Introduction to Applied Mathematics and Informatics in Drug Discovery (AMIDD)

How were new medicines discovered?

Yao, Peter, Petra, Carlos, Jan

Content

1. Target-based screening
2. Phenotypic based screening
3. Examples of discovered drugs
4. Conclusion

How were new medicines discovered?

David C. Swinney[*‡] *and Jason Anthony*[*]

NATURE REVIEWS | DRUG DISCOVERY
VOLUME 10 | JULY 2011 | 507

Target-Based Screening

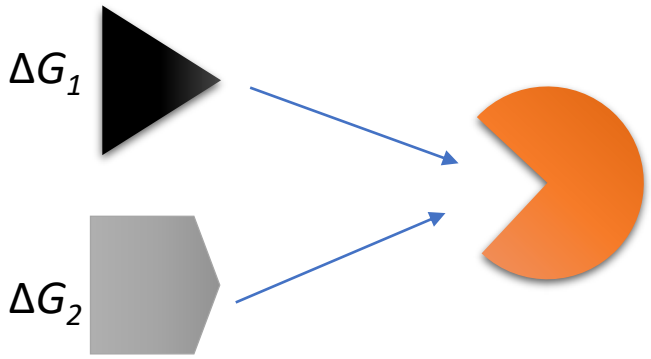
Jan Weinreich

Target-Based Screening

Biological Hypothesis:
Target (e.g. protein) plays key role in a disease pathogenesis

→ Identify molecules that modify target activity (hit molecules)
→ Lead optimization
→ Preclinical development
→ ...

*"**Rational basis for discovery of new medicines**"* [1]



Example for MMOA: Minimize Free Energy ΔG

[1] D. Swinney , J. Anthony, Nat. Rev. Drug Discov., 10, 507–519, (2011)

"The target-based approach can very effectively develop novel treatments for a validated target, but the process of target validation is complex and associated with a high degree of uncertainty"

Frank Sams-Dodd

Target-based drug discovery: is something wrong? [2]

[2] F. Sams-Dodd, Drug Discovery Today, 10, 2, (2005)

+	-
• Use knowledge of MMOA to improve binding affinities to target (QM/MM) → Systematic improvement of drug candidate (lead optimization)	• Chain: *in silico*→ *in vitro* → *in vivo* might break!
• Mol. approach & empirical rules like Lipinski's rule of five → small molecule screening	• Small sampling of Chemical Compound space
• Biologic based approach	• Target validation often difficult → expensive
• High-throughput screening (*in silico* & *in vitro*)	• High target affinity does not necessarily mean high therapeutic efficiency
• Target based approach most successful for cancer, infectious diseases etc.	• "One target view" often too simple & full dynamics of all interactions has to be considered

Target based Screening responsible for decline in attrition rates?

-	+
• Since 90s focus on target based approach [1]	• Machine Learning improves (virtual) screening throughput [3,4]
• Uncertainty of the physiological role of the target in the intact organism [2]	• Laboratory automation/lab-on-a chip advances in Microfluidics [5]
• If validity of target not given, programs should have been terminated earlier point (interim analysis) [2]	
• Companies often use same targets because of a lack of druggable and validated targets [2]	

[1] D. Swinney , J. Anthony, Nat. Rev. Drug Discov., 10, 507–519, (2011)

[2] F. Sams-Dodd, Drug Discov. Today, 10, 2, (2005)

[3] A. Lee, M. Brenner, Proc. Nat. Ac. of Sci., 48 ,13564-13569 (2016)

[4] M. Rupp, O. A. von Lilienfeld, K. Burke, J. Chem. Phys. 148, 241401 (2018)

[5] P. Dittrich,A. Manz., Nat. Rev. Drug. Discov., 5, 210–218 (2006)

Phenotypic-Based Screening

Petra Stankovic

Phenotypic Based Screening

- Phenotypic screening is one of the four preclinical strategies used to identify potential drug candidates
- It identifies substances such as small molecules or peptide that alter the phenotype of a cell or of any other observed organism
- Empirical approach

Advantages

- Assays (analytic procedures) do not require prior understanding of *MMOA* (Molecular Mechanism of Action)
- The translation of the activity in such assays into therapeutic impact, in a given disease state, is more effective than in artificial target-based approach

