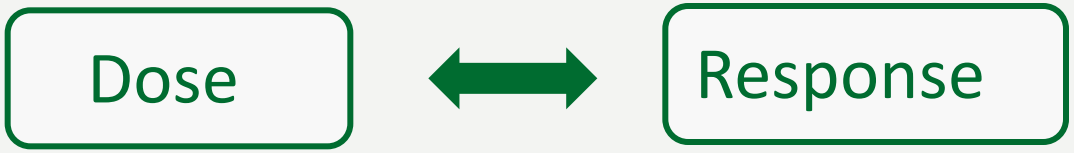
LMU LUDWIG-MAXIMILIANS-UNIVERSITÄT MÜNCHEN

Fundamental Relationships in Toxicology

M1 – Methods in Toxicology



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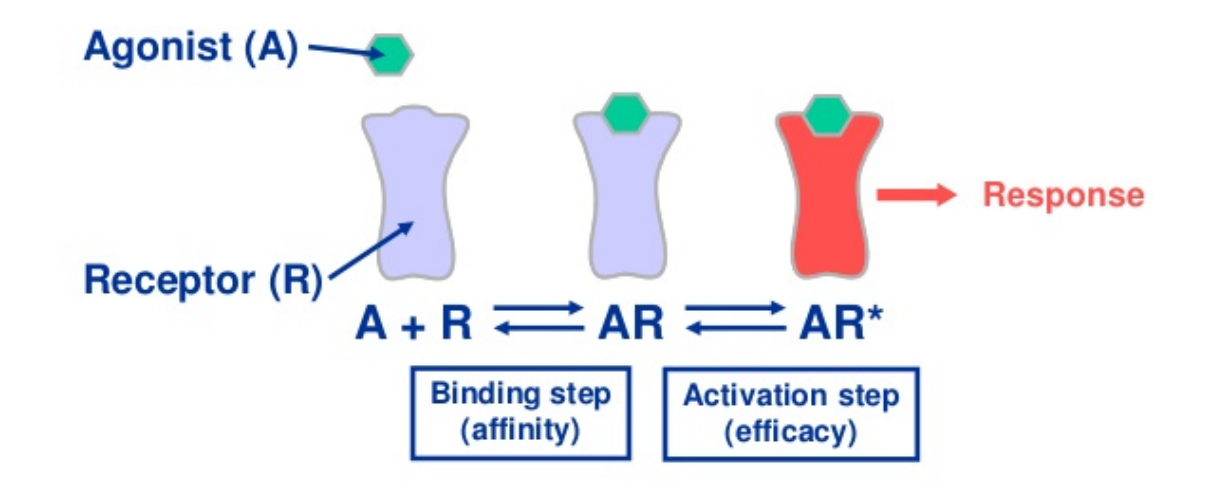
Affinity vs. Efficacy

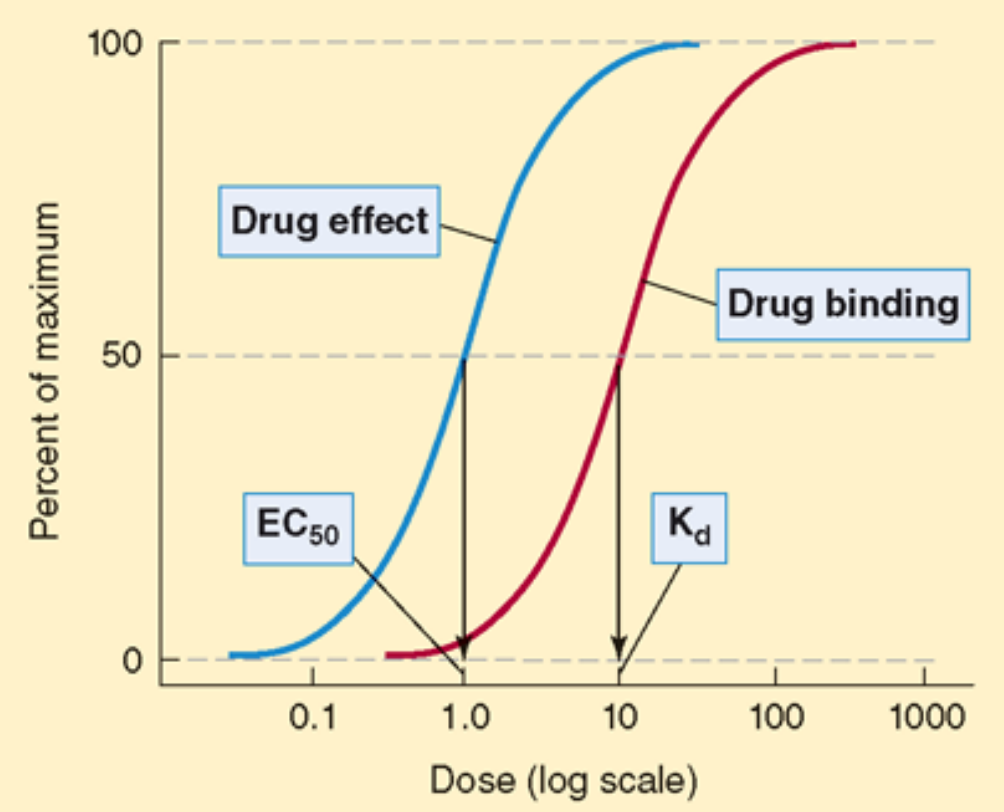
- Affinity: ability of the toxicant to bind to its receptor
- Efficacy: ability of a toxicant to evoke a cellular response



https://www.slideshare.net/PharmacologyEducationProject/introductory-receptor-pharmacology201415jap

- Agonist: high affinity and intrinsic activity/efficacy
- Antagonist: certain affinity but no intrinsic activity/efficacy

