2

THE EVOLUTION OF CLASSICAL DRUG DESIGN

Early beginings

- Eber Papyrus, an Egyptian document which is one of the oldest known medical texts (1550 B.C.)
- Covers subjects like the heart, respiratory disease, cancer, mental diseases, prediction and prevention of pregnancy, intestinal disease, abscesses, skin problems, etc.…
- Practical recipes mingle with enchantments and rites to exorcise demons ….
- Example: inhalation of the smoke of heated plants for the treatment of asthma

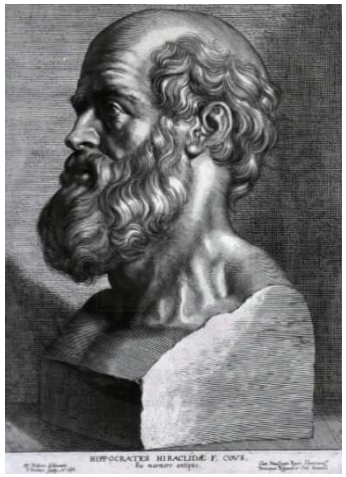
Fragmento do Papiro de Ebers

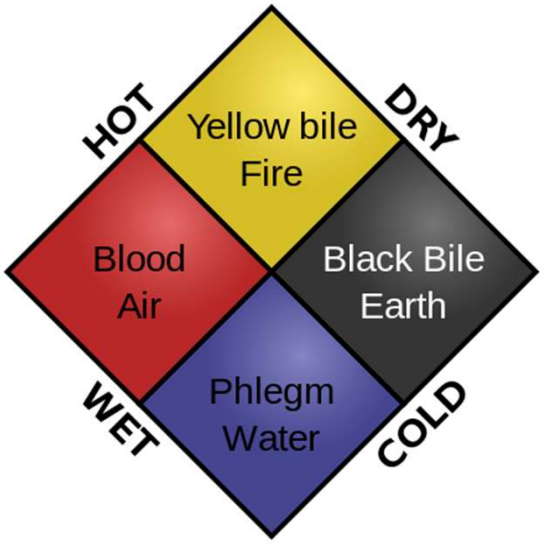
The Eber papyrus

- The Eber Papyrus includes around 800 medical “recipes”.
- Recipes for appeasing the gods…
- Empiric knowledge and common sense lead to the discovery of many plant extracts with medicinal properties which are still in use today…
- Opiate alkaloids, ephedrine, cannabis, etc.…

Greek Medicin

- Non-theistic interpretation
- Fundamentally theoretical
- Theory of the 4 humors
- Very little emphasis on medicinal plants





Traditional medicines

- Chinese
- Indian
- Arabic

Still practiced to this day. They have provided many compounds and active principles of pharmacological interest. Mix of practical knowledge with theories of disease and organism “equilibrium” based on scientifically unsound concepts like “energies”, “fluxes”, “chakras”, etc…

Paracelso

"Just as a women can be recognized and appraised on the basis of their shape, drugs can easily be identified by appearance. God has created all diseases, and he has also created na agent or drug for every disease. They can be found everywhere in nature, because nature is the natural pharmacy.."
-- "Doctrine of Signatures", Paracelsus

Paracelso (1493-1541)

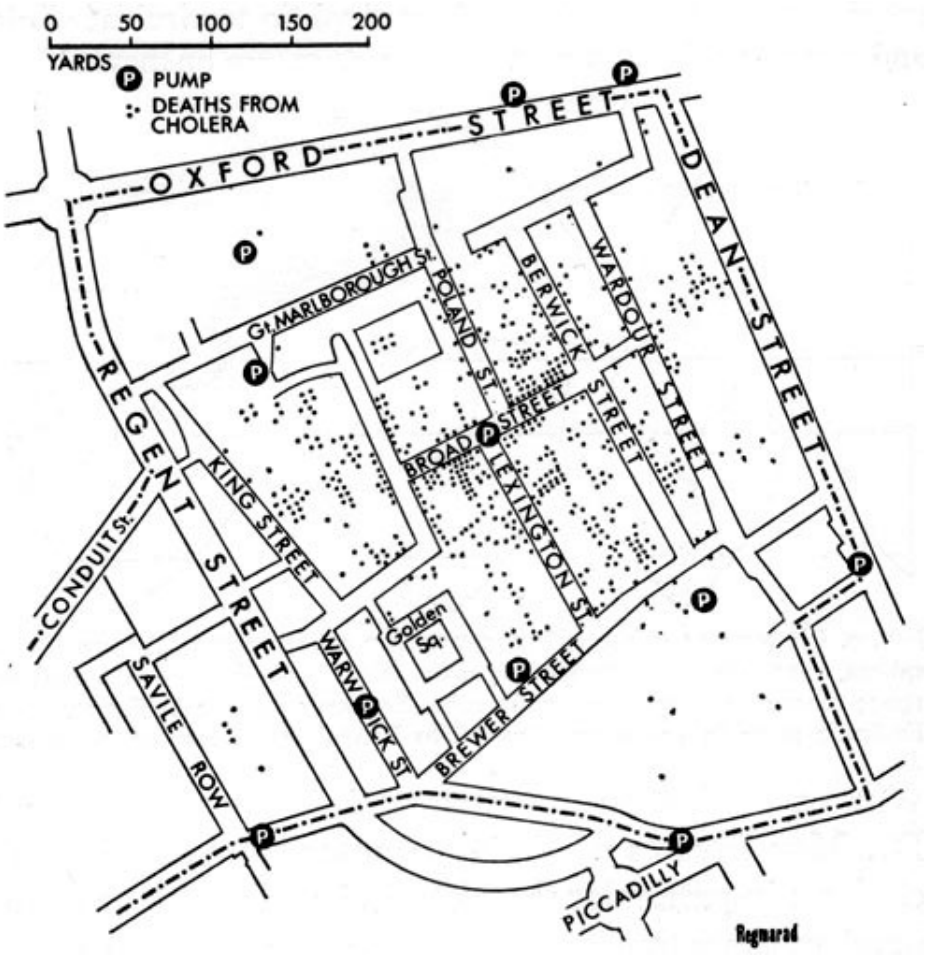
- He recognizes the existence of active principles in medical preparations
- Hypothesizes on the possibility of a match between active principle and disease
- Predicts the advent of rational drug design!

Success cases in disease prevention

- Vaccination (Jenner 1798, Pasteur 1864)
- Scurvy (Lind, 1763)
- Anti-septics (Lister, 1867)
- Control of the 1854 cholera outbreak 1854 (John Snow, 1854)

In spite of these achievements, therapeutic medicine is practically non-existent until late XIX century.

Cholera outbreak, 1854



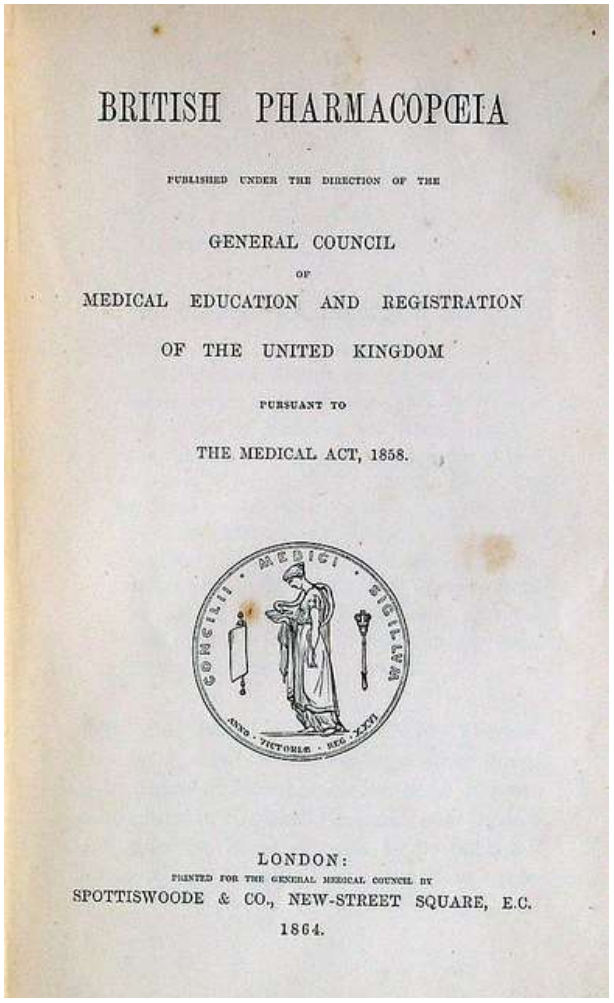
London Cholera Outbreak, 1854

Oliver Wendell Holmes (1809-1894)

“I firmly believe that if the whole Materia Medica, as now used, could be sunk to the bottom of the sea, it would be all the better for mankind – and the worse for the fishes”

-- Oliver Wendell Holmes, 1860

The status of pharmacology in the XIX century



BRITISH PHARMACOPŒIA

PUBLISHED UNDER THE DIRECTION OF THE

GENERAL COUNCIL

OF

MEDICAL EDUCATION AND REGISTRATION

OF THE UNITED KINGDOM

PURSUANT TO

THE MEDICAL ACT, 1858.

