

Impact of ADME/TOX in Drug Discovery: Past, Present and Future

Li Di

Wyeth Research

Princeton, NJ

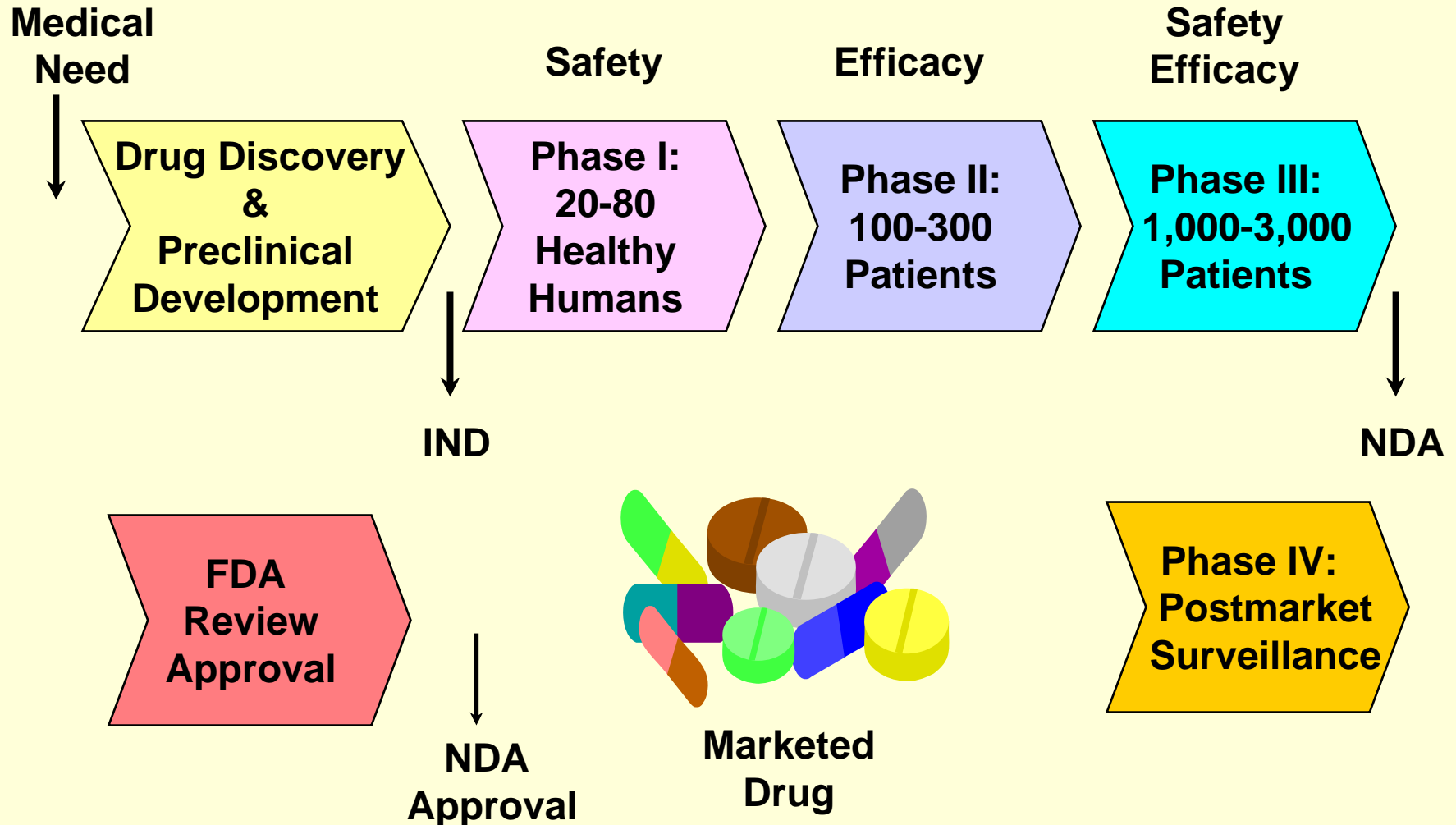
CoSMoS

Chapel Hill, NC,

July 30 - August 1, 2007

Wyeth
Research

Stages of Drug Discovery and Development Process



Drug Discovery and Development Cascade

High Throughput Screening	1,000,000
HTS Hits	2,000
HTS Actives	1,000
Discovery Program	50
Drug Candidate	10
Drug	1

12 years, \$800 million, 10% Success Rate

M.M. Hann & T. I. Oprea, Curr Opin Chem Bio, 2004, 8(3), 255-263

M. Dickson, J. P. Gagnon, Nature Rev Drug Disc. 2004, 3, 417 - 429

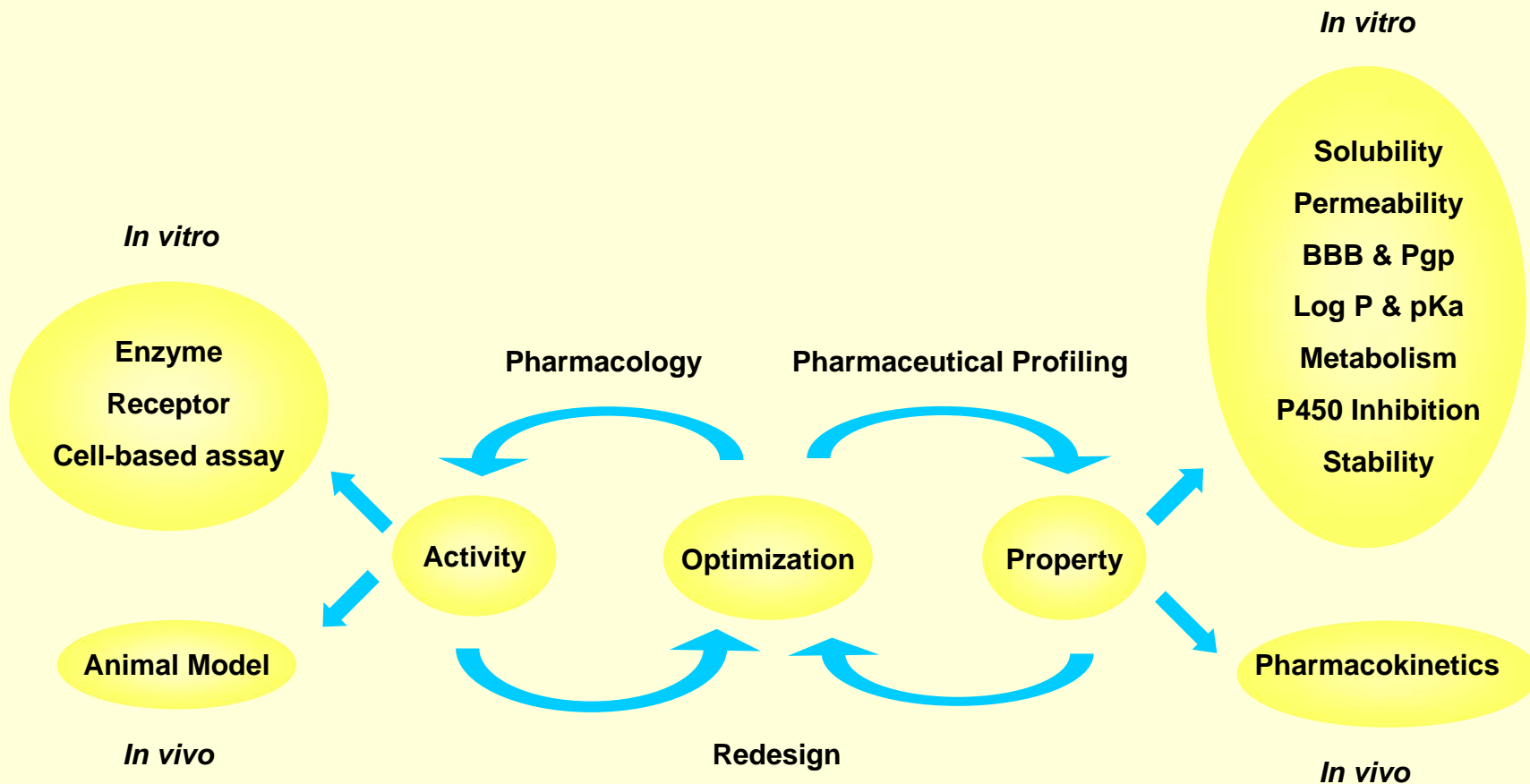
Importance of Pharmaceutical Profiling

<u>Reason for Failure</u>	<u>Percentage</u>
Toxicity	22 %
Lack of Efficacy	31 %
Market Reasons	6 %
Poor Biopharmaceutical Property	41%

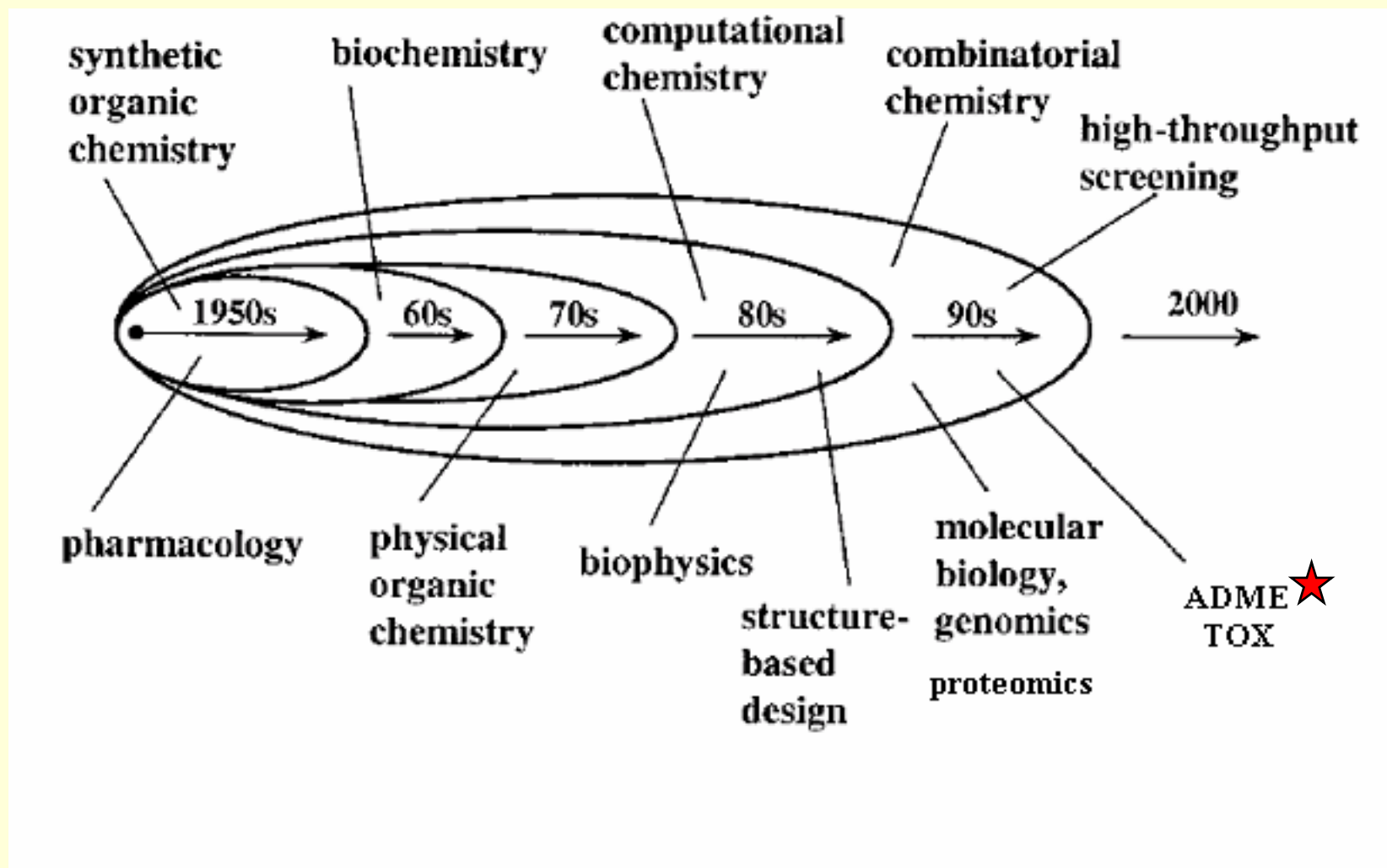
ADME-TOX Properties

- **A**bsorption
- **D**istribution
- **M**etabolism
- **E**xcretion
- **T**oxicity

Successful Drug = Potency + Properties

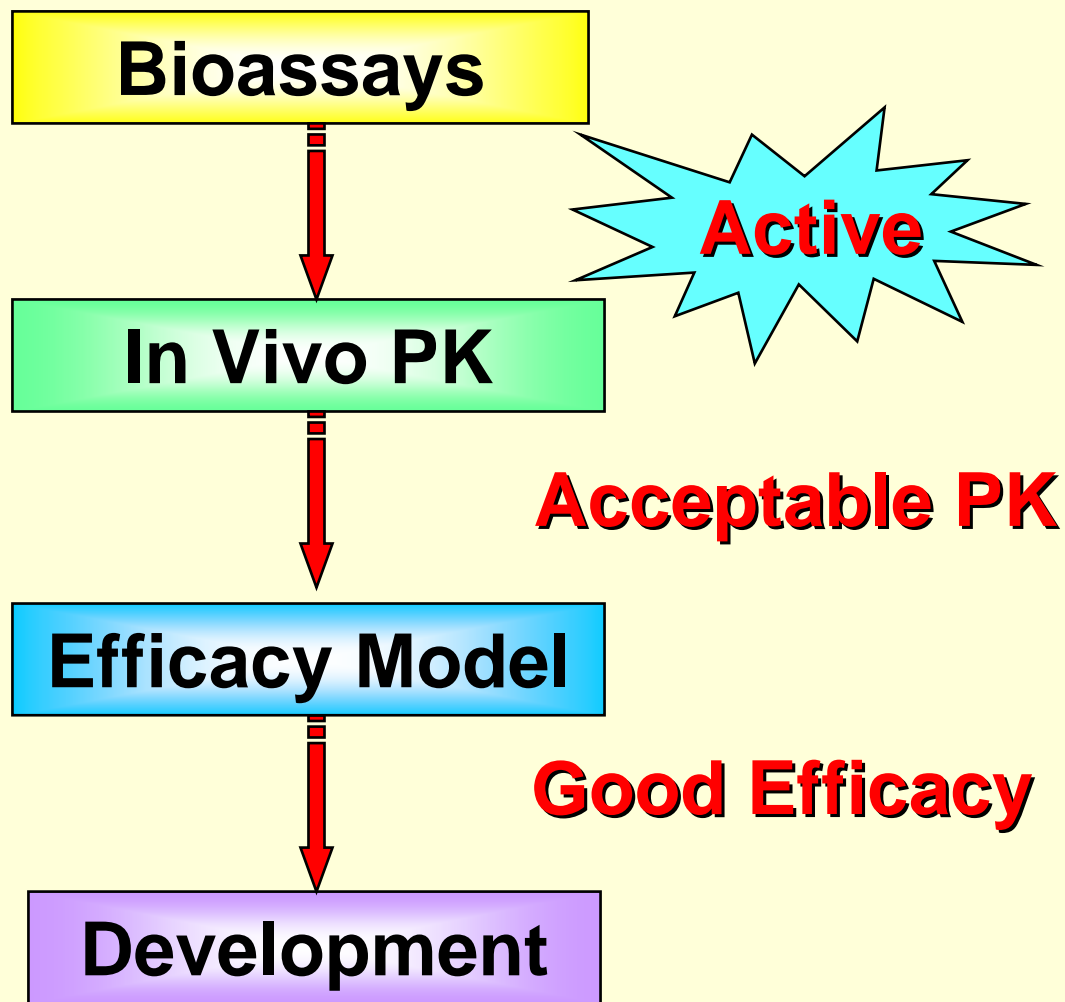


ADME/TOX Screening: New Tool for Drug Discovery



Modified from Han van de Waterbeemd, et al, *J. Med. Chem.*, 1313-1333, 44(9), 2001