Dose-response-curve



https://accesspharmacy.mhmedical.com/content.aspx?sectionid=95700976&bookid=1568

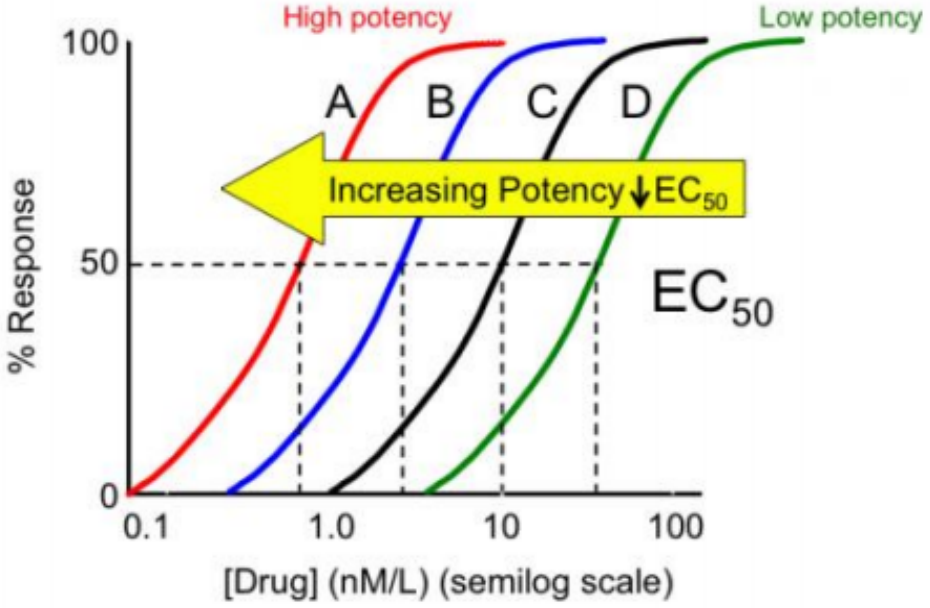
— occupancy/affinity:

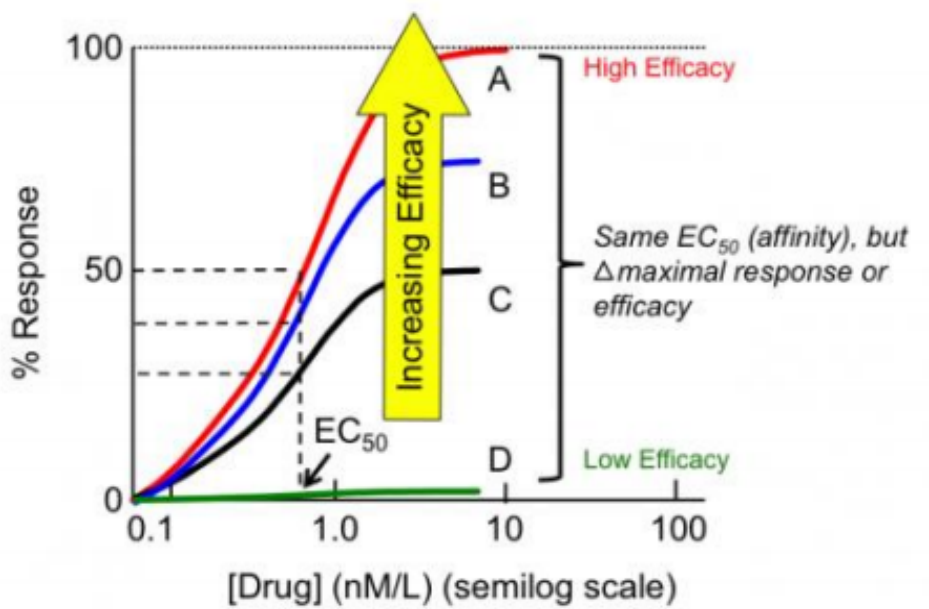
K_d: concentration, at which 50% of receptors are occupied

— response/potency:

EC_{50}: concentration, that causes 50% of the maximal effect

Potency and Efficacy



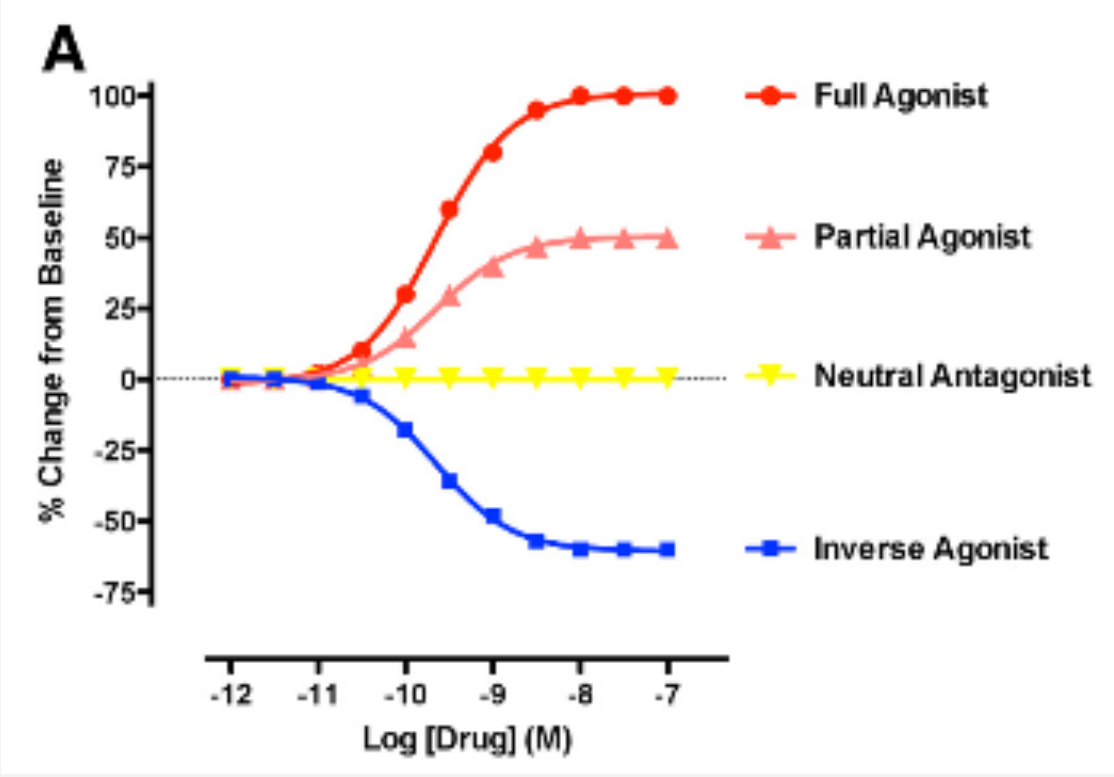


http://tmedweb.tulane.edu/pharmwiki/doku.php/basic_principles_of_pharm

- Potency: refers to the amount of drug needed to produce a given effect → EC_{50}
- Efficacy: determines the maximum effect that a drug can produce regardless of dose → E_{max}