LONDON:

PRINTED FOR THE GENERAL MEDICAL COUNCIL BY

SPOTTISWOODE & CO., NEW-STREET SQUARE, E.C.

1864.

- The first edition of *British Pharmacopeia* (1864), lists 311 preparations:
  - 187 extracts plant, of only 9 are pure substances, and almost none of them with pharmacological action
  - 103 inorganic chemical substances: iron, iron sulfur, sodium bicarbonate and many toxic salts of arsenic, lead and mercury
  - Some synthetic compounds, like diethyl ether and chloroform
  - Some animal products

Dawning of the pharmaceutical industry (end of XIX century)

- Biomedicine & Pharmacology
- Development of Synthetic Chemistry
- European Chemical Industry
- Trade of medical supplies

All of these gave a decisive boost to the search for new synthetic compounds.

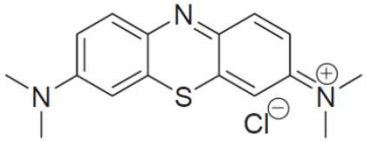
Pharmacology & Biomedicine

- Cell Theory (Virchow, 1858)
- Dorpat Pharmacological Institute (Buccheim, 1847)
- Physiology (Claude Bernard)
- Microbial theory of disease (Pasteur, 1878)
- Direct observation of pathogens (Koch)
- Beginning of chemotherapy (Paul Erlich)

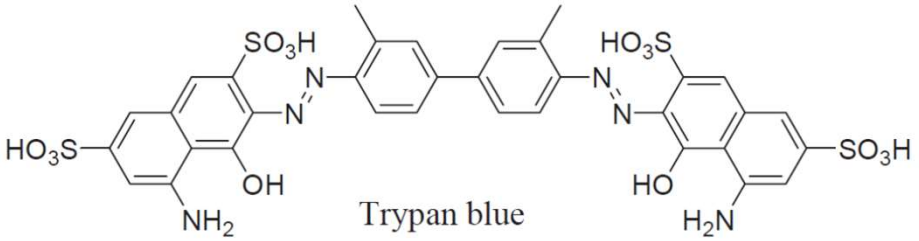
Ehrlich and the dawn of rational drug design

- Study of the selective affinity of dyes for the different cellular structures
- Searching for chemical compounds with therapeutic activity
- Concept of "receptor” e “magical bullet”
- Diphtheria anti-toxin
- Anti-syphilitic drugs (Salvarsan and Neosalvarsan)
- Theory of antibody action
- The first organized effort to modify the activity of a lead compound through systematic chemical modifications
- Ehrlich coined the word *chemotherapy*

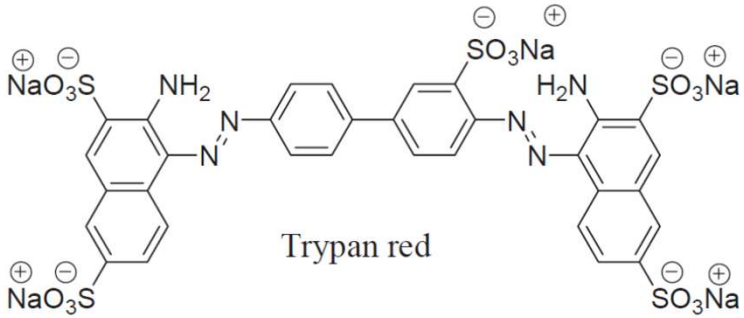
Paul Ehrlich
(1854-1915)



Methylene blue

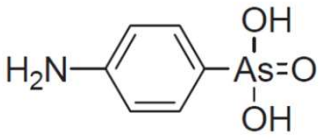


Trypan blue



Trypan red

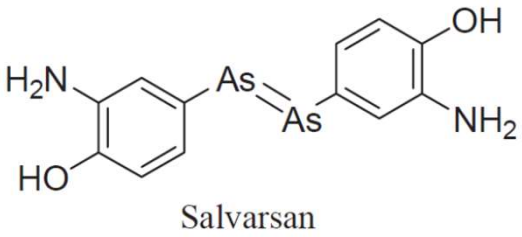
Dyes studied by Erlich



Atoxyl

African trypanossomiasis

Rabbit animal model of syphilis

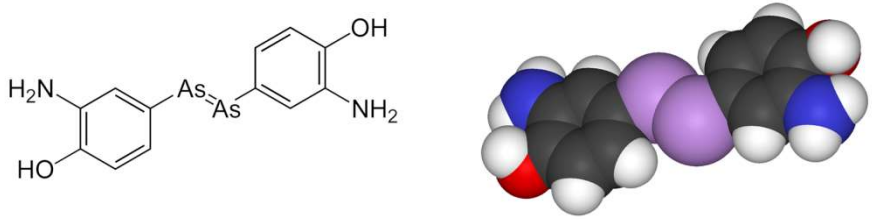


Salvarsan

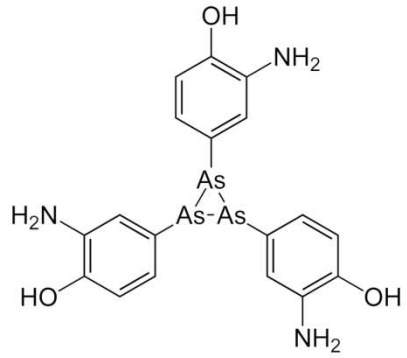
Syphilis

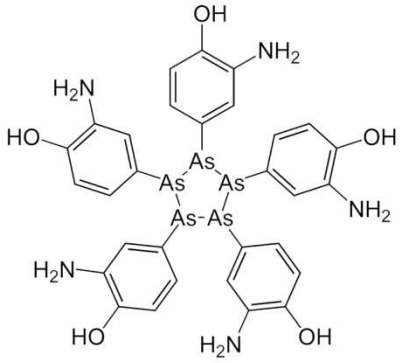
Arsphenamine (Salvarsan)

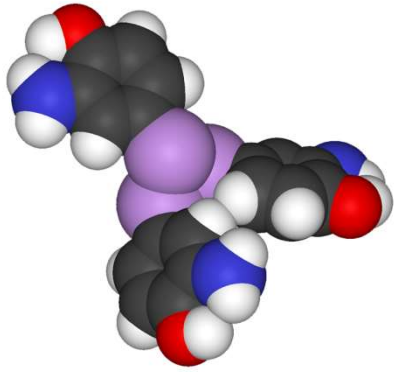
Before 2005

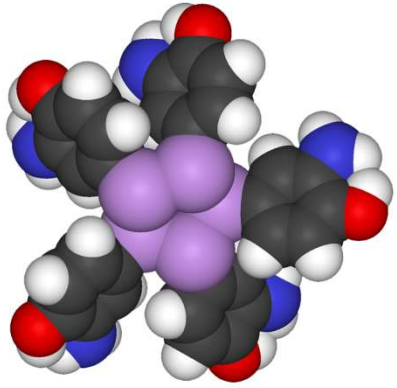


Structures resolved in 2005 using Mass Spectrometry





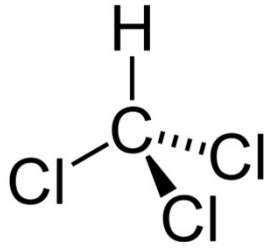


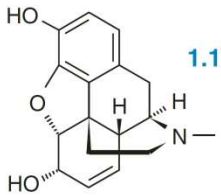


Lloyd NC, Morgan HW, Nicholson BK, Ronimus RS (2005). ".*Angew. Chem. Int. Ed. Engl.* **44** (6): 941–4

