New Technology in The Design and Formulation of Anti-TB Drugs

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- Computer Aided Drug Design of Anti-TB Drugs
- New Technology in Formulation of Anti-TB Drugs

Computer Aided Drug Design:

Rational Drug Design:

The ***goal*** is to use ***what is known*** about a disease or an infectious agent ***to create*** safer, more effective drugs that act specifically to prevent the disease.

What is known about TB?

Causative organism: ***Mycobacterium tuberculosis***

Current Treatment: **Isoniazid (INH), Rifampicin, Ethambutol**

Problems with current treatment: **Emergence of multi-drug resistant TB (MDR-TB)**

Isoniazid: Action Mechanism

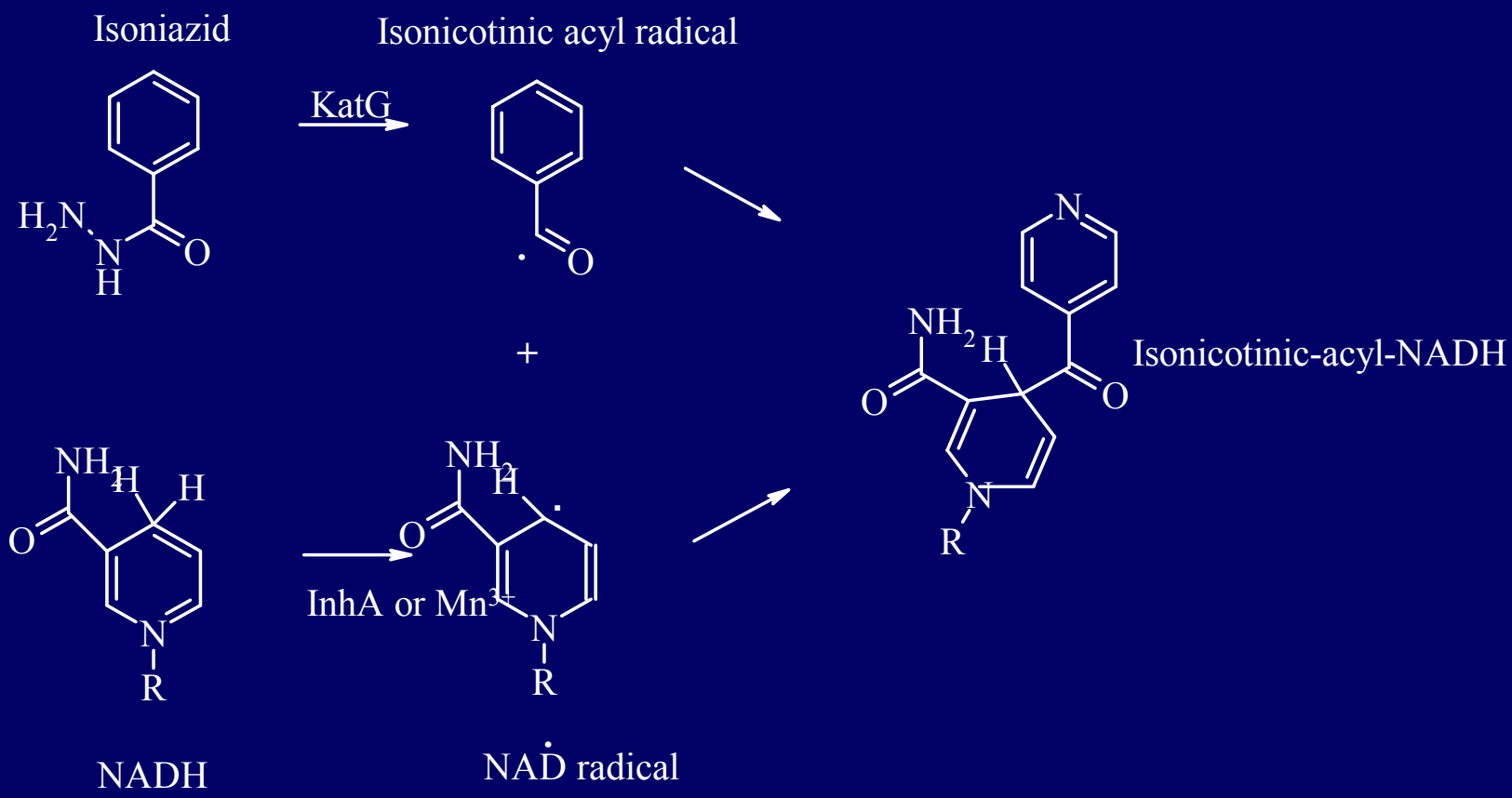
Destruction of mycobacterium cell wall?