Types of Agonists

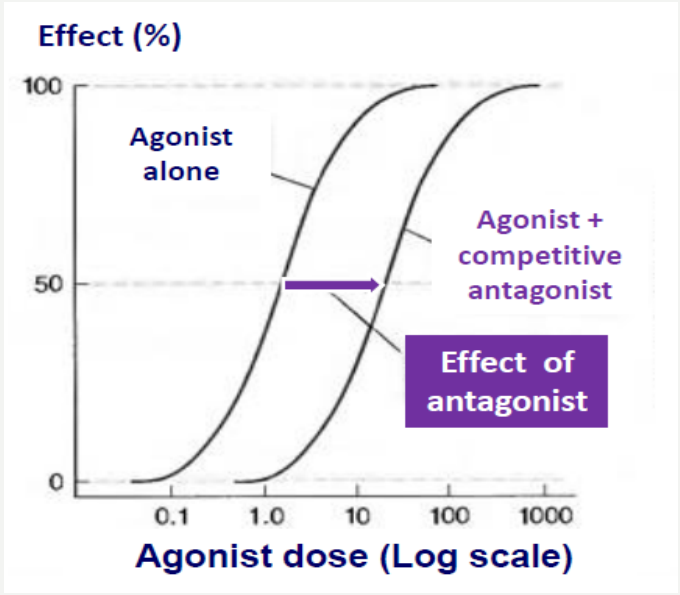
- Full agonist: activate the receptor with a maximum response (e.g. morphine)
- Partial agonist: have only partial efficacy at the receptor relative to a full agonist (e.g. buprenorphine)



Agnostic about in Vivo Inverse Agonism of Agouti-Related Peptide, Malcolm J.

- Neutral Antagonist: intrinsic/basal level of activity in absence of any ligand
- Inverse agonist: decreases the activity below basal level (e.g. rimonabant)

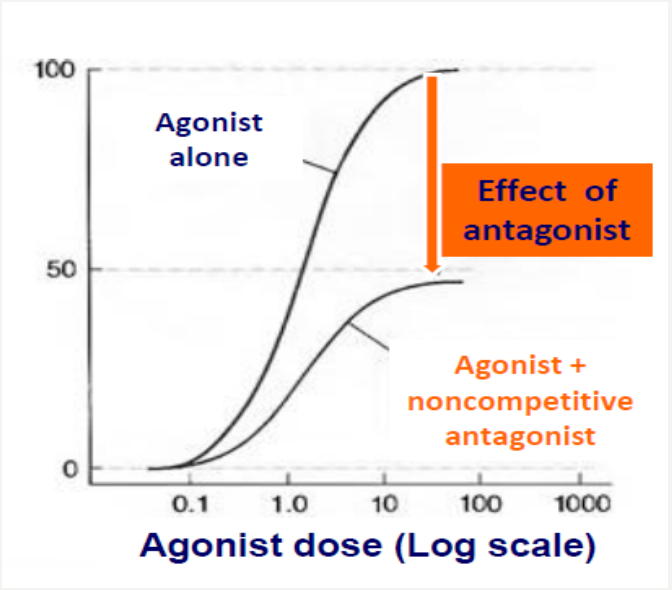
Types of Antagonists



http://tmedweb.tulane.edu/pharmwiki/doku.php/basic_principles_of_pharm

- **Competitive antagonist**:
  bind to receptor at the same binding site as the agonist but without activating it

EC_{50} ↑
E_{max} = const.

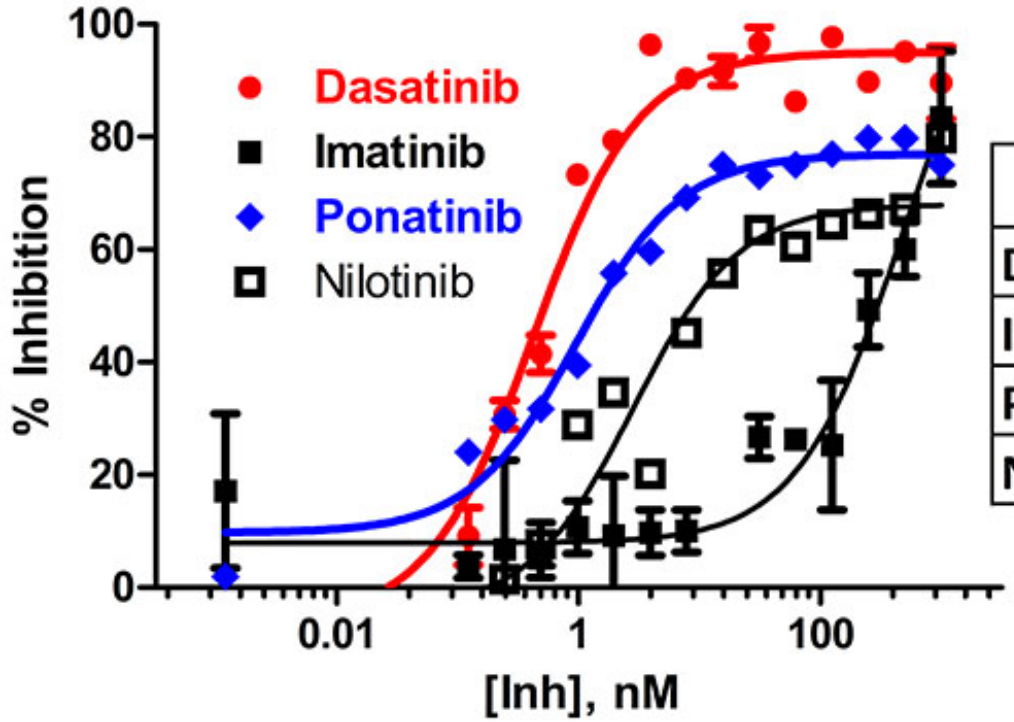


http://tmedweb.tulane.edu/pharmwiki/doku.php/basic_principles_of_pharm

- **Non-competitive antagonists**:
  1. bind irreversibly to the same site as the agonist
  2. allosteric modulation: bind to a different site as the agonist → change in the binding affinity of the ligand (enhanced or decreased)

