

# **“Target Validation”**

An overview for sceptics

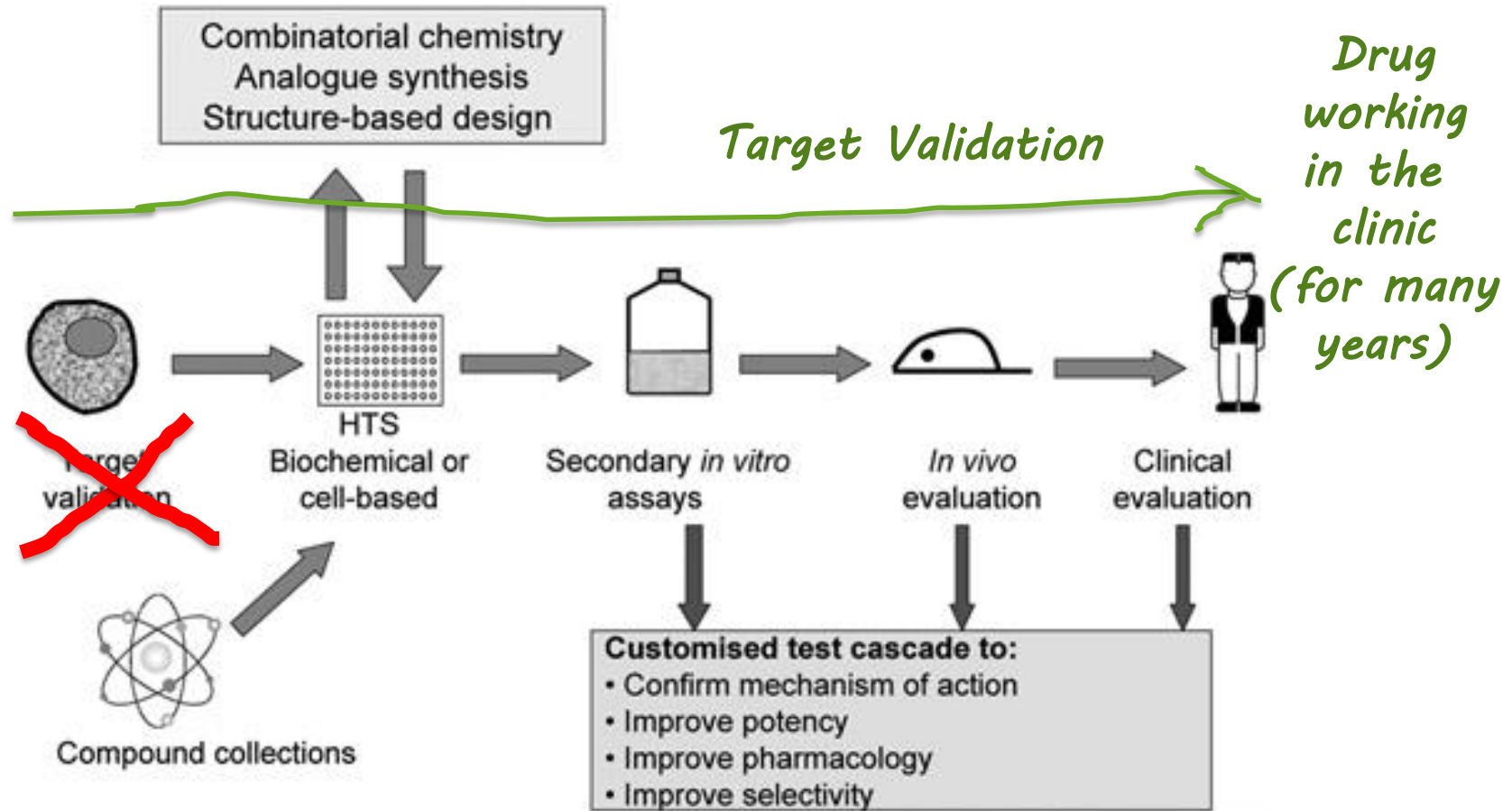
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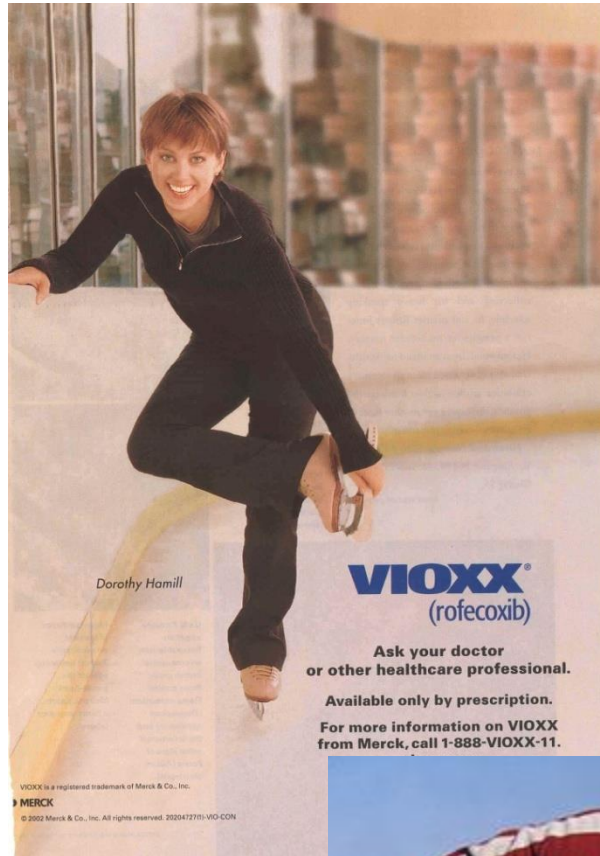
**Dr Michael R. Barnes**

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Queen Mary University of London

[m.r.barnes@qmul.ac.uk](mailto:m.r.barnes@qmul.ac.uk)

# What is Target Validation?





Dorothy Hamill

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**Cox-2 : “A validated target”**

**Should safety  
be part of the  
target validation  
process?**



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# Target Validation: The first rule

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*"All things are poison, and nothing is without poison;  
only the dose permits something not to be poisonous"*



Paracelsus, 1493-1541

# What makes an “ideal” drug target

## Disease Validation

- Target has proven disease-modifying function
- Modulation of the target is less important under physiological conditions

## Tractability

- If the druggability is not preceded, a 3D-structure for the target protein or a close homolog should be available for a druggability assessment.
- Target has a favorable ‘assayability’ enabling high throughput screening

## Safety & Efficacy

- Target expression is not uniformly distributed throughout the body
- A target/disease-specific biomarker exists to monitor therapeutic efficacy
- Favorable prediction of potential side effects according to phenotype data (e.g. in k.o. mice or genetic mutation databases).

# What makes a good target? : The Bayer view

Disease with high unmet medical need

Identification of a molecular drug target

## Target assessment

### Molecular target assessment (experimental)

Characterization of the molecular mechanisms addressed by the target  
(*ex vivo*, *in vitro*; e.g. siRNA, overexpr. of cells)

Modification of disease by target modulation in a relevant *in vivo* model?  
(e.g. using k.o./transgenic mice)

### Drugability assessment (theoretical)

SMOL binding domain existing?  
Extracellular domain (for BIOL) existing?  
Crystal structure available?  
High-throughput assay feasible?

### Ideas on target-related/stratification biomarkers

Ensure early proof-of-concept

## Adverse events evaluation

Tissue  
selectivity of  
expression

Phenotype data

Clinical data  
(if existing)

Drug class  
related adverse  
events

## IP/ competitors

Freedom to  
operate  
(FTO)  
analysis

Options for  
commercial-  
ization

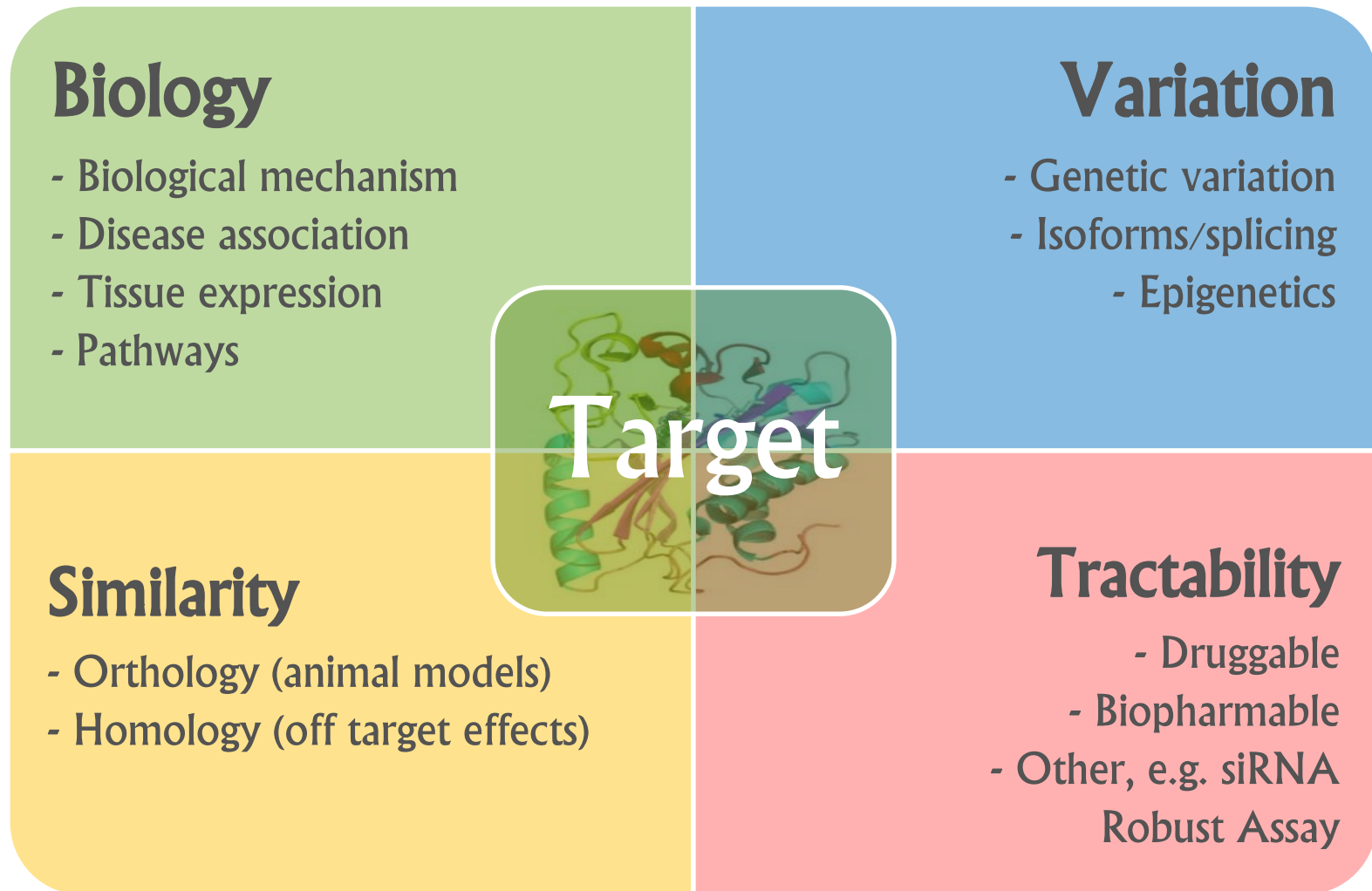
Common  
mechanism  
potential of  
target

Options for  
generation  
of IP



Gashaw, et al 2012  
Drug Discov Today.  
Feb;17 Suppl:S24-30.

# Perspectives on target validation





# Perspectives on target validation

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## Biology

- Biological mechanism
- Disease association
- Tissue expression
- Pathways



Target



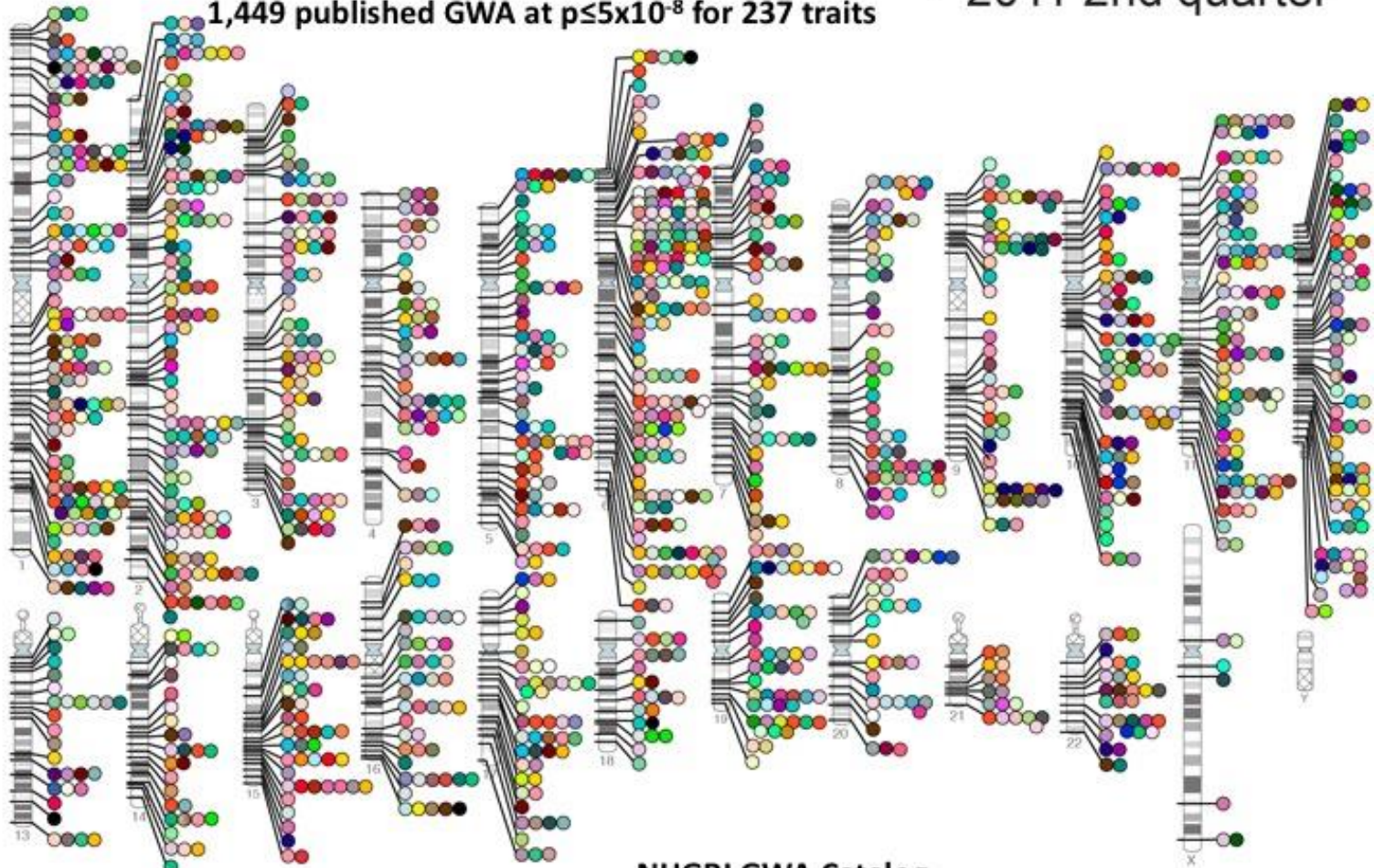
# What is a Genome Wide Association Study (GWAS)?