- Direct disodering effect on the mycolic acid cell wall?
- Inhibition of enzymes involved in mycolic acid biosynthesis ?

Isoniazid: Resistance Mechanism

- Loss of catalase and peroxidase (KatG enzyme) activities;
- Mutation of other enzymes (such as InhA, AlpC, KatE)

INH is a prodrug (Johnsson and Shultz, 1994)… KatG required to form its active metabolite: Isonicotinic-acyl-NADH (INADH)



However, is KatG really responsible for the activity/resistivity?

Absence or mutation of KatG results in resistance to INH. (Zhang *et. al*., 1992, 1993).

However, Quemard, *et. al*., 1995 suggested that there must be other resistance mechanism as only 20-30% clinical isolates of *M. tuberculosis* lose the catalase-peroxidase activity.

Mutation in InhA

INADH binds to InhA to inhibit the enzyme activities which is important in the biosynthesis of mycolic acid cell wall. (Quemard *et al*., 1995, Rozwarski,*et al*., 1998).

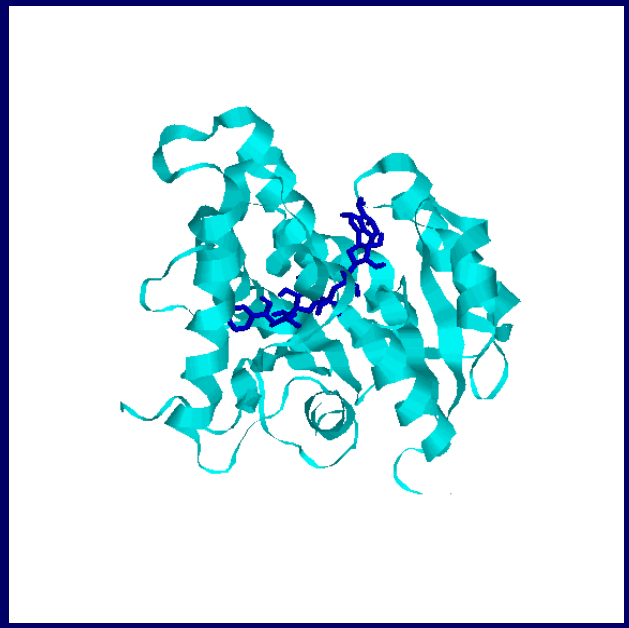
Mutation of the enzyme might explain the resistance towards INH treatment.(Banerjee *et al*., 1994)

Our Studies:

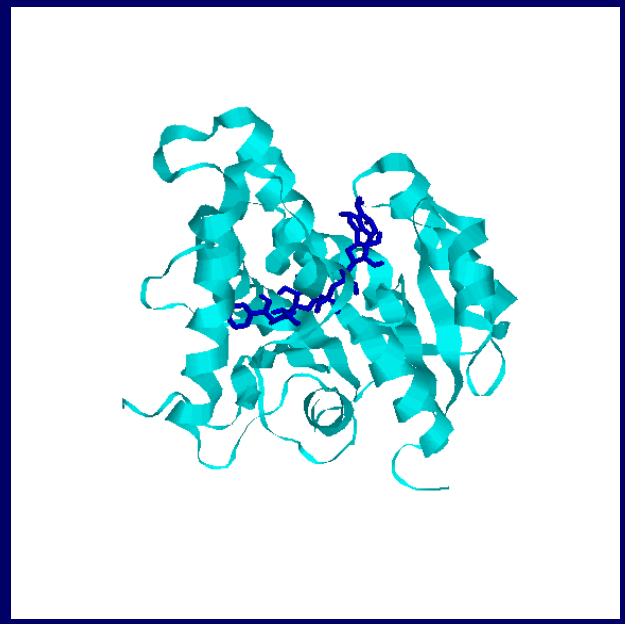
- Effect of INH on the cell wall
- Effect of INADH on InhA enzyme; wild and mutant-type.
  - Where is the binding site?
  - How strong is the binding?
- Repeat the experiment on INH and the various derivatives.
  a. Is there any correlation with the MIC value determined from experiment?

