generate dose–response curves in the primary assay for each HIT**
  - Looking for reversibility
- **Assess surviving HITS in a secondary assay (if available)**
  - Not necessarily a HT assay
  - Effect of the compounds in a functional response
  - e.g. second messenger assay or in a tissue-or cell-based bioassay
  - Provide assurance that compounds are able to modulate more intact systems
- **Chemistry analysis**
  - Cluster compounds into groups (could form the basis of lead series)
  - Identify structure/activity relationships
  - Synthesis aspects assessed