

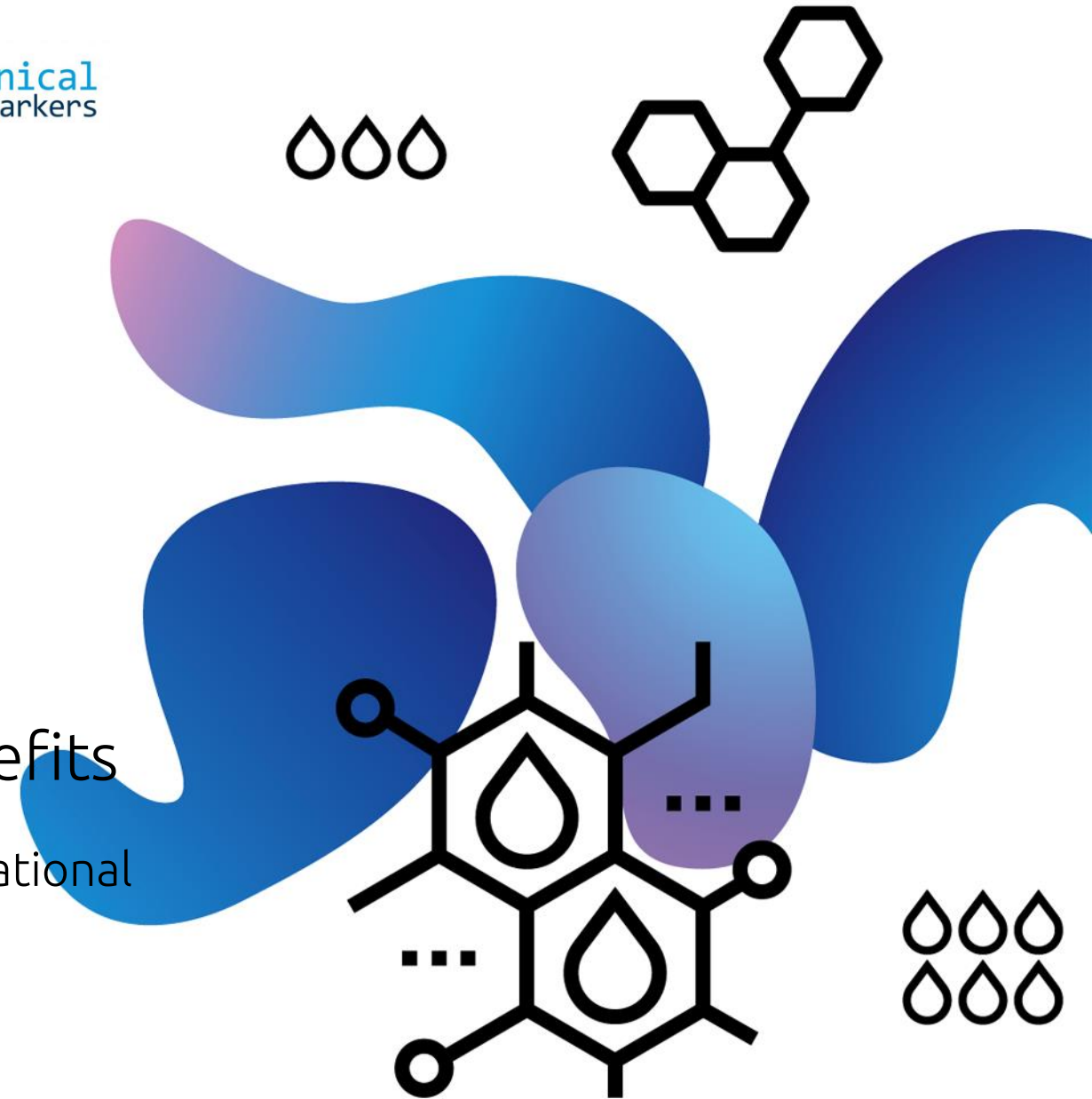


# Biomarkers in the pharmaceutical industry.

Translating research into clinical benefits

Peter Groenen, Sr. Director, Sr. Group leader Translational Biomarkers

COST CliniMARK– Summer School – 24 September 2019





# Idorsia Pharmaceuticals Ltd.

Founded June 16, 2017 as a demerger from Actelion-  
JnJ



# 50 minutes of roller coaster



# Disclaimer:

Everything presented and explained here today are my own words, thoughts and opinions. They do not represent those of Idorsia or any other organization I work or have worked for and are properly cited when derived from a third party.

Also

My discussion may include predictions, estimates or other information that might be considered forward-looking. While these forward-looking statements represent my current judgment on what the future holds, they are subject to risks and uncertainties that could cause actual results to differ materially. You are cautioned not to place undue reliance on these forward-looking statements, which reflect our opinions only as of the date of this presentation. Please keep in mind that I am not obligating myself to revise or publicly release the results of any revision to these forward-looking statements in light of new information or future events.



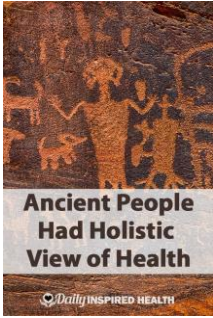
# The history of medicine and pharmaceuticals in 5 minutes



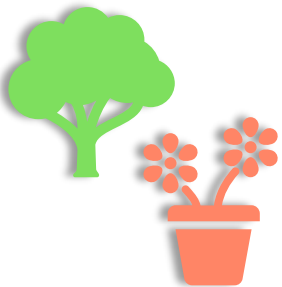
**“IT'S FAR MORE IMPORTANT TO KNOW WHAT  
PERSON THE DISEASE HAS THAN WHAT DISEASE  
THE PERSON HAS.”**

**HIPPOCRATES  
470-360 BC**

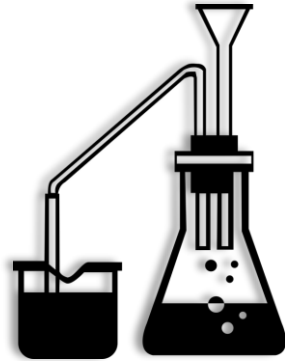
# History of the pharmaceutical industry in 3 steps



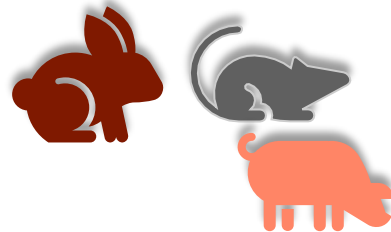
The first generally accepted use of plants as healing agents was depicted in the cave paintings discovered in the Lascaux caves in France, which have been radiocarbon-dated to between 13,000 and 25,000 BC.



- Until 1847 empirical research in humans
- Herbs, plants and minerals



- Since 1847 Pharmacology as a scientific discipline
- Since 1859 Chemical synthesis
- Empirical research in humans and animals



- Since the 1970's:
- Molecular biology
- Experimental research
- Industrialization of R&D: targeted rationalized drug discovery and development

Interim Question  
Which Drug has  
saved most lives until  
now?



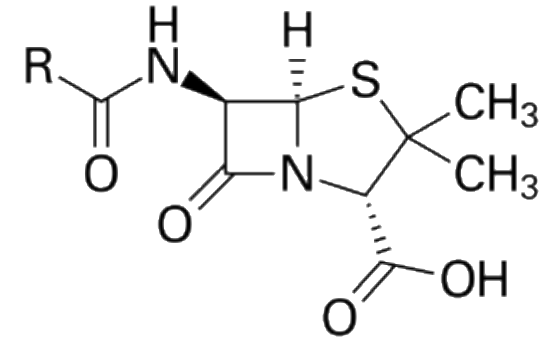
# A life saver

A lot of drugs work very well fortunately

## Penicillin – 1942

Penicillin was first identified in 1928 (Fleming), but started to be used in 1942. (First isolation and *in vivo* efficacy demonstrated in 39 by Florey & Chain, first patient treated in 42). Turning point in human history and led the way in the treatment of numerous bacterial diseases.

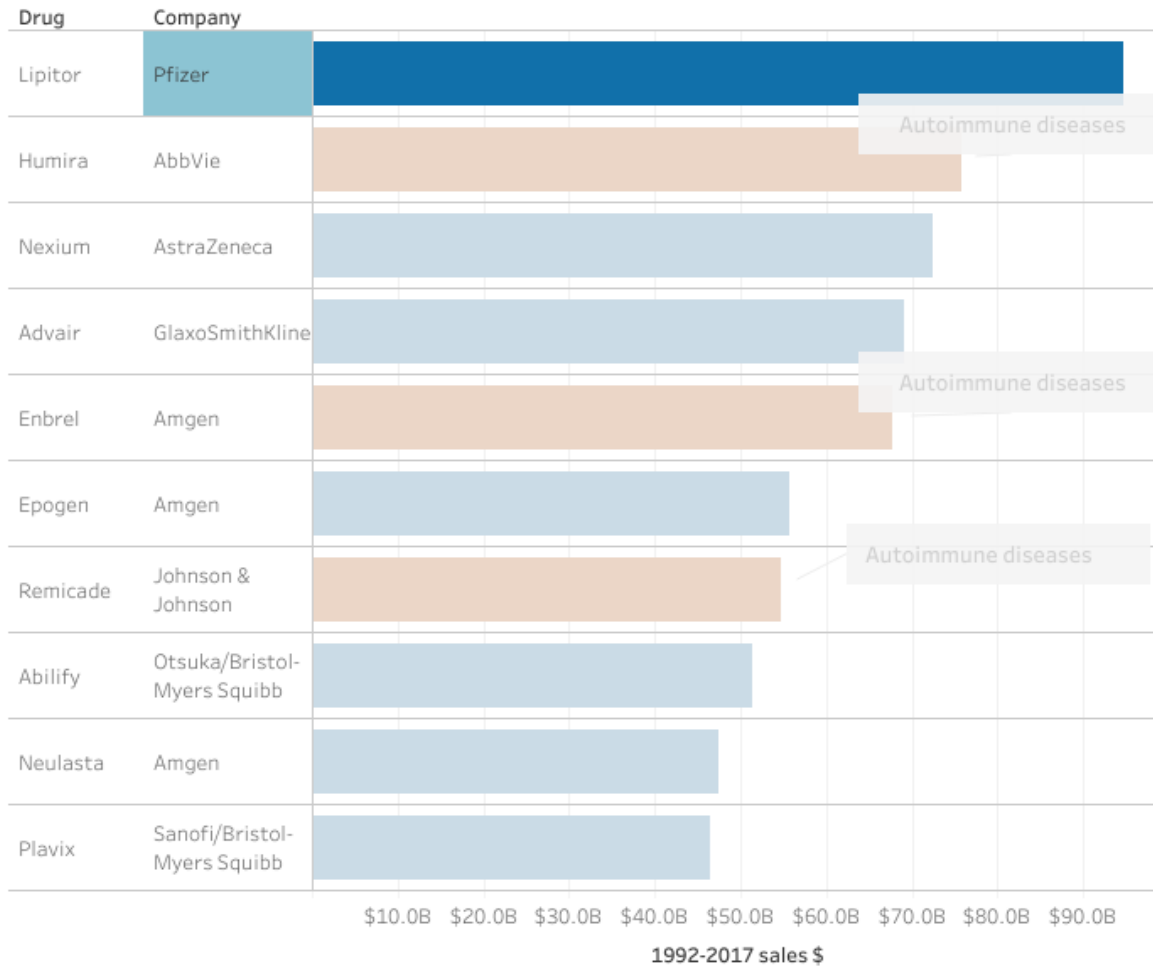
It has been calculated that the antibiotic has saved over 80 million lives and without its discovery and implementation, 75% of people today would not be alive because their ancestors would have succumbed to infection.



Interim Question  
Which Drug was the  
biggest seller ever?

# Best selling drugs to date (in 2017)

Best-selling U.S. drugs over 25 years



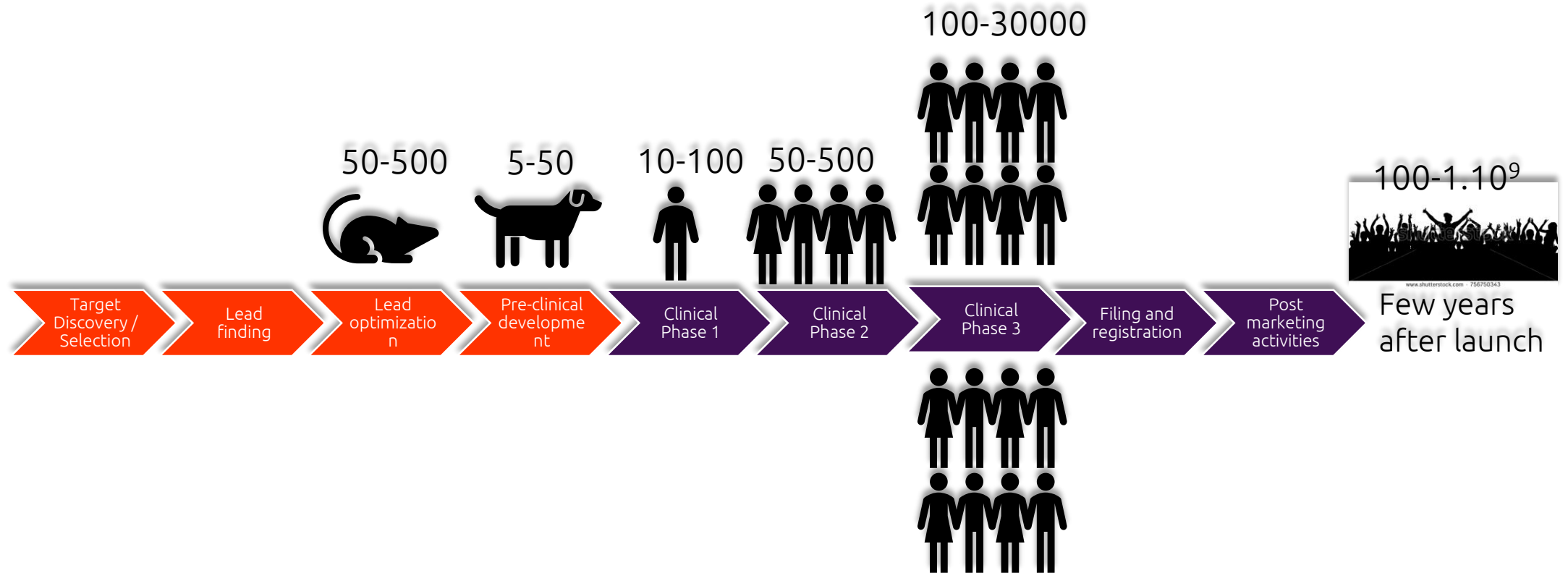
Humira is probably going to take the 1<sup>st</sup> spot in 2020

# The modern pharmaceutical R&D



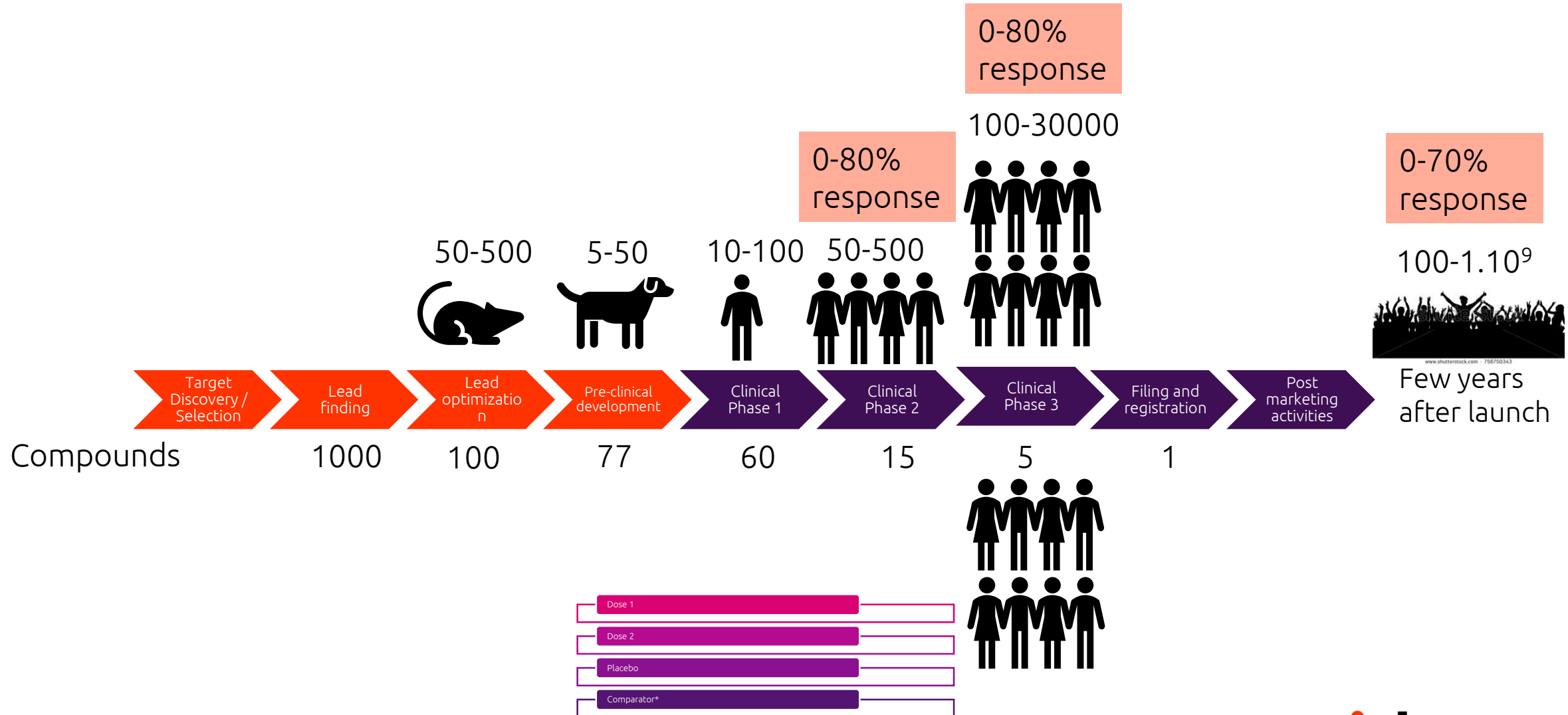
# Current Pharmaceutical R&D

## Numbers



# Current Pharmaceutical R&D

## Success rates



# Success rates per organ system

Table 1 | **Average success rate, sales and share of the total number of R&D projects\***

Anatomical Therapeutic Classification (ATC1)	Number of projects	Average sales (US\$ million)	Average POS (%)	Percentage of total projects		
				1990–1999	2000–2007	Change <sup>‡</sup>
L: Antineoplastic and immunomodulating agents	6,566	105.3	1.80	21.77	29.77	+8.00
Including L01: Antineoplastic agents	5,094	92.0	1.29	16.55	23.43	+6.88
N: Nervous system	3,817	43.5	2.85	14.46	15.55	+1.09
B: Blood and blood-forming organs	822	72.9	3.81	4.11	2.38	–1.73
J: Anti-infectives for systemic use	4,737	82.4	3.92	18.85	18.41	–0.44
M: Musculoskeletal system	1,472	22.6	4.19	6.49	5.10	–1.39
A: Alimentary tract and metabolism	2,046	14.8	4.46	7.26	8.82	+1.56
R: Respiratory system	1,165	13.3	4.81	5.07	4.10	–0.97
C: Cardiovascular system	2,139	45.6	4.86	10.72	6.15	–4.57
D: Dermatologicals	859	4.4	6.64	3.63	3.13	–0.50
G: Genitourinary system and sex hormones	865	21.0	11.75	3.95	2.86	–1.09
Other (H+P+S) <sup>§</sup>	945	11.2	19.79	3.70	3.73	+0.04

POS, probability of success; R&D, research and development. \*The top ten areas in terms of activity are defined according to the top level of the ATC system. †All differences are statistically significant ( $P$ -value < 5 %) except for class J and the residual class 'Other'. §H represents systemic hormonal preparations, excluding sex hormones and insulins; P represents antiparasitic products, insecticides and repellents; S represents sensory organs. Source: analysis of the Pharmaceutical Industry Database (BOX 1).

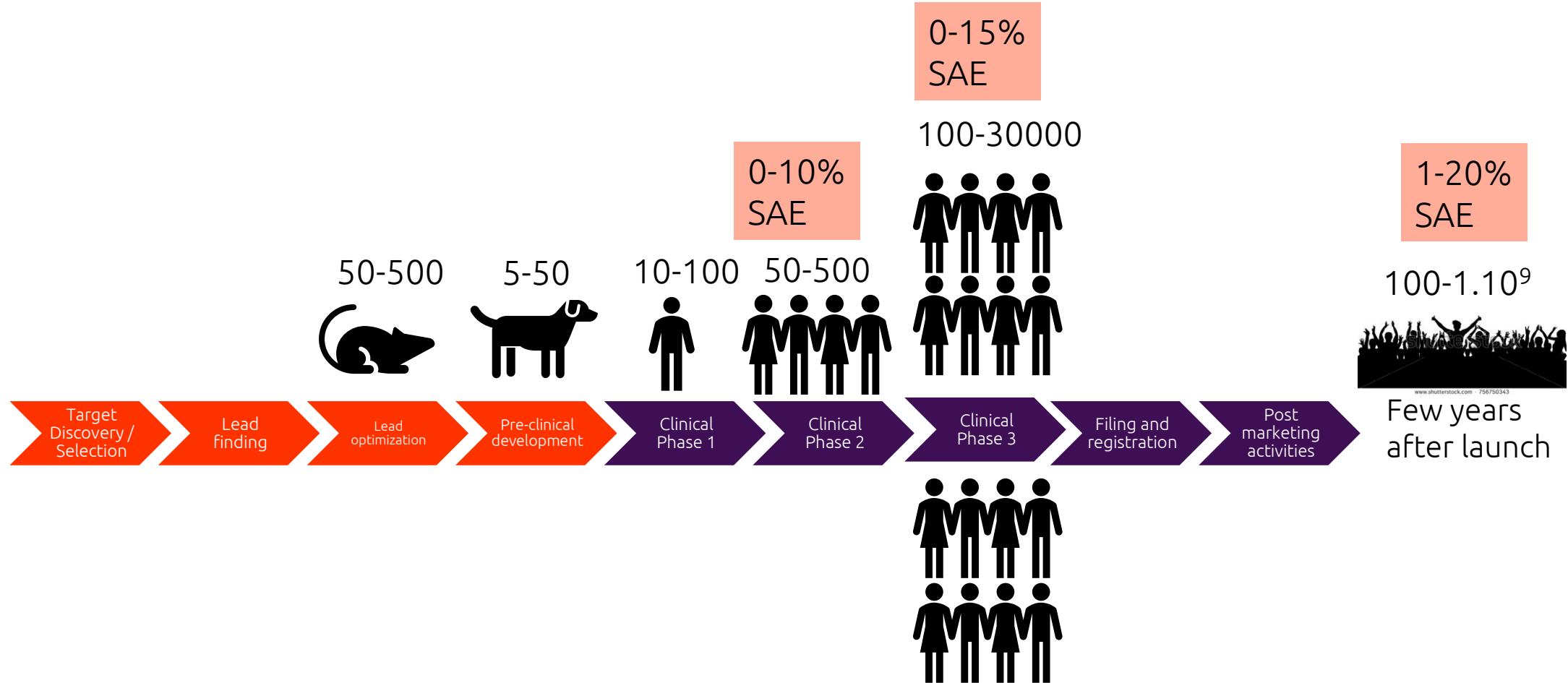
<https://www.nature.com/articles/nrd3405>

From Pammolli et al. Nature Reviews Drug Discovery 2011



# Current Pharmaceutical R&D

## Side effects





# Successful Drug discovery and development is team work!

© The New Yorker Collection 1980 Eldon Dedini from cartoonbank.com.  
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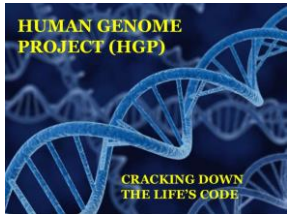
*"Ssh, gentlemen. I believe Watkins is on the verge of a new miracle drug."*



When and why did  
we start talking  
about biomarkers?

# Biomarkers & trends

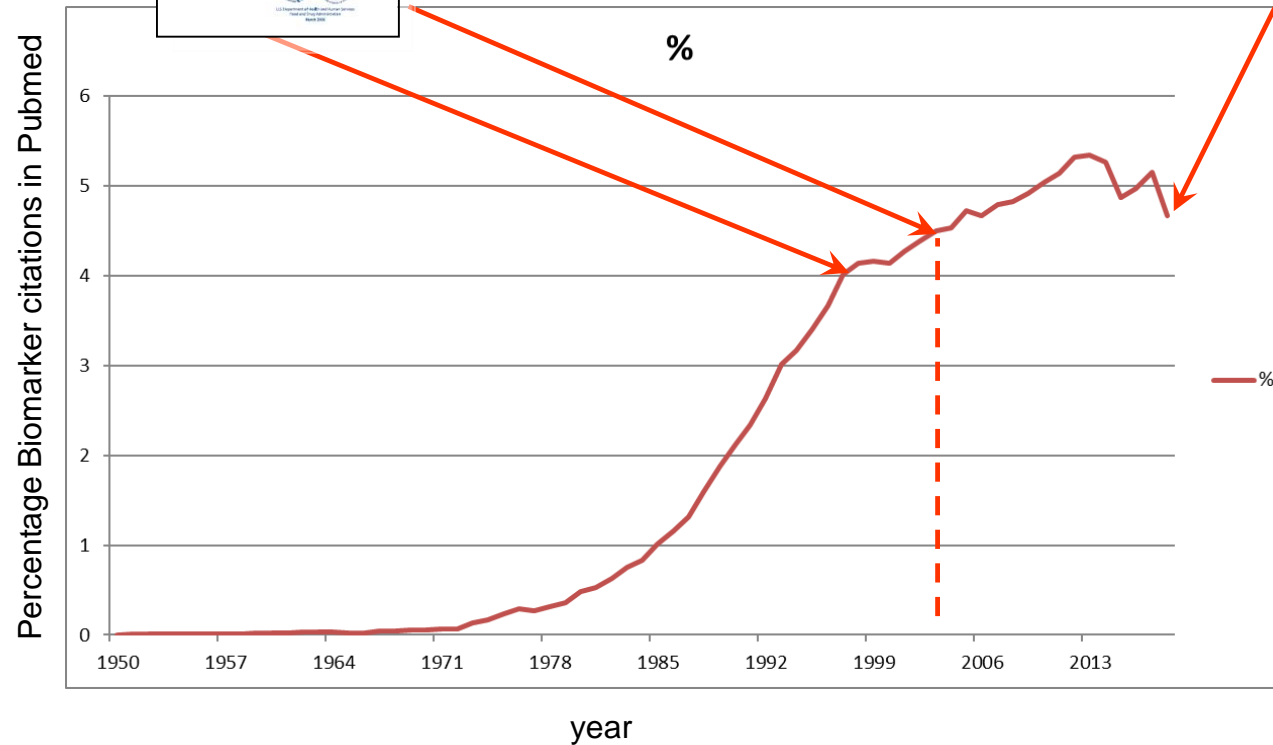
Has biomarker research reached a plateau?



Start  
Human  
Genome  
project

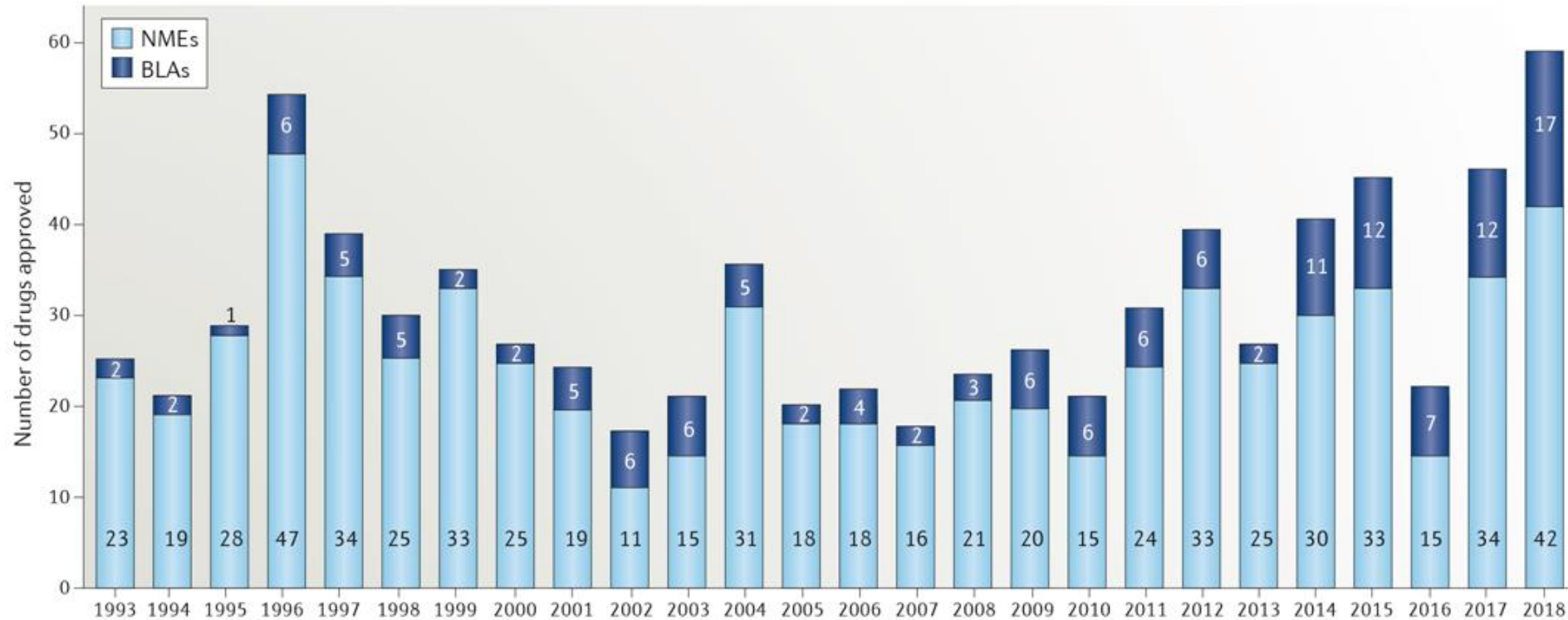


White paper from the FDA



Reduced interest in 2018?