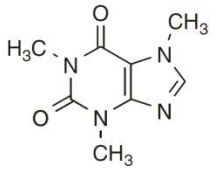
Synthetic chemistry and the development of new drugs

- The first synthetic compounds finding medical use were anesthetics rater than therapeutic agents
- Diethyl ether synthesis in 1540
- Humphrey Davy synthesizes nitrous oxide (N_2O) in 1799
- These compounds where used as anesthetics starting from 1840, as well as chloroform
- The chemical industry of dyes gave a decisive boots to synthetic organic chemistry
- Valence theory and benzene structure (von Kekulé, 1865)

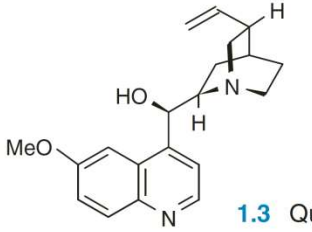




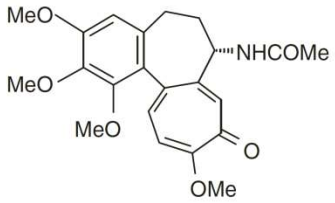
1.1 Morphine



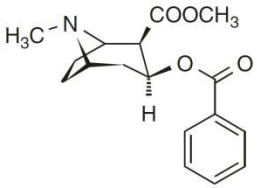
1.2 Caffeine



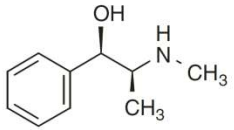
1.3 Quinine



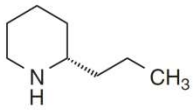
1.4 Colchicine



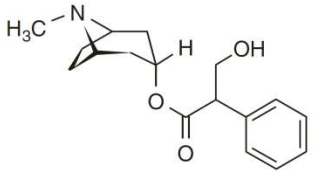
1.5 Cocaine



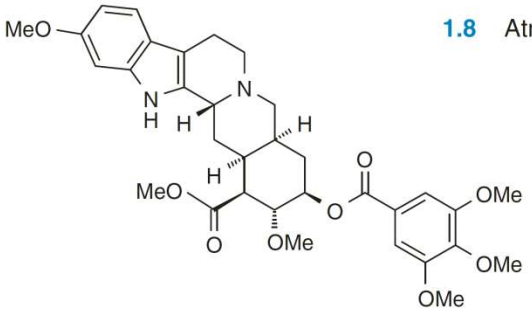
1.6 Ephedrine



1.7 Coniine



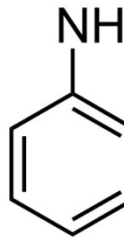
1.8 Atropine (racemate)



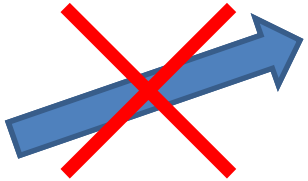
1.9 Reserpine

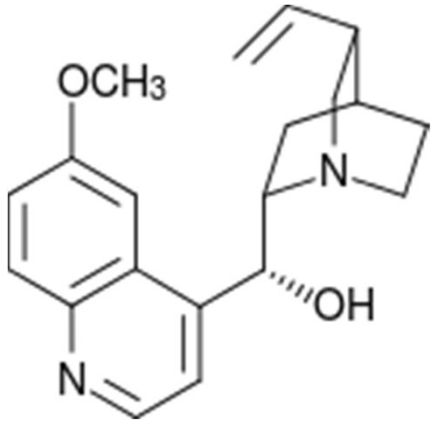
Serendipity: the discovery of *mauvein*

Perkin 1856



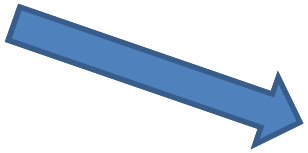
Anilin

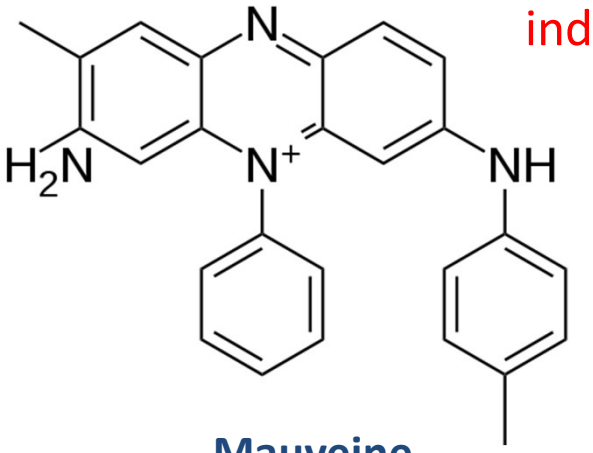




Quinine

Strong purple dye, its discovery kick started the synthetic dye industrv.

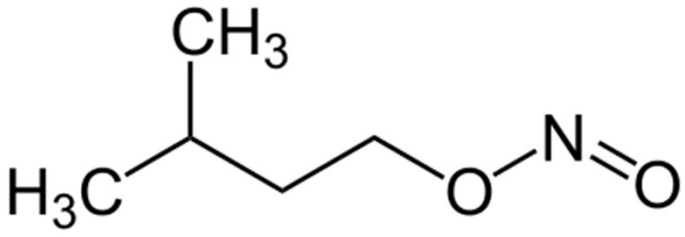




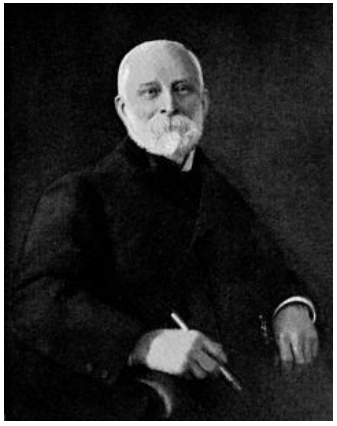
Mauveine

A failed attempt to synthesize quinine from aniline lead to the discovery of the first synthetic dye, mauveine!

The first synthetic drug: amyl nitrite



Frederick Guthrie
(1833–1886)



Lauder Brunton
(1844-1916)

- Synthesis by Guthrie in 1859
- Very powerful vasodilator
- Used by Brunton in 1864 for treating angina pectoris
- 40 years would pass before another synthetic drug was created

XX Century

- Racional Drug Design
- Molecular Genetics
- Genomics (and other “omics”)
- High throughput screening
- Structural methods (NMR, X-ray, etc)
- Molecular Modelling
- Systems Biology

Pharmacology at the turn of XX century

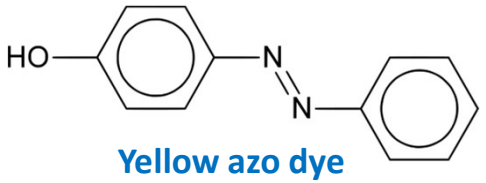
- Several convergent approaches:
- Animal models
- Target identification
- Advances in synthetic chemistry
- Molecular structure and bonding theories
- Birth of quantitative Enzymology
- First attempts at racional drug design

Synthetic chemistry dominates drug discovery, but the ideas of Fischer and Erlich raise interest in the analysis of the *targets* of pharmacologically active substances.

At this time, mechanism is almost always overlooked, with the main focus on the optimization of therapeutic effect, in what it is mostly a trial and error approach (irrational drug design).

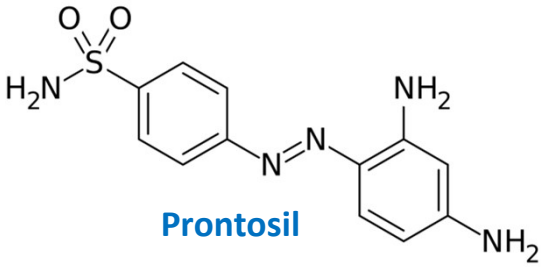
An example of “classic” drug design: sulfa drugs

- Gerhard Domagk from IGFarben researches some azo dyes with antibacterial properties and low toxicity in humans.



