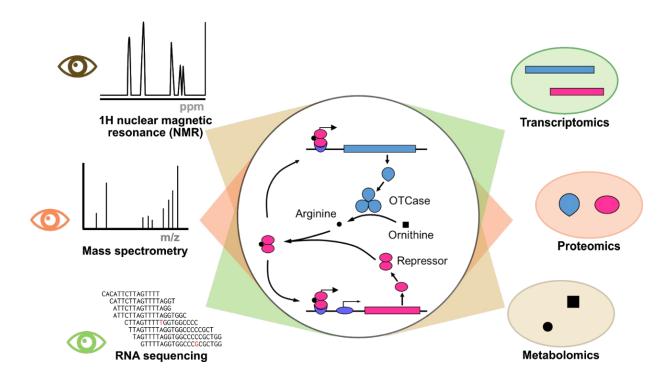# AMIDD 2024 Lecture 10: Biological networks and omics
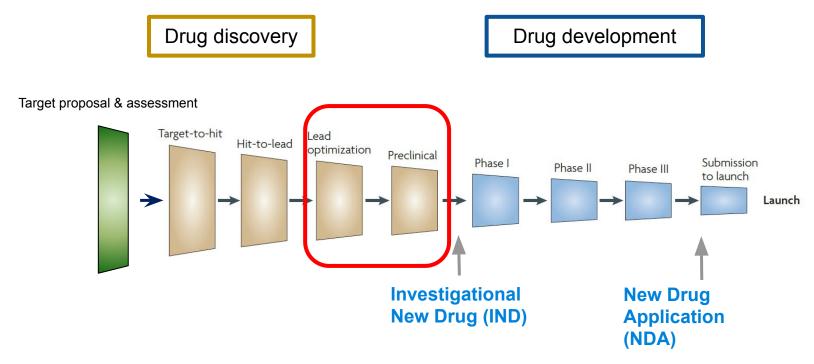
Dr. Jitao David Zhang, Computational Biologist
[1] *Pharmaceutical Sciences, Pharma Research and Early Development, Roche Innovation Center Basel, F. Hoffmann-La Roche*
[2] *Department of Mathematics and Informatics, University of Basel*

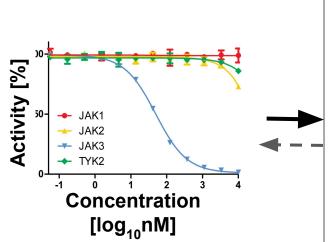# Translational research makes molecules into medicines

Adapted from Paul *et al.* "How to Improve R&D Productivity: The Pharmaceutical Industry's Grand Challenge." Nature Reviews Drug Discovery, 2010

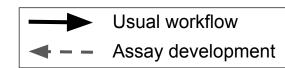# Classical workflow of efficacy and toxicity assessment

Biochemical & biophysical assays

Cellular assays (*in vitro*)

Animal experiments (*in vivo*)

→ Usual workflow

⇠ - - Assay development
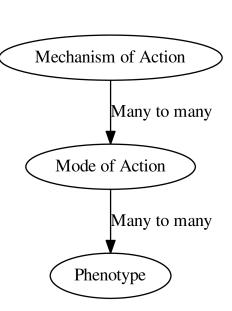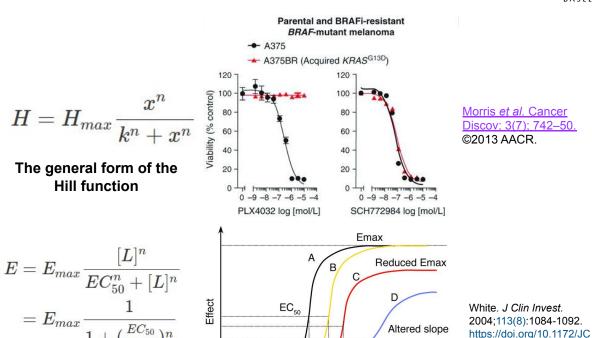
Mechanism of Action & Mode of Action

- **Mechanism of Action**: The specific biochemical interaction through which a drug substance produces its pharmacological effect, *at the molecular level*.