RESULTS:

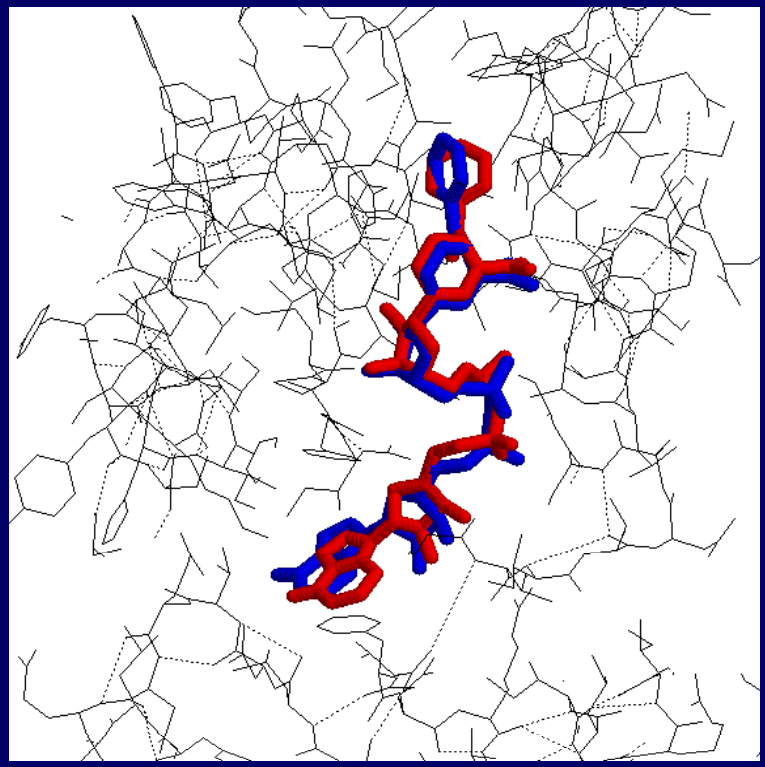
Binding Site of INADH



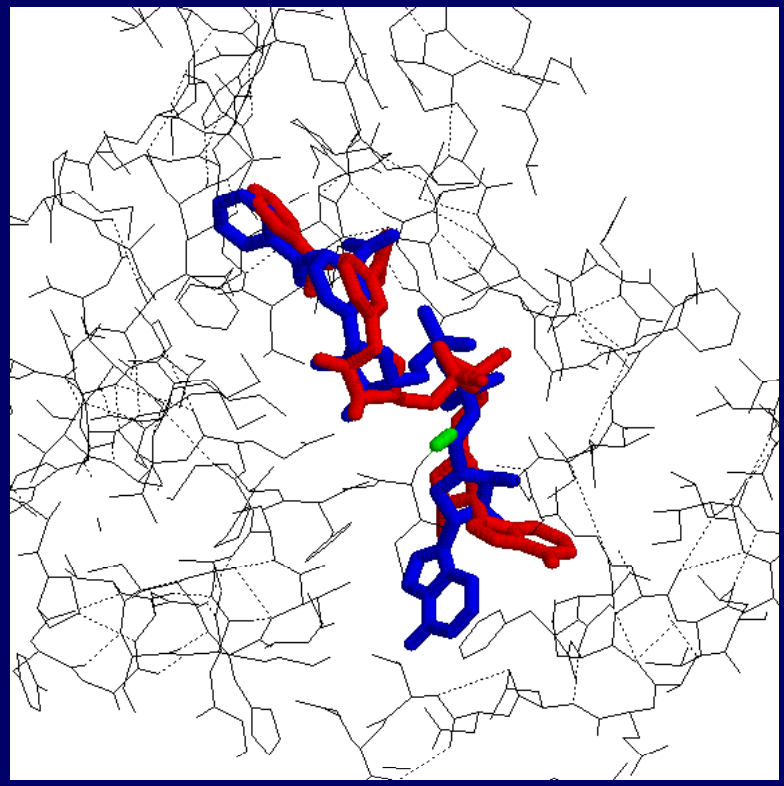
Wild Type



Mutant Type



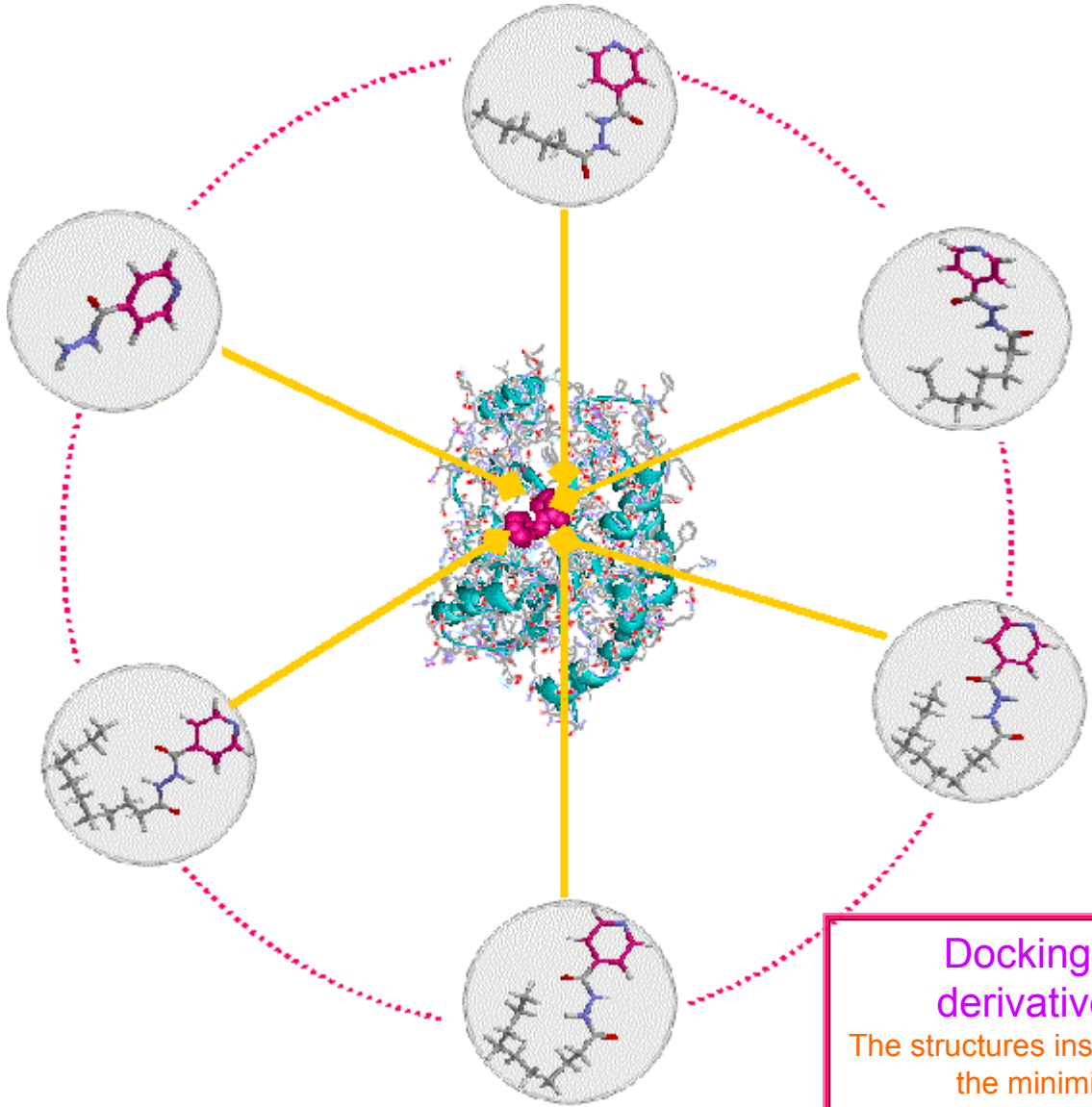
Wild Type



Mutant Type

Table 1: The predicted energy calculated from Docking of Isocotinic Acyl NADH (INADH)

Enzyme Type	*No in cluster*	*Tot. runs*	*Free energy (kcal/mol)*	*Final docked energy (kcal/mol)*	*Final inter-molecular energy (kcal/mol)*	*Final intra-molecular energy (kcal/mol)*	*RMSD from crystal Struct.*
Wild InhA	**25**	**100**	**-11.57**	**-18.91**	**-19.04**	**0.13**	**1.511 *A**
Mutant InhA	**100**	**100**	**-10.4**	**-17.74**	**-17.87**	**0.14**	**2.289 *A**