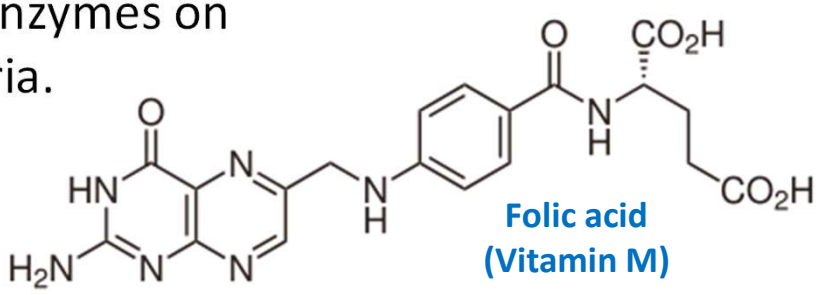
Yellow azo dye

- Sulfonamidochrysoidine is marketed in 1935 under the trade name *Prontosil* by Bayer, the first commercially available antibacterial drug and starting point for the family of sulfonamide compounds produced in the following years.



Prontosil

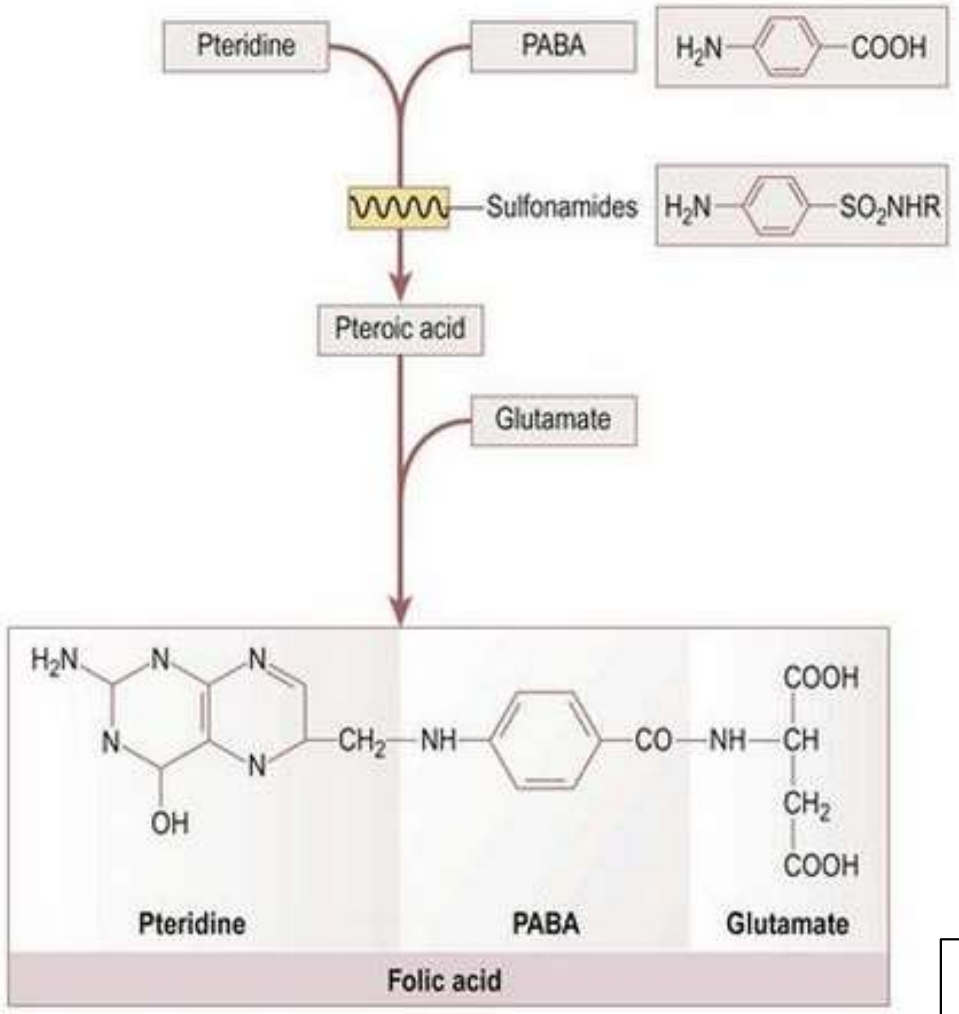
- In 1940, D.D.Woods discovers that sulfonamides are competitive inhibitors of DHPS, one of the enzymes on the folic acid biosynthesis pathway in bacteria.

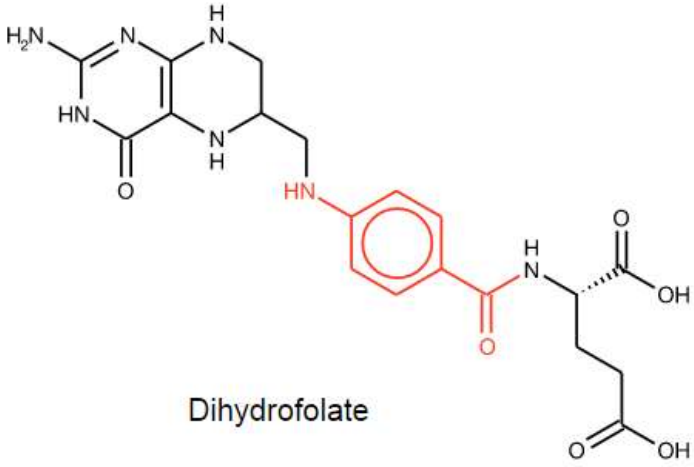


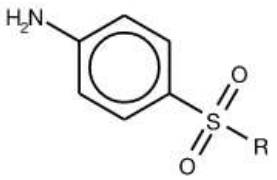
Folic acid
(Vitamin M)

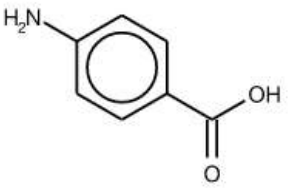
DHPS - Dihydropteroate synthase

Sufanilamides are structural analogues of *p*-aminobenzoic acid









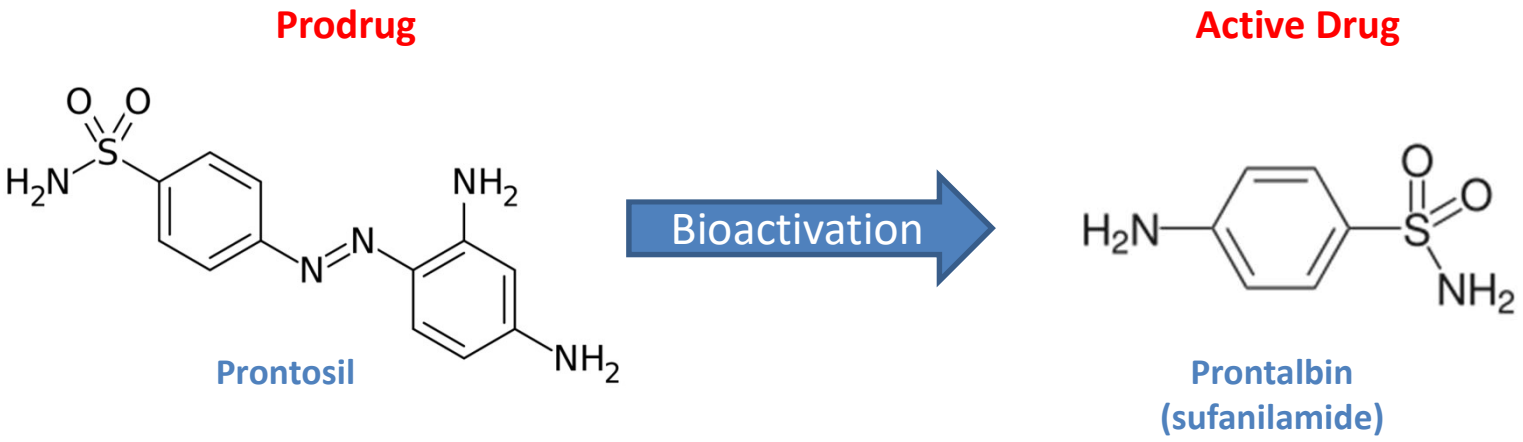
Sufanilamides are structural analogues of PABA (competitive inhibition)

PABA – *p*-aminobenzoic acid

Prontosil is a prodrug

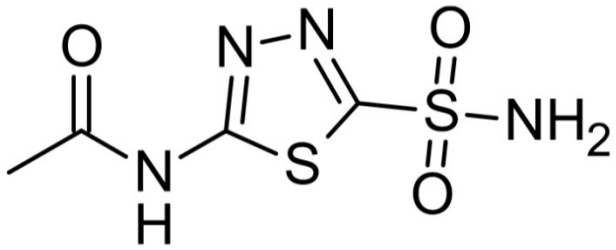
- In late 1935, working at the Pasteur Institute in Paris in the laboratory of Dr. Ernest Fourneau, Jacques and Thérèse Tréfouël, Dr. Daniel Bovet and Federico Nitti discovered that Prontosil is metabolized to sulfanilamide, a much simpler, colorless molecule, reclassifying Prontosil as a prodrug
- Sulfanilamide was market by Bayer under the trade name *Prontalbin*

- These findings help establish the concept of **bioactivation**, the process by which a **prodrug** is metabolized in the body to an active drug.



