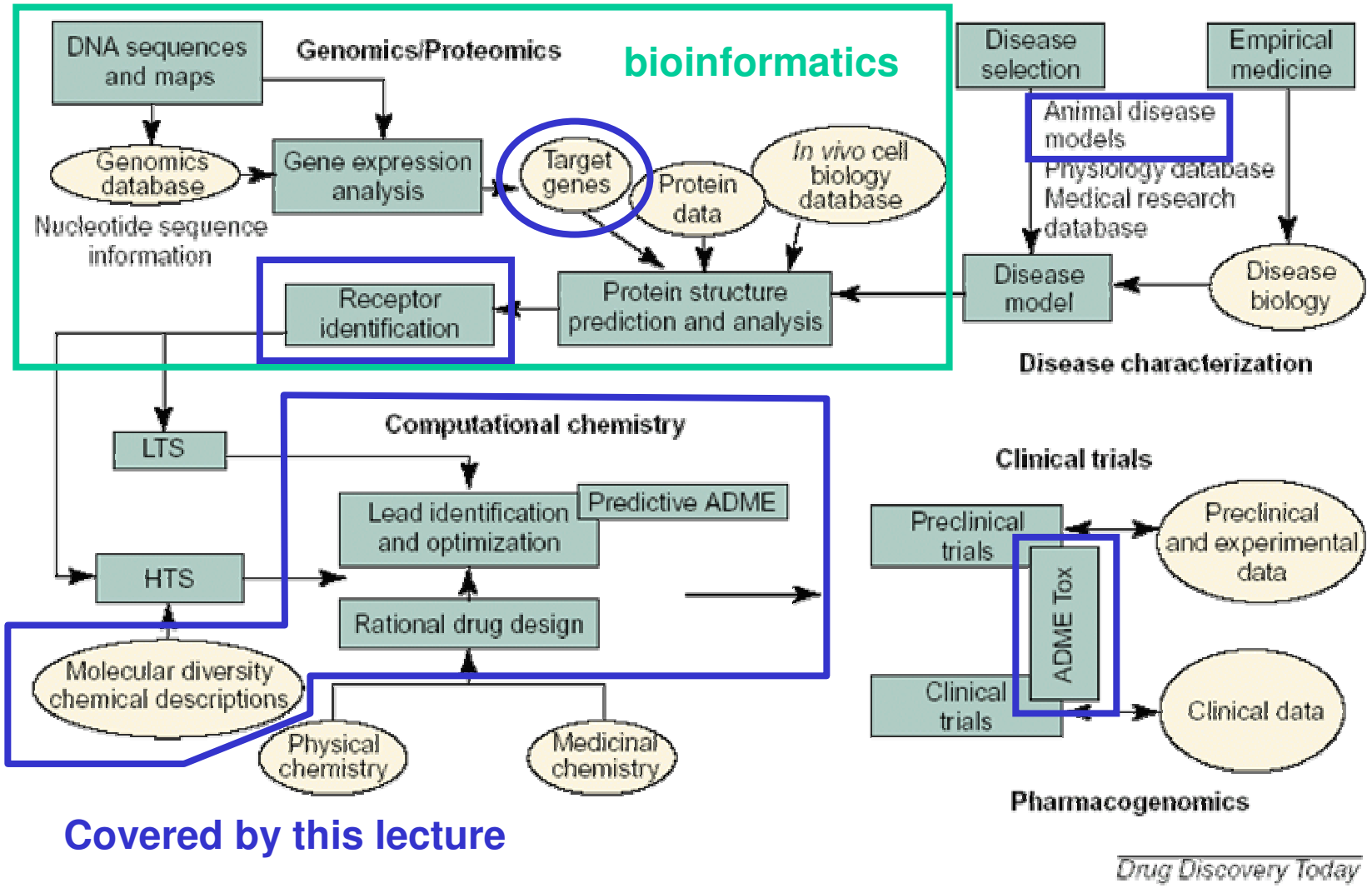
# Modern Methods in Drug Discovery

Aims of this course:

- comprehensive knowledge about all processes in the *drug discovery pipeline*
- in particular *in silico* methods of *drug design*

# flow of information in a *drug discovery pipeline*




**Covered by this lecture**

# Related topics not covered by this lecture

medicinal chemistry
organic synthesis
biopharmaceutical aspects
  (tissue models, non-oral administration)
clinical aspects
molecular modelling theory
homology modelling theory
docking basics and applications
computational chemistry
genome, proteome, metabolome
bioethics and patent law

# Required knowledge

Use of tools for sequence analysis, e.g. BLAST, CLUSTALW
Use of visualizing tools, e.g. BALL, VMD, SPDBV, JMOL

**recommended courses:**

Softwarewerkzeuge der Bioinformatik
Computational Chemistry
Bioinformatics I + II

**Actual applications during the excerices and homework:**

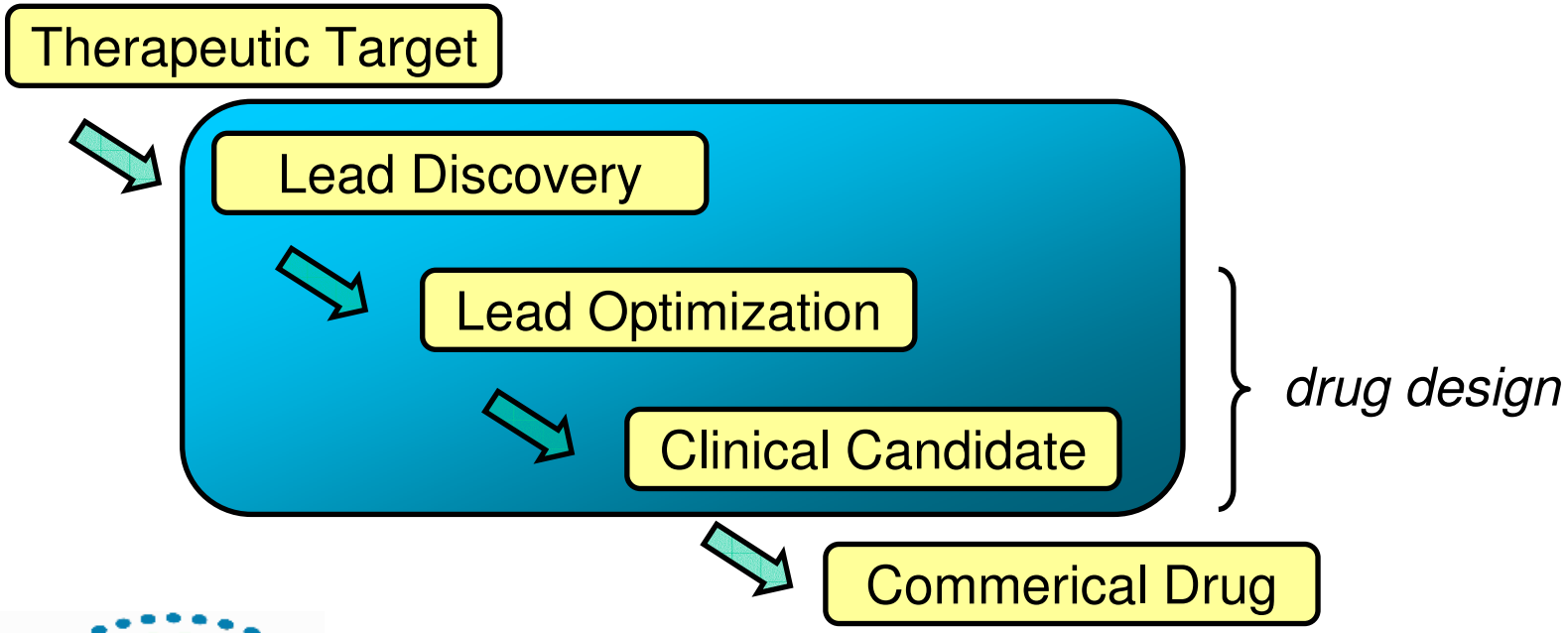
multiple sequence alignment, homology in sequences
simple homology modelling
protein-ligand interactions

SMILES and SMARTS notation of chemical structures
using SMARTS with Open Babel

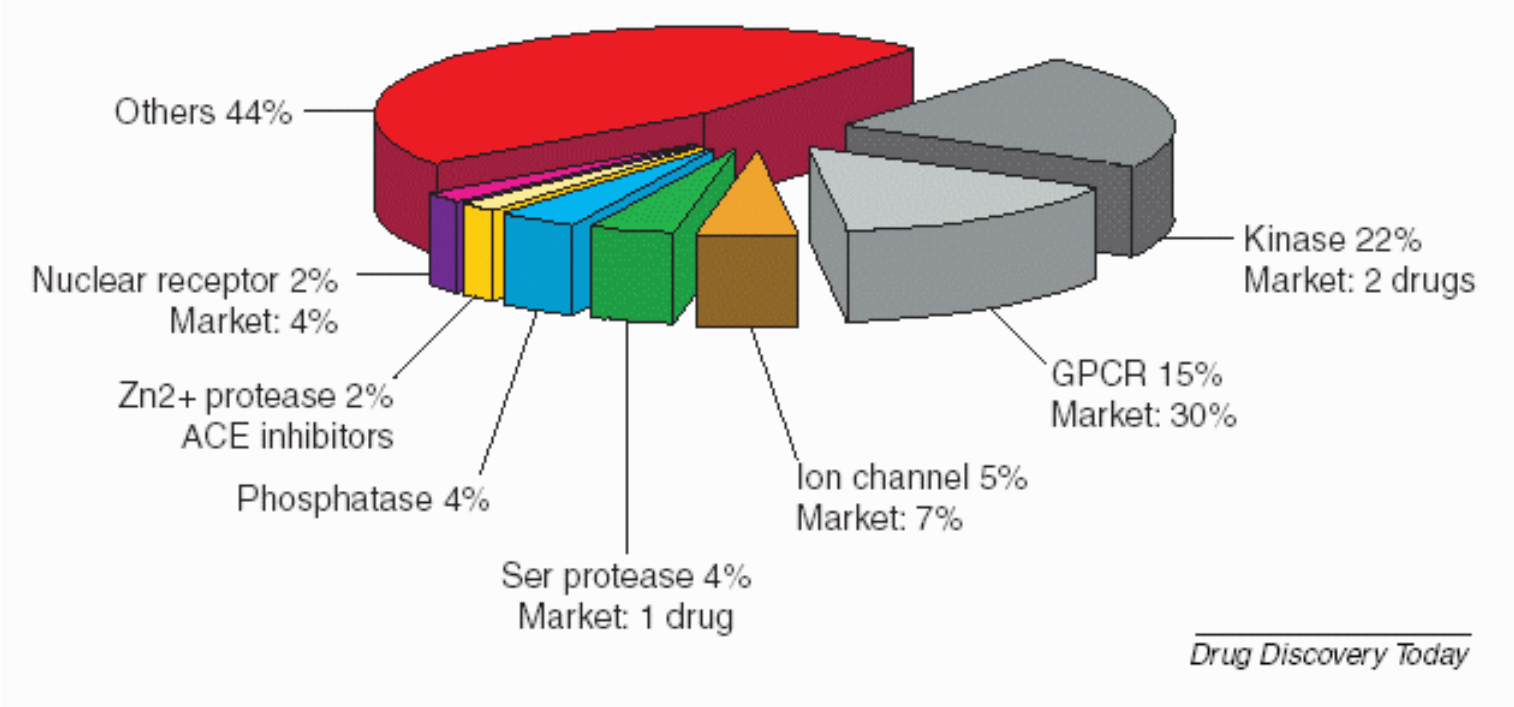
substance database queries (PubChem, ChEMBL,...