- **Mode of Action**: Functional or anatomical changes, *at the cellular level,* resulting from the exposure of a living organism to a substance.

- For instance, a mechanism of action of a drug can be "*binding to Monoacylglycerol lipase (MAGL)*" while its mode of action would be "*regulating endocannabinoid signaling*" and "*reducing inflammation*".

- In lead optimization (LO) and early development, our goal is to understand both the mechanism of action and the mode of action *in vitro*, *in vivo*, and in human. The term *MoA* is used to refer both.

Mechanism of Action → Many to many → Mode of Action → Many to many → Phenotype

The Hill function is a common model of *in vitro* pharmacology

- The Hill function is one of the mostly useful non-linear functions to model biological systems.

- In its general form, H_{max} indicates the maximal value to which the function is asymptotic, n is the shape parameter (known as the Hill's coefficient), and k is the reflection point, often abbreviated as XC_{50} (X=I, E, C, …), the half-saturation constant.

- The Michaelis-Menten model is a special case of the Hill function ($n=1$).

$$H = H_{max} \frac{x^n}{k^n + x^n}$$

The general form of the Hill function

$$E = E_{max} \frac{[L]^n}{EC_{50}^n + [L]^n}$$
$$= E_{max} \frac{1}{1 + \left(\frac{EC_{50}}{[L]}\right)^n}$$

Modelling the dose-dependent effect

Parental and BRAFi-resistant
BRAF-mutant melanoma

- A375
- A375BR (Acquired *KRAS*^{G13D})

Suppose it is an antiviral drug, compared with curve B, what does curve A, C, and D suggest?
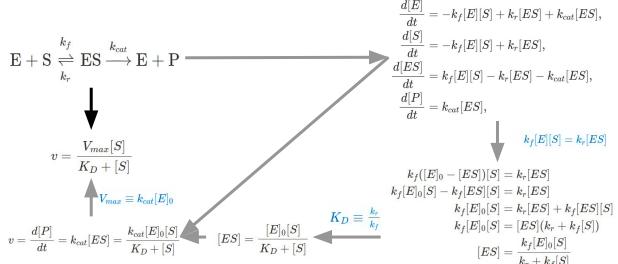
Biological networks interact with drugs and manifest its efficacy and safety

Cell membrane

Nuclear membrane

Gene expression modulated by TFs

Primary target (on-target)

Ligand/Drug

Secondary target (off-target)

Secondary target (off-target)

Protein translation

Phenotype

Transcription Factors (TFs)

Reaction Rate Equations: a compartment/ODE model of biological chemical reaction

RRE simulation of the Michaelis-Menten model

$$E + S \underset{k_r}{\overset{k_f}{\rightleftharpoons}} ES \overset{k_{cat}}{\longrightarrow} E + P$$

$$\frac{d[E]}{dt} = -k_f[E][S] + k_r[ES] + k_{cat}[ES],$$

$$\frac{d[S]}{dt} = -k_f[E][S] + k_r[ES],$$

$$\frac{d[ES]}{dt} = k_f[E][S] - k_r[ES] - k_{cat}[ES],$$

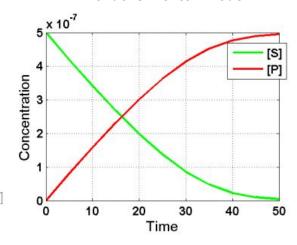$$\frac{d[P]}{dt} = k_{cat}[ES],$$

$$k_f[E][S] = k_r[ES]$$

$$v = \frac{V_{max}[S]}{K_D + [S]}$$

$$V_{max} \equiv k_{cat}[E]_0$$

$$K_D \equiv \frac{k_r}{k_f}$$

$$v = \frac{d[P]}{dt} = k_{cat}[ES] = \frac{k_{cat}[E]_0[S]}{K_D + [S]}$$

$$[ES] = \frac{[E]_0[S]}{K_D + [S]}$$

$$k_f([E]_0 - [ES])[S] = k_r[ES]$$
$$k_f[E]_0[S] - k_f[ES][S] = k_r[ES]$$
$$k_f[E]_0[S] = k_r[ES] + k_f[ES][S]$$
$$k_f[E]_0[S] = [ES](k_r + k_f[S])$$

$$[ES] = \frac{k_f[E]_0[S]}{k_r + k_f[S]}$$

$$[ES] = \frac{k_f[E]_0[S]}{k_f(\frac{k_r}{k_f} + [S])}$$

Source: Systems Engineering Wiki (tue.nl)

RRE is a set of ODEs, with each ODE representing one chemical species. Solution of the *j*th equation at time *t* is a real number representing the concentration of species *j* at time *t*.