Disadvantages

- Assays (analytic procedures) do not require prior understanding of *MMOA* (Molecular Mechanism of Action)
- The translation of the activity in such assays into therapeutic impact, in a given disease state, is more effective than in artificial target-based approach

NMEs that were discovered through phenotypic screening

- The first-in-class small-molecule *Nme*s (New molecular entities) that were discovered came from two directions :
  1. Intentional targeting of a specific phenotype (25 *NME*s)
  2. Through serendipity (3 *NME*s)
- The newly discovered molecules were used to identify *MMOA*s for the physiological phehomena, e.g. oxazolidinone antibiotics such as linezolid (infectious disease)

- The focus was on using specific chemical classes in which prior knowledge contributed to matching them with the phenotype
- Random library screening was successful for ezetimibe (cardiovascular), Pemirolast and sirolimus (immune modulation), Retapamulin and linezolid, (infectious disease)
- The process of identification of new *MMOAs* also led to the discovery of e.g. aripiprazole and varenicline

Box 2 | **Biochemical efficiency**

The dose of a drug required to achieve the desired physiological response depends on its biochemical efficiency[10,11]. This is defined as 'binding affinity/functional response', which is equivalent to K_i/EC_{50} (effector concentration for half-maximal response). Good biochemical efficiency enables efficacy at lower drug concentrations and increases the therapeutic index. It is a property of many approved medicines[10,11].

There are many factors that can influence the shift in dose–response curves between binding and functional assays, including:

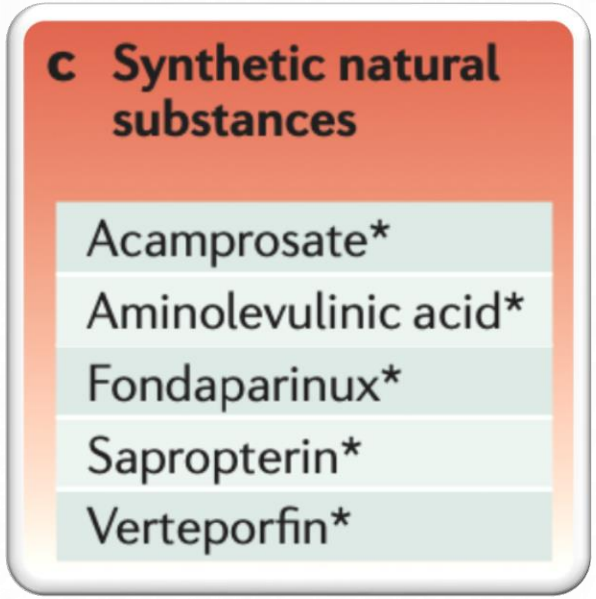
- Pharmacokinetics and ADME (absorption, distribution, metabolism and excretion) properties
- Assay relevance (is the functional assay appropriate for the target? Are the assays technically accurate?)
- The involvement of the target in the functional readout and biology
- The molecular mechanism of action (MMOA)

Although all of these factors can and do contribute to the relationship between binding affinity and the functional response, the role of the MMOA is not always considered. The concept of biochemical efficiency was introduced to quantify this possibility[10,11]. When biochemical efficiency is used as a measure of an optimal MMOA, it is important that the other mitigating factors are eliminated. For example, when evaluating biochemical efficiency, the assays must be run in the absence of serum (or plasma) to eliminate the shift in IC_{50} (half-maximal inhibitory concentration) owing to serum protein binding.