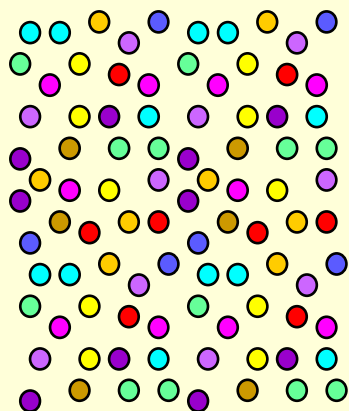
Screening Paradigm in the Past



Activity was the sole driver for discovery programs

Past: Activity-Driven Screening

Compounds



Bioassays



Very Active

$K_i = 7 \text{ nM} !$

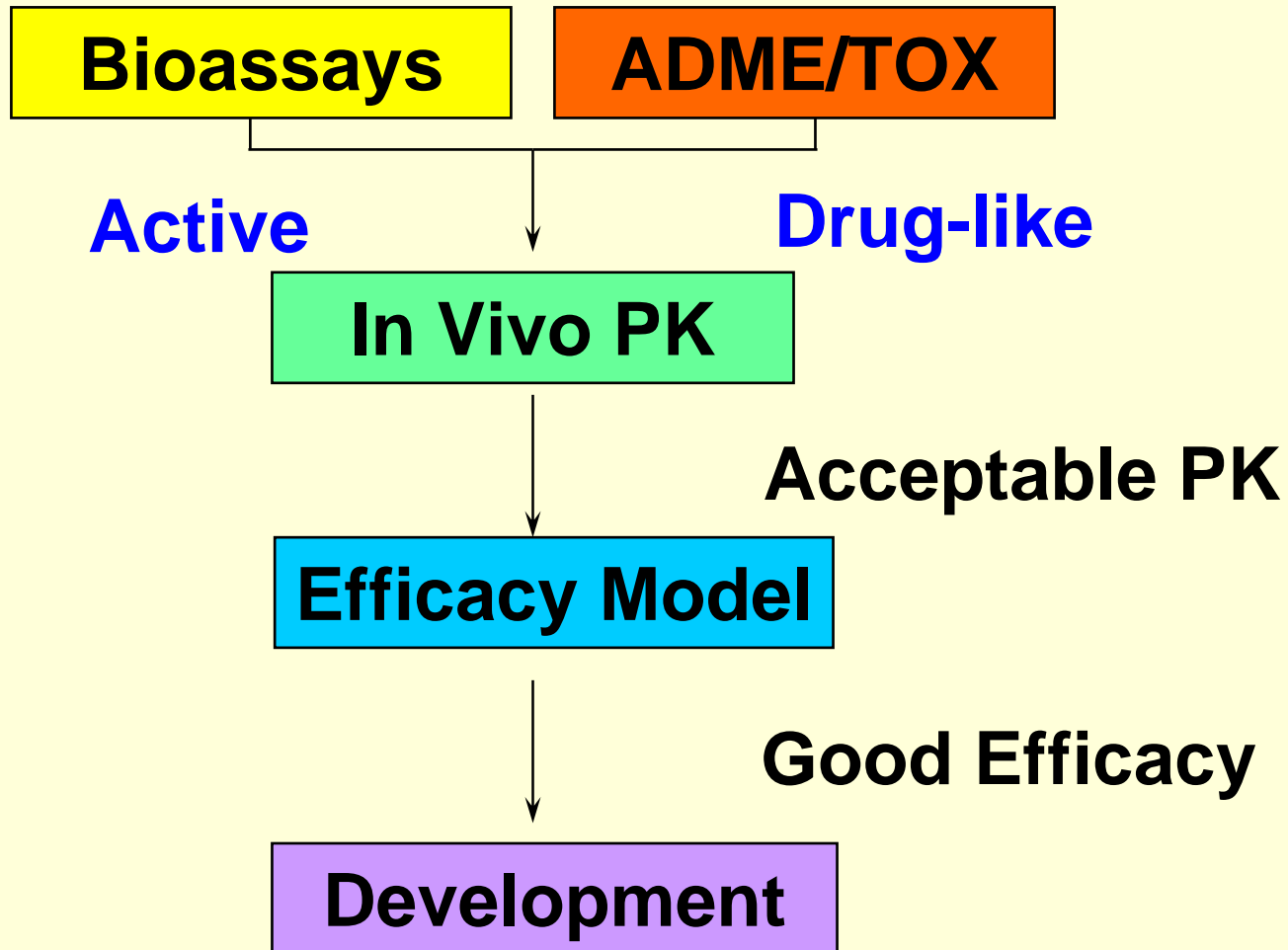


$F\% = 0$

Inactive
Terminated

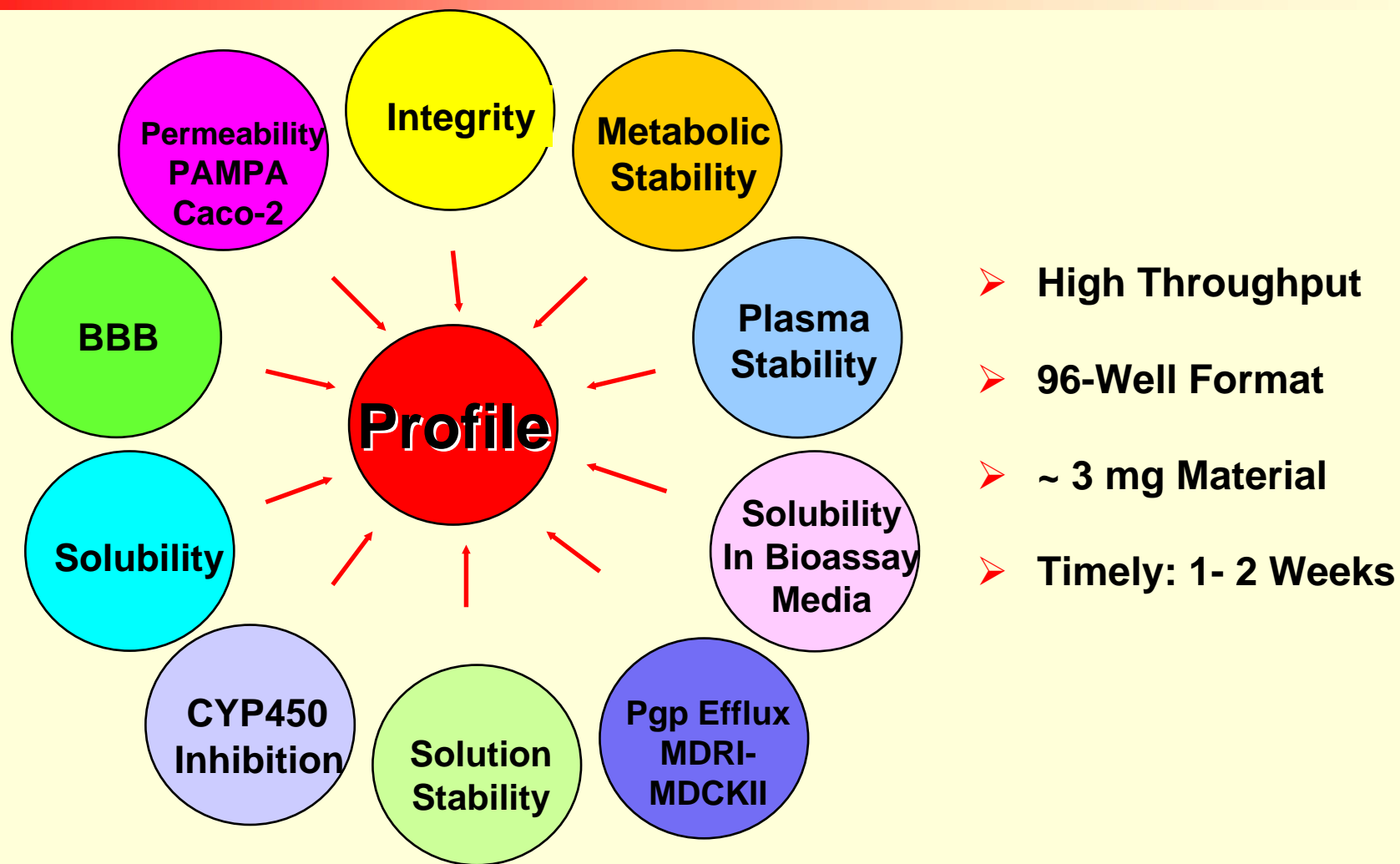
Long Cycle Time
High Cost
Low Success Rate

Present Drug Discovery Screening Paradigm



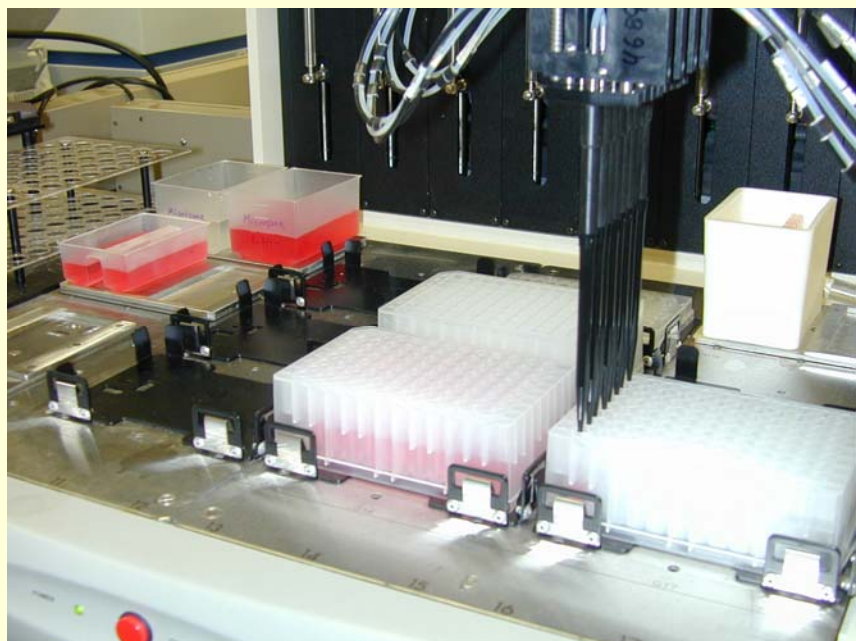
ADME/TOX: Advancement Criteria

Wyeth Pharmaceutical Profiling Assays



Modified from Edward H. Kerns, J. Pharm. Sci., 2000, 90(11), 1838-1858

Robotic Sample Preparation



Packard Robot
Microsomal Stability
CYP450 Inhibition



Tecan Robot
PAMPA, BBB
Solubility

High Throughput Sample Analysis



LC-MS-MS

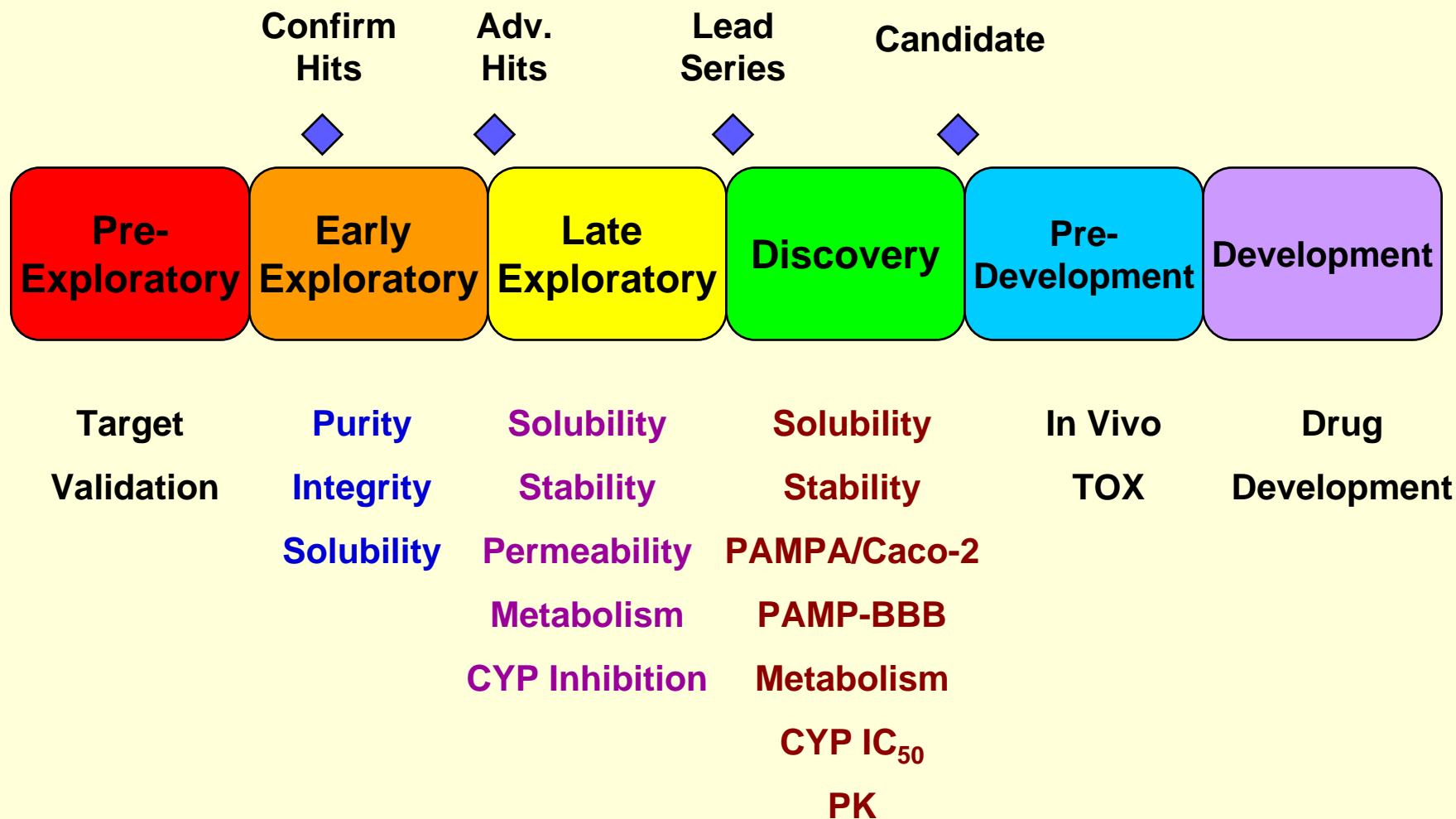


Fluorescent Plate Reader



UV/Vis Plate Reader

ADME/TOX Screening in Drug Discovery



Ensure Quality of Development Candidates

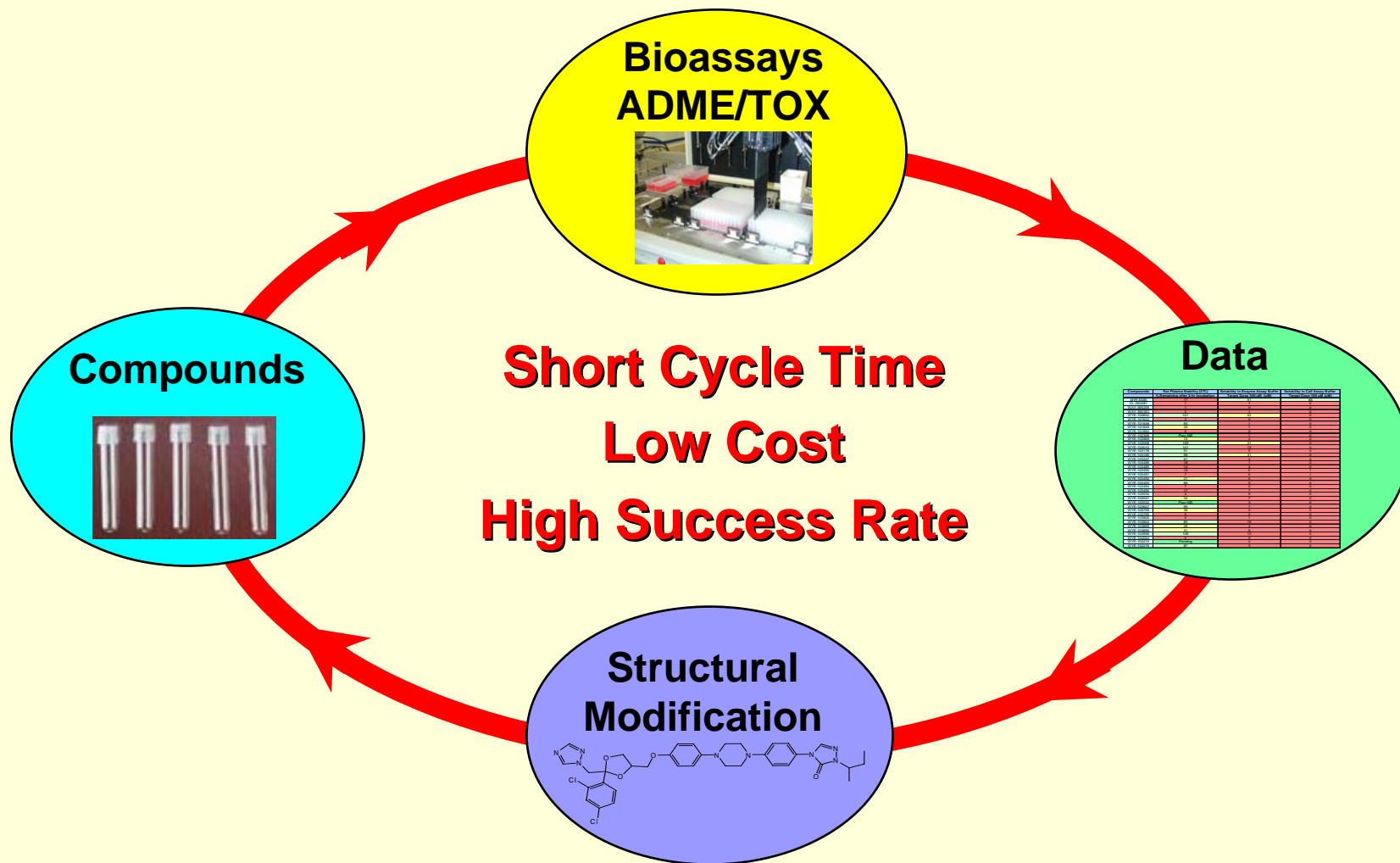
Industry Development ADME/TOX Criteria

- **Solubility > 100 $\mu\text{g/mL}$**
- **Oral Bioavailability > 20%**
- **CYP IC_{50} > 10 μM or $\text{C}_{\text{max}} / \text{K}_i < 0.1$**
- **hERG $\text{IC}_{50} / \text{C}_{\text{max}} > 30$**