Simulation of biological networks with ordinary differential expression

Given the reaction

$$S + E \underset{k_2}{\overset{k_1}{\rightleftharpoons}} C \overset{k_3}{\longrightarrow} P + E$$

Given the initial values and rate constants

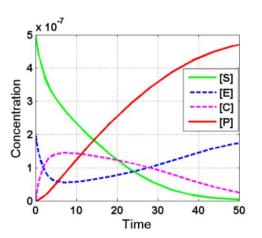- $S(0) = 5e^{-7}$
- $E(0) = 2e^{-7}$
- $C(0) = P(0) = 0$
- $k_1 = 1e^6$
- $k_2 = 1e^{-4}$
- $k_3 = 0.1$

According to the law of mass action

$$\frac{d[S]}{dt} = -k_1[E][S] + k_2[C],$$

$$\frac{d[E]}{dt} = -k_1[E][S] + (k_2 + k_3)[C],$$

$$\frac{d[C]}{dt} = k_1[E][S] - (k_2 + k_3)[C],$$

$$\frac{d[P]}{dt} = k_3[C],$$

It is possible to simulate the concentration changes by time *deterministically*.

See Systems Engineering Wiki (tue.nl) for MATLAB/COPASI codes and
Stochastic Modelling for Systems Biology by Darren J. Wilkinson

Simulating behavior of complex ODE systems with COPASI

- COPASI, freely available at http://COPASI.org/, supports both **ordinary differential equation (ODE)** based simulation as well as stochastic kinetic simulation.

- Such tools are important for detailed analysis of enzymatic reactions, for instance in the presence of drugs and/or disease-relevant mutation.
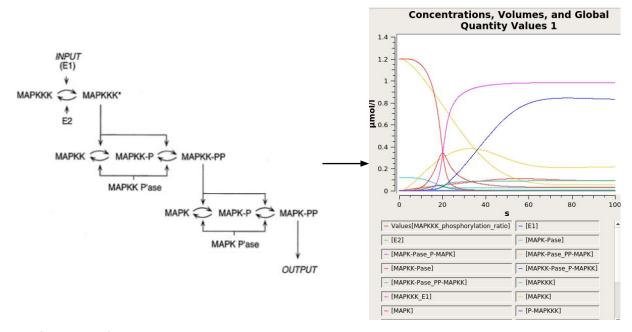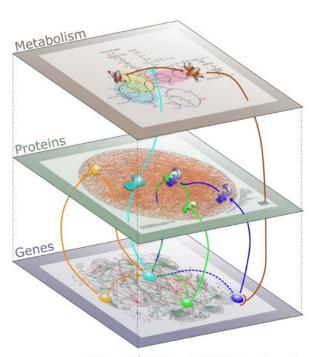

ODE-based simulation of dynamics

Figure: Huang and Ferrell, PNAS, 2006. Resources to learn more about stochastic modelling: MIT OpenCourseWare by Jeff Gore, and Stochastic Processes: An Introduction, Third Edition by Jones and Smith. Tutorials also available on the website of European Bioinformatics Institute (EBI)