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- ✖ A genome-wide association study is an approach that involves rapidly scanning genetic variants (markers) **across genome** ( $\approx 0.5\text{M}$  or  $1\text{M}$ ) of **many people** ( $>2\text{K}$ ) to find genetic variations associated with a particular disease or trait.
- ✖ A large number of subjects are needed because
  - (1) associations between causal variants in common diseases are expected to show **low odds ratios**, typically below 1.5
  - (2) In order to obtain a reliable signal, given the very large number of tests that are required, associations must show a high level of significance to survive the multiple testing correction
- ✖ Such studies are particularly useful in finding genetic variations that contribute to common, complex diseases, such as asthma, cancer, diabetes, heart disease and mental illnesses
- ✖ GWAS studies are a source of target validation in humans

# GWAS: Human disease validation

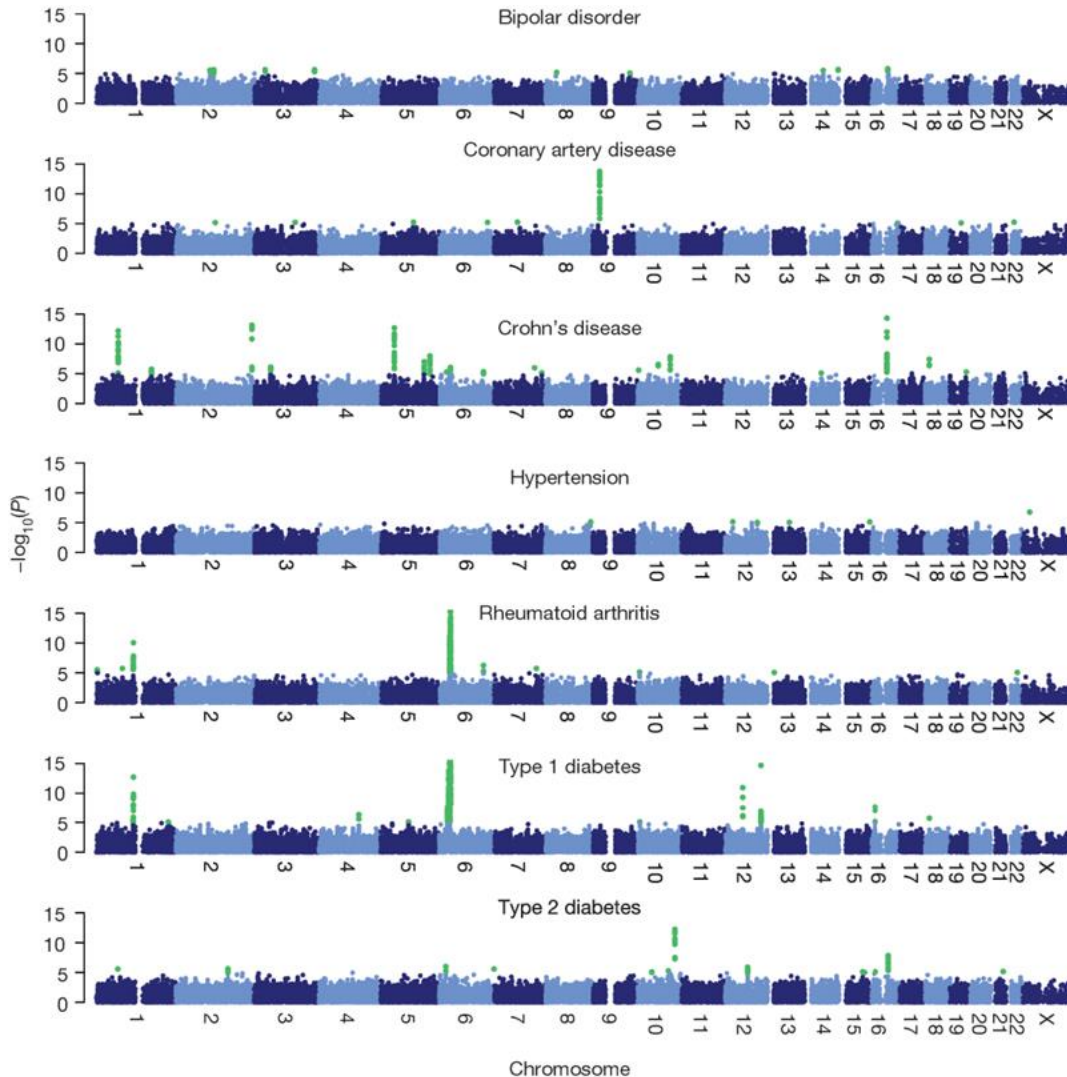
Published Genome-Wide Associations through 06/2011, 2011 2nd quarter  
1,449 published GWA at  $p \leq 5 \times 10^{-8}$  for 237 traits



NHGRI GWA Catalog  
[www.genome.gov/GWASudies](http://www.genome.gov/GWASudies)

Data from NHGRI GWAS catalog ([www.genome.gov/gwastudies/](http://www.genome.gov/gwastudies/))

# Genetics: Mining Genome Wide Association Studies (GWAS)



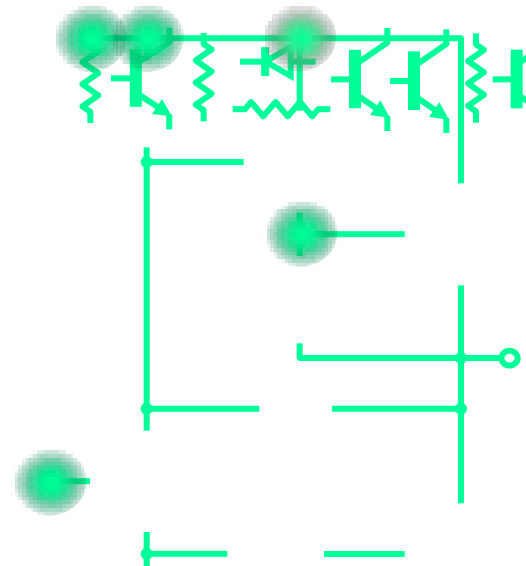
WTCCC (2007) Nature 447(7145):661-78

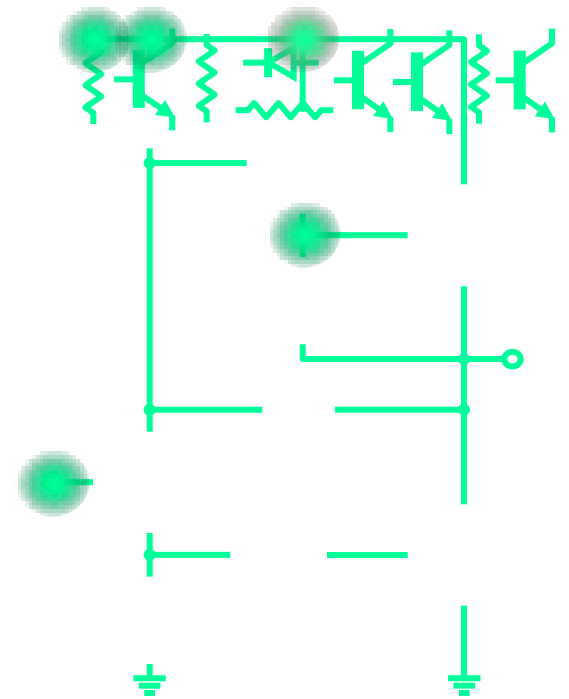
Wellcome trust case/control consortium

GWAS for seven common diseases

- + Genome-wide corrected P values  $< 10^{-5}$  in green
- + Crohn's disease (CD) shows multiple genome wide significant associations
- + Several CD genes were involved in the process of autophagy, suggesting deregulation of this pathway?

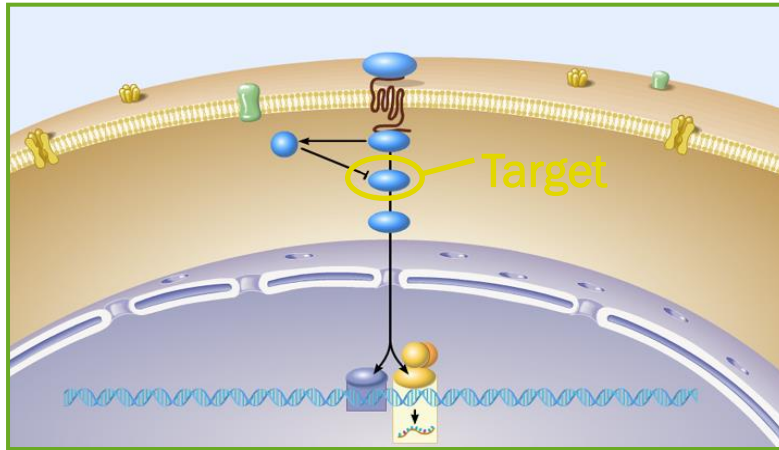
# Target validation in a pathway context

- ✖ An appealing and widespread metaphor for **biological systems** is that of **electronic circuits**
    - + The genome, as '**parts list**,' comprises the components
      - ✖ Do we *really* know all the parts?
    - + Next, we try to understand the connectivity or '**wiring diagram**' of a given system
    - + Finally, we want to be able to specify and predict the system's **dynamic behavior**
- 





# Pathway Informatics & Target Discovery

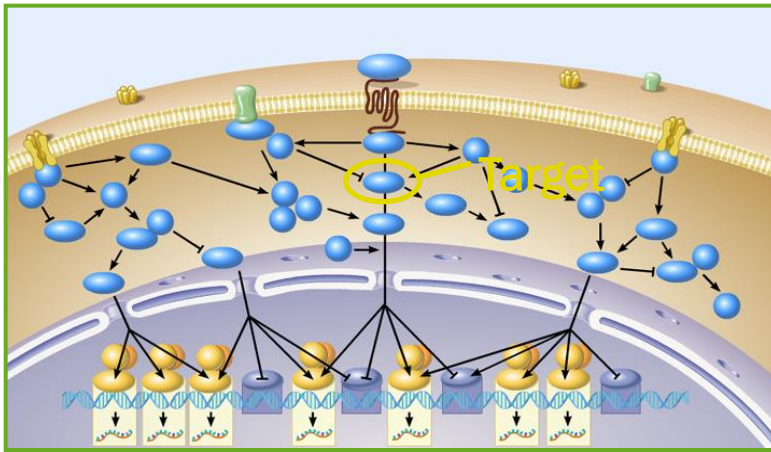


Target discovery focused on identification of a pathway and macromolecular target involved in disease. The assumption is:

- One target  $\longrightarrow$  One consequence

In reality:

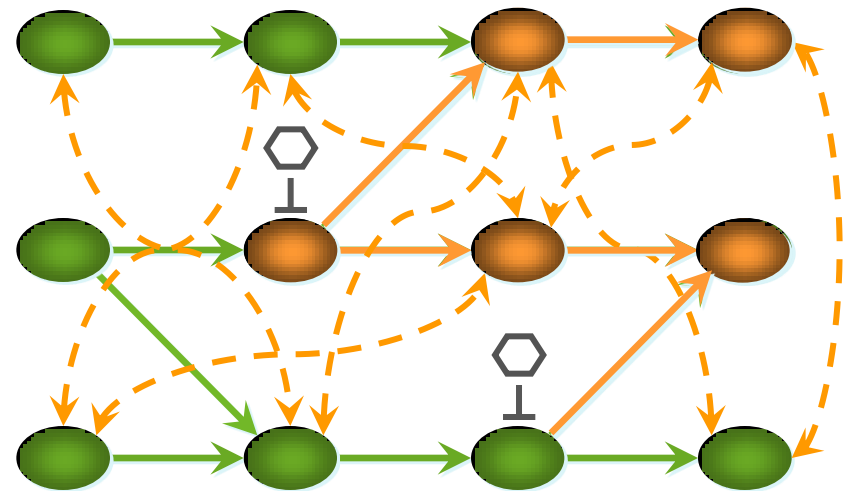
The target is usually one component of a complicated biochemical network



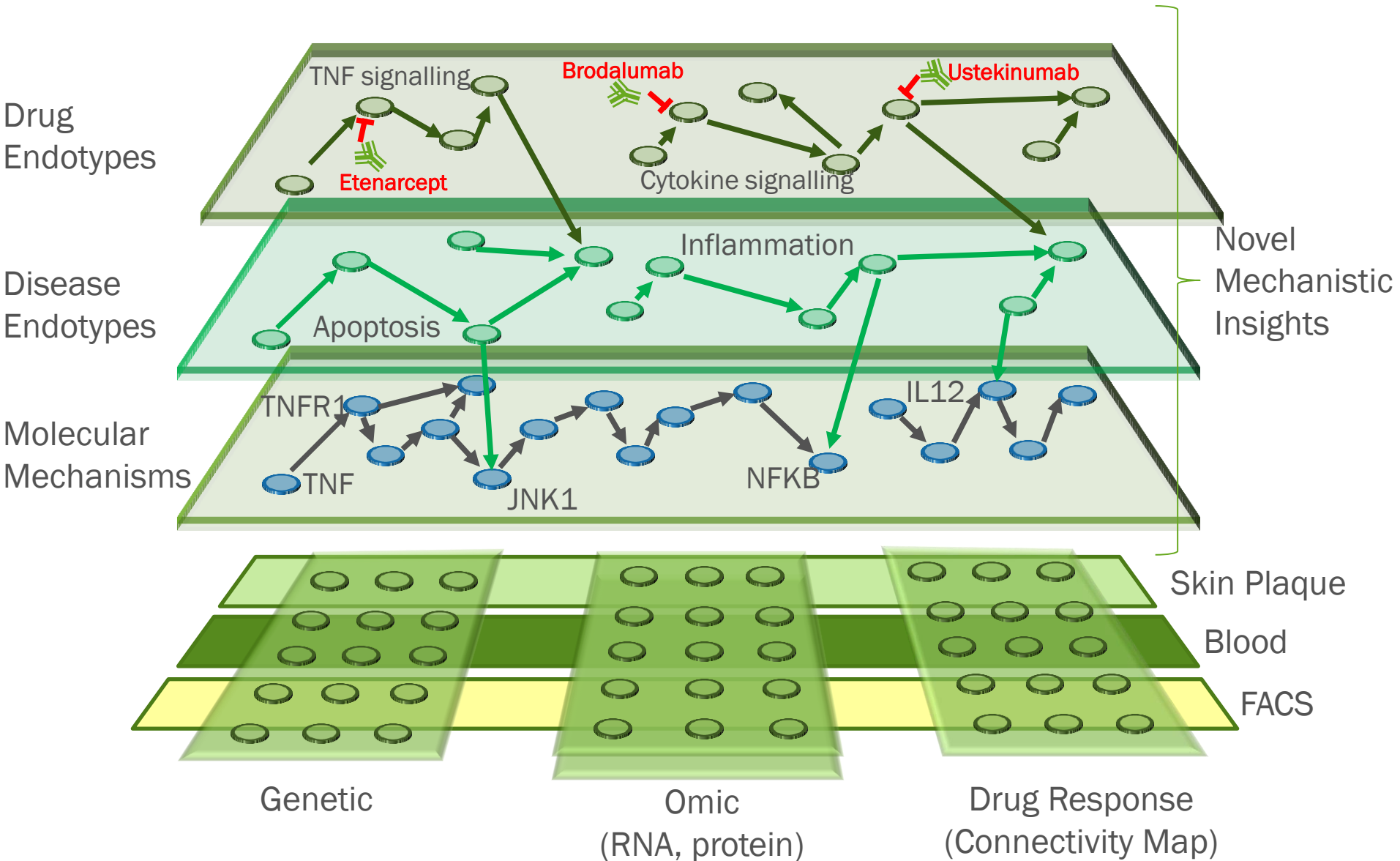
- A target acting as a single critical node may control or influence many processes.
- Network interactions can be redundant. “Work arounds” limit efficacy of a drug.
- Drugs can interact with multiple targets.
- Efficacy and safety are often a consequence of interaction with multiple targets

# Pathway Pleiotropy & Redundancy

- ✗ In drug discovery, **pleiotropy** and **redundancy** are crucial considerations
  - + Intervention is easier in 'simple' pathways
  - + Crosstalk between pathways introduces complications to aspects of drug action
    - ✗ **Safety**: unexpected target-related effects
    - ✗ **Efficacy**: may call for combination therapy or polypharmacology
  - + More holistic effects may be a challenge




# Genomics: A Systems Approach





# STITCH: Pathway Analysis (<http://stitch.embl.de/>)

 **STITCH**

[Input Page](#) | [Downloads](#) | [Help/Info](#)

search by name

chemical structure

protein sequence

multiple names

multiple sequences

list of names: (examples: #1 #2 #3)

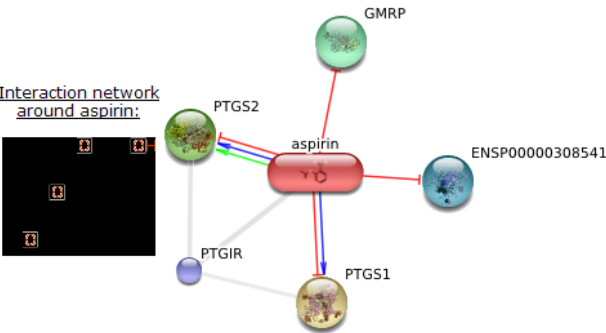
... or upload a file:

organism: auto-detect

please enter your protein or chemical of interest...

### STITCH: Chemical-Protein Interactions

Interaction network around aspirin:



```
graph TD; aspirin((aspirin)) --- PTGS2((PTGS2)); aspirin --- PTGIR((PTGIR)); aspirin --- PTGS1((PTGS1)); aspirin --- GMRP((GMRP)); aspirin --- ENSP00000308541((ENSP00000308541));
```

#### What is STITCH?

STITCH is a resource to explore known and predicted interactions of chemicals and proteins. Chemicals are linked to other chemicals and proteins by evidence derived from experiments, databases and the literature.

STITCH contains interactions for over 68,000 chemicals and over 1.5 million proteins in 373 species.