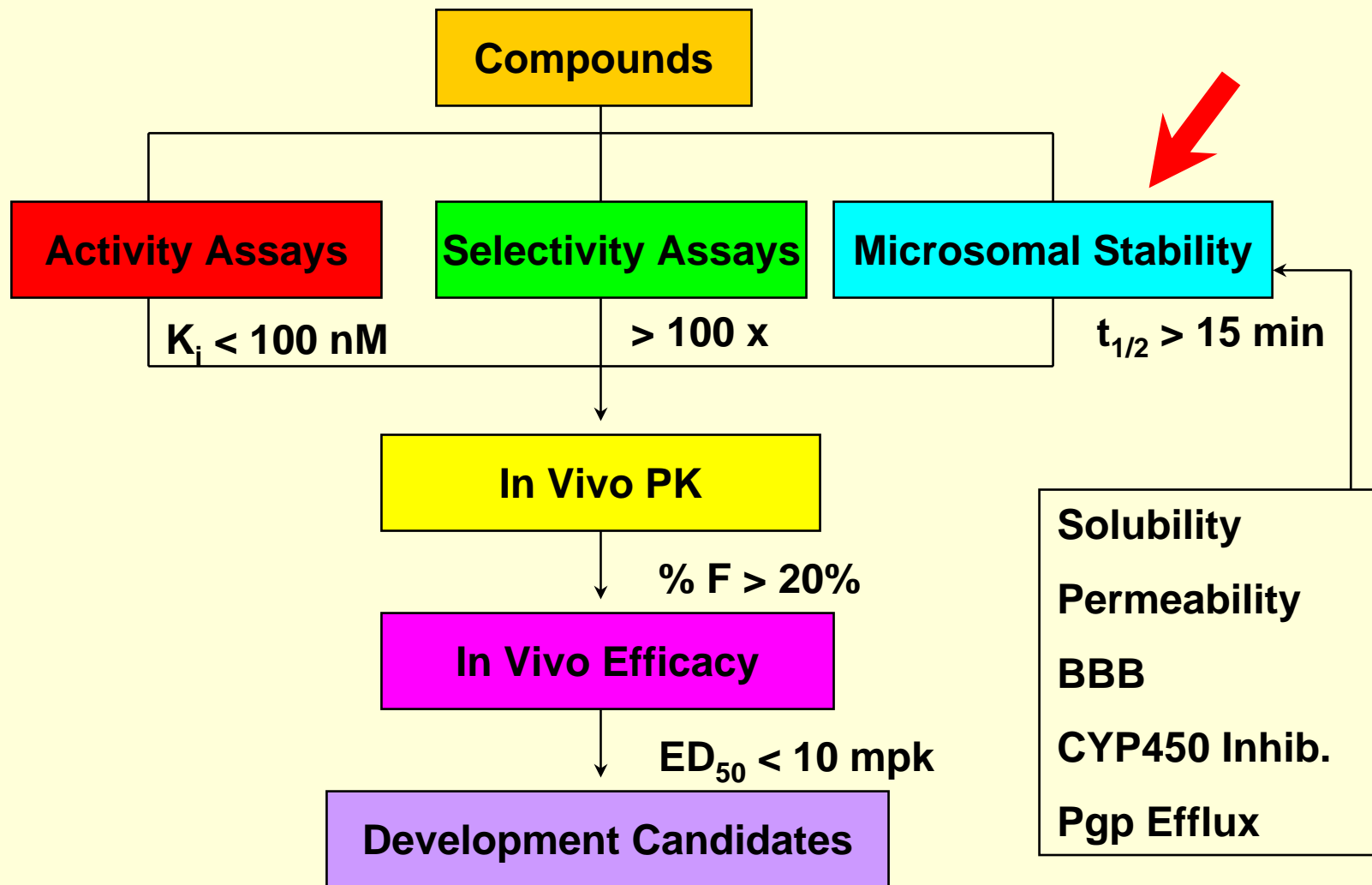
“You can’t manage it, if you can’t measure it. ”

**Robert R. Ruffolo
Wyeth R&D President**

Present: Activity and Property Driven Screening



Property Screening is Incorporated into Compound Selection



Selection criteria: Active, Selective and Stable

Critical Factors for Successful ADME/TOX Profiling

- **Throughput**
- **Speed**
- **Assay Design**
- **Timing**

Throughput: Past - Low; Present - High

Past

- 500 / year
- Limited information
- Not enough for SPR
- Not for selection
- Profile only actives

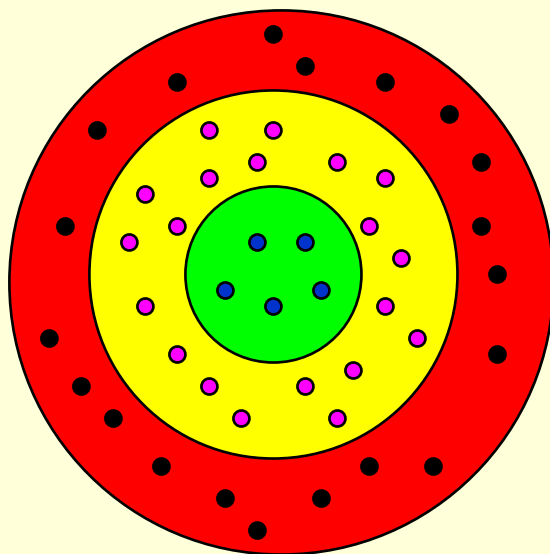
Present

- 50,000 / year
- Rich information
- Guide SPR
- Selection criteria
- Profile all

Past: Long Turnaround Time

Present: Short Turnaround Time

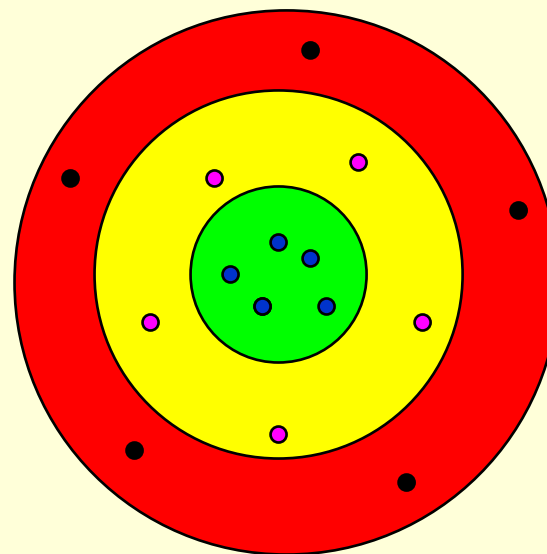
Turnaround Time = 1 month



40 Compounds

2 months

Turnaround Time = 1 week



10 Compounds

2 weeks

■ unstable

■ moderate

■ stable

Assumes Discovery Team makes 5 compounds / week

Past and Present Assay Design

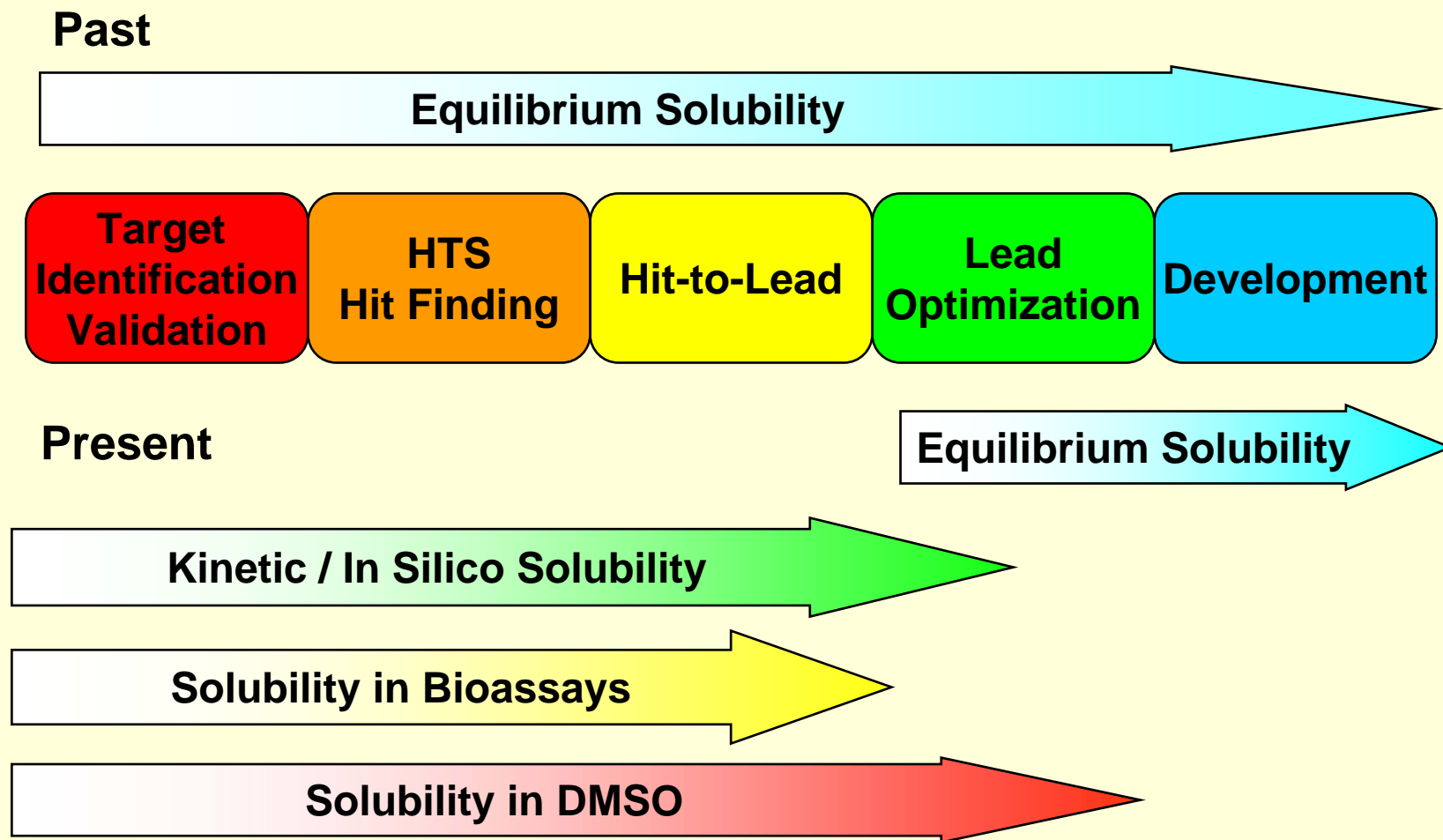
- **Past**

- ▶ One size fits all
- ▶ Multiple time points
- ▶ Multiple concentrations (IC50)