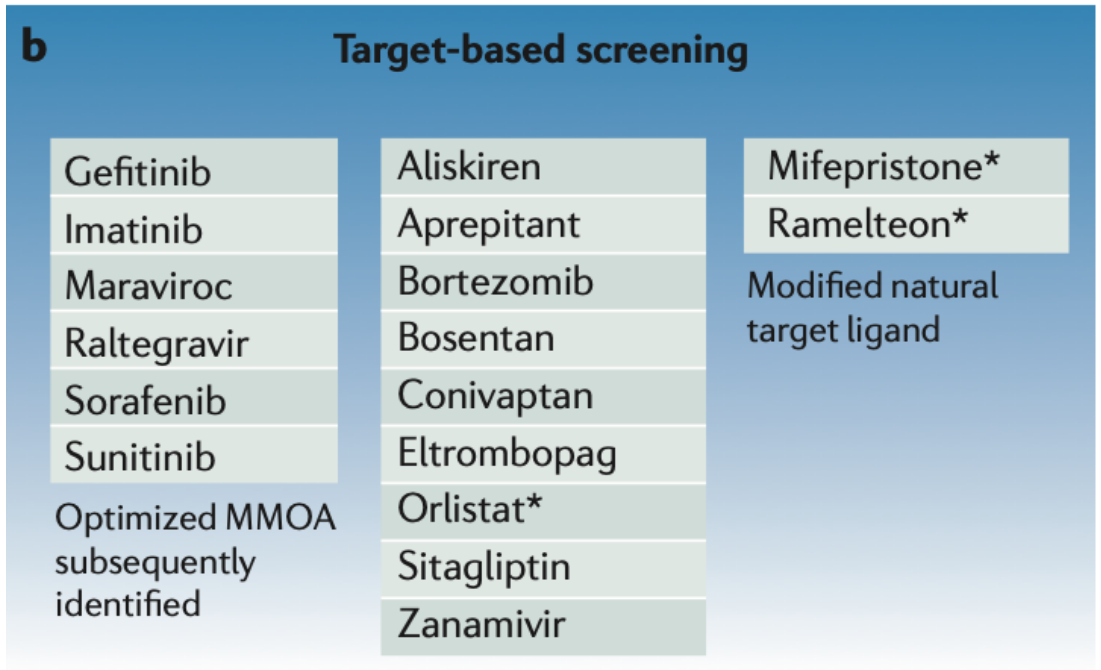
BOX 2
biochemical efficiency

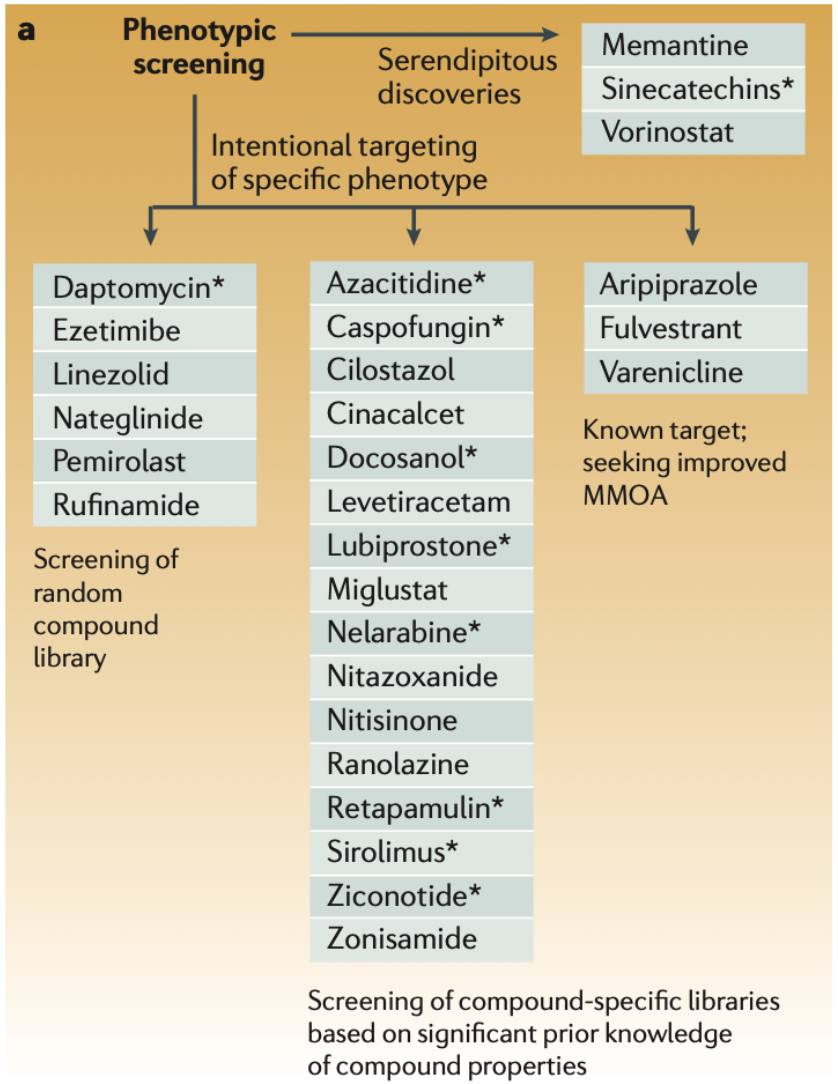
NMEs that were developed as synthetic and/or modified versions of natural substances, or discovered by screening such substances

