Drug Design 1

Oliver Kohlbacher
Summer 2009
2. Basic Pharmaceutics

Abt. Simulation biologischer Systeme
WSI/ZBIT, Eberhard-Karls-Universität Tübingen
Overview

- Definitions and terms
- Galenics
- Pharmacology
  - Pharmacokinetics
  - Pharmacodynamics
  - Molecular Mechanisms of Action
    - Terms
    - Lock-and-Key Principle
    - Examples
Pharmaceutics is often split into four areas:

- **Pharmaceutical Biology**
  is concerned with the use of naturally derived products for therapy.

- **Pharmaceutical Chemistry**
  is concerned with chemical structure, synthesis, properties, and stability of drugs.

- **Pharmacology and Toxicology**
  are concerned with the interactions between exogenous substances and biological systems.

- **Pharmaceutical Technology**
  is concerned with turning a drug into a medicinal product.
Pharmacological Basics

• Effect of a substance on the body
  - Useful/desired - medicine
  - Harmful - poison

• Paracelsus: 'the dose makes the poison'!
  ⇒ each drug has its own characteristic, dose-dependent effect

• Effect described by
  - **Quality** (type of effect)
  - **Strength**
  - **Duration**

• Effects are
  - **Reversible** or **irreversible**
  - **Local** or **systemic** (throughout the organism)
# Pharmacological Basics

## Interaction

Interaction between drug and organism occurs in both directions.

- **What does the organism do to the drug?**
  - Absorption (uptake)
  - Distribution, storage
  - Elimination (excretion and metabolism)

- **What does the drug do to the organism?**
  - Dose/concentration
  - Receptor response (susceptibility)
  - Non-receptor-mediated actions
Overview of the Different Areas

Pharmaceutical Phase
- Application
- Dissolution

Pharmacokinetic Phase
- Absorption
- Distribution
- Place of Action (Receptors)
- Storage
- Biotransformation
- Excretion

Pharmacodynamic Phase
- Pharmacological Effect
- Clinical Effect
- Toxic Effect

After: Mut, p. 5
Pharmaceutical Technology

- **Definition:** the science of preparing and dispensing drugs

- **Goals:**
  
  Achieve the desired target concentration at the place of action

- This is achieved by the right *dosage form*

- There is a plethora of different dosage forms for the various *routes of administration*
## Dosage Forms

<table>
<thead>
<tr>
<th>Oral</th>
<th>Topical</th>
</tr>
</thead>
<tbody>
<tr>
<td>• Tablets</td>
<td>• Cream</td>
</tr>
<tr>
<td>• Capsules</td>
<td>• Ointment</td>
</tr>
<tr>
<td>• Solutions</td>
<td>• Gel</td>
</tr>
<tr>
<td>• Powder</td>
<td>• Nose drops</td>
</tr>
<tr>
<td>• ...</td>
<td>• Eye drops</td>
</tr>
<tr>
<td>Parenteral</td>
<td>• Spray</td>
</tr>
<tr>
<td>• Infusion</td>
<td>• Transdermal patch</td>
</tr>
<tr>
<td>• Injection</td>
<td>• ...</td>
</tr>
<tr>
<td>• ...</td>
<td></td>
</tr>
</tbody>
</table>
Dosage Forms

- Distinguished by the type of release
  - Immediate release
  - Controlled release (sustained release)

- Determined by
  - Disintegration of the dosage form
    - Capsule vs. tablet
    - Coatings (film coating, sugar coating)
  - Dissolution of the drug
    - Particle size
    - Solubility
  - Diffusion of the drug
Disintegration of an aspirin tablet in water
Routes of Administration

• There is a plethora of different routes of administration (RoA): FDA recognizes 111 different RoAs!