# From the 2018 FDA drug approvals you may think we're uphill again



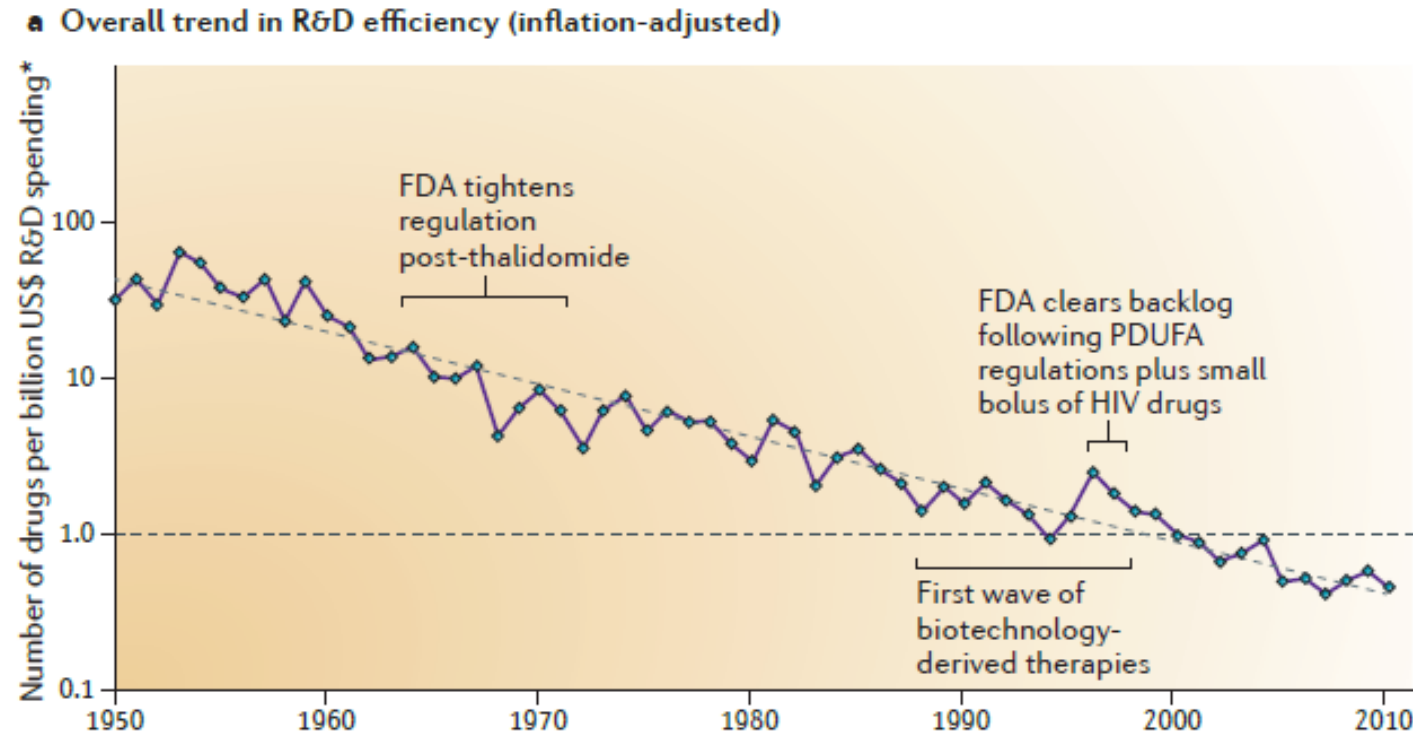
*From Mullard, Nature Reviews Drug Discovery (2019)*

<https://www.nature.com/articles/d41573-019-00014-x>



# Eroom's law

...Moore's law in reverse: halving the output every 9 years



From Scannell, *Nature Reviews Drug Discovery* (2012)

<https://www.nature.com/articles/nrd3681>

# Pharmaceutical R&D in the 21st Century: professional gambling or industrialized science?

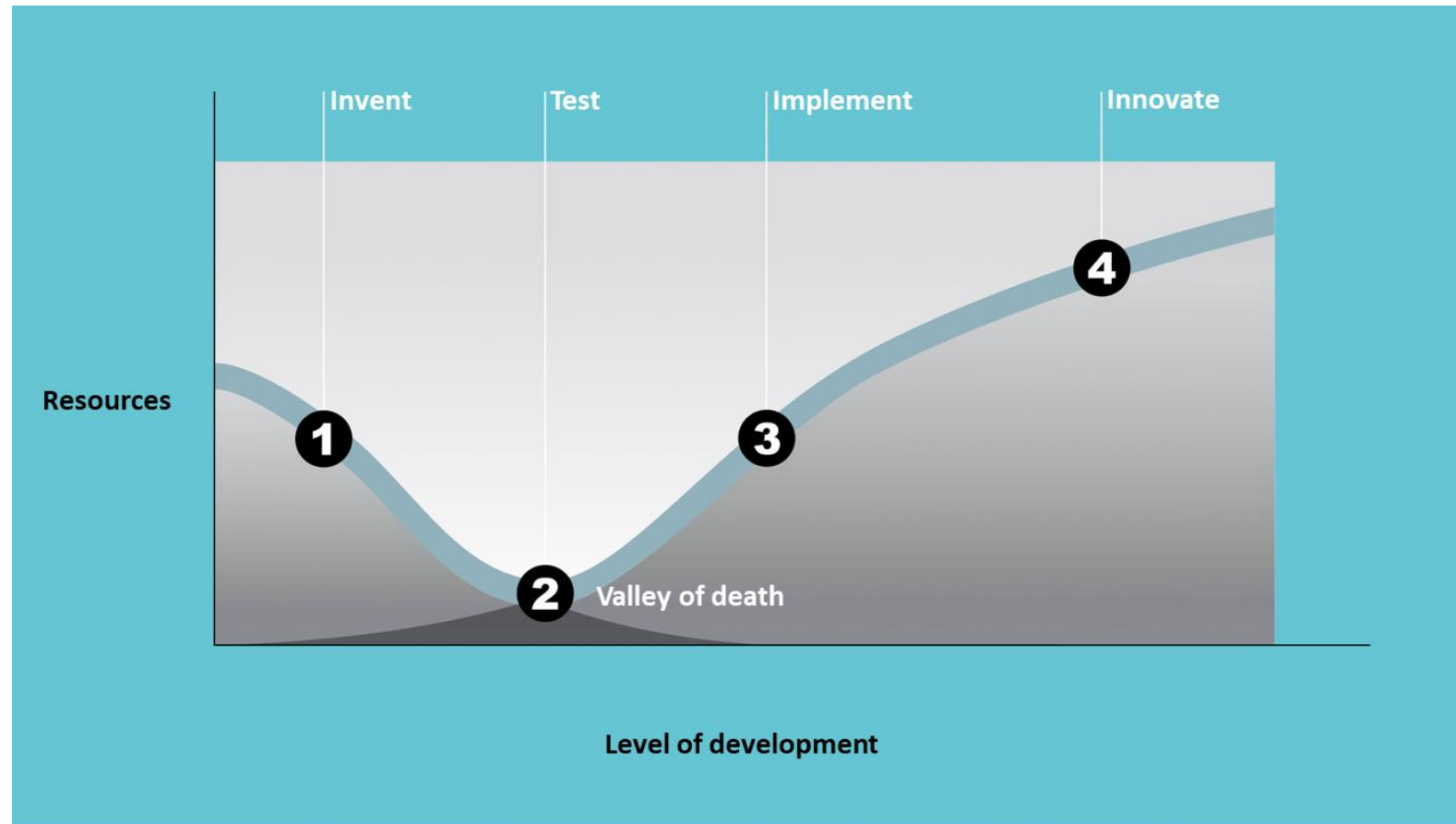


# Valley of Death

From Research inventions to Clinical Benefit



From Butler, Nature 2008



Foundation Meeting DayOne, Basel, 2016

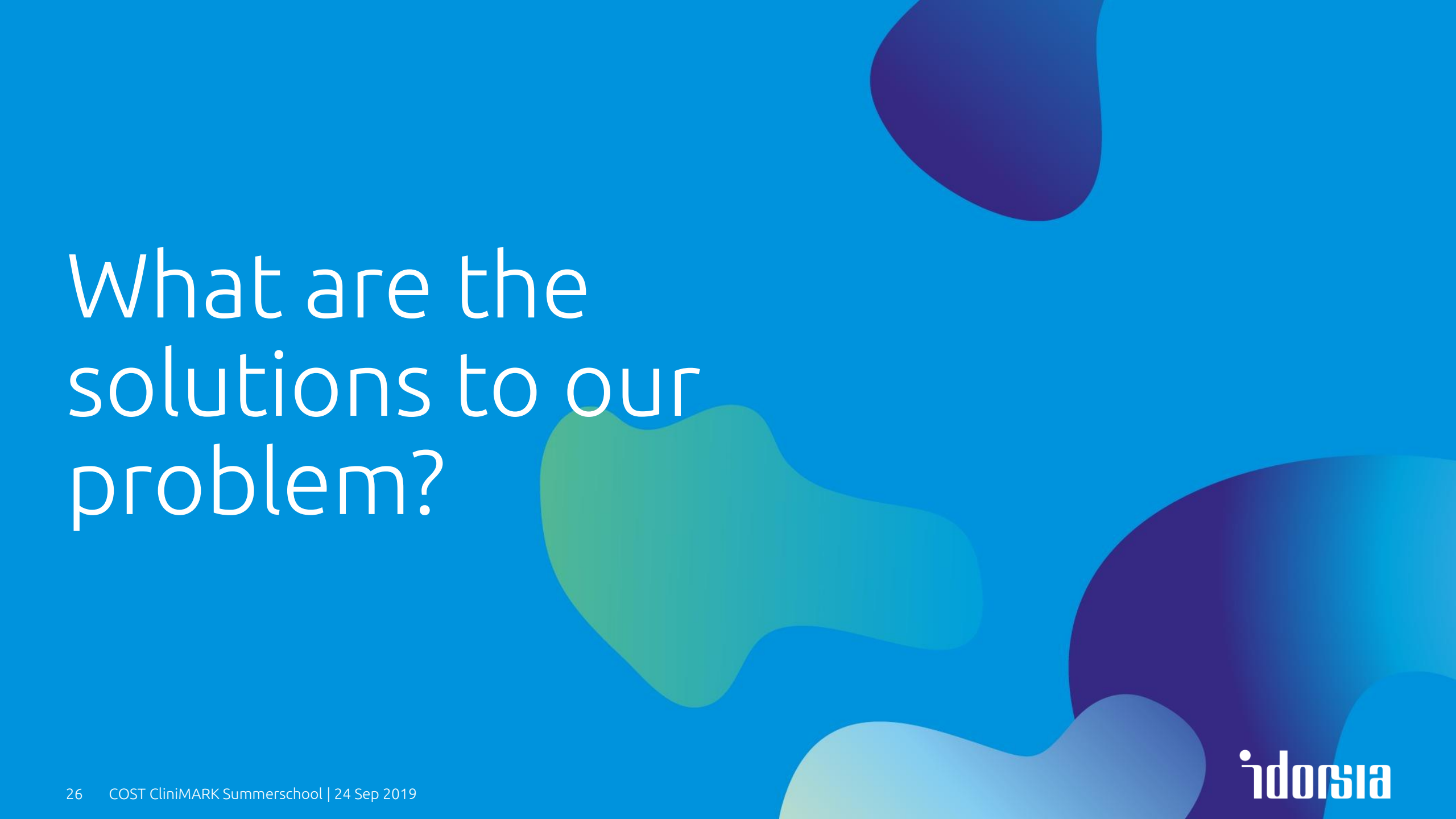


"I go home today. They cured me using this new miracle drug. I'm afraid it'll be years before it's approved for humans."



# But...also medical practice can be inaccurate and imprecise

- Traditionally clinical diagnosis is based on symptomology
- Clinical endpoints are often not objective (think about pain)
- Outside of Oncology most diseases (except Mendelian disorders with a clear genetic cause) do not use molecular information for diagnosis or classification
- Most of the current medications are not curative, they treat symptoms, not the cause
- Dosing of drugs are mostly based on an average response and an average safety level
- Adjusted dosing is mostly empirical



What are the  
solutions to our  
problem?

# Wanted: new toolkit

“A new product development toolkit - containing powerful new scientific and technology methods such as **animal or computer-based predictive models, biomarkers for safety and effectiveness and new clinical evaluation techniques** - is urgently needed to improve predictability and efficacy along the critical path from lab concept to commercial product.”

FDA

<https://www.fda.gov/science-research/critical-path-initiative/critical-path-opportunities-reports>



FDA report, 2004



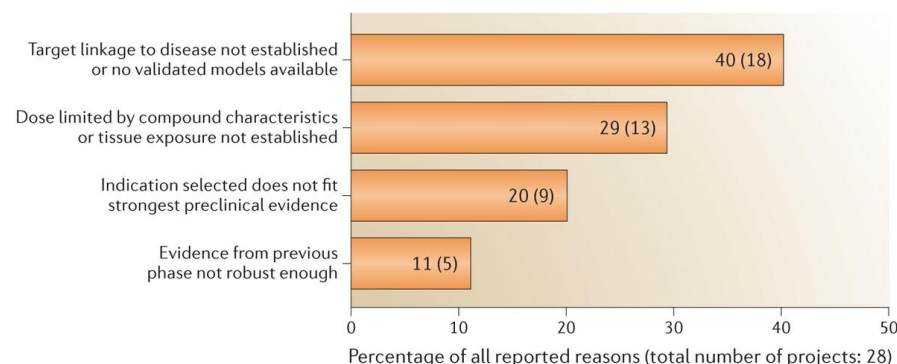
Janet Woodcock, CDER FDA

**idorsia**

# AstraZeneca's painful soul searching of project failures

The answer to it was implementing the 5R's

## a Reasons for lack of clinical efficacy



## Right target

- Strong link between target and disease
- Differentiated efficacy
- Available and predictive biomarkers

## Right tissue

- Adequate bioavailability and tissue exposure
- Definition of PD biomarkers
- Clear understanding of preclinical and clinical PK/PD
- Understanding of drug-drug interactions

## Right safety

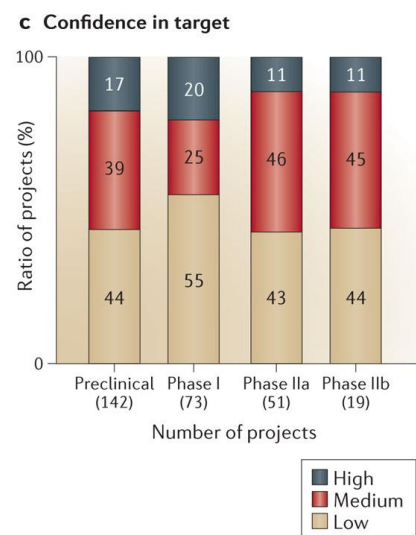
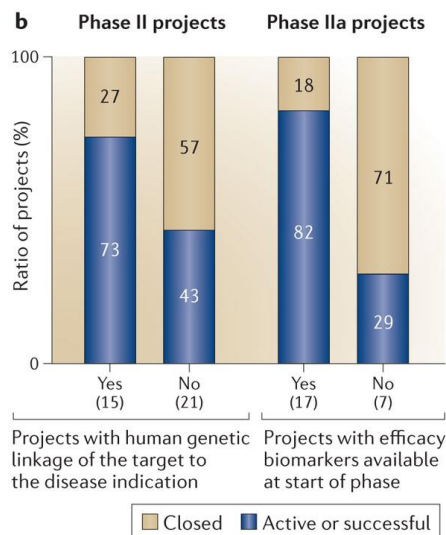
- Differentiated and clear safety margins
- Understanding of secondary pharmacology risk
- Understanding of reactive metabolites, genotoxicity, drug-drug interactions
- Understanding of target liability

## Right patients

- Identification of the most responsive patient population
- Definition of risk-benefit for given population

## Right commercial potential

- Differentiated value proposition versus future standard of care
- Focus on market access, payer and provider
- Personalized health-care strategy, including diagnostic and biomarkers



# Pfizer's similar answer after a similar exercise

Pfizer's three pillars of survival



- **Exposure** at the target site of action over a desired period of time
- **Binding** to the pharmacological target as expected for its mode of action
- **Expression** of pharmacological activity commensurate with the demonstrated target exposure and target binding

From Morgan et al., Drug Discovery Today 2012

<https://www.sciencedirect.com/science/article/pii/S1359644611004776>



Interim Question:  
Do you know one of  
the bigger problems  
causing drugs not to  
work (or cause SAEs)?



# Compliance!!!

- Compliance has always been a big problem for oral administered drugs like antibiotics, statins, anti inflammatory drugs, anti depressants, anti psychotics e.o.

# Systematic Biomarker Approaches

Problem statement: what question are we trying to solve?



# Current definitions and semantic standards from traditional biomarkers

## Biomarker Definition

- “A characteristic that is objectively measured and evaluated as an indicator of normal biological processes, pathogenic processes, or pharmacologic responses to a therapeutic intervention”

## Procedural Definitions

- Validation: technical, for the device and/or assay
- Qualification: the actual clinical utility

## Biomarker types

- Susceptibility/risk
- Diagnostic
- Monitoring
- Prognostic
- Predictive
- Pharmacodynamic/response
- Safety.

## Biomarker use

- Fit for Purpose
- Context of Use (COU)



Validation Parameters	Definitive Quantitative Assay	Relative Quantitative Assay	Quasi Quantitative Assay	Qualitative
Accuracy	Yes	Yes*	n.a.	n.a.
Precision	Yes	Yes	Yes	n.a.
Selectivity	Yes	Yes	Yes	Yes
Specificity	Yes	Yes	Yes	Yes
LLOD	Yes	Yes	Yes*	n.a.
Working range	Yes	Yes	Yes*	n.a.
Calibrators curve	Yes	Yes	n.a.	n.a.
Parallelism	Yes	Yes	n.a.	n.a.
Robustness	Yes	Yes	Yes	Yes
Stability	Yes	Yes	Yes	Yes
Examples of assay**	LC-MS (small molecule)	RIA, EIA, LC-MS, HPLC	EIA, FACS, qPCR, WB	IHC, WB

\* estimation

\*\* list not exhaustive

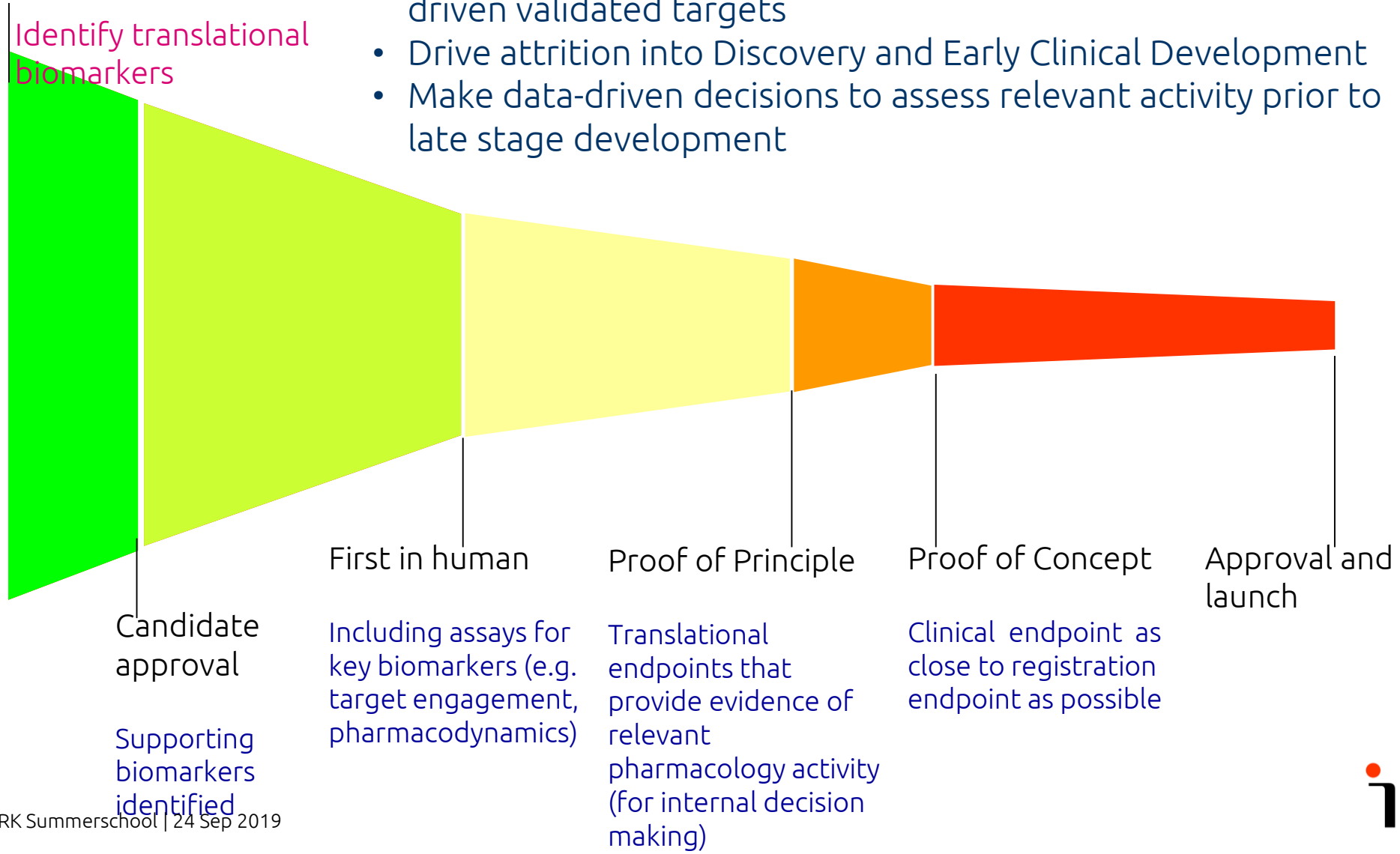
<https://www.ncbi.nlm.nih.gov/books/NBK326791/>

# Key decision points in pharmaceutical R&D

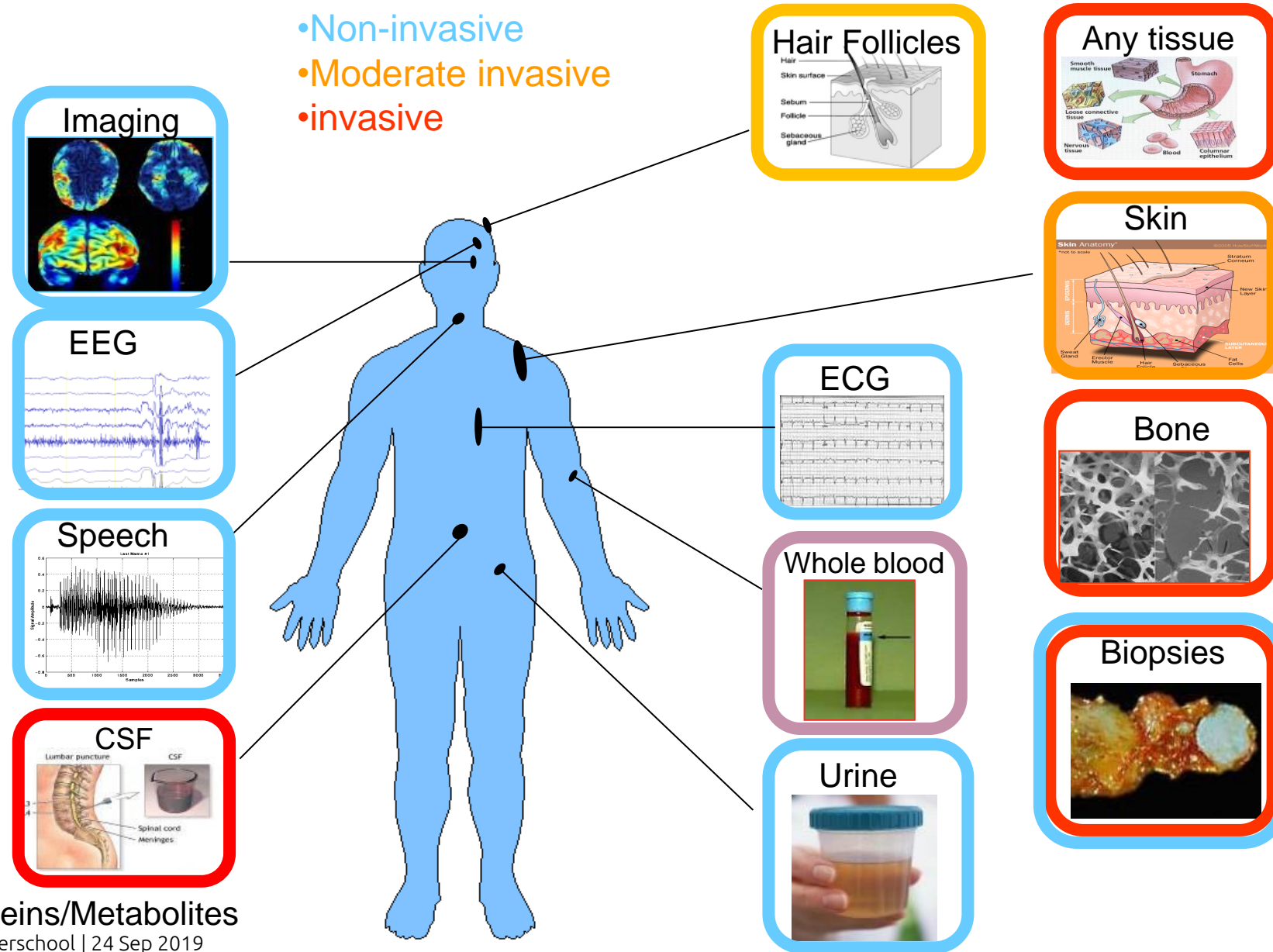
Lead op

Identify translational biomarkers

- Tools to enable earliest possible assessment hypothesis-driven validated targets
- Drive attrition into Discovery and Early Clinical Development
- Make data-driven decisions to assess relevant activity prior to late stage development



# Biomarker considerations: the reality of standard of care



Proteins/Metabolites



# Fit-for-purpose principle

Biomarkers have different purposes in the R&D process and require different levels of control

	Exploratory biomarker	Decision-making biomarker	Regulatory decision-making biomarker
Category	A	B	C
Validation	Level I	Level II	Regulatory guidance
Importance	Low / middle	High	Very high
Scope	Actelion	Actelion	Regulation Authorities
Examples	PD biomarker, stratification	PD biomarker, stratification, valid biomarker (primary or secondary endpoints)	Surrogate endpoint

# Case Example 1

Pharmacodynamic Response  
Biomarker

# Biomarker support for Lead Optimization to Clinical Proof of Concept

Editorial

Special Focus Issue: Bioanalysis of Biomarkers: Part 2

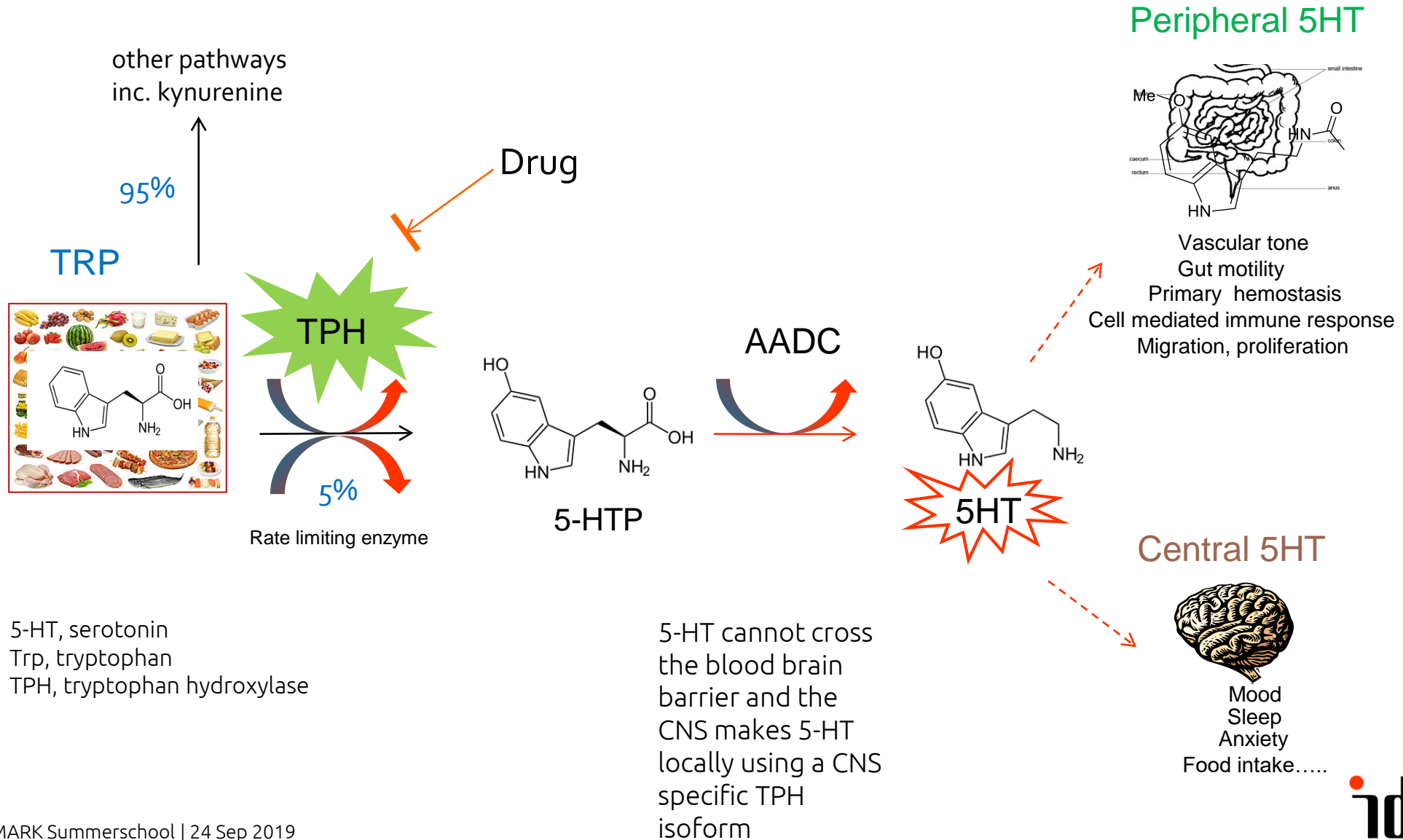
*For reprint orders, please contact: [reprints@futuremedicine.com](mailto:reprints@futuremedicine.com)*

Bioanalysis

## Biomarker measurements: how far have we come and where are we heading?

“...for successful translation of biomarkers into the clinic, it is imperative that biomarker researchers do not approach their work either with only biological or analytical aspects in mind...”