Docking of INH & its derivatives into InhA.

The structures inside the grey boxes are the minimised structures.

INH – isoniazid n n=no of carbon in the hydrocarbon chain.

Docking of INH and its derivatives onto InhA

Ligand	*Predicted Free Energy of Binding* (kcal/mol)
INADH	-11.57
INH	-5.53
INH5	-5.29
INH7	-4.95
INH8	-5.49
INH9	-5.87

The MIC values of the INH derivatives were found to be comparable if not lower than the parent molecule. This show that M.tuberculosis is susceptible to the derivatives regardless its hydrophobicity.

The conclusion from modeling studies:

- Metabolism of INH to INADH by KatG necessary for the activity. (Results support experimental evidence – Rouse et al., 1996)
- INADH bound in wild-type InhA in almost the same conformation as found by X-Ray Diffraction Data.
- Mutation of InhA results in lower binding free energy, thus lower activity (resistance)

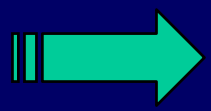
New technology in the formulation of anti-TB drugs

Problems in the current treatments:

- 8 million new cases every year (WHO)
- Directly-Observed Treatment, Short-course (DOTS)
- Involve less than 25% of those sick with TB

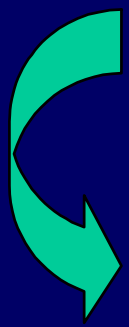
Difficulty in adopting the programme due to:

- cost of implementation
- require a trained team to supervise
- long duration of course
- inconsistent drug supply