• Roughly three main groups of RoAs:
  - **Topical**: applied locally, where it should act
  - **Parenteral**: systemic effect desired, not given through gastrointestinal tract (e.g., injection)
  - **Enteral**: systemic effect desired, given through the gastrointestinal tract (e.g., tablets)

• Key differences:
  biological barriers the drug needs to overcome
Routes of Administration

- Intranasal
- Pulmonary
- Parenteral
  - Intramuscular (i.m.)
  - Subcutaneous (s.c.)
  - Intravenous (i.v.)
- Enteral
  - Sublingual (s.l.)
  - Oral (p.o.)
  - Rectal (p.r.)
- Transdermal
Pharmacokinetics

Pharmacodynamics

- Intensity of Effect
- Duration of Effect
- Dose

Pharmacokinetics

- Absorption
- Distribution
- Metabolism
- Excretion
Pharmacokinetics - Terms

- Concerned with understanding (and modeling) absorption, distribution, metabolism and excretion of a drug
  (ADME - Absorption, Distribution, Metabolism, Elimination)

- Basics of pharmacokinetic models
  - Body modeled as consisting of individual compartments (e.g., blood, intestine, muscles)
  - Central compartment: blood (plasma)
  - Each compartment is characterized by
    - Drug concentration \( c \)
    - Distribution volume \( v \)
  - Drug can cross biological barriers from one compartment to another
  - Simulation of these processes with methods from chemical kinetics
Parenteral Administration

- Systemic effect, but not through gastrointestinal tract, typically by direct injection
- Different types of injections
  - **Intravenous** (i.v.) (A)
    - Rapid systemic effect
    - Rapid dilution of the drug
  - **Intraarterial** (i.a.) (B)
    - Rapid systemic effect
    - Little dilution
  - **Intramuscular/subcutaneous** (i.m., s.c.)
    - Directly into muscle tissue or under the skin
    - Drug has to diffuse to the next blood vessel
      - Depot effect
      - Slow onset of the effect
Enteral Administration

- Most common RoAs, easiest for patients
- Absorption through gastrointestinal (GI) tract
- Different parts of the GI tract differ in their surface area
- Strong dependence on stomach content
Enteral Absorption

- pH varies within the GI tract (stomach: 1—3, small intestine: 7—8)
- Absorption of acids/bases occurs preferentially in different parts of the GI tract
- Drug has to be stable at low pH or protected from stomach juices (coatings)
- Absorption occurs mainly through the mucous membrane of the small intestine (huge surface)
Excretion

Excretion of drugs through

- **Kidneys** → urine (renal excretion)
  Mostly for hydrophilic drugs
  Prefers < 300 Da

- **Liver** → bile (biliary excretion)
  Mostly for lipophilic drugs
  Prefers > 500 Da

- **Lungs** → exhaled air (pulmonary excretion)
  Diffusion-limited, gaseous substances
Absorption and Elimination

**Absorption** and **Elimination** processes are illustrated in the diagram. The **Central Compartment** is central to these processes, with pathways for **Biliary** and **Renal** elimination. **Enteral** absorption and **Parenteral** administration are also depicted, along with the **Blood-Brain Barrier** and **Membranes of the GI Tract**.
Blood-Brain Barrier

- Brain requires huge amounts of nutrients and oxygen
- Blood vessels in the brain are surrounded by an additional cell layer, the blood-brain barrier (BBB)
- Protects the brain from toxins and infections
- Molecules larger than 400 - 500 Da cannot penetrate this layer
- Major hurdle to the delivery of drugs acting on the central nervous system (CNS)

http://institut.cochin.inserm.fr/research/scientific-departments/infectious-diseases/team-p.o.-couraud/team-biology-of-brain-endothelium/images/Fig-1.gif
Blood-Brain Barrier

- **Paracellular aqueous pathway**: Water-soluble agents
- **Transcellular lipophilic pathway**: Lipid-soluble agents
- **Transport proteins**: Glucose, amino acids, nucleosides, Vinca alkaloids, Cyclosporin A, AZT
- **Receptor-mediated transcytosis**: Insulin, transferrin
- **Adsorptive transcytosis**: Albumin, other plasma proteins
Biological Membranes