#### Mini-Tutorial

When searching for multiple names, you can enter a mixture of protein and chemical names. For example, you can [paste](#)

```
tyrosine
phenylalanine
phenylalanine hydroxylase
```

If you click "GO!", STITCH will ask you to select a species from those that have phenylalanine hydroxylase. Next, you will be presented with a long list of matching names. Make sure that the items you want to see are checked, and click continue. You will be taken to the evidence view, but you should switch to the actions view to see that phenylalanine hydroxylase catalyzes the reaction from phenylalanine to tyrosine.

#### Status



STITCH (Search Tool for Interactions of Chemicals) is a sister project of the protein-protein interactions server [STRING](#). The database of chemicals is based on [PubChem](#). Up-to-date genomes and proteomes are maintained at [UniProtKB/Swiss-Prot](#) and [Ensembl](#).

To be informed about the latest developments, please subscribe to the [STRING/STITCH blog](#).

STITCH references: [Kuhn et al. 2008](#)

Miscellaneous: [Access Statistics](#), [Robot Access Guide](#), [Medusa Network Viewer](#), [Supported Browsers](#).

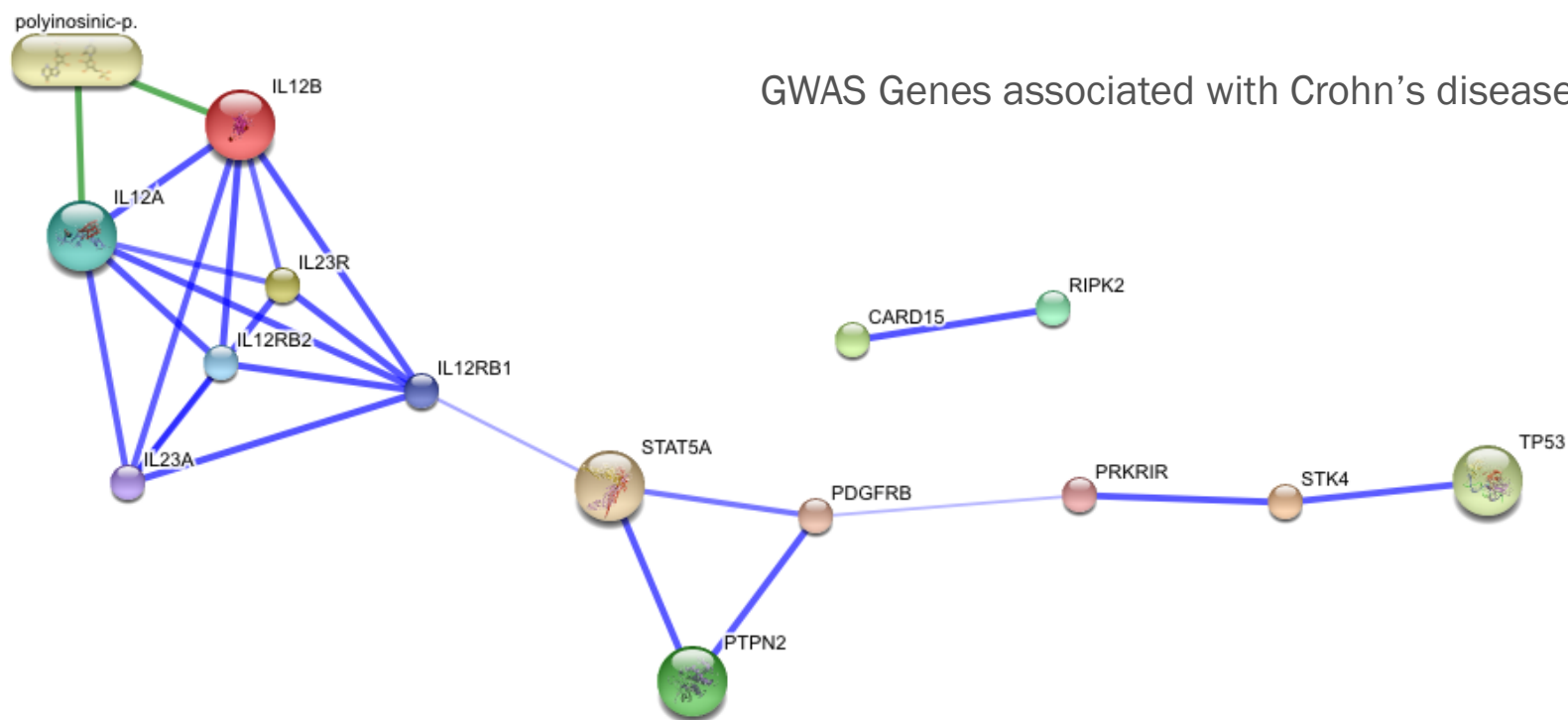
**What's New?** This is the first public version of STITCH. You can also access the [beta version of STITCH 2](#).

## SIMPLE INTERFACE

- Add multiple genes
- Identify interactions
- Identify known drugs
- Expand network

# STITCH: Pathway Analysis (<http://stitch.embl.de/>)



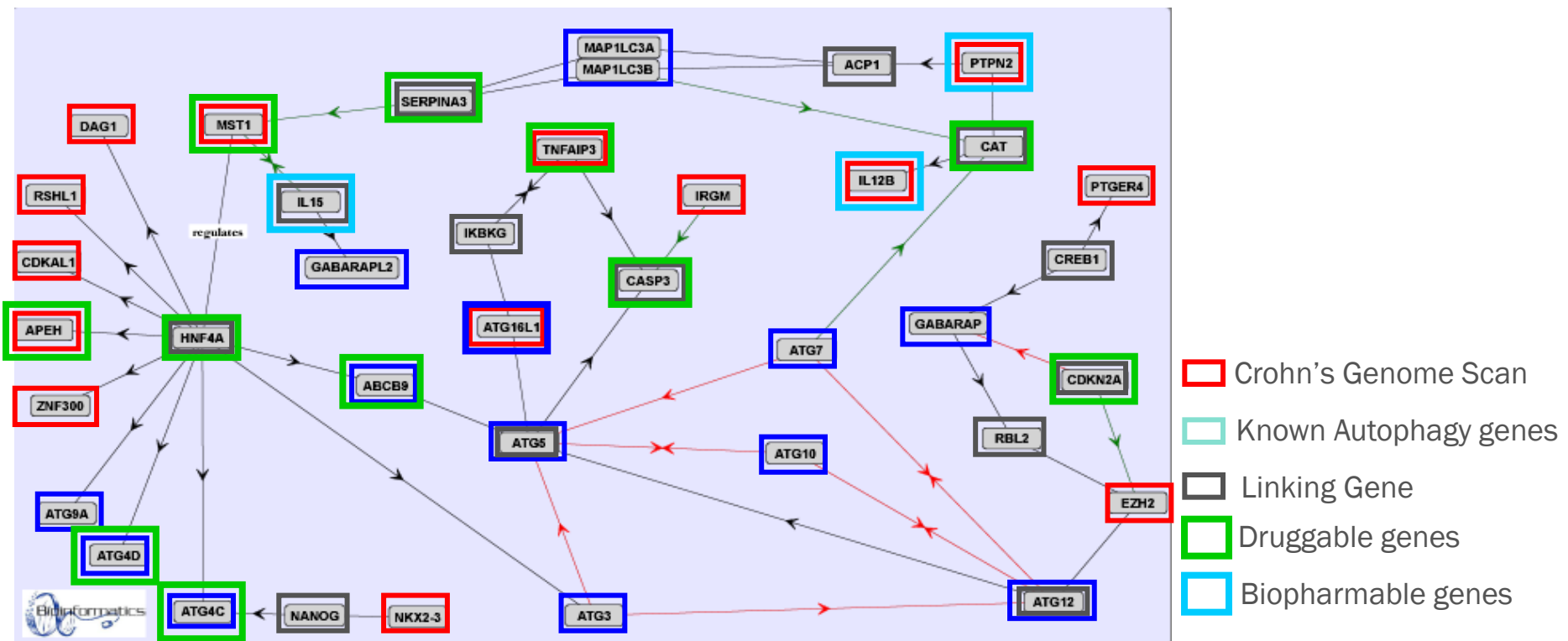
This is the **confidence view**. Stronger associations are represented by thicker lines.  
Protein-protein interactions are shown in blue, chemical-protein interactions in green and interactions between chemicals in red.

Confidence view Evidence view Actions view Interactive view

CC Chemical links + Add nodes - Remove nodes Save

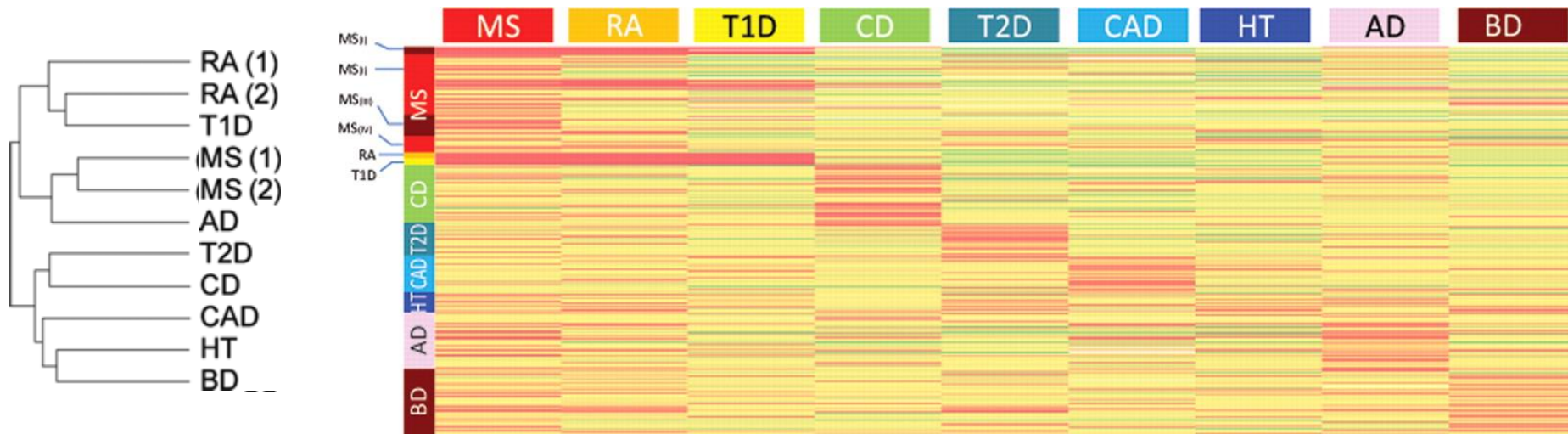
# New Drug Mechanisms from Pathways: Crohn's disease GWAS

- ✗ Wellcome trust genome scan identified and replicated 23 genes in CD
  - + 14/23 associated genes were linked to the autophagy mechanism
  - + Reduced autophagy identified as a key mechanism in Crohn's disease
    - ✗ Rapamycin is a known drug which downregulates autophagy
    - ✗ Rapamycin is now in phase II trial for Crohn's disease

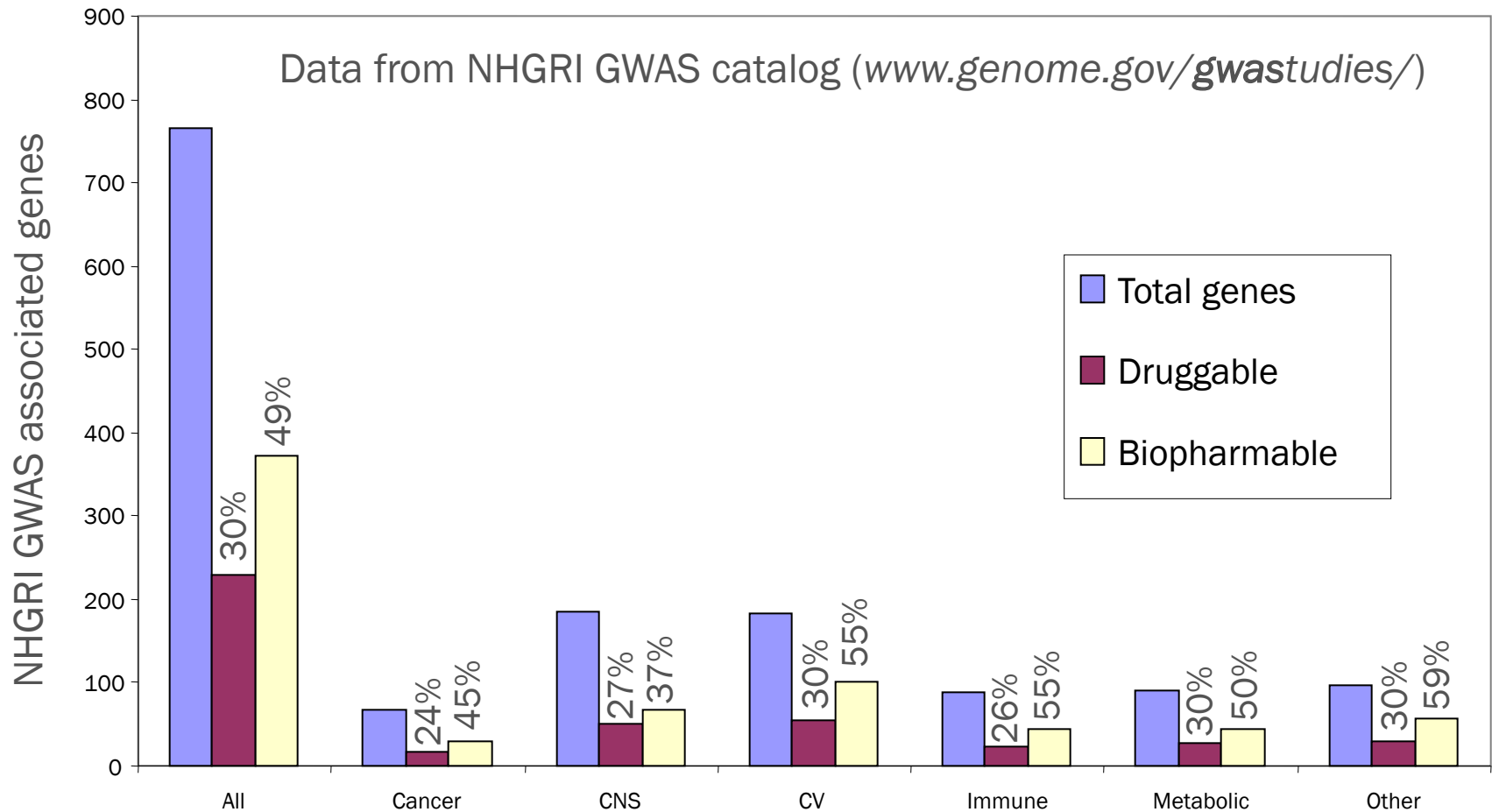


# COMMON DISEASES SHOW COMMON PATHOLOGIES

- ✗ Pathway analysis applied to the seven WTCCC GWAS studies and a GSK Alzheimer's GWAS
- ✗ Common modules emerged and clustered across all the disease GWAS, hinting at linked pathology
  - + Suggests repositioning opportunities?



# Therapeutic opportunities from GWAS



# GWAS: Confirming drug indications

**Table 1 Selected examples of matches between GWAS trait and drug indication<sup>a</sup>**

| Drug name or class    | Most advanced development phase (for the indication) | Gene    | Drug indication                    | GWAS trait              | GWAS reference |
|-----------------------|------------------------------------------------------|---------|------------------------------------|-------------------------|----------------|
| Statins               | Launched                                             | HMGCR   | Hypercholesterolemia               | LDL Cholesterol         | 1              |
| Ustekinumab           | Approved                                             | IL12B   | Psoriasis                          | Psoriasis               | 13             |
| Ustekinumab           | Phase 2                                              | IL12B   | Crohn's disease                    | Crohn's disease         | 2              |
| Anti-IL2 receptor mAb | Phase 2                                              | IL2RA   | Ulcerative colitis                 | Crohn's disease         | 2              |
| AMG -785/<br>CDP-7851 | Phase 2                                              | SOST    | Bone regeneration/<br>osteoporosis | Bone mineral<br>density | 14             |
| Znt8 agonists         | Preclinical                                          | SLC30A8 | Type 2 diabetes                    | Type 2 diabetes         | 15             |

<sup>a</sup>Examples are ranked from most advanced drug (launched) to less advanced (preclinical). The associated gene between each GWAS and the drug is shown. The drug indication and the phase of development for each drug are derived from the Pharmaprojects database. In each example the GWAS trait is identical (rows 1, 2, 3 and 6) or closely related (rows 4 and 5) to the drug indication. For the full list, see **Supplementary Table 3**. In many cases, more drugs for the gene are listed in the database at different phases. The GWAS references are from the catalog of GWAS data (<http://www.genome.gov/gwasstudies>).