failure  Multidrug-resistant tuberculosis

Improving bioavailability, acceptability and tolerance of antituberculosis drugs

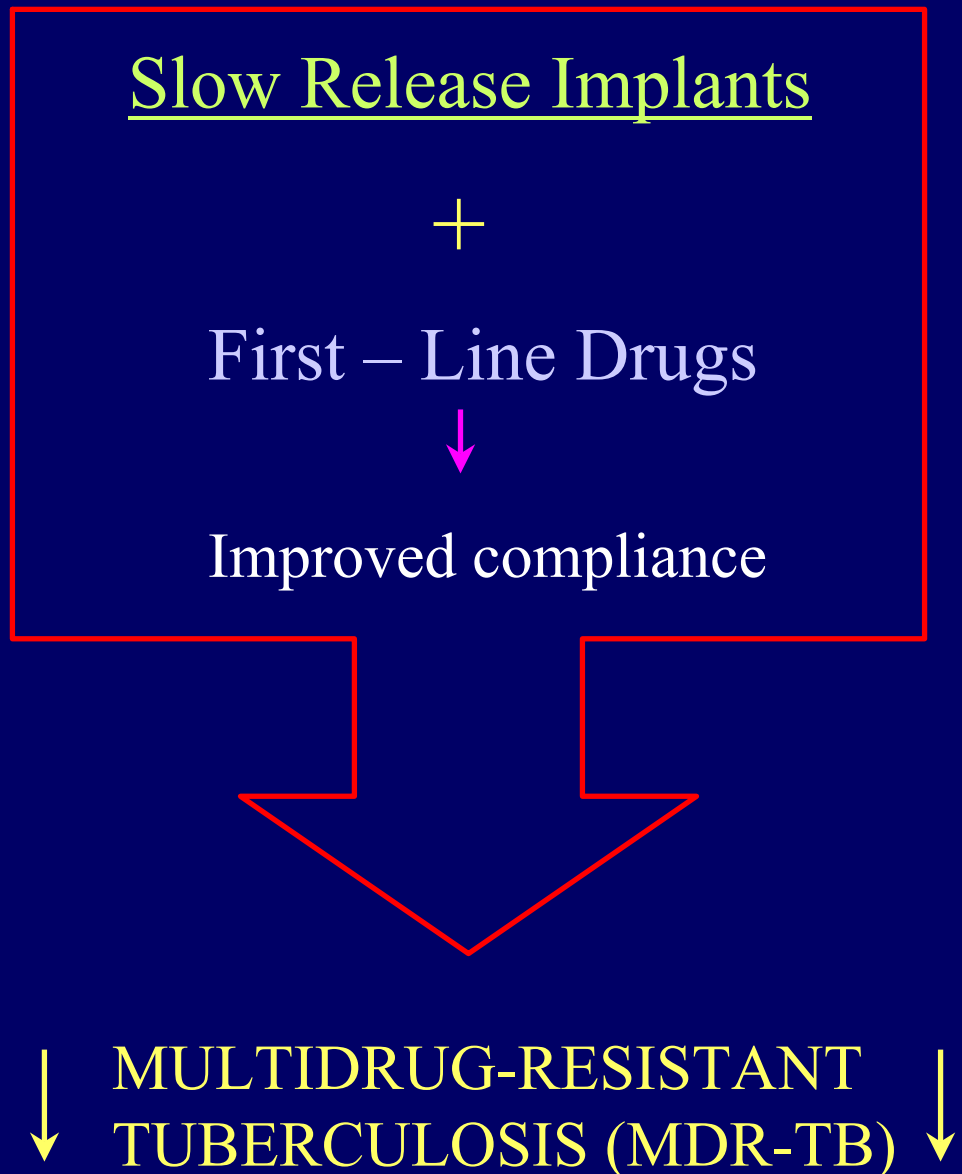
Impracticality of repetitive dosing



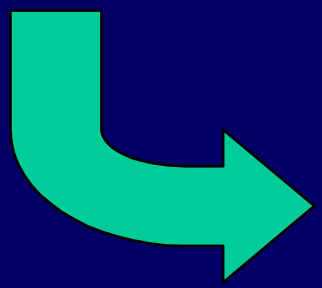
Slow Release Implants

Biodegradable polymers
eg. PLGA

IMPROVED COMPLIANCE


Slow Release Implants
+
First – Line Drugs
Improved compliance
MULTIDRUG-RESISTANT
TUBERCULOSIS (MDR-TB)

- MDR-TB treated with second-line drugs including aminoglycosides, thioamides, fluoroquinolones, cycloserine and para-aminosalicylic acid (PAS)
  - ➢ more toxic



Liposomes

❖ Anti-TB drugs targeting using liposomes need the knowledge of

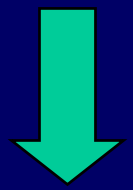
- carrier characteristics eg. size, surface charge, surface groups
- targeted cells eg. cell membrane composition, fluidity
- interactions of drugs with phospholipids

❖ Able to additionally prolong the release by grafting macromolecules onto liposomes