- **Present**

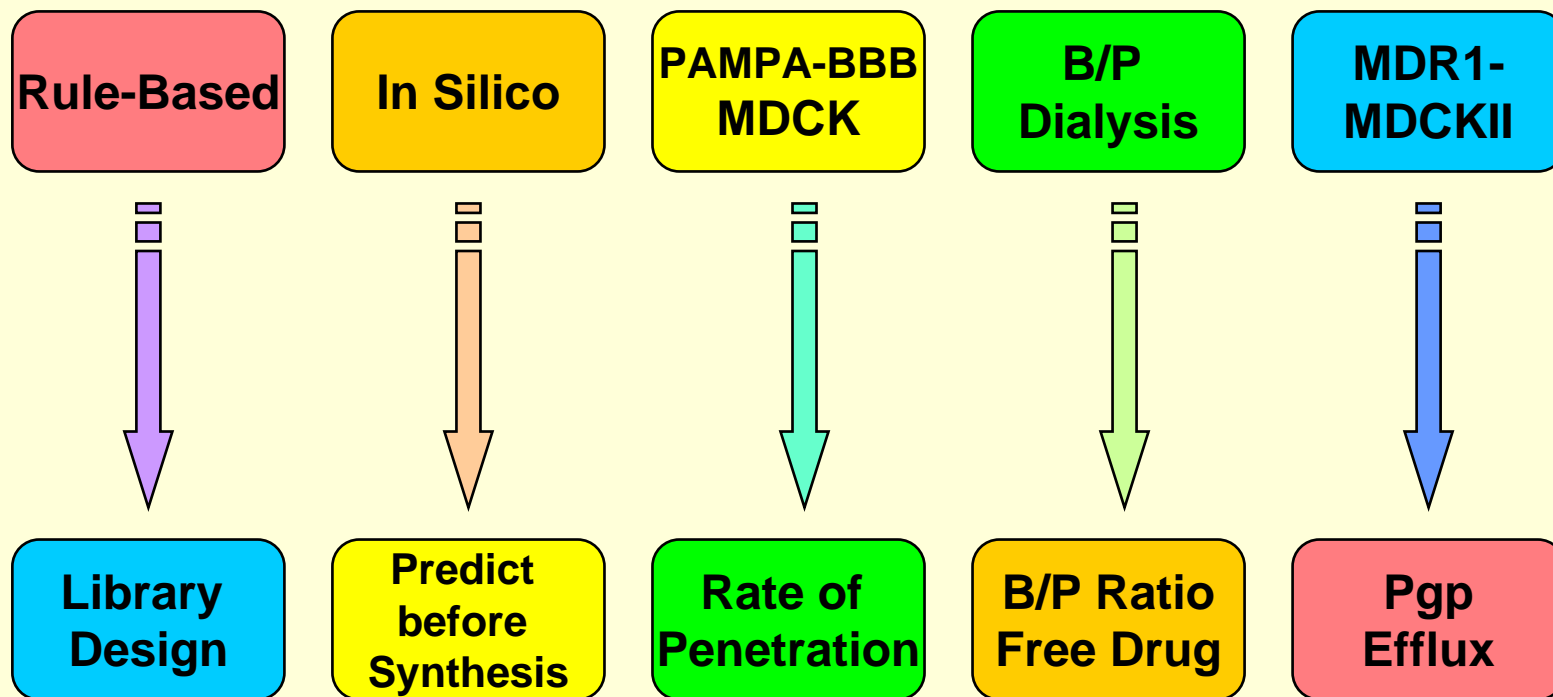
- ▶ Tier approach
- ▶ Custom assays

Solubility Assays in Drug Discovery



Different Assays at Different Stages for Various Purposes

Tier Approach to Blood-Brain Barrier



Determine Underlining Mechanisms

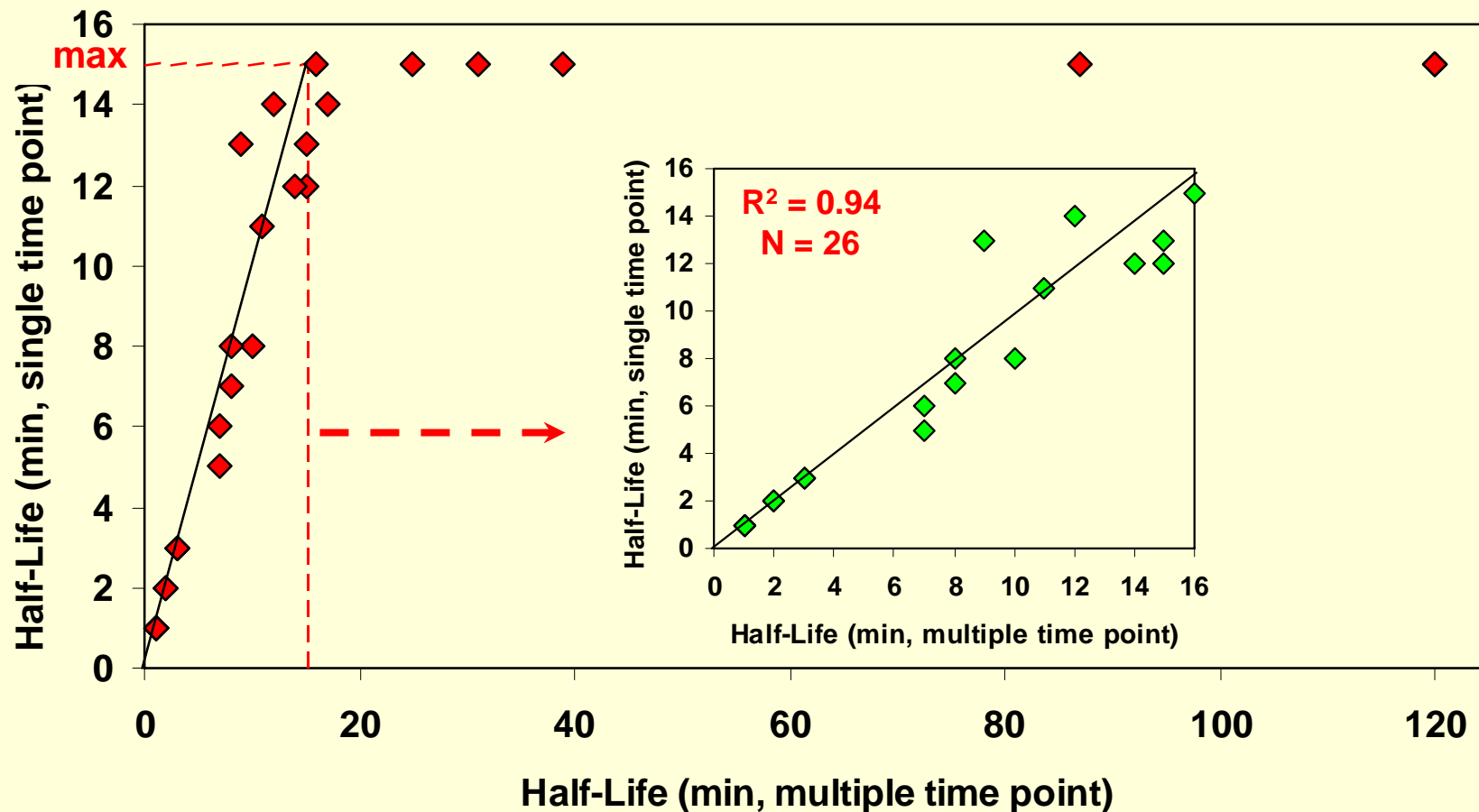
Microsomal Stability: Single Time Point vs. Multiple Time Point

Validation Data	t1/2 (mins)	t1/2 (mins)
Compounds	Wyeth	Literature*
Midazolam	3	4
Verapamil	6	10
Diltiazem	15	21
Zolpidem	> 30	44
Tenoxicam	> 30	38

* Scott Obach, Drug Met. Disp., 1999, 27(11), 1350-1359

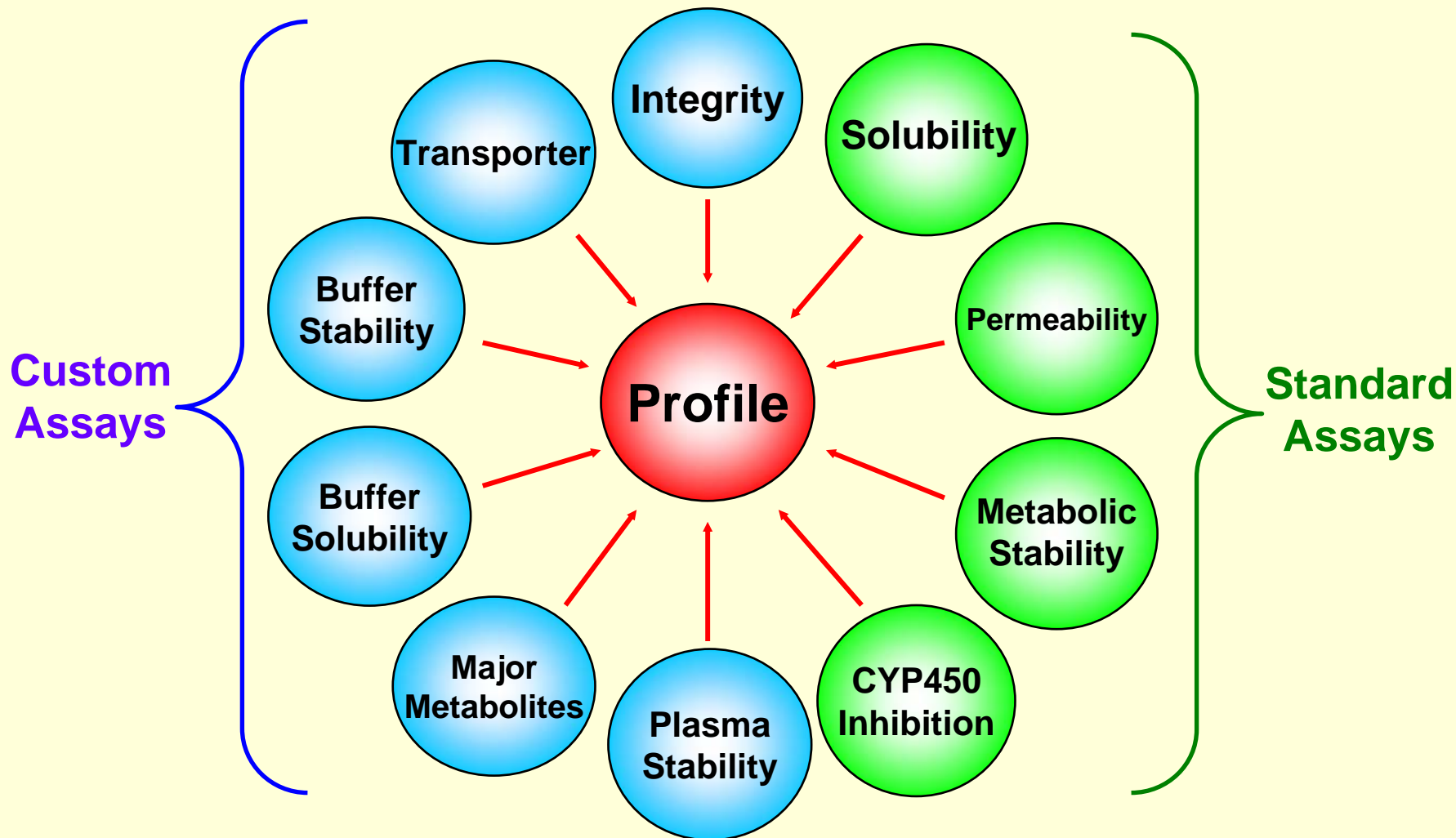
Good correlation between single time point and multiple time point

Correlation of Single Time Point and Multiple Time Point for Wyeth Compounds (5 min incubation)



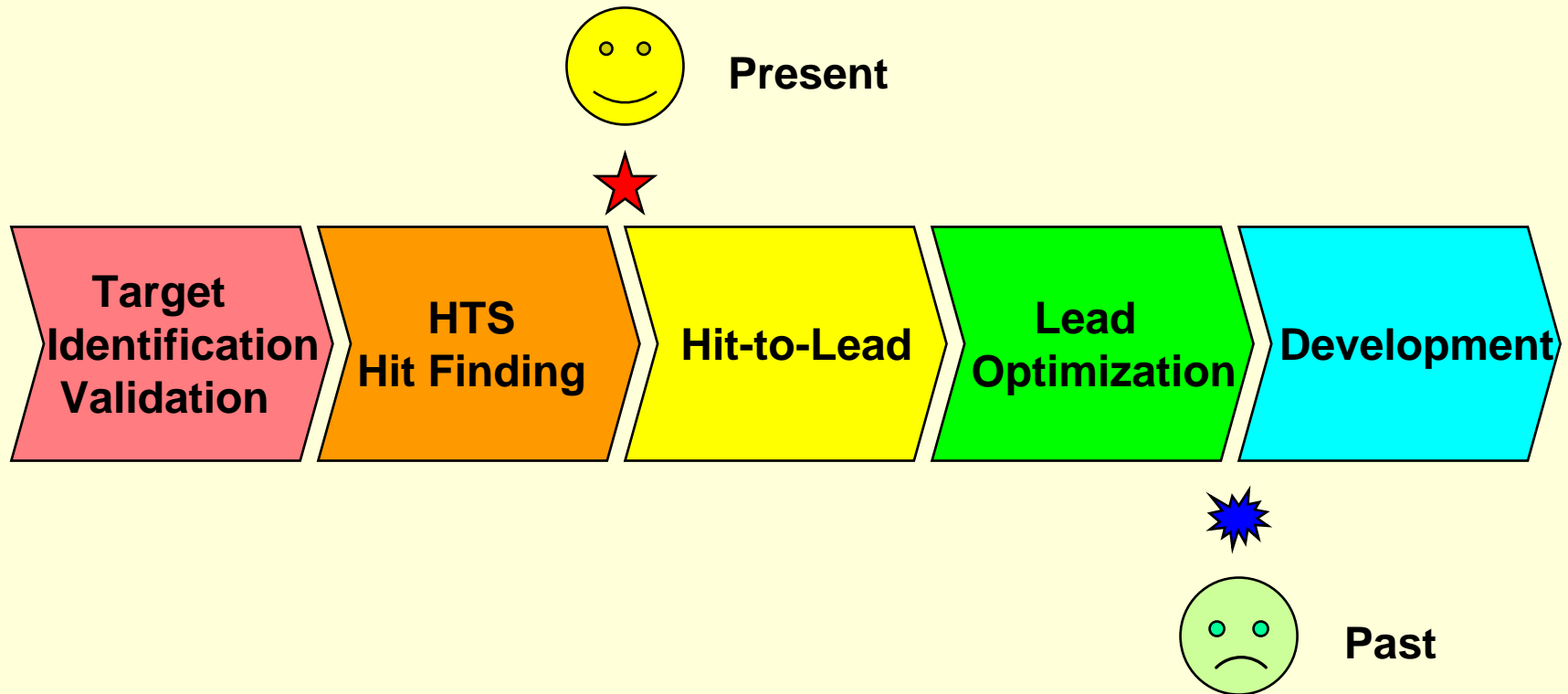
Li Di, Ed Kerns, et al, J. Pharm. Sci. 2004, 93, 1537-1544.

Standard and Custom Assays

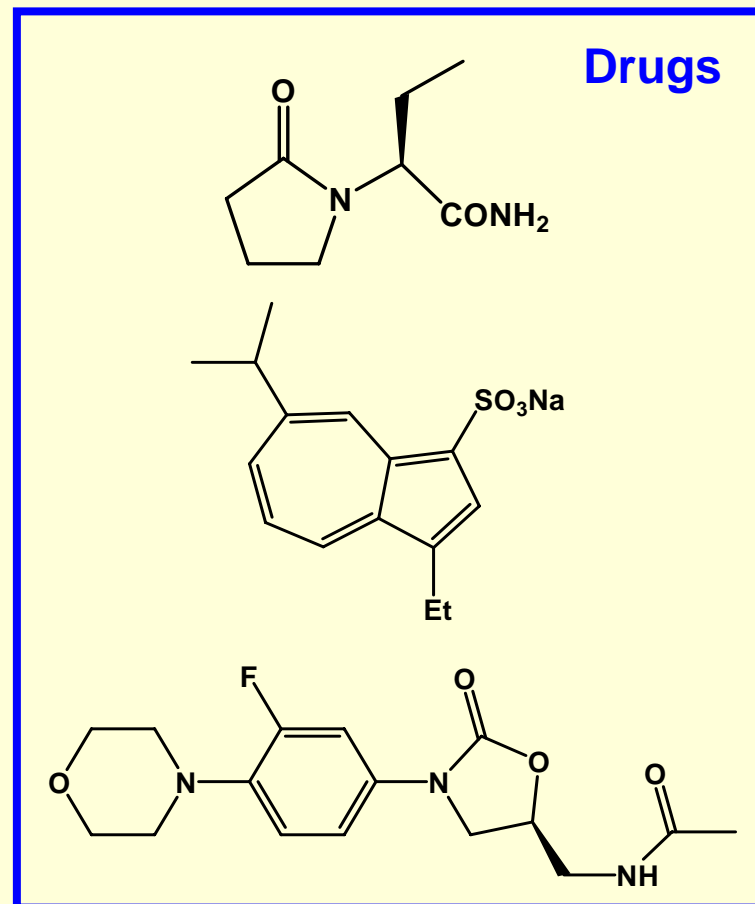
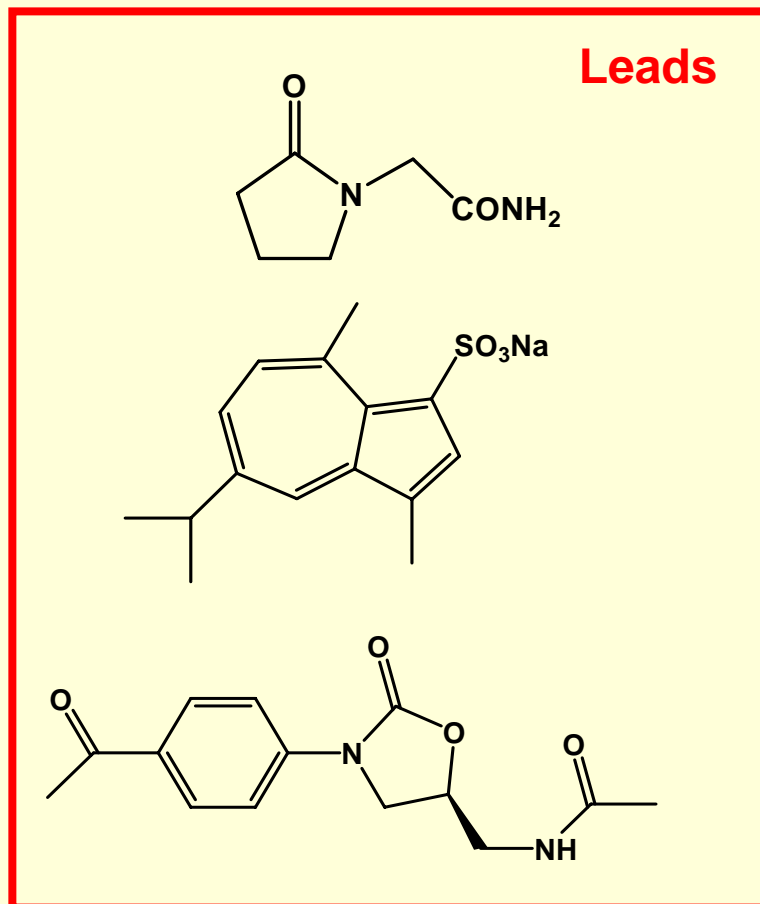


Informed Decisions

Timing: Past-Too Late; Present-Early



Drug Structure Similar to Lead Structure



Great opportunity to improve the properties is at exploratory stage

Hit-to-Lead: Lead Profiles

Project X	Hit	Lead 1	Lead 2	Desirable Profile
Enzyme IC 50 (nM)	542	161	198	< 1000
Cell Assay (nM)	30000	19047	4823	< 10000
Selectivity	> 100	> 100	> 100	> 10
MW	344	316	350	< 450
clog P	3.9	3.2	4.6	< 4.5
TPSA	62	75	62	< 80
Aq Sol. (ug/mL)	14	28	6	> 60
PAMPA(10^{-6} cm/s)	12	2.6	4.7	> 1.0
CYP3A4	-4	2	24	< 15
CYP2D6	-6	0	6	< 15
CYP2C9	9	13	8	< 15
RLM $t_{1/2}$ (min)	< 1	3	> 30	>15
MLM $t_{1/2}$ (min)	> 30	14	> 30	>15
HLM $t_{1/2}$ (min)	12	2	> 30	>15
hERG (%)	25	39	22	< 50%
Definable Series	Yes	Yes	Yes	Yes
Definable SAR	Yes	Yes	Yes	Yes
Novelty	Yes	Yes	Yes	Yes