EC_{50} = const. E_{max} ↓

IC_{50} value



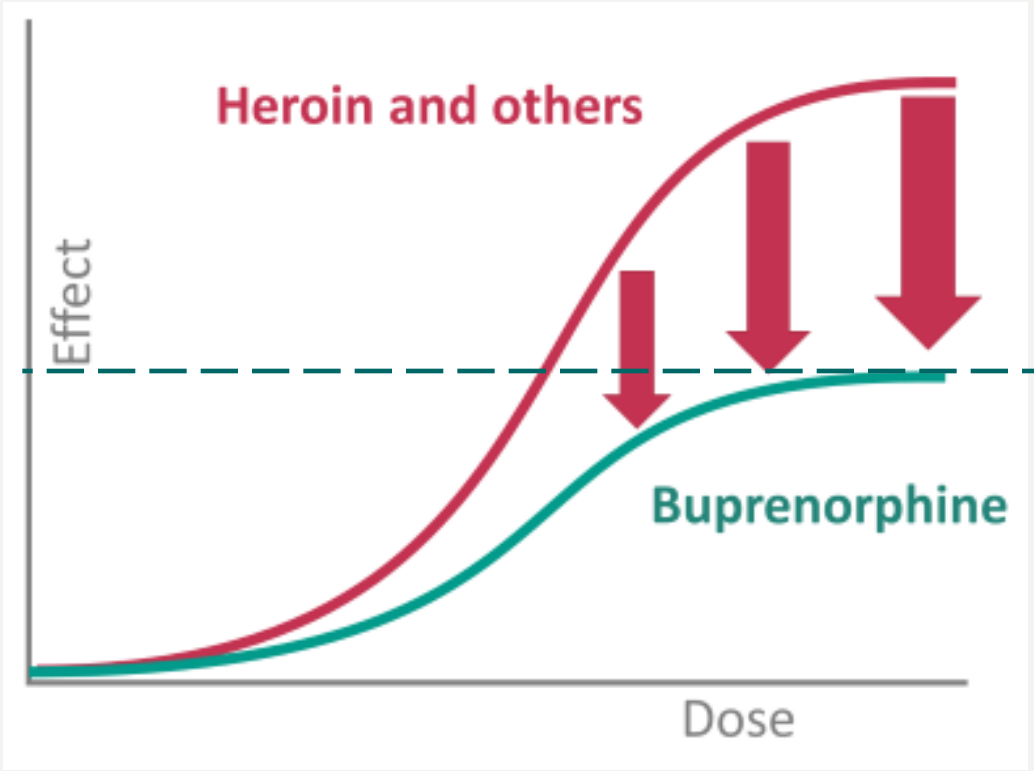
	IC_{50} (nM)
Dasatinib	0.42
Imatinib	474.90
Ponatinib	0.95
Nilotinib	2.85

https://www.sigmaaldrich.com/technical-documents/protocols/biology/transcreener-residence-time-protocol.html

- IC_{50} value: concentration of antagonist, that is required for 50% inhibition of receptor or enzyme

Partial agonists as antagonists

Clinical relevance



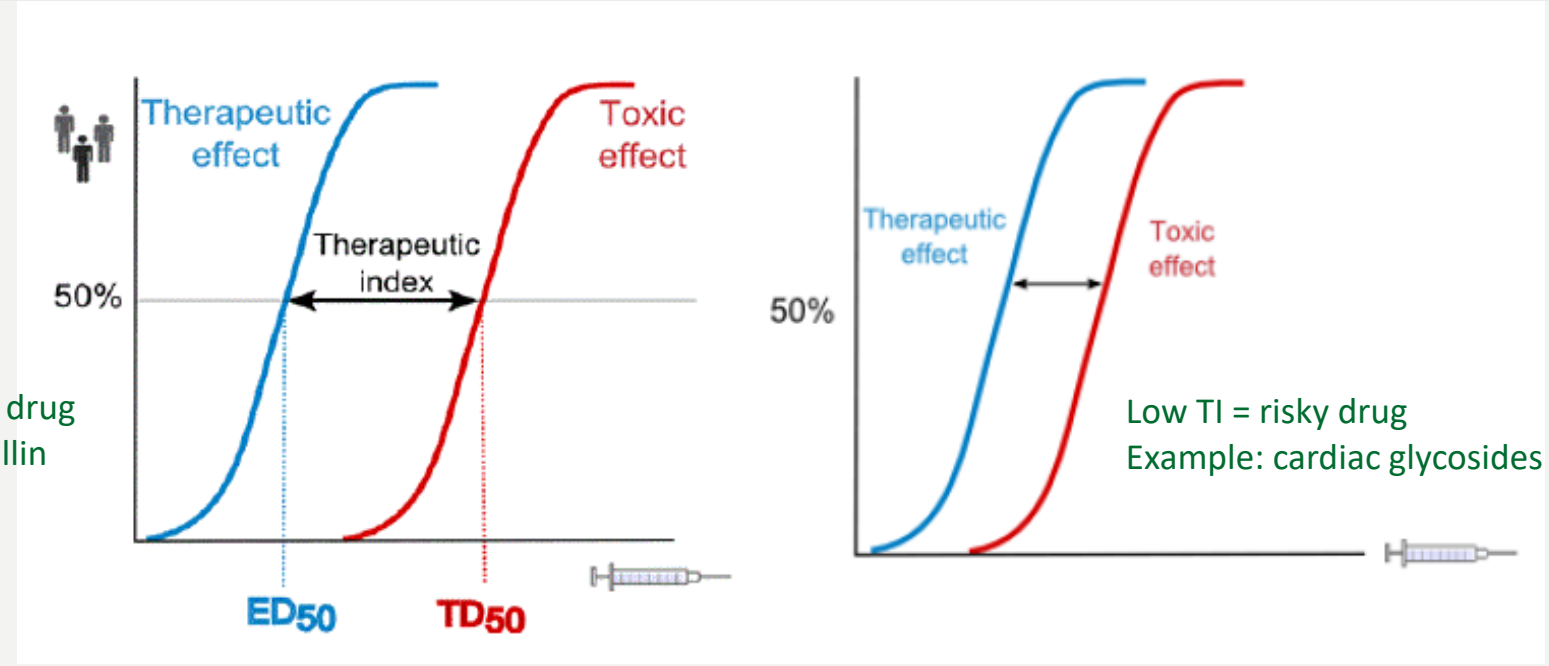
https://psychopharmacologyinstitute.com/clinical-psychiatry/substance-use-disorders/buprenorphine-opioid-use-disorder-mechanism-action/

- Full agonist + partial agonist → partial agonist acts as an antagonist: decrease in receptor activation

Ceiling effect

- Clinically used to treat opiate dependence (milder effects + lower abuse potential) → ceiling effect

Therapeutic Index



High TI = "safe" drug
Example: Penicillin

Low TI = risky drug
Example: cardiac glycosides

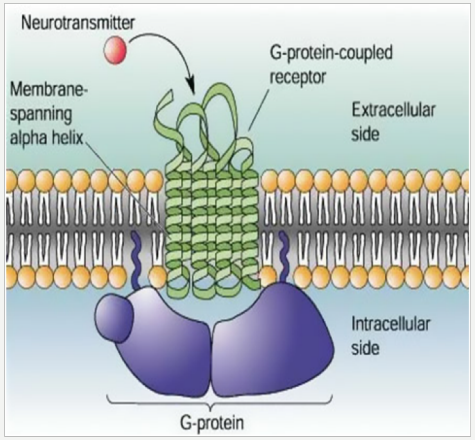
http://tmedweb.tulane.edu/pharmwiki/doku.php/basic_principles_of_pharm