Lipid bilayers

- Polar “head regions” on the outside
- Nonpolar fatty acid residues aggregate on the inner side
- Hydrophilic on the outside
- Lipophilic on the inside
Transport Across Membranes

Possible mechanisms

- **Passive**
  - Diffusion
  - Pores

- **Active**
  - Carrier
  - Vesicular
Transport across Membranes
Active Transport

- Achieved by specific transporters
- Transporters are membrane-bound proteins/protein complexes
- Active transport consumes energy (ATP)
- Depending on the direction, transporters can be responsible for uptake/absorption (influx) or elimination (efflux)

Examples
- amino acid uptake in the GI tract
- Efflux transporters responsible for drug resistance
Gastrointestinal Absorption

**Passive Transport**
- Pores / Ion Channels
- Membrane

**Active Transport**
- Influx
  - ATP
- Efflux
  - ATP

- Para-cellular
- Lumen
- Cytosol
Absorption and Elimination

- **Central Compartment**
- **Biliary**
- **Renal**
- **Enteral**

- **Absorption**
- **Elimination**

- **Parenteral**

- **Blood-Brain Barrier**

- **Membranes of the GI Tract**
Pharmacokinetics - Models

- Concentration as function of time
- Compartments connected by fluxes $k_{12}, k_{21}$ (≡ reaction rates)

$\implies$ Kinetic differential equations
Pharmacokinetics - Models

\[ \frac{dc}{dt} = -k_0 \]  \text{0}^{\text{th}} \text{ order reaction}

\[ \frac{dc}{dt} = -k_1c \]  \text{1}^{\text{st}} \text{ order reaction}
One-Compartment Model

- Single homogenous compartment (central compartment)
- Drug distributed equally over the compartment
- Elimination with rate $k$
- Typically 1st order kinetics

\[
\frac{dc}{dt} = -kc
\]
Two-Compartment Model

- Kinetic model
- Distribution between plasma and a tissue (peripheral compartment)
- Distribution defined by rates $k_{12}$, $k_{21}$ and volumes of distribution
- Elimination with rate $k_E$
Bateman Equation

- Simultaneous absorption and elimination of 1st order
- Independent processes, thus we obtain the **Bateman equation**

\[
c = c_0 \frac{k_r}{k_r - k_e} \left( e^{-k_e t} - e^{-k_r t} \right)
\]

with
- Absorption rate/elimination rate \( k_r / k_e \),
- Initial concentration \( c_0 \)
- Time \( t \)
Pharmacodynamics

- Duration of Effect
- Intensity of Effect
- Dose

Pharmacodynamics

- Absorption
- Distribution
- Metabolism
- Excretion
Pharmacodynamics

Receptor theory

- Based on ideas of John Newport Langley (1852-1926), Paul Ehrlich (1854-1915), Alfred Joseph Clark (1885-1951)

- Drugs bind to ‘receptors‘ to cause an effect

- In general, receptors are places where the action occurs; mostly they are proteins, though

- Binding of the drug to the receptor is usually reversible and then follows mass action
Dose-Response Relationships

- „The dose makes the poison“
  - Typically, an increase in dose increases the effect
  - Above a certain dose, additional toxic effects may be observed
  - Strength/duration of effect depends on many factors, e.g., genotype, age, body mass, ...
  - Difficult to quantify strength of the response

⇒ Dose-response relationships measured for collectives
  - $ED_{50}$: median effective dose
  - $LD_{50}$: median lethal dose
Therapeutic Index

- **Therapeutic index** or therapeutic ratio is the ratio between the concentration causing a toxic effect and the concentration causing a therapeutic effect.
- It is a measure of a drug’s safety.
Pharmacodynamics

- **Effect:**
  - Intensity
  - Duration

- **MEC:** *minimal effective concentration*

- **MTC:** *minimal toxic concentration*
Modes of Action

- **Receptor theory**
  
  Effect = interaction of the drug with a receptor

- **Paul Ehrlich** (1913)
  
  „Corpora non agunt nisi fixata“
  
  „a drug will not work unless it is bound“

⇒ Binding required for action
Emil Fischer 1894:

“Um ein Bild zu gebrauchen, will ich sagen, dass Enzym und Glucosid wie Schloss und Schlüssel zu einander passen müssen, um eine chemische Wirkung aufeinander ausüben zu können.”