# Serotonin biosynthesis and homeostasis



# Peripheral Serotonin synthesis as a drug target

- Carcinoid syndrome
  - Carcinoid syndrome is caused by secretion of large quantities of 5-HT and other hormones from neuroendocrine tumors.
  - Activation of 5-HT<sub>2b</sub> results in mesenteric and cardiac fibrosis leading to valvulopathy.
  - The TPH1 inhibitor telotristat ethyl was FDA approved for treatment of carcinoid syndrome in 2017 (Marketed by Lexicon Pharmaceuticals Ltd as **Xermelo**).
- Pre-clinical and translational studies indicate therapeutic potential of peripheral 5-HT synthesis inhibitors in diseases including
  - Lung fibrosis
  - Pulmonary arterial hypertension
  - Ulcerative colitis
  - Obesity

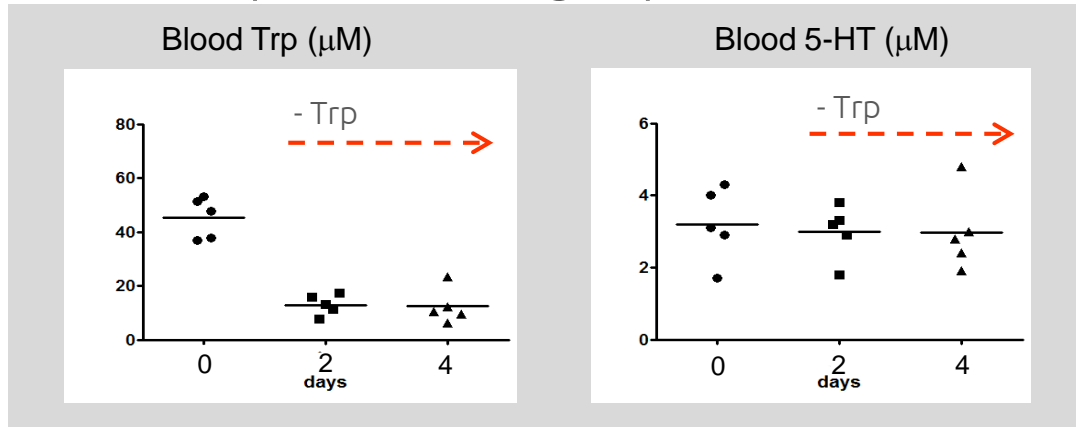


3 years of unsuccessful chemistry to find promising leads



# Blood 5-HT has a long half-life

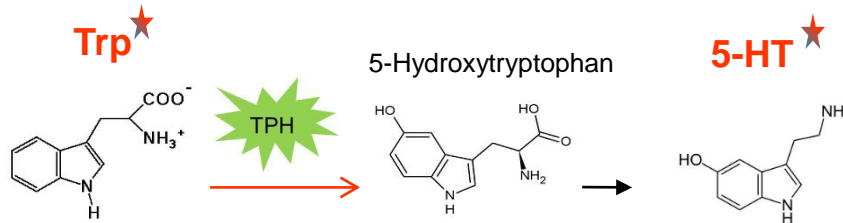
- Blood 5-HT rat  $t_{1/2} = 3$  d, dog  $t_{1/2} = 6$  d



Need for a tracer

Measure 5-HT synthesis/  
*In vivo* TPH1 activity

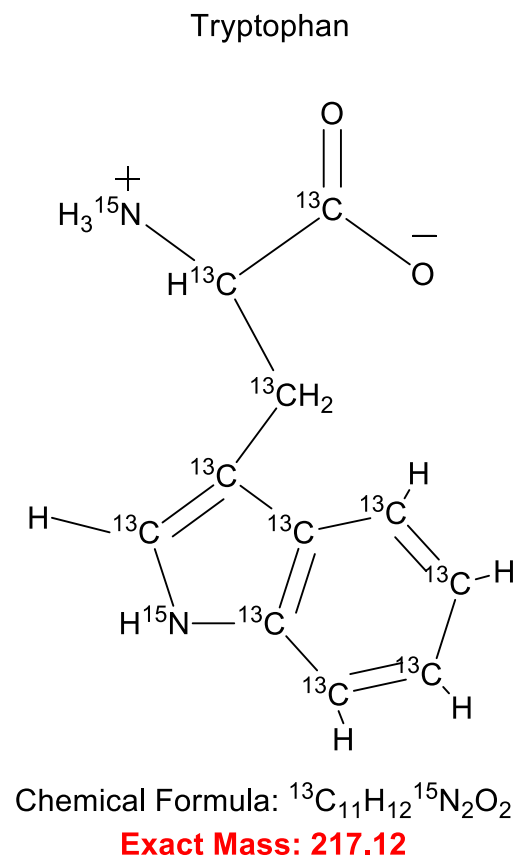
Trp★



5-HT, serotonin  
Trp, tryptophan  
TPH, tryptophan hydroxylase

# Tracer selection is important










- $^{13}\text{C}$ ,  $^{15}\text{N}$  labels, No deuteriums to avoid altered metabolism
- > 2 labels to avoid interference from naturally occurring +1 (10%) and +2 (1%) isotopes ( $^{13}\text{C}$ )
- Commercial availability/cost



$^{13}\text{C}_{11}, ^{15}\text{N}_2$ -TRP was the only  
One that fit all the criteria at the time

Cost per rat in an experiment ~20CHF  
Estimated cost per human ~ 2000CHF

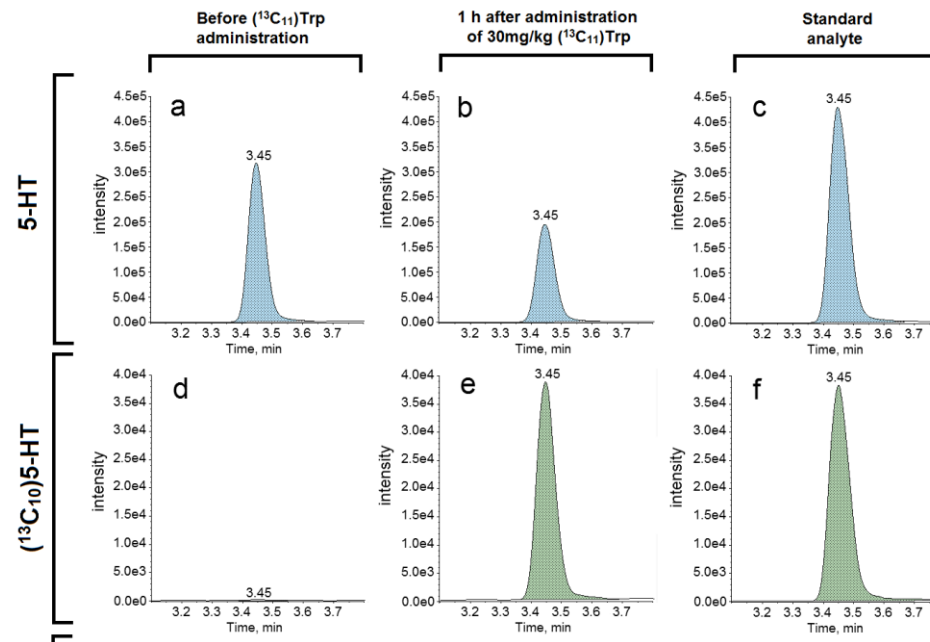
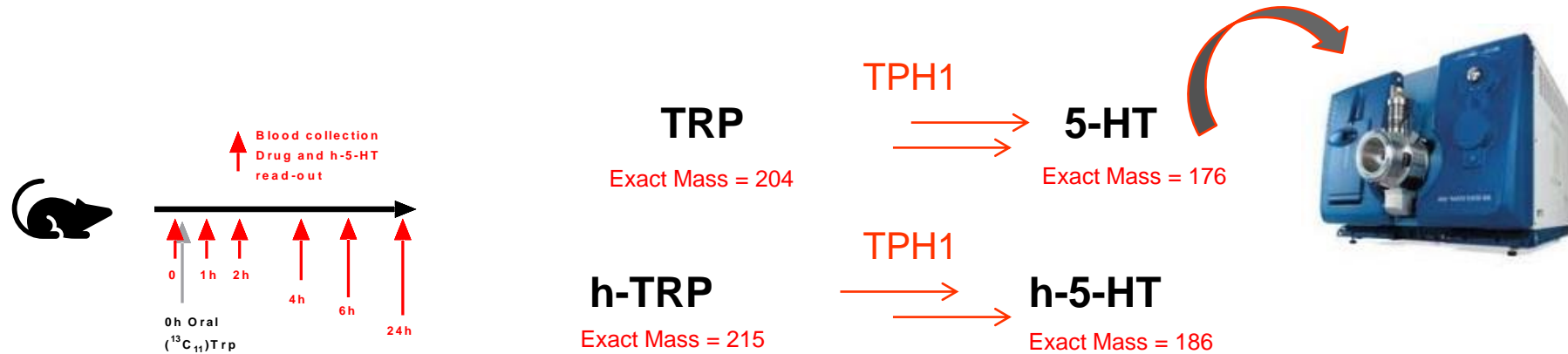
# Stable isotope tracer approach

Tracer	Unaltered metabolism	Not radioactive	Best data quality
Fluoro-TRP			
$^{14}\text{C}$ -Me-TRP			
Heavy-TRP			

Heavy-Trp =  $^{13}\text{C}_{11}$ -Trp

MeTrp PET tracer is used clinically to measure 5-HT synthesis, but results are confounded by Trp uptake

# Heavy serotonin measurement

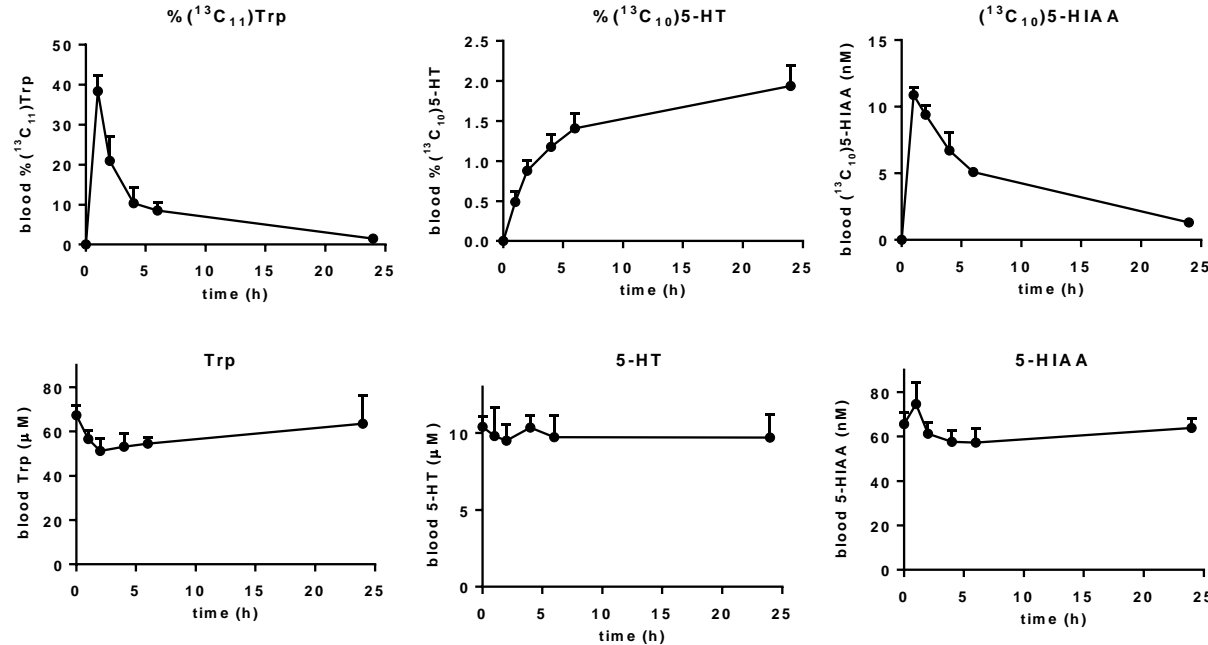
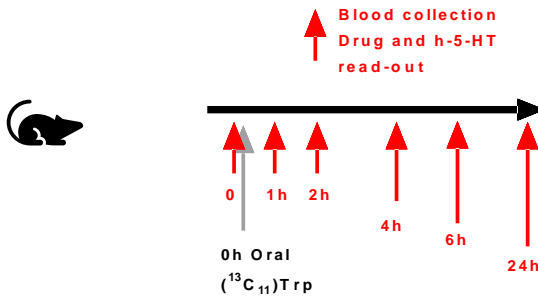


5-HT, serotonin  
Trp, tryptophan  
TPH, tryptophan hydroxylase

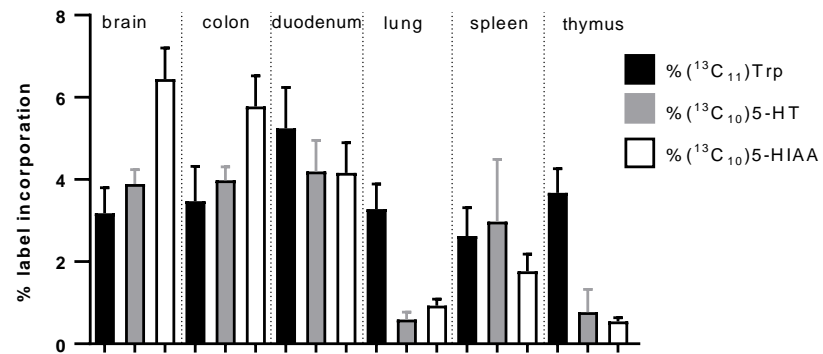
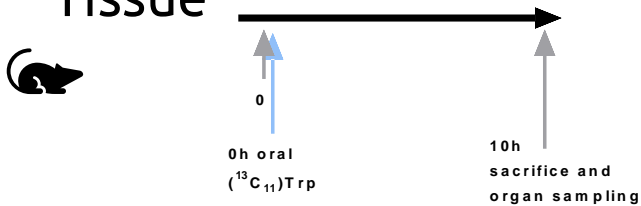


# h-5-HT synthesis in healthy rats

## Blood



## Tissue

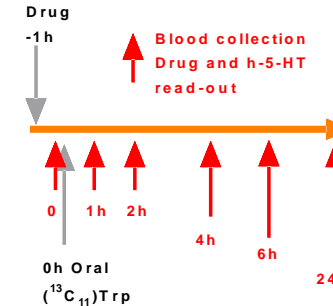


5-HT, serotonin  
Trp, tryptophan  
5-HIAA, 5-hydroxyindole acetic acid

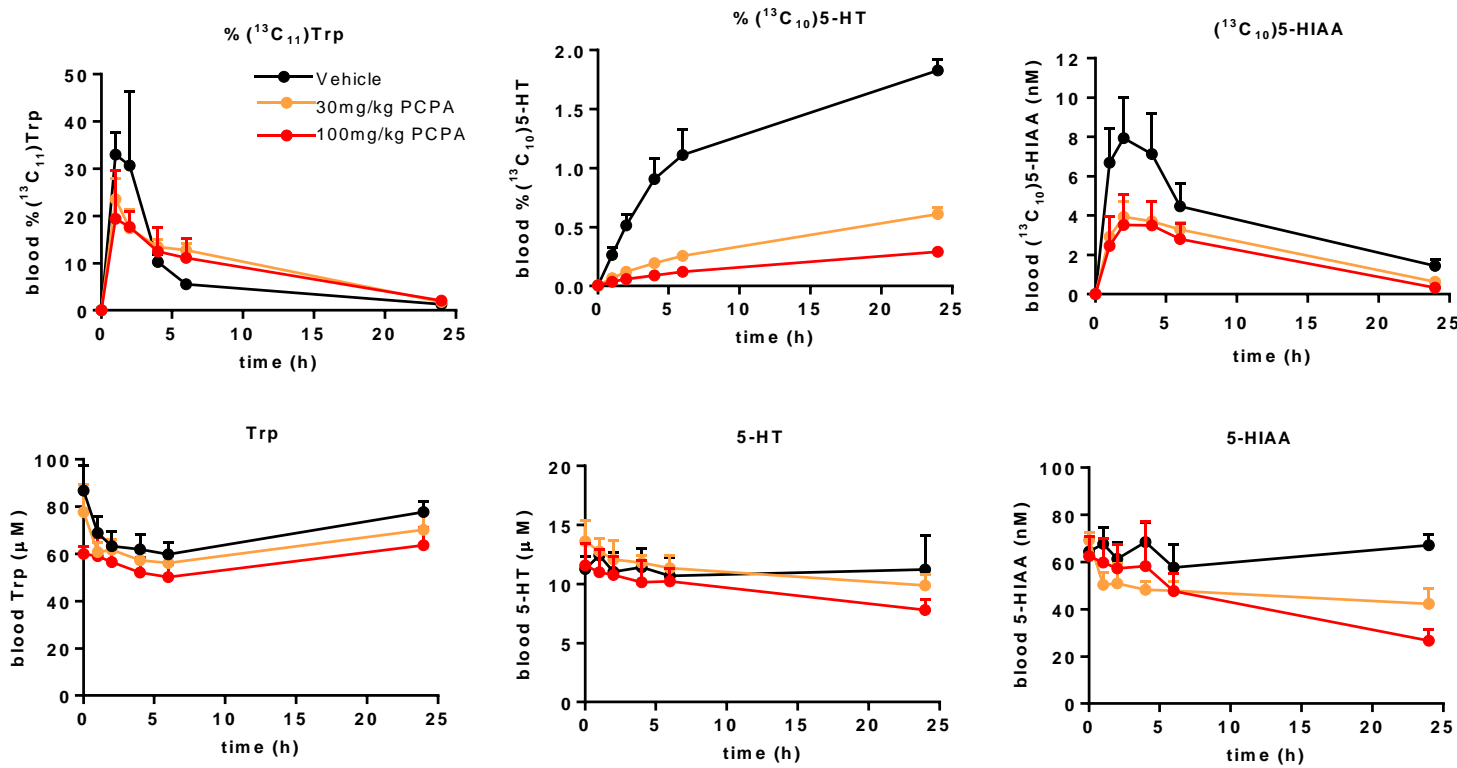
Welford, Vercauteren (2016) SciRep

# TPH inhibitor pharmacodynamics with oral h-Trp

- PCPA is a non-specific brain penetrating TPH inhibitor
- h-5-HT is the most sensitive marker of TPH inhibition in rats



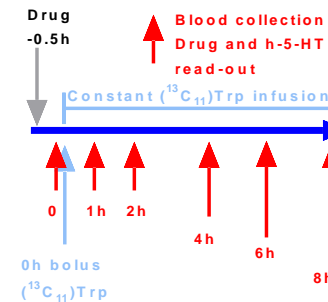
5-HT, serotonin  
Trp, tryptophan  
5-HIAA, 5-hydroxyindole acetic acid  
PCPA, para-chlorophenylalanine



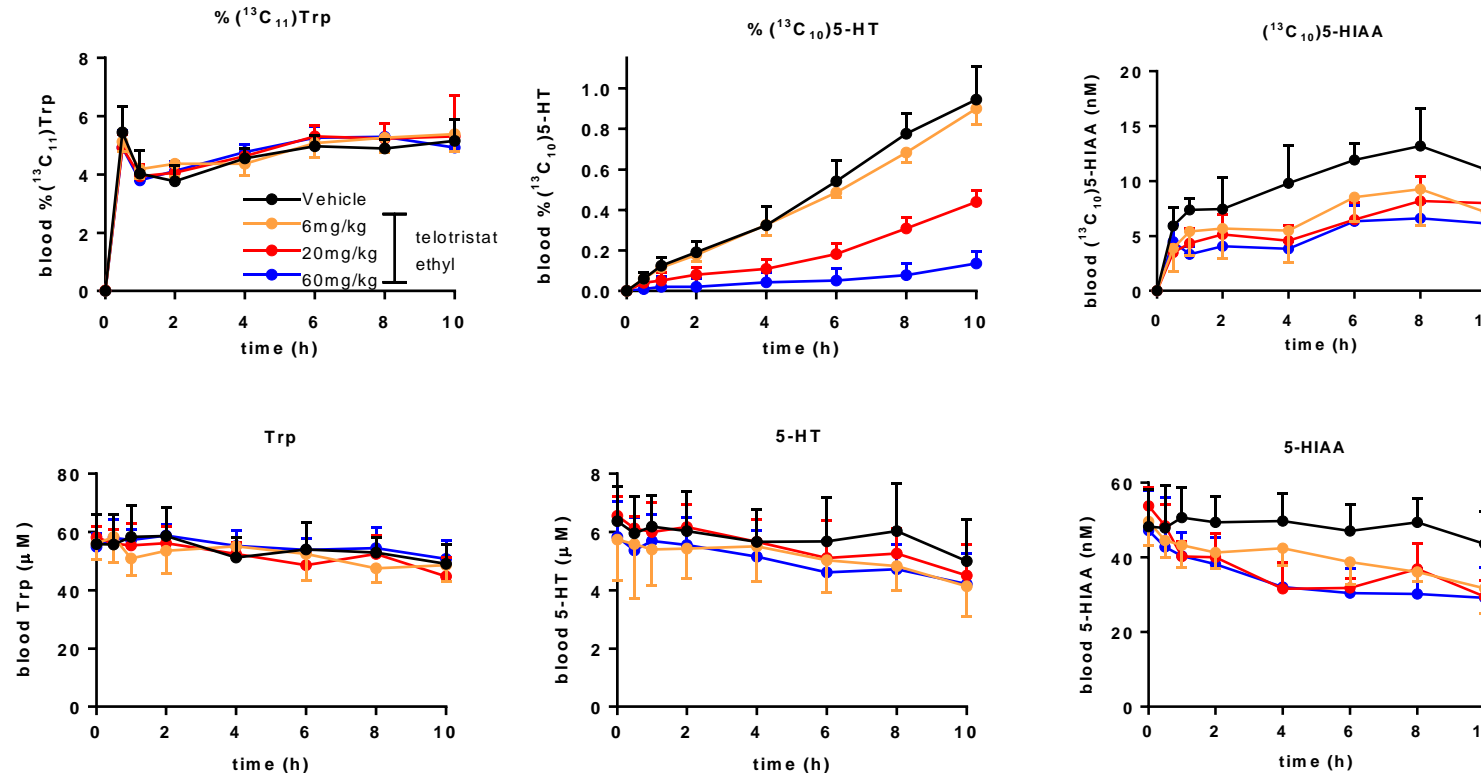
n = 4  
AV±SD

# TPH inhibitor pharmacodynamics with infused h-Trp

- Telotristat ethyl dose dependently reduces h-5-HT production in rats
- h-Trp infusion allows estimation of duration of action