# Repositioning opportunities from GWAS

**Table 2** Selected examples of mismatch between GWAS trait and drug indication<sup>a</sup>

| Drug name         | Most advanced development phase (for the indication) | Gene    | Current drug indication                          | GWAS trait (new potential drug indication)        | GWAS references |
|-------------------|------------------------------------------------------|---------|--------------------------------------------------|---------------------------------------------------|-----------------|
| Denosumab/AMG-162 | Launched/registered                                  | TNFSF11 | Osteoporosis/bone cancer                         | Crohn's disease                                   | 2               |
| RPI-78M           | Phase 3                                              | IL27    | Adrenoleukodystrophy                             | Crohn's disease/inflammatory bowel disease        | 2,16            |
| Nepicastat        | Phase 2                                              | DBH     | Cocaine addiction/post-traumatic stress disorder | Smoking cessation                                 | 7               |
| Biib-033          | Phase 1                                              | LINGO-1 | Multiple sclerosis                               | Essential tremor                                  | 4,5             |
| AMG-557           | Phase 1                                              | ICOSLG  | Systemic lupus erythematosus                     | Crohn's disease/celiac disease/ulcerative colitis | 17–19           |
| Cwp-231           | Preclinical                                          | TCF4    | Cancer                                           | Fuchs's corneal dystrophy                         | 20              |

<sup>a</sup>Examples are ranked from most advanced drug (launched) to less advanced (preclinical). The associated gene between each GWAS and the drug is shown. The drug indication and the phase of development for each drug are derived from the Pharmaprojects database. For the full list, see **Supplementary Table 4**. In many cases, more drugs for the gene are listed in the database at different phases. The GWAS references are from the catalog of GWAS data (<http://www.genome.gov/gwasstudies>).



# Target biology informs drug discovery



- Target pathway
- Near targets/pathways
- Target distribution
- Functional genomics
- Tool compounds

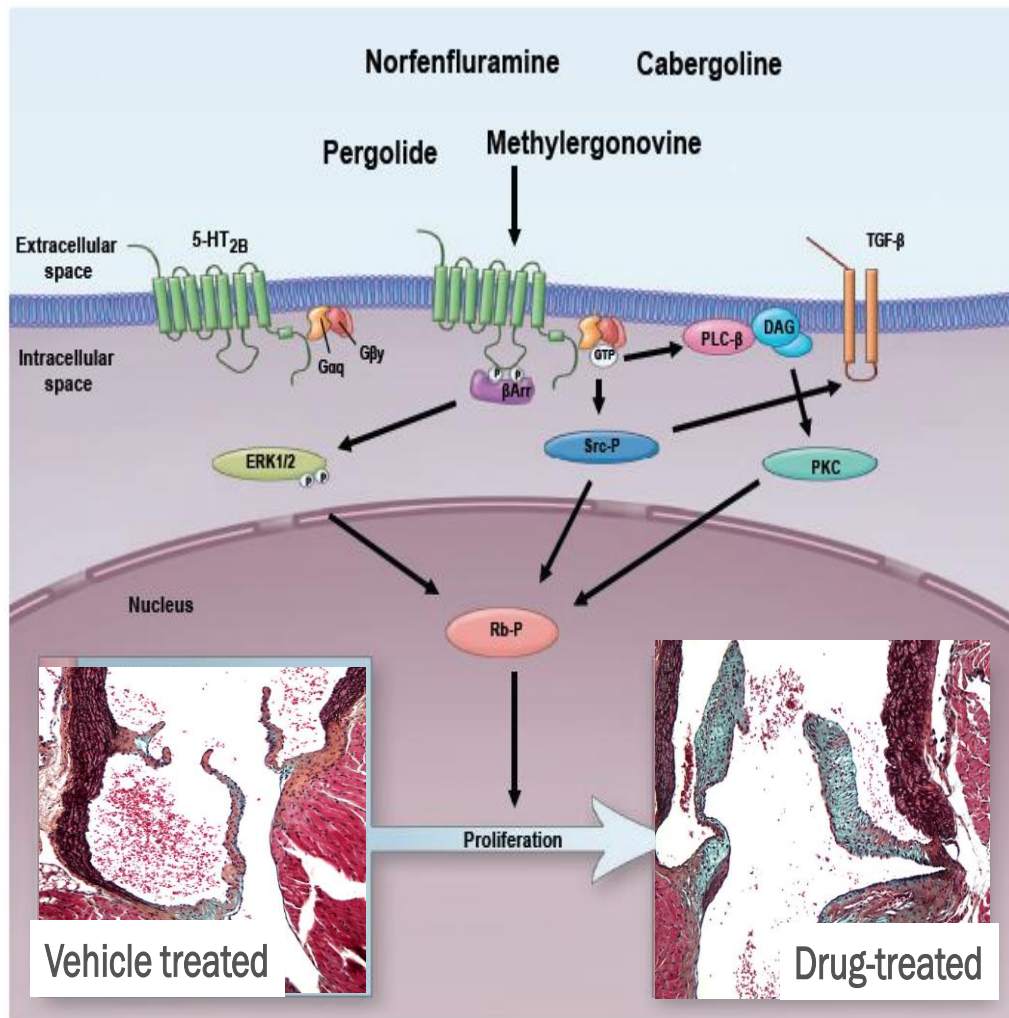
Candidate Selection  
therapeutic index ?  
- (Toxic dose/Effective dose)

Drug Development

- Species
  - sensitive?
  - relevant?
- Endpoints?
- Timing?

Risk Assessment  
Extrapolation from  
animal model to man  
(relevance, sensitivity)

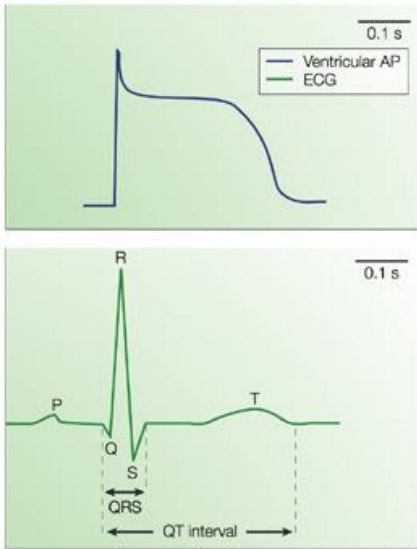
# Common liabilities: On Target



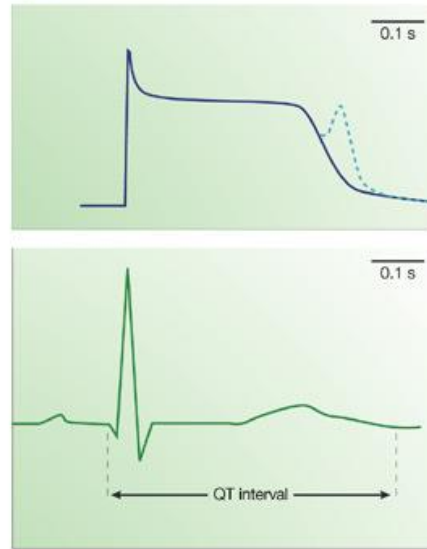
- ✗ e.g. Neuropsychiatric HTR2B agonists cause heart valve pathology
- ✗ Proliferation of cardiac fibroblasts on the tricuspid valve, known as cardiac fibrosis
- ✗ Emphasises ideal of restricted expression of target to disease tissue

# Common liabilities: Off Target

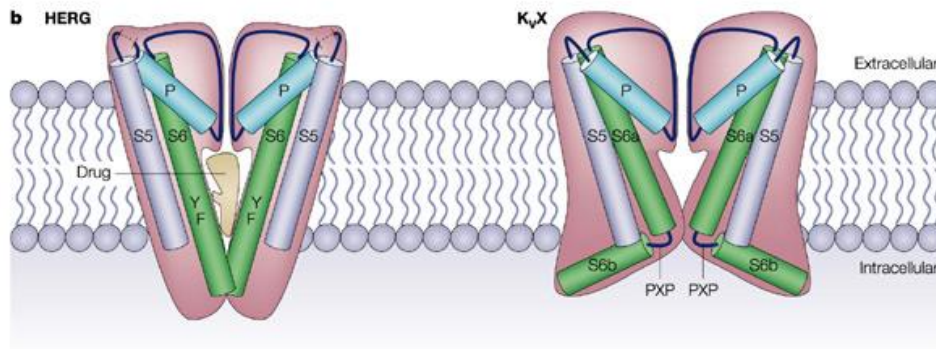
**a Normal**



**LQTS**



**b HERG**

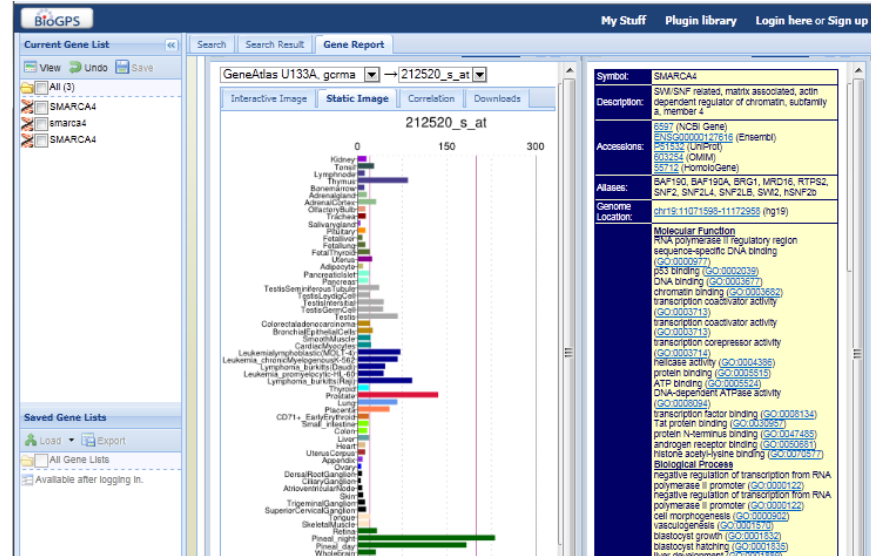


- ✗ Many drugs, eg. Antihistamines induce Long QT syndrome by HERG action
- ✗ HERG channels contain two aromatic residues (Y652 and F656) in the S6 helices (green), which line the pore cavity and can therefore form cation–interactions with multiple drugs.
- ✗ HERG also has a larger cavity than other KV channels, and so can accommodate a wide size range of drugs

# Target Expression

BioGPS

✖ [www.biogps.org](http://www.biogps.org)



EBI Gene Expression Atlas

✖ Comprehensive view of public expression data

✖ View the Novartis data in BioGPS here

+ <http://www.ebi.ac.uk/gxa/experiment/E-AFMX-5>

# Target Expression : Allen Brain Atlas

✕ <http://www.brain-map.org/>

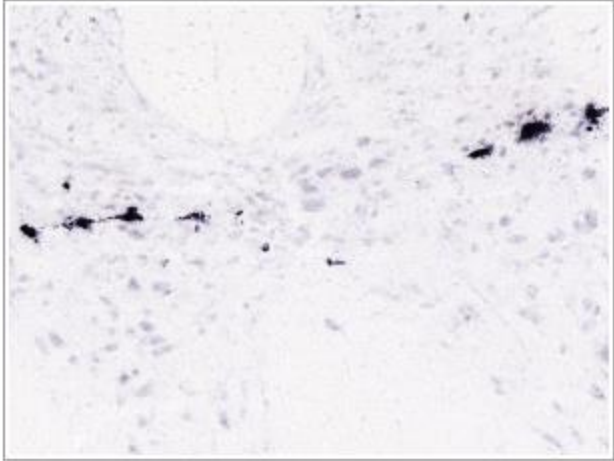
## ALLEN BRAIN ATLAS

A growing collection of online public resources integrating extensive gene expression and neuroanatomical data, complete with a novel suite of search and viewing tools.

Get started with tutorials offering introductory overviews and guided tours.

|                                                                                                                      |                                                                                                                    |
|----------------------------------------------------------------------------------------------------------------------|--------------------------------------------------------------------------------------------------------------------|
| <b>Mouse Brain</b><br>A genome-wide, high-resolution atlas of gene expression throughout the adult mouse brain       | <b>Human Brain</b><br>A multi-modal, multi-resolution atlas detailing gene expression across the adult human brain |
| <b>Developing Mouse Brain</b><br>A detailed atlas of gene expression across mouse brain development                  | <b>Developing Human Brain</b><br>A detailed atlas of gene expression across human brain development                |
| <b>Mouse Connectivity</b><br>A high-resolution map of neural connections in the mouse brain                          | <b>Non-Human Primate</b><br>A detailed atlas of gene expression across postnatal primate brain development         |
| <b>Mouse Spinal Cord</b><br>A genome-wide, high-resolution atlas of gene expression throughout the mouse spinal cord | <b>Glioblastoma</b><br>A unique platform for exploring human glioblastoma at the cellular and molecular levels     |
| <b>Mouse Diversity</b><br>Gene expression data in the mouse brain across genetic backgrounds and sex                 | <b>Sleep</b><br>Gene expression data in the mouse brain for five conditions of sleeping and waking                 |

### HIGHLIGHTS- Updated June 30, 2012



*Cart is expressed in the intermediate gray matter in the spinal cord of the postnatal day 56 mouse.*

[More highlights](#)

### ANNOUNCEMENTS- Updated June 7, 2012

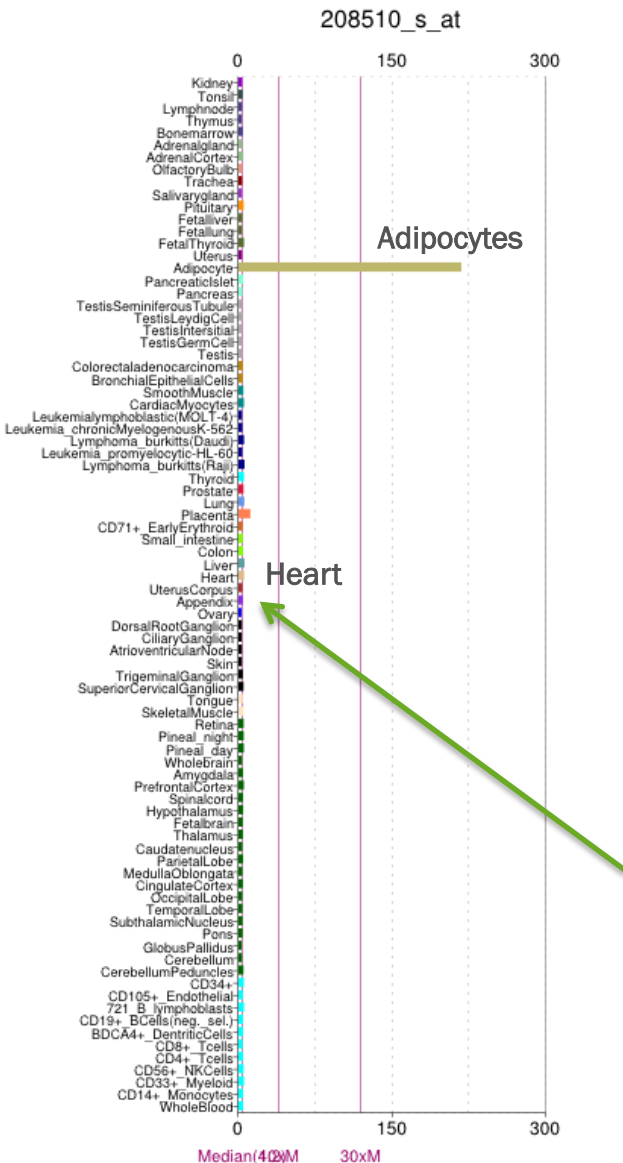
[What's New - Latest Data Release June 7, 2012](#)

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# Target Expression : PPARG agonists



- ✖ Type II Diabetes target
- ✖ Nuclear hormone receptor
- ✖ Highly expressed in adipocytes
- ✖ Rosiglitazone (Avandia) was a blockbuster for GSK....
- ✖ ....until emergence of cardiotox
- ✖ Seems unlikely on the basis of expression!
- ✖ Could it be an off-target effect?