- A small fraction of first-in class Nmes were developed as synthetic versions of natural substances
- Some were anticoagulant drugs (sapropterin) others treat alcoholism (acamprosate), and Verteporfin is used to eliminate blood vessels in the eye



In some cases, natural substances provided starting points for small-molecule phenotypic screening (a) and target-based discovery (B)





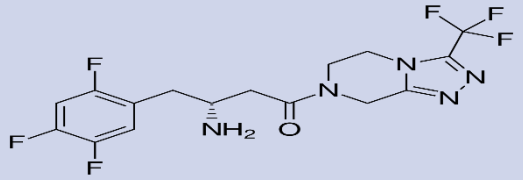
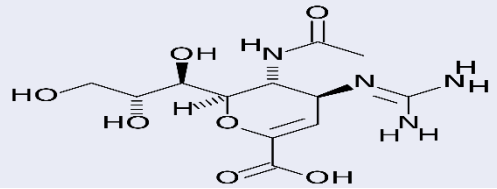
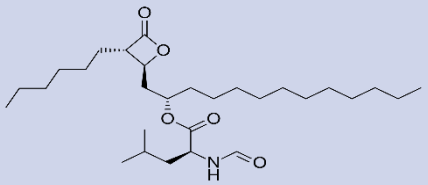
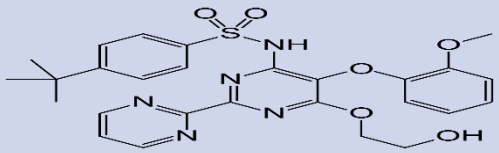
Conclusion

- 36% of first-in-class small-molecule NMEs originated from natural substances
- The results are consistent with other studies such as “Natural Products as sources of new drugs over the last 25 years” conducted by Newman and Cragg
- That natural substances were prevalent, was noticeable in discovery of NMEs through target-based approach as well

Target-based screening first-in-class drugs

Carlos Tejera

Examples: Target-based first-in-class drugs

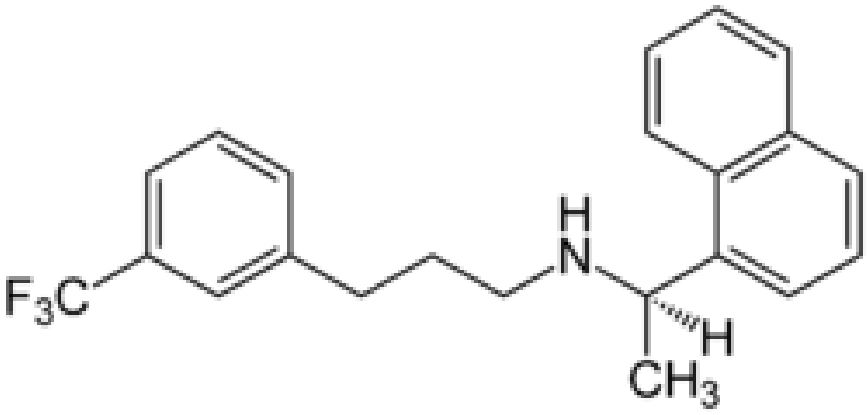
Drug	Therapeutic area	Target type	MMoA	Molecular structure
Sitagliptin	Metabolic	Enzyme	Equilibrium binding	
Zanamivir	Infectious desease	Enzyme	Equilibrium binding	
Orlistat	Metabolic	Enzyme	Inhibition	
Eltrompobag	Immune	Receptor	Non-competitive agonist	
Bosetan	Cardiovascular	Receptor	Equilibrium binding	