- **Toxic response curve:** TD_{50} = dose producing death in 50 % of cases
- **Therapeutic Index:** measure of drug safety

$$TI = \frac{Median\ toxic\ dose}{Median\ effective\ dose} = \frac{TD_{50}}{ED_{50}}$$

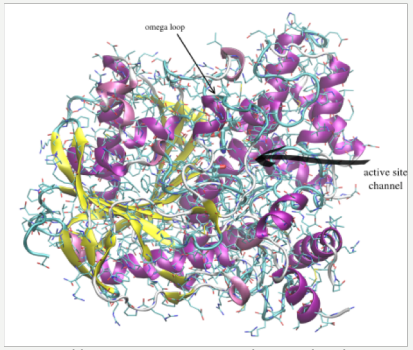
Targets for drugs/toxicants

- **Receptors**



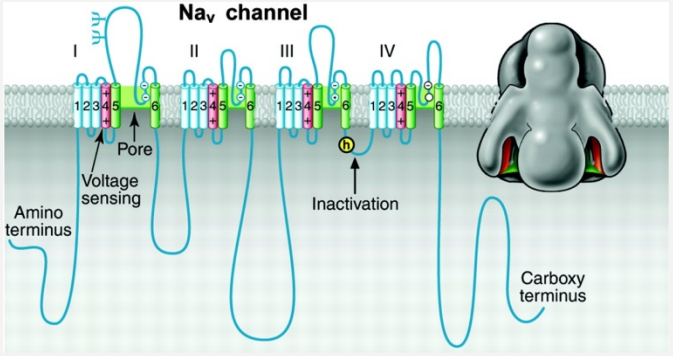
https://medcraveonline.com/JSRT/JSRT-02-00065

- **Enzymes**



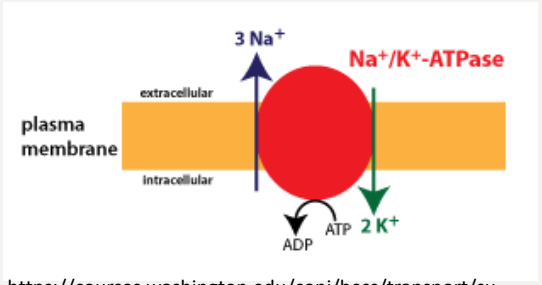
http://www.ncbr.muni.cz/group/lcc/acetylcholinesterase.html

- **Ion channels**



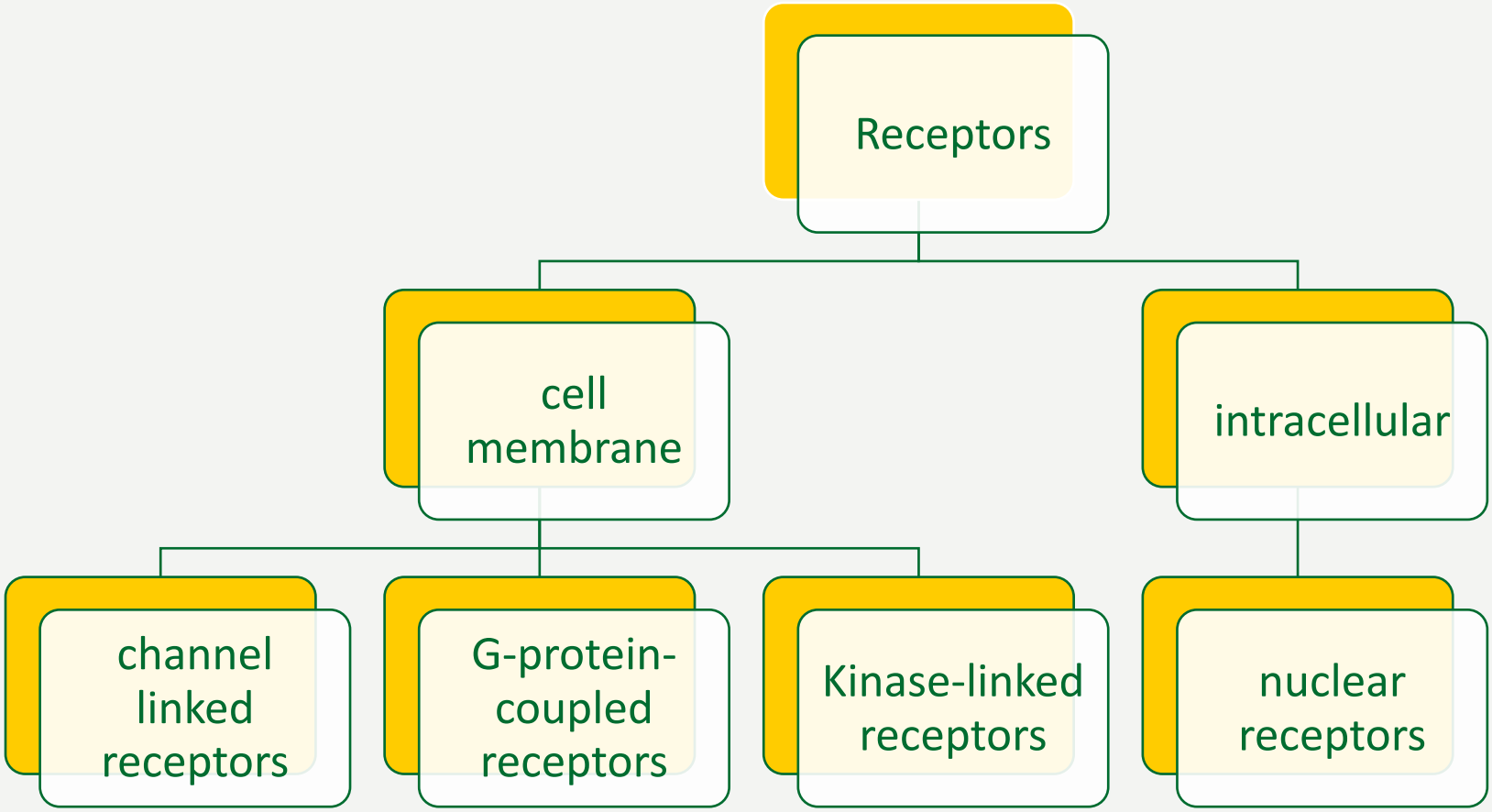
http://pharmrev.aspetjournals.org/content/57/4/387