# Target Expression : PPARG agonists

Genes 1-5 of 5 total found • [Download all results](#) • [JSON](#) [XML](#)

Legend:  - number of studies the gene is **over** expressed in

| Gene  | Ontology              |              |        |                |        |                 |
|-------|-----------------------|--------------|--------|----------------|--------|-----------------|
|       | cardiovascular system | blood vessel | artery | umbilical vein | heart  | heart component |
| PPARA | 8<br>1                | 1            |        | 1              | 6      | 5<br>1          |
| PPARD | 5<br>3                | 1            |        | 1              | 3      | 3<br>1          |
| RXRA  | 4<br>1                | 2<br>1       | 2<br>1 | 1              | 2<br>1 | 1<br>1          |
| RARG  | 4<br>1                | 1            |        | 1              | 2      | 3<br>1          |
| PPARG | 2<br>1                | 2<br>1       | 2<br>1 | 1              |        | 1               |

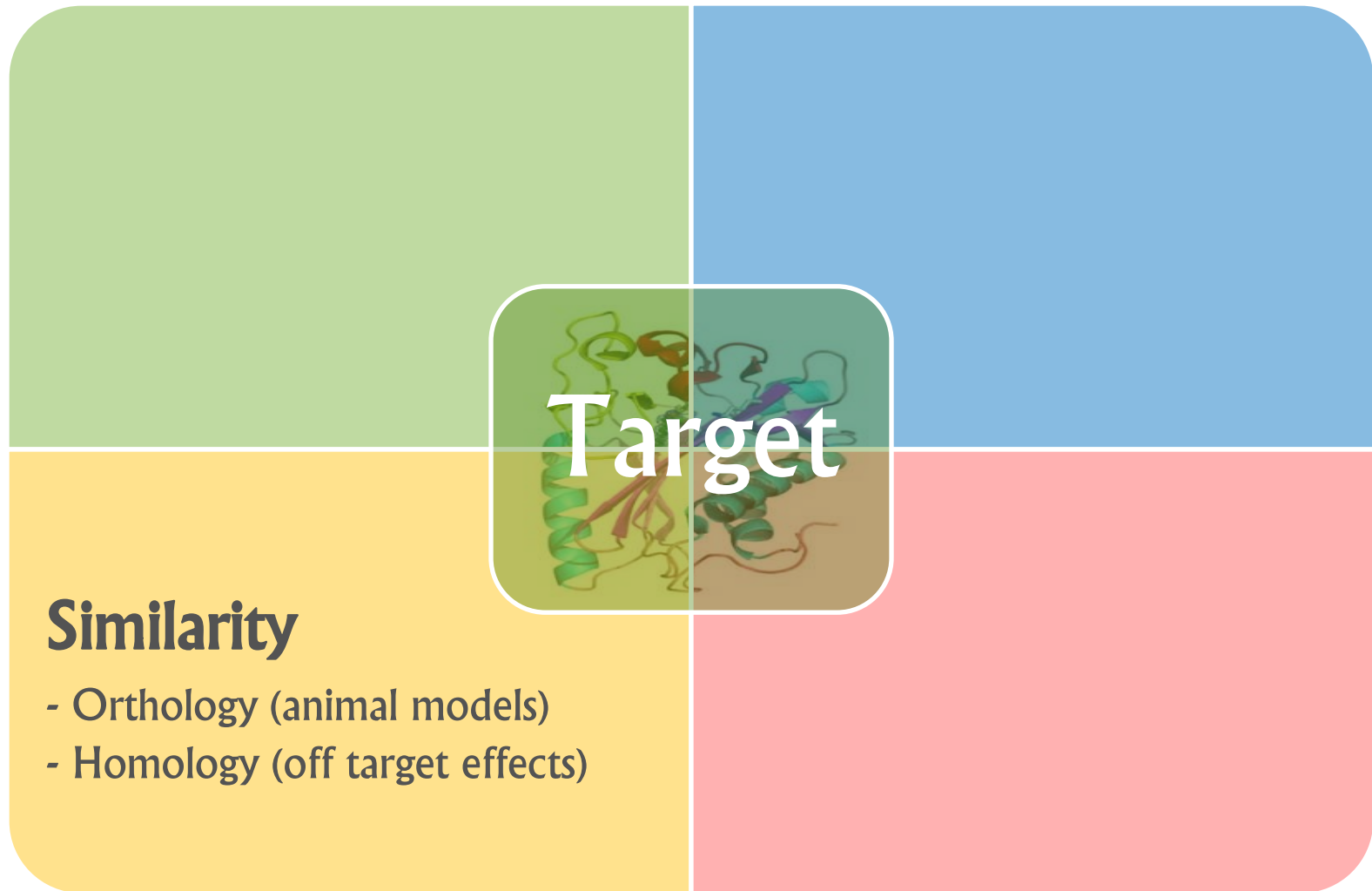
Processing time: 1.55 secs.

- ✗ ChEMBL query shows Avandia activity at PPARG homologues
- ✗ Notably all are expressed at higher levels than PPARG in heart according to EBI Gene Expression Atlas



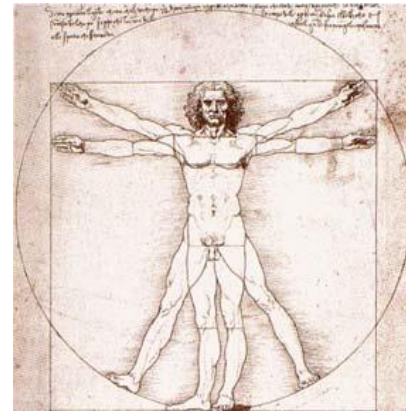
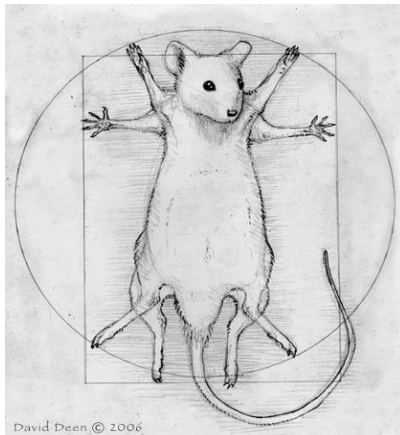
# Perspectives on target validation

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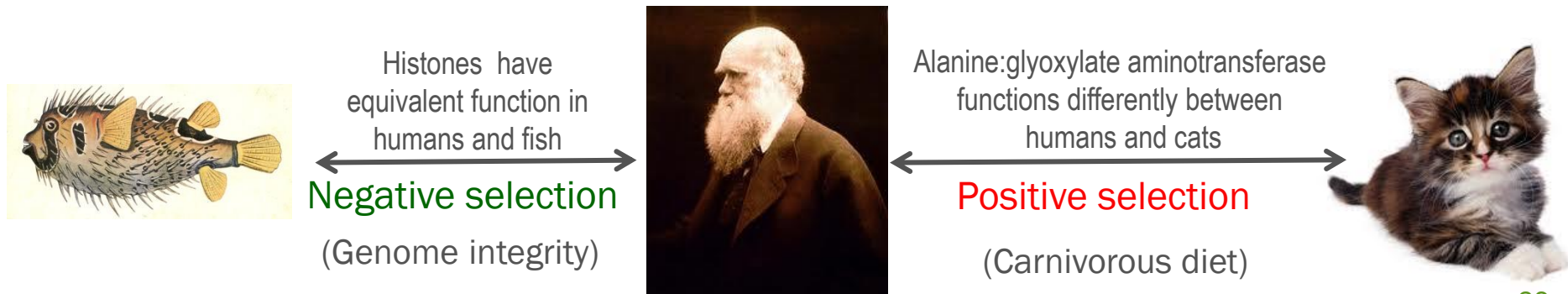
# What Makes a Good Animal Model?

- ✖ A good animal model should have the following characteristics:
  - + Should closely reproduce the disease or condition under study
  - + Should be an industry/academic accepted model
  - + The data should be robust enough to be duplicated by a 3rd party
  - + Should show statistical significance – feasible to use sufficiently large number of animals to demonstrate statistical significance
  - + The data endpoints should be convincing enough to justify transition into man



# Selecting Species for Preclinical Studies

- ✗ Preclinical studies in animal models aim to accurately predict drug action and safety in Man
  - + Preclinical studies are only valid if the function of the gene is equivalent (orthologous) between the model and man
- ✗ Three types of evolutionary selection pressure:
  - + **Negative selection:** Deleterious mutations are selected against to conserve function
  - + **Neutral selection:** Neutral mutations are unaffected by selection
  - + **Positive selection:** Advantageous mutations are selected for. Positive selection is indicative of functional change



# Molecular Evolution in Drug Discovery

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- ✖ Evolution of targets between and within species
  - + Loss of classic dose-response paradigms
- ✖ Humanisation of antibodies
  - + Intra-species variation
  - + Immunogenicity in animals may differ to human
- ✖ Evolution Increases uncertainty in the preclinical transition to man
  - + Recently underlined in the disastrous TGN1412 trial
- ✖ A good understanding of the preclinical model is imperative for a safe transition to man

# Marmoset Models in Neuroscience Drug Discovery

- *Callithrix jacchus*
- Key animal model in psychiatry
  - Tiny radio implant measures EEG/EMG/EOG traces 24/7
  - Records sleep levels.....

Response  
to threats...



.....social and  
grooming  
behaviour



## GSK Orexin Receptor Antagonists

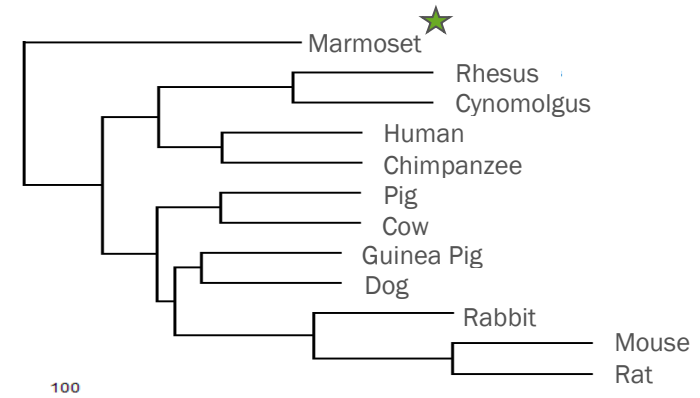
- Orexin signalling is key in satiety, sleep and mood
- Neuroscience programmes in Sleep & Mood
  - Orexin Receptor antagonist pharmacology appeared normal in Marmosets.....

# Orexin receptor evolutionary analysis predicts receptor pharmacology

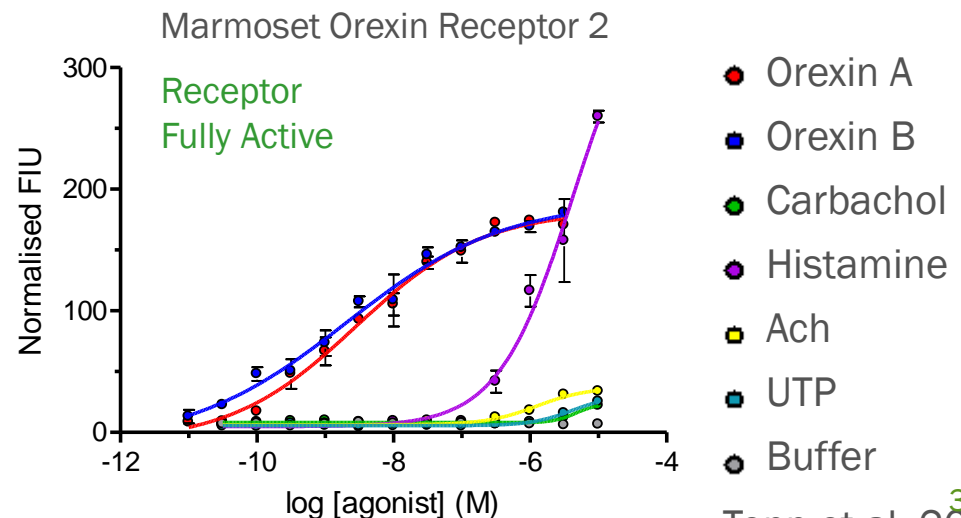
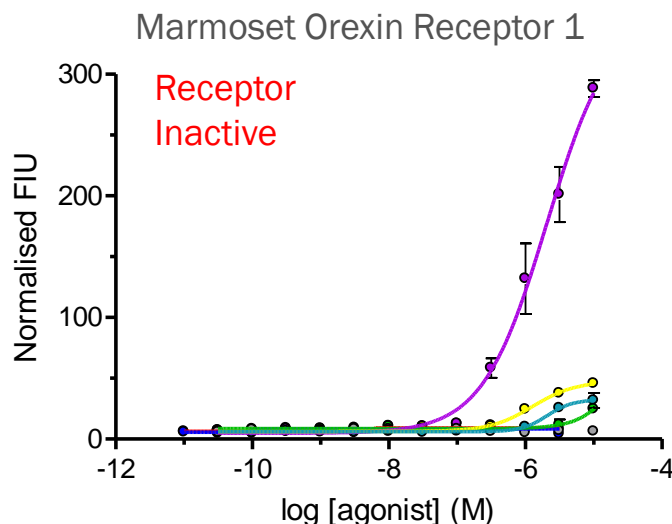
## Orexin Receptor Antagonist Development

- Marmoset Orexin R1 protein highly divergent
- Orexin R2 & ligands also showed modest sequence divergence in Marmoset
- Orexin R1 receptor pharmacology investigated
  - Marmoset Orexin R1 shown inactive *in vitro*
- Demonstrates that Orexin action in Marmoset is mediated entirely by Orexin R2

### Orexin Receptor 1 protein phylogeny

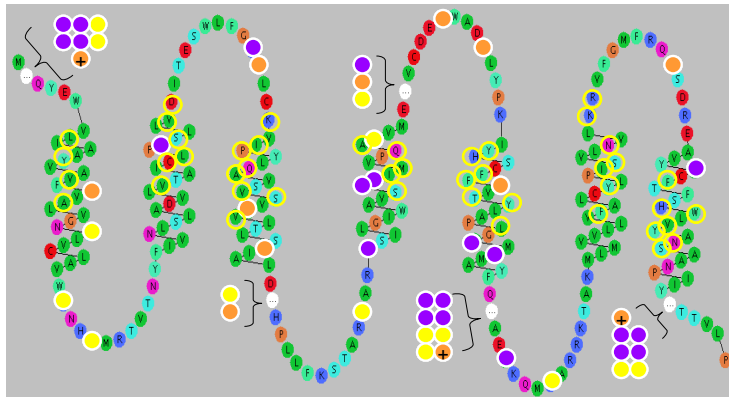


## in vitro response of Marmoset Orexin receptors to Orexin ligands A & B (FLIPR)



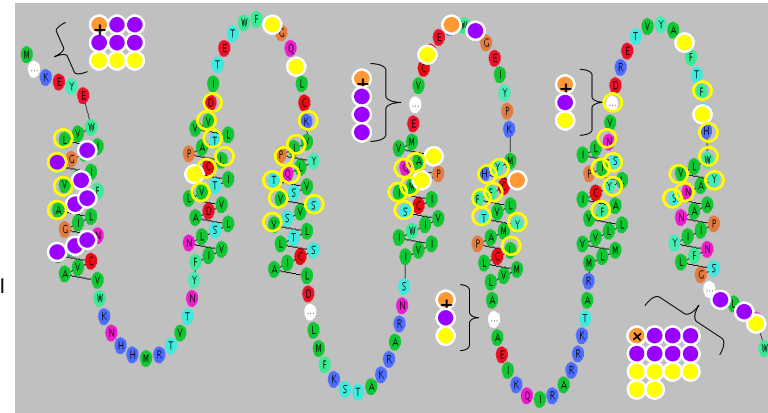
# EVOLUTION OF Orexin Signalling in Marmosets

Orexin Receptor 1



- Yellow circle: Unique to marmoset
- Purple circle: Unique to one other mammal
- Orange circle: Changes in >1 mammal
- White circle: Structural key residue