Modelling biological networks
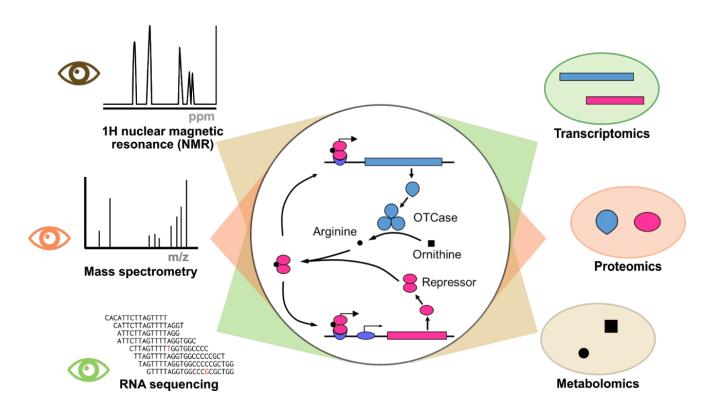
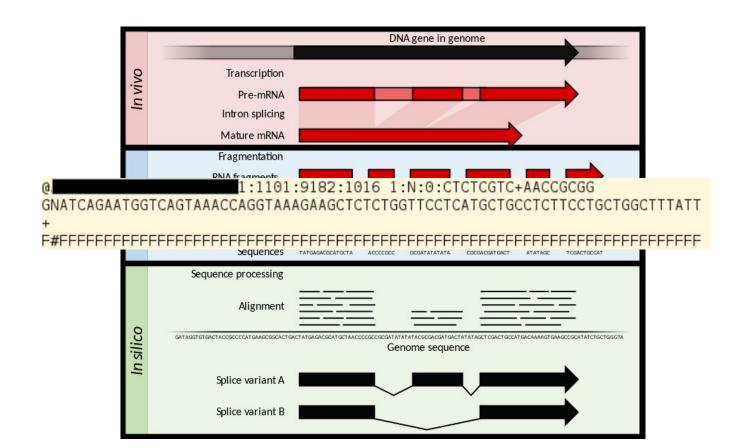Stéphane CHÉDIN & Jean LABARRE, www-dsv.cea.fr

Garg, Abhishek, Kartik Mohanram, Giovanni De Micheli, and Ioannis Xenarios. 2012. "Implicit Methods for Qualitative Modeling of Gene Regulatory Networks." In *Gene Regulatory Networks: Methods and Protocols*, edited by Bart Deplancke and Nele Gheldof, 397–443. Methods in Molecular Biology. Totowa, NJ: Humana Press.
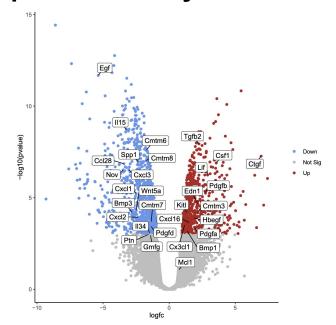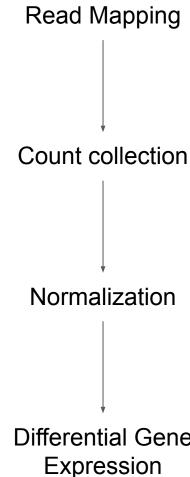
Biological networks can be studied with omics technologies

Principle of next-generation RNA sequencing (NGS)

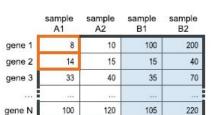**We can reveal compound's effect on gene expression by performing differential gene expression analysis**

Read Mapping

Count collection

Normalization

Differential Gene Expression Analysis

Visualization (e.g. volcano plot)

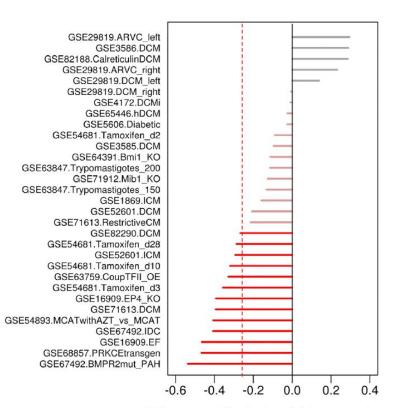# Advantages and challenges of using RNA sequencing to study MoA

Advantages:

1. Since patient samples can be also profiled with omics technologies, it is possible to compare a compound's effect with the changes induced by disease progression (right).