- **Transporters**



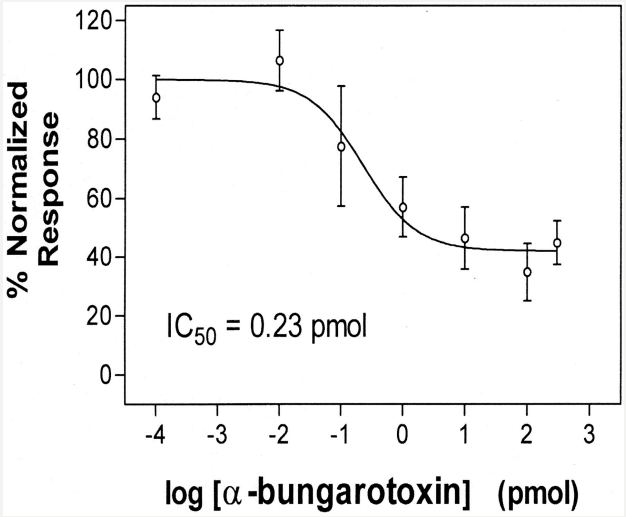
https://courses.washington.edu/conj/bess/transport/summary/NaKpump.png

Receptors


Receptors
cell membrane
intracellular
channel linked receptors
G-protein-coupled receptors
Kinase-linked receptors
nuclear receptors

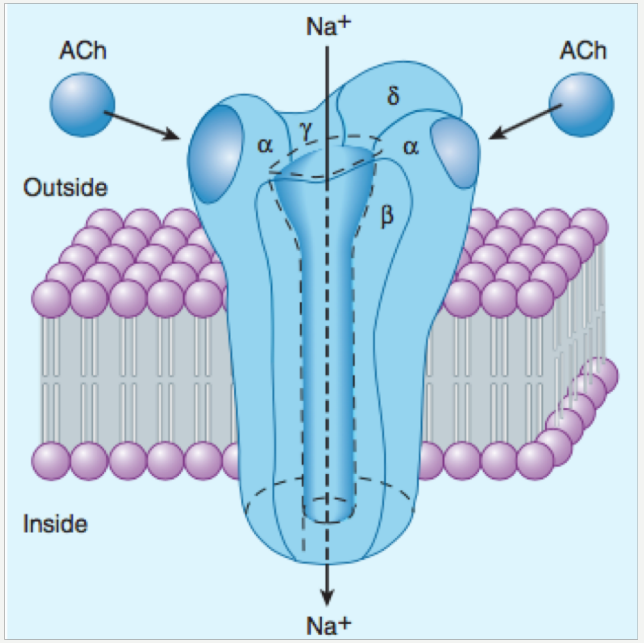
Channel linked receptors

- coupled directly to an ion channel
- crucially important for synaptic transmission in the CNS
- e.g. nicotinic Ach-receptor
- toxin: α-Bungarotoxin



http://jpet.aspetjournals.org/content/296/2/260

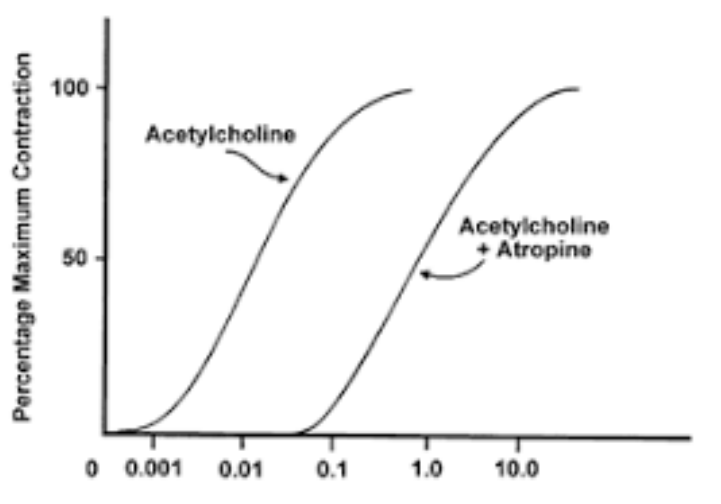
https://www.hongkongsnakeid.com/many-banded-krait



http://www.tomhsiung.com/wordpress/2014/12/ligand-and-voltage-gated-channels-receptors/

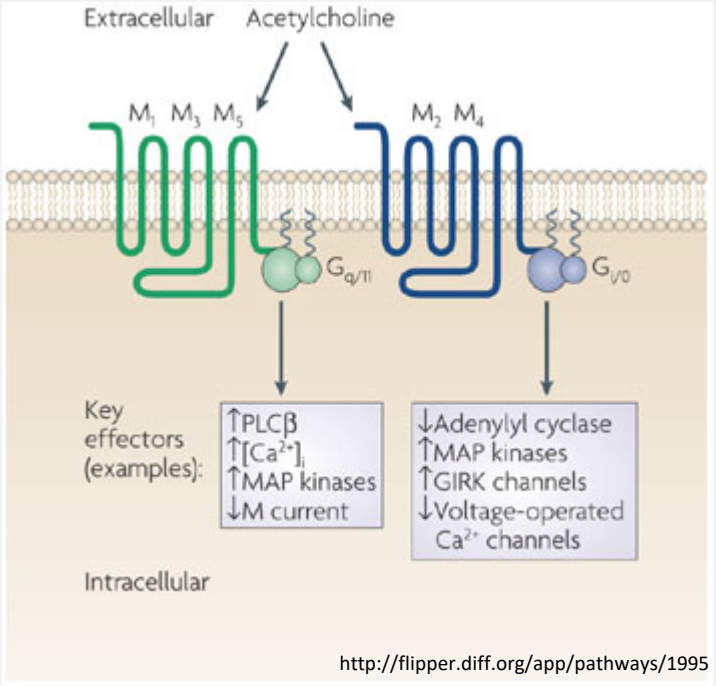
G-protein coupled receptors

- site of action of about 45% of drugs
- coupled to intracellular effector mechanism via G-proteins → enzyme activation or opening of ion channel
- e.g. muscarinic Ach-receptor
- ligand: atropine



https://www2.courses.vcu.edu/ptxed/m2/powerpoint/download/Damaj%20DR%20Curves.PDF

Competitive antagonist:

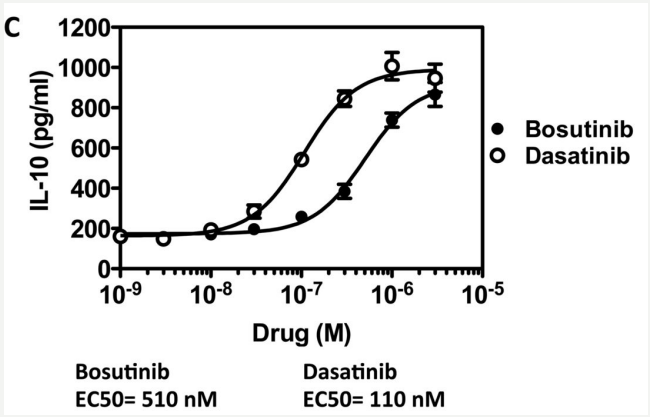


http://flipper.diff.org/app/pathways/1995

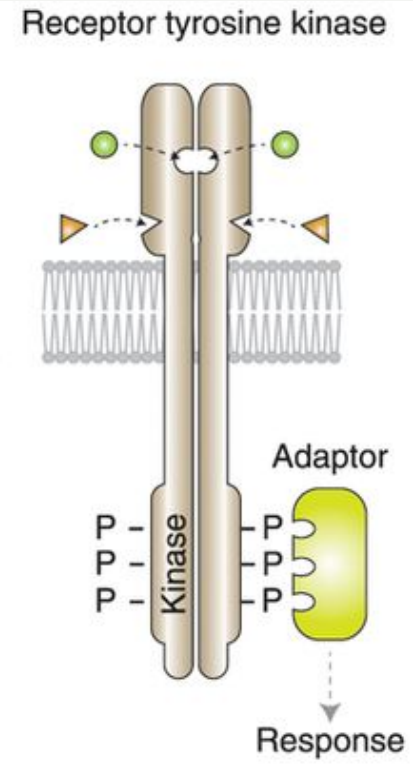
Kinase-linked receptors

- linked directly to an intracellular protein kinase that triggers a cascade of phosphorylation reactions
- receptor for cytokines, growth factors and hormones
- e.g. growth factor receptor
- ligand: dasatinib

Dasatinib is more potent in terms of IL-10 production



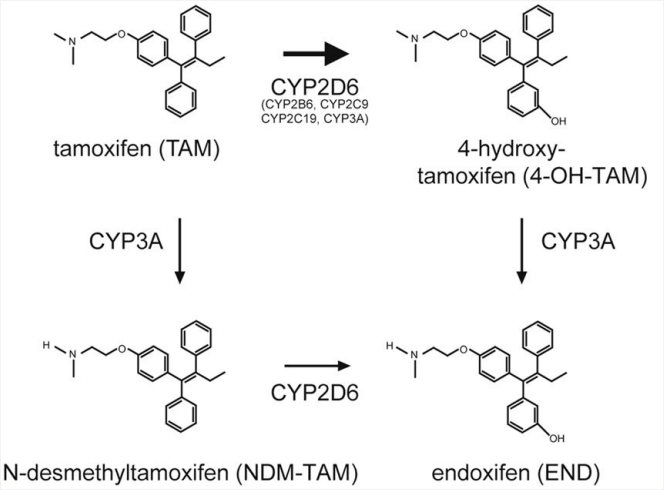
http://www.biochemj.org/content/465/2/271.figures-only



https://www.nature.com/articles/nbt.3028

Nuclear receptors

- intracellular receptors
- act as transcription factors that promote or inhibit synthesis of new proteins
- e.g. oestrogen receptor
- Ligand: tamoxifen

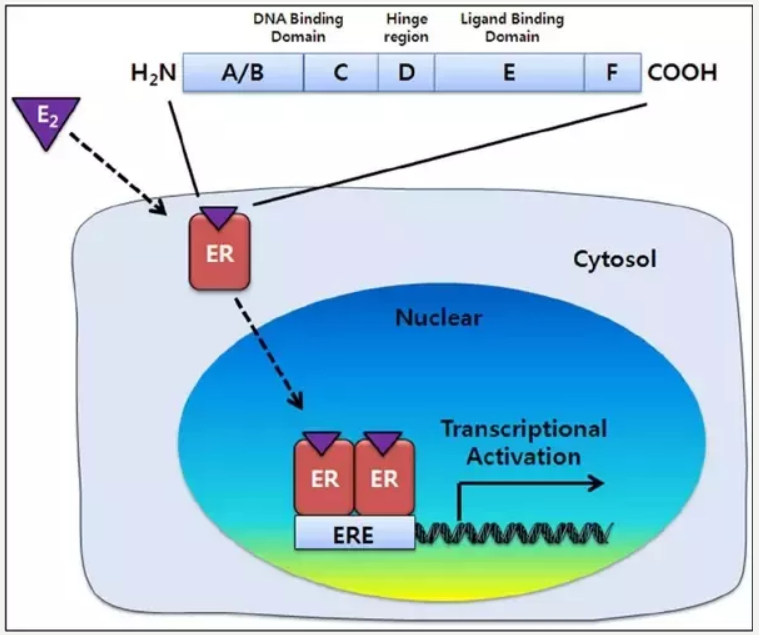


https://www.nature.com/articles/s41598-018-24085-9

K_d(4-OH-TAM) <<< K_d(TAM)

⬇

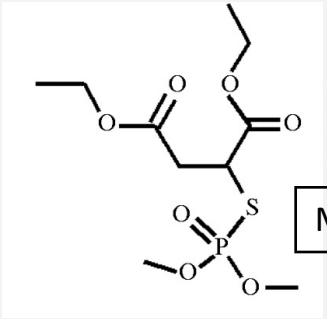
30 to 100 times greater affinity of 4-OH-TAM



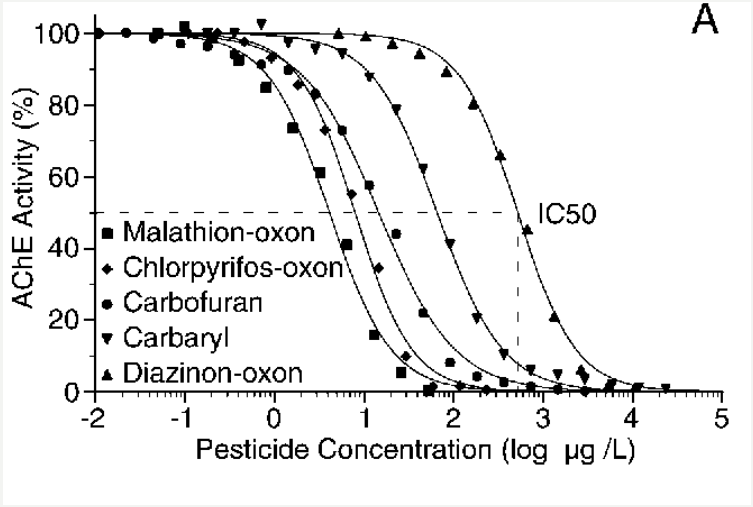
https://www.quora.com/Are-nuclear-receptors-attached-to-DNA-or-to-the-nuclear-membrane