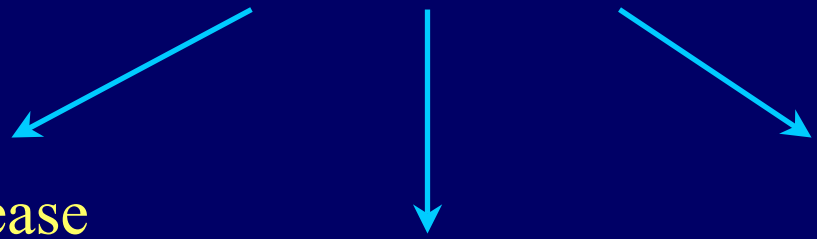
- Using lung as the target of delivery by aerosol,

  - dose can be reduced
  - abolish painful injections for drugs not absorbed via GI tract
  - act at the site of infection directly
  - sustained-release rate delivery

Another lipidic carrier that can increase the bioavailability of drugs:



Polyglycolised glycerides



controlled-release

taste-masking

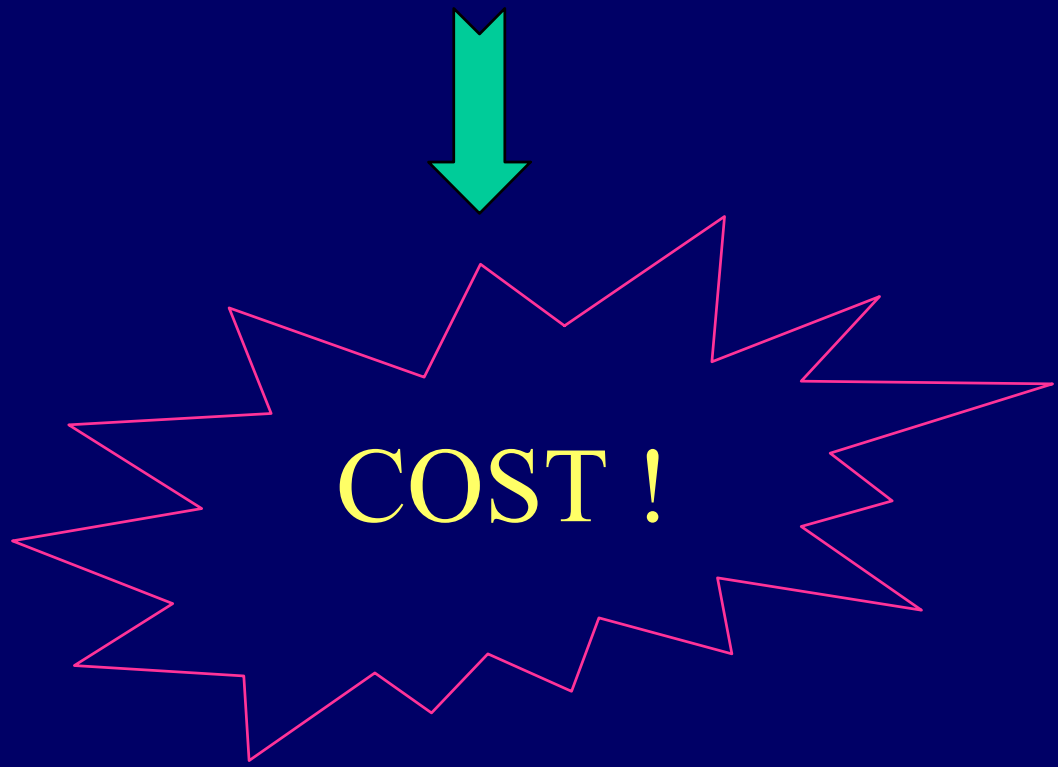
avoid first-pass metabolism

- Other formulations include

- Niosomes (non-ionic surfactant vesicles)
  - → increase bioavailability
  - → controlled-release delivery
  - → cheap and stable alternative to liposomes

- pH-sensitive nano and microparticles
  - → deliver at the site of absorption

Rate limiting step



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