# What is *drug discovery* ?

rational and targeted search for new drugs

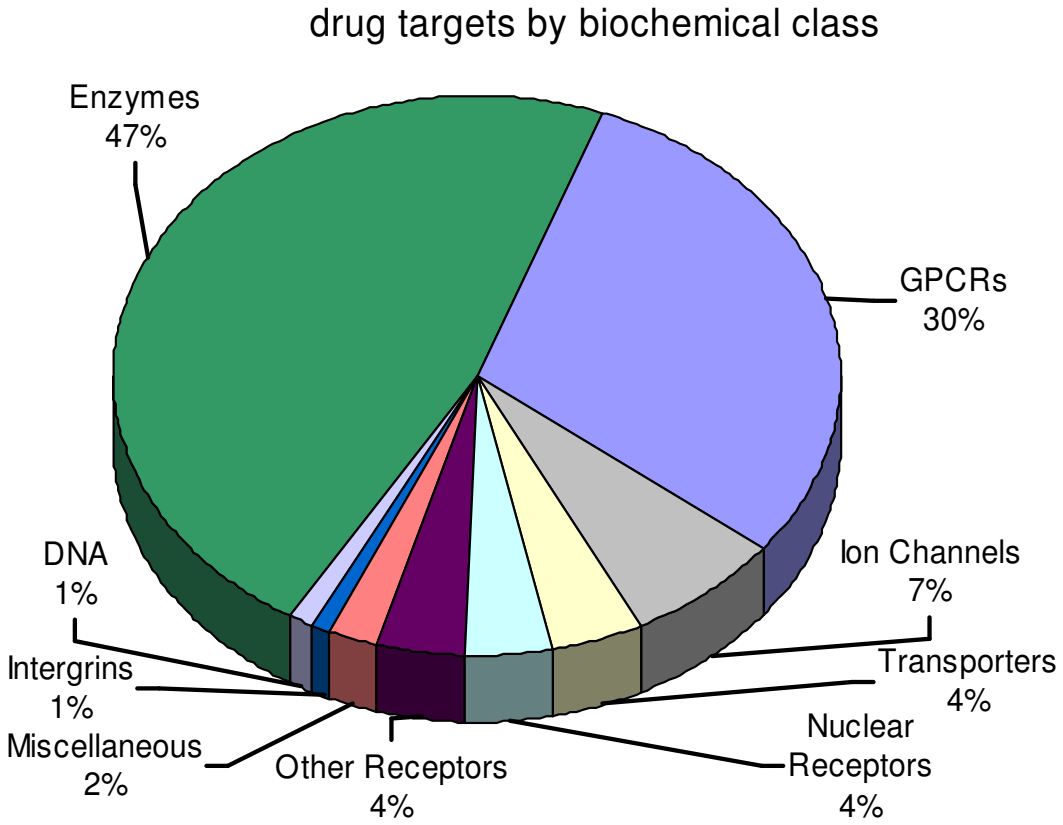


# typical targets (I)




contribution to the human genome and marketed drugs

# typical targets (II)




Fractional content of marketed drugs according to their biochemical targets

data: Hopkins & Groom, *Nat.Rev.Drug.Disc.* **1** (2002) 727

# preliminary schedule (lectures)

1. Introduction, overview, recap of chemical structures
2. typical diseases
3. properties of drugs and their mode of action
4. Substance databases and bioisosteric compounds
5. QSAR, statistics and descriptors
6. ADME models
7. metabolism and toxicology
8. target identification, animal models
9. cytochrome P450, polymorphisms, transporters
10. more complex diseases malaria, obesity
11. in silico prediction of molecular properties
12. current trends, disease vs. lifestyle drugs, doping

# preliminary schedule (exercises)

Biweekly in the CIP-Pool (building E 2.1 room 003 )
computer account and access card required

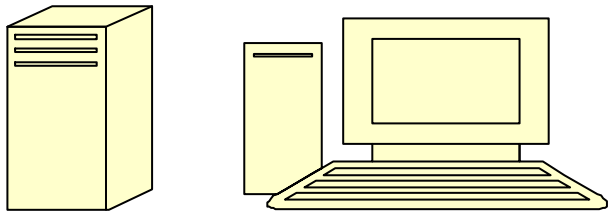
- chemical structures of drugs
- enzyme-ligand interactions, analysis of .pdb files
- substance databases and SMARTS queries
- QSAR, statistics and descriptor handling
- ortholog targets in model organisms
- further online tools

# Compound Databases

## existing substance libraries

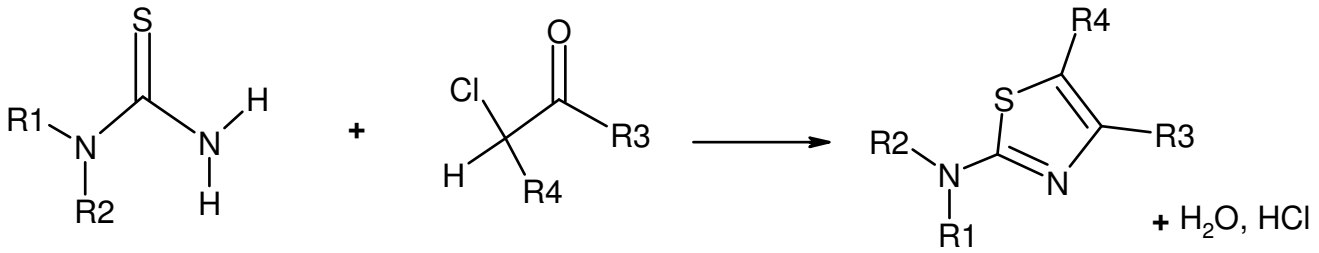
| | | |
|---|---|---|
| ACD | >100,000 chemicals | commercial |
| World Drug Index | 58,000 compounds | |
| USAN | <10,000 in clinical trial | |
| virtual library | ≈100,000 compounds | company, in house |

| | | | |
|---|---|---|---|
| PubChem | > 3,000,000 compounds | NCBI | academic |
| ChEMBL | > 1,213,000 compounds | EMBL | |
| ZINC | >73,126,243 compounds | UCSF | |

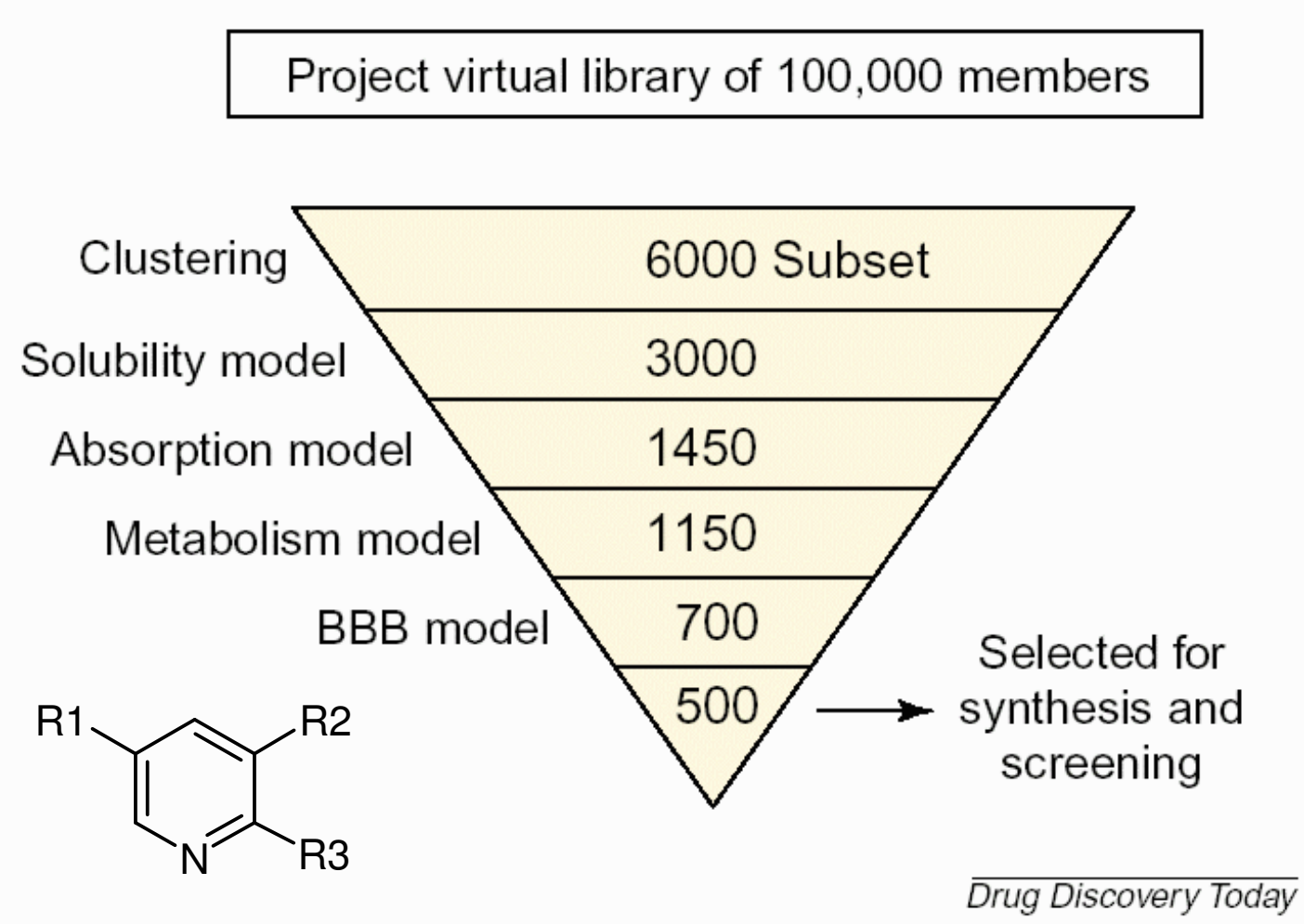


Investment per new chemical entity: >500,000 $
New chemical entities per year: ca. 15

# Methods of Combinatorial Synthesis for High Throughput Screening (HTS)



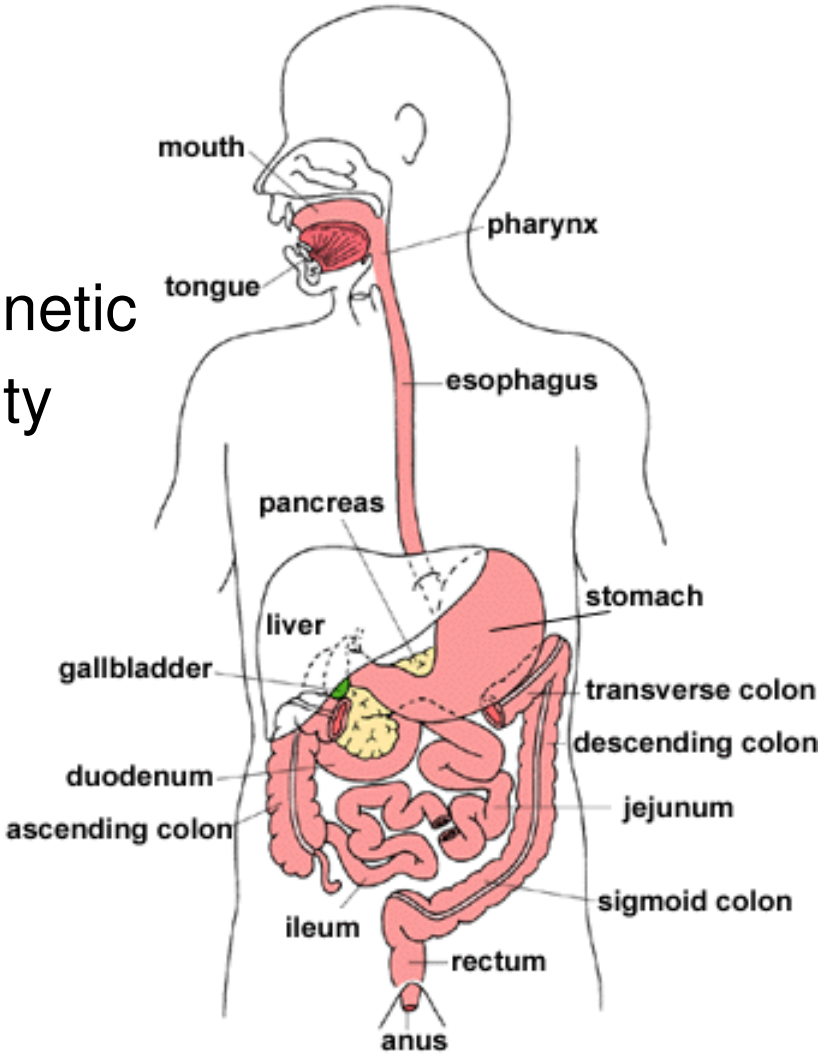

# Selection of compounds for High Throughput Screening (HTS)



# Predictive ADME

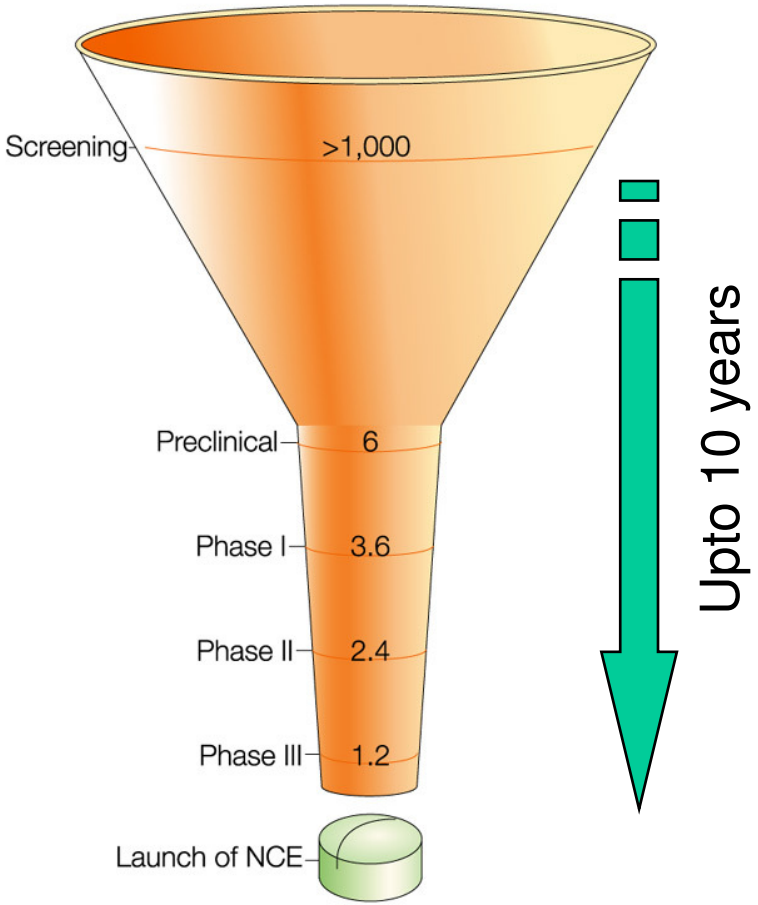
Absorption
Distribution
Metabolism
Elimination

} Pharmacokinetic Bioavailability



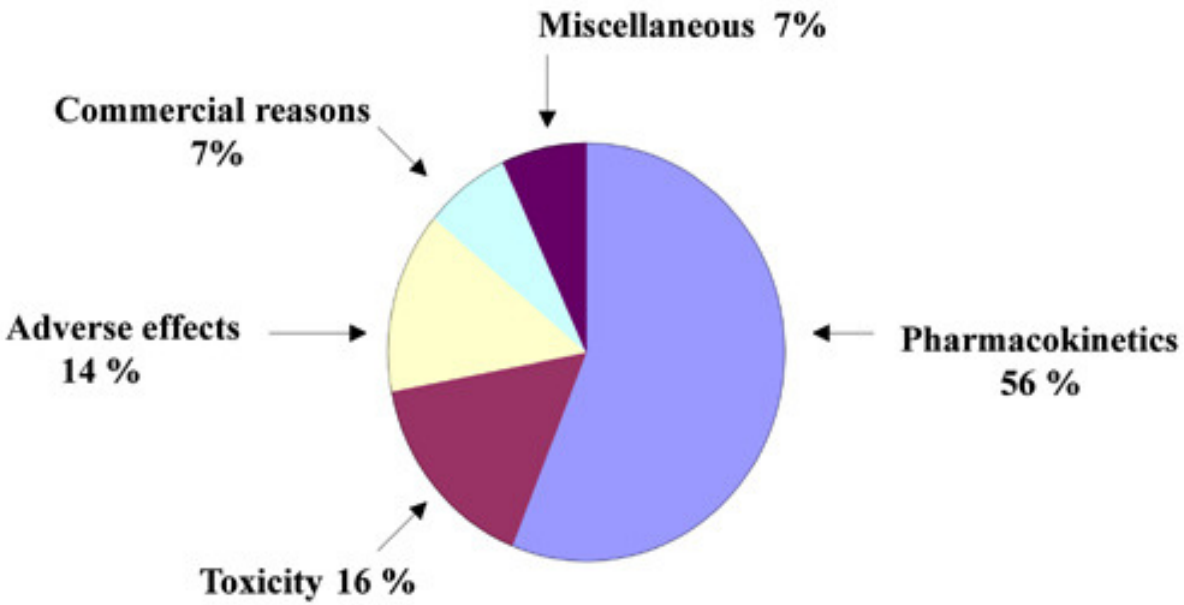
# From the pipeline until the commerical launch

For each actual marketed drug (*new chemical enitity, NCE*) there have been more than 1000 substances that underwent screened *in vitro*. Without the use of available computer-based ADMET filters, this number would be even larger.



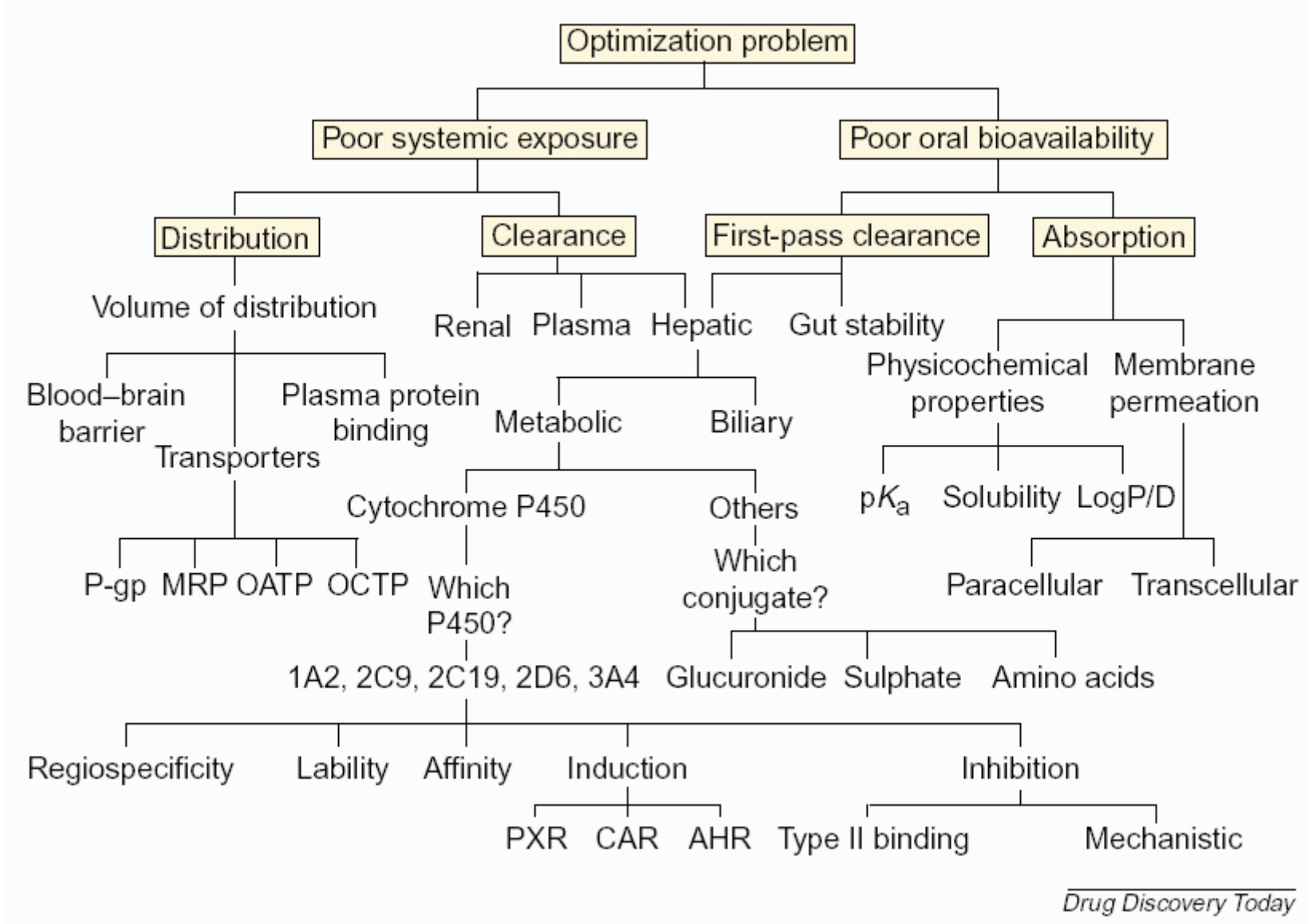

Nature Reviews | Drug Discovery

# Why is the prediction of ADME parameters that important ?




Reasons that lead to failure or withdrawl of a potential drug

# pharmacokinetics and bioavailability



# (Some) descriptors based on molecular properties used to predict ADME properties

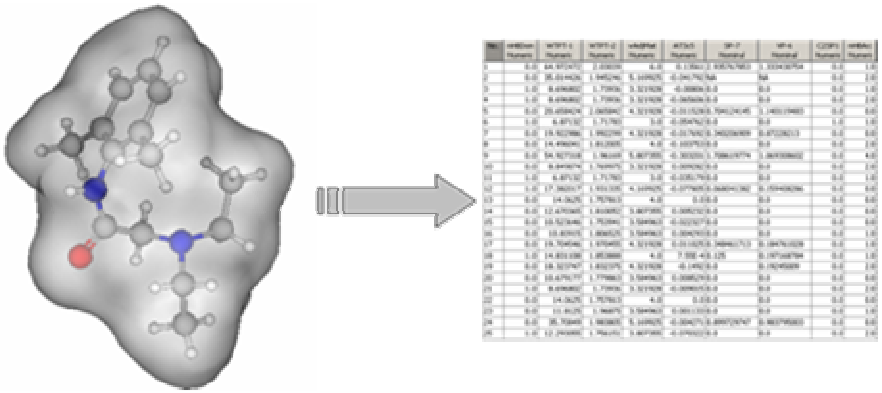
logP  water/octanol partitioning coefficient

Lipinski‘s rule of five

topological indices

polar surface area

similary / dissimilarity



QSAR quantitative structure activity relationship

QSPR quantitative structure property rel.

# metabolism

(bio-)chemical reactions of xenobiotics in the body

*First pass effect:*

Extensive metabolization of mainly lipophilic molecules, such with MW>500, or those that have a specific affinity to certain transporters, during the first passage through the liver

Phase I:

Oxidation, reduction and hydrolysis → esp. cytochrome P450 enzymes

Phase II:

Conjugation with small molecules (e.g. glutamine)

Phase III:

elimination by transporters

# cytochrome P450 enzymes

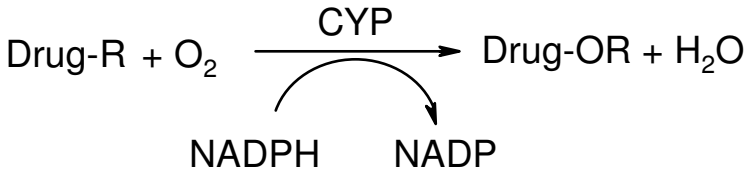
Flavin monooxygenase isoenzme

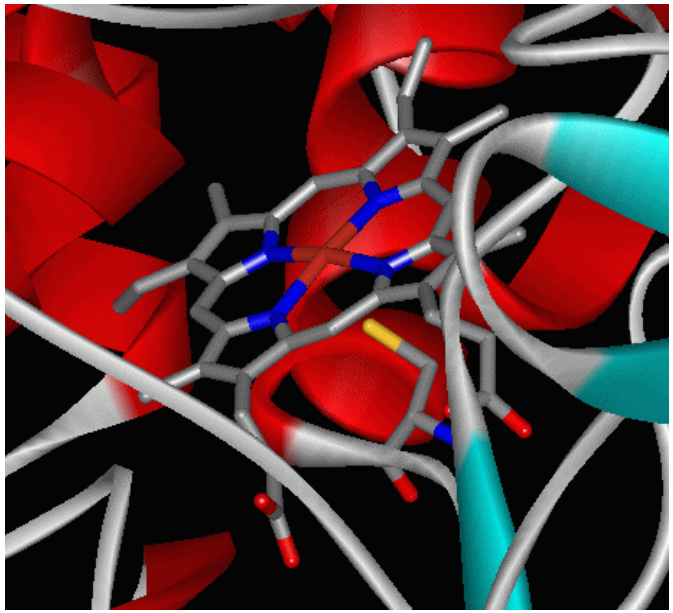
Alcohol dehydrogenase

Aldehyde oxidase

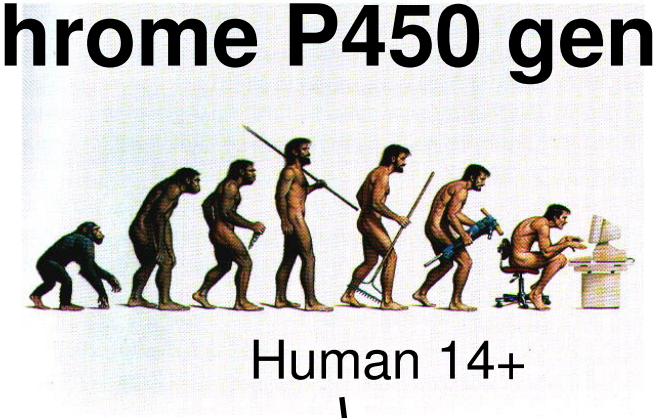
Monoamine dehydrogenase (MAO)

} Redoxactivity is enabled by a iron-containing porphyrin in the active site



# cytochrome P450 gene families

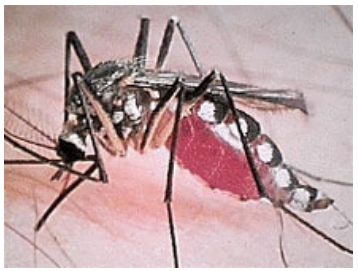


Human 14+

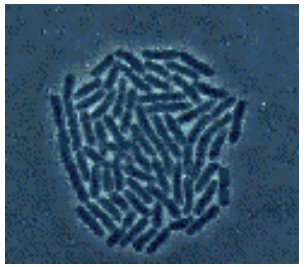
Molluscs 1

CYP450

Plants 22

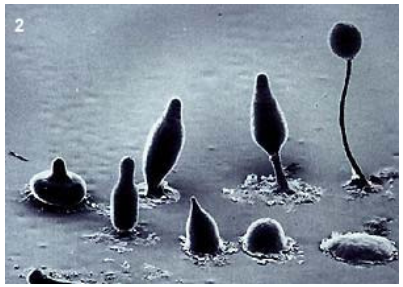


Insects 3

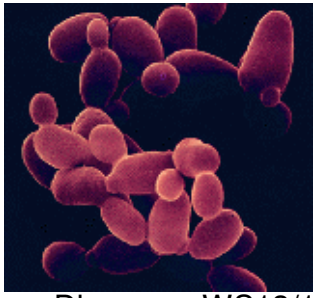


Bacteria 18

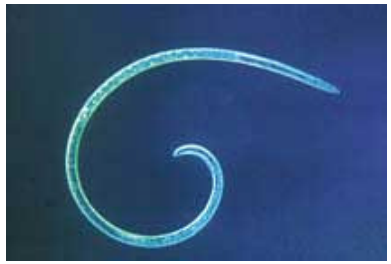
Fungi 11



Yeasts 2



Nematodes 3



# cytochrome P450 polymorphism

„Every human is (more or less) different “

Determination of the phenotype by the actual activity or the amount of the expressed enzyme.

In contrast, the genotype is determined by the individual DNA sequence.

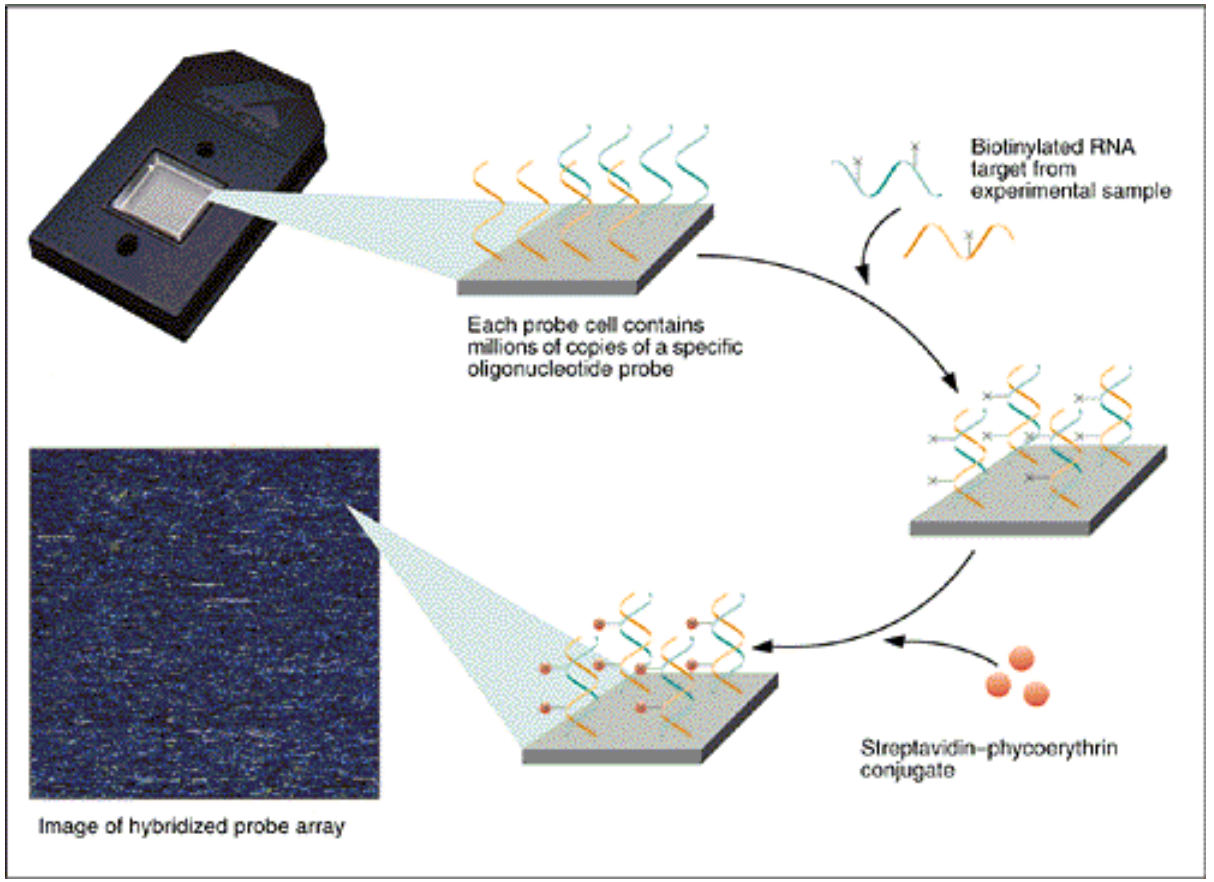
Thus, the same genotype enables several different phenotypes

According to their metabolic activity of CYP there is a classification into normal (*extensive metabolizer*), weak (*poor metabolizer*), und accelerated (*ultra-rapid metabolizer*) metabolism.

Lit: K. Nagata et al. *Drug Metabol. Pharmacokin* **3** (2002) 167

# genotyping of CYP P450 alleles

By using immobilized, synthetic copies of P450 nucleotides, the Affymetrix company (USA) has developped mircoarrays (gene chips) that allow the identification of all clinically relevant alleles.



# Prediction of molecular properties (I)

The keynote of *rational drug design*

The general question is:

What is the connection between the biological space (activity) and the chemical space (structure) ?

How are we able to make structure-based prediction ?

→ QSAR and QSRP, regression analysis

→ decision trees, machine learning algorithms

→ other statistical methods

# Prediction of molecular properties (II)

What are molecular properties?

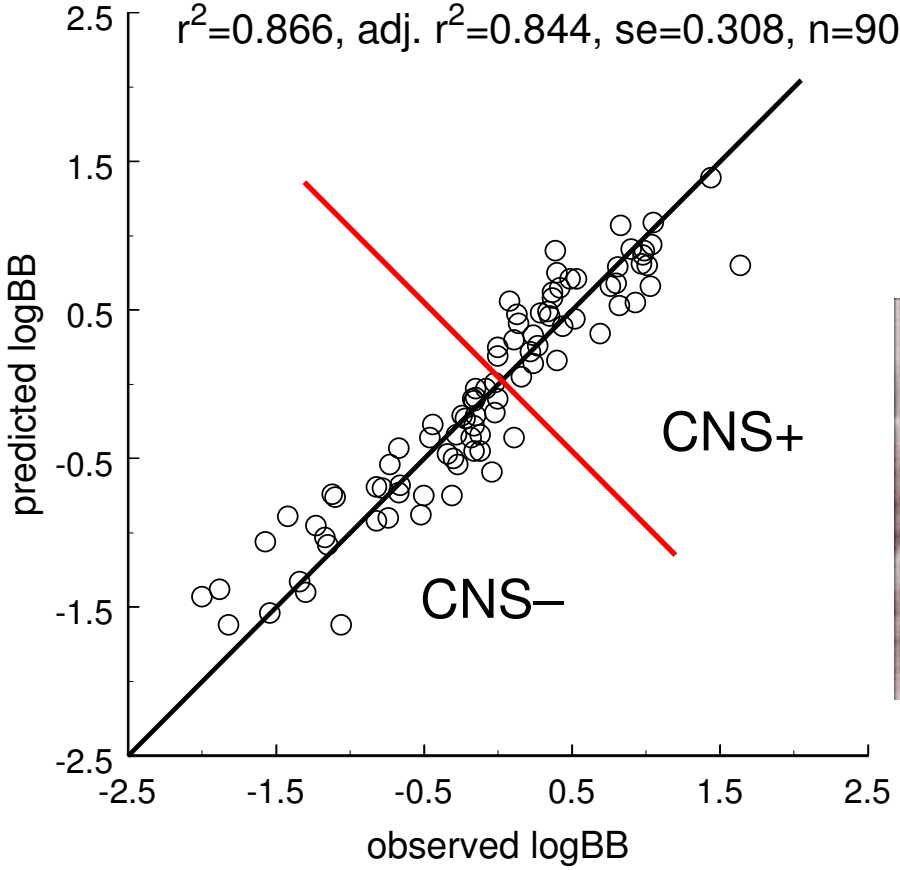
observables

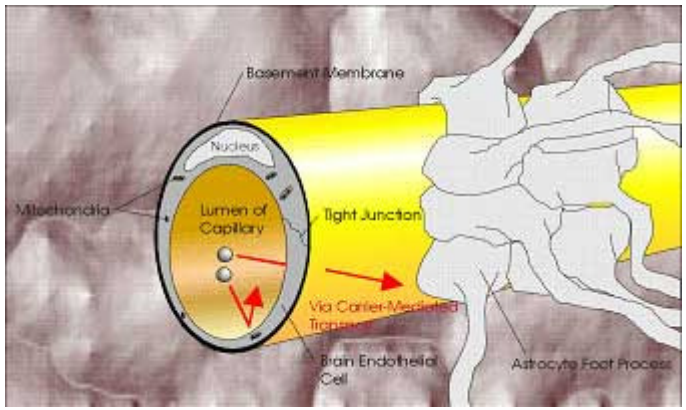
- molecular weight MW (from the sum formula $C_{12}H_{11}N_3O_2$)
- melting point
- boiling point
- vapour pressure
- solubility (in water)
- charge
- dipole moment
- polarizability
- ionization potential
- electrostatic potential

Directly computable from the electronic wave function of a molecule

# BBB-model with 12 descriptors

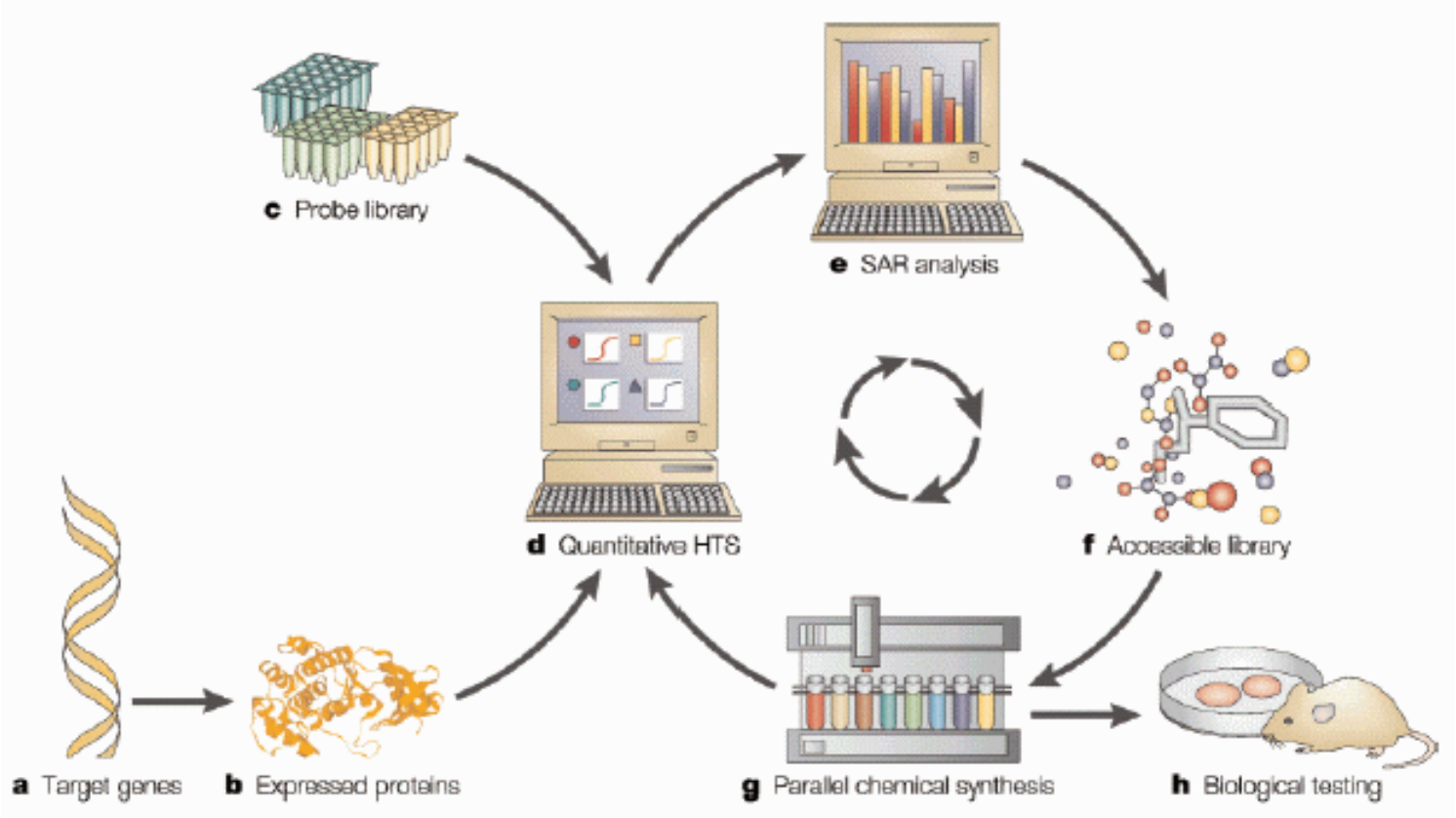
Descriptors mainly from QM calculations: electrostatic surface, principal components of molecular geometry, H-bond properties




Lit: M. Hutter *J.Comput.-Aided.Mol.Des.* **17** (2003) 415.

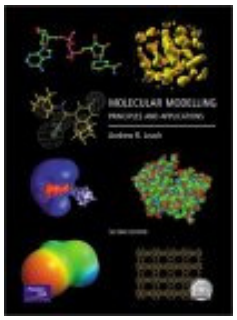
# Cycle of optimization in the drug discovery pipeline



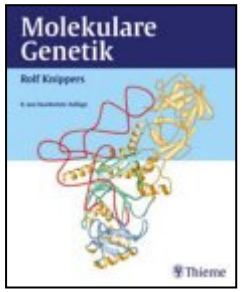

Source: D.K. Agrafiotis et al. *Nature.Rev.Drug.Discov.* **1** (2002) 337.

# Accompanying books and further reading (I)

Andrew R. Leach*
Molecular Modelling. Principles and Applications
2nd edition, Prentice Hall, 2001

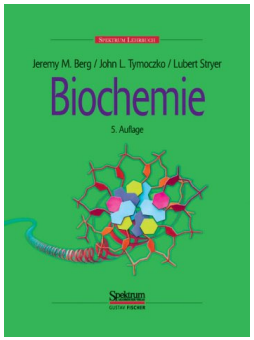



Rolf Knippers*
Molekulare Genetik
8. Auflage, Thieme, 2001




The Merck Index*
13th edition, Merck & CO., Inc., 2001


J.M. Berg, L. Stryer*
Biochemie, Spektrum Verlag
Biochemistry, W.H. Freeman & Co Ltd.

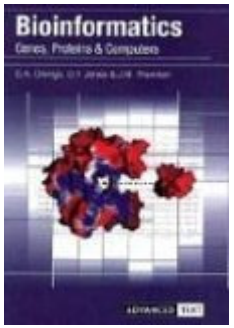



*Available in the „Semesterapparat“

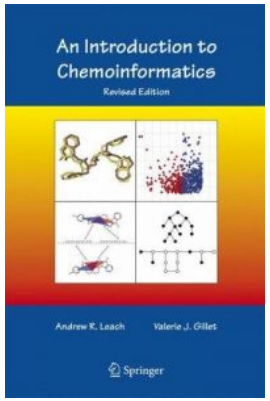
# Accompanying books and further reading (II)

Gerhard Klebe*
Wirkstoffdesign
2. Auflage, Spektrum Akad. Verlag, 2009


C.A. Orengo, D.T. Jones, J.M. Thornton*
Bioinformatics
Genes, Proteins & Computers
1st ed., Bios Scientific Publishers, 2003




A.R. Leach, V. Gillet*
An Introduction to Chemoinformatics
revised ed., Springer, 2007




*Available in the „Semesterapparat“

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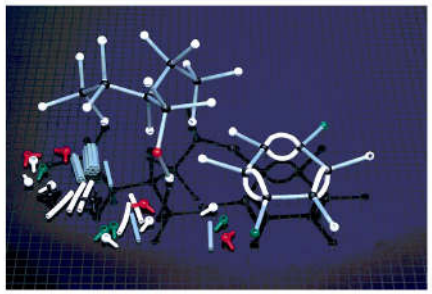
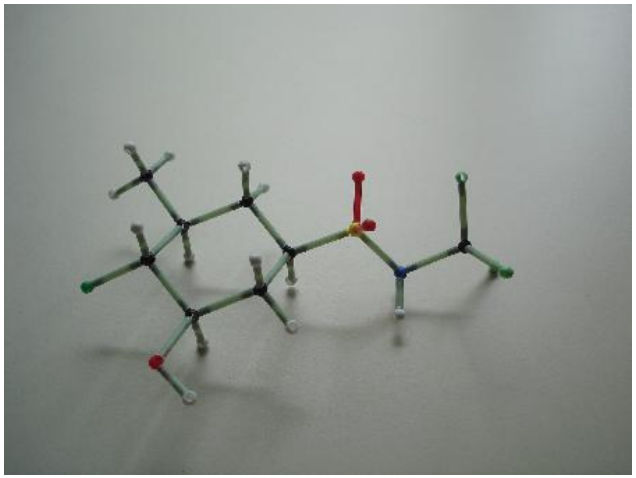
# Further hands-on tools

Molecular model sets / Molekülbaukasten





Commerically available at various price ranges

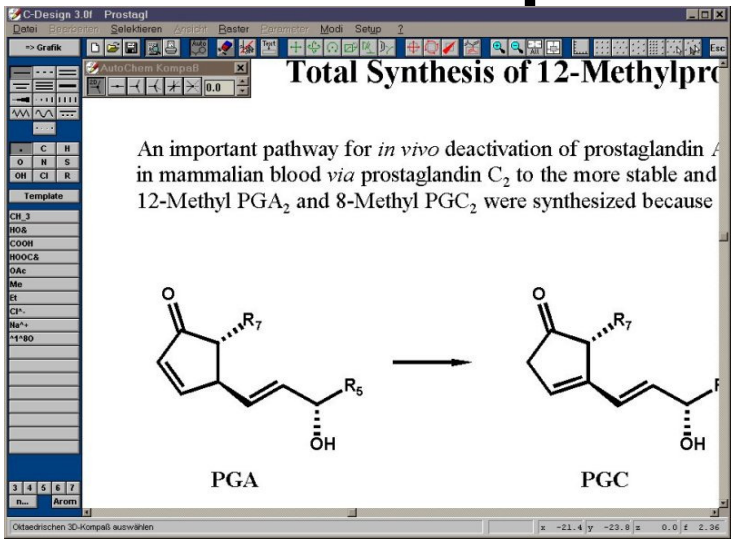
General remark: The lecturer does not endorse any of the mentioned books/software/products. Enquiries are welcome.

# Other useful software to make nice pictures

Chemical structures
and other objects:

C-Design 3.0f

Windows-Platform

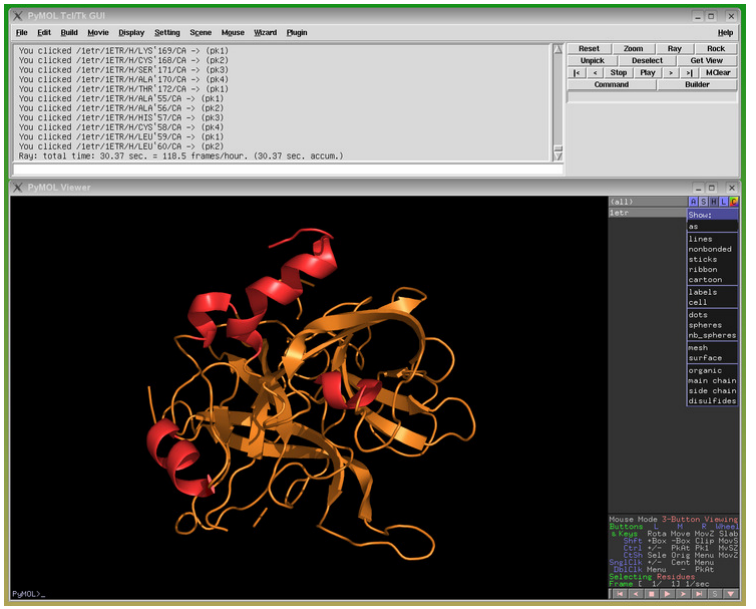



Protein structures:

PyMOL

www.pymol.org

Linux, Mac OS X, Windows



# Requirements to obtain the certificate and the credit points

1. 50% of all accomplishable points from the home work. Two thirds (66.7%) of all assignments (ca. 6) must be returned. The assignments have to be handed in until the beginning of the next exercise unit.
2. 50% of all accomplishable points from the final exam taking place at the end of the lecture period. If necessary, repeated (written) exam or oral exam.

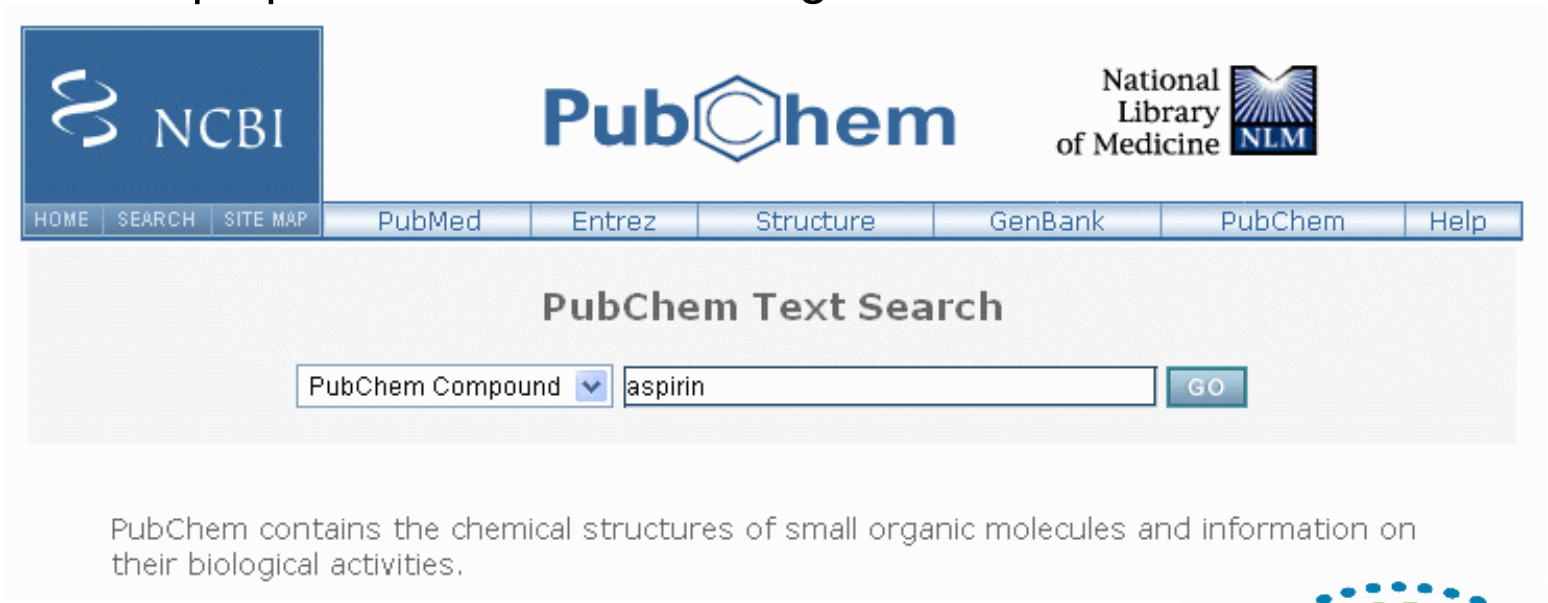
# 1st assignment (I)

Refer to a prescription medicine of your own choice

Write down the active ingridient

Try to find out its molecular structure:

http://pubchem.ncbi.nlm.nih.gov/

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# 1st assignment (II)

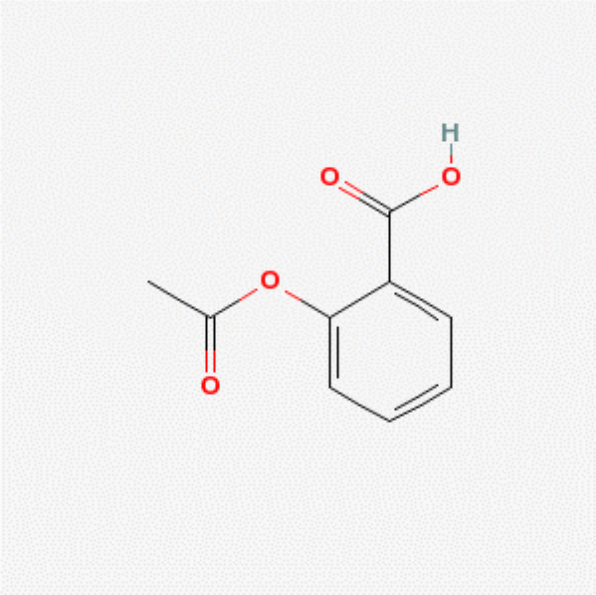
NCBI

PubChem

National Library of Medicine NLM

HOME | SEARCH | SITE MAP | PubMed | Entrez | Structure | GenBank | PubChem | Help

**Compound Summary:**




**CID:** 2244

**Substances:**

All: 52 Links
Same: 10 Links
Mixture: 42 Links

**BioActivity:** 66 Links

**Protein Structures:** 2 Links

**NLM Toxicology:** Link

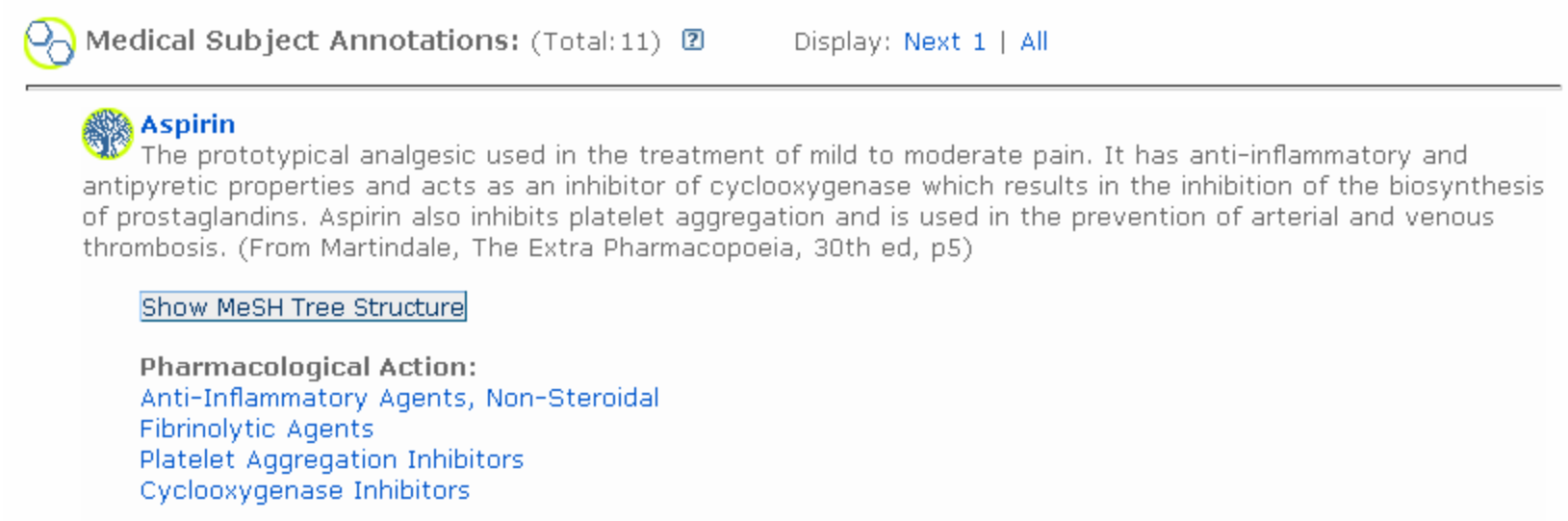
**Related Compounds:**

Same, Connectivity: 2 Links

**Similar Compounds:** 30 Links

**Structure Search**

# 1st assignment (III)




Explain why the medicine has a completely different name compared to the actual substance.

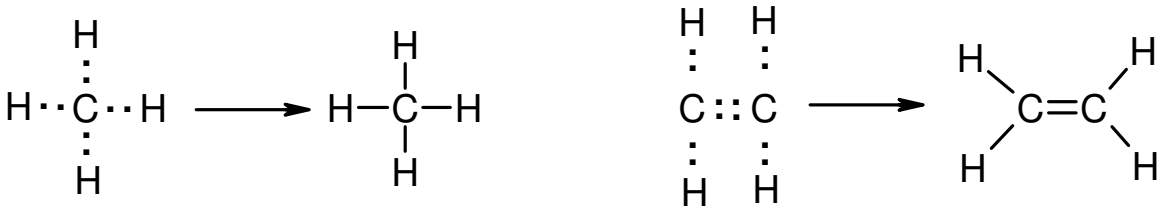
Try to find out some information about its molecular target:

e.g. using PubMed http://www.ncbi.nlm.nih.gov

or consult the Merck Index.

# Representation of chemical structures (I)

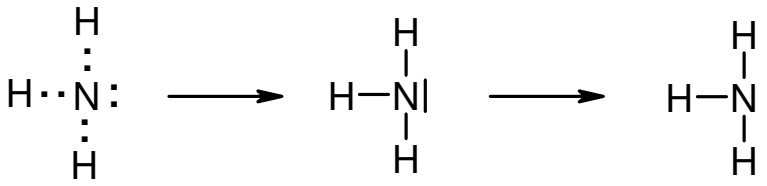
The valence electrons of the atoms are pairwise grouped together



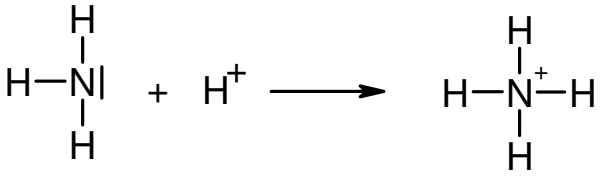
Lewis structures reflect covalent bonds between atoms in a molecule

# Representation of chemical structures (II)

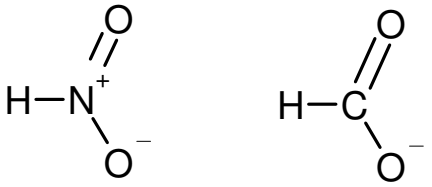
(electron) lone pairs are often not shown for clarity

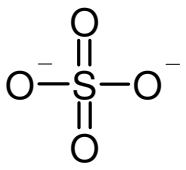


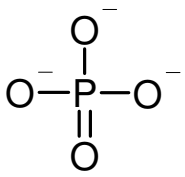
octet rule and hypervalent atoms



Equal bond lengths !

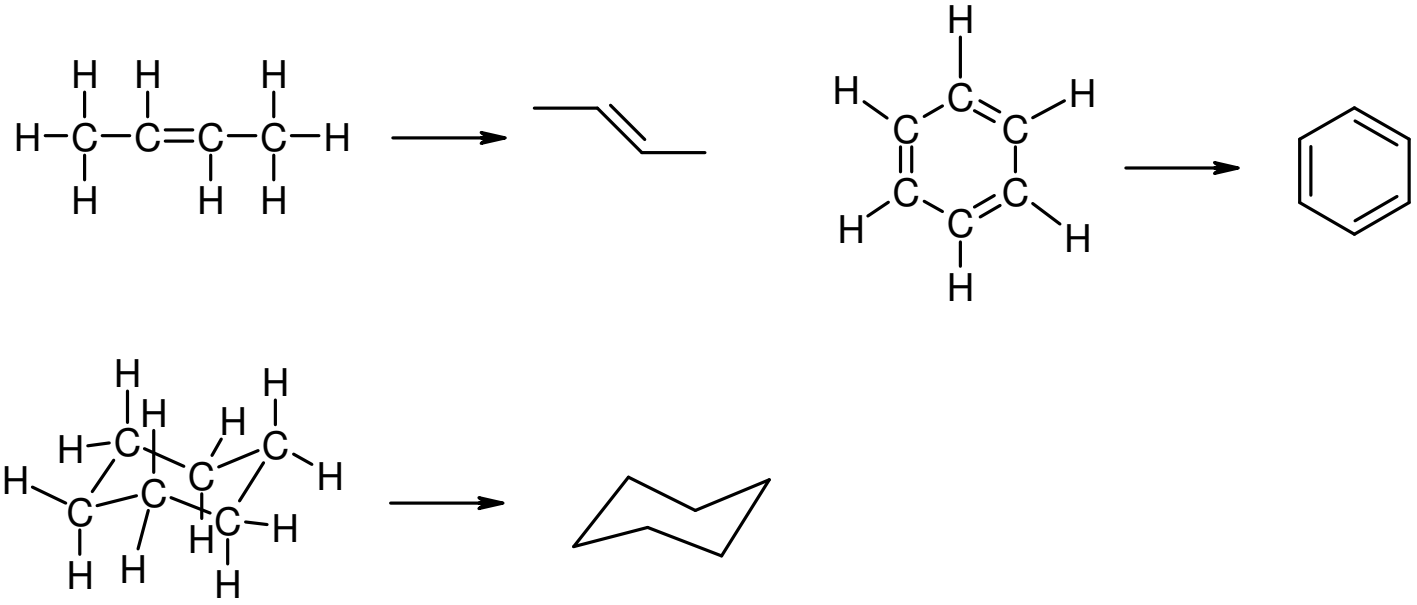






# Representation of chemical structures (III)

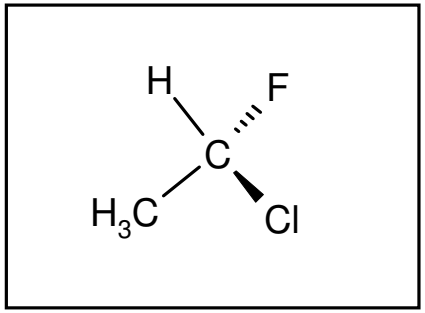
Also carbon atoms are often omitted



Corners and end of lines denote carbon atoms saturated with the appropriate number of hydrogen atoms

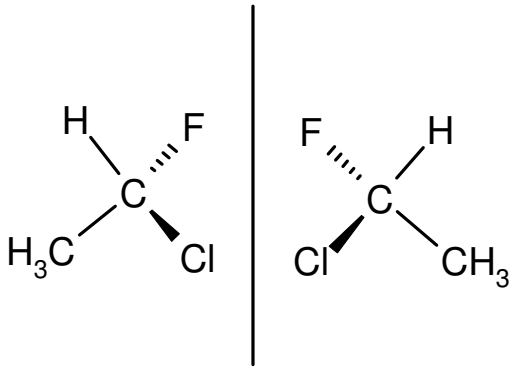
# Representation of chemical structures (IV)

Stereochemistry



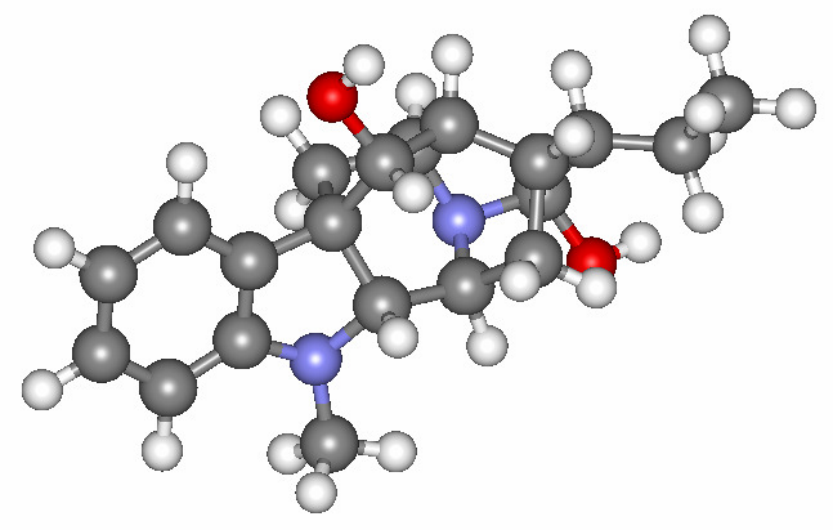

Solid wedges denote atoms in front of the plane,dashed wedges denote atoms behind

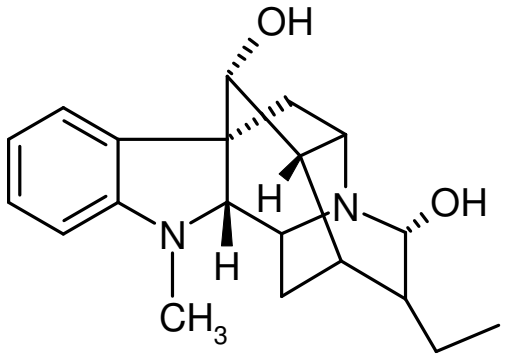
Four different substituents at a carbon atom cause chirality



# Representation of chemical structures (V)

Particular for more complex molecules, these structural drawings provide more clarity than a picture of an actual 3D representation does.

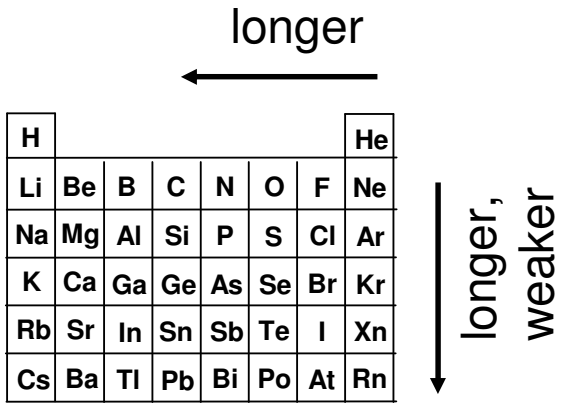





Exercise: Construct this molecule using a molecular model set. Specify the chiral carbon atoms.

# Bond distances and bond dissociation energies (I)

| bond | distance [Å] | $D_o$ [kJ/mol] (homolytic cleavage) | |
|---|---|---|---|
| H–H | 0.742 | 432 | |
| C–H | 1.09 ± 0.01 | 411 ± 7 | |
| C–C | 1.54 | 345 | |
| C=C | 1.34 - 1.40* | 602 ± 21 | *aromatic bond |
| C≡C | 1.20 | 835 | |
| C–N | 1.47 | 305 | |
| C=N | 1.35 | 615 | |
| C≡N | 1.16 | 887 | |
| C–O | 1.43 | 358 | |
| C=O | 1.20 | 526 | |
| C–Si | 1.85 | 318 | |
| C–P | 1.84 | 264 | |
| C–S | 1.82 | 272 | |
| C=S | 1.60 | 577 ± 21 | |



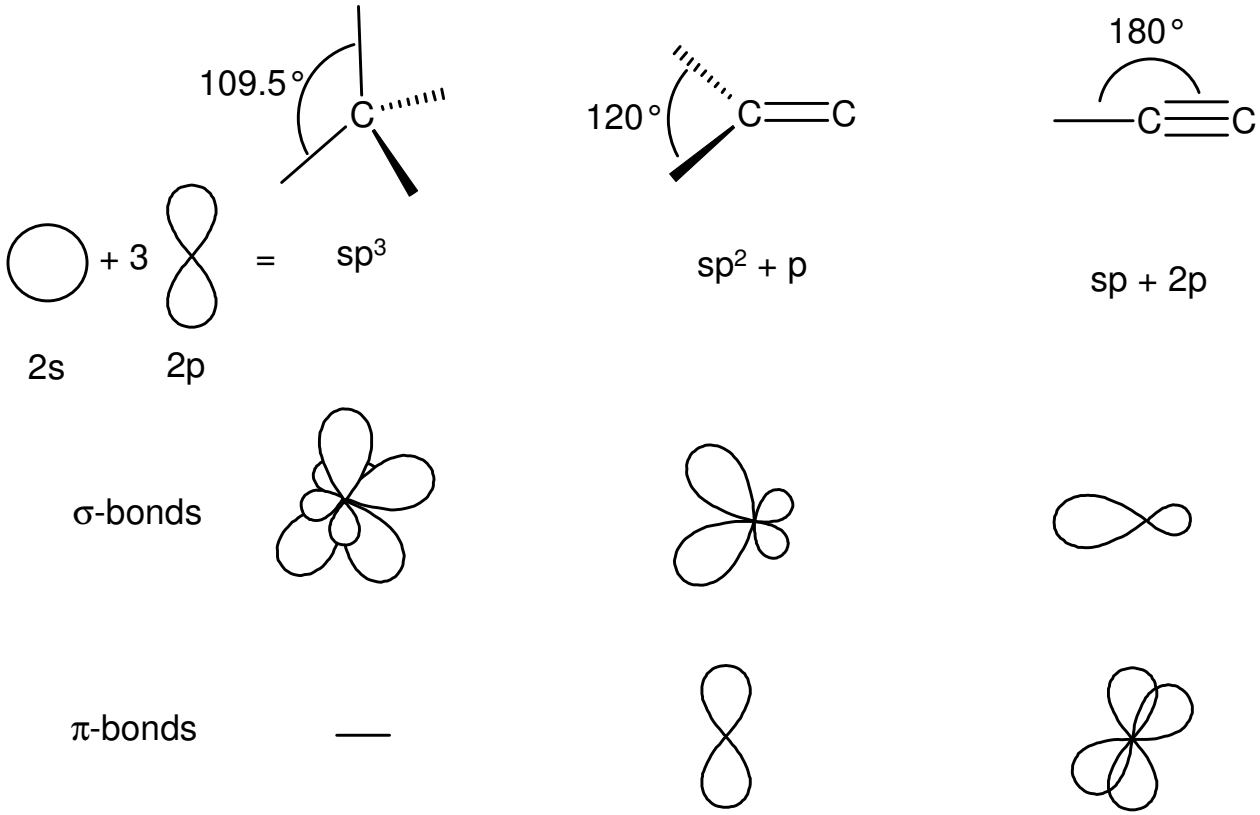

Adapted from: J.E.Huheey *Inorganic Chemistry,* Wiley.

# Bond distances and bond dissociation energies (II)

| bond | distance [Å] | $D_o$ [kJ/mol] | |
|---|---|---|---|
| C–F | 1.35 | 485 | |
| C–Cl | 1.77 | 327 | |
| C–Br | 1.94 | 285 | |
| C–I | 2.14 | 213 | |
| C–H | 1.09 | 411 | non-polar hydrogen |
| O–H | 0.96 | 459 | polar hydrogens, exchangable in polar solvents |
| N–H | 1.01 | 386 ± 8 | |
| S–H | 1.34 | 363 ± 5 | |
| N–N | 1.45 | 247 ± 13 | reason: N, O, and S are more electronegative than C; heterolytic cleavage that leads to ions |
| N=N | 1.25 | 418 | |
| N–O | 1.40 | 201 | |
| N=O | 1.21 | 607 | |
| P–O | 1.63 | ≈335 | |
| P=O | ≈1.50 | ≈544 | |

# Bond angles (I)

Strongly dependend on the hybridization



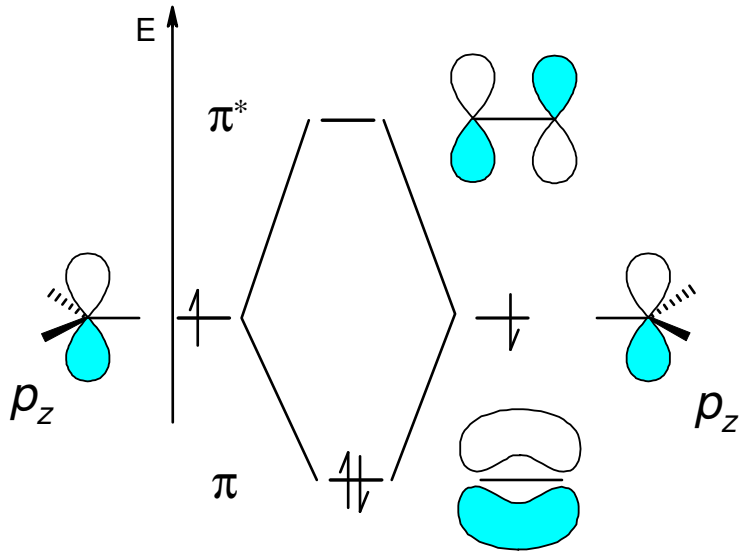

The C–C σ-bond is formed by overlap of the 1s orbitals

These are hybrizided atomic orbitals. Do not confuse with molecular orbitals (=linear combination of atomic orbitals)

# Molecular Orbitals

MO = linear combination of atomic orbitals (LCAO)

$\pi$-bond of ethylene $H_2C{=}CH_2$

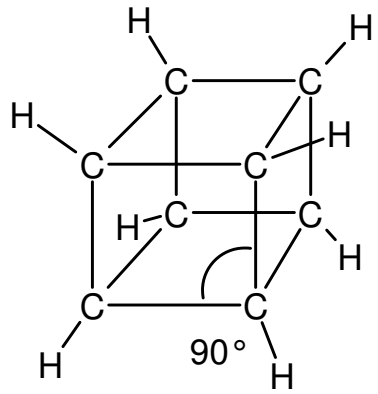



The two combinations usually result in one bonding and one anti-bonding MO

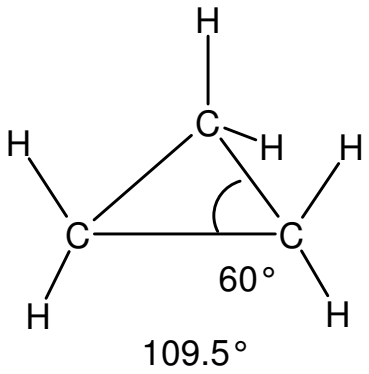
# Bond angles (II)

Extreme deviations from ideal bond angles

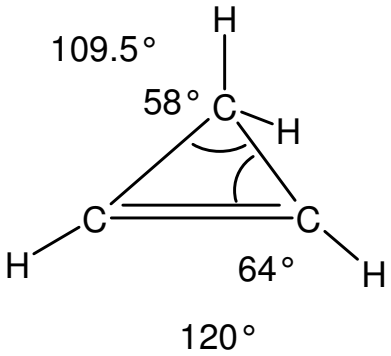
cubane




cyclopropane




cyclopropene

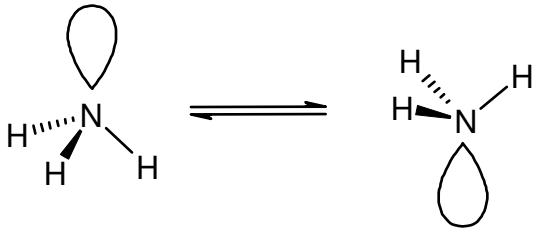



gives rise to strain energy in small rings

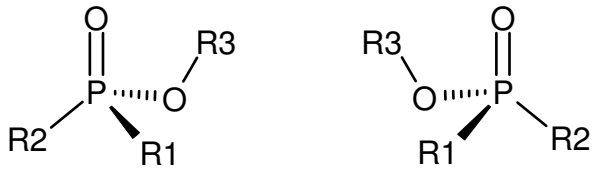
→ problems in force fields. More than one atom type per hybridization needed.

# Chiral atoms

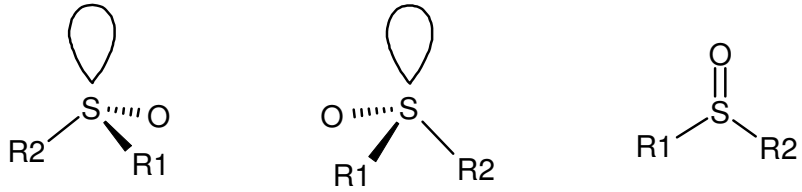
Further elements showing chirality/stereochemistry
the lone pair behaves like a substituent




Fast exchange at room temperature,
but slow at 77K



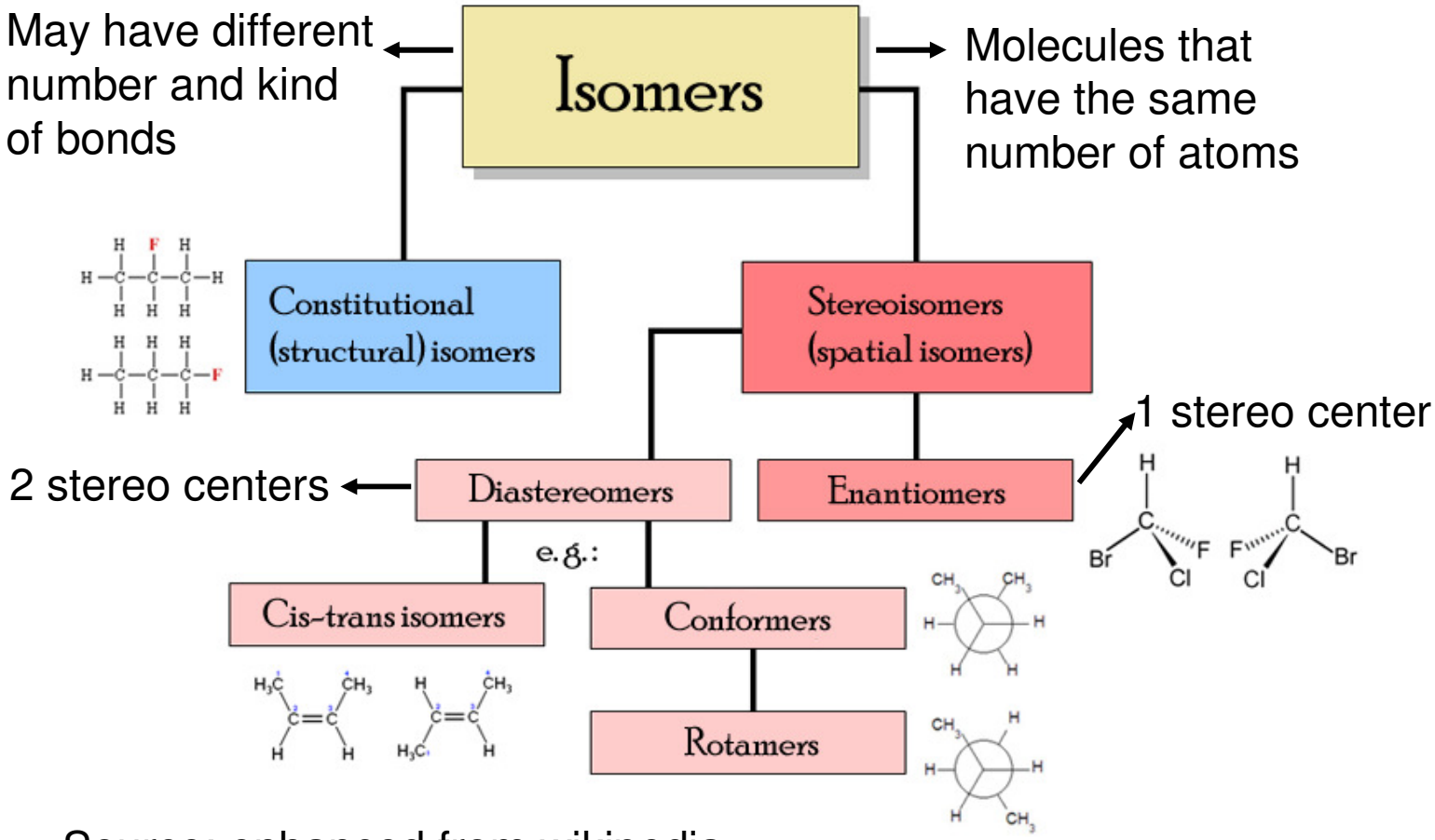

phosphorus inverts even slower




sulfoxides,
sulfinic esters, etc

Furthermore: As, Si, ..., compounds with transition elements,
esp. octahedral metal complexes

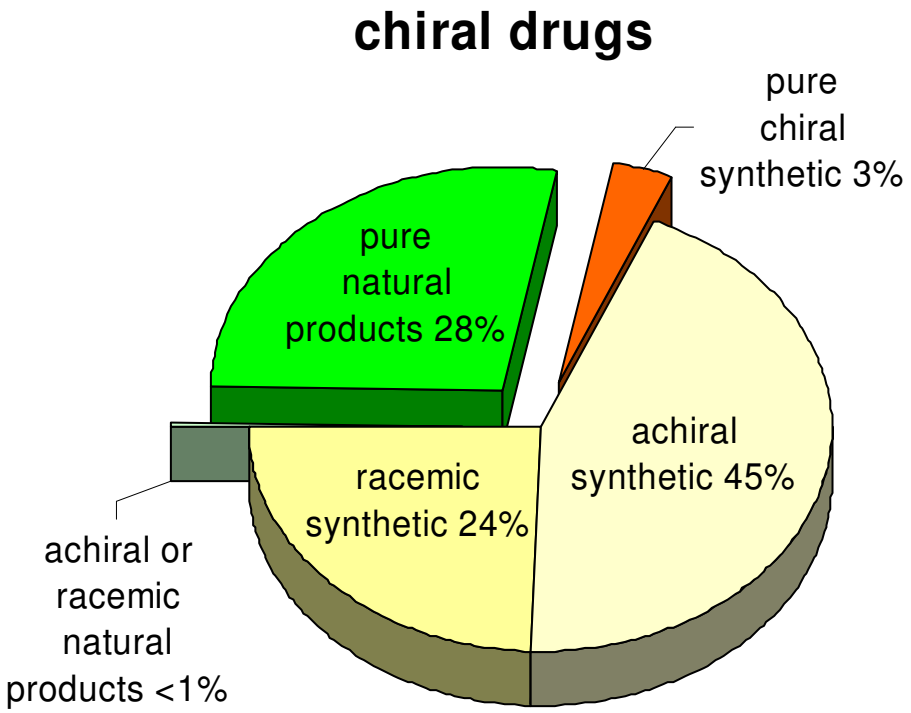
# Isomers




Source: enhanced from wikipedia

Exercise: Which kind of computational method(s) allow(s) to calculate differences in energy between the respective isomers ?

# Is stereochemistry important ?




Data from 1982: Böhm, Klebe & Kubinyi, *Wirkstoffdesign*