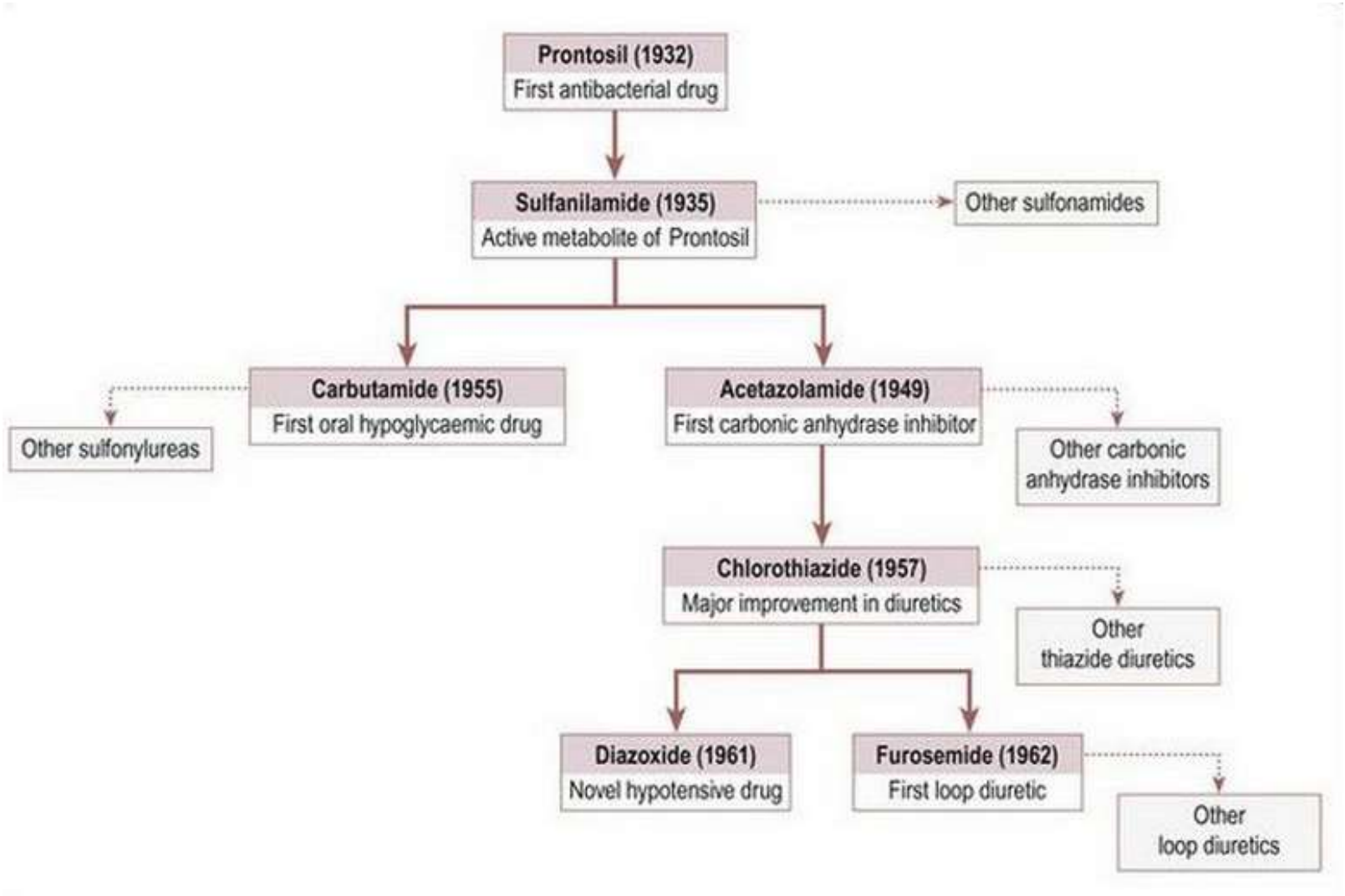
Serendipitous side effect: carbonic anhydrase inhibition

- Some sulfonamides were found to be diuretic (unexpected side effect)
- The discovery of carbonic anhydrase in 1940, and its role in bicarbonate secretion, lead to the experimental demonstration of the inhibitory effect of some sulfonamides on this enzyme.
- Modification of the structure of diuretic sulfonamides led to the making of the first commercially available carbonic anhydrase inhibitor, *acetazolamide*, marketed as a diuretic drug under the trade name *Diamox* (1952).



Acetazolamide

Sulfa family tree



Lessons from the sulfa story

Transition from the *synthetic chemistry* paradigm to *therapeutic target* paradigm (target-derived drug design).

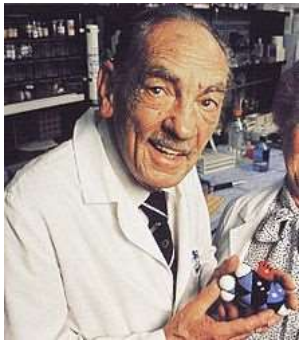
The active drug may be a metabolic product of a prodrug.

Serendipity in the discovery of new drugs: the diuretic action was an unsought side effect of sulfa drugs, but the researchers were able to recognize its utility.

"Chance favors only the prepared mind."
-- Louis Pasteur

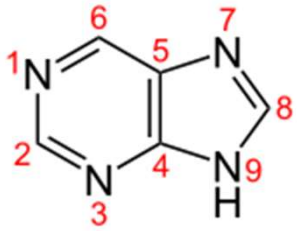
The “anti-metabolic principle”

- George Hitchings and Gertrude Elion worked together a the Wellcome Research Labs (1944). Development of inhibitors of folic acid biosynthesis
- Search for potential anti-metabolites for the purine and pyrimidine biosynthetic pathways
- Discovery of the enzyme DHFR (dihydrofolate reductase)
- Discovery of DHFR inhibitors with specificity towards particular microbial species.
- Development of several drugs with anti-bacterial, anti-cancer and immunosuppressive action
- Development of allopurinol, a Xanthine Oxydase inhibitor effective in gout treatment.
- They received the 1998 Nobel Prize in Phys. & Med.

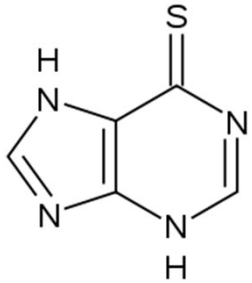


George Hitchings
(1905-1998)

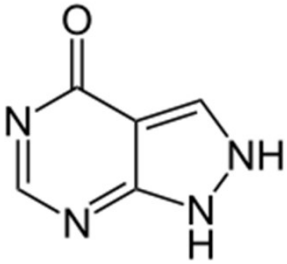
Gertrude Elion
(1918-1999)



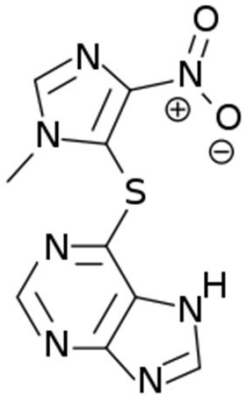
Purine



Mercaptopurine



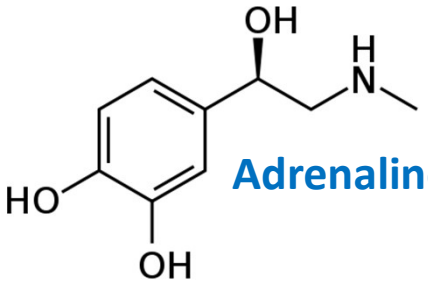
Allopurinol



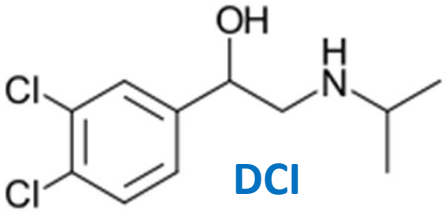
Azathioprine

Receptor pharmacology

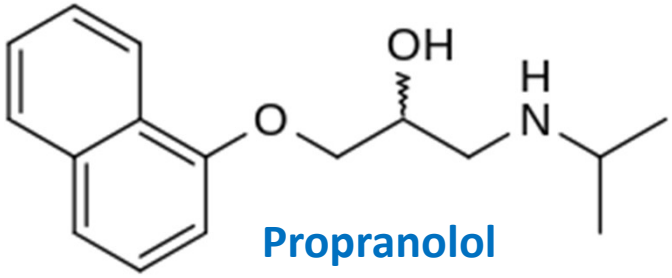
- James Black develops the first beta-blocker in 1960, pronethalol
- Pronethalol was found to be carcinogenic in mice, and was quickly replaced by *propranolol*
- This was the first a drug designed based on a previous specification of its target (the *β-adrenergic receptor*).
- Propranolol was marketed in 1964 under the name *Inderal*
- Propranol is an non-selective *antagonist* of the β-adrenergic receptors that revolutionized management of angina pectoris and later became the world's best selling drug