Potency & Selectivity

Calculated Properties

ADME/TOX
Properties

Chemical Series
Novelty

Decision Making

Goals of ADME/TOX Screening from Hit-to-Lead

- **Solubility**

- ▶ Diagnose erroneous SAR
- ▶ Optimize bioassay protocol

- **Stability**

- ▶ Intrinsic liabilities of chemotypes
- ▶ Guide structural modification

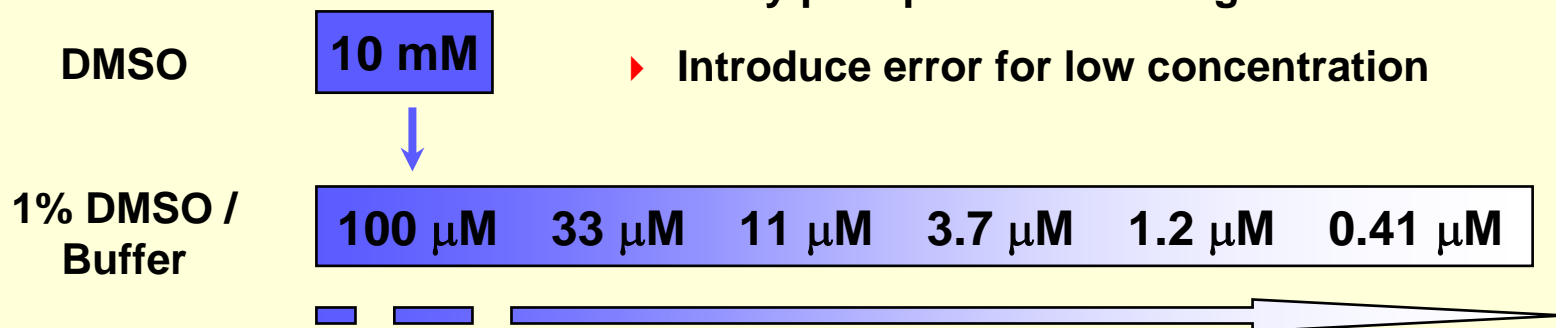
- **Metabolism, CYP Inhit., Permeability**

- ▶ Liability in core structure
- ▶ Areas to focus on in optimization

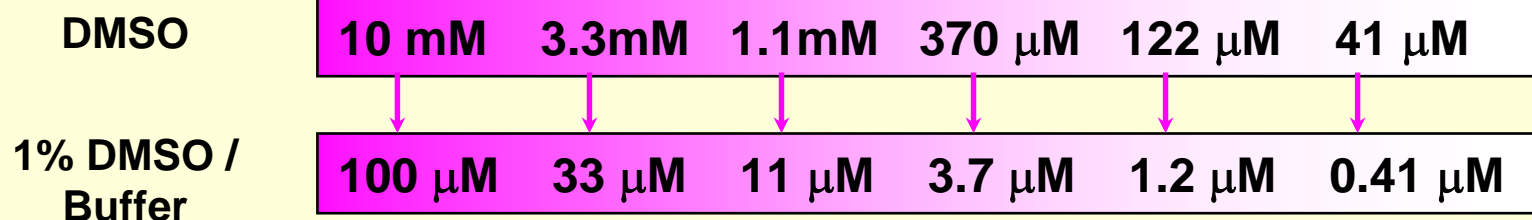
Strategies for Serial Dilution: Dilute in DMSO

From Aqueous

- ▶ Carry precipitation from high concentration
- ▶ Introduce error for low concentration



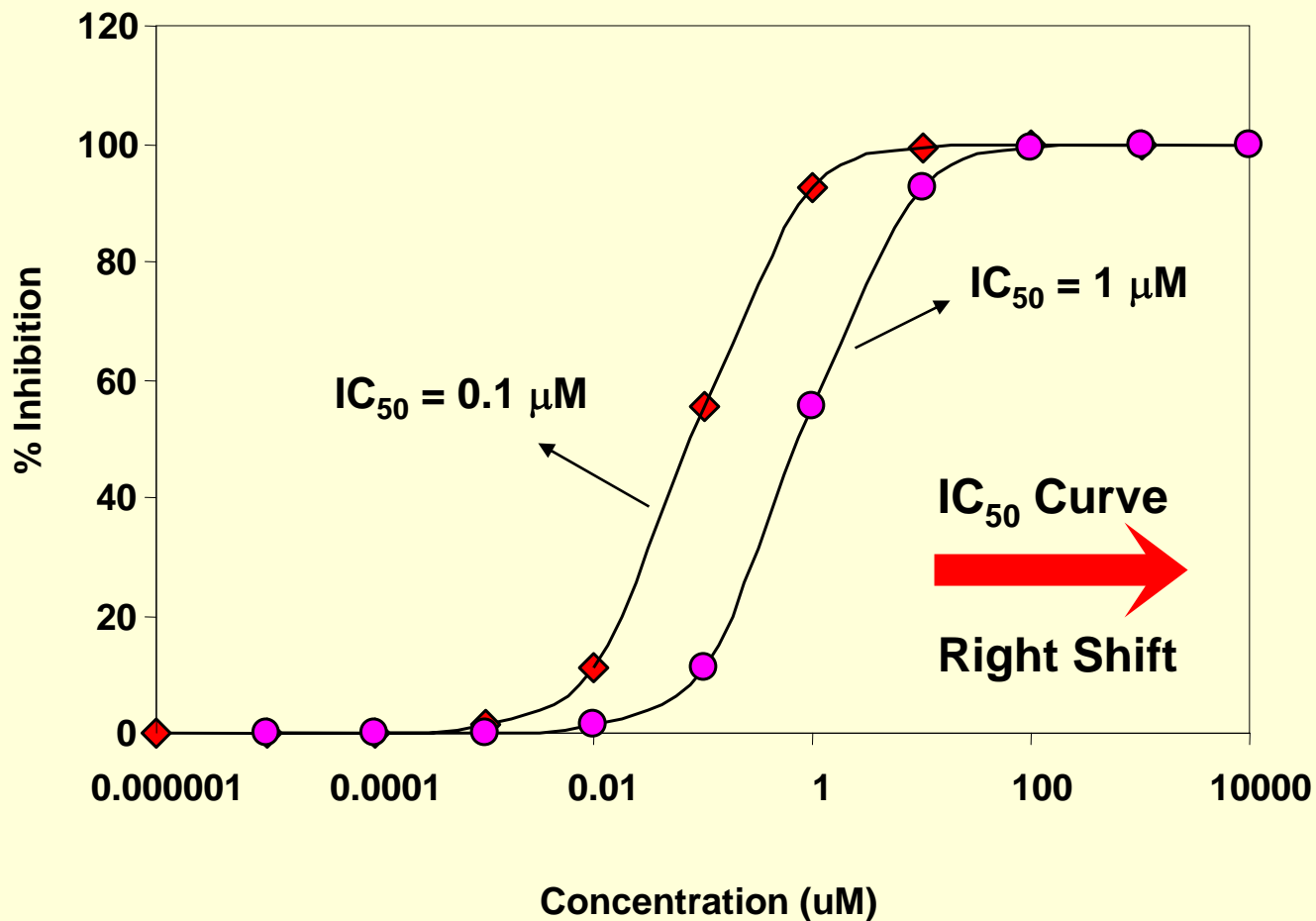
From DMSO



- ▶ High concentration might still precipitate, but will not affect low concentration

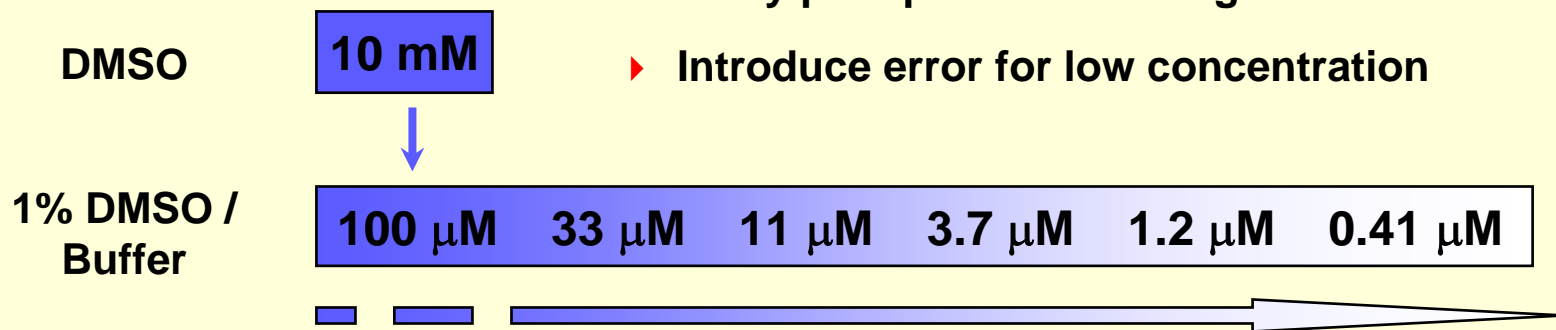
Right Shift of IC_{50} due to Low Solubility

When all the concentrations in assay buffers are lower:



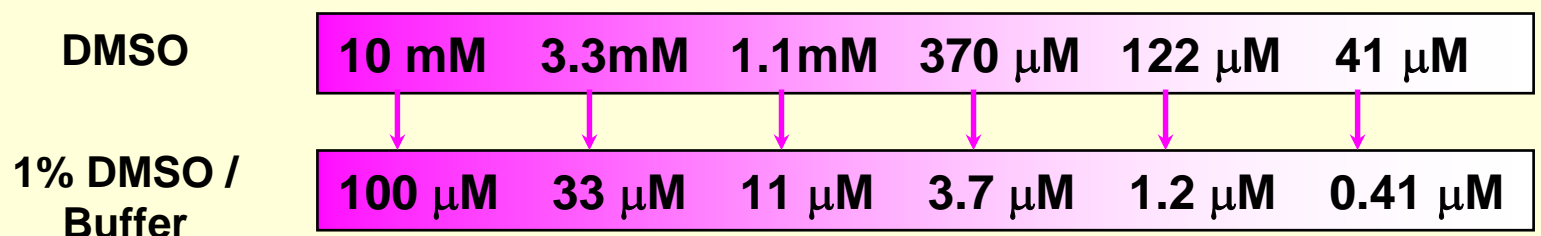
Strategies for Serial Dilution: Dilute in DMSO

From Aqueous



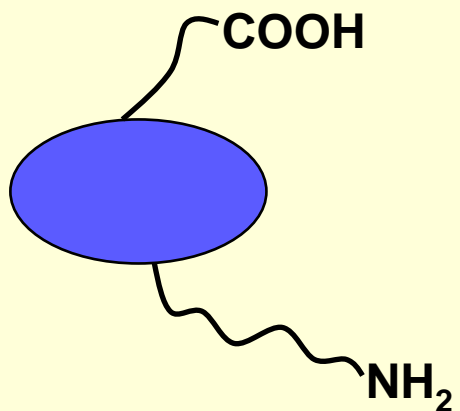
- ▶ Carry precipitation from high concentration
- ▶ Introduce error for low concentration

From DMSO



- ▶ High concentration might still precipitate, but will not affect low concentration

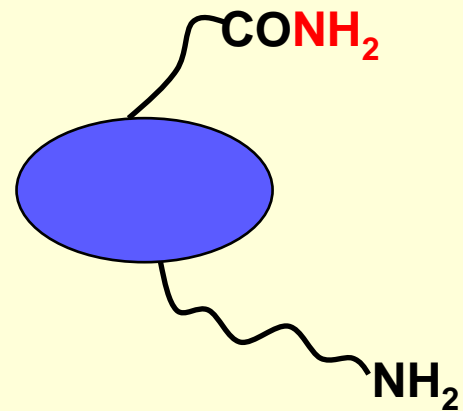
Stability Affects Assay Results



Low oral bioavailability

Poor Permeability

IV Administration

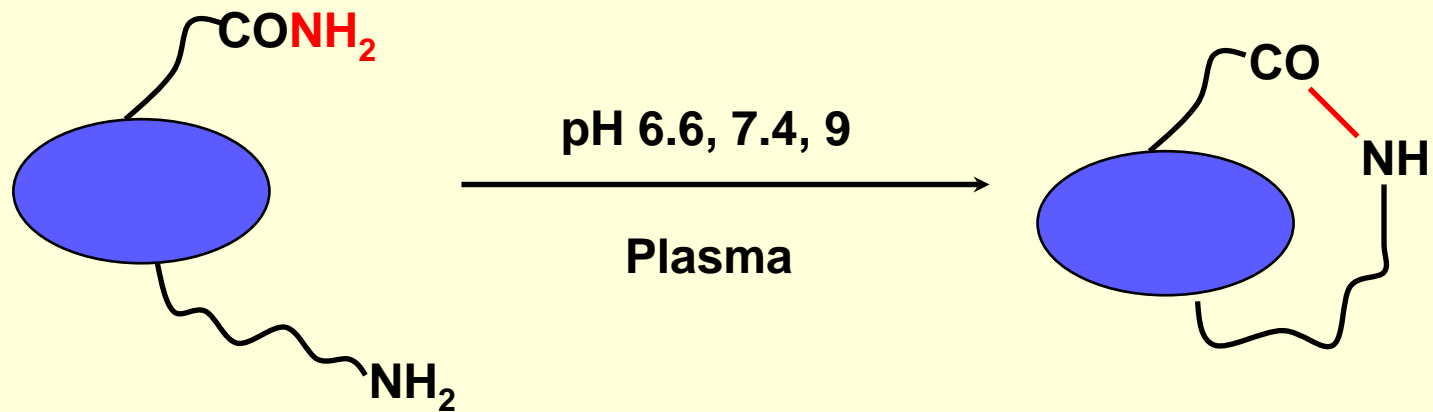


Very active in vitro

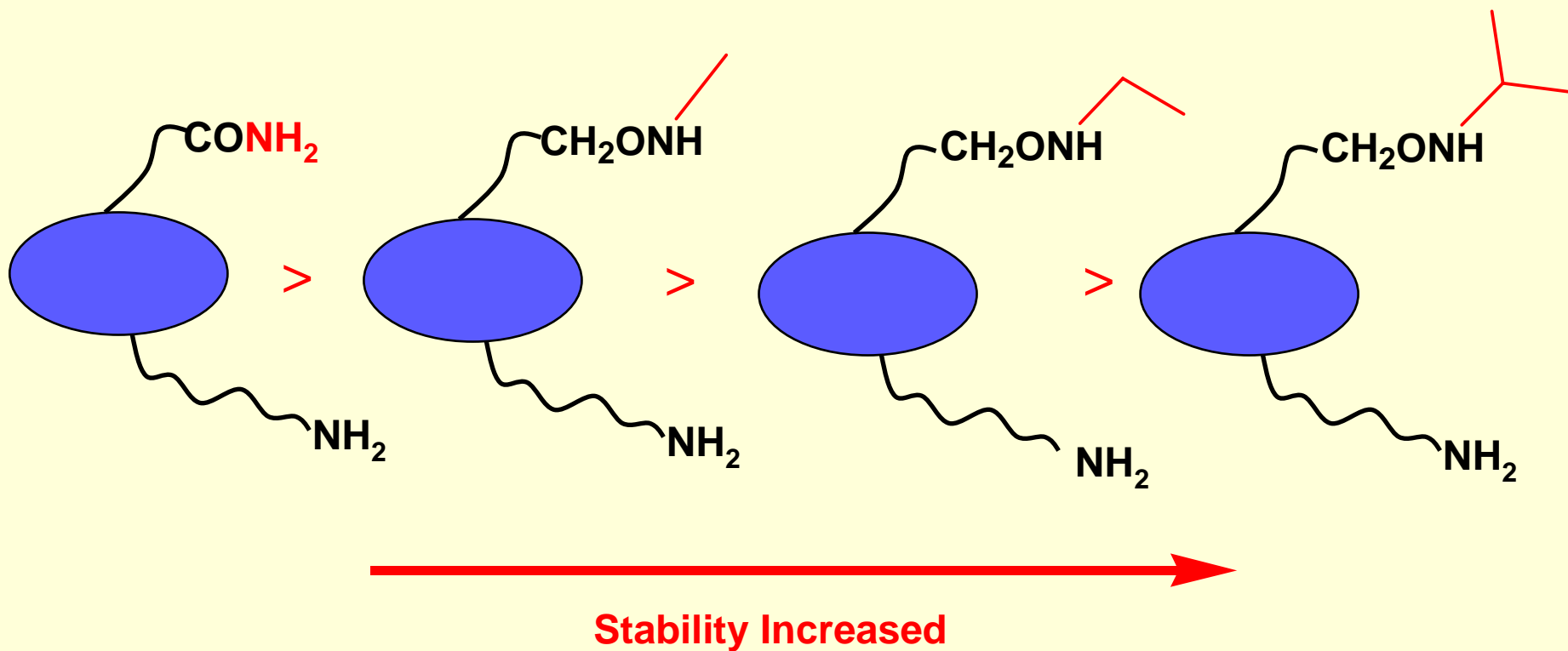
Very active in vivo

But.....