Enzymes

- site of action of about 30% of drugs
- inhibition of the active site may be competitive or long lasting and irreversible
- e.g. acetylcholinesterase
- toxicant: organophosphates

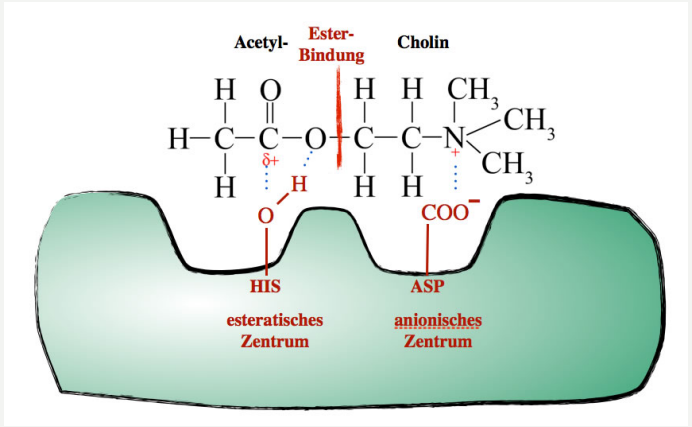


Malathion-oxon



https://www.researchgate.net/figure/The-concentration-dependent-inhibition-of-rat-brain-AChE-by-the-OP-pesticides_fig4_8369600

Malathion-oxon: lowest IC_{50} → highest potency



http://www.u-helmich.de/bio/lexikon/A/acetylcholinesterase.html

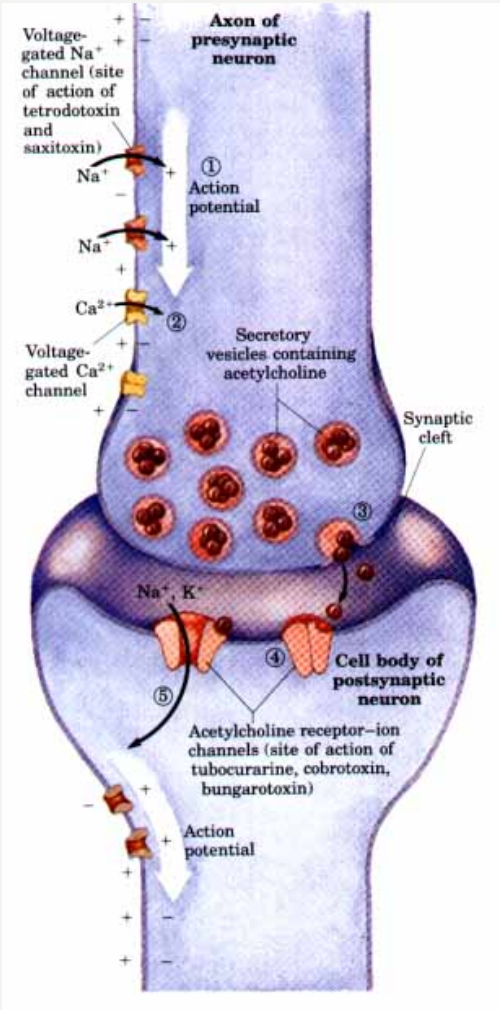
Ion channels

- ion channels that are activated by changes in the electrical membrane potential near the channel
- found in excitable tissues along the axons and synapses
- e.g. voltage gated sodium channel
- toxin: aconitine

Aconitum napellus

https://www.chemistryworld.com/podcasts/aconitine/1017356.article

LD for humans:
0,028mg/kg orally
→ 2,24mg for a 80kg person



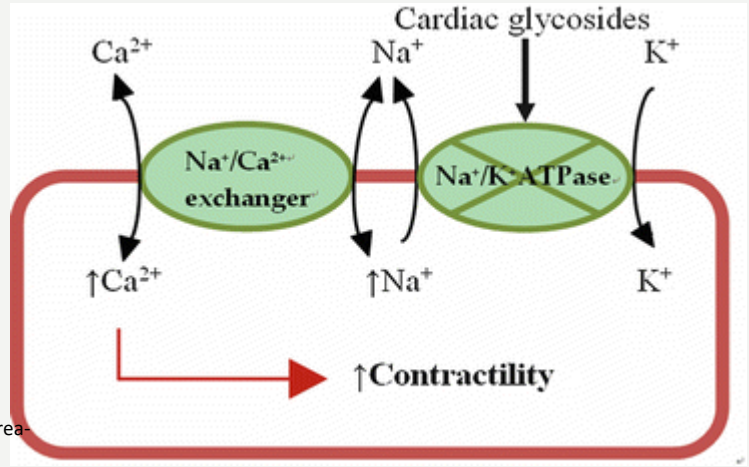
http://www.bioinfo.org.cn/book/biochemistry/chapt22/bio7.htm

Transporters

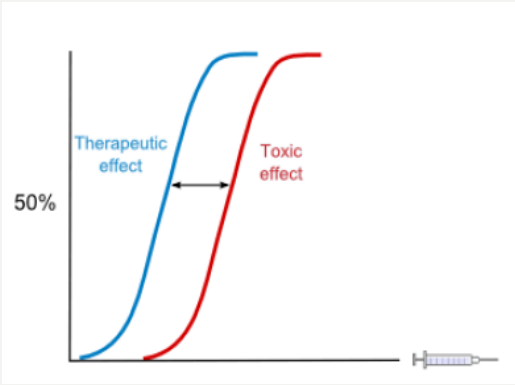
- specialized proteins that carry ions or molecules across the cell membrane
- transport may be passive or active
- e.g. Na^+/K^+-ATPase
- drug: cardiac glycoside

Digitalis pupurea

https://www.crocus.co.uk/plants/_/digitalis-purpurea-dalmatian-purple/classid.2000027281/



https://link.springer.com/referenceworkentry/10.1007%2F978-3-642-22144-6_159



TI (digoxin) ≈ 2:1

Thank you for your attention !