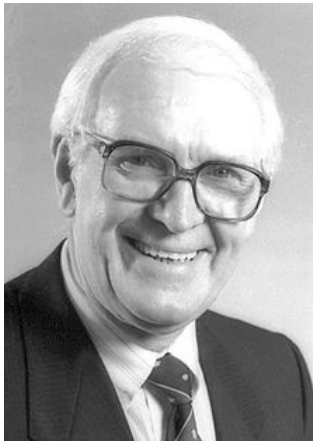
Adrenaline



DCI



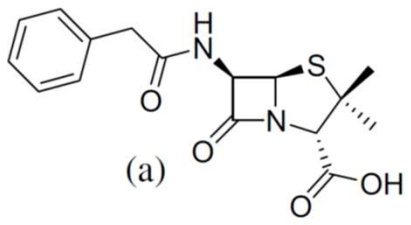
Propranolol



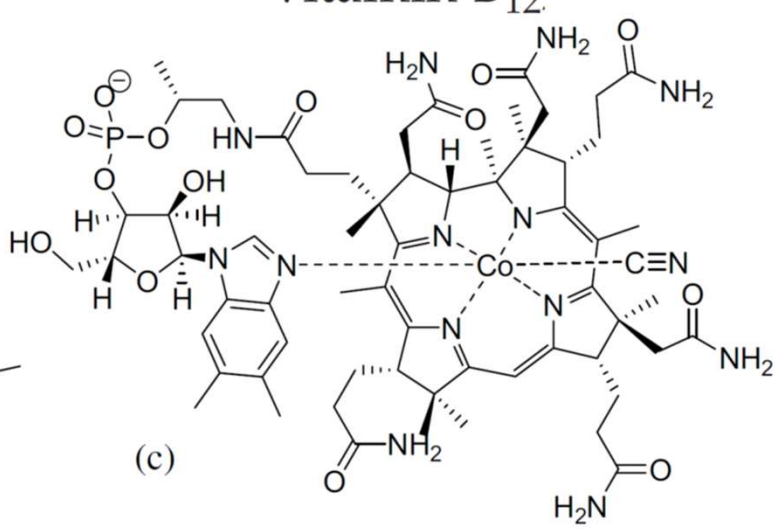
James Black
(1924-2010)

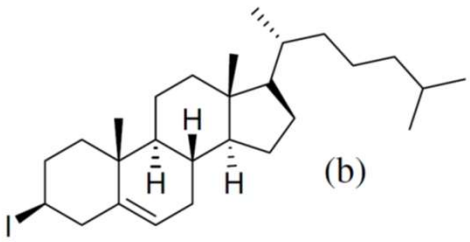
Structural chemistry

benzylpenicillin

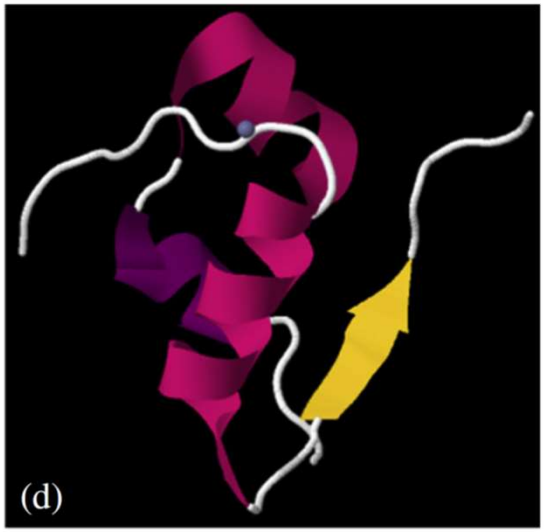


vitamin B_{12}





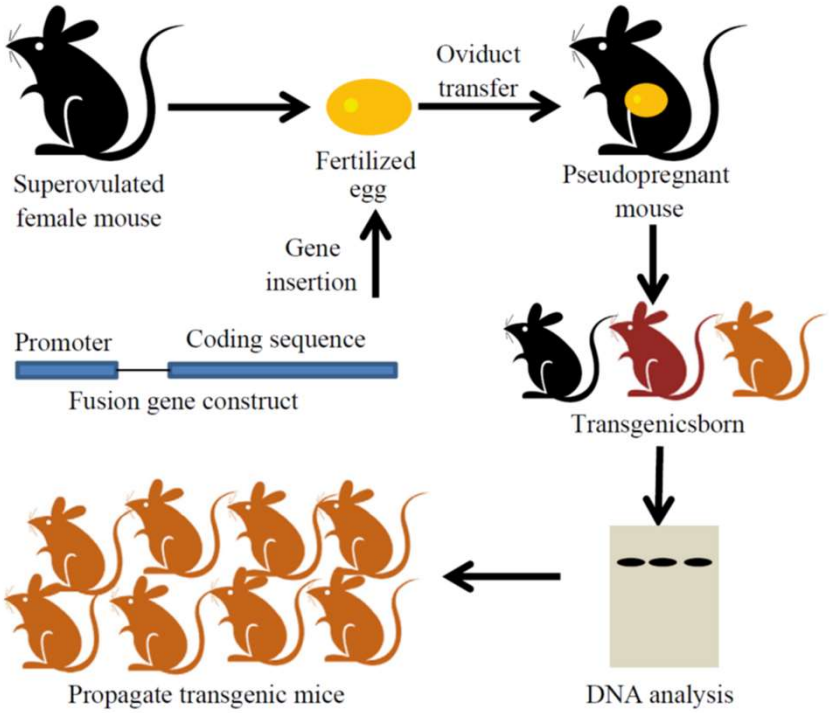
cholesteryl iodide,



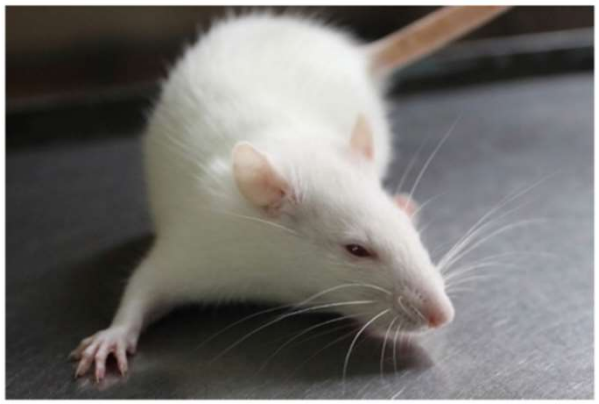
insulin

- The development of crstllographic methods in the early XX century permitted the discovery of many chemical structures, from simple to complex
- Quantum mechanics provide the theoretical framework to understand chemical bonding and reactivity

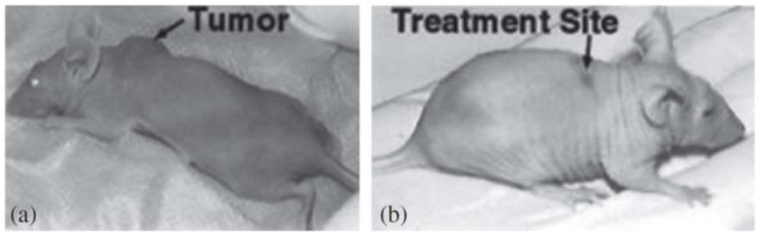
Animal models



Transgenic animal models

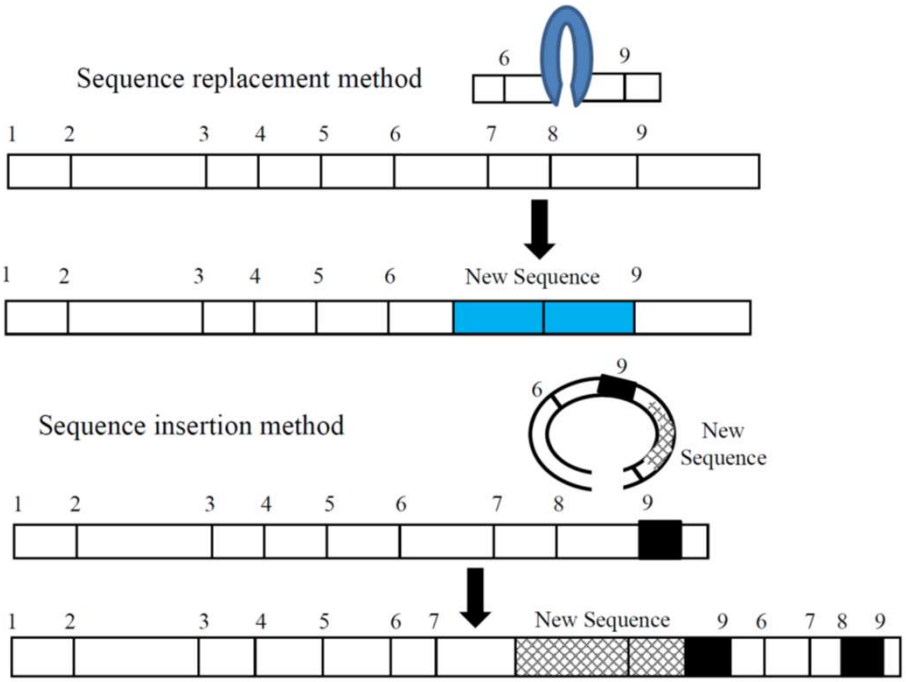


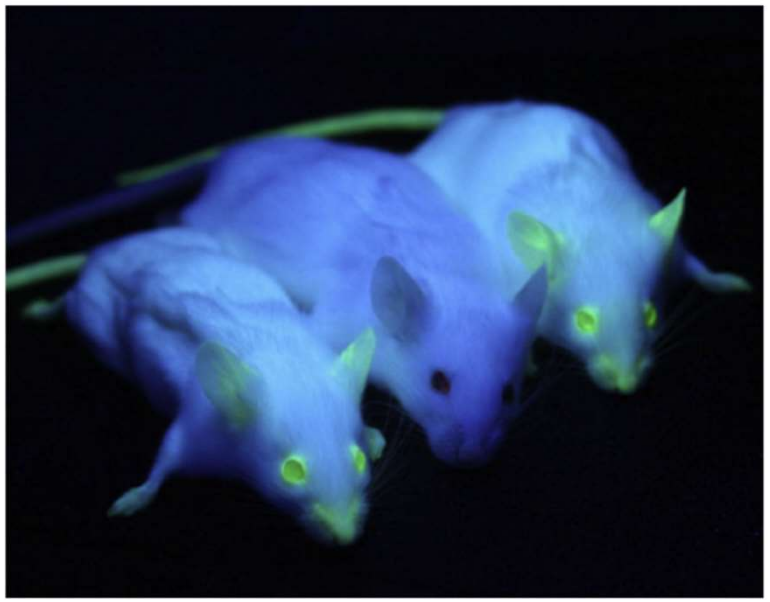
Wistar rat



Nude mice

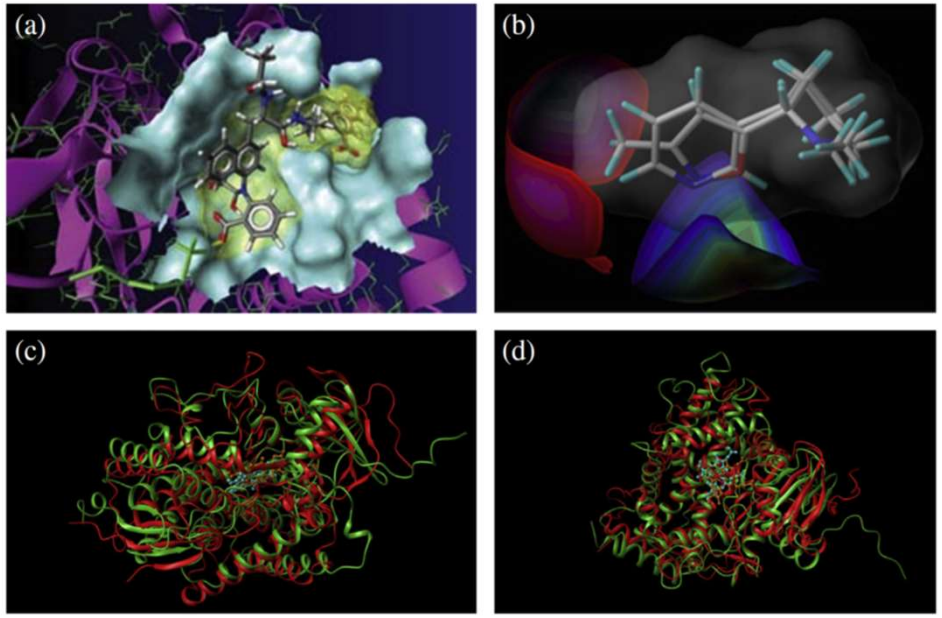
Molecular Genetics





Transgenic insertion of GFP

Computational methods



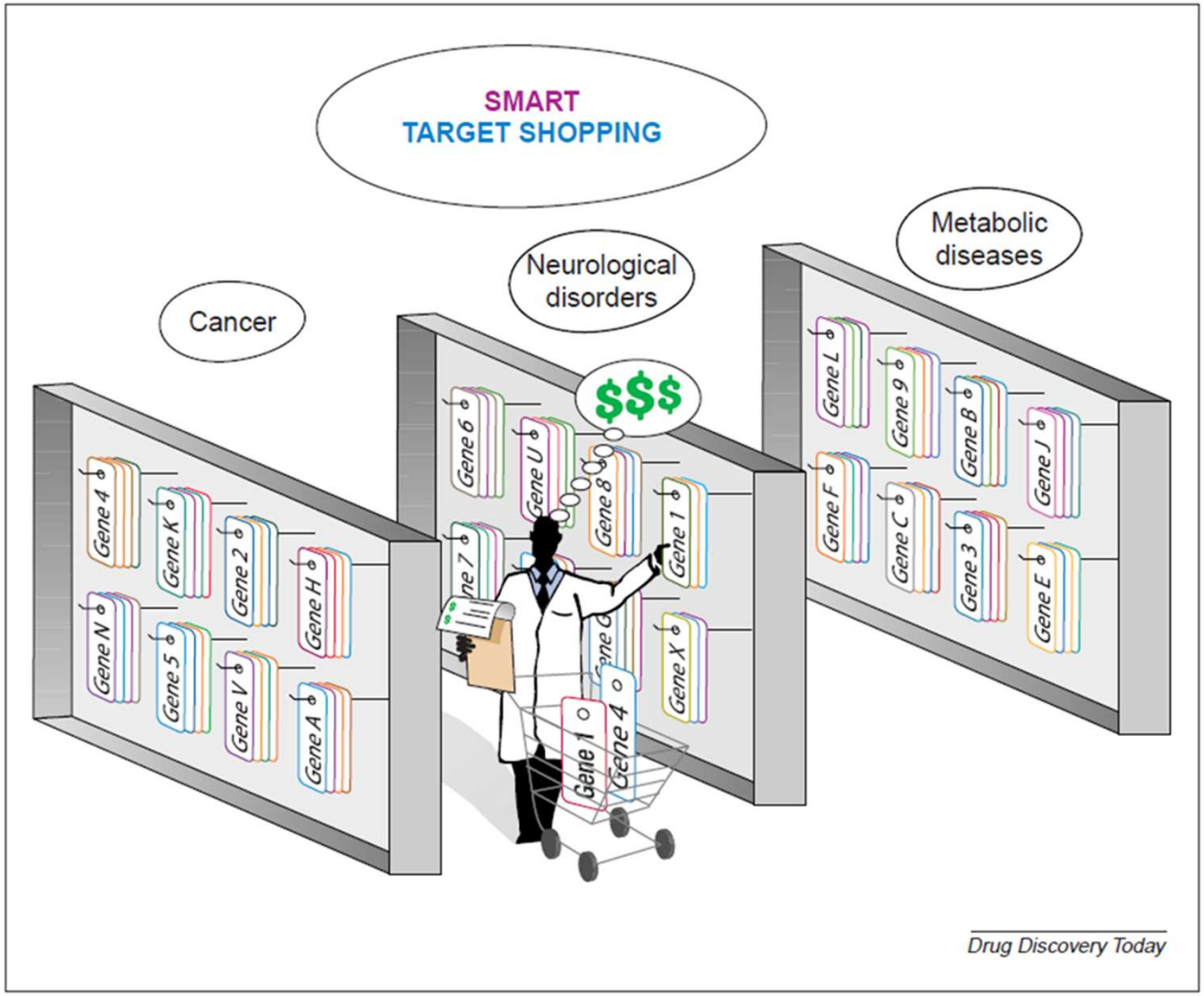
IBM 709 (1958)

A new paradigm: Structure-based drug design

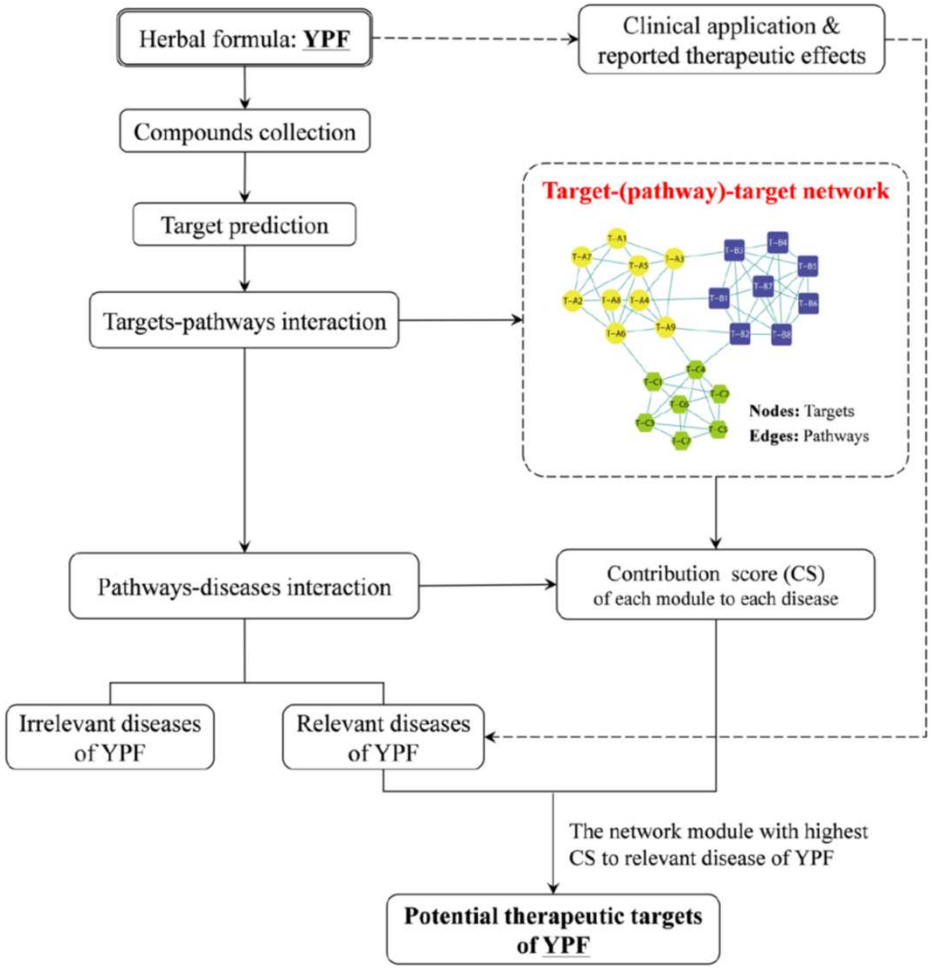
XX century's last quarter has witnessed multiples advances in several areas of crucial impact in drug development:

- DNA structure and protein synthesis mechanism, leading to molecular genetic techniques
- Big advances in the methods for the elucidation of the 3D structure of proteins
- Complete draft of the Human Genome and development of new bioinformatics tools to analyze it.
- High-throughput screening methods for the discovery of new lead compounds
- Advances in both hardware, methods and algorithms for the computational modeling of proteins, ligands and their interactions (docking, virtual screening, molecular dynamics, QM)
- Real and virtual fragment libraries, fragment-based design, click chemistry, cheminformatics methods

Systems Biology and Smart Target Finding

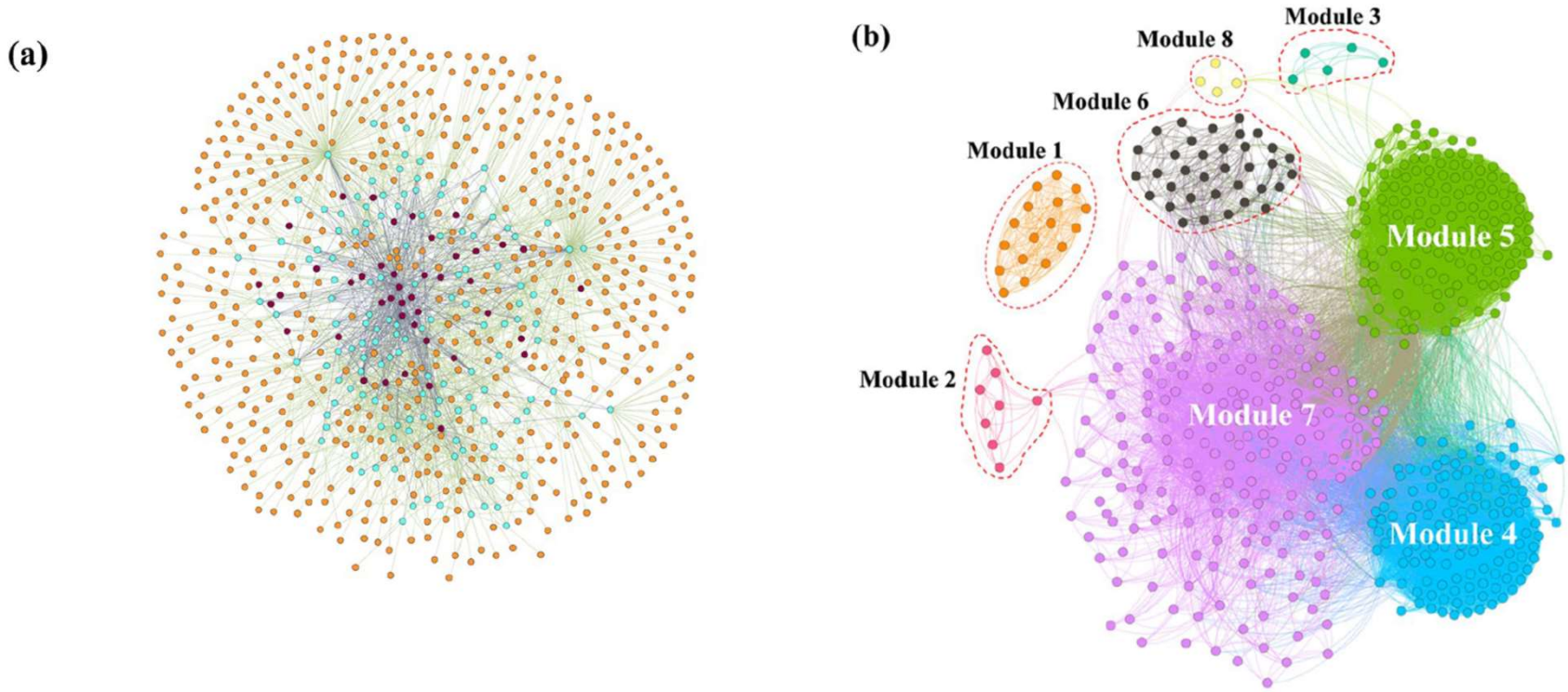


Network Pharmacology



Zuo (2018) *Scientific Reports* ***8****:11418*

Network Pharmacology



Zuo (2018) *Scientific Reports* ***8****:11418*