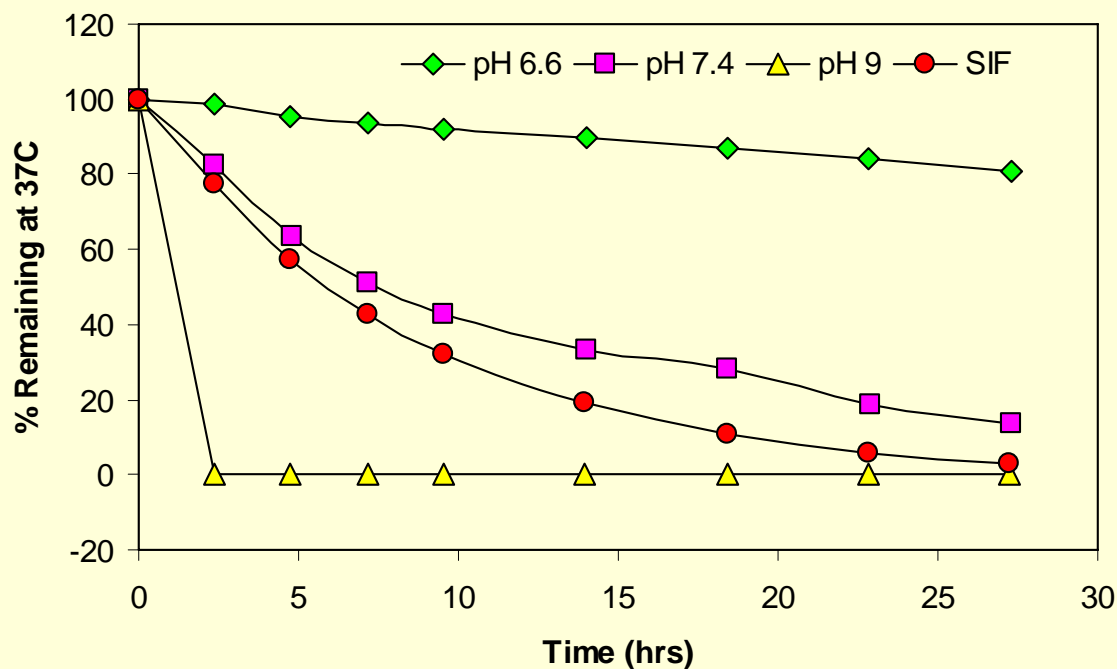
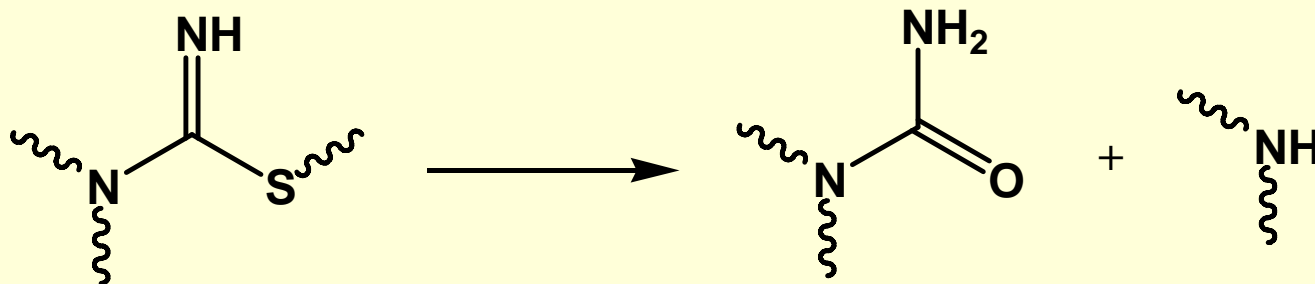
Compound Rapidly Cyclized



Guide Structural Modification



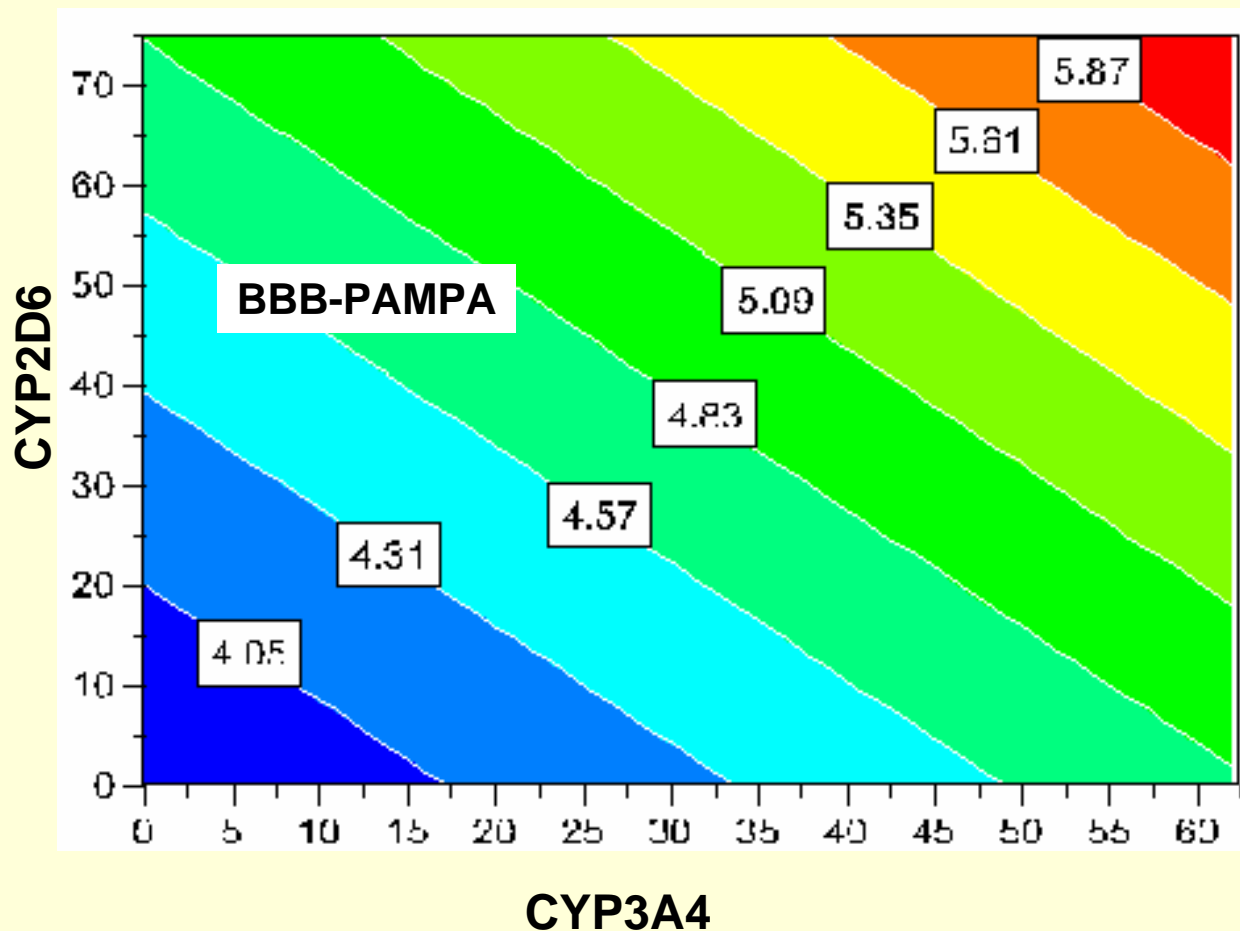
Isothiourea Rapidly Hydrolyzed in Buffer



Project was terminated

Identify Series with High Potential for Optimization of BBB and CYP

MVA

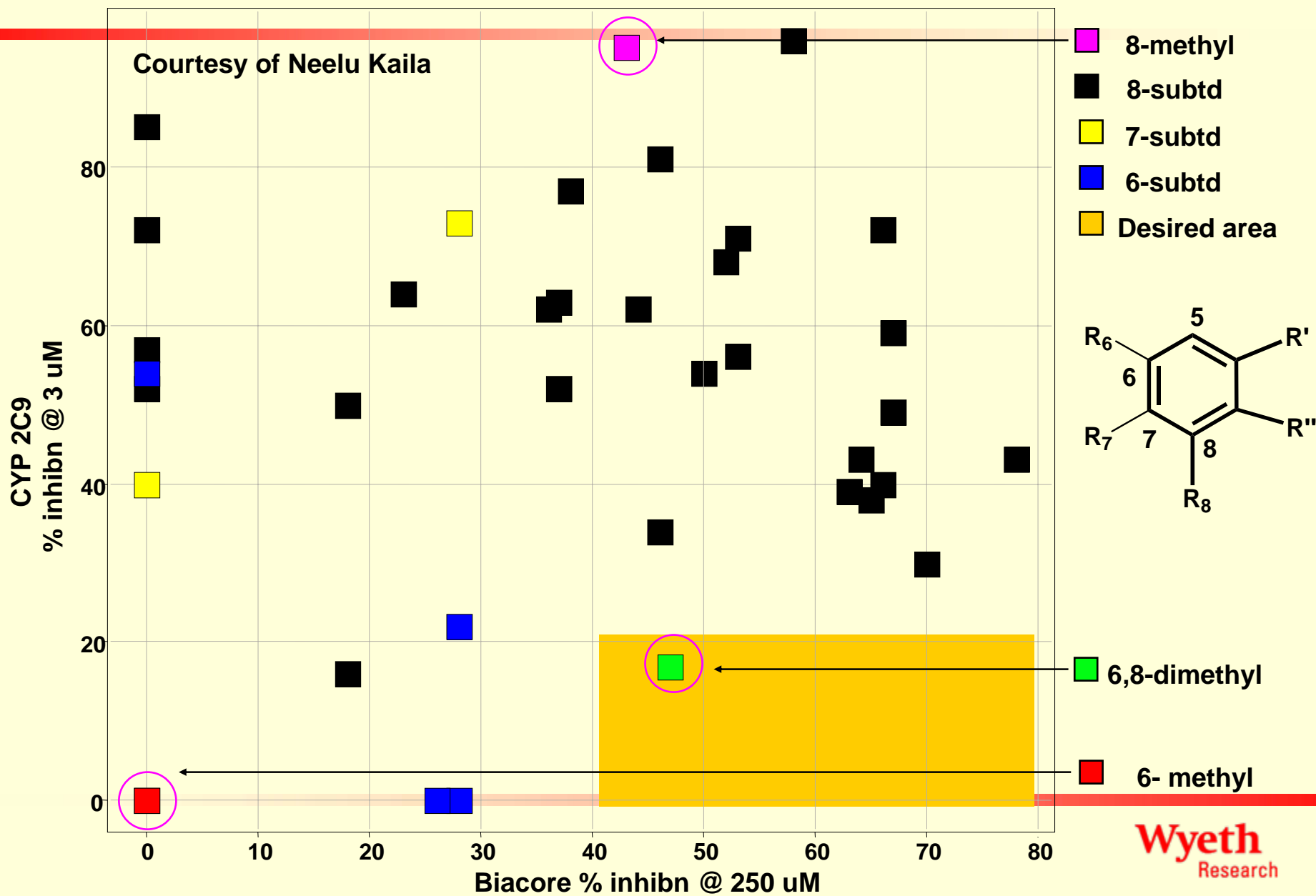


- Reverse amide series
- BBB-PAMPA values > 4 and acceptable CYP 3A4/CYP 2D6 inhibition can be obtained

Courtesy of Adam Gilbert

Parallel Optimization of Potency and CYP Inhibition

6,8 Disubstituted analogs may help get away from CYP inhibition



Guide Structural Modification

Screen solubility of 500 compounds

Lead

- ▶ Solubility = 0 $\mu\text{g/mL}$
- ▶ % F = 0 (Tw/MC)
- ▶ Efficacy: Corn Oil

Candidate

- ▶ Solubility > 6 mg/mL
- ▶ % F > 20 % (Tw/MC)
- ▶ Efficacy: Tw/MC

Quality Development Candidate

Guide Structural Modification

Screen ~1000 metabolic stability and ~100 Pgp

Lead

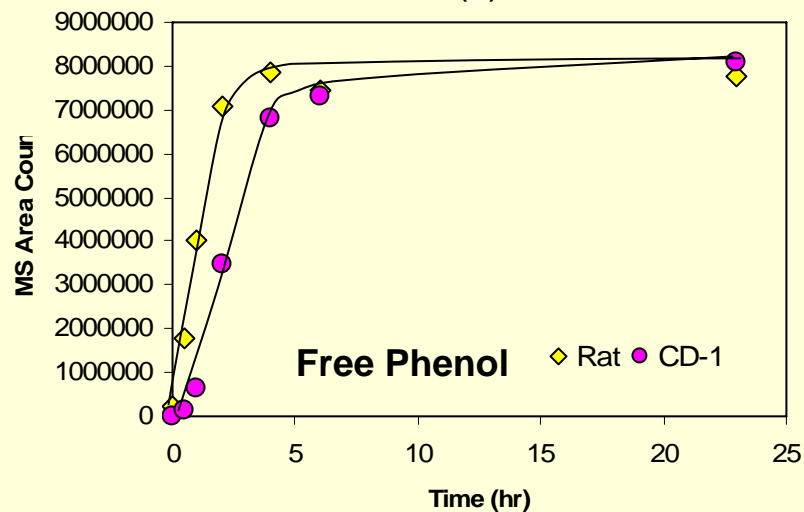
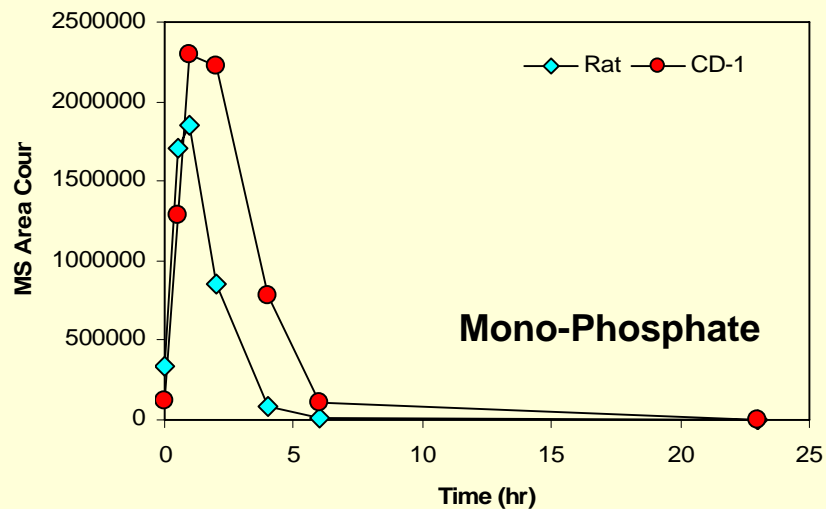
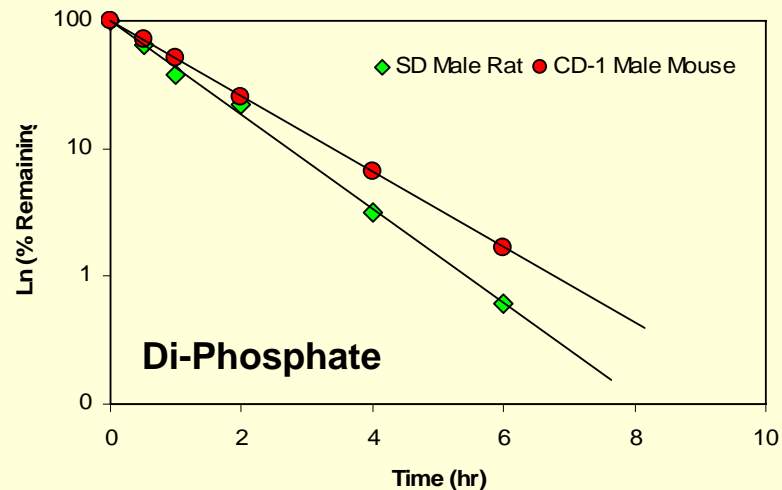
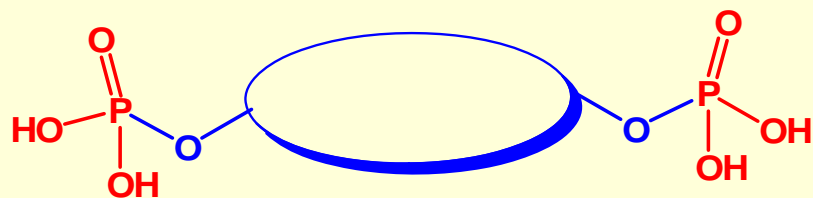
- ▶ Met. $t_{1/2} < 5$ min
- ▶ Pgp efflux > 10
- ▶ B/P ratio < 0.02
- ▶ No Efficacy

