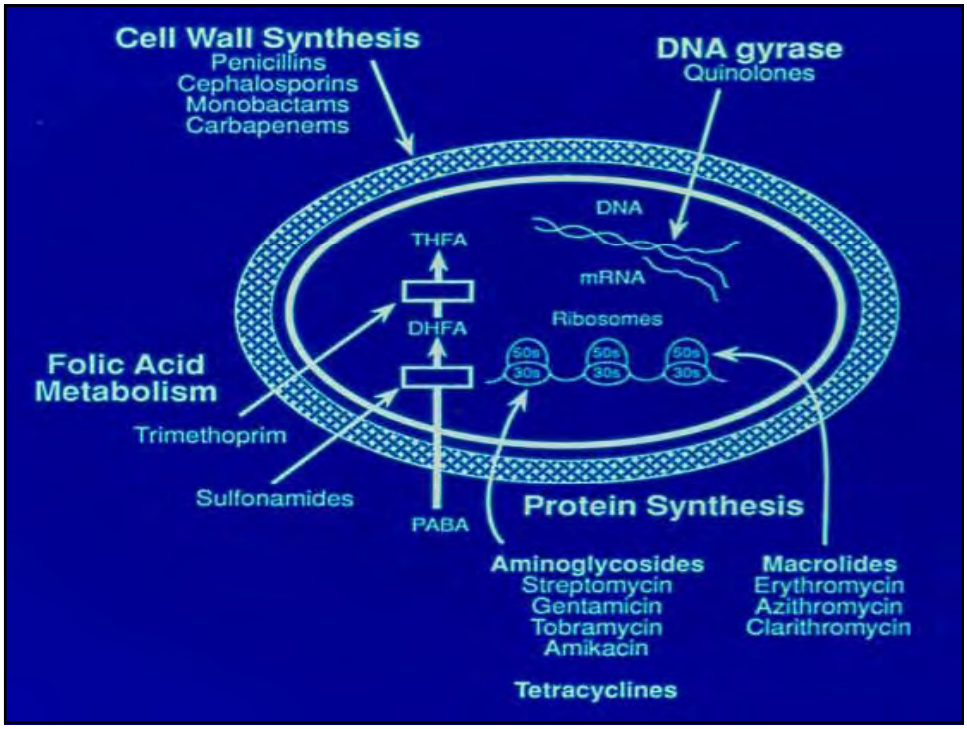


Beta-lactam antibiotics

Penicillins

Target - Cell wall - interfere with cross linking
Actively growing cells

Bind to **Penicillin Binding Proteins**

Enzymes involved in cell wall synthesis

Activity of an Antibiotic

Affinity for target

Permeability properties
(ability to get to the target)

Stability to bacterial enzymatic degradation

Bacterial modifications:

1 – Mutate target - ? More than one protein
Importance of the target –
? Essential

2. Permeability – Size/charge considerations
? Substrate for an efflux pump

3. Selection for mutants that destroy the antibiotic

WHO discovered the penicillins??

Abess Hildegarde von Bingen ?

"Good things that grow on the sides of trees…."

Fleming –

Florey – WWII….