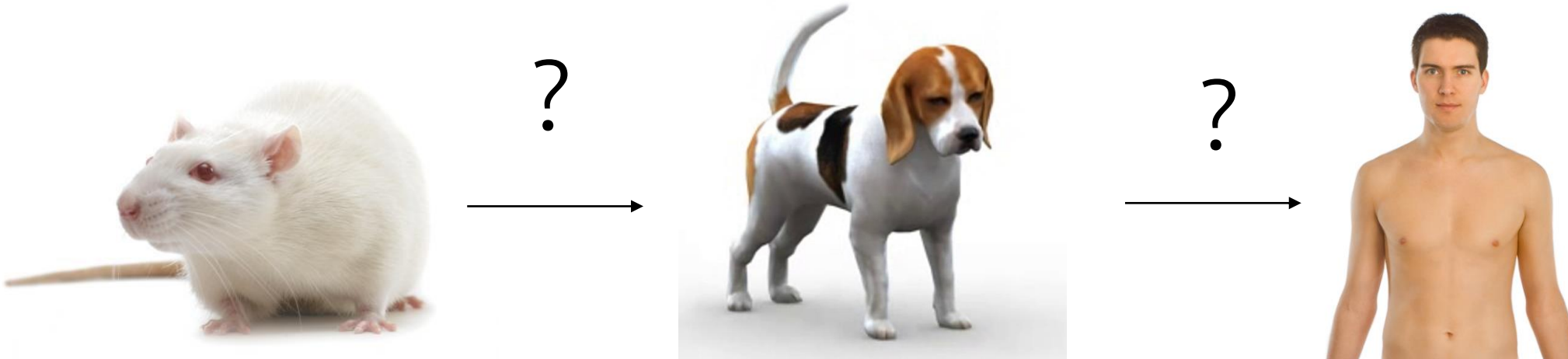


5-HT, serotonin  
Trp, tryptophan  
5-HIAA, 5-hydroxyindole acetic acid



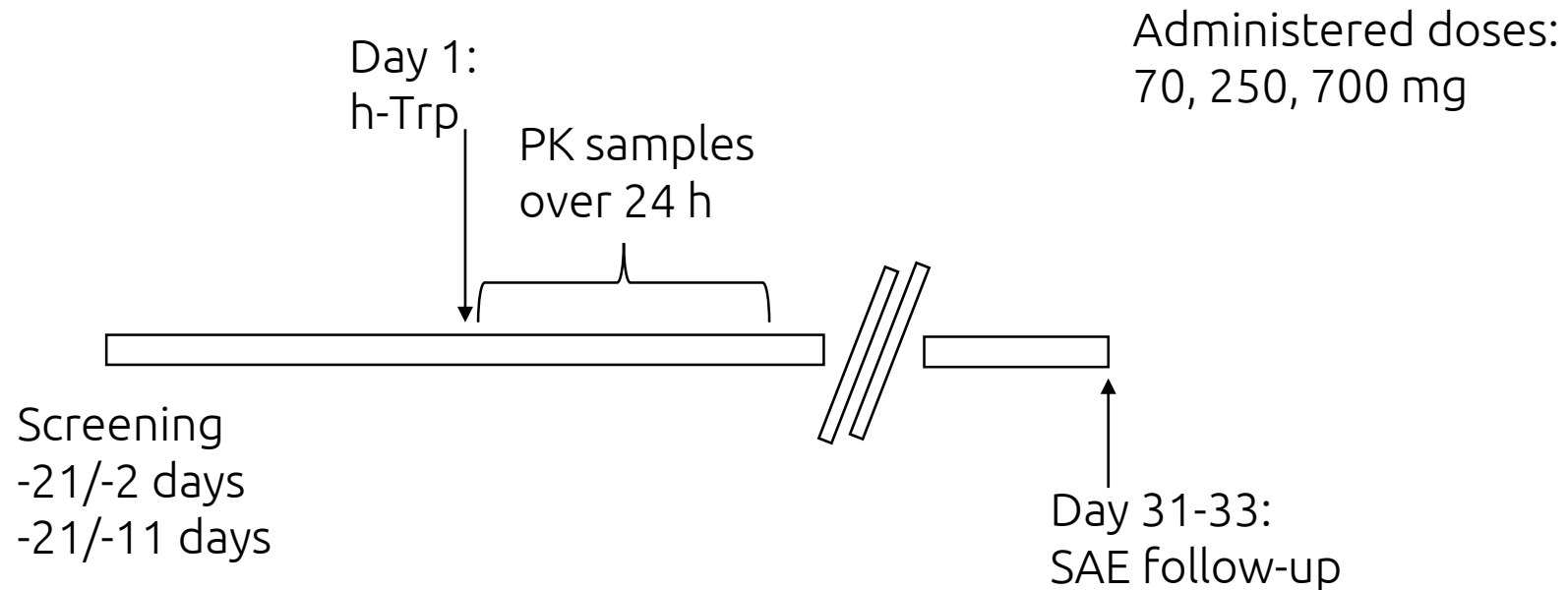
n = 6  
AV $\pm$ SD

# Translating the TPH1 inhibition effects

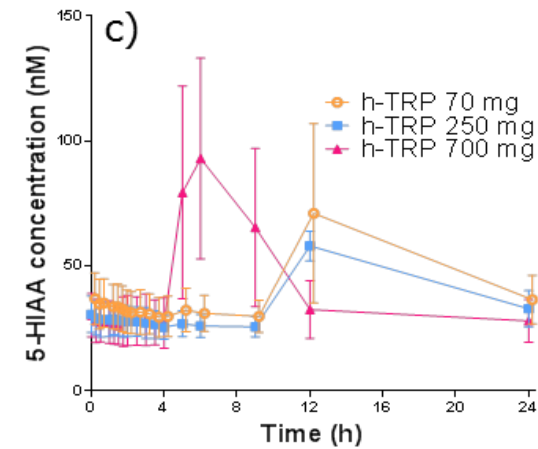
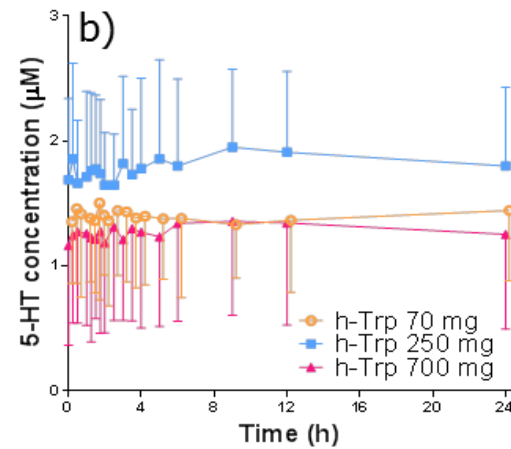
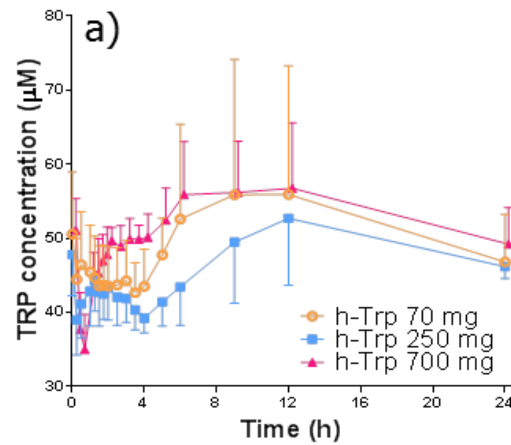
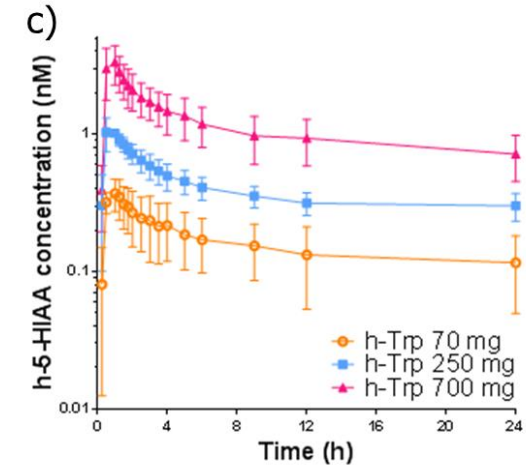
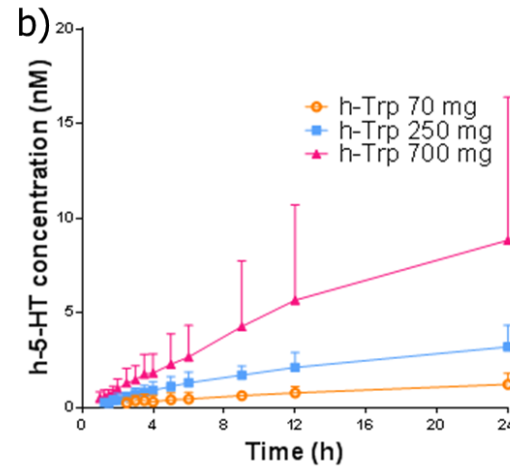
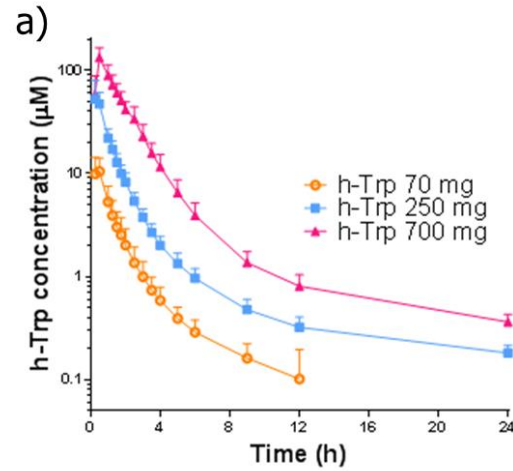


# Heavy tryptophan study in man: Design

- 6 healthy subjects per group: 3 males and 3 females
- Subjects were overnight fasted up to 4 h post dose (lunch)
- The 70 and 250 mg cohorts were administered on the same day, the 700 mg cohort several months later



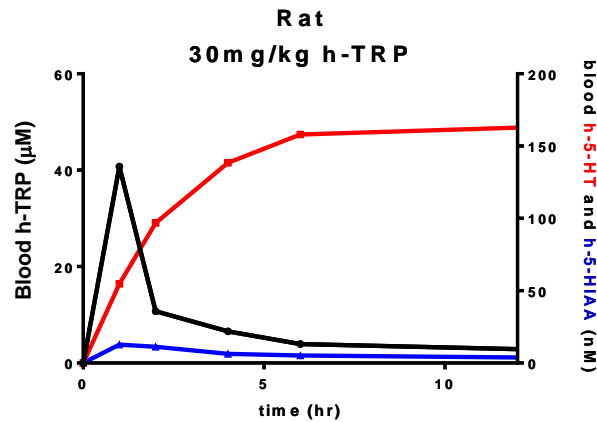
# h-5-HT synthesis in healthy subjects



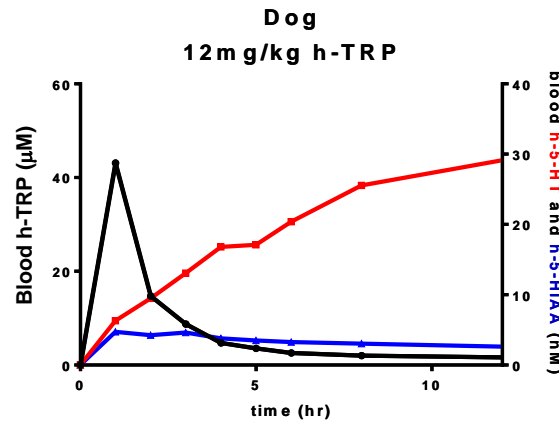


# h-5-HT production changes dramatically with species

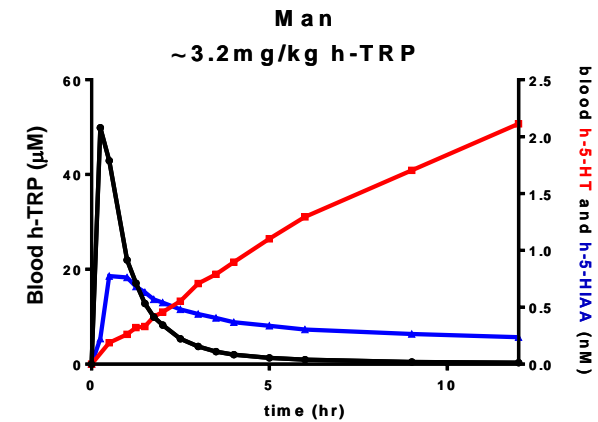
- h-5-HT changes more dramatically than 5-HT between species
- In man synthesis is linear despite rapidly changing h-TRP, i.e. h-5-HT production in a single compartment will not explain the data



Weight 0.3 kg  
Trp 73  $\mu\text{M}$   
5-HT 13  $\mu\text{M}$



15 kg  
54  $\mu\text{M}$   
5.0  $\mu\text{M}$



80 kg  
48  $\mu\text{M}$   
1.7  $\mu\text{M}$

5-HT, serotonin  
Trp, tryptophan  
5-HIAA, 5-hydroxyindole acetic acid

# Acknowledgements

**R Welford**

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E Fournier

**O Nayler**

P Sieber

S Roux

M Probst

J Segrestaa

C Kohl

M Machacek (Lyo-X)

**M Gehin-Beurne**

& team

J Dingemanse

R Karge

# SCIENTIFIC REPORTS

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## Serotonin biosynthesis as a predictive marker of serotonin pharmacodynamics and disease-induced dysregulation

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The biogenic amine serotonin (5-HT) is a multi-faceted hormone that is synthesized from dietary tryptophan with the rate limiting step being catalyzed by the enzyme tryptophan hydroxylase (TPH). The therapeutic potential of peripheral 5-HT synthesis inhibitors has been demonstrated in a number of clinical and pre-clinical studies in diseases including carcinoid syndrome, lung fibrosis, ulcerative colitis and obesity. Due to the long half-life of 5-HT in blood and lung, changes in steady-state levels are slow to manifest themselves. Here, the administration of stable isotope labeled tryptophan (heavy "h-Trp") and resultant *in vivo* conversion to h-5-HT is used to monitor 5-HT synthesis in rats. Dose responses for the blockade of h-5-HT appearance in blood with the TPH inhibitors L-para-chlorophenylalanine (30 and 100 mg/kg) and telotristat etiprate (6, 20 and 60 mg/kg), demonstrated that the method enables robust quantification of pharmacodynamic effects on a short time-scale, opening the possibility for rapid screening of TPH1 inhibitors *in vivo*. In the bleomycin-induced lung fibrosis rat model, the mechanism of lung 5-HT increase was investigated using a combination of synthesis and steady state 5-HT measurement. Elevated 5-HT synthesis measured in the injured lungs was an early predictor of disease induced increases in total 5-HT.

The biogenic amine serotonin (5-HT) is a biochemical messenger and regulator that signals through 13 receptors which are distributed throughout the nervous system and peripheral organs<sup>1–3</sup>. 5-HT was initially described as a vasoconstrictor and in the periphery it also plays a role in vasodilation, hemostasis, intestinal motility<sup>4</sup>, wound healing<sup>5</sup> and inflammatory responses<sup>6</sup>. 5-HT is synthesized in two steps from dietary L-tryptophan (Trp) and accounts for 1–3% of Trp metabolism<sup>7</sup>. The first and rate limiting step of 5-HT production is hydroxylation catalyzed by the non-heme iron pterin-dependent oxygenase Tryptophan hydroxylase (TPH) (Fig. 1a). There are two TPH isoforms, TPH2 is mainly expressed in the central nervous system and enteric neurons, while TPH1 is expressed in the periphery and pineal gland<sup>8</sup>. The second step in 5-HT synthesis is rapid decarboxylation of 5-hydroxytryptophan (5-HTP) by the enzyme aromatic amino acid decarboxylase (DDC). Peripheral 5-HT is said to be largely synthesized by TPH1 expressed in the enterochromaffin cells lining the gut, where it is initially stored in secretory granules via the action of vesicular monoamine transporters<sup>1</sup>. 5-HT can then be released into the extracellular space and either signal through receptors or be taken up in to other cell types that express the 5-HT re-uptake transporter SERT such as epithelial cells, smooth muscle cells and platelets. Platelet 5-HT has a half-life of at least 3 days and platelets are able to buffer plasma 5-HT complicating its measurement<sup>4,9,10</sup>. 5-HT is further metabolized intracellularly to 5-hydroxyindole acetic acid (5-HIAA) by a combination of the mitochondrial enzyme monoamine oxidase A (MAO-A) and an aldehyde dehydrogenase (AD). 5-HIAA is excreted in the urine and can be monitored as a surrogate of 5-HT level, with 24-hour urinary 5-HIAA used as a diagnostic for carcinoid syndrome<sup>11</sup>.

Increasing evidence implicates TPH1 in a number of peripheral diseases including lung fibrosis<sup>12,13</sup>, ulcerative colitis<sup>14</sup>, pulmonary hypertension<sup>15</sup>, osteoporosis<sup>16</sup>, irritable bowel syndrome<sup>17</sup> and obesity<sup>18,19</sup>. Recently, the

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## ARTICLE

## Assessment of Peripheral Serotonin Synthesis Using Stable Isotope-Labeled Tryptophan

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Serotonin (5-HT) is synthesized from dietary tryptophan (Trp) and plays an important role in numerous diseases of the central nervous system and periphery. Stable isotope tracers enable safe monitoring of metabolic rates. Here we demonstrate measurement of peripheral 5-HT synthesis in healthy subjects by monitoring the produced [<sup>13</sup>C<sub>10</sub>]-5-HT (h-5-HT) in EDTA-whole blood from three doses of orally administered [<sup>13</sup>C<sub>10</sub>]-Trp (h-Trp) tracer. h-Trp was rapidly absorbed and distributed in a multiphasic manner, followed by a slower terminal elimination phase. The h-5-HT synthesis rate was dependent on h-Trp dose, appeared linear up to 12 hours postdose, and could be reliably assessed for the two highest doses. The human data was compared to similar studies in rats and dogs, finding larger interspecies differences in the h-5-HT synthesis rate than in 5-HT levels. In future studies, the h-5-HT synthesis rate can be used to assess disease-dysregulated 5-HT synthesis or quantify the pharmacodynamics of 5-HT synthesis inhibitors.

## Study Highlights

## WHAT IS THE CURRENT KNOWLEDGE ON THE TOPIC?

Inconvenient 24-hour urine collection and measurement of the 5-HT metabolite 5-HIAA is frequently used as a diagnostic for carcinoid syndrome and has also been used as a biomarker of TPH inhibition.

## WHAT QUESTION DID THIS STUDY ADDRESS?

Development of methodology enabling a specific and rapid assessment of 5-HT synthesis rate in humans in a short study. Comparison of h-5-HT synthesis rate between species.

## WHAT DOES THIS STUDY ADD TO OUR KNOWLEDGE?

The pharmacokinetics and metabolism of Trp/5-HT were investigated and compared to those in dog and rat.

## HOW MIGHT THIS CHANGE CLINICAL PHARMACOLOGY OR TRANSLATIONAL SCIENCE?

This study provides a new tool to screen and diagnose patients that may benefit from TPH1 inhibition and/or to better quantify the pharmacodynamics of a TPH1 inhibitor.

Serotonin (5-HT) is a pleiotropic hormone whose diverse functions encompass activation of 15 different receptors with roles in both the central nervous system (CNS) and the periphery.<sup>1,2</sup>

5-HT is synthesized from dietary tryptophan (Trp) through a two-step enzymatic pathway.<sup>2</sup> Hydroxylation by the rate-limiting tryptophan hydroxylase (TPH) is followed by rapid decarboxylation of the 5-hydroxytryptophan intermediate giving 5-HT. 5-HT is then further oxidized to 5-hydroxyindoleacetic acid (5-HIAA) by a monoamine oxidase and aldehyde dehydrogenase (Figure 1). TPH expression has been identified in a range of peripheral tissues including gut, aorta, stomach, and thymus. In the periphery, 5-HT is produced by TPH1.<sup>3</sup> In the CNS, a second isoform TPH2 is responsible for the hydroxylation step.

The majority of peripheral 5-HT is formed and stored in enterochromaffin cells in the gut, where it is initially packed into granules by vesicular monoamine transporters.<sup>2</sup> The packaged 5-HT can be released into the lamina propria, where it can interact with the enteric nervous system or be transported to the portal

circulation. More than 99% of circulating 5-HT is sequestered in platelets by the serotonin transporter SERT, protecting it from liver and lung metabolism.<sup>4,5</sup> The half-life of platelet 5-HT in healthy male subjects was found to be 5–6 days.<sup>6</sup> Platelet 5-HT is a vasoconstrictor and can modulate lung function, cardiac development, and immune responses.<sup>7</sup>

Dysregulation of 5-HT synthesis is observed in several severe conditions. Beneficial effects of TPH inhibitors and/or *Tph1* knockout have been observed in a number of preclinical models including colitis,<sup>8</sup> obesity,<sup>9</sup> lung fibrosis,<sup>10</sup> and pulmonary arterial hypertension.<sup>11</sup> Carcinoid syndrome is caused by the secretion of large quantities of 5-HT and other hormones from neuroendocrine tumors.<sup>12</sup> 5-HT activation of the 5-HT<sub>2A</sub> receptor is thought to be causative of the mesenteric and cardiac fibrosis leading to the valvulopathy<sup>13,14</sup> observed in many of the patients with heart valve disease and/or carcinoid syndrome. Telotristat ethyl recently became the first approved 5-HT synthesis inhibitor for treatment of carcinoid syndrome patients refractory to somatostatin analogs.<sup>15,16</sup>

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# Case example 2

## Diagnostic Biomarker

# Plasma lyso-sphingomyelin and glucosylsphingosine as biomarkers for Niemann-Pick disease type C

# NP-C is a sphingolipidosis

Crocker 1961 J. Neuro Chem

TABLE 4.—COMMON CLINICAL FORMS OF NIEMANN-PICK DISEASE

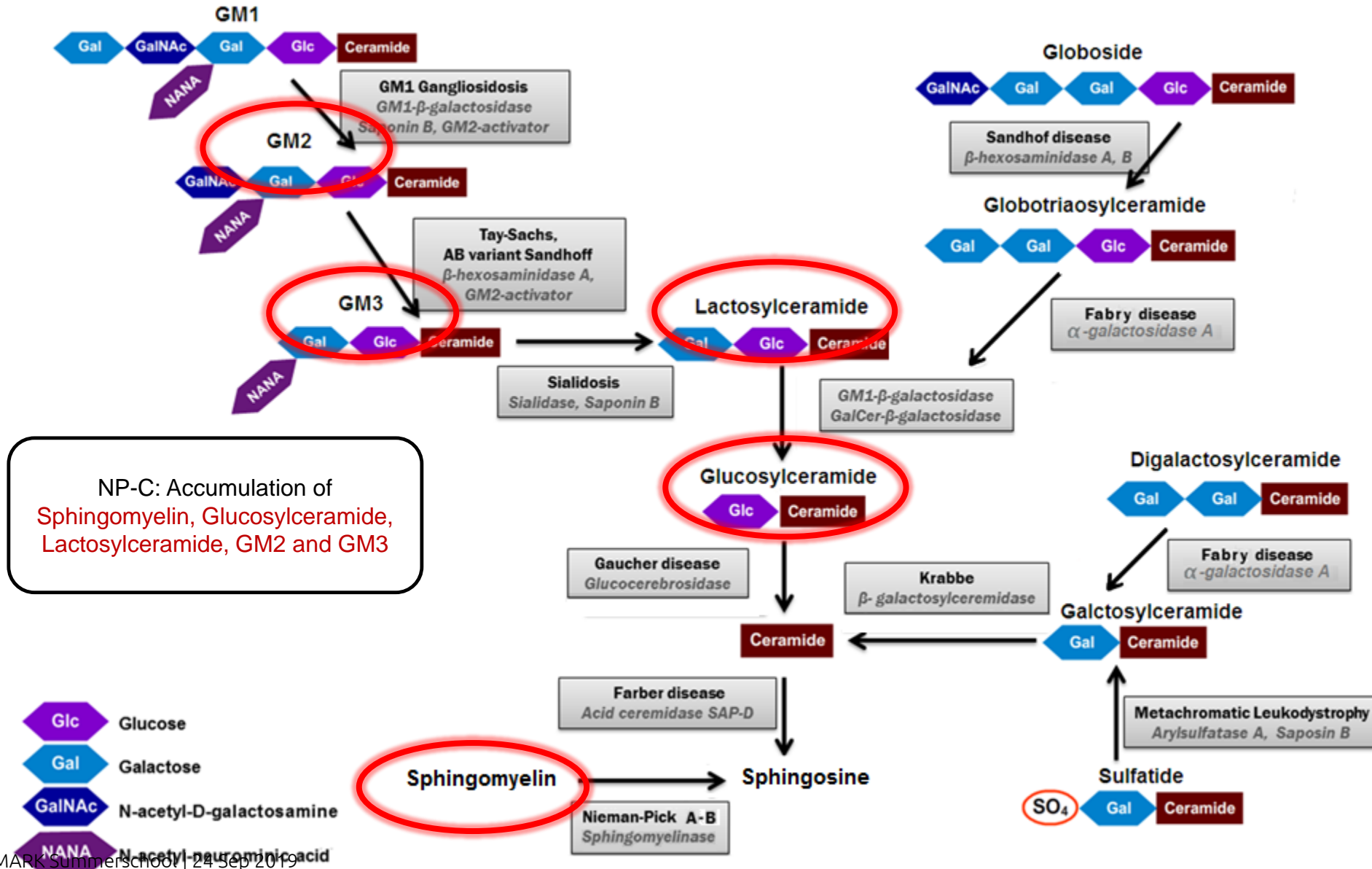
Enlargement of liver and spleen	Chest X-ray	Onset of CNS symptoms	Elevation of lipids Blood      Tissue	Age at death (Yr.)	Ethnic background
Group A. 'CLASSICAL' NIEMANN-PICK DISEASE (6 families)					
++++	++	Early infancy (often CRS)	++      +++++	1-2	Often Jewish
Group B. HEAVY VISCERAL INVOLVEMENT, NORMAL CNS (3 families)					
++++	++	None seen at 4-13 yrs.	++      +++++	—	Mixed
Group C. MODERATE COURSE (5 families)					
++	0	Late infancy	0      ++	3-5	Non-Jewish
Group D. NOVA SCOTIAN GROUP (4 families)					
Usually +++	0	Early to middle childhood	0      ++	12-20	Catholic

*“The clinical picture in Niemann-Pick disease, as the syndrome is conventionally defined, is variable. The table summarizes the cumulative experience in this hospital with patients who show a constitutional affliction by major visceral **sphingomyelin accumulation** in the usual type of “foam cell”.*”

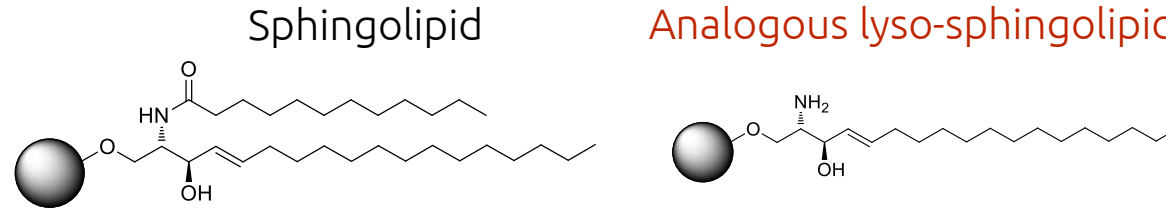


# Degradation of sphingolipids

Vanier (2010) Orphanet  
Figure adapted from  
©www.lysosomalstorageresearch.org



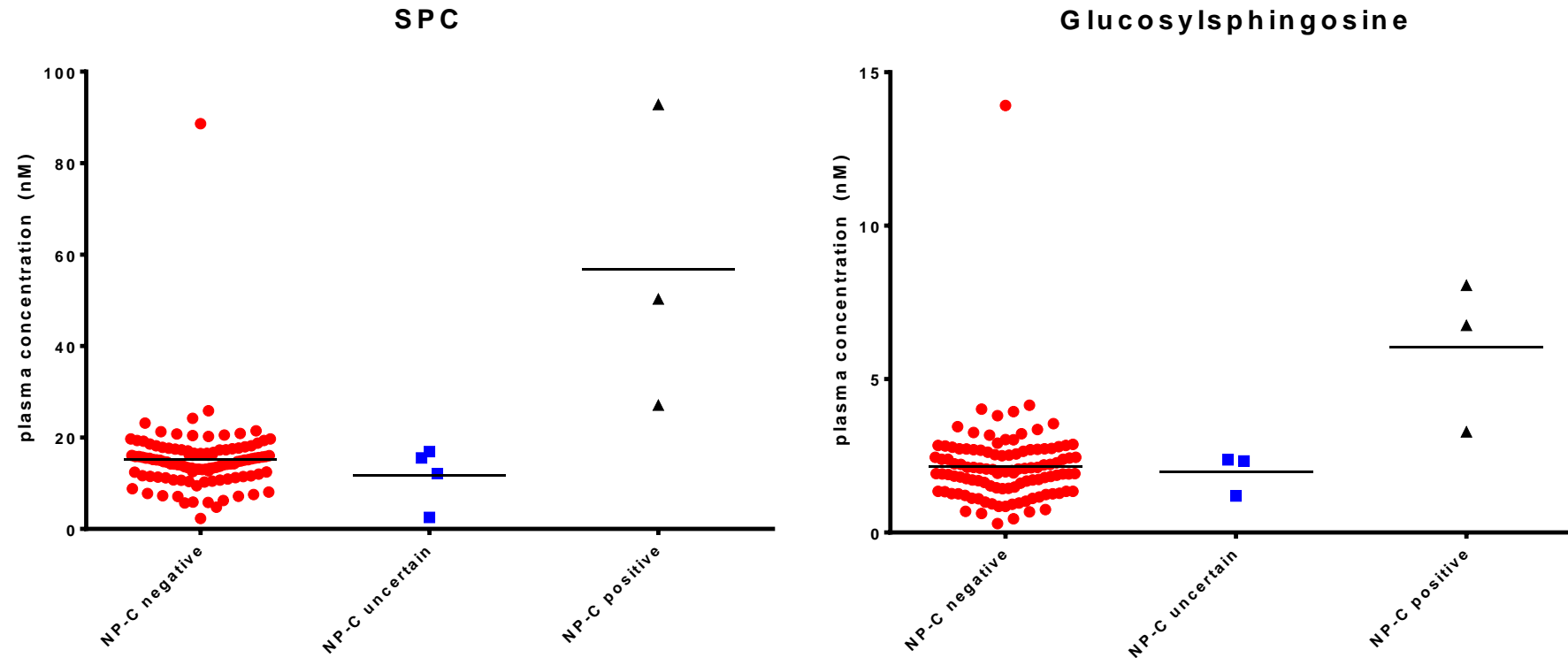
# Plasma lyso-sphingolipids in LSDs



Disease	Sphingolipid(s) accumulated in organs	Lyso-sphingolipid in plasma	References for lyso-form as a biomarker
<b>Fabry</b>	Gb3	lysoGb3 <b>&gt;10-fold</b>	Aerts et al. (2008) ,Krüger et al (2012), Togawa et al. (2011)
<b>Krabbe</b>	Galactosylceramide	Psychosine <b>10-fold</b>	Zhu et al. (2012), Svennerholm et al. (1980)
<b>Gaucher</b>	Glucosylceramide	Glucosylsphingosine <b>100-fold</b>	Dekker et al. (2011),
<b>Tay-Sachs/Sandhoff</b>	GM2	lysoGM2 <b>10-fold</b>	Kodama et al. (2011)
<b>NP-C</b>	Sphingomyelin Glucosylceramide Lactosylceramide GM2 - GM3	Lyso-sphingomyelin (SPC) <b>x-fold?</b> Glucosylsphingosine (GlcSph) <b>x-fold?</b> Lactosylsphingosine <b>x-fold?</b> lysoGM2 - lysoGM3 <b>x-fold?</b>	

Vanier (2010). Orphanet J. Rare Dis.  
Sandhoff (2013). Biochem. Soc. Trans.

# Feasibility study : ZOOM study



- ▶ ZOOM was a **multi-center** genetic screening study on the occurrence of NP-C in **adult** patients with neurological/psychiatric symptoms.
- ▶ Of the screened patients, 3 of 250 were identified as NP-C positive based on genetics and filipin staining

Bauer et al. Hum. Mol. Genet. (2013)

# Technical LC- MS/MS Assay validation

# technical assay validation

## Source documents

- EMA guideline on analytical method validation (*EMEA/CHMP/EWP/192217/2009*) (2011)
- FDA: Guidance for Industry Bioanalytical Method Validation (2001)
- Jennings et al. “Recommended principles for validating clinical molecular pathology tests” (2009) *Arch. Path Lab. Med.*
- Houghton, R., et al., *Generic approach to validation of small-molecule LC-MS/MS biomarker assays*. Bioanalysis, 2009. **1**(8): p. 1365-74.