2. Well-designed omics study can reveal strong and subtle effects of the compound (the example with splicing modifier).
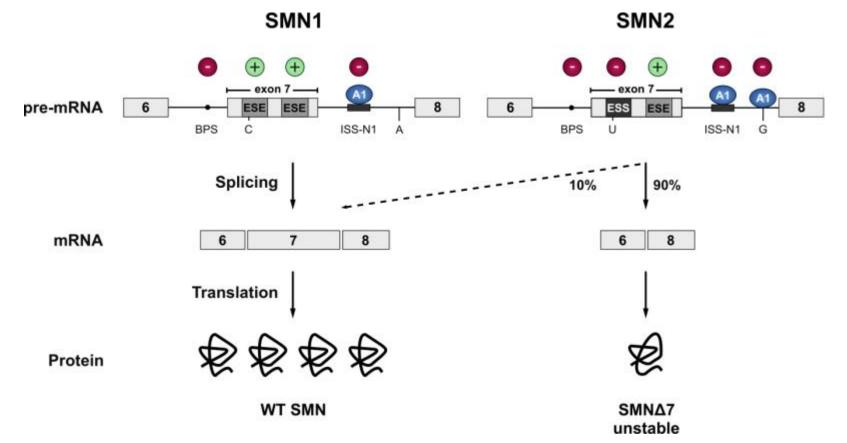
Challenges:

1. Data from biological models that poorly reflect human disease can do more harm than benefits.

2. Curse of dimensionality.

Pathway regulation by beneficial compounds and in cardiomyopathy: the correlation

Splicing of SMN1 and SMN2 genes: patients with mutations in SMN1 gene suffer from Spinal Muscle Atrophy (SMA)
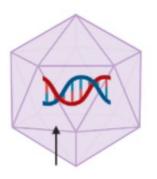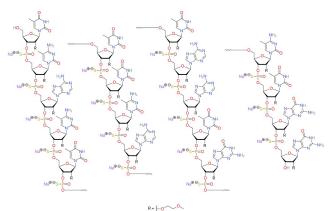
Three drugs of different modalities are approved to treat SMA
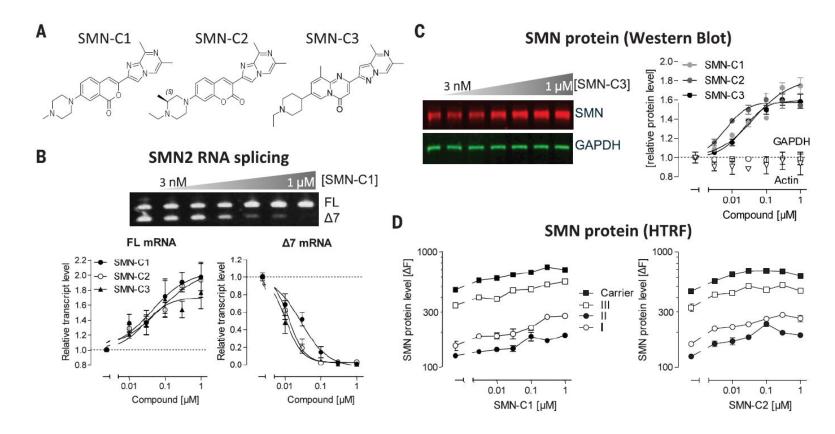
AAV9 capsid

SMN1 gene

Onasemnogene
Abeparvovec/
Zolgensma

Nusinersen sodium/ Spinraza
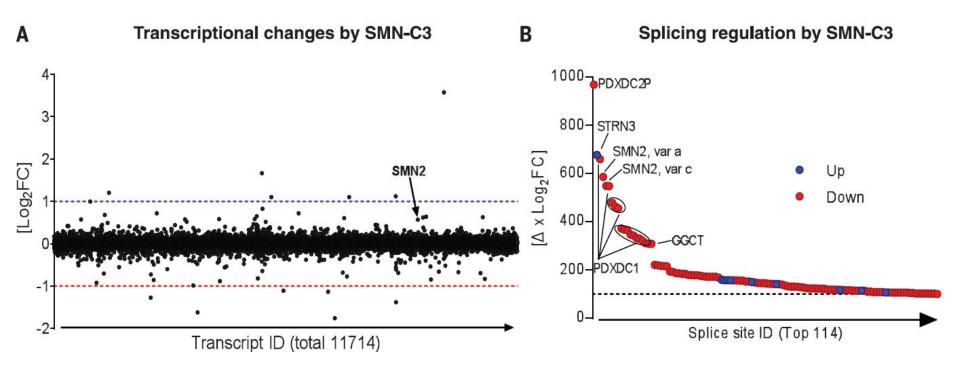(CHEMBL3833342)