Phenotypic-based Screening first-in-class drugs

Peter Ruthemann

Examples: Phenotypic-based screening

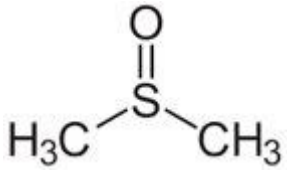
Drug (trade name; company)	Therapeutic area	Target type	Molecular mechanism of action
Discovered through phenotypic screening			
Aripiprazole (Abilify; Bristol-Myers Squibb/Otsuka Pharmaceutical)	CNS	Receptor	Conformational/partial agonist
Azacitidine (Vidaza; Celgene/Pfizer)	Cancer	Enyzme	Irreversible inhibition
Caspofungin (Cancidas; Merck)	Infectious disease	Enzyme	Noncompetitive inhibition
Cilostazol (Pletal; Otsuka)	Cardiovascular	Enzyme	Inhibition
Rufinamide (Inovelon; Novartis)	CNS	Unknown	Unknown
Sinecatechins (Veregen; Medigene)	Infectious disease	Unknown	Unknown
Sirolimus (Rapamune; Pfizer)	Immune modulation	Enzyme	Conformational/inhibition
Varenicline (Chantix; Pfizer)	CNS	Ion channel	Conformational/partial agonist
Vorinostat (Zolinza; Merck)	Cancer	Enzyme	Equilibrium kinetics
Ziconotide (Prialt; Elan Pharmaceuticals)	Pain and/or CNS	Ion channel	Equilibrium kinetics
Zonisamide (Excegran; Dainippon Pharmaceuticals)	CNS	Unknown	Unknown



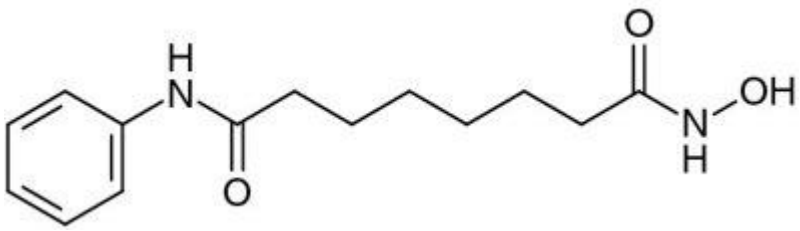
Cinacalcet

Ex.1: Cinacalcet

- Allosteric modulator of Ca^{2+} sensitive GPCR receptor
- Increases sensitivity of receptor to Ca^{2+}
- Strong affinity to receptors in thyroid gland
- Therapy: Excess segregation of parathyroid hormone