Orexin Receptor 2

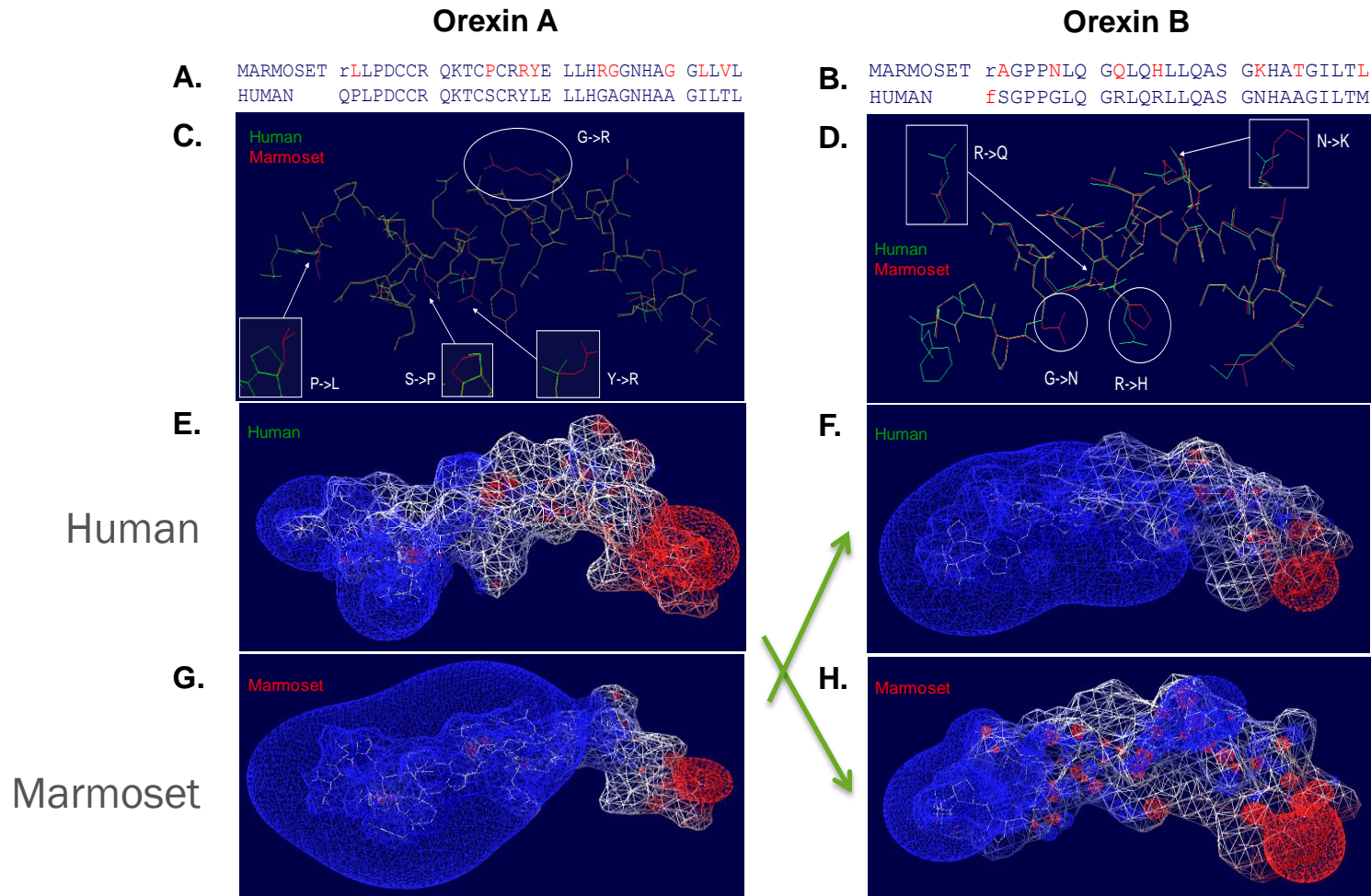


- ✗ Analysis of Orexin receptor structure showed most changes in Orexin R1 are intracellular (signalling?), changes in Orexin R2 are mostly extracellular (ligand binding?)
- ✗ Evolutionary selection analysis was performed on the Orexin receptor and ligand genes using PAML (Yang, 1997)
  - + Both receptor & ligand lineages show evidence of positive selection
  - + Positive selection at specific-site residues is restricted to the mature peptide regions of the orexin ligands A & B.
    - ✗ Suggests altered ligand function to compensate for receptor loss?



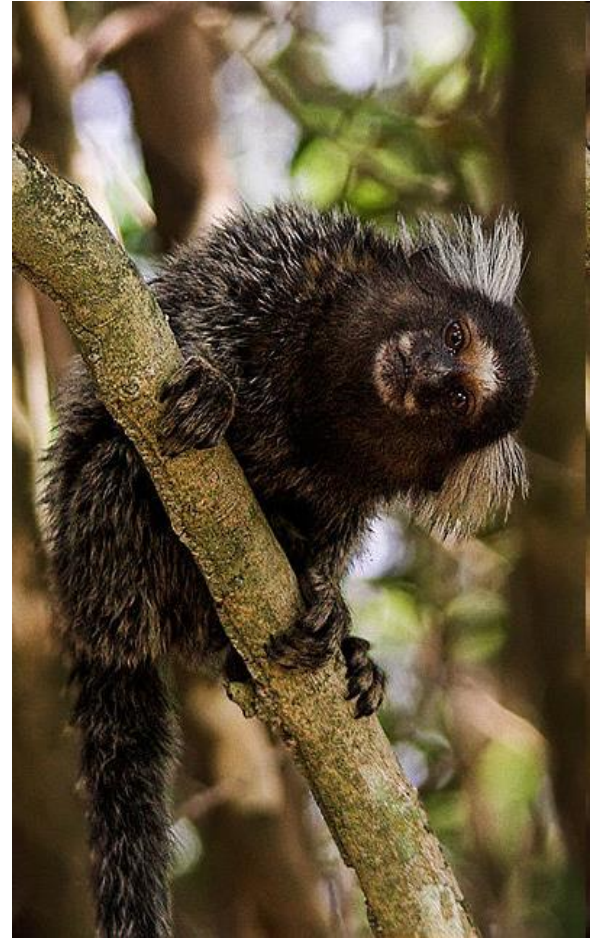
# Structural & electrostatic modelling of Marmoset and Human orexin ligands

- ✖ Orexin A most active against Orexin R1 (which is inactive in Marmoset)
- ✖ Marmoset Orexin A has evolved similar electrostatic properties to human Orexin B



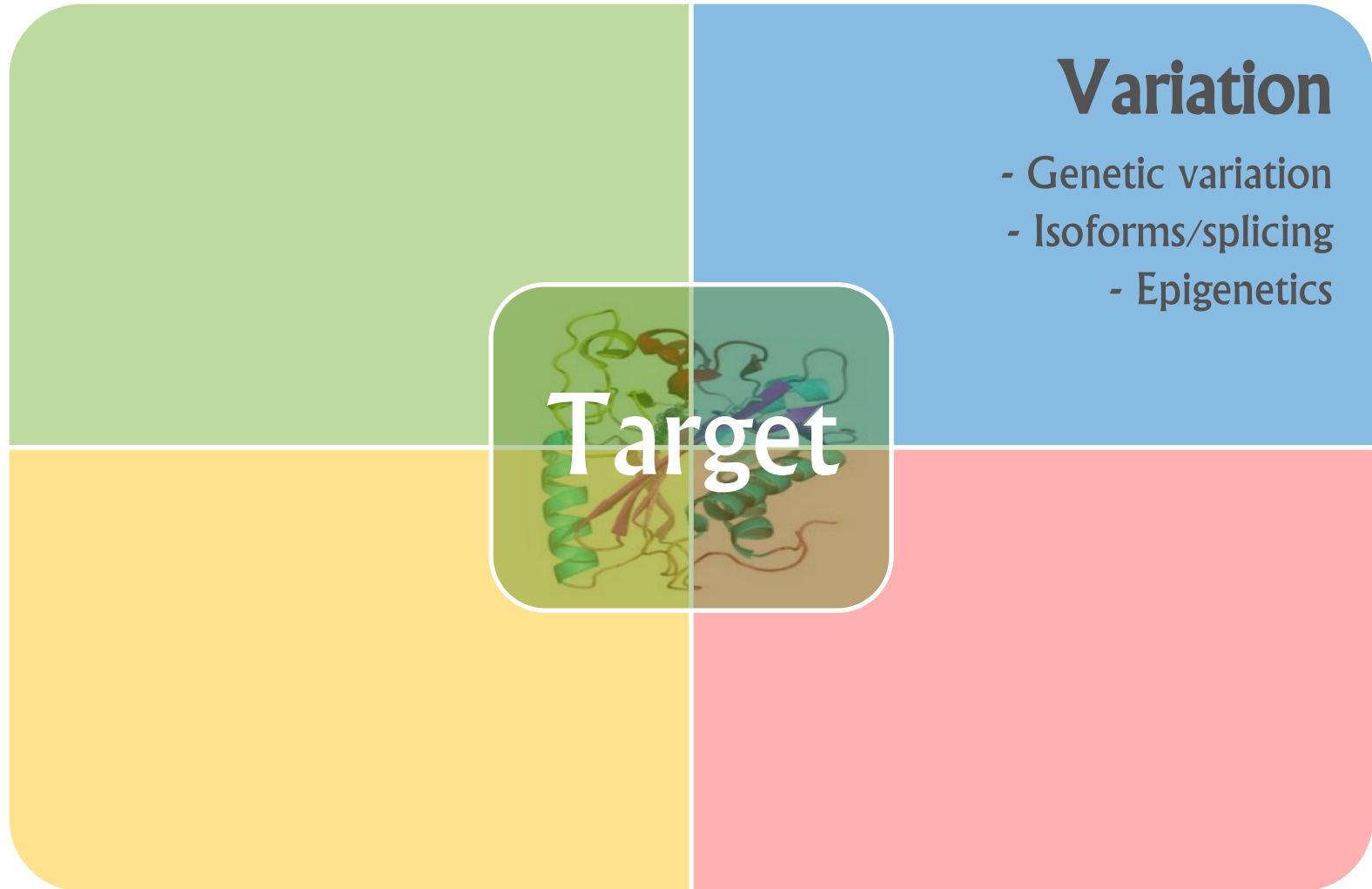
# Conclusions: Marmoset orexin signalling

- Orexin ligands and receptors have both diverged significantly in Marmosets
  - Suggests co-evolution of ligand & receptor
  - Orexin A ligand may have evolved to act on remaining active orexin receptor 2
- Orexin antagonist results in marmosets should be treated with caution
  - No divergence seen in old world primates
  - These would be better models
- Evolutionary biology and phylogenetics are key tools for drug discovery

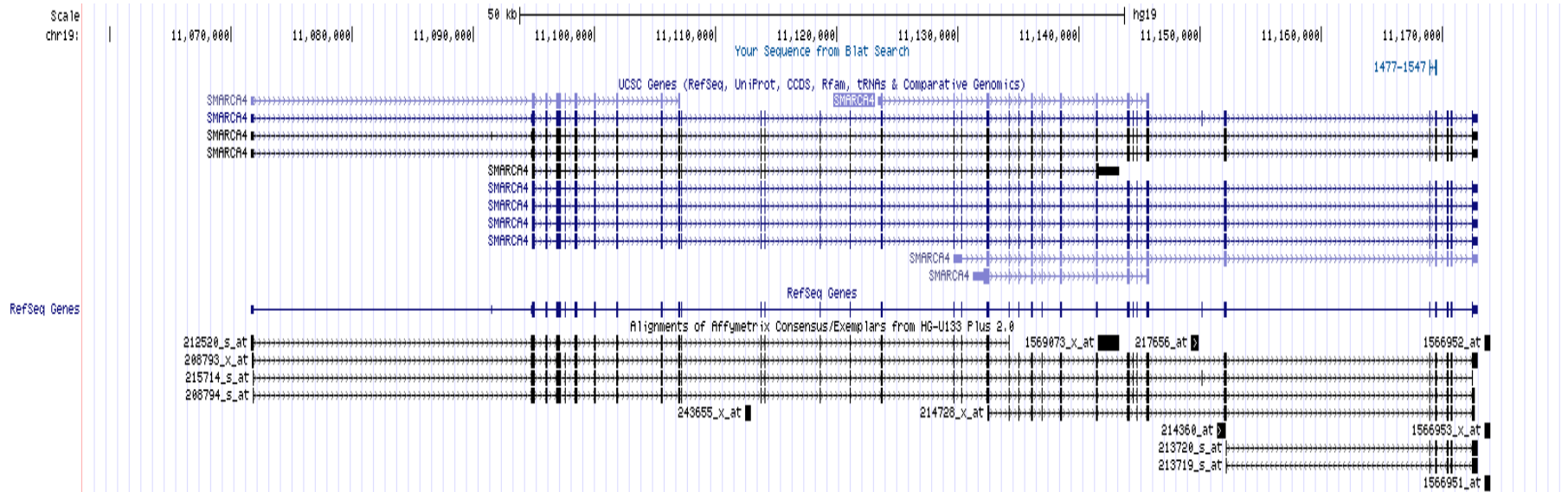


# Perspectives on target validation

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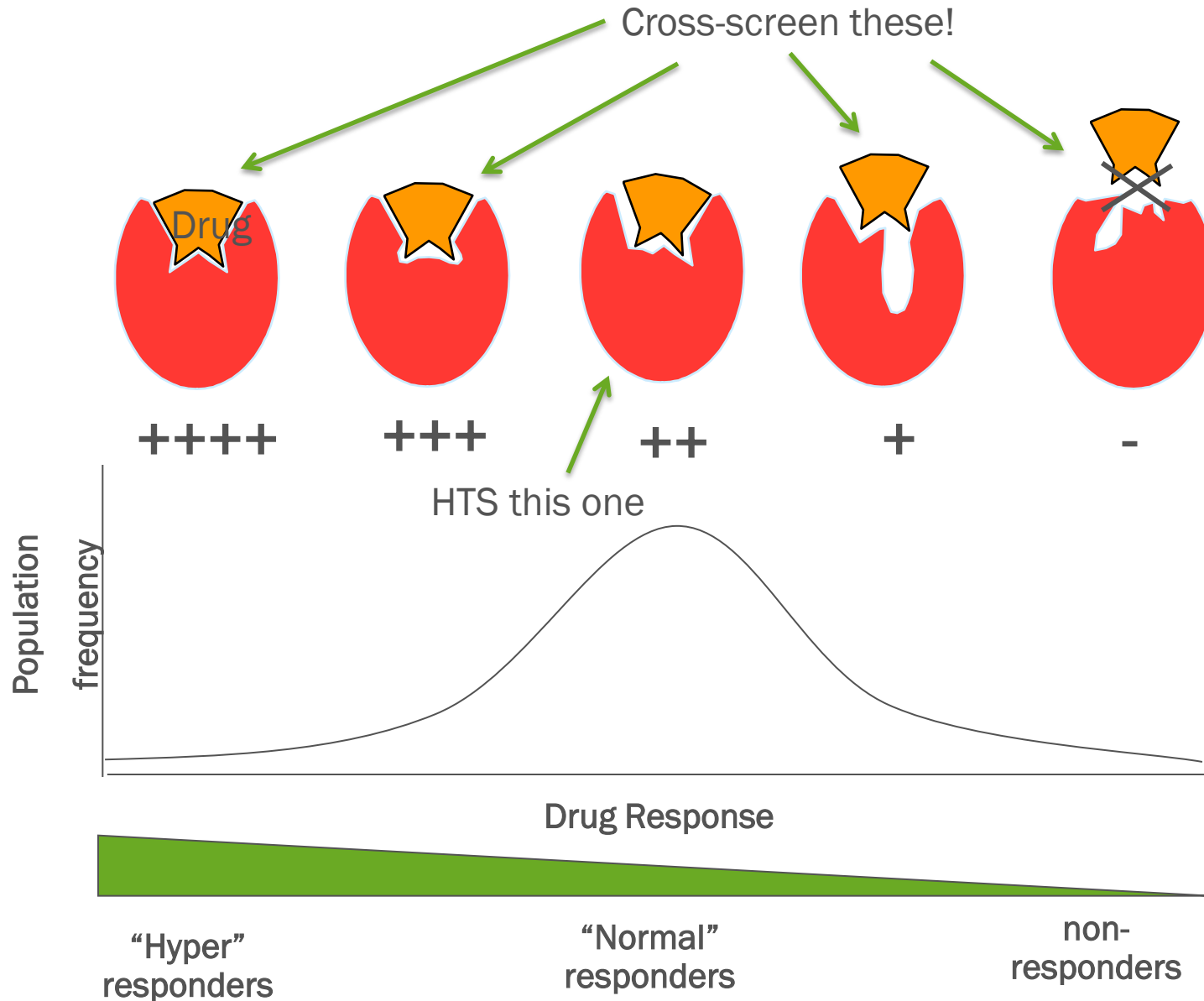