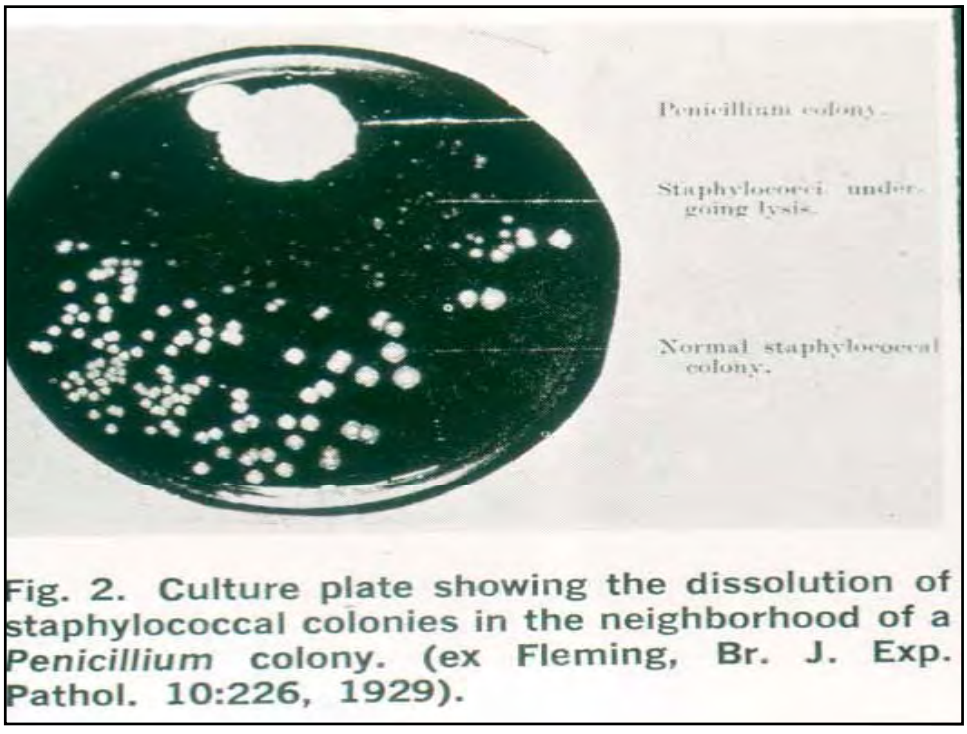
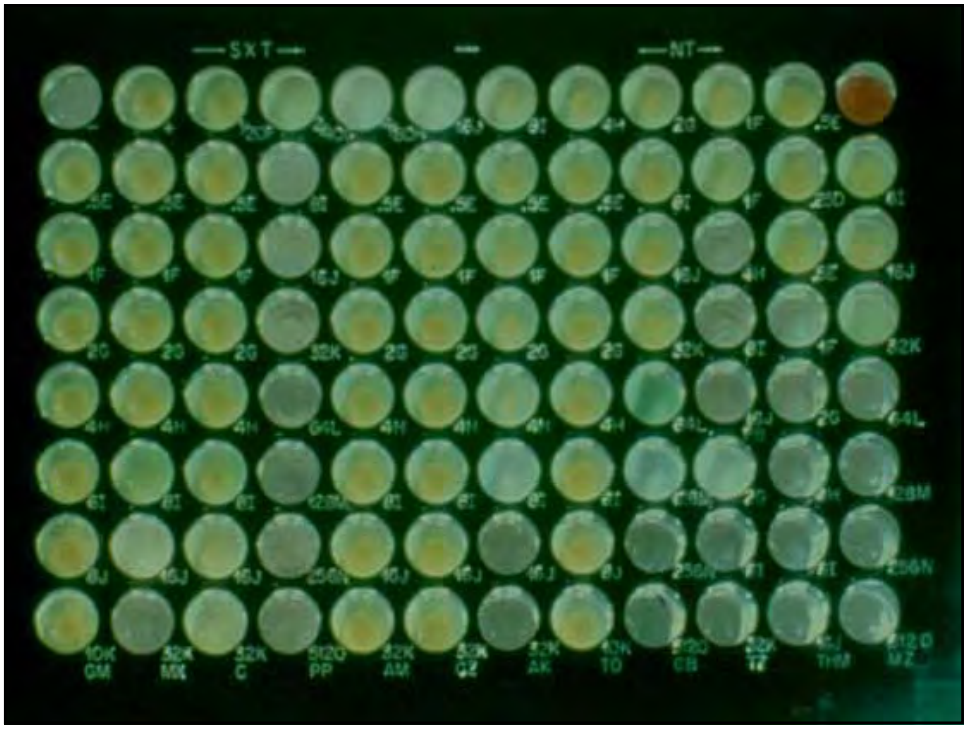


Fig. 2. Culture plate showing the dissolution of staphylococcal colonies in the neighborhood of a *Penicillium* colony. (ex Fleming, Br. J. Exp. Pathol. 10:226, 1929).