# Validation plan: CAL and qc samples

Houghton *et al* *Bioanalysis*.2009

Analyte	QC2 plasma [nM]	QC3 plasma spiked [nM]	QC4 plasma spiked [nM]
SPC	$X_1$	$100 + X_1$	$360 + X_1$
GlcSph	$X_2$	$10 + X_2$	$36 + X_2$

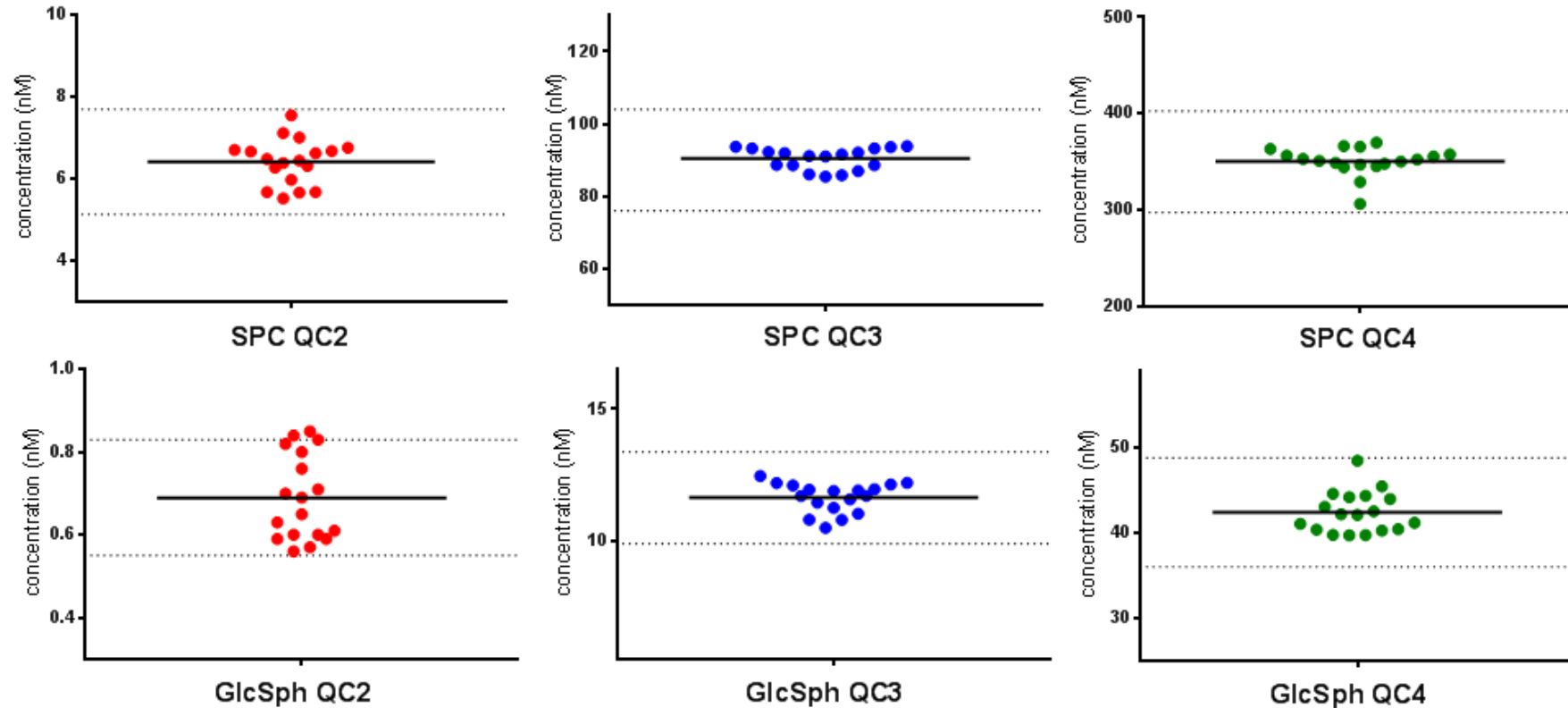
Analyte	CAL1	CAL2	CAL3	CAL4	CAL5	CAL6	CAL7	CAL8	CAL9
	[nM]	[nM]	[nM]	[nM]	[nM]	[nM]	[nM]	[nM]	[nM]
SPC	5	10	25	50	100	160	250	360	500
GlcSph	0.5	1	2.5	5	10	16	25	36	50

CAL samples are prepared by spiking standard SPC and GlcSph in 10 fold diluted plasma (PBS)



aliquots  
in barcoded storage tubes  
frozen at  $-20^{\circ}\text{C}$ .

# precision: 3 batches



## SPC

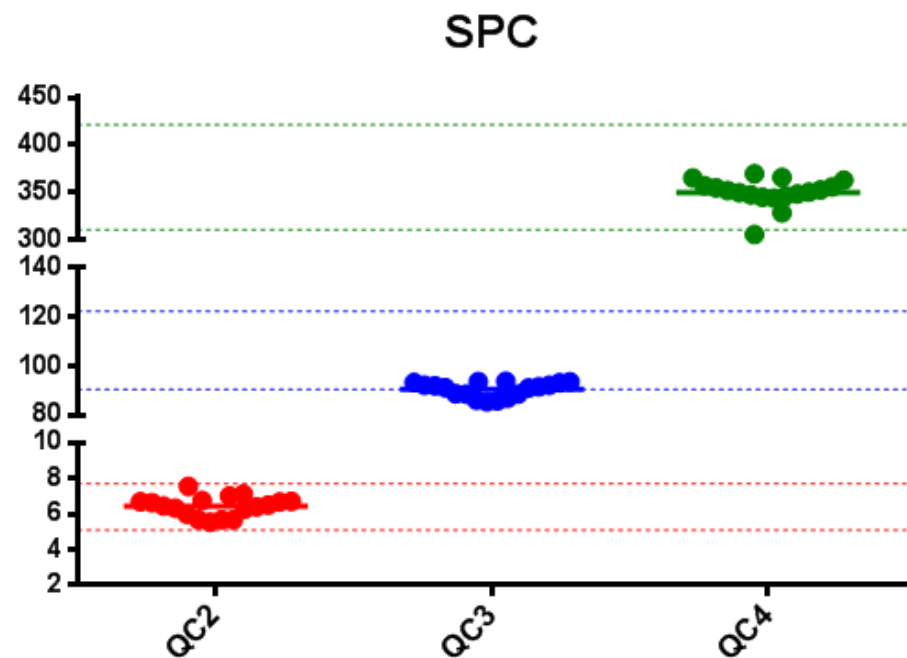
	QC2	QC3	QC4
CV [%]	8.6	3.2	4.2

## GlcSph

	QC2	QC3	QC4
CV [%]	15.0	4.8	5.7

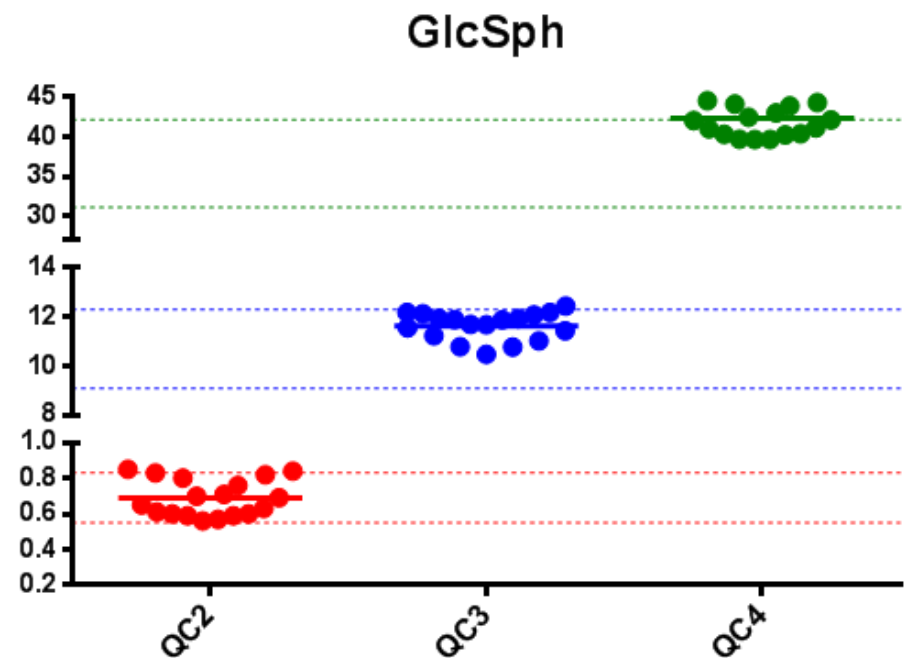


# Accuracy: 3 batches



SPC

	QC2	QC3	QC4
accuracy [%]	100.0	85.0	95.7



GlcSph

	QC2	QC3	QC4
accuracy [%]	100.0	108.8	115.6

# Robustness: Validation on different LCMS platforms

SHIMADU



QTRAP  
API4000



DIONEX



QTRAP  
5500



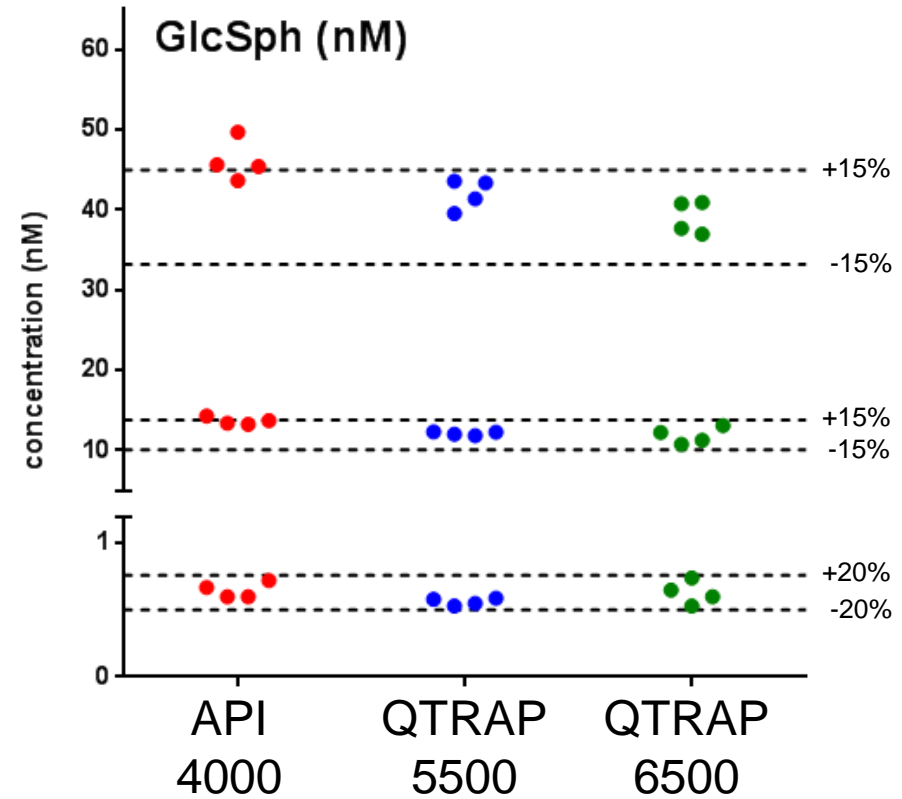
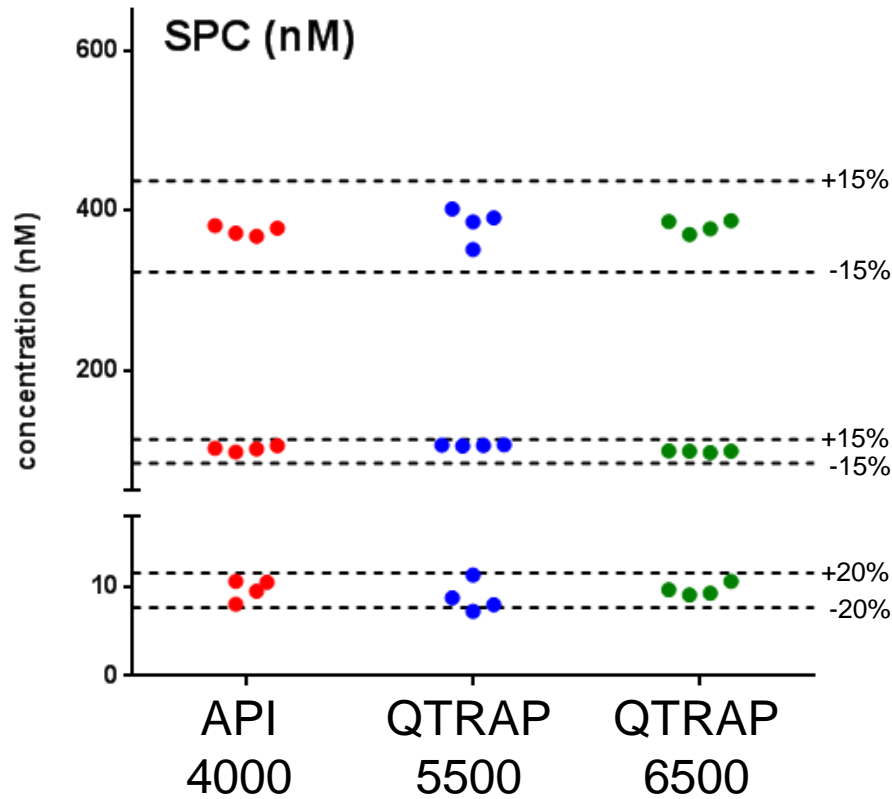
DIONEX



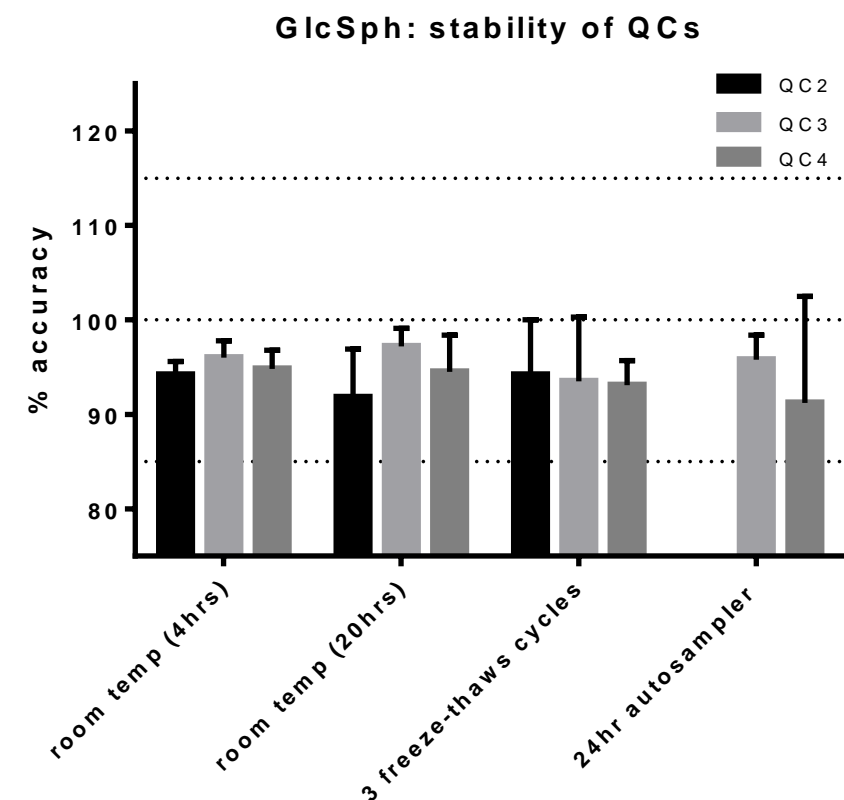
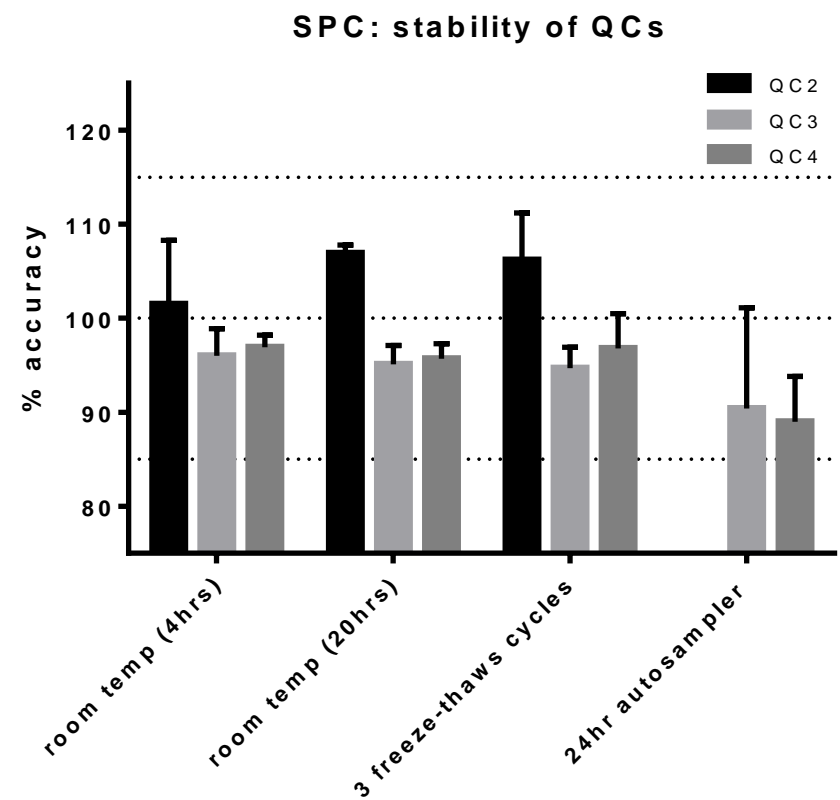
QTRAP  
6500



# Robustness: QCs on different LCMS platforms

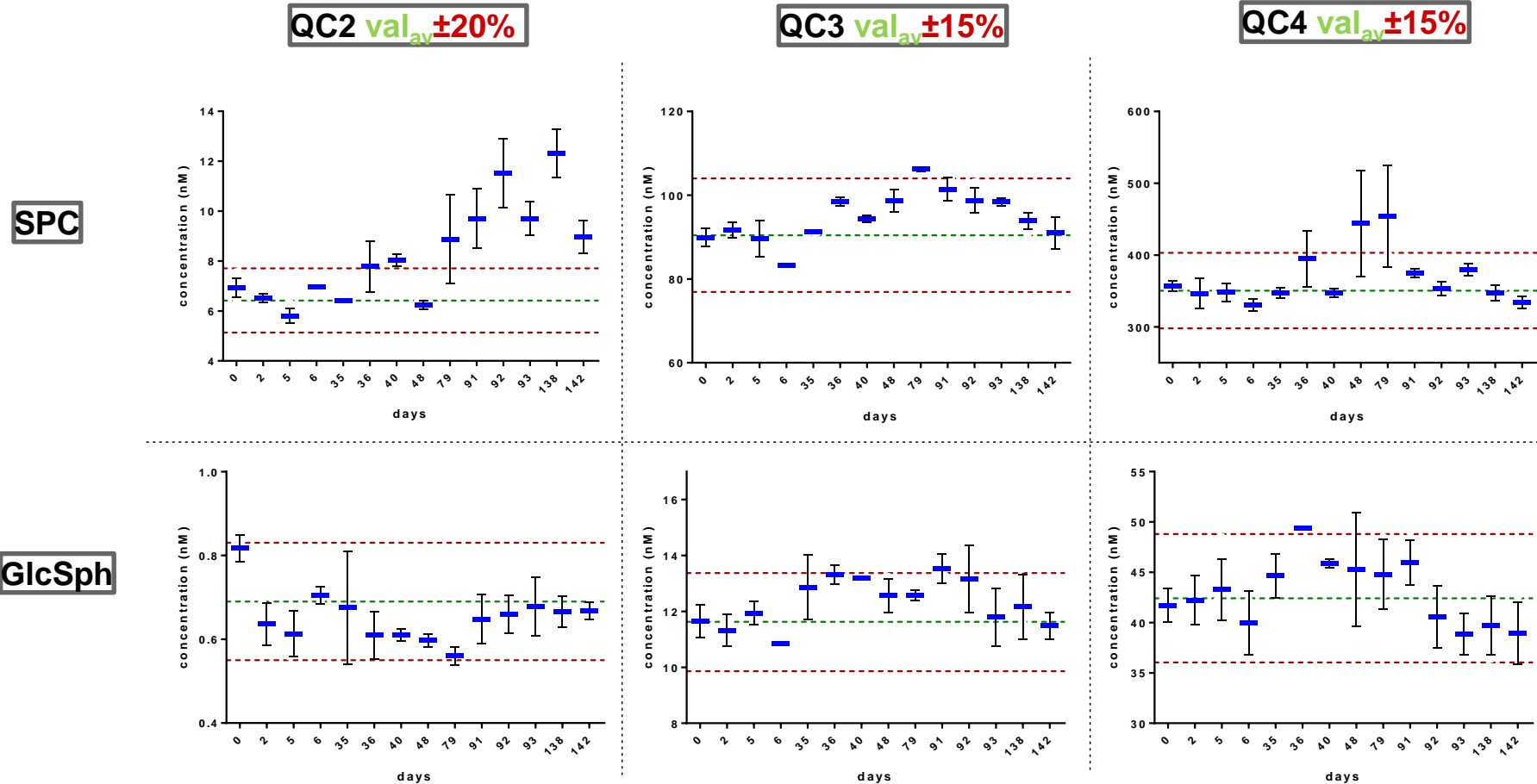


# Short term Stability of Qcs



mean±SD (n = 3)

# Long term Stability of Qcs

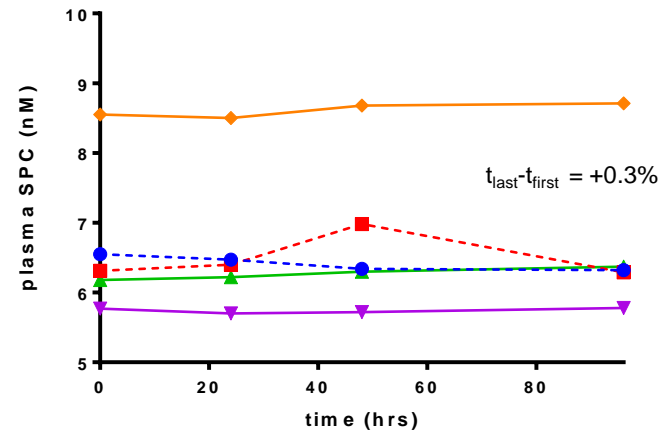


-Issue observed with QC2 for SPC at >50days when stored at -20°C  
 -But, 2yr old samples stored at -80°C are in the normal range

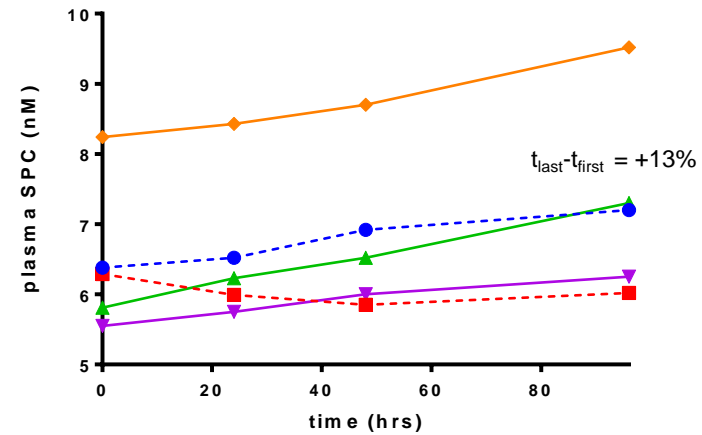
# Plasma stability: 5 donors, 96hrs

SPC

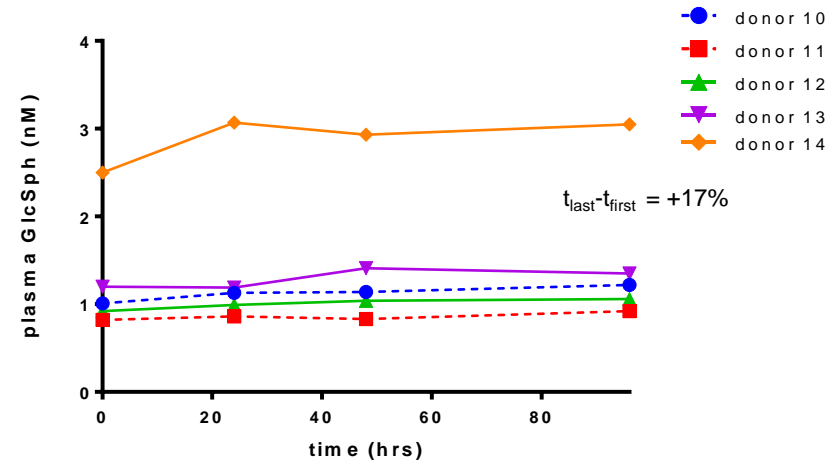
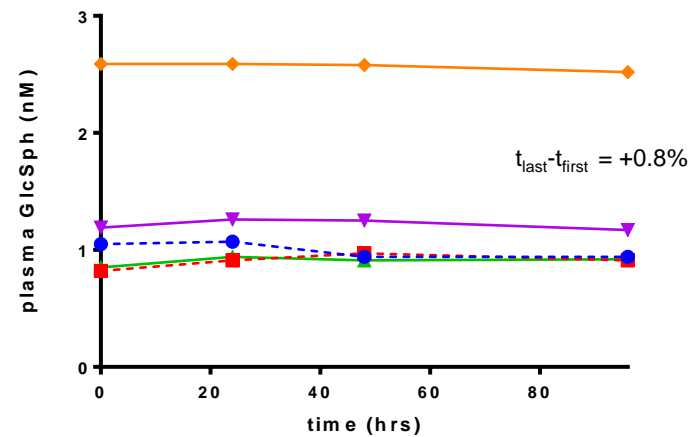
4°C



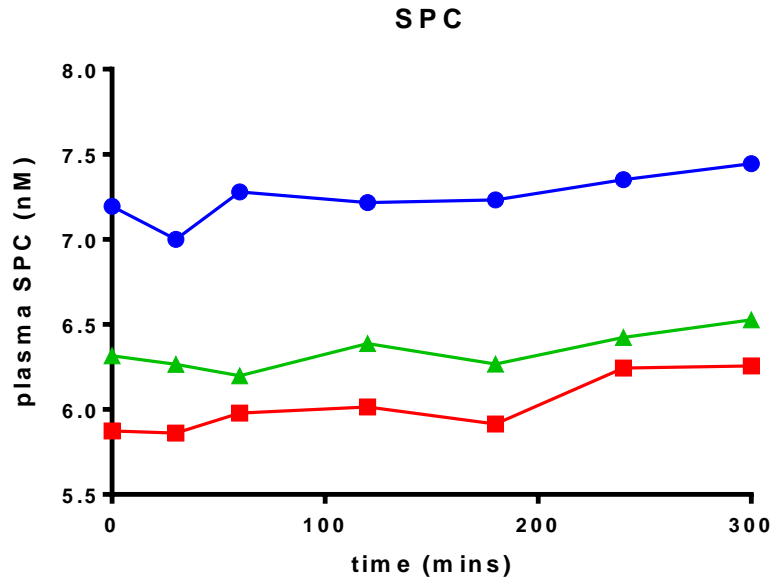
Room temp



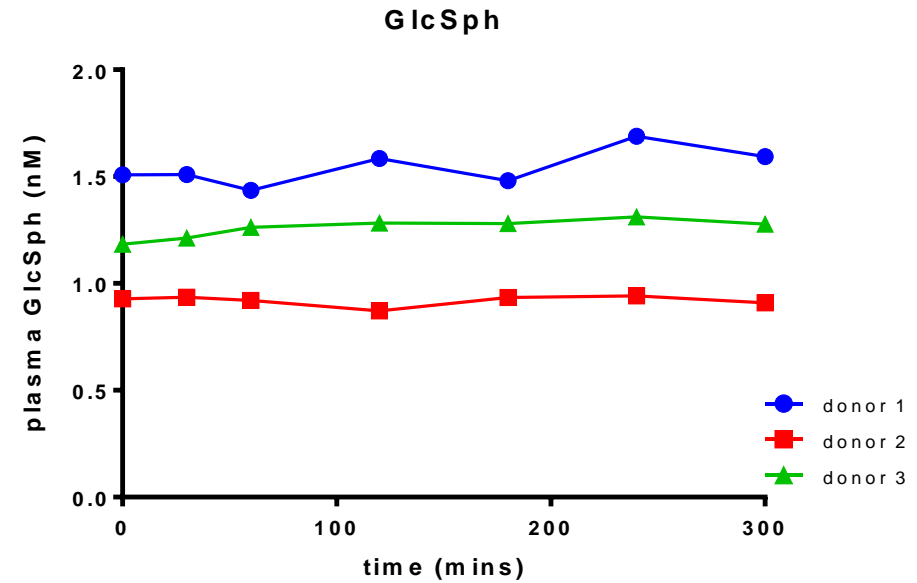
GlcSph



# Whole Blood stability: 3 donors, 5 hours, room temperature



$$t_{\text{last}} - t_{\text{first}} = +4.5\%$$



$$t_{\text{last}} - t_{\text{first}} = +3.8\%$$



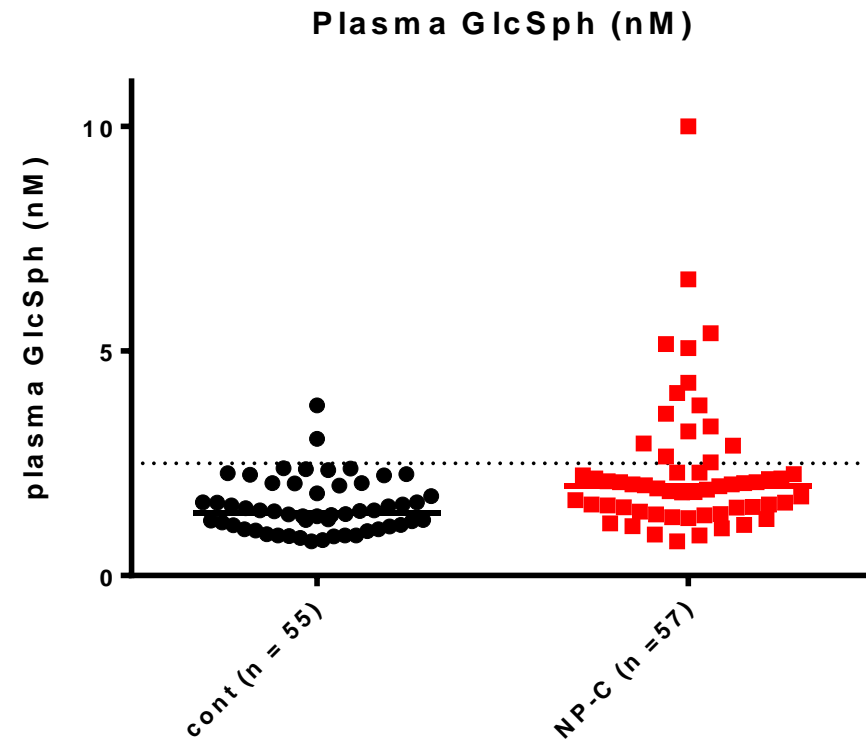
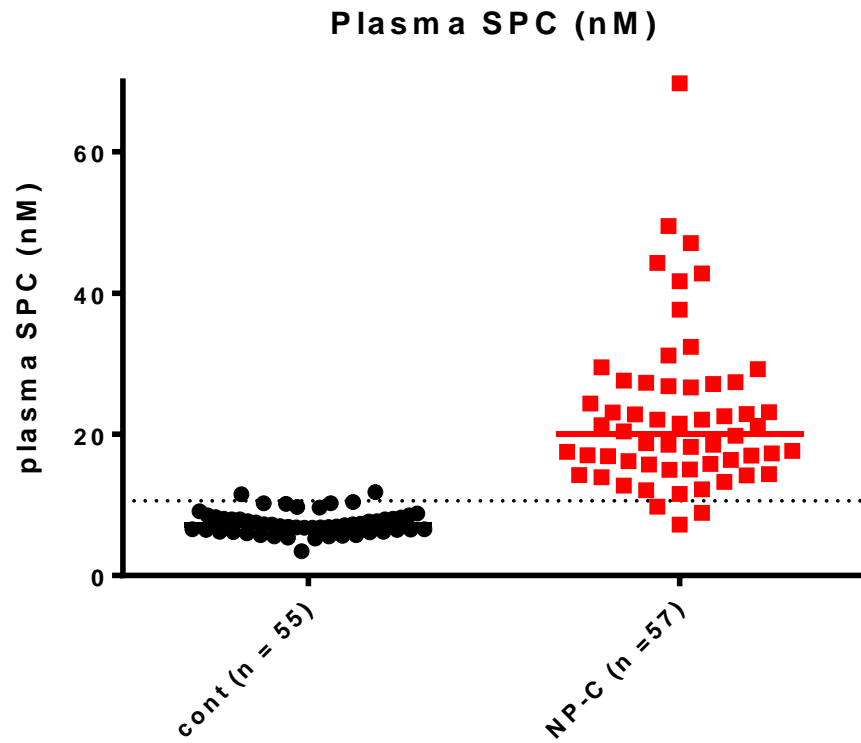
# Application to NP-C patient samples