# Target Expression and Splicing



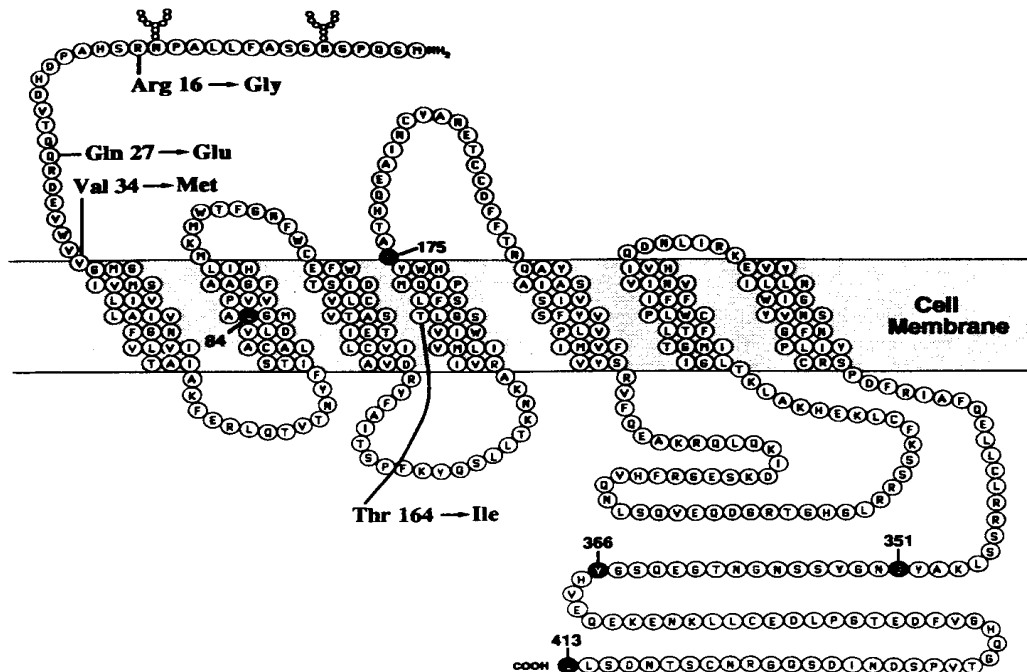
- ✗ Target validation needs to consider the most biologically relevant transcript
- ✗ UCSC genome browser presents a view of target splice isoforms and gene expression probes (“Affy U133Plus2” track)

# The Classical PGX paradigm – Target polymorphism and Therapeutic Response



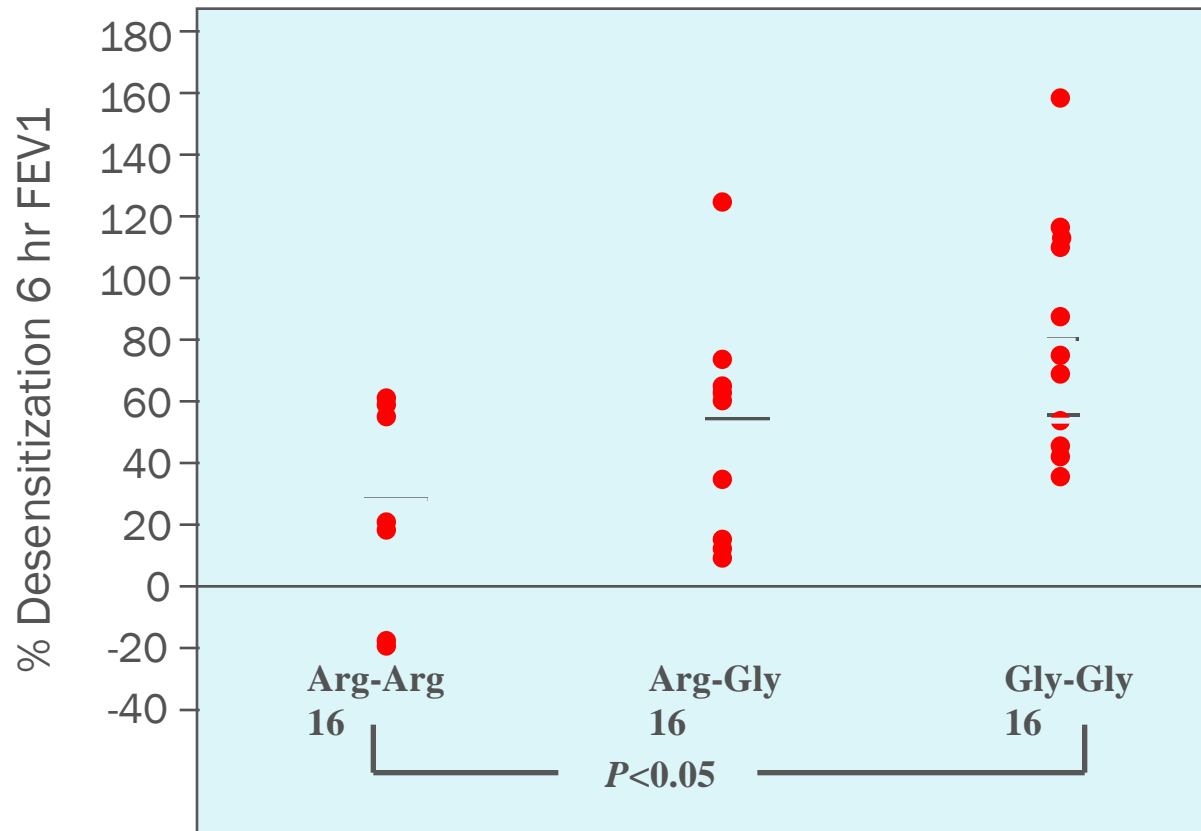
# $\beta_2$ -Adrenergic Receptor Polymorphisms

- ✗ beta2-adrenoceptor variants can lead to desensitization with regular formoterol therapy (Tan et al. 1997)





# Drug Target Pharmacogenetics – Formoterol and Desensitization



Adapted from: Tan *et al.* *Lancet* 1997;350:995–999.

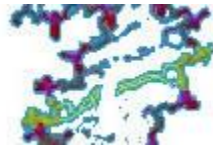
# Getting a view of target variation



## Uniprot

- ✗ “Chemist friendly”
- ✗ Good context
- ✗ Not comprehensive

**dbSNP**  
Short Genetic Variations



## NCBI dbSNP

- ✗ Comprehensive / Exhaustive
- ✗ Rare variants (<1% freq) may not be an issue for target validation, but could be a safety issue

# Perspectives on target validation

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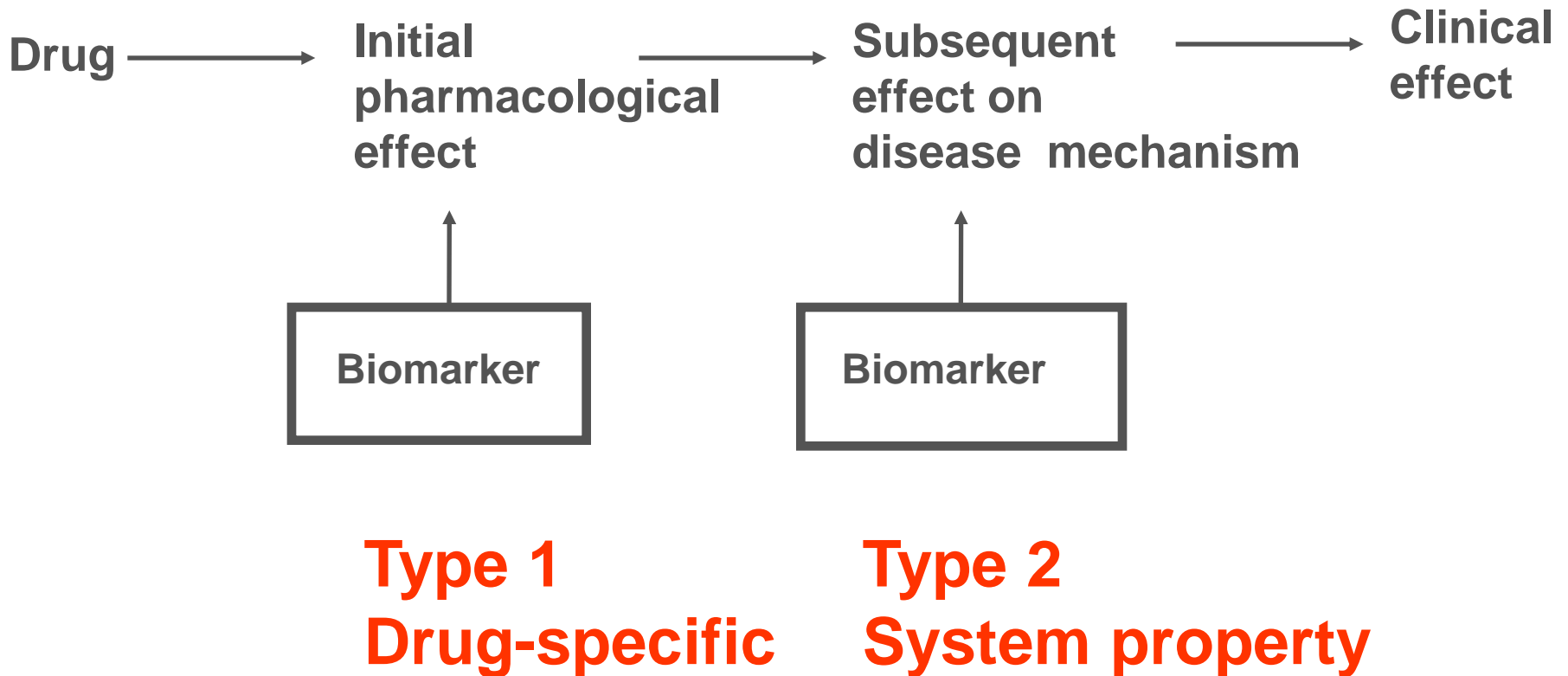
# Druggability

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✖ See Anna's Talk

# Robust Assays need Biomarkers

- ✖ Biomarkers are critical to demonstrate efficacy



# Types of Biomarker in Drug Discovery

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## Type 1

- ✖ confirmation of primary pharmacology
- ✖ supports the predictions of preclinical models
- ✖ allows PK /PD relationship assessment
- ✖ but is not really closer to predicting efficacy
- ✖ drug specific
- ✖ easy to validate

## Type 2

- ✖ biological / disease response
- ✖ an effect downstream from primary pharmacology and which is likely to result in clinical benefit
- ✖ potential diagnostic or surrogate
- ✖ system not drug specific
- ✖ because of system complexity may need to be an experimental model
- ✖ hard to validate



# So you have a target? What next?

---

## Understand target biochemistry

- ✗ kinetics – will help to inform assay development and drug design
- ✗ substrate specificity – may aid design of inhibitors
- ✗ inhibitors / agonists – may be starting point of medicinal chemistry
- ✗ protein structure – aids rational design and modelling

## Clone and express gene(s) to develop systems to assay target

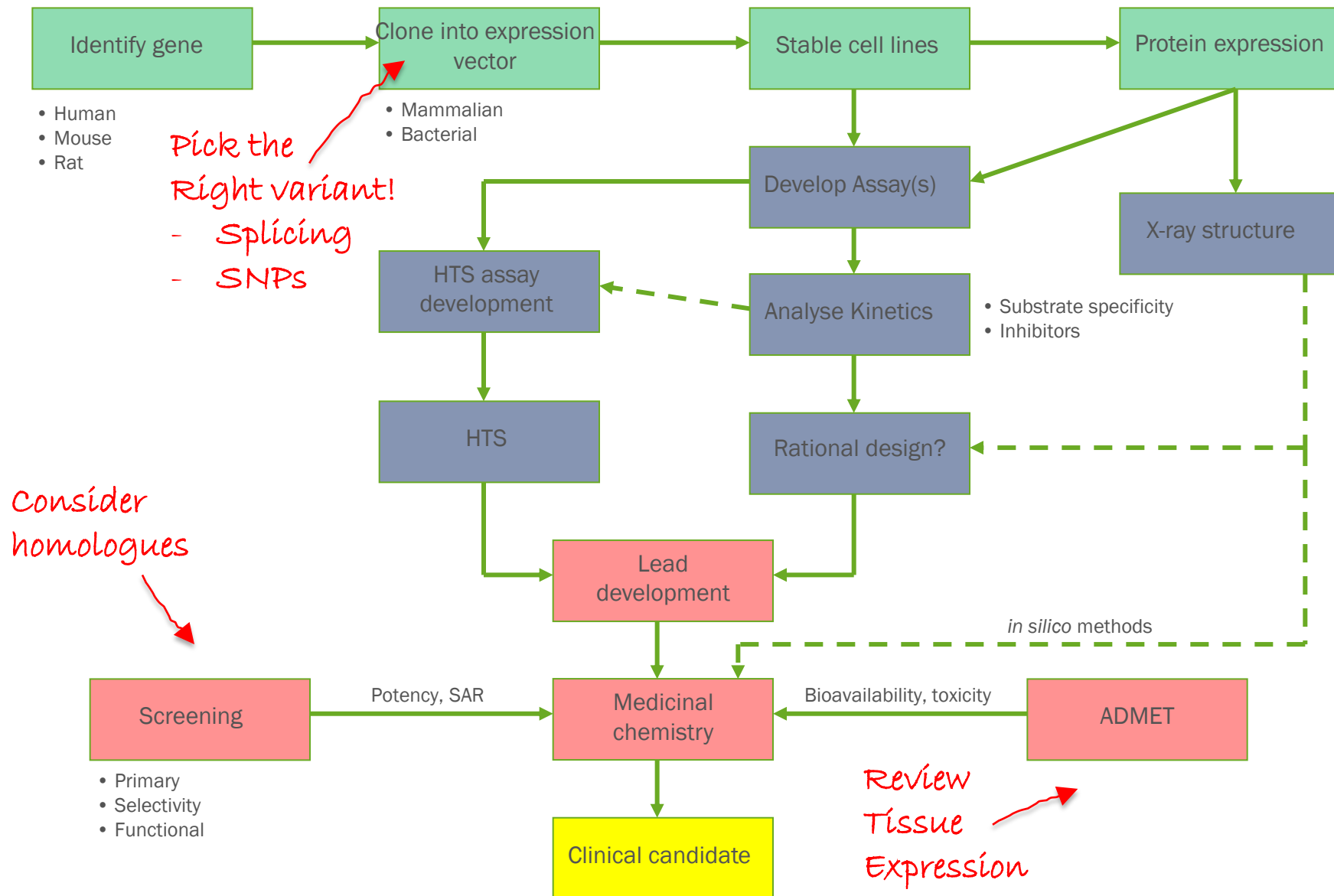
- ✗ mammalian cells – use to develop whole cell assays
- ✗ yeast or *E. coli* – used to express protein

## Assay development

- ✗ primary – target assay, kinetic or binding, solution-based (?)
- ✗ whole cell - may be primary assay
- ✗ functional - required to assess effect of target inhibition
- ✗ selectivity – required to determine specificity of leads

## High throughput screening (HTS)

# Defining a critical path to the clinic



# Time for some wet work

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# Time for some wet work....



**assay depot**

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# Seeking the Holy Grail of Drug Discovery

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## PROOF OF CONCEPT

# POC: The Holy Grail of Target Validation

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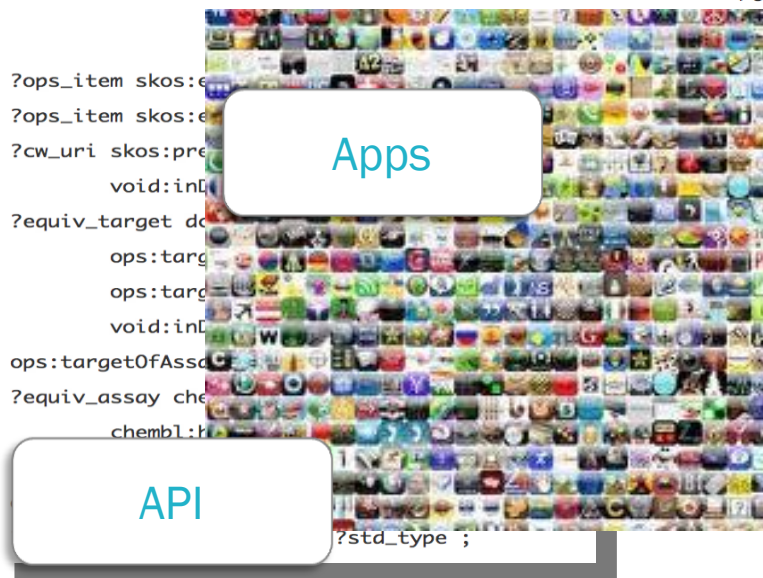
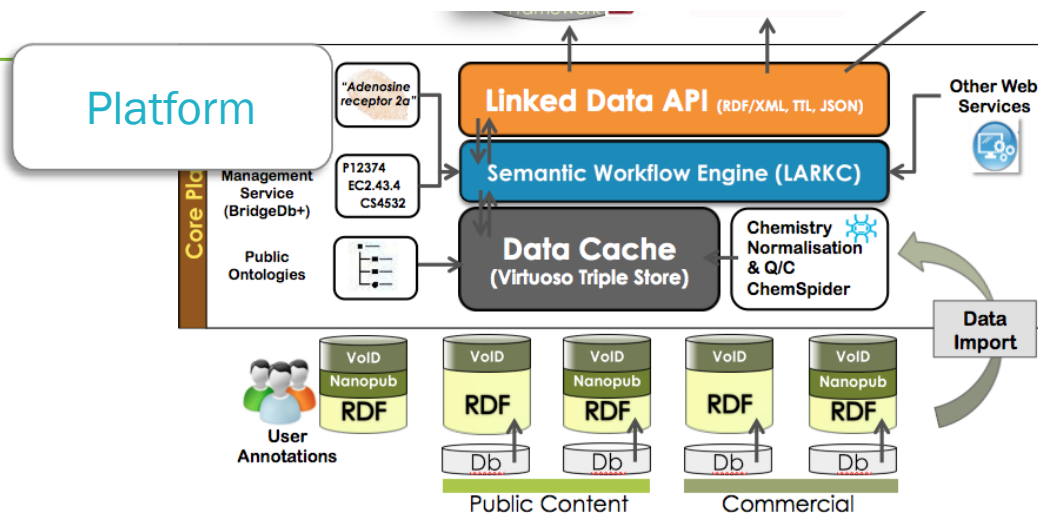
- ✖ PoC is a *critical* stage in drug discovery
  - + At PoC huge resources are committed to take a drug to market
  - + PoC is usually judged at Phase II
- ✖ Proof of Concept should show the following:
  - + That human data reflects the data generated in the animal models
  - + No unexpected side effects
  - + demonstrate efficacy in the target disease
  - + Ideally secondary benefits or indications
- ✖ PoC studies need to satisfy the following:
  - + Endpoints of study should be minimum size to demonstrate efficacy with statistical significance
  - + Placebo effect can not be ignored.