DMSO



Vorinostat

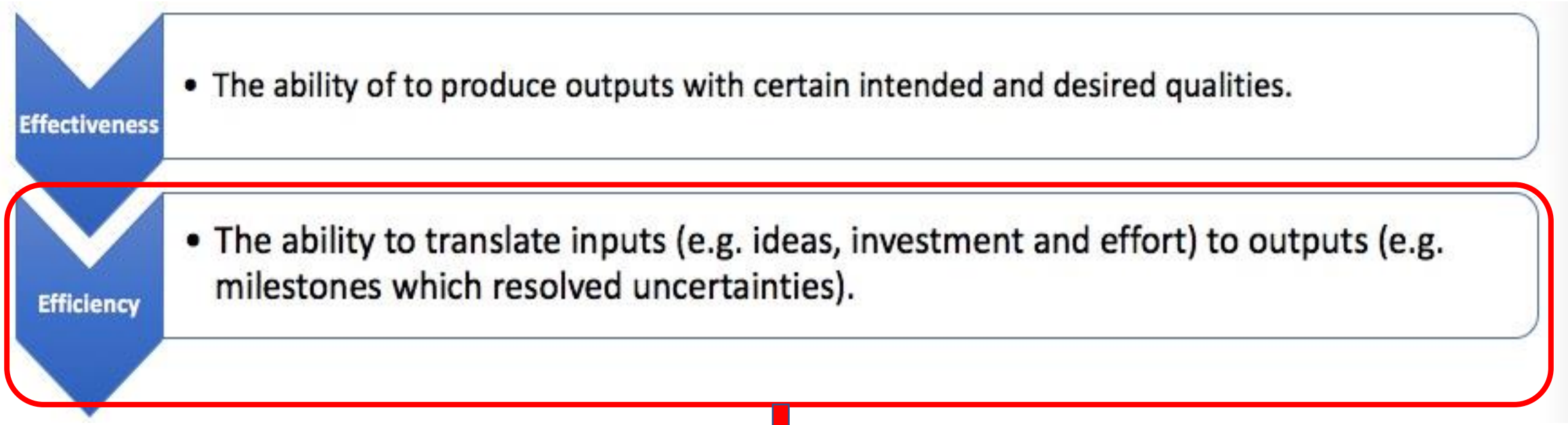
Ex.2: Serendipity: Vorinostat

- Murine cells differentiated transfection procedure
- Traced back to DMSO
- Lead optimisation: Vorinostat
- Therapy: T-cell lymphoma
- Epigenetic regulator

Summary

Yao Wei

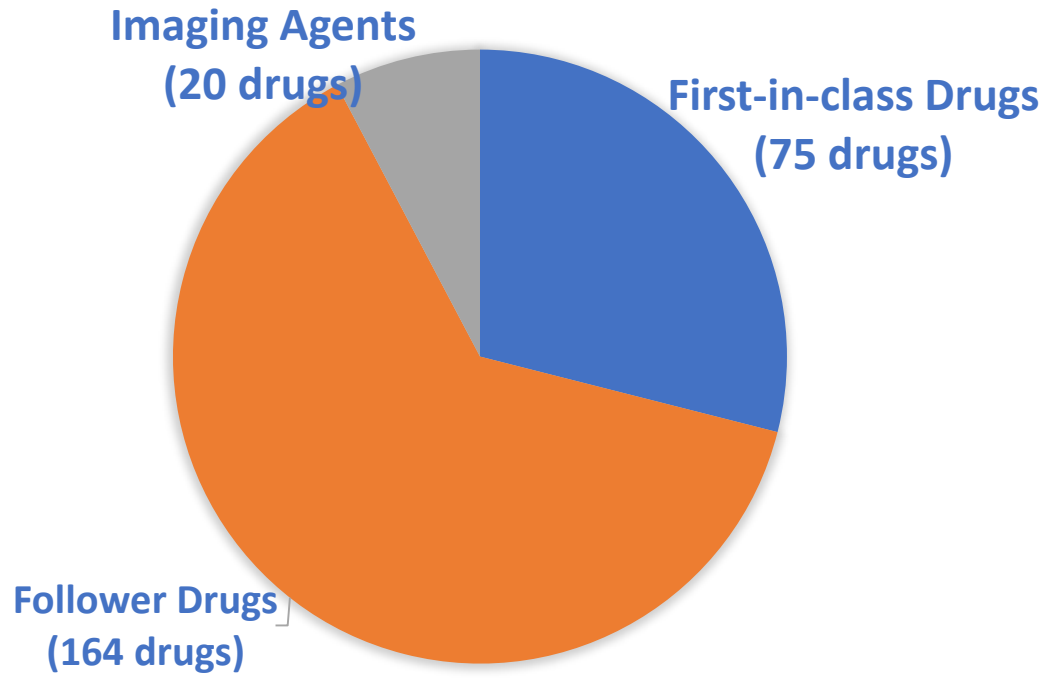
In Pharmaceutical Research and Development:

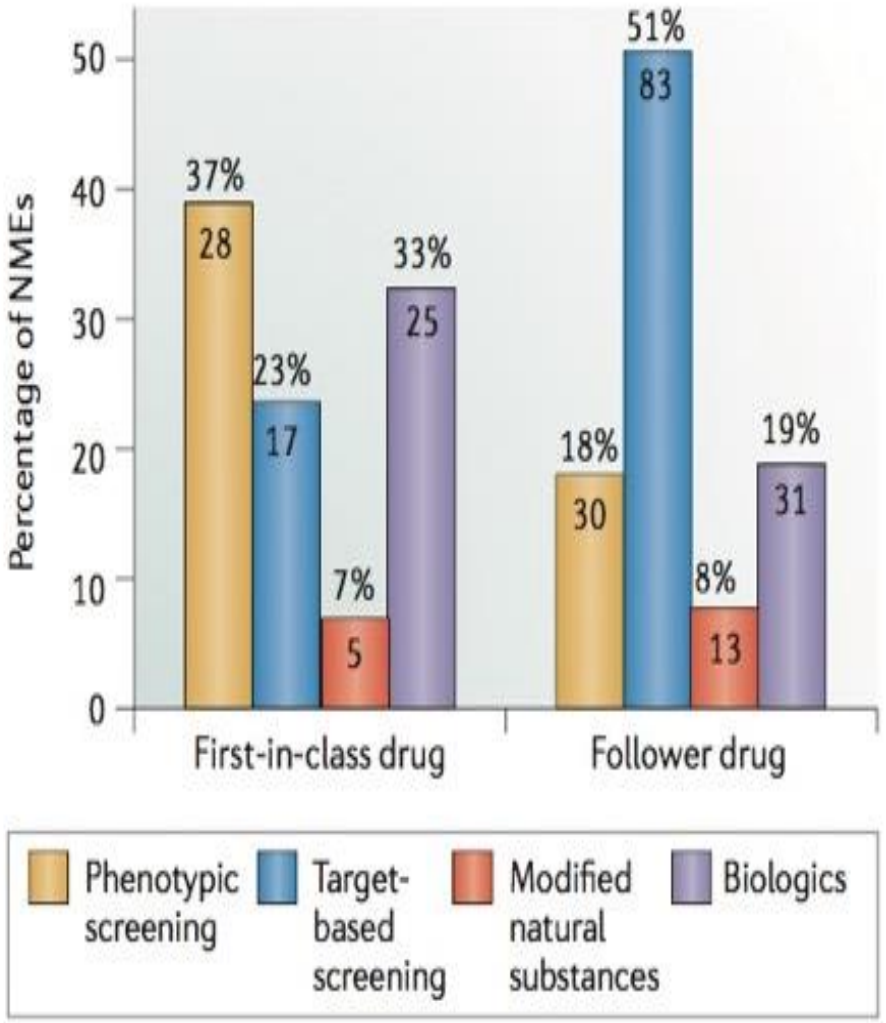


- The probability of **technical success** is a key variable.
- The **target selection** may be one of the most important determinants of attrition and overall R&D productivity.

This paper:

- Introduced two main target selection strategies: **target-based approaches and phenotypic-based approaches**.
- Analyzed **259 agents** which were approved by the US Food and Drug Administration between 1999 and 2008.





The distribution of new drugs discovered between 1999 and 2008, according to the discovery strategy

- **First-in-class drugs**

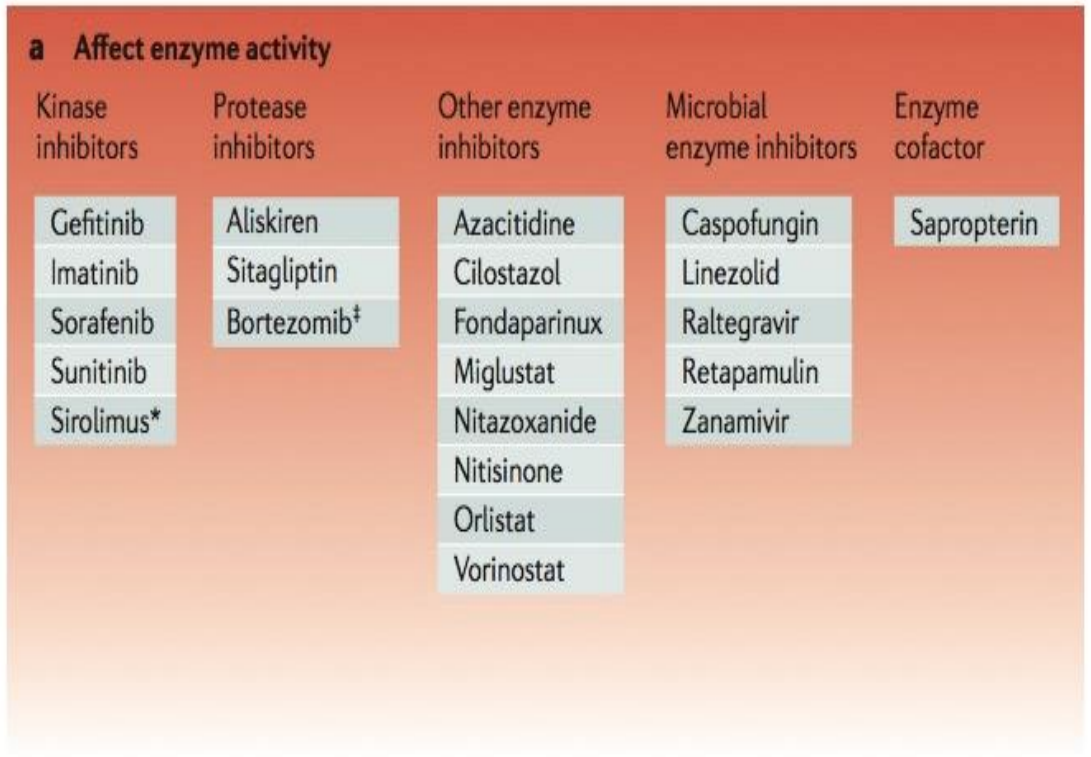
  - 28 agents were developed via phenotypic-based screening, while, 17 agents were developed via the target-based screening;