Risdiplam/ *Evrysdi*
(CHEMBL4297528)

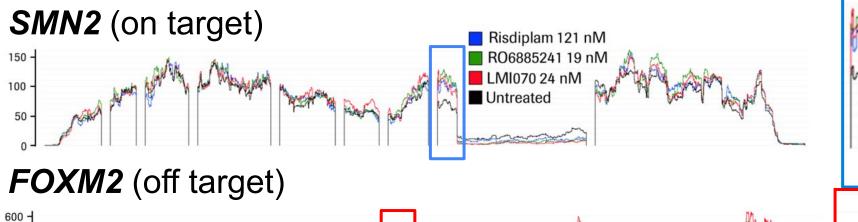# Small molecules were identified as RNA splicing modifiers

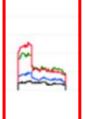# RNA sequencing confirms the specificity of SMN-C3

A Transcriptional changes by SMN-C3

B Splicing regulation by SMN-C3

RNA sequencing confirms the superior safety profile of SMN-C3 over other compounds

SMN2 (on target)

FOXM2 (off target)

Legend:
- Risdiplam 121 nM (blue)
- RO6885241 19 nM (green)
- LMI070 24 nM (red)
- Untreated (black)
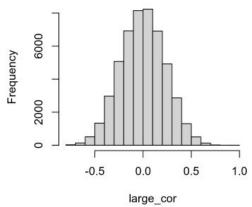
Given enough tests, there will be significant results
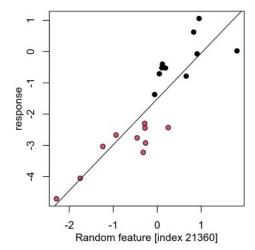

Histogram of large_cor

```r
set.seed(1887)
patient_group <- gl(2,10)
response <- c(rnorm(10, 0), rnorm(10, -3))
random_features_large <- matrix(rnorm(20*50000), nrow=20)
large_cor <- cor(response, random_features_large, method="spearman")
hist(large_cor)
```

```r
largest_cor_ind <- which.max(large_cor)
{
  compactPar()
  plot(random_features_large[, largest_cor_ind],
       response,
       bg=patient_group,pch=21,
       xlab=sprintf("Random feature [index %d]", largest_cor_ind))
  abline(lm(response ~ random_features_large[, largest_cor_ind]))
}
```
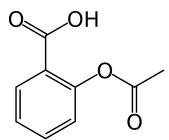
The road of MoA understanding can be 120 year long

Aspirin trademarked in 1899

Dai *et al*, Cell, 2019

Acetylation blocks cGAS activity and inhibits self-DNA-induced autoimmunity

- Acetylation suppresses cGAS activity
- Aspirin directly acetylates cGAS
- Aspirin inhibits cGAS-mediated interferon production
- Aspirin alleviates DNA-induced autoimmunity in AGS mouse models and patient cells
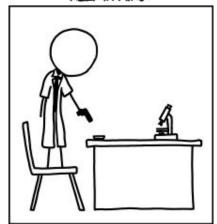
MoA understanding can be a long process full of surprises

Summary

1. In lead optimization and early development, we are interested in MoA of drug candidates *in vitro*, *in vivo*, and in human.

2. We can study MoA by modeling biological networks, for instance with ODE-based models and its variants.

3. We can also study MoA by performing omics experiments and analysing the data with statistical, machine-learning or AI tools. It is helpful to keep both advantages and challenges in mind.

WHEN YOU SEE A CLAIM THAT A COMMON DRUG OR VITAMIN "KILLS CANCER CELLS IN A PETRI DISH,"

KEEP IN MIND:

SO DOES A HANDGUN.

https://xkcd.com/1217/