Candidate

- ▶ Met. $t_{1/2} > 30$ min
- ▶ Pgp efflux < 2.5
- ▶ B/P ratio > 0.60
- ▶ Efficacious

Enhanced Success

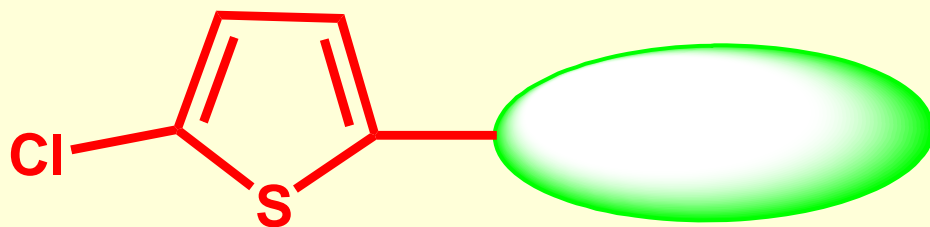
Screening of Prodrugs



Identify prodrug with desirable properties **Wyeth** Research

Develop Structure-Property Relationships

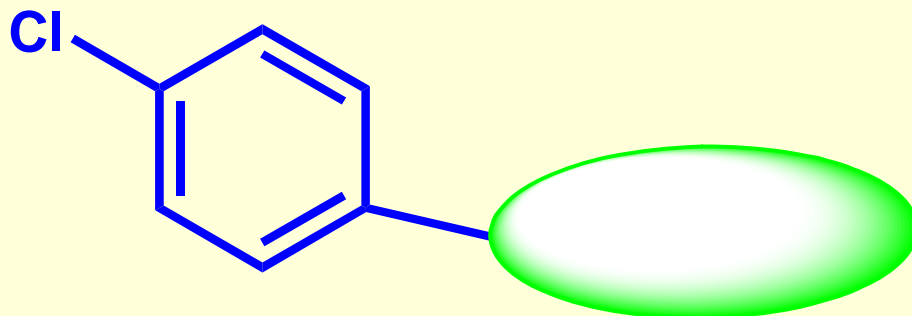
2-Cl Thiophene



ED ₅₀	5-13 nM
Rat Stab.	< 1 min
Mouse Stab.	< 1 min
Human Stab.	4 min

Inactive in vivo

p-Cl Benzene

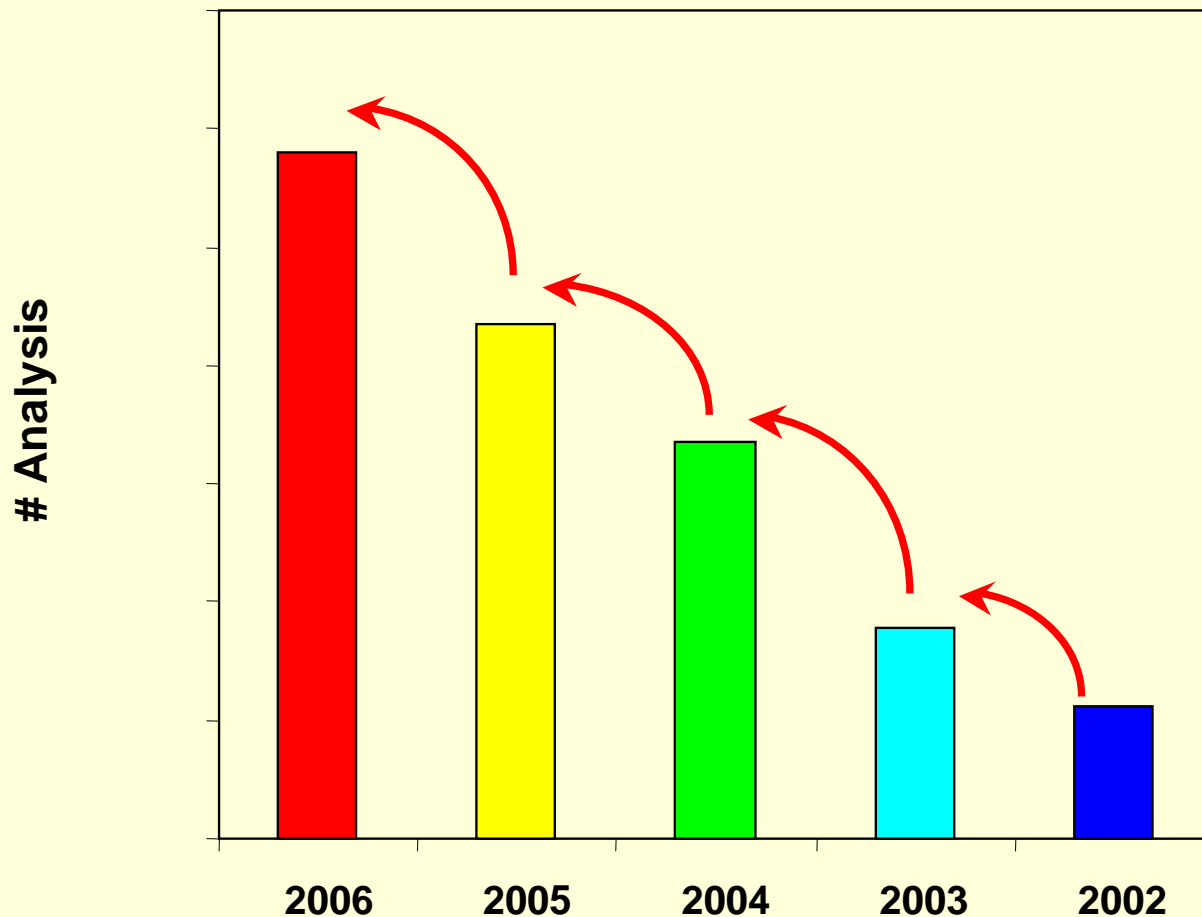


ED ₅₀	12 nM
Rat Stab.	21 min
Mouse Stab.	13 min
Human Stab.	11 min

Active in vivo

Improved Stability and In Vivo Performance

Microsomal Stability: >30% Increase Per Year



Success Breeds Success

Future Prospective

- **More predictive software**
- **Miniaturization**
- **Predictive toxicity**
- **Earlier profiling**

Enhance Predictability and Intelligence of Software

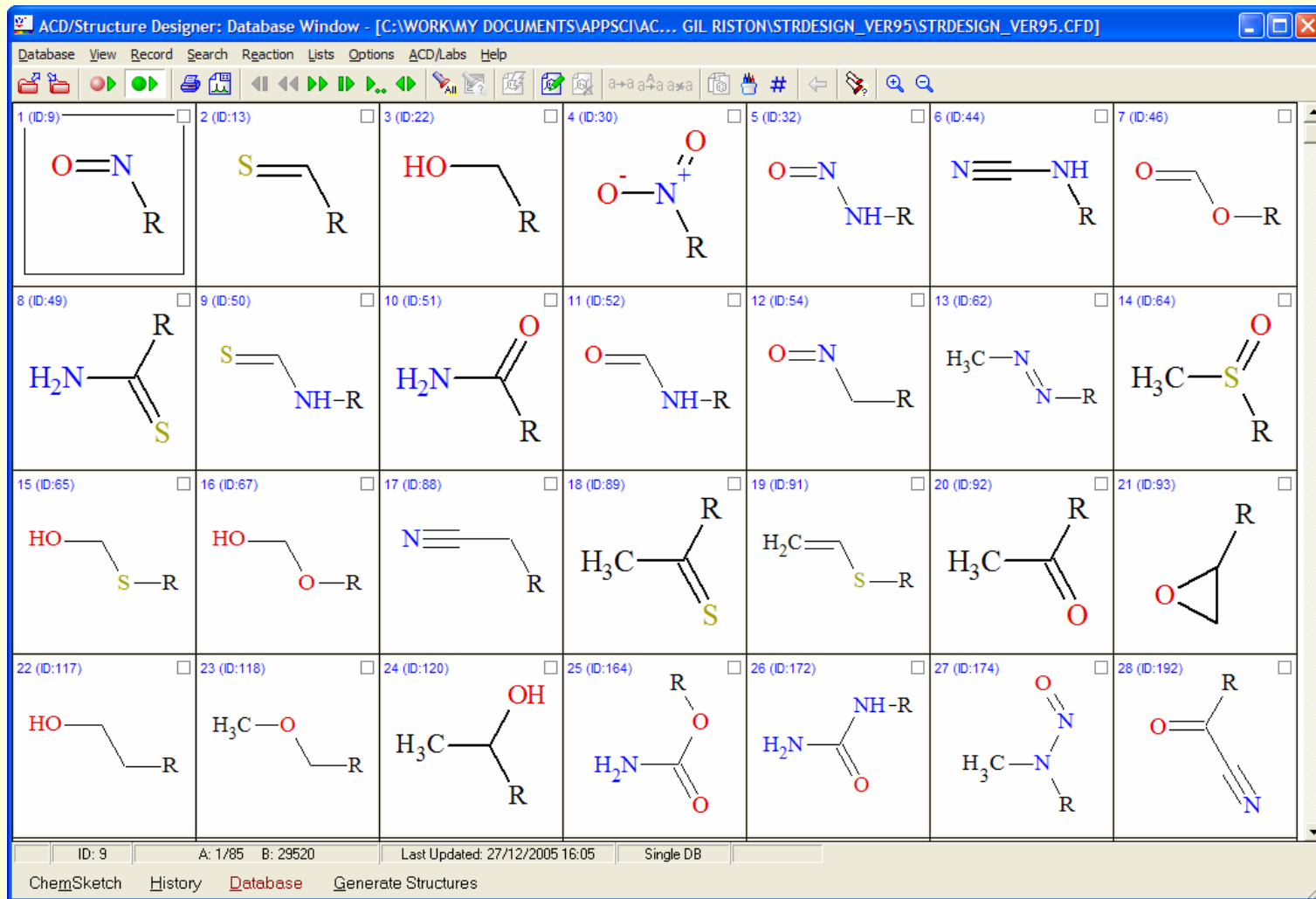
- **Predictability**

- ▶ Accuracy
- ▶ Coverage
- ▶ Custom models

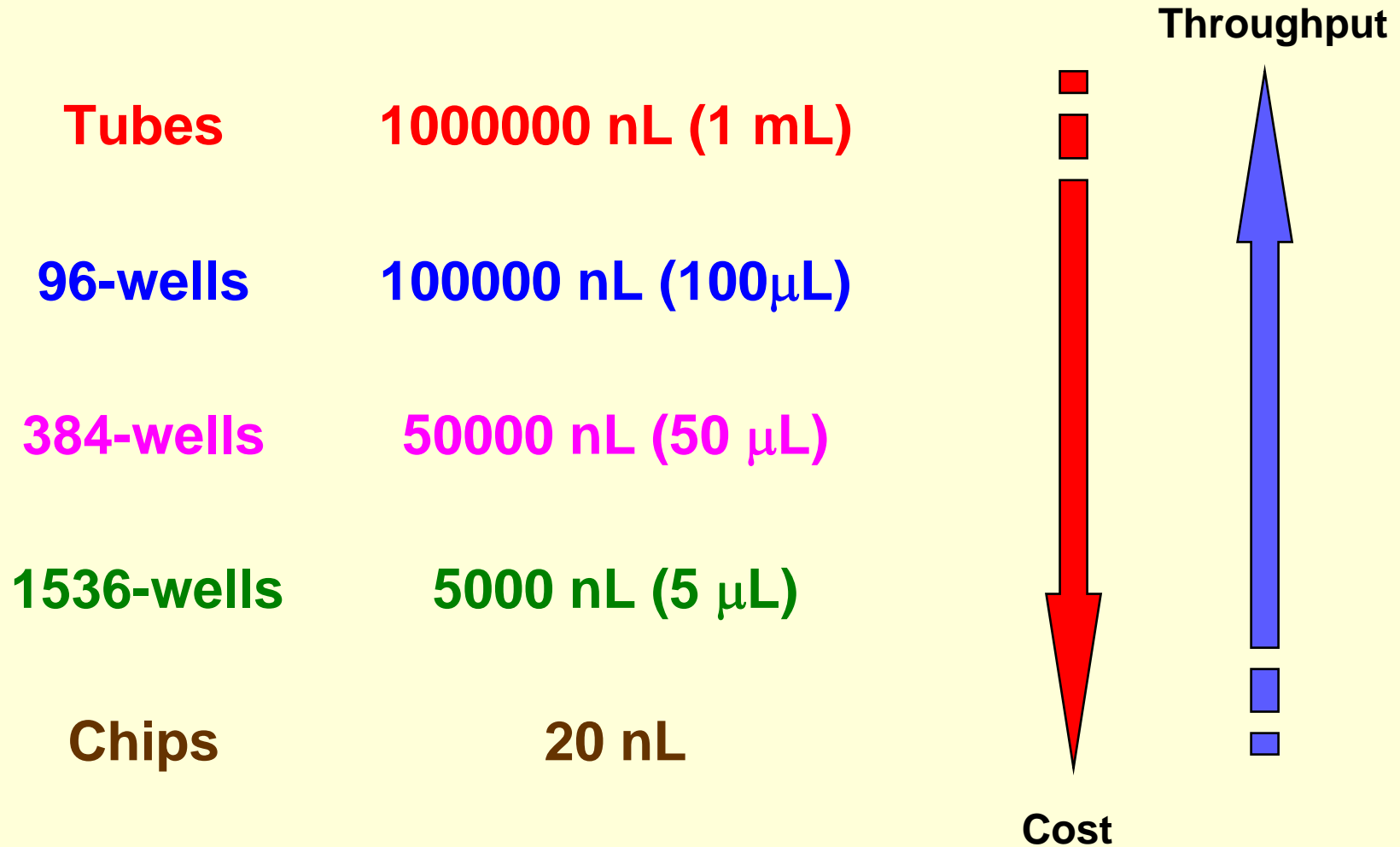
- **Intelligence**

- ▶ Propose substituents
- ▶ Design new compounds

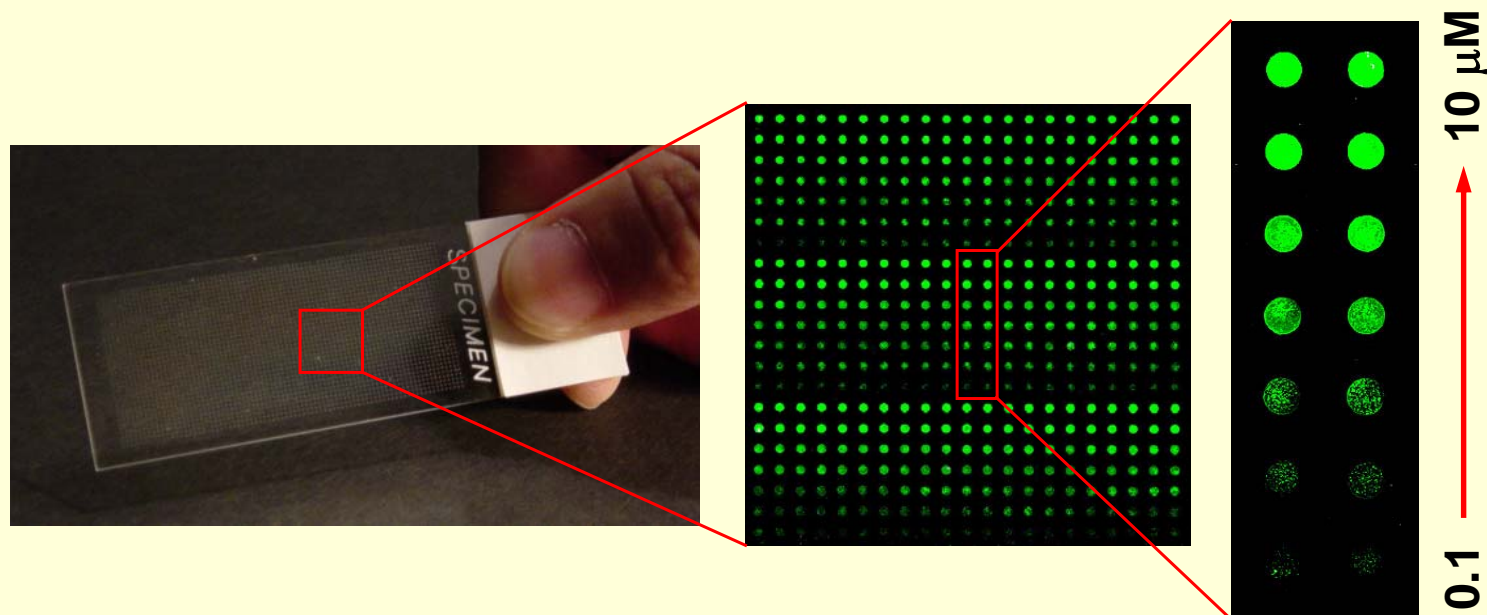
ACD Structure Designer: Propose Substituents