# Tools for Target Validation



# Open PHACTS



Convergence Meeting: Semantic Interoperability for Clinical Research & Patient Safety in Europe

"What is the selectivity profile of known p38 inhibitors?"



"Let me compare MW, logP and PSA for known oxidoreductase inhibitors"



"Find me compounds that inhibit targets in NFkB pathway assayed in only functional assays with a potency <1  $\mu$ M"



ChEMBL

DrugBank

Gene  
Ontology

Wikipathways

GeneGo

ChEBI

UniProt

UMLS

GVKBio

ConceptWiki

ChemSpider

TrialTrove

TR Integrity

Convergence Meeting: Semantic  
Interoperability for Clinical Research &  
Patient Safety in Europe

57

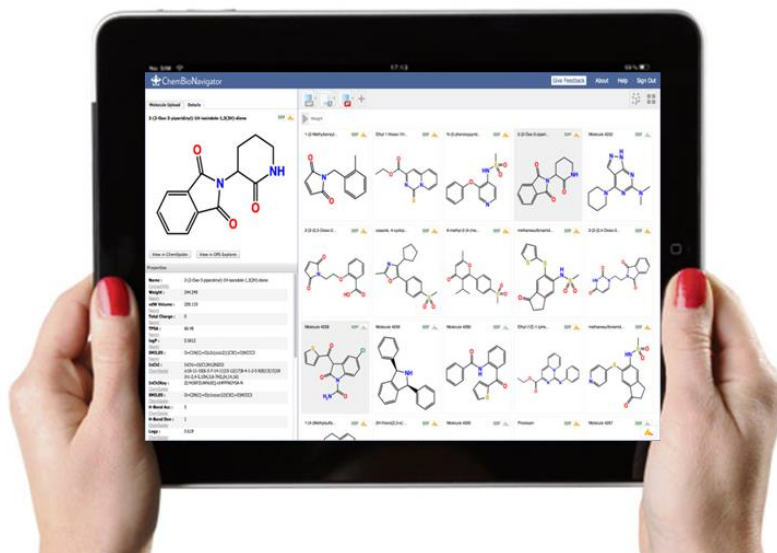
# Open Phacts Apps ([www.openphacts.org](http://www.openphacts.org))

Pharmatrek

The Pharmatrek interface shows a sidebar with filters for 'p38 alpha homo' and 'Mitogen-activated protein kinase 14 (Homo sapiens)'. The main area displays a grid of 35 targets and 546 molecules. A 'LIGANDS' section on the left lists three specific ligands: 21111-quinazolinone, 5-(2-chloro-4-fluorophenyl)-1-(2,6-dichlorophenyl)-3,4-dihydro-7-(4-piperidinyl)-, 21111-quinazolinone, 5-(2-chloro-4-fluorophenyl)-1-(2,6-dichlorophenyl)-3,4-dihydro-7-(4-piperidinyl)-, and 21111-quinazolinone, 5-(2-chlorophenyl)-1-(2,6-dichlorophenyl)-3,4-dihydro-7-(4-piperidinyl)-.

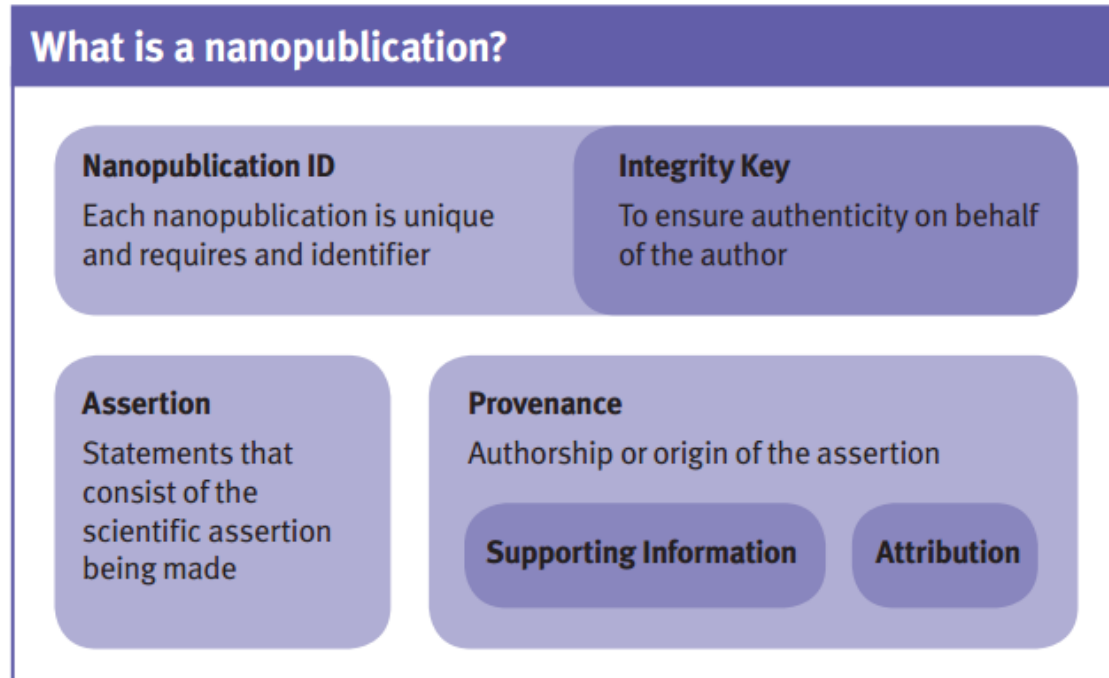
Utopia Docs

The Utopia Docs interface displays a chemical structure of 2-acetolactate and a table of related compounds. The table lists various compounds and their associated enzymes, such as 2-acetolactate, 2-acetolactate mutase, and 2-acetolactate decarboxylase. The interface also includes a 'Back to overview' button and a 'Look up' button.



ChemBioNavigator

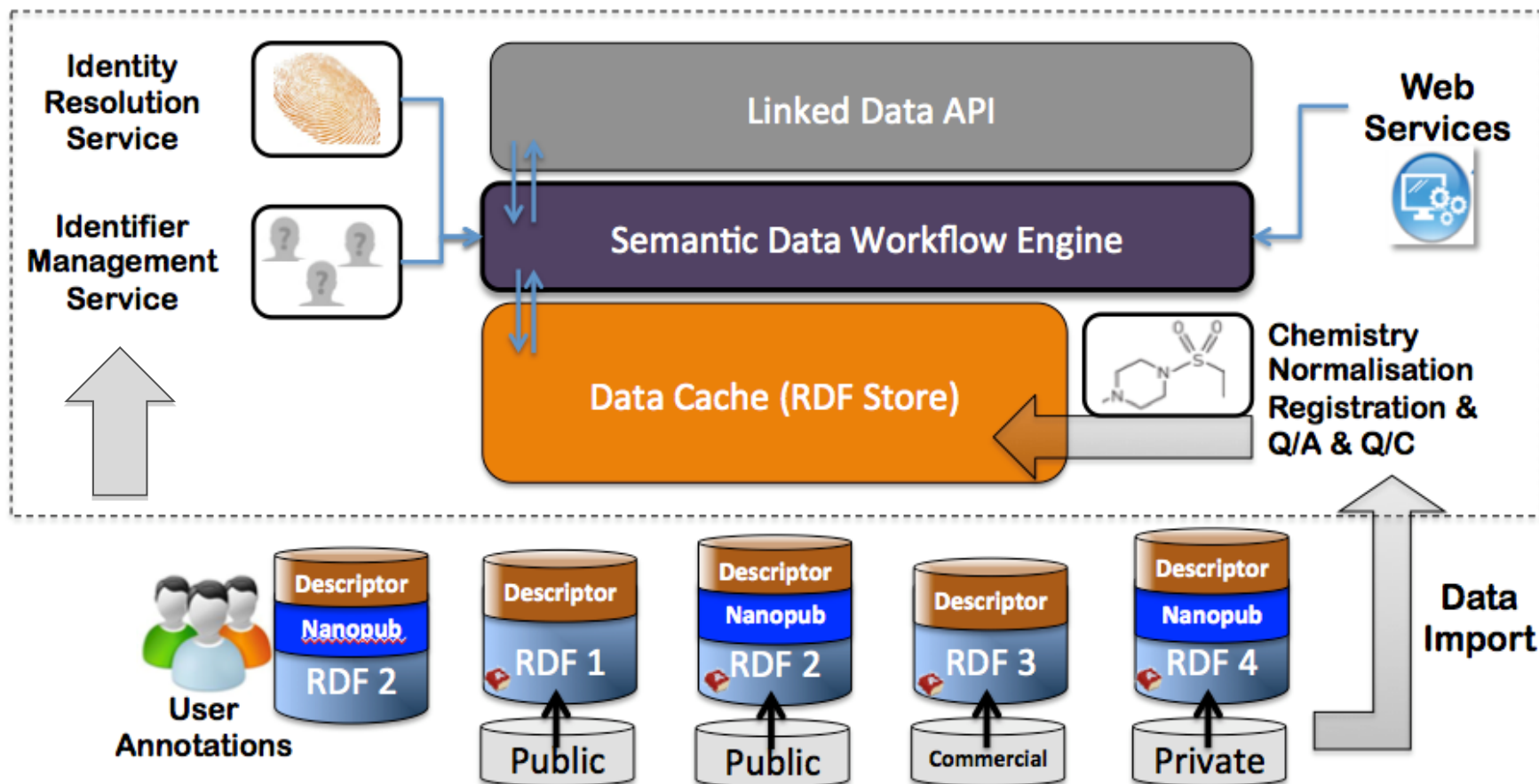
# Semantic interoperability approach



## Principles

- Respect data providers
- Make it easy for application developers

# Semantic interoperability approach





# Semantic Resources – Data sets

814,535,923 triples

| Source                 | Version                        | Supplier                            | Downloaded  | Initial Records                                    | Triples     | Properties                             |
|------------------------|--------------------------------|-------------------------------------|-------------|----------------------------------------------------|-------------|----------------------------------------|
| ChEMBL                 | ChEMBL 13<br>RDF (11-Jun-2012) | Maastricht                          | 08 Aug 2012 | 1,149,792<br>(~ 1,091,462 compounds, 8845 targets) | 146,079,194 | 17 (for compounds)<br>13 (for targets) |
| DrugBank               | Aug 2008                       | Bio2Rdf<br>(www4.wiss.fu-berlin.de) | 08 Aug 2012 | 19,628<br>(~14,000 targets, 5000 drugs)            | 517,584     | 74                                     |
| SwissProt              | 2012_07<br>(July 11, 2012)     | SIB                                 | 07 Aug 2012 | 536,789                                            | 156,569,764 | 78                                     |
| ENZYME                 | July 11, 2012                  | SIB                                 | 07 Aug 2012 | 6,187                                              | 73,838      | 2                                      |
| ChEBI                  | Release 94                     | EBI                                 | 08 Aug 2012 | 35,584                                             | 905,189     | 2                                      |
| ChemSpider<br>ACD Labs |                                | ChemSpider                          | 08 Aug 2012 | 1,194,437                                          | 161,336,857 | 22 ACD<br>4 CS                         |
| ConceptWiki            |                                | NBIC                                | 07 Aug 2012 | 2,828,966                                          | 3,739,884   | 1                                      |

# Tools for Target Validation



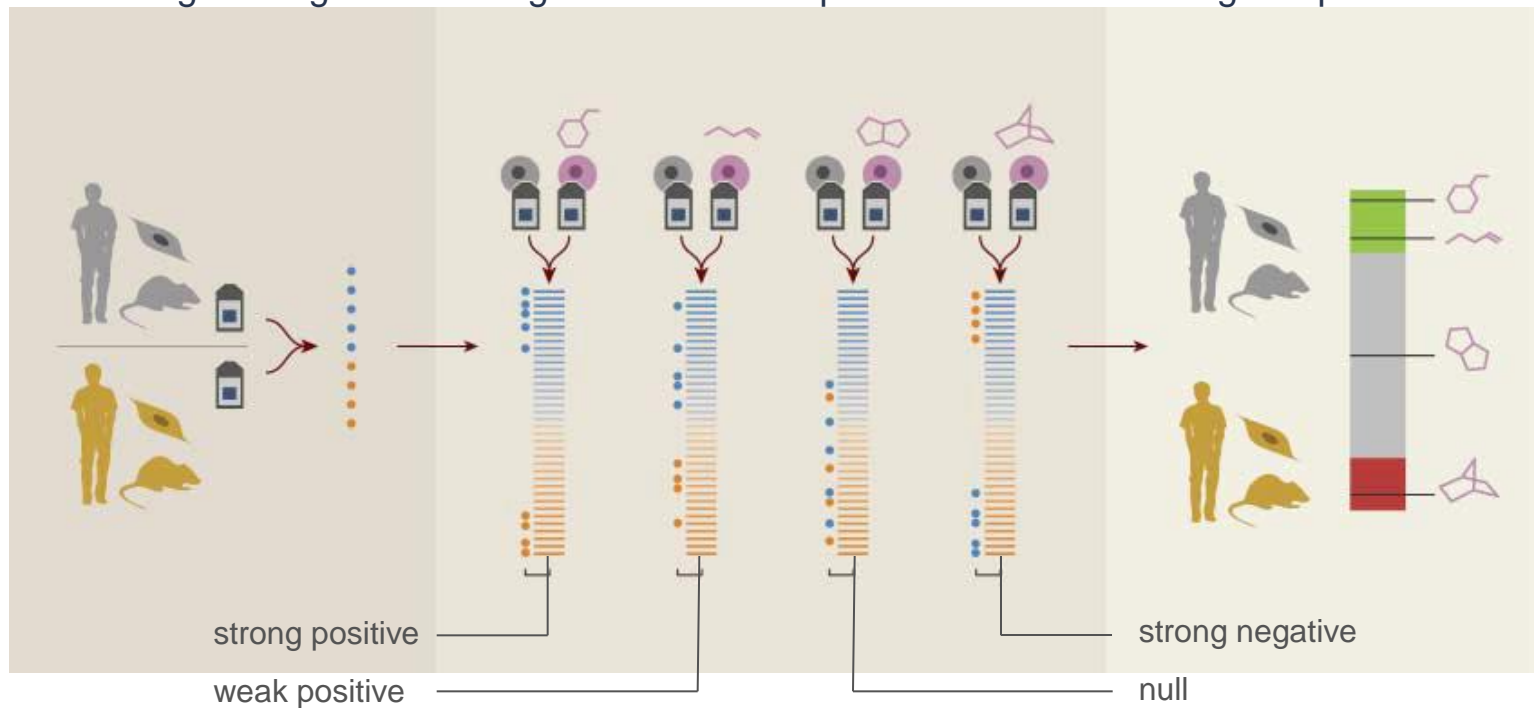
# Connectivity Map

Search database by comparing a query signature with reference signatures

Query signature – list of up- and down-regulated genes

Reference signatures - ranked gene lists for compounds

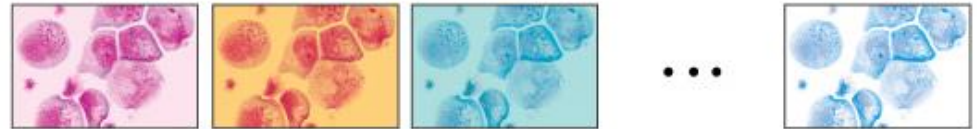
Output – lists of high and low scoring compounds



# CMAP v2.0

- ✖ Early access portal
- ✖ Drugs of interest
  - + Methotrexate
- ✖ siRNA of interest
  - + TNF/TNFR
  - + IL12/IL12RB
  - + IL17/IL17RA
  - + Pathway components
- ✖ Lincsccloud.org

15 Cell Types



Chemical & Genetic Perturbations