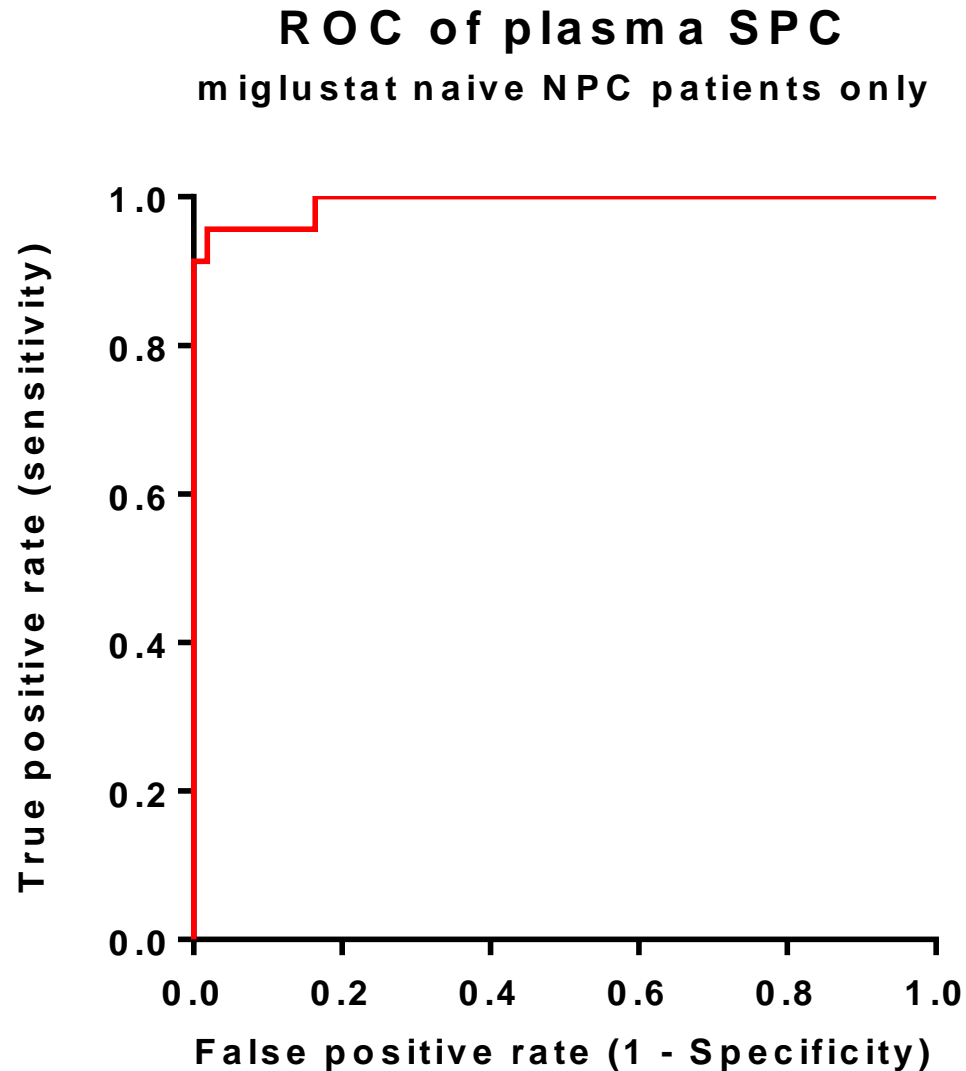
# Retrospective case control analysis

- 1 sample per subject
- NP-C samples
  - From 3 centers
- Control samples
  - From 2 centers and one commercial provider

Group	Control	NP-C
<b>Number of values</b>	55	57
<b>Male (%)</b>	42	44
<b>Female (%)</b>	58	56
<b>Age (yrs) 25% percentile</b>	9	10.5
<b>Median age (yrs)</b>	14	15
<b>Age (yrs) 75% percentile</b>	23	22
<b>Receiving miglustat (%)</b>		44
<b>Miglustat naive (%)</b>		40
<b>Unknown miglustat status (%)</b>		16

# SPC and GlcSPH





- ▶ 0-50 years, miglustat naïve patients, n = 23
- ▶ Controls, n = 55
- ▶ Area = 0.992
- ▶ Cut-point of 10.6nM separates control from NP-C with a sensitivity of 96% and a specificity of 95%

# PPV and NPV

- Positive predictive value (PPV)- “What is the likely hood that the patient has the disease when the test result is positive”
- Negative predictive value (NPV)- “The probability that the patient does not have the disease when the result is negative”

prevalence	sensitivity	specificity		PPV	NPV
0.05	0.9565	0.9565		0.5365	0.9976
0.01	0.9565	0.9565		0.1817	0.9995
0.00001	0.9565	0.9565		0.0002	1.0000
0.05	1	0.97		0.6369	1.0000
0.01	1	0.97		0.2519	1.0000
0.00001	1	0.97		0.0003	1.0000

- Context of use and the decisions to be made are important for understanding usefulness of a test (note sens and spec also need to be determined in right setting)
- In the case of NP-C combination of neurological symptoms, BMs and genetics
- In broad screening there will be many false positives even with a high specificity diagnostic assay

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**Villa Metabolica**  
**Mainz, Germany**

Dr Eugen Mengel

**Universitätsklinikum**  
**Münster, Germany**

Dr Thorsten Marquardt

Dr Janine Reunert

RESEARCH ARTICLE

# Plasma Lysosphingomyelin Demonstrates Great Potential as a Diagnostic Biomarker for Niemann-Pick Disease Type C in a Retrospective Study

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**Data Availability:** The authors confirm that all data underlying the findings are fully available without restriction. All relevant data are within the paper and its Supporting Information files.

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**Competing Interests:** RW, MG, SK, OM and PG are employees of Actelion Pharmaceuticals Ltd; CML, EM, YA, JR and TM have received travel reimbursements, consulting fees or honoraria from Actelion Pharmaceuticals Ltd, Allschwil, Switzerland. The competing interests do not alter the authors' adherence to PLOS ONE policies on sharing data and materials.

## Abstract

Niemann-Pick disease type C (NP-C) is a devastating, neurovisceral lysosomal storage disorder which is characterised by variable manifestation of visceral signs, progressive neuropsychiatric deterioration and premature death, caused by mutations in the *NPC1* and *NPC2* genes. Due to the complexity of diagnosis and the availability of an approved therapy in the EU, improved detection of NP-C may have a huge impact on future disease management. At the cellular level dysfunction or deficiency of either the NPC1 or NPC2 protein leads to a complex intracellular endosomal/lysosomal trafficking defect, and organ specific patterns of sphingolipid accumulation. Lysosphingolipids have been shown to be excellent biomarkers of sphingolipidosis in several enzyme deficient lysosomal storage disorders. Additionally, in a recent study the lysosphingolipids, lysosphingomyelin (SPC) and glucosylsphingosine (GlcSph), appeared to be elevated in the plasma of three adult NP-C patients. In order to investigate the clinical utility of SPC and GlcSph as diagnostic markers, an in-depth fit for purpose biomarker assay validation for measurement of these biomarkers in plasma by liquid chromatography-tandem mass spectrometry was performed. Plasma SPC and GlcSph are stable and can be measured accurately, precisely and reproducibly. In a retrospective analysis of 57 NP-C patients and 70 control subjects, median plasma SPC and GlcSph were significantly elevated in NP-C by 2.8-fold and 1.4-fold respectively. For miglustat-naïve NP-C patients, aged 2–50 years, the area under the ROC curve was 0.999 for SPC and 0.776 for GlcSph. Plasma GlcSph did not



# Collaboration with Rare Disease Centers

Avoid the stereo typing, pharma is working intensively with the community



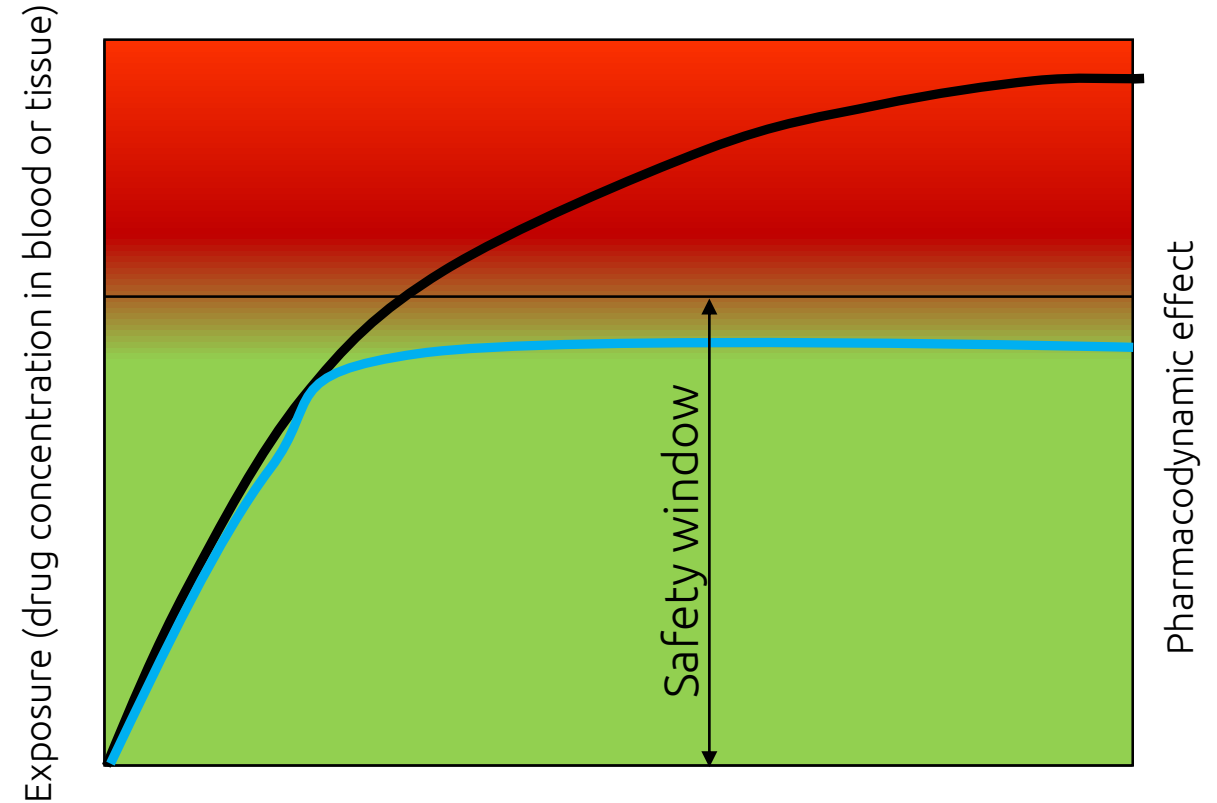
# Case example 3

Pharmacodynamic Response  
Biomarker: Target Engagement

# For drugs it's all about the dose and the effect

## Therapeutic vs. Safety window

- Paracelsus: "All things are poison, and nothing is without poison, the dosage alone makes it so a thing is not a poison."
- Searching for new drugs requires establishing a clear safety window. This preferably is much bigger than the therapeutic window

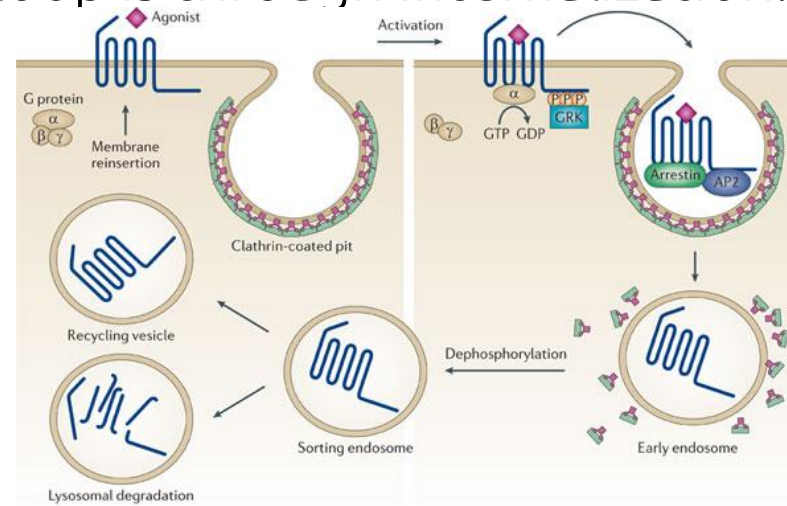


# Pharmacodynamic and Target engagement in the search for a potent CRTH2 antagonist for seasonal allergic rhinitis

# Developing a flow cytometer assay for receptor internalization

Regulation of signalling of GPCRs is complex

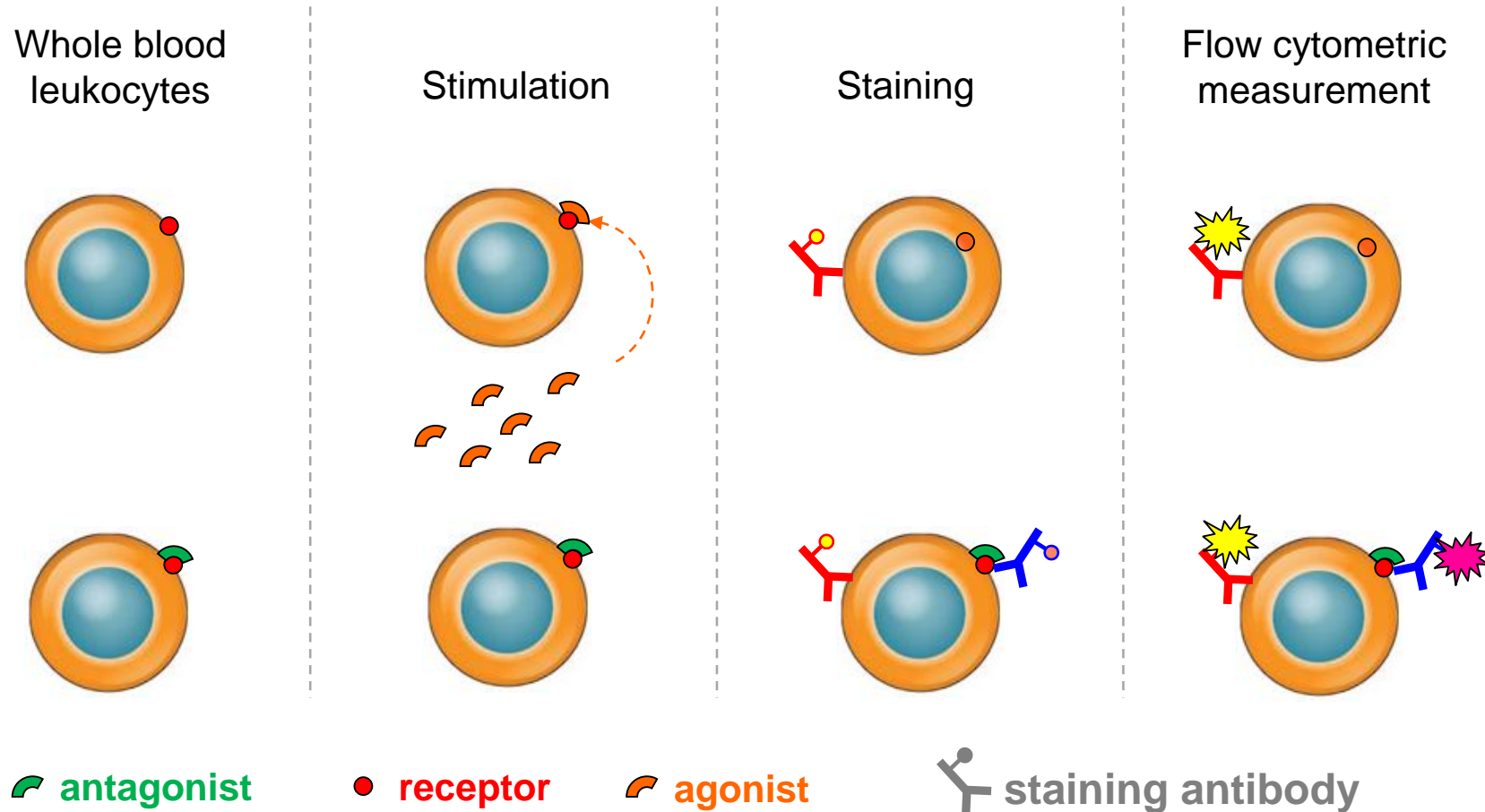
- GPCR signalling is a complex process. One of the ways some GPCR signalling is controlled in a feedback loop is through internalization.



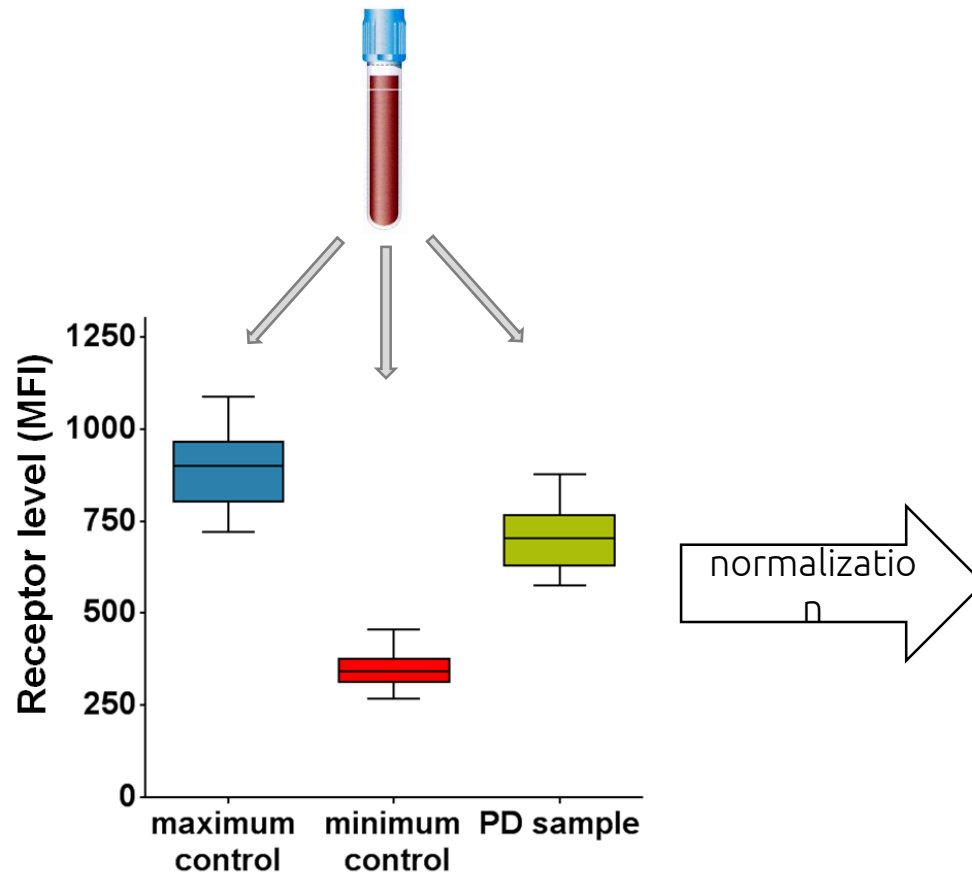
From: Ramachandran et al. *Nature Reviews Drug Discovery* 2012

- Receptor internalization is a very proximate target engagement biomarker and in principle also a pharmacodynamic, downstream signalling marker
- Measuring receptor levels ex vivo reveals a dynamic view on relation between exposure (PK) and effect (PD)

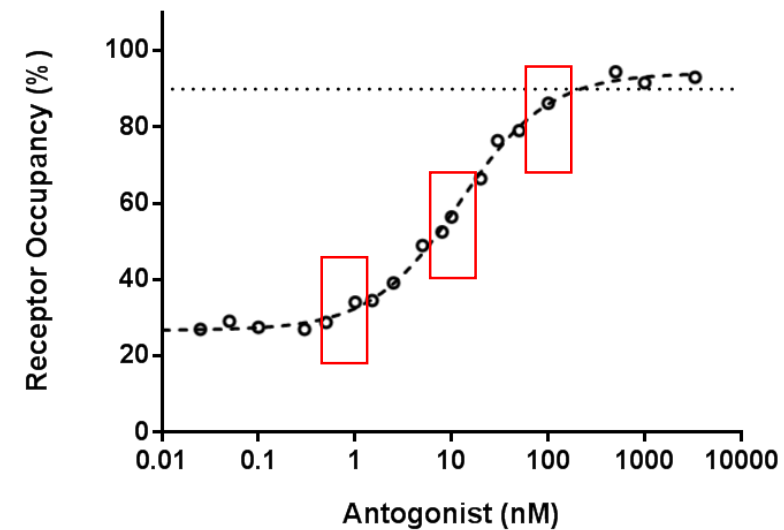
# Receptor Occupancy Assay Principle



# Normalization



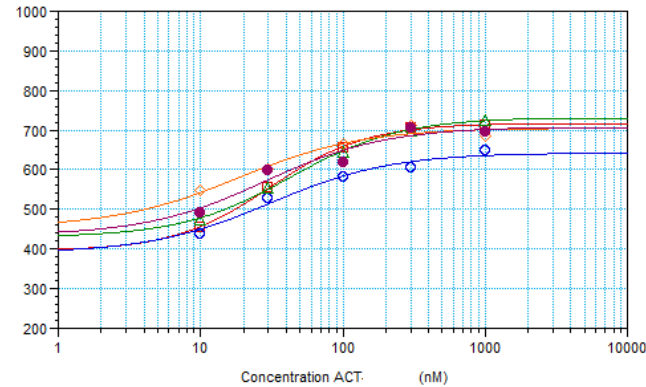
Pharmacodynamic (PD) effect



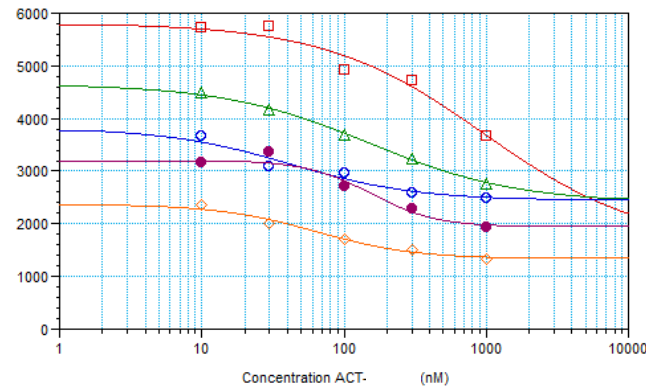


# Biomarker Selection - Precision

CRTH2 internalization



CD11b upregulation



CRTH2  
internalization

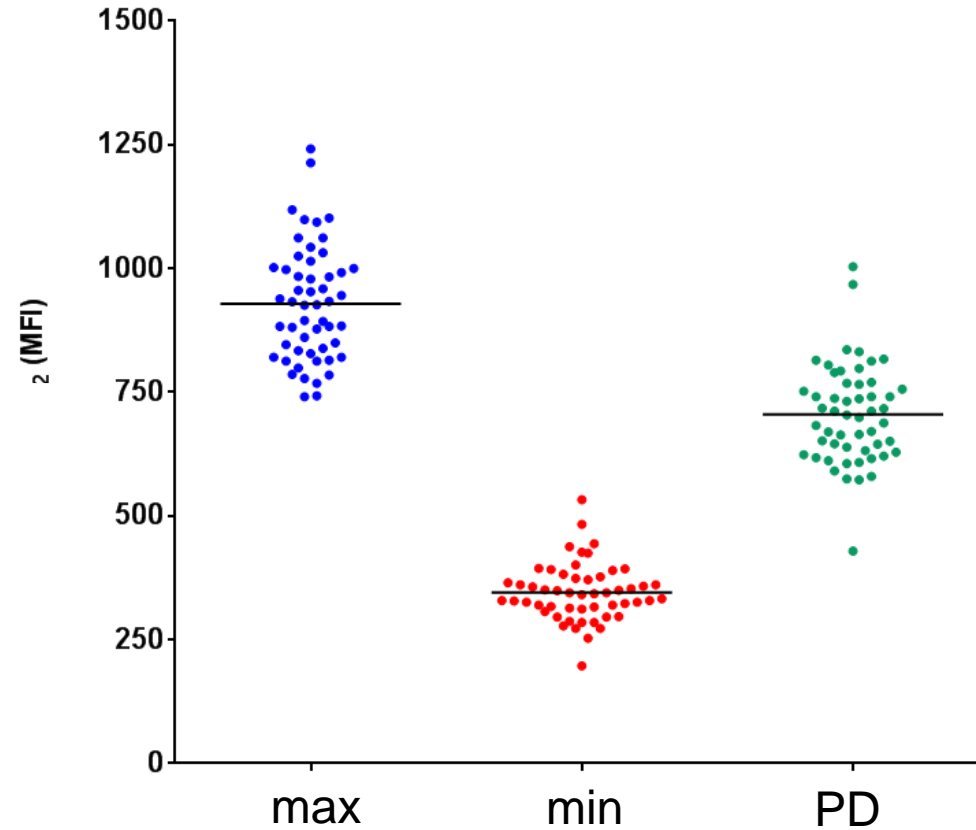
CD11b  
upregulation

	CRTH2 internalization	CD11b upregulation
Donor 1	30	256
Donor 2	33	40
Donor 3	41	104
Donor 4	15	67
Donor 5	27	168
Mean	29.2	127.0
SD	9.5	86.6
CV%	33%	68%

# Biomarker Selection - Feasibility

## Actelion Blood Donation Day:

- feasibility
- throughput
- normalization



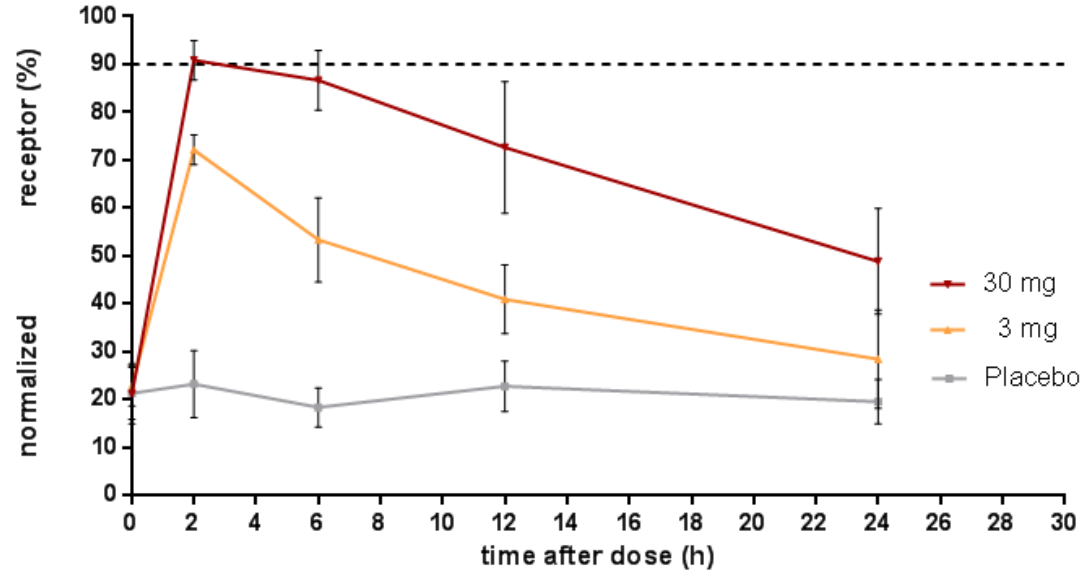
GPCR internalization assay on PBMCs.  
applying intrinsic normalization to calculate the PD effect.

# Fit-for-Purpose Assay Validation

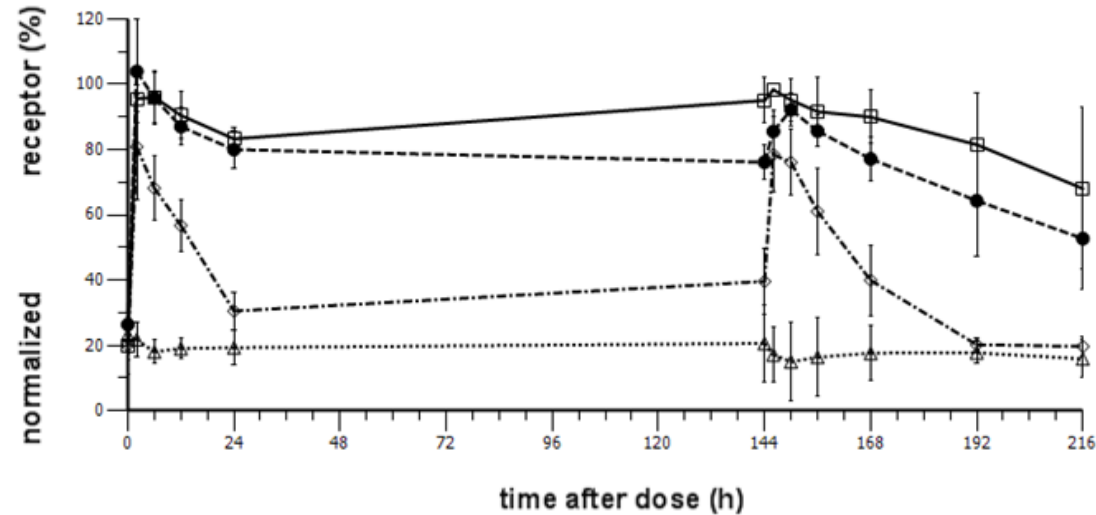
Validation parameters		FACS	Results	
		ex-vivo stimulation		
Selection of ; and		✓	102%	99%
Specificity of the anti- <small>GPCR</small> antibody		✓	unstained < isotyp control < minimum level	
Precision	intra-assay	✓	4%	5%
	inter-assay	✗	---	
	intra-subject	✗	---	
	inter-subject	✓	12%	15%
Accuracy		✗	---	
Calibration curve		✗	---	
Assay range	LLOQ	✓	6 nM	
	HLOQ	✓	140 nM	
Stability	Reagent (-80°C)	✓	over 2 months	
	Reagent (freeze / thaw)	✓	over 5 cycles	
Robustness	blood samples	✓	24h at 4°C	
	prepared samples	✓	> 48h	24h
	combined	✓	12h + 24h	2h + 24h

Outcome: reliable PD biomarker for GPCR antagonist efficiency

# Phase I PD Biomarker Results



SAD



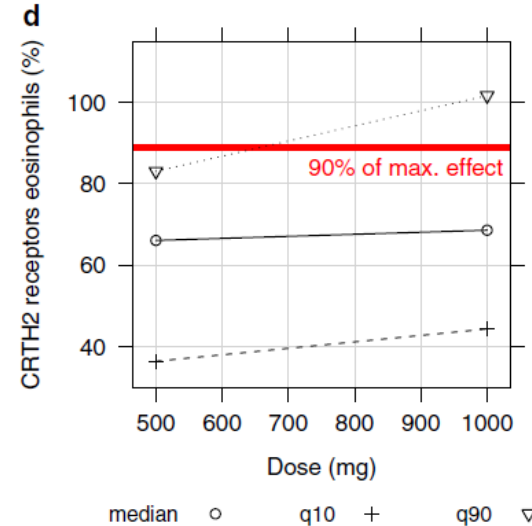
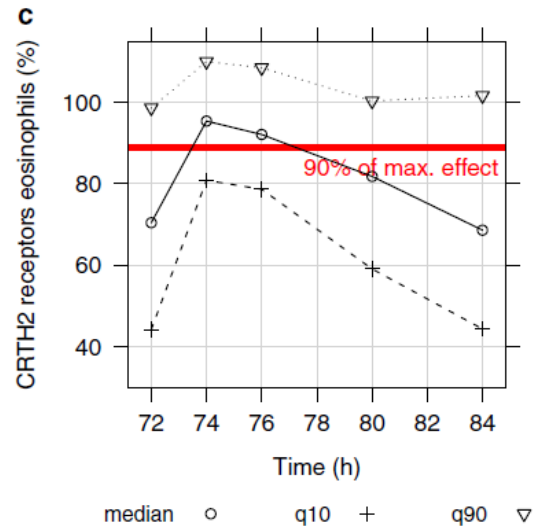
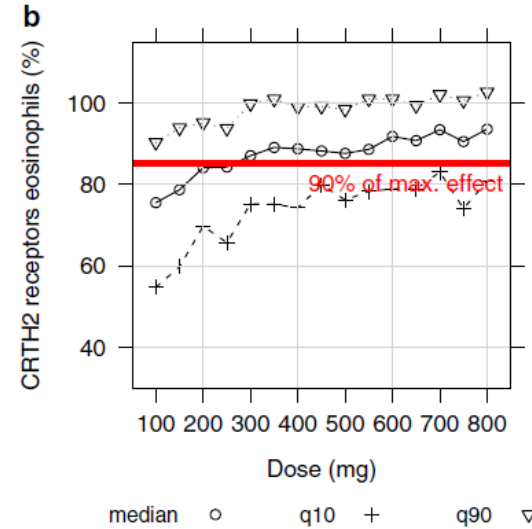
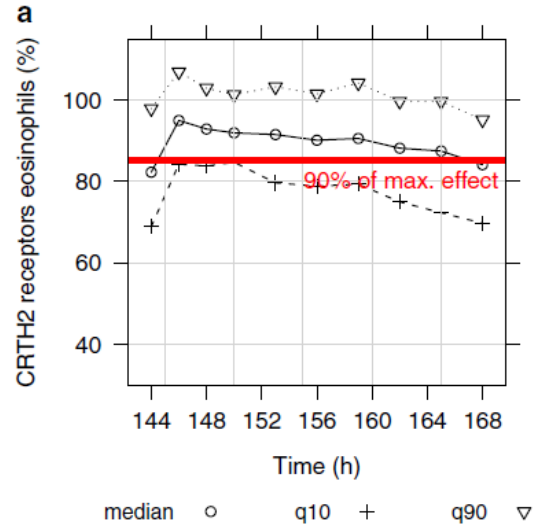
MAD

800 mg o.d.  
100 mg o.d.  
10 mg o.d.  
Placebo o.d.

# PK/PD modelling

PK/PD model  
determined  
dosing regimen

GO!



Failed trial in  
Seasonal Allergic  
Rhinitis (SAR)

# acknowledgements

## **Idorsia**

Daniel Strasser

Herve Farine

Virginie Sippel

Martin Holdener

Martine Gehin-Beurne

Patricia Sidharta

Jasper Dingemanse

Andreas Krause

Jochen Zisowsky



## Pharmacokinetic/Pharmacodynamic Modelling of Receptor Internalization with CRTH2 Antagonists to Optimize Dose Selection

Andreas Krause<sup>1</sup> · Jochen Zierecky<sup>1</sup> · Daniel S. Strauss<sup>2</sup> · Martine Gehin<sup>1</sup> ·  
Patrick N. Sidharta<sup>1</sup> · Peter M. A. Groenen<sup>2</sup> · Jasper Dierckx<sup>1</sup>

Published online: 21 December 2015  
© Springer International Publishing Switzerland 2015

### Abstract

**Background and Objective** The chemottractant receptor-homologous molecule expressed on T helper-2 cells (CRTH2) is a G-protein-coupled receptor for prostaglandin D<sub>2</sub> (PGD<sub>2</sub>), a key mediator in inflammatory disorders. Two selective and potent CRTH2 antagonists currently in clinical development, ACT-453859 and astipiprant, were compared with respect to their (predicted) clinical efficacy.

**Methods** Population pharmacokinetic (PK) and pharmacodynamic (PD) models were developed to characterize how plasma concentrations (PK) of ACT-453859, its active metabolite ACT-463036 and astipiprant related to their effect on blocking PGD<sub>2</sub>-induced internalization of CRTH2 on eosinophils (PD). Simulations were used to identify doses and dosing regimens leading to 90 % of maximum blockade of CRTH2 internalization at trough.

**Results** A combined concentration of ACT-453859 and its metabolite ACT-463036, with weights proportional to potency (based on an eosinophil shape change assay), enabled good characterization of the PD effect. The modelling and simulation results facilitated decision making by suggesting an ACT-453859 dose of 400 mg once daily (or

100 mg twice daily) for clinically relevant CRTH2 antagonism.

**Conclusion** Pharmacometric quantification demonstrated that CRTH2 internalization is a useful new biomarker to study CRTH2 antagonism. Ninety percent of maximum blockade of CRTH2 internalization at trough is suggested as a quantitative PD target in clinical studies.

### Key Points

CRTH2 [chemottractant receptor-homologous molecule expressed on T helper-2 cells] internalization is a useful new biomarker for CRTH2 antagonists, a new class of compounds with promising characteristics for treatment of allergic diseases.

This biomarker is of great help for designing future clinical studies and selecting efficacious doses of such drugs.

This biomarker can accelerate development of this class of drugs.

**Electronic supplementary material** The online version of this article (doi:10.1007/s40262-015-0354-3) contains supplementary material, which is available to authorized users.

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### 1 Introduction

The chemottractant receptor-homologous molecule expressed on T helper-2 cells (CRTH2) is a G-protein-coupled receptor expressed on the surface of blood-borne cells (T helper-2 lymphocytes, eosinophils and basophils) [1]. The receptor mediates the activation and chemotaxis of

△ Adis

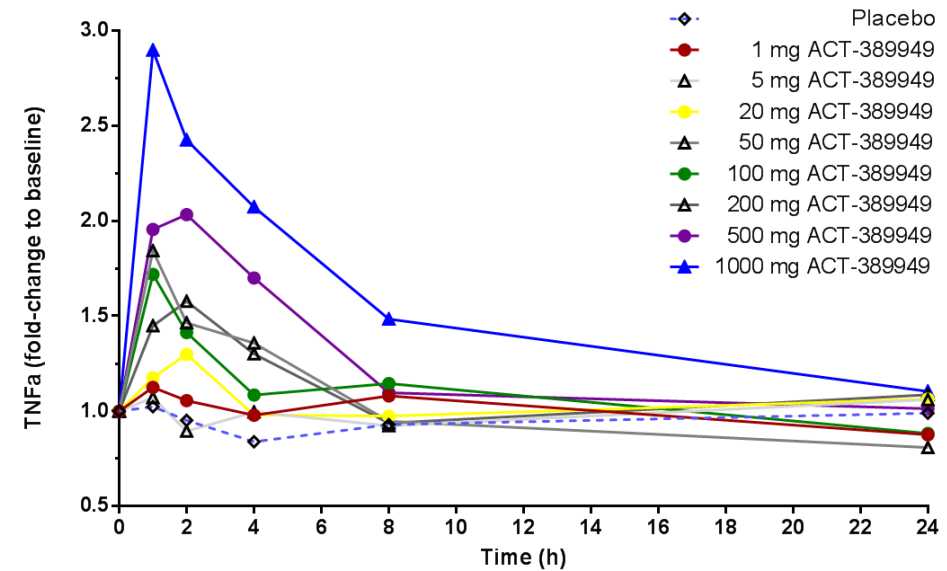
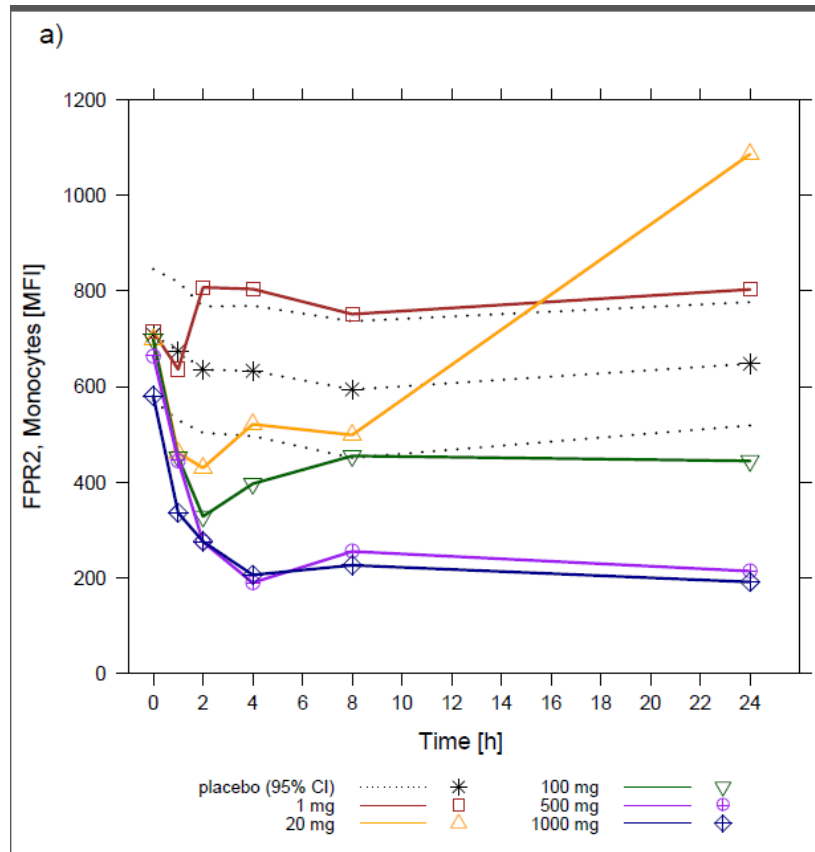


# Another GPCR story

## Pharmacodynamic Biomarkers and Target Engagement

# Results: SAD

SAD dose responsiveness of FPR2 internalization and TNFa

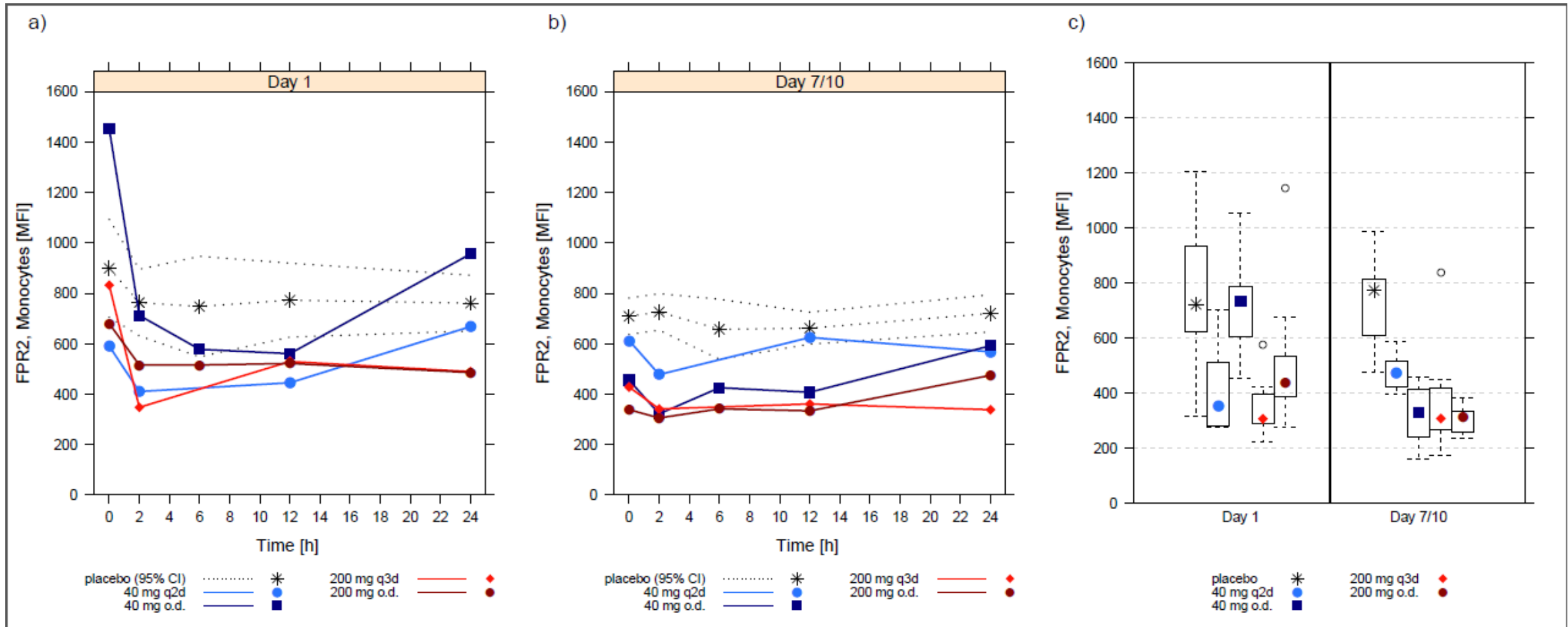


# Dosing schedules and Doses Tested

Study drug administration

<div> <div>Qd3 (200mg) ↓ ↓ ↓ ↓ ↓</div> <div>Qd2 (40mg) ↓ ↓ ↓ ↓ ↓ ↓ ↓ ↓</div> <div>Daily (40, 200 800) ↓ ↓ ↓ ↓ ↓ ↓ ↓ ↓ ↓ ↓ ↓ ↓</div> </div>													
Day:	D1	D2	D3	D4	D5	D6	D7	D8	D9	D10	D11	D12	D13 EOT
Biomarker Sampling	0 2 12 24	2 6 12 24	0	0	0 2 12 24	0	0 2 12 24		0	0 2 12 24	0	0	0 LPS
White blood cell count	0 1 2 6 24									0 1 2 6			Sputum

# target engagement marker



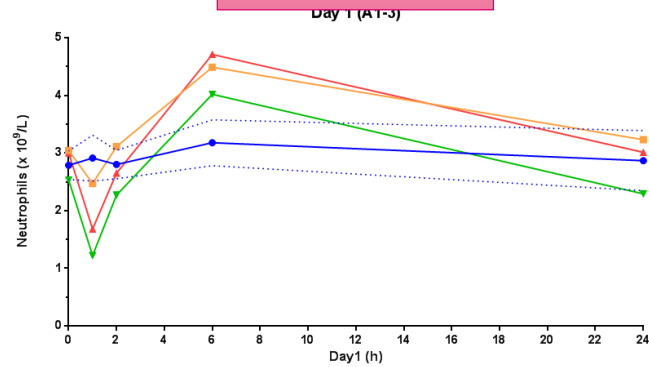
# results

## Hematological effect on blood cell counts

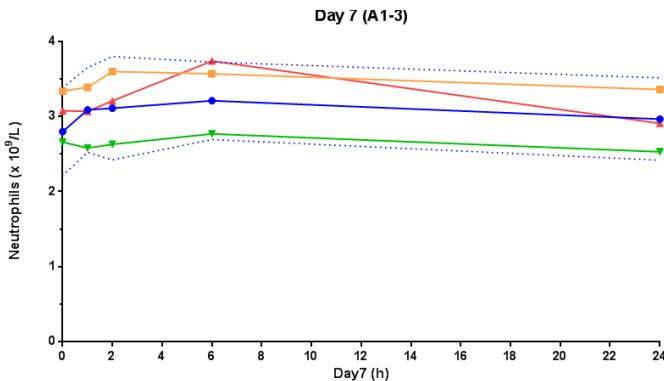
neutrophil

S

DAY 1

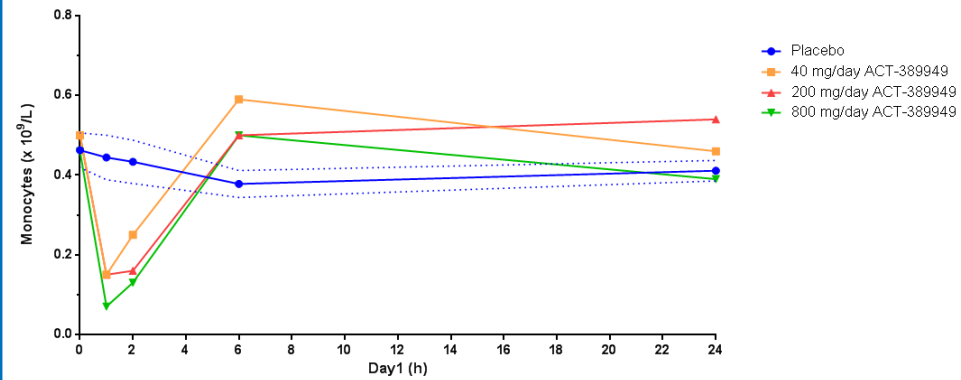


DAY 7

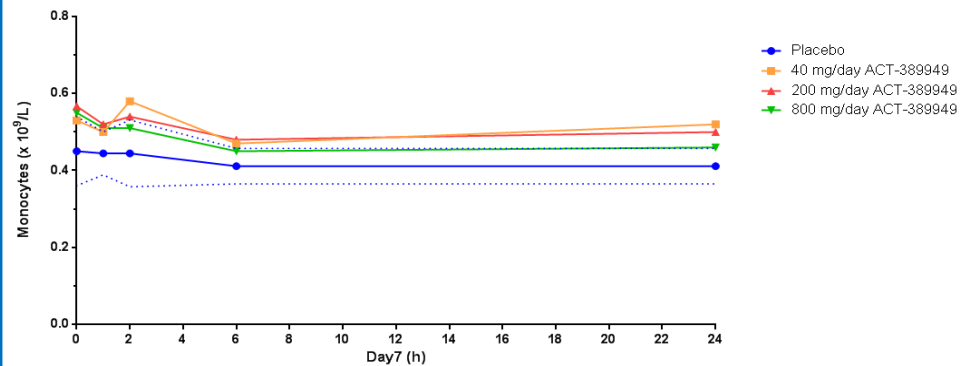


Monocytes

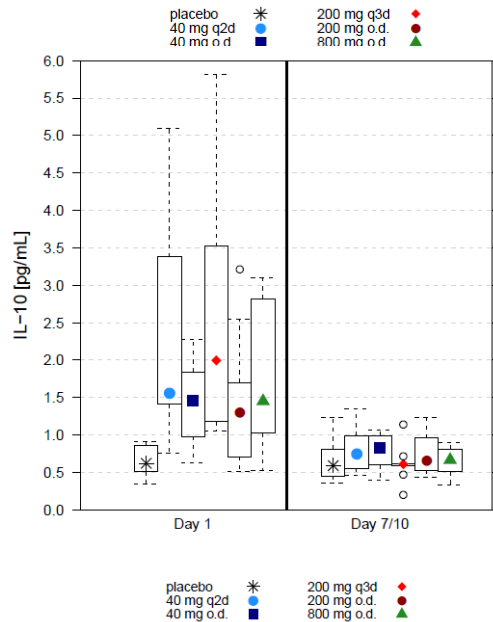
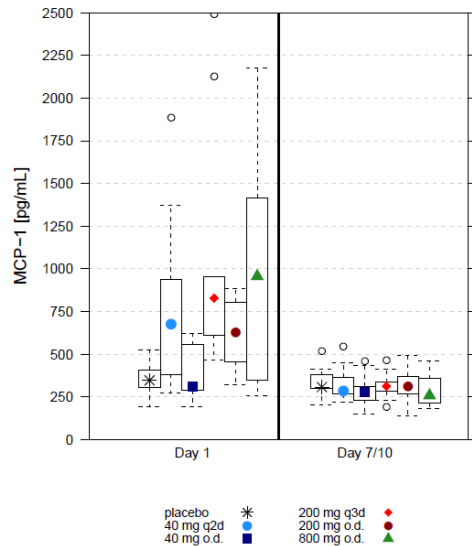
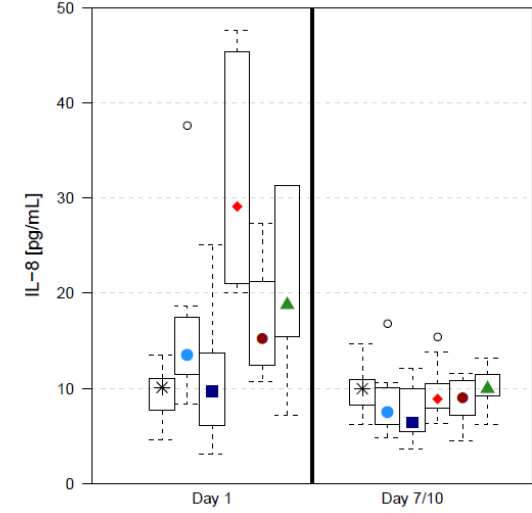
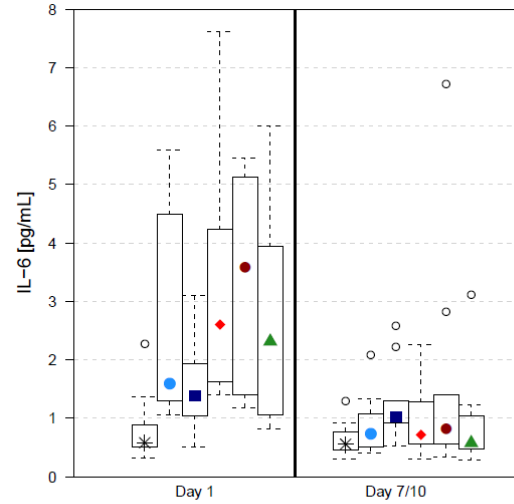
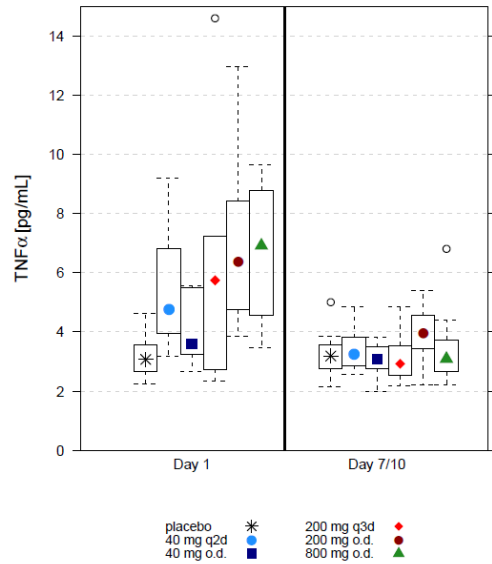
Day 1 (A1-3)



Day 7 (A1-3)



# Downstream marker: cytokines



STOP!

## CLINICAL TRIALS

# Biomarker-guided clinical development of the first-in-class anti-inflammatory FPR2/ALX agonist ACT-389949

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Received 13 July 2016; Revised 19 September 2016; Accepted 4 October 2016

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\*Last authors contributed equally to this study.

**Keywords** biomarker, cytokines, dose finding, FACS, first in class, flow cytometry, FPR2/ALX, inflammation, LPS challenge, sputum

### AIMS

The main objectives of these two phase I studies were to investigate safety and tolerability as well as the pharmacokinetic/pharmacodynamic profile of the novel potent and selective formyl peptide receptor type 2 (FPR2)/Lipoxin A<sub>4</sub> receptor (ALX) agonist ACT-389949. A challenge model was used to assess the drug's anti-inflammatory potential, with the aim of selecting a dosing regimen for future patient studies.

### METHODS

Two double-blind, randomized phase I studies investigated the safety, tolerability, pharmacokinetics and pharmacodynamics of ACT-389949 at different doses and dosing regimens. Drug exposure was correlated with target engagement markers such as receptor internalization and cytokine measurements. The effect of FPR2/ALX agonism on neutrophil migration was studied in a lipopolysaccharide (LPS) inhalation model.

### RESULTS

ACT-389949 was well tolerated. Maximum concentrations were reached around 2 h after dosing, with a mean terminal half-life of 29.3 h [95% confidence interval (CI) 25.5, 33.7]. After multiple-dose administration, exposure increased by 111% (95% CI 89, 136), indicating drug accumulation.

Administration of ACT-389949 resulted in a dose-dependent, long-lasting internalization of FPR2/ALX in to leukocytes. Pro- and anti-inflammatory cytokines were dose-dependently but transiently upregulated only after the first dose. No pharmacological effect on neutrophil count was observed in the LPS challenge test performed at steady state.

### CONCLUSIONS

FPR2/ALX agonism with ACT-389949 was shown to be safe and well tolerated in healthy subjects. Receptor internalization and downstream mediators pointed towards a desensitization of the system, which may explain the lack of effect on neutrophil recruitment in the LPS challenge model.





Biomarkers in the lab  
versus biomarkers on site

The case for assay  
transfer and monitoring

# Method familiarization

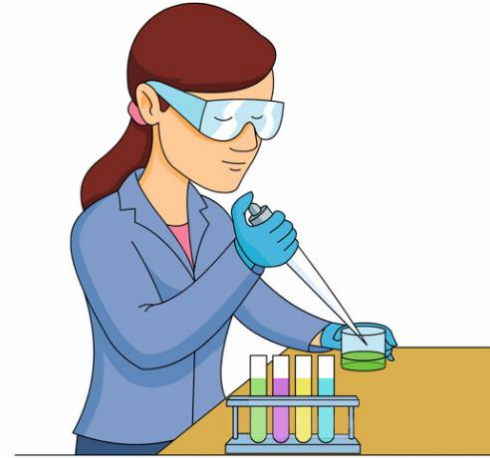
1



## Presentation

- assay protocol
- results obtained
- system suitability criteria
- highlighting of the critical steps

2



## Training session in the lab

- 2 fresh EDTA blood samples
- 2 to 3 different drug levels
- 1 vehicle

Team

- 5 technicians for the clinical site
- 2 technicians for the flow cytometry facilities
- 2 lab managers

Is it not going to  
cost even more?

# Costs is not the right question

## DEEP DIVE

### How biomarkers cost Bristol-Myers the lung cancer market



Credit: [Flickr, A4](#)

One striking example which has been a dramatic difference between to similar concepts

Anti PD1 (immune checkpoint inhibitor)

BMS: Opdivo  
Merck: Keytruda

Having a biomarker early in the process to differentiate patients on PD1 levels turned to be a costly, yet profitable. Failure costed BMS significantly.

# Probability of clinical success significantly improves with biomarkers

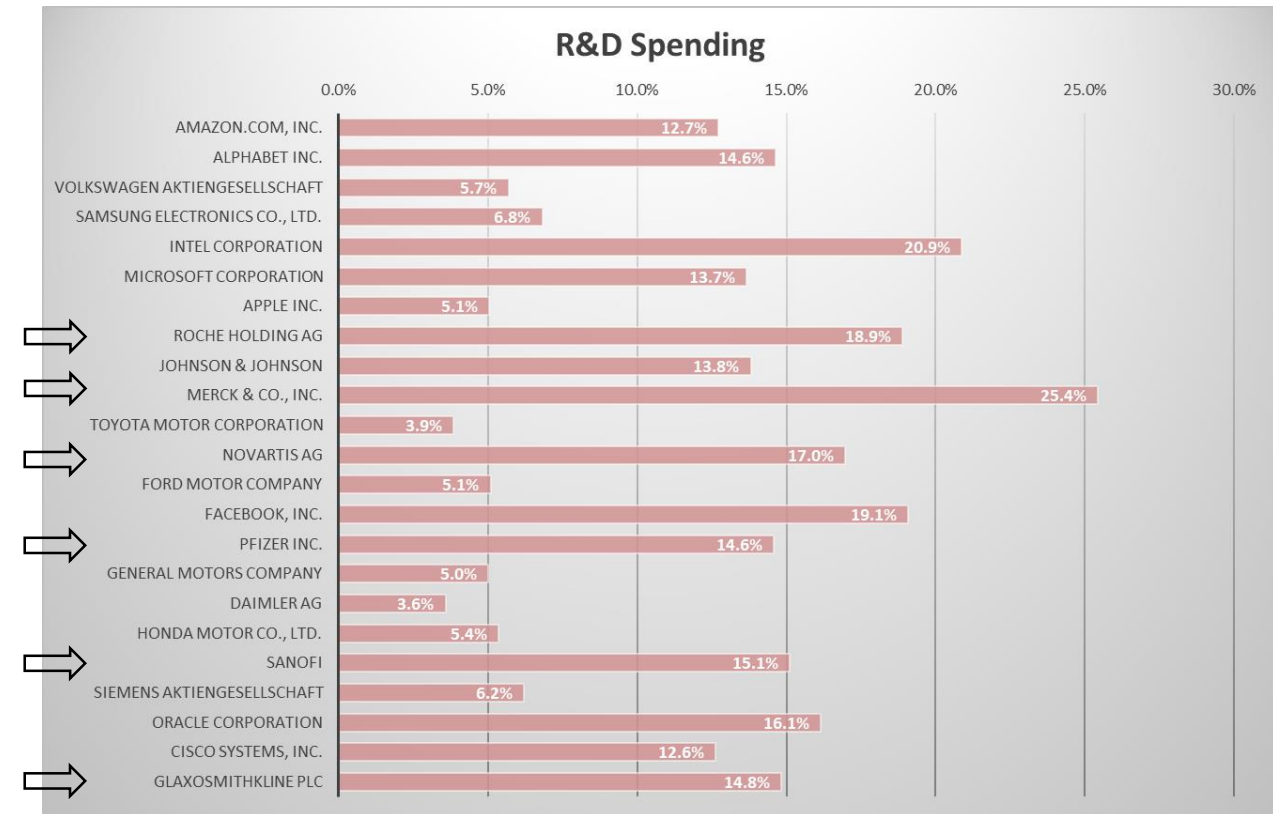
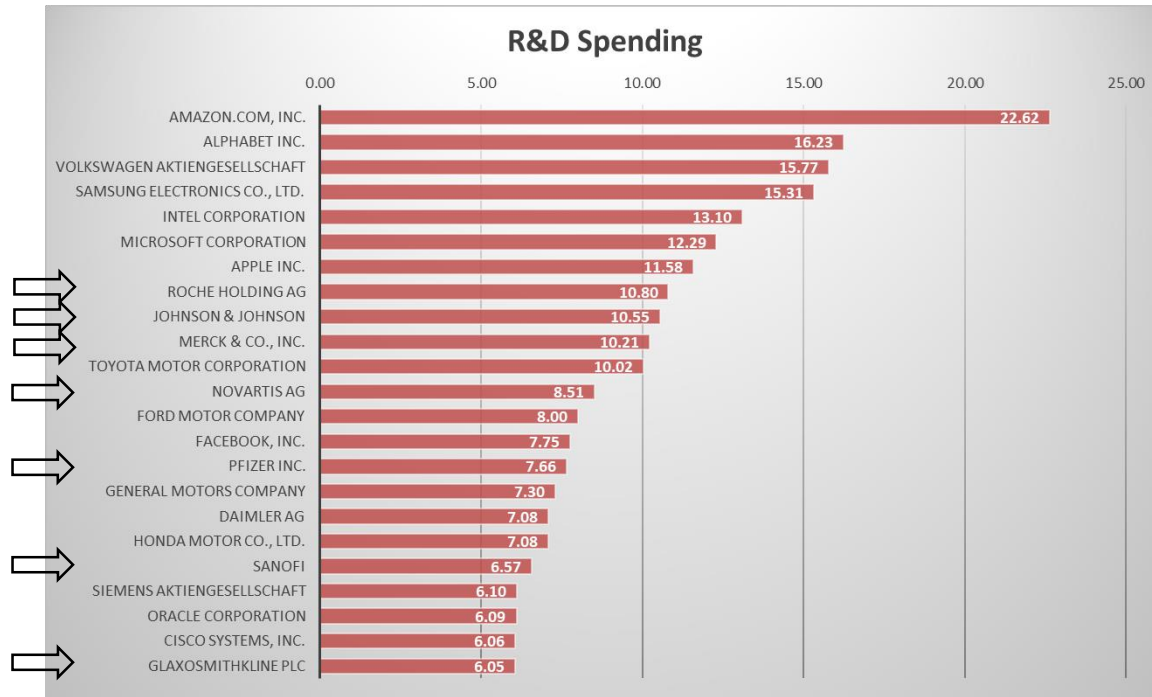
Table 3. POS of drug development programs with and without biomarkers, using data from January 1, 2005, to October 31, 2015, computed using the phase-by-phase method. These results consider only trials that use biomarkers in patient stratification. Since for the majority of trials using biomarkers (92.3%) their status is observed only on or after January 1, 2005, the choice of the time period is to ensure a fair comparison between trials using and not using biomarkers. SE denotes standard error

		Biomarkers									
		Phase 1 to Phase 2			Phase 2 to Phase 3			Phase 3 to approval			Overall
Therapeutic group		Total phase transitions	POS <sub>1,2</sub> , %	(SE, %)	Total phase transitions	POS <sub>2,3</sub> , %	SE, %	Total phase transitions	POS <sub>3,APP</sub> , %	(SE, %)	POS, % (SE, %)
Oncology	No biomarker	9349	28.0	(0.5)	4773	17.4	(0.5)	1159	33.6	(1.4)	1.6 (0.2)
	With biomarker	1136	43.5	(1.5)	742	38.8	(1.8)	77	63.6	(5.5)	10.7 (1.9)
	All	10485	29.7	(0.4)	5515	20.3	(0.5)	1236	35.5	(1.4)	2.1 (0.2)
Metabolic/ endocrinology	No biomarker	1532	44.5	(1.3)	1438	33.9	(1.2)	1086	52.0	(1.5)	7.9 (0.8)
	With biomarker	7	57.1	(18.7)	2	50.0	(35.4)	15	20.0	(10.3)	5.7 (13.9)
	All	1539	44.6	(1.3)	1440	34.0	(1.2)	1101	51.6	(1.5)	7.8 (0.8)
Cardiovascular	No biomarker	1241	39.6	(1.4)	1027	37.9	(1.5)	962	62.2	(1.6)	9.3 (1.0)
	With biomarker	7	85.7	(13.2)	5	100.0	(0.0)	2	100.0	(0.0)	85.7 (13.2)
	All	1248	39.9	(1.4)	1032	38.2	(1.5)	964	62.2	(1.6)	9.5 (1.0)
CNS	No biomarker	2181	40.4	(1.1)	2050	30.2	(1.0)	1141	51.1	(1.5)	6.2 (0.6)
	With biomarker	42	54.8	(7.7)	42	28.6	(7.0)	15	53.3	(12.9)	8.3 (6.4)
	All	2223	40.7	(1.0)	2092	30.2	(1.0)	1156	51.1	(1.5)	6.3 (0.6)
Autoimmune/ inflammation	No biomarker	2506	38.9	(1.0)	2106	25.4	(0.9)	964	63.7	(1.5)	6.3 (0.6)
	With biomarker	9	55.6	(16.6)	14	35.7	(12.8)	5	60.0	(21.9)	11.9 (16.8)
	All	2515	39.0	(1.0)	2120	25.5	(0.9)	969	63.7	(1.5)	6.3 (0.6)
Genitourinary	No biomarker	359	34.3	(2.5)	287	28.9	(2.7)	212	66.5	(3.2)	6.6 (1.5)
	With biomarker	5	80.0	(17.9)	0	N.A.	N.A.	0	N.A.	N.A.	N.A. N.A.
	All	364	34.9	(2.5)	287	28.9	(2.7)	212	66.5	(3.2)	6.7 (1.5)
Infectious disease	No biomarker	1961	39.7	(1.1)	1453	34.7	(1.2)	1069	75.1	(1.3)	10.4 (0.9)
	With biomarker	6	66.7	(19.2)	27	44.4	(9.6)	9	100.0	(0.0)	29.6 (16.8)
	All	1967	39.8	(1.1)	1480	34.9	(1.2)	1078	75.3	(1.3)	10.5 (0.9)
Ophthalmology	No biomarker	180	52.2	(3.7)	274	34.7	(2.9)	207	74.9	(3.0)	13.6 (2.8)
	With biomarker	1	0.0	(0.0)	3	33.3	(27.2)	0	N.A.	N.A.	N.A. N.A.
	All	181	51.9	(3.7)	277	34.7	(2.9)	207	74.9	(3.0)	13.5 (2.8)
Vaccines (infectious disease)	No biomarker	733	40.8	(1.8)	761	32.9	(1.7)	609	85.4	(1.4)	11.4 (1.3)
	With biomarker	0	N.A.	N.A.	5	0.0	(0.0)	0	N.A.	N.A.	N.A. N.A.
	All	733	40.8	(1.8)	766	32.6	(1.7)	609	85.4	(1.4)	11.4 (1.3)
Overall	No biomarker	20042	34.7	(0.3)	14169	26.8	(0.4)	7409	59.0	(0.6)	5.5 (0.2)
	With biomarker	1213	44.5	(1.4)	840	38.6	(1.7)	123	60.2	(4.4)	10.3 (1.6)
	All	21255	35.2	(0.3)	15009	27.4	(0.4)	7532	59.0	(0.6)	5.7 (0.2)

<https://academic.oup.com/biostatistics/article/20/2/273/4817524>

From Wong et al. Biostatistics 2018

# Pharma is still the one of the biggest R&D investors



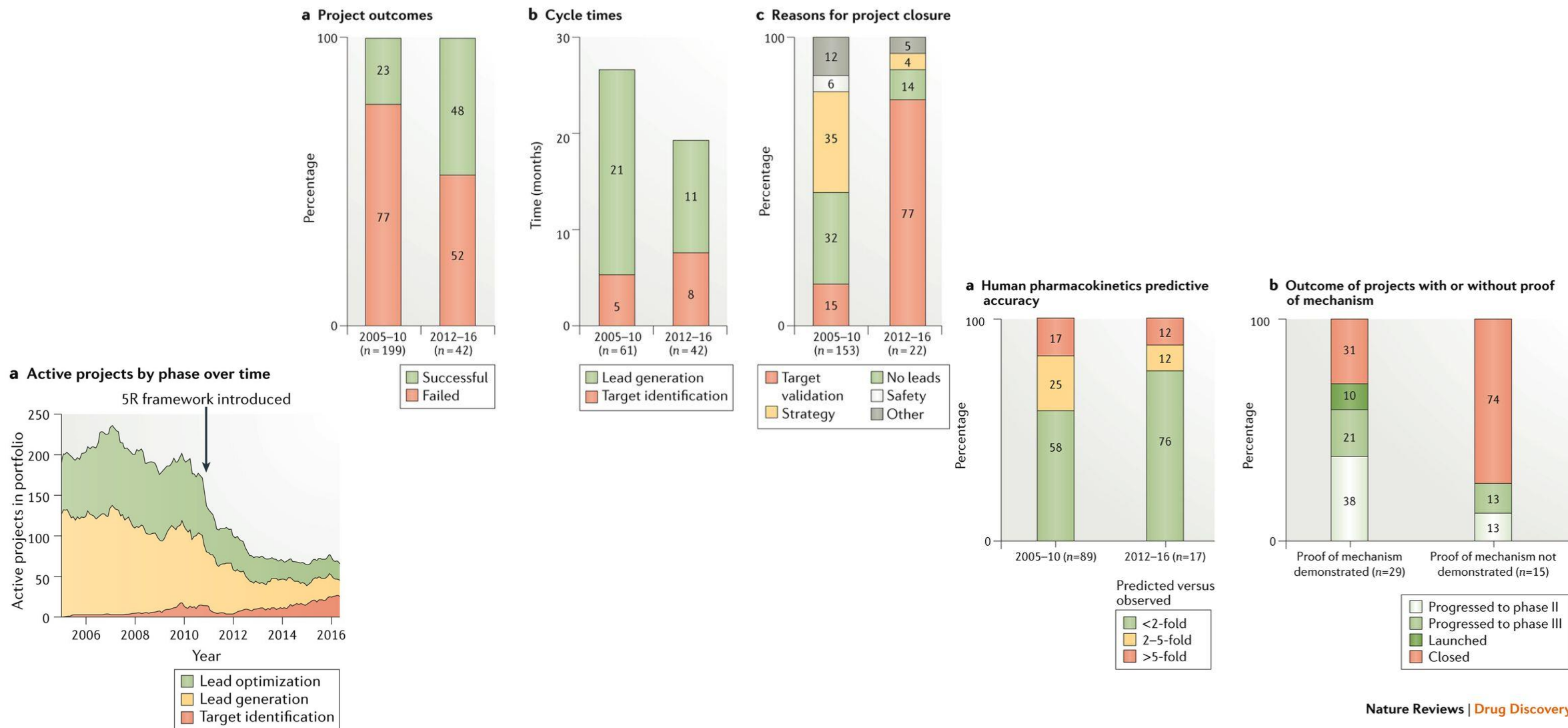
In 2017:

Top 20 Pharma spent \$93.7 Billion on R&D  
while public spending only reached: \$51.2 Billion



We have been  
improving for more  
than a decade, are  
we doing better?

# AstraZeneca, after implementing the 5Rs

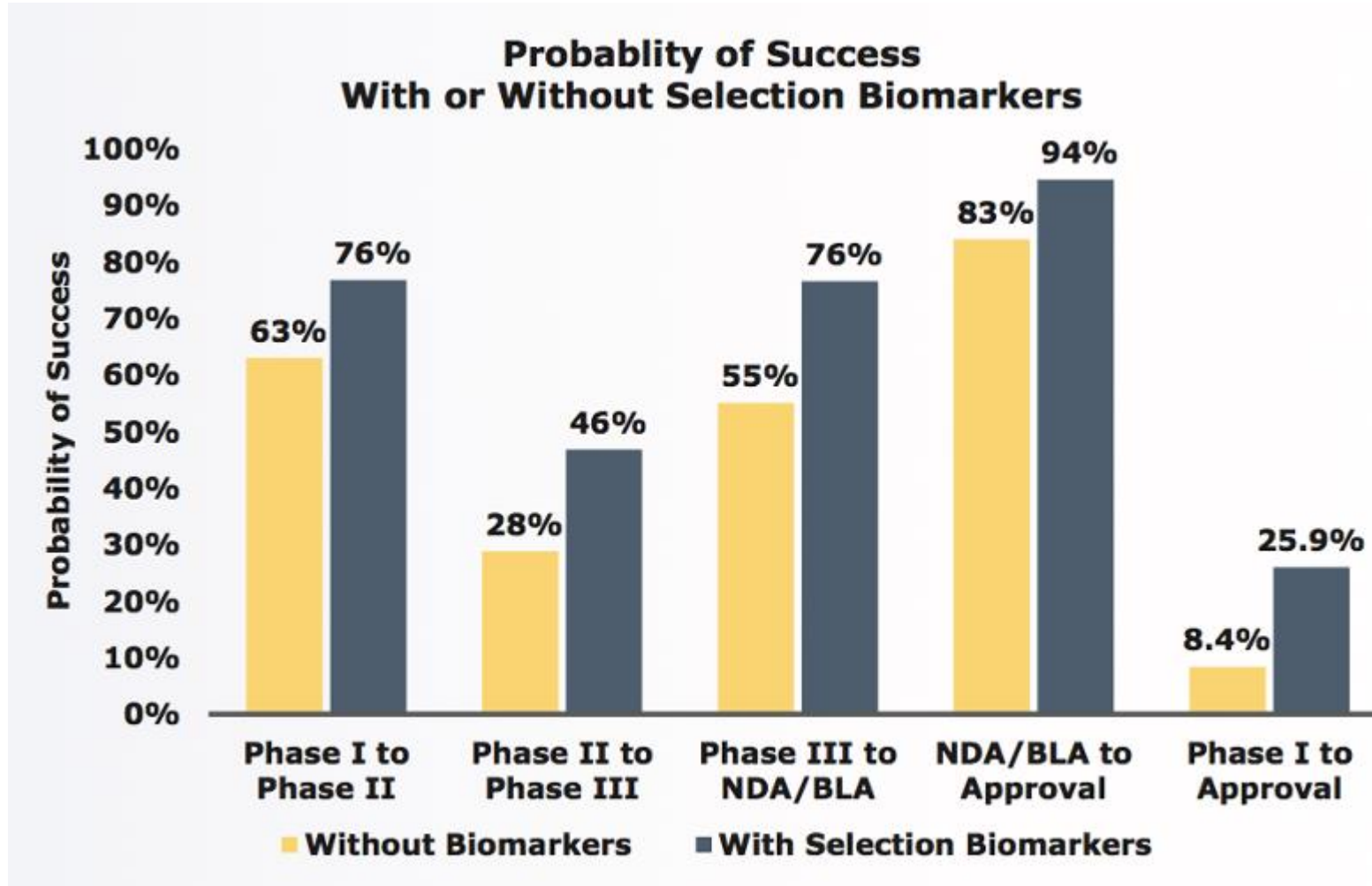


From Morgan et al, Nature Reviews Drug Discovery 2018

<https://www.nature.com/articles/nrd.2017.244>



# Biomarkers increase the %POS



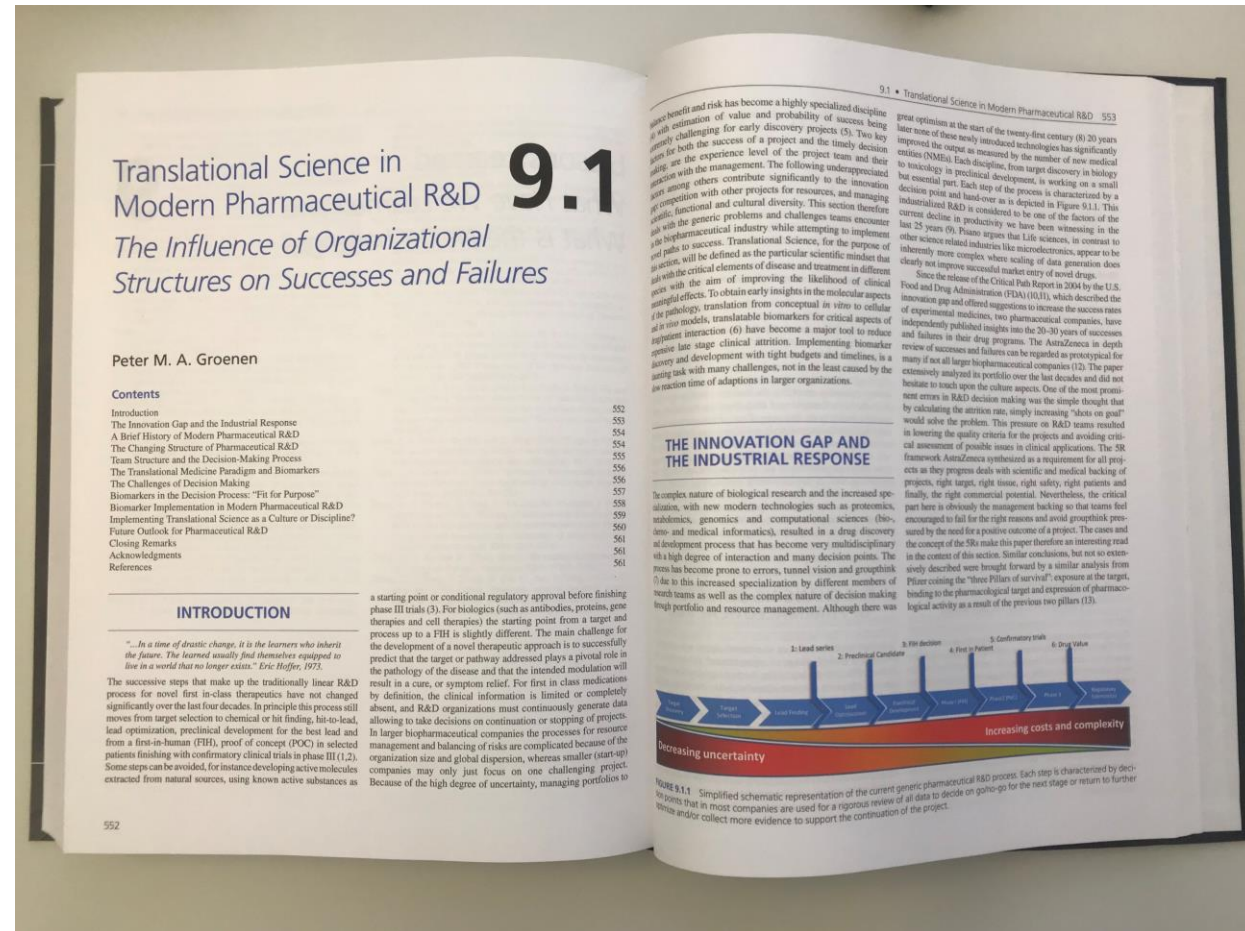
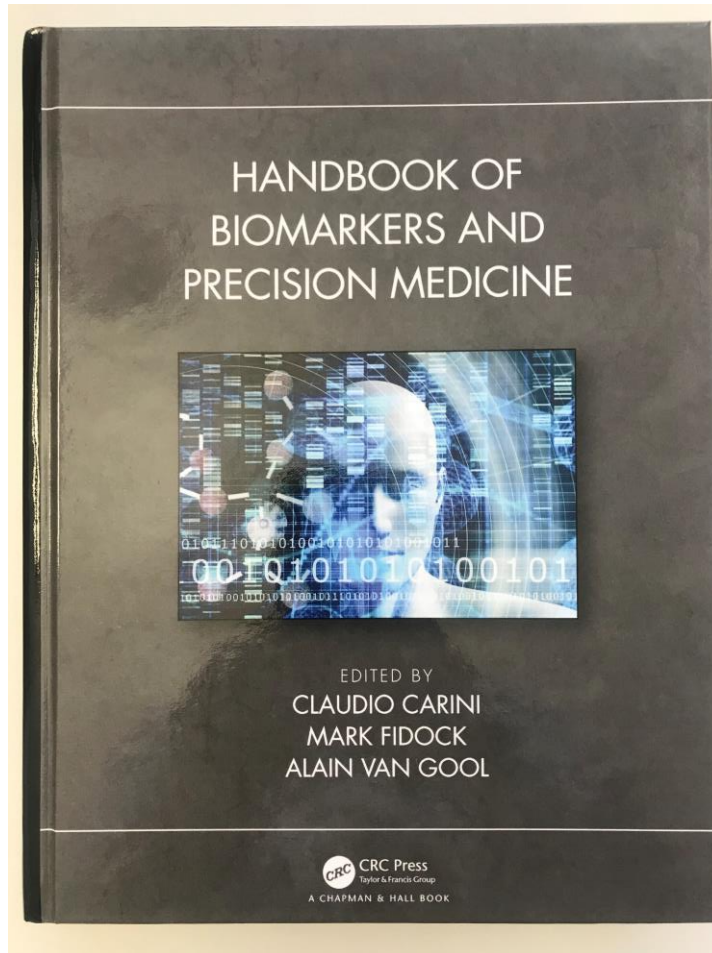
Source : Health Decisions CRO+ reports 2015

# Summary

- Limited biomarker clinical uptake is symptomatic for our current modern biomedical research
- Translation of biomarker research into clinical benefits needs radical improvement on several levels
- Specifically the pharma industry is also in great need for novel biomarkers in the path from discovery to clinical benefit
- Robust scientific methodology does not solve all problems, but is key to success and to avoid waste
- Remember: an invention does not equal innovation!
- Biomedical innovation is team work along the healthcare chain and between all parties, profit and non-profit: let's avoid polarizing discussions.

# Cultural Changes Needed....

Please read this book!



# Outlook for the Future





# The digital revolution in health care

**digital biomarkers**

Digit Biomark 2019:392–402  
DOI: 10.1159/000502000  
Received: May 2, 2019  
Accepted after revision: July 8, 2019  
Published online: August 16, 2019

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**Viewpoint – Review Article**

## Traditional and Digital Biomarkers: Two Worlds Apart?

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Giovanni Nisato<sup>e</sup> Marc Zinggeler<sup>f</sup> Noé Brasier<sup>g</sup> Katja Baerenfaller<sup>h</sup>  
Thomas Brenzikofer<sup>d</sup> Laurenz Baltzer<sup>i</sup> Christian Vogler<sup>j</sup> Leo Gschwind<sup>k</sup>  
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**Keywords**  
Traditional biomarkers · Digital biomarkers · Semantics · Biomarker classification · Digital health

**Abstract**  
The identification and application of biomarkers in the clinical and medical fields has an enormous impact on society. The increase of digital devices and the rise in popularity of health-related mobile apps has produced a new trove of biomarkers in large, diverse, and complex data. However, the unclear definition of digital biomarkers, population groups, and their intersection with traditional biomarkers hinders their discovery and validation. We have identified current issues in the field of digital biomarkers and put forth suggestions to address them during the DayOne Workshop with participants from academia and industry. We have found similarities and differences between traditional and digital biomarkers in order to synchronize semantics, define unique features, review current regulatory procedures, and describe novel applications that enable precision medicine.

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- Imagine when compliance and dosing could be followed real time!