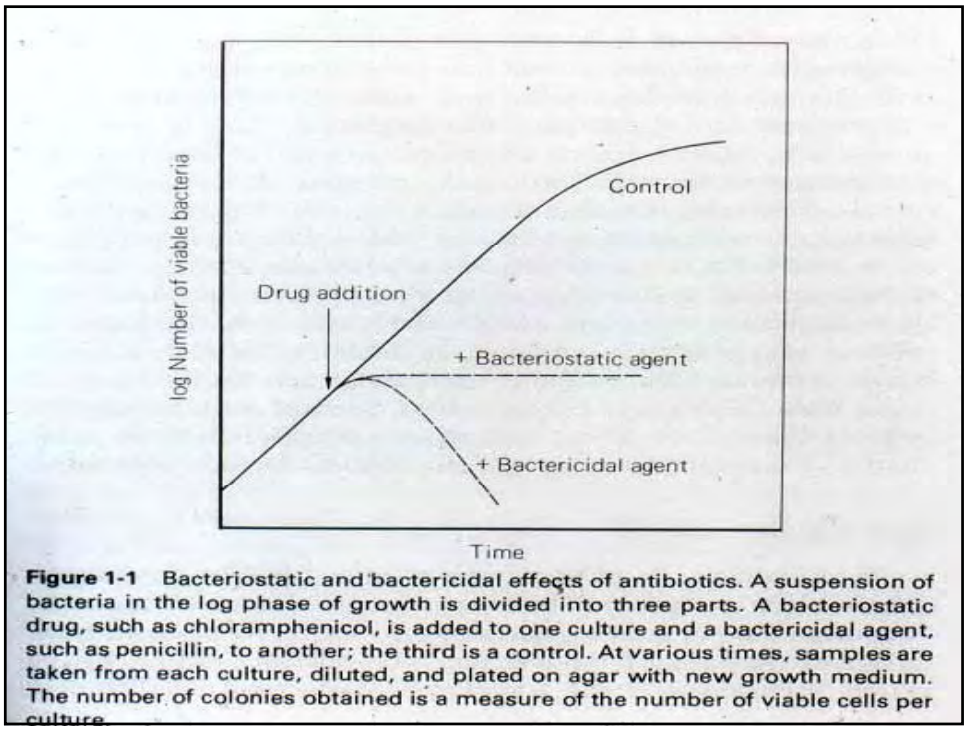
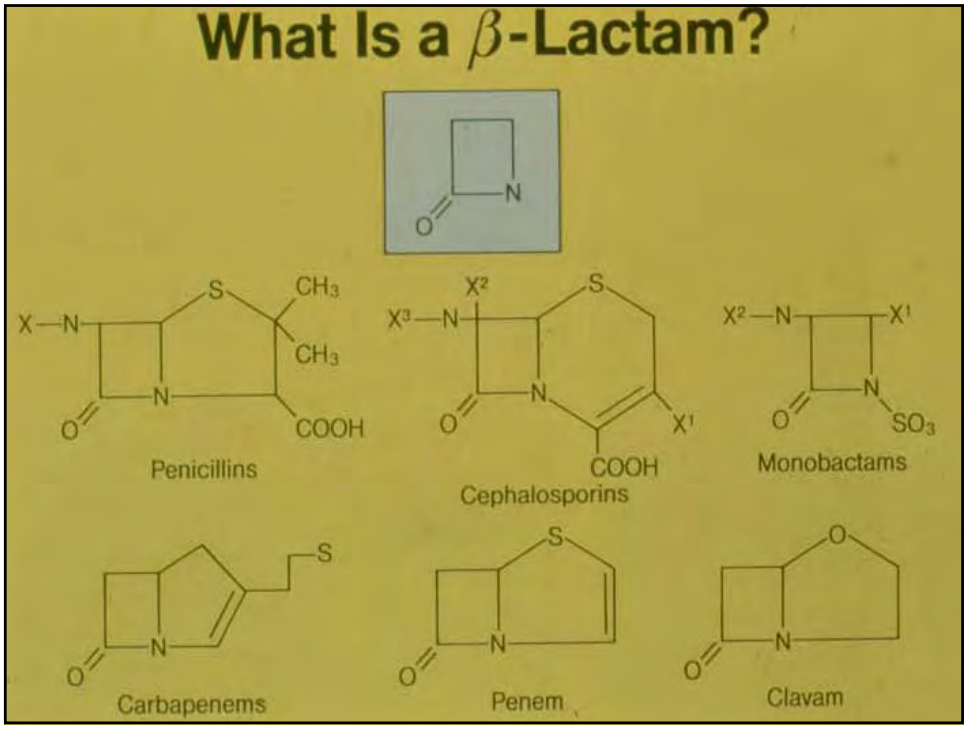
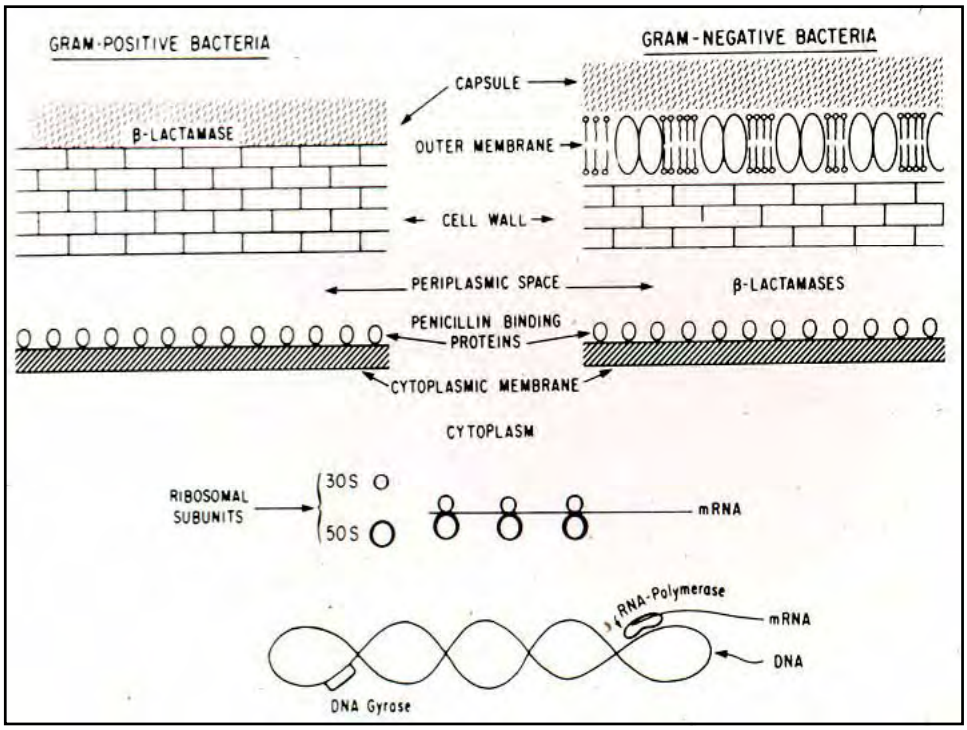
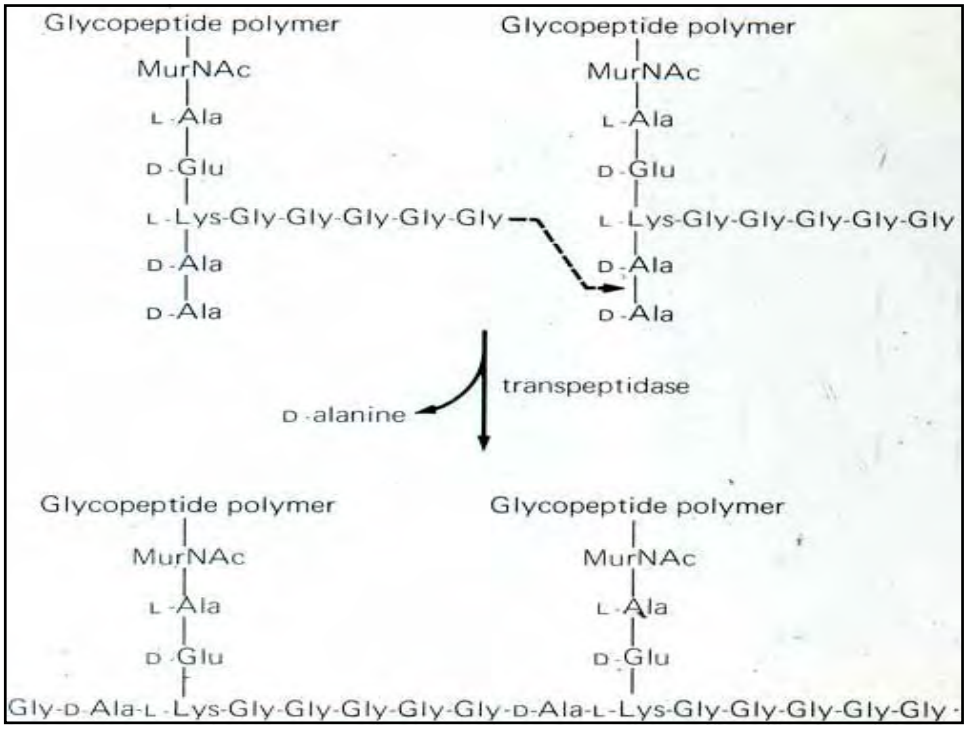


Figure 1-1 Bacteriostatic and bactericidal effects of antibiotics. A suspension of bacteria in the log phase of growth is divided into three parts. A bacteriostatic drug, such as chloramphenicol, is added to one culture and a bactericidal agent, such as penicillin, to another; the third is a control. At various times, samples are taken from each culture, diluted, and plated on agar with new growth medium. The number of colonies obtained is a measure of the number of viable cells per culture.


What Is a β-Lactam?
Penicillins
Cephalosporins
Monobactams
Carbapenems
Penem
Clavam


GRAM-POSITIVE BACTERIA
GRAM-NEGATIVE BACTERIA
CAPSULE
β-LACTAMASE
OUTER MEMBRANE
CELL WALL
PERIPLASMIC SPACE
β-LACTAMASES
PENICILLIN BINDING PROTEINS
CYTOPLASMIC MEMBRANE
CYTOPLASM
RIBOSOMAL SUBUNITS
30S
50S
mRNA
RNA-Polymerase
mRNA
DNA
DNA Gyrase



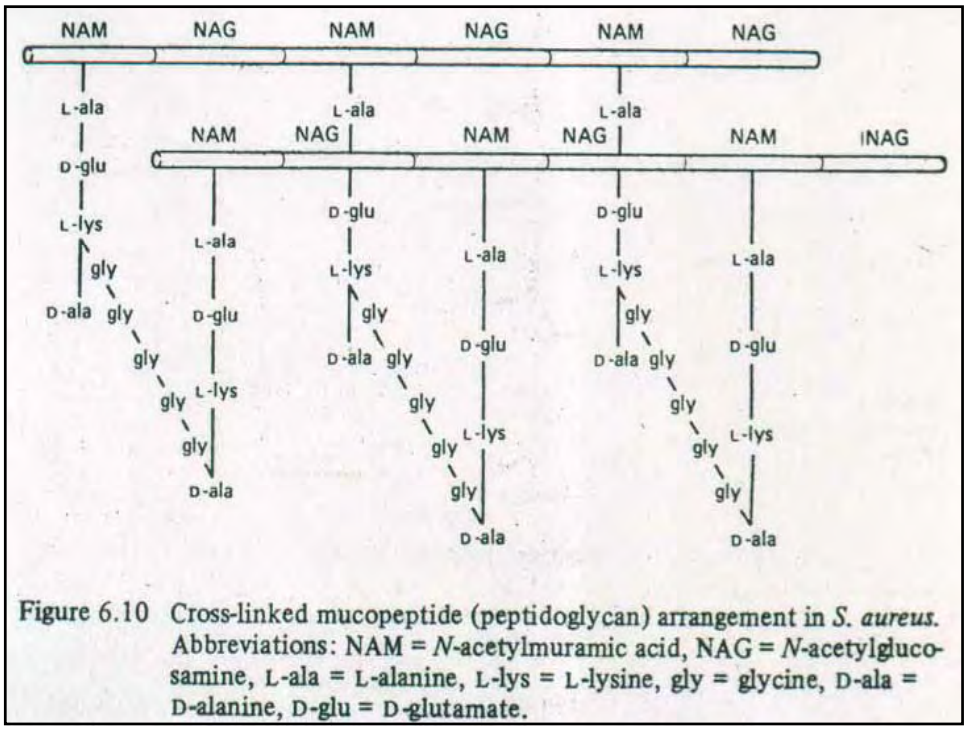


Figure 6.10 Cross-linked mucopeptide (peptidoglycan) arrangement in *S. aureus*. Abbreviations: NAM = *N*-acetylmuramic acid, NAG = *N*-acetylglucosamine, L-ala = L-alanine, L-lys = L-lysine, gly = glycine, D-ala = D-alanine, D-glu = D-glutamate.

Penicillin binding proteins

Transpeptidases

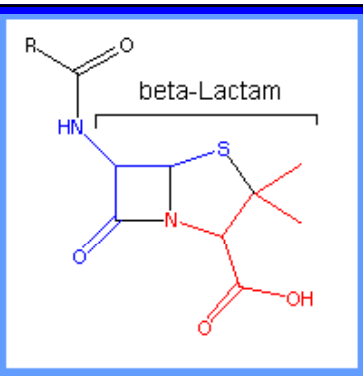
Carboxypeptidases

Differ in Gram (+) and in Gram (-) bacteria

Differ in abundance

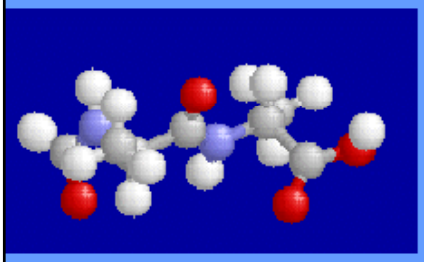
Similarity in structure of cell wall peptide and penicillin

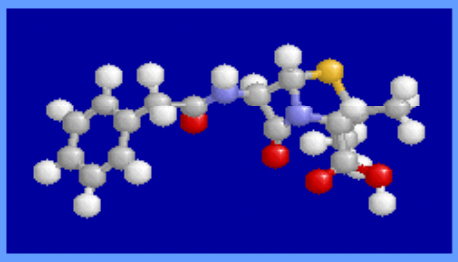
D-alanine-alanine



Trace red oxygen and blue nitrogen backbone

Penicillin





Ophardt, c. 2003

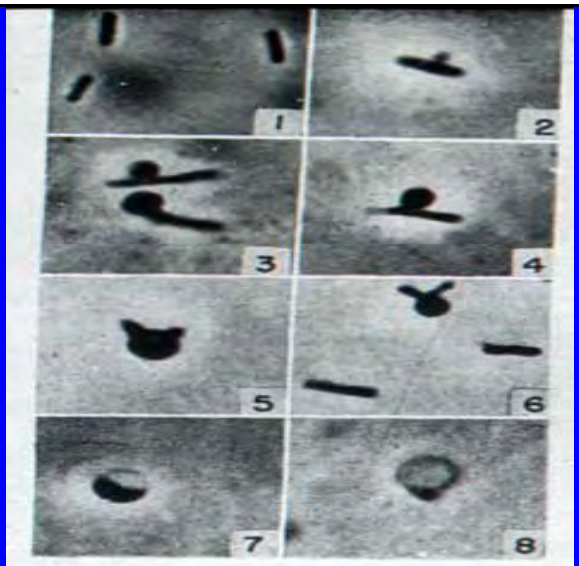
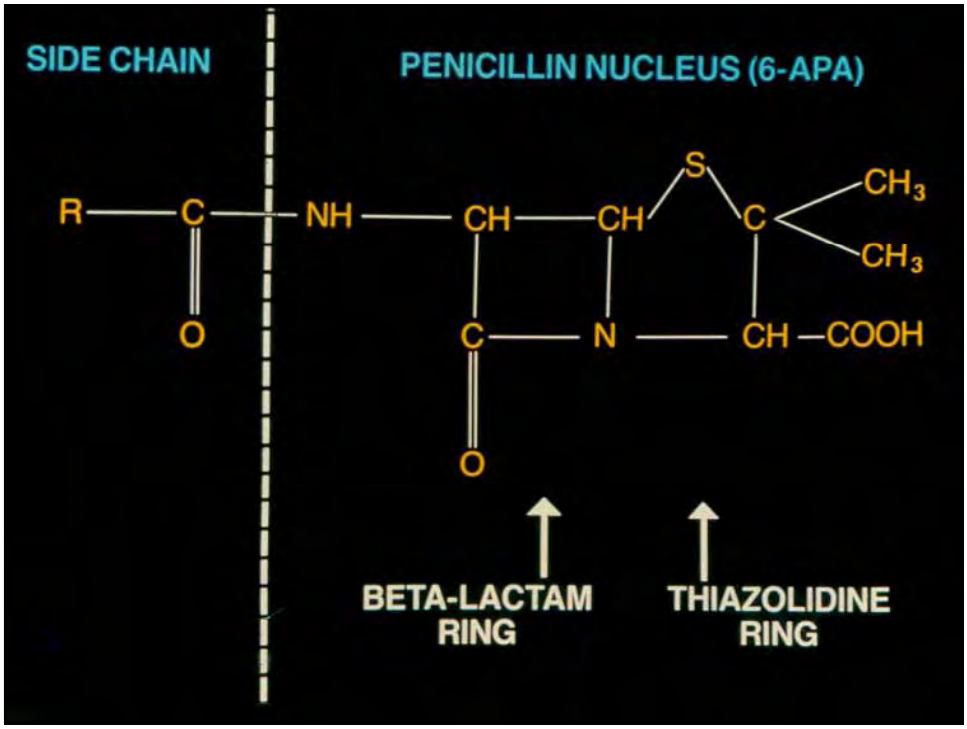


Fig. 2. Sequential phases of penicillin-induced lysis of *E. coli* B: 1, Bacteria immediately after addition of penicillin; 2, 3, and 4, emergence of globular extrusions; 5 and 6, "rabbit ear" forms; 7, partially vacuolized globular structure; 8, "ghost" form of a cytoplasmic membrane.

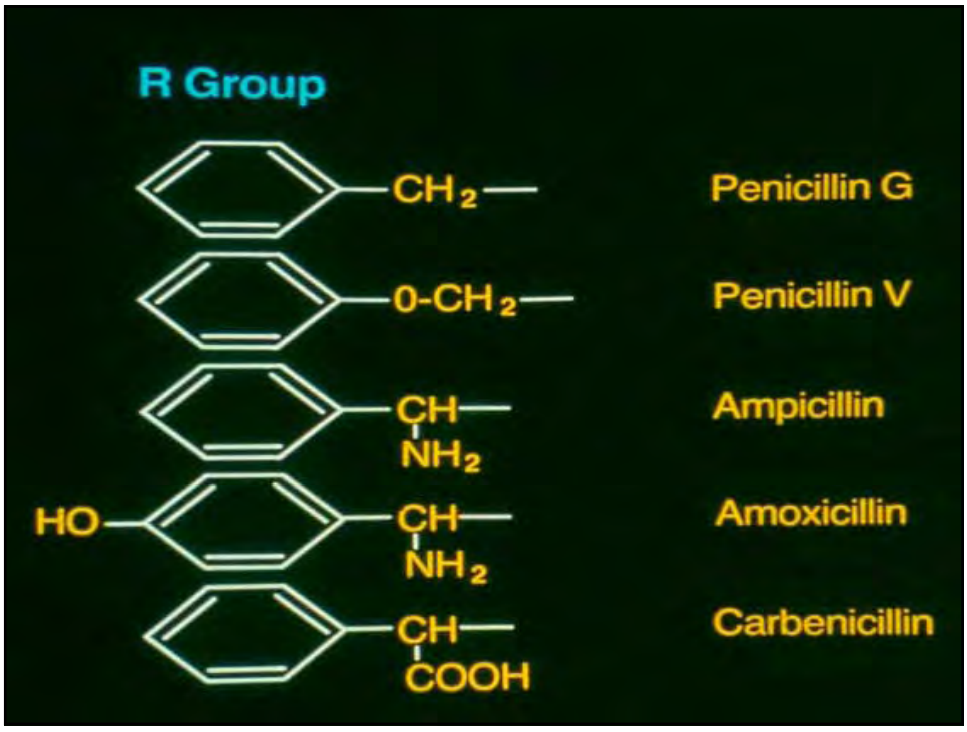


Activity of the beta-lactam antibiotic:

Affinity for critical PBP's (number of copies of the target)

Ability to get to the target (permeability properties – more of an issue for Gram negs)

Stability to beta-lactamases - degradation



Beta-lactamases - cleave the beta-lactam ring -
inactivate the drug -
Open ring - can't bind to the target

Co-evolved with the penicillin binding proteins

Share a ser-X-X-lys - binding site for interactions

Gram positives - Secreted into the environment
Gram negatives - Secreted into the periplasmic space

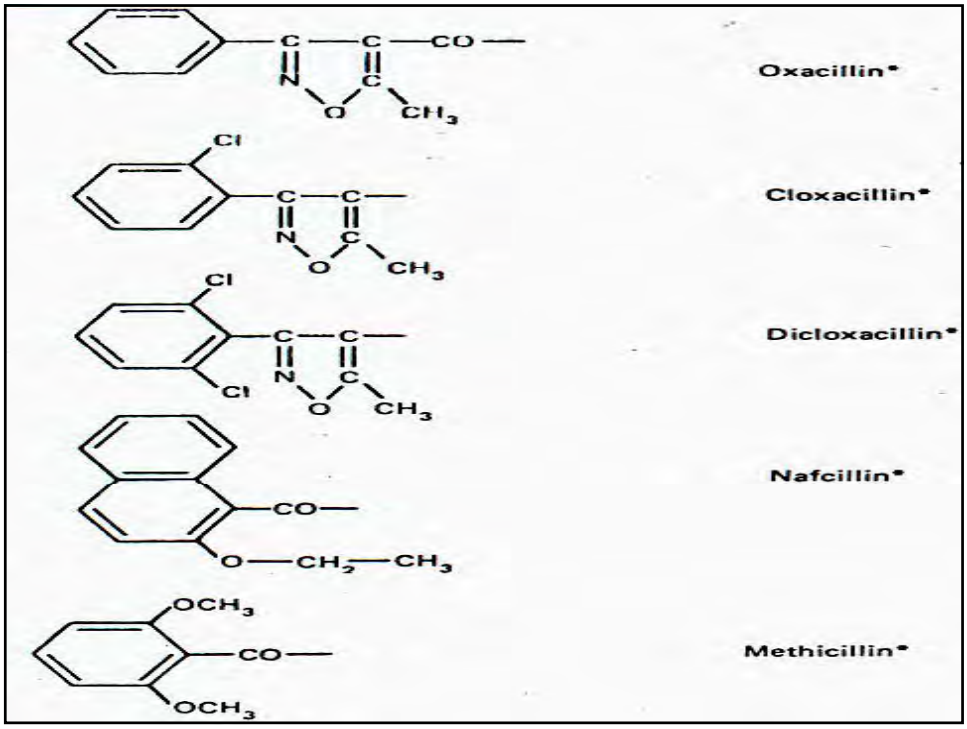
ANTI-staphylococcal penicillins

"semi-synthetic"

Add bulky side chains to provide

STERIC HINDRANCE to protect the Beta-lactam nucleus –

Gram positives – secrete bla's – "cloud"

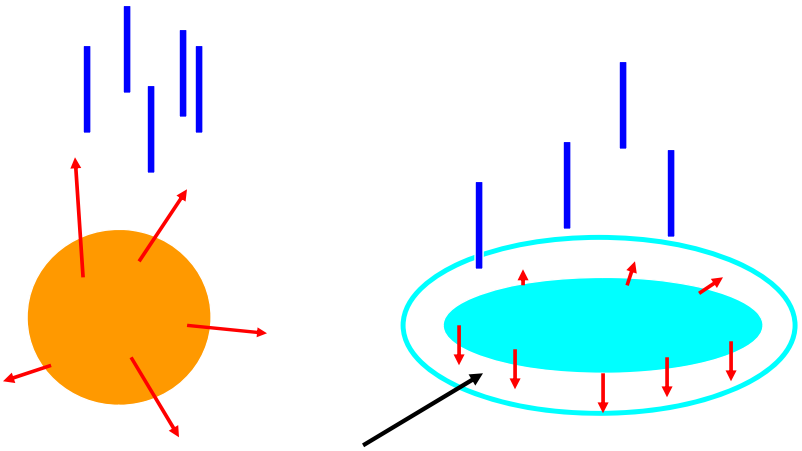


Anti-staphylococcal penicillins

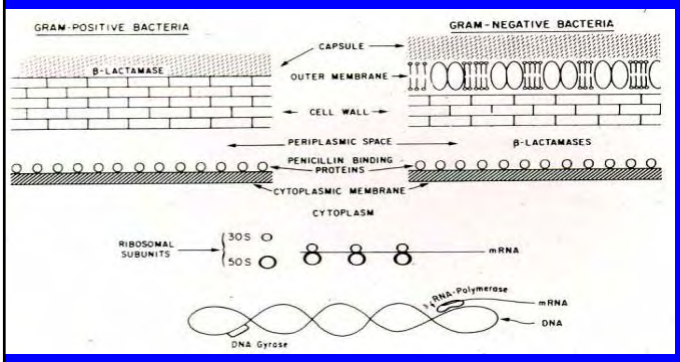
Strategy - Add a bulky side group to block beta-lactamase

(Methicillin) - renal toxicity
Nafcillin
Oxacillin
Cloxacillin (di-clox) - oral drugs

Beta-lactamase secretion in Gram positive and Gram negative bacteria



Periplasmic Space – Gram Negative



Capsule – No barrier

Peptidoglycan – No barrier

Outer wall – Porins Control access

Cytoplasmic Membrane – Lipid Bilayer

Beta-lactamases

Regulation - Consititutive - Chromosomal (*E*.coli)

Plasmid mediated -
copy number dependent

Inducible - chromosomal - SPACE organisms - as a model

2-component signaling - (*ampD, ampE, ampR*)
- Sensor
- Response regulator
- Transcriptional activator

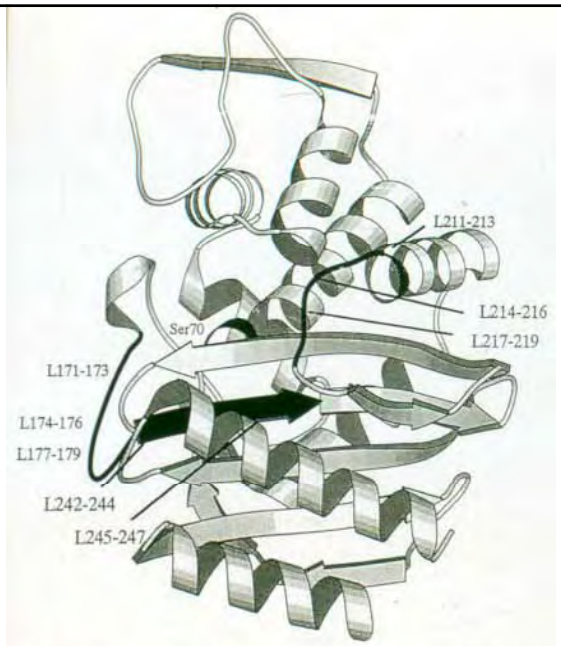
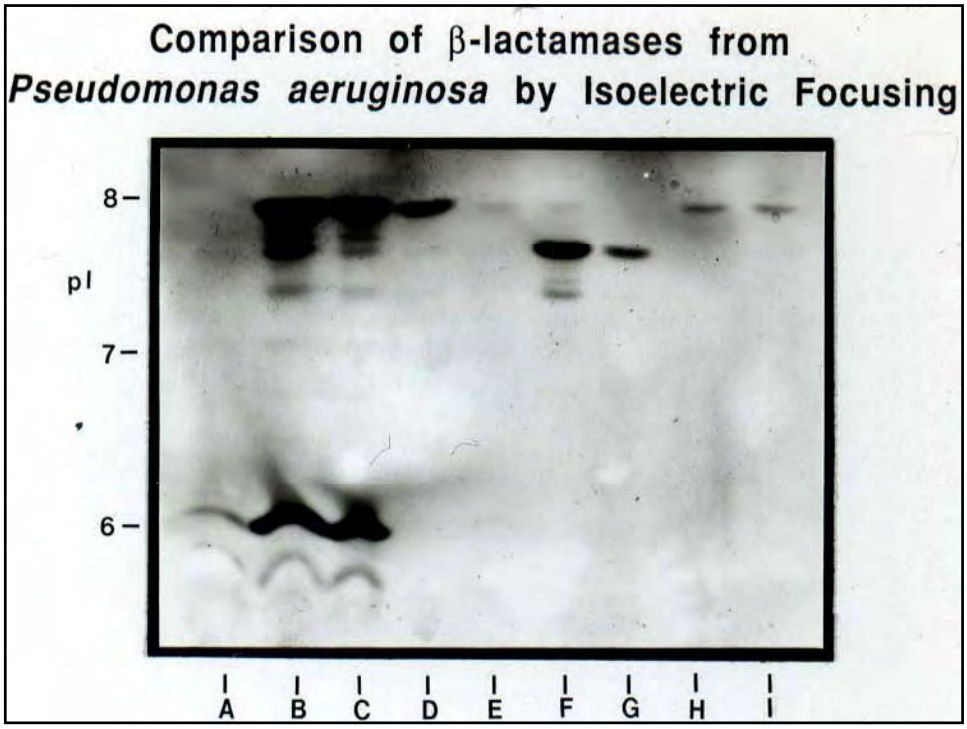


FIG. 2. Ribbon diagram of TEM-1 β-lactamase. The locations of the randomized library positions are indicated. The figure was prepared by using MOLSCRIPT (16).



Drugs in clinical use:

Penicillin G, VK

Ampicillin (+) clavulanic acid (beta-lactamase inhibitor)

(oral or parenteral)

Piperacillin - anti-Pseudomonas (+tazobactam)
(parenteral)

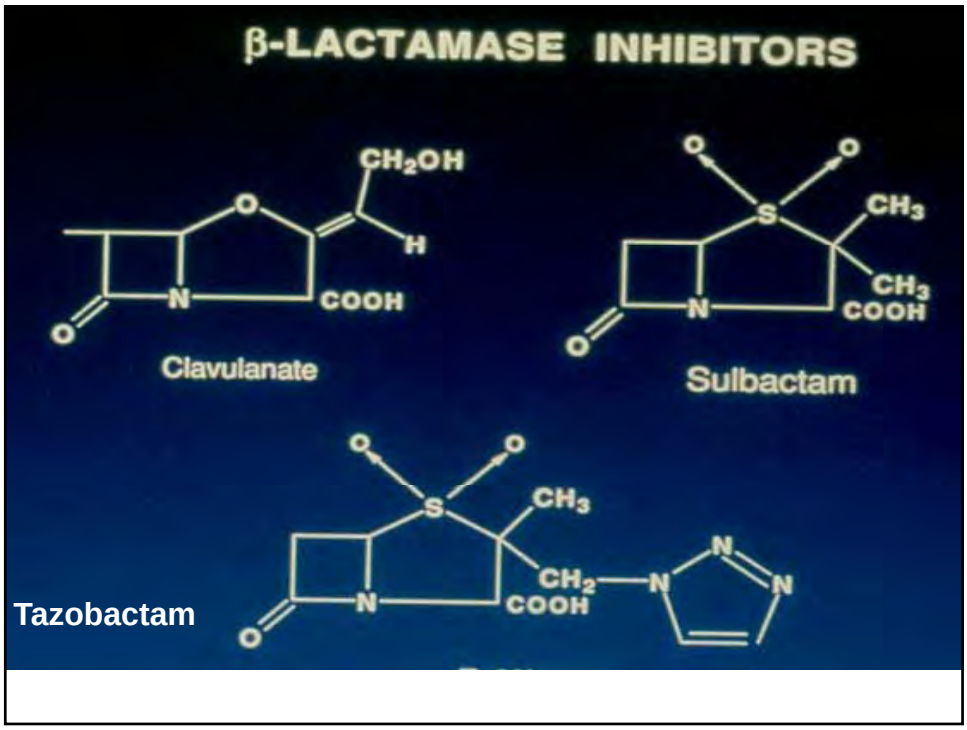
Spectrum - gram positive and gram negative -
Not inherently beta-lactamase stable
Spectrum - dependent upon permeability properties

Add a beta-lactamase inhibitor