(“To use an image, I suggest that enzyme and glucoside need to fit to each other like a lock and a key to exert a chemical effect onto each other.”)

Lock-and-Key Principle
Lock-and-Key Principle

Retinol bound to retinol-binding protein (RBP)

PDB: 1CRB
Chirality

- Proteins are chiral (made from chiral amino acids!)
- Interaction of proteins with different enantiomers of the same ligand different!
- Example:
  Propranolol (beta blocker)
  S-enantiomer 100-times more active than R-enantiomer
- Eudismic ratio:
  Ratio of affinities/effect strengths of two enantiomers

Propranolol
Infamous example: Thalidomide
- R-enantiomer is an excellent hypnotic
- S-enantiomer is teratogenic!

A mixture of both enantiomers was marketed under the name Contergan

Also, enantiomers interconvert in vivo

In the 1950s severe birth defects were observed in the children of women taking thalidomide („children of thalidomide“)

Contergan was taken from the market in 1961

Terms

Receptor
Protein causing a response upon binding of a ligand. Usually an enzyme or ion channel.

Ligand
A (usually small) molecule binding to a receptor.

Enzyme
Biocatalyst, transforming a substrate into a product.

Inhibitor
Ligand that inhibits substrate binding directly (competitive) or indirectly (allosteric). Inhibition can be reversible or irreversible.

Ion channel
Integral membrane protein forming a pore across the membrane that allows the flow of ions through across the membrane.

Transporter
Integral membrane protein that allows the transport of various substances across the membrane, usually requiring energy.
Terms

Agonist
Ligand of a receptor causing the desired response (e.g., neurotransmitter binding to a neuroreceptor).

Antagonist
Ligand of a receptor that prevents binding of an agonist directly or indirectly, but does not cause a receptor response.

Partial Agonist
Weak agonist that has a high affinity to the receptor and thus acts as an antagonist, as well (good binding, but weak response).

Inverse Agonist
Ligand that stabilizes the inactive form of the receptor.

Functional Antagonist
Substance preventing receptor response through a different mechanism (same effect as an antagonist, different mechanism).
Mechanism of Action

Receptor possesses one or more binding sites
Mechanisms of Action

Binding of an **agonist** activates the receptor.
Mechanisms of Action

- Binding of an **antagonist** blocks binding site
- Prevents binding of agonists and thus the activation
Binding

Binding described by

- Association and dissociation constants $K_A / K_D$

\[ L + R \rightleftharpoons LR \]

\[ K_D = \frac{c(L)c(R)}{c(LR)} = 1 / K_A \]

- $K_D$ has units of concentration
- Corresponding Gibbs free enthalpy $\Delta G$: \[ \Delta G = RT \ln K_A \]
- A smaller (more negative) $\Delta G$ implies stronger binding of the ligand
Binding

- **Half maximal inhibitory concentration $IC_{50}$**
  
  Inhibitor concentration where 50% of the enzyme is inhibited

  $$c(LR) \approx c(R) \Rightarrow IC_{50} \approx c(L) = K_D \frac{c(LR)}{c(R)}$$

- $IC_{50}$ has units of concentration
- Identical to inhibition constant $K_i$

- Strong inhibitors have low $IC_{50}$
  
  „nanomolar inhibitors“: $IC_{50} \sim O(10^{-9} \text{ mol/l})$

- Weaker inhibitor have accordingly higher $IC_{50}$
  
  „micromolar inhibitors“: $IC_{50} \sim O(10^{-6} \text{ mol/l})$
Enzymes as Receptors