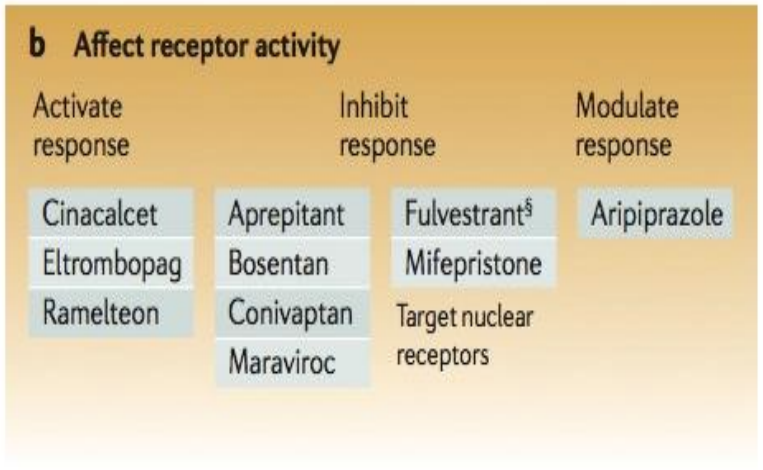
  - The phenotypic-based screening was used before the target-based screening. So there is a lag time for introducing new technologies and strategies.

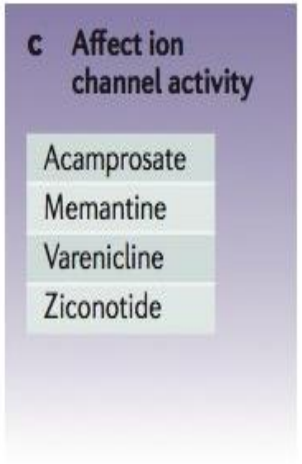
- **Follower drugs** :

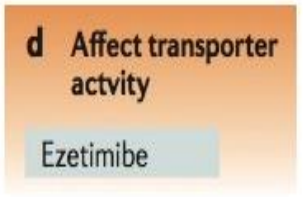
  - Target based screening contributed for 83 agents, while, phenotypic based screening contributed for 30 agents;

  - Drug developers take knowledge of a previously identified MMOA to effectively use target-based tools.









- The MMOAs of First-in-class drugs:

a) Affect enzyme activity

- In almost half of the first-in-class drugs
- MMOAs: reversible, irreversible, competitive, and noncompetitive inhibition, blocking activation and stabilizing the substrate.

b) Affect receptor activity

- Most of the receptors are G protein-coupled receptors
- MMOAs: agonism, partial agonism, antagonism and allosteric modulation.

c) Affect ion channel activity

- MMOAs: uncompetitive antagonism and partial agonism

Outlook:

- Nowadays, because of advances in genetic and molecular technology, would lead to an increase in new medicines.
- **Molecular mechanism of action** is a key factor for the success of all approaches.
- Further efforts to understand the predictability/translation of the assays to human disease and the challenges of clinical development for a molecule with a limited understanding of the molecular mechanism of action will lead to a greater realization of the value of phenotypic drug discovery and ultimately increase the chance for success.

Thank you for your attention