Future: Miniaturization

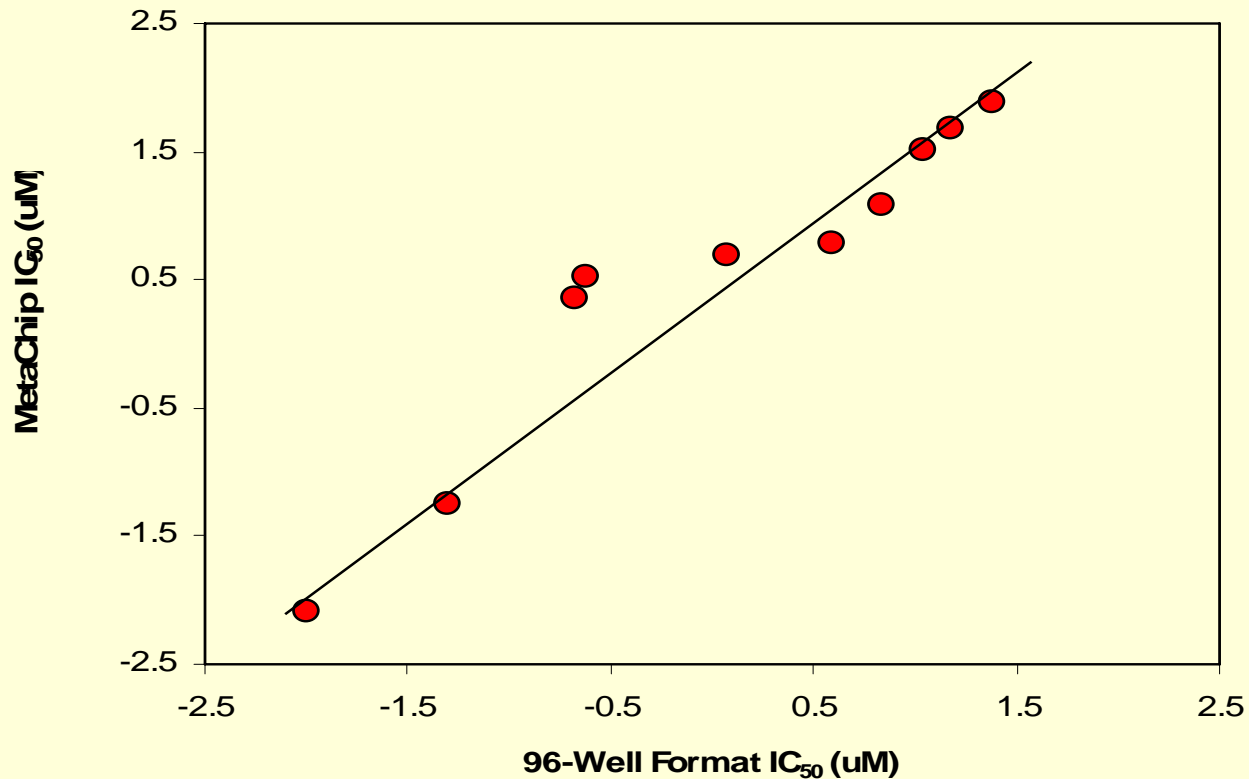


Future Miniaturization: Metachip from Solidus

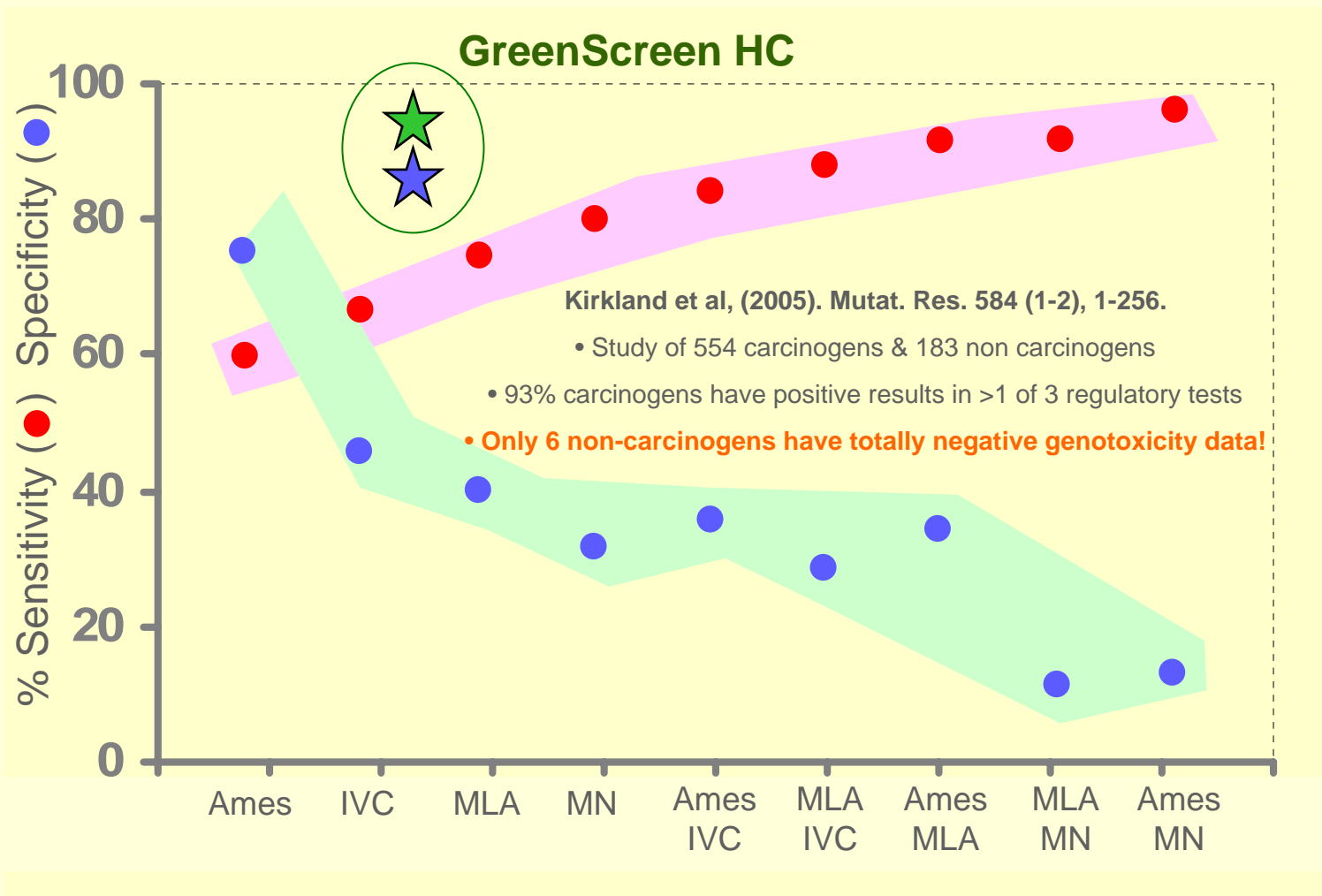


- **> 1000x volume reduction (20 nL): savings**
- **11,200 spots / slide: throughput and speed**

Correlation Between MetaChip and 96-Well Format for CYP3A4 Inhibition



More Predictive TOX Assays



Peter McCulloch, Gentronix

Early Metabolite Identification

- **In Silico**

- ▶ MetaSite

- ▶ Admensa

- **LC-MS-MS**

- **LC-NMR**

Impact of Pharmaceutical Profiling on Attrition

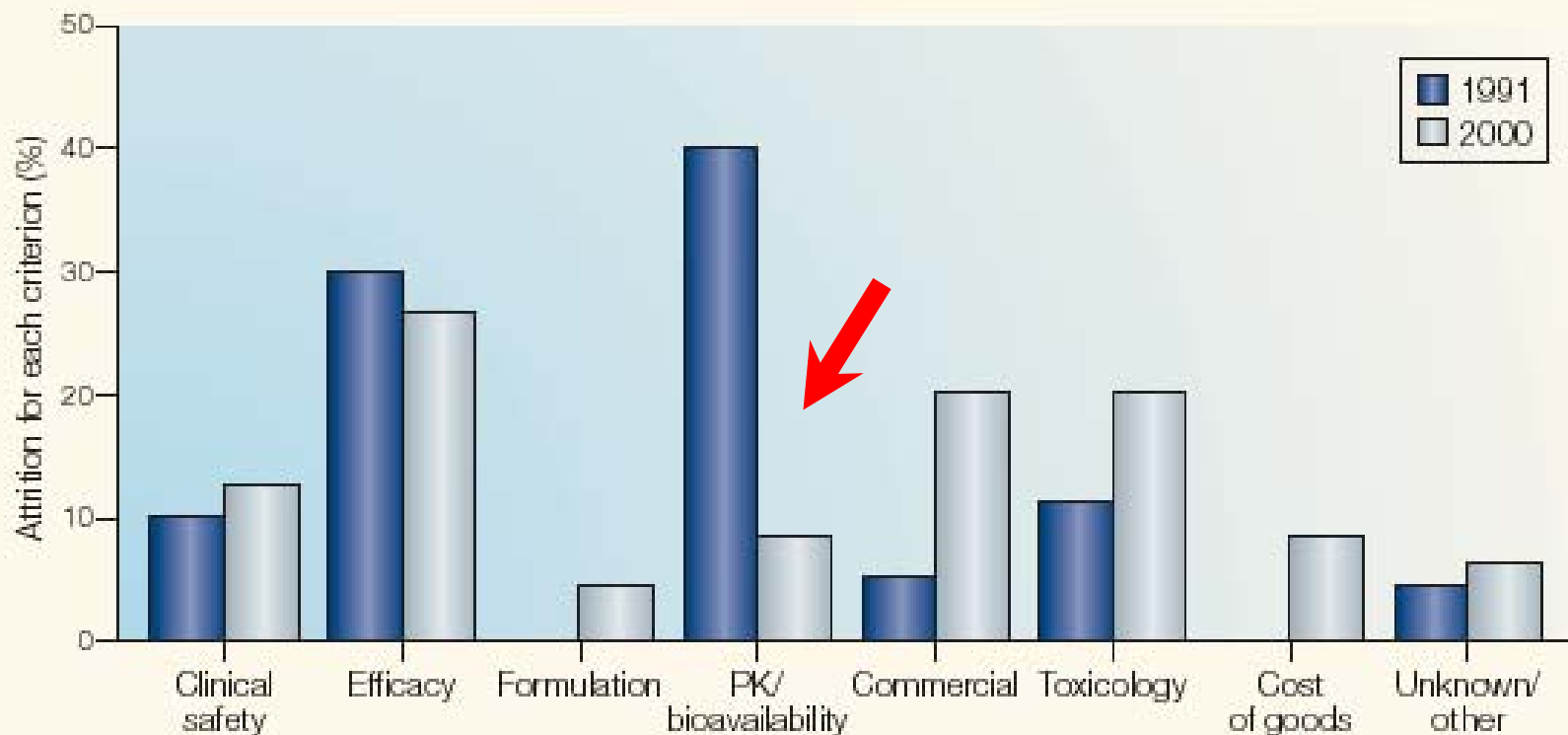
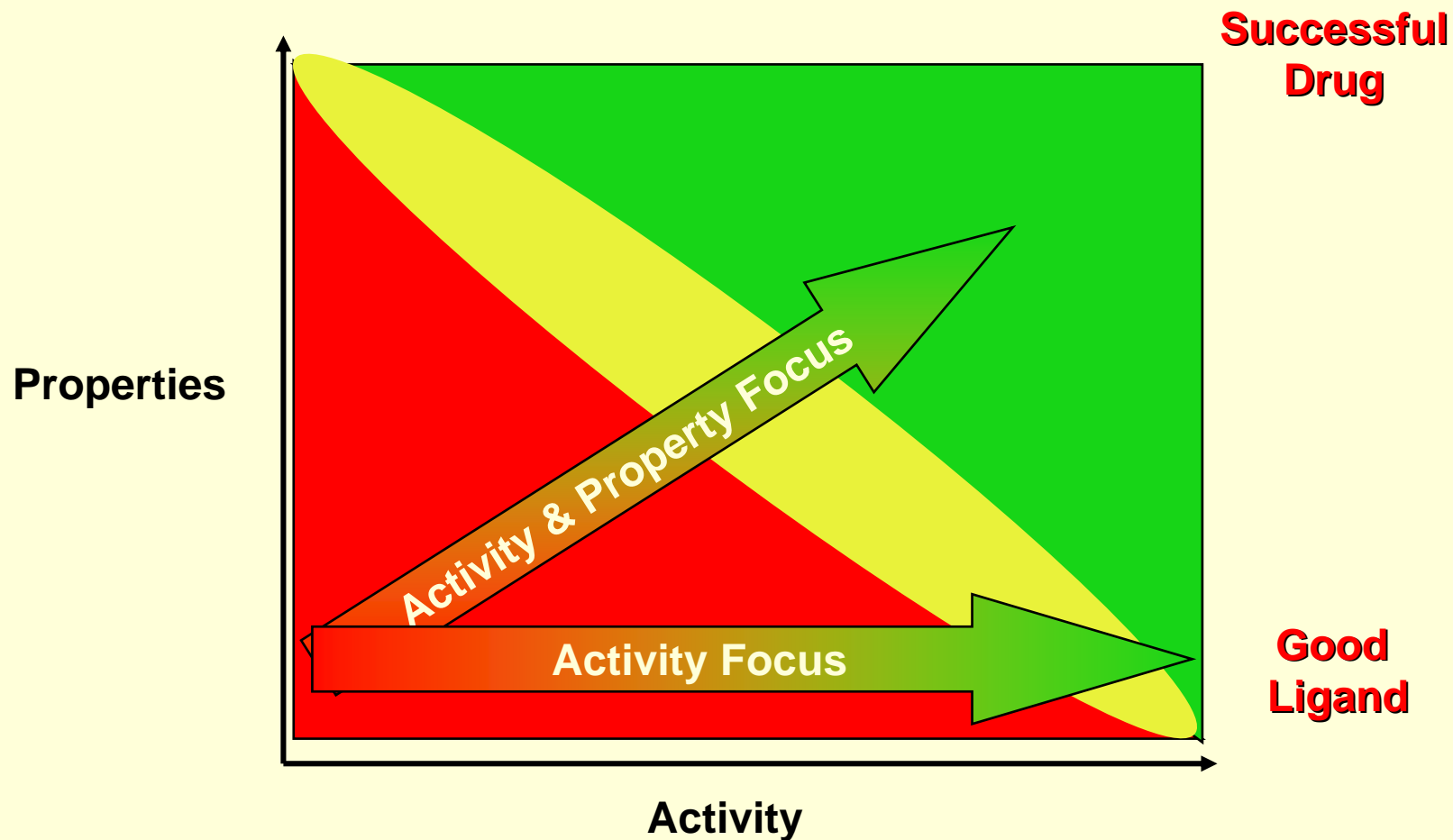


Figure 3 | Reasons for attrition (1991–2000). PK, pharmacokinetics.

Conclusions



Acknowledgments

- **Princeton:** Edward Kerns, Susan Petusky, Susan Li, Zhen Lin, Hong Jin, Ian Bezar
- **Pearl River:** Yelena Pyatski, Deanna DeGrandi, Joe Marini, Barry Press
- **Collegeville:** Scott Brecker
- **Cambridge:** Nelson Huang
- **Chem./Bio.:** Neelu Kaila, Adam Gilbert, Baihua Hu, Jay Wroble, Jeremy Levin, Jeff Pelletier, Jonathon Gross, Martin DeGrandi, Lee Jennings, Mike Malamas, Paul Dollings, Tim Lock, Paige Mahaney, Bill Moore, Andy Fensome, Yuren Wang, Derek Cole, Al Robichaud, John Butera, Boyd Harrison, John Ellingboe
- **Leadership:** Oliver McConnell, Guy Carter, Magid Abou-Gharbia