- Enzymes catalyze biochemical reactions
- **Signal transduction**: chain of enzymatic reactions
- Disrupt signal transduction: interrupt this chain
  ⇒ Drugs are usually **inhibitors**
  - Substrate-like
  - Cannot be converted to product
- Drug design often starts from **transition state** of the reaction
- Examples
  - Acetylsalicylic acid: irreversible inhibitor of cyclooxygenase (COX)
  - Penicillin: inhibition of transpeptidase
**Acetylsalicylic Acid**

- **Prostaglandins** play a key role in pain, inflammation and related processes
  - Fever mediators
  - Sensitization of pain receptors (nociceptors)
- Acetylsalicylic acid (ASA) inhibits prostaglandin-G/H-synthases (COX-1/2, cyclooxygenases)
- ASA and related compounds are so-called **non-steroidal anti-inflammatory drugs** (NSAIDs)
- COX-1 and COX-2 are involved in the synthesis of prostaglandins from arachidonic acid:
Acetylsalicylic Acid

- Acetylsalicylic acid is an irreversible inhibitor of COX-1 and -2
- COX-1/-2 both contain a serine residue in their active site (Ser516 and Ser530, respectively)
- Acetylsalicylic acid *acetylates* this serine residue *irreversibly*
- Arachidonic acid can no longer bind at the active site
  ⇒ COX is inactivated
  ⇒ Prevents synthesis of prostaglandins
Acetylsalicylic Acid

COX-2 with bound substrate (arachidonic acid, blue)
Acetylsalicylic Acid

- COX-1 have COX-2 multiple functions unrelated to pain and inflammatory processes
- Inhibiting COX-1 and -2 can cause severe adverse effects
- Known adverse effects of ASA:
  - Heartburn
  - Stomach bleeding
  - Gastric ulcers
Acetylsalicylic Acid

- Most severe adverse effect: irritation of stomach lining
- **Cause**: COX-1 involved in synthesis of prostacyclin, that protects stomach lining
- **Solution**: specific inhibition of COX-2, which is more important in inflammation, without inhibiting COX-1
- Some specific inhibitors of COX-2 have been approved: Rofecoxib, Celecoxib
Celecoxib

- Selectivity is based on an additional pocket in COX-2 (and not present in COX-1) accepting the toluyl group
- Crystal structure of related compound SCC-558 illustrates the binding mode
- SCC-558 binds 1900x better to COX-2 than to COX-1

Kurumbail et al., Nature (1996), 386, 644
PDB:6COX
Merck Sharp & Dohme (MSD) Announces Voluntary Worldwide Withdrawal of VIOXX

Dear Healthcare Professional:

MSD today announced a voluntary worldwide withdrawal of VIOXX (rofecoxib), its arthritis and acute pain medication. The Company's decision, which is effective immediately, is based on new, three year data from a prospective, randomized, placebo-controlled clinical trial, the APPROVe [Adenomatous Polyp Prevention on VIOXX] trial.

The trial, which is being stopped, was designed to evaluate the efficacy of VIOXX 25mg in preventing recurrence of colorectal polyps in patients with a history of colorectal adenomas. In this study, there was an increased relative risk for confirmed cardiovascular (CV) events, such as heart attack and stroke, beginning after 18 months of treatment in the patients taking VIOXX compared to those taking placebo. The results for the first 18 months of the APPROVe study did not show any increased risk of confirmed CV events on VIOXX, and in this respect are similar to the results of two placebo-controlled studies described in the current labeling for VIOXX.
Penicillin

- Penicillin belong to the group of **β-lactam antibiotics**
- Target: synthesis of the bacterial cell envelope
  - Cell envelope consists of murein (peptidoglycan)
  - Inhibition of murein synthesis
    ⇒ no proliferation of bacteria

Penicillin structure contains a **four-membered β-lactam ring**
Penicillin

\[ \text{M} : \text{N-Acetylmuraminsäure,} \]
\[ \text{G} : \text{N-Acetylglucosamin; } \text{Aminosäuren der Glycinbrücke und des Tetrapeptids} \]
\[ \text{UDP} = \text{Uridindiphosphat, } P = \text{Phosphat} \]
Penicillin

Terminales d-Alanyl-d-alanin-Dipeptid

Transpeptidase

d-Ala

Transpeptidase

R' - N - C - C - O - Ser

Transpeptidase

R =

C = O

H - C - C - S - C - H - C - O - COO⁻

H - C - H - C - O - C - N - C - H - C - O

Transpeptidase

Ser

R' - N - C - C - N - C - H - C - O

R' - N - C - C - N - C - H - C - O
G-Protein-Coupled Receptors

- **GPCR (G-protein-coupled receptor)**
- **Name:** act on G proteins, i.e. proteins binding GDP/GTP
- **Class of membrane proteins of identical architecture**
  - Seven transmembrane helices
  - Agonist activates G protein complex on the opposite side of the membrane
- **Examples:**
  numerous receptors for neurotransmitters, histamine, opiates, LSD, THC

GPCRDB: [www.gpcr.org](http://www.gpcr.org)

Mut, S. 63 ff
BKK S. 80
G-Protein-Coupled Receptors
G-Protein-Coupled Receptors

- GPCRs are among the most promising drug targets
- Example: Serotonin receptors
- Serotonin = 5-hydroxytryptamine, 5-HT
- Neurotransmitter of the CNS
- Many different GPCRs bind 5-HT
- There are several GPCR drugs against
  - Antihypertension
  - Migraine
  - Depression
  - Nausea caused by chemotherapy and radiation therapy (Serotonin antagonists, e.g., Tropisetron, Granisetron)
Ion Channels

- Embedded in membrane
- Ions may pass through the channel
- Channels controlled by
  - Ligands
  - Receptors
  - Voltage (electrostatic potential)
- Important examples
  - Antihypertensives (block Ca$^{2+}$ channels)
  - Local anesthetics
  - Tranquilizer (Benzodiazepine)
  - Tetrodotoxin (poison of the puffer fish)

Ion Channels

- **Example:** nicotine receptor
- **Acetylcholine receptors**
  - Stimulate the CNS
  - Stimulate sympathetic ganglia
    - Increased blood pressure
    - Increased heart rate
    - Reduced esophageal tonus
      ⇒ increases likelihood of stomach ulcers
- **In large doses:** respiratory paralysis
Antisense Drugs

- Oligonucleotides complementary to viral mRNA
- Doublestrand formation inhibits translation, reproduction
- Problem: delivery/stability
Antisense Drugs

• **Vitravene** (INN: Fomivirsen, ISIS Pharma) is the first (and up to now only) antisense drug on the market
  
• Used to treat infection of cytomegalovirus retinitis (a common complication with AIDS)
  
• Intravitreal injection (directly into the eye)
  
• Mechanism: binds to viral mRNA
  
• Approved in the USA in 2001
  
• No longer on the market in Germany due to low demand

Grillone, Drugs Today (2001), 37, 245
Targets Currently in Use

- Enzymes 47%
- GPCRs 30%
- DNA 1%
- Integrins 1%
- Miscellaneous 2%
- Ion channels 7%
- Transporters 4%
- Other receptors 4%

Novel Targets

- *Novel targets*: targets for which drugs have been approved for the first time
- Number of novel targets rather small, about two per year
- Vast majority of new drugs binds to well-known targets
- Genomics did not yet keep its promise to provide a sudden wealth of new targets
  - Knowing the target sequences alone is not sufficient for target selection
  - Drug discovery process takes too long, so new drugs based on genomics couldn’t really be on the market yet
Summary

- Pharmacology studies the effect of drugs
- Galenics is concerned with the administration of drugs
- Both are essential for a drug’s success
- Drug action is complex and can be understood on the molecular level, but also in the context of the whole organism
- Popular target classes are
  - Enzymes
  - Receptors
  - Ion channels
  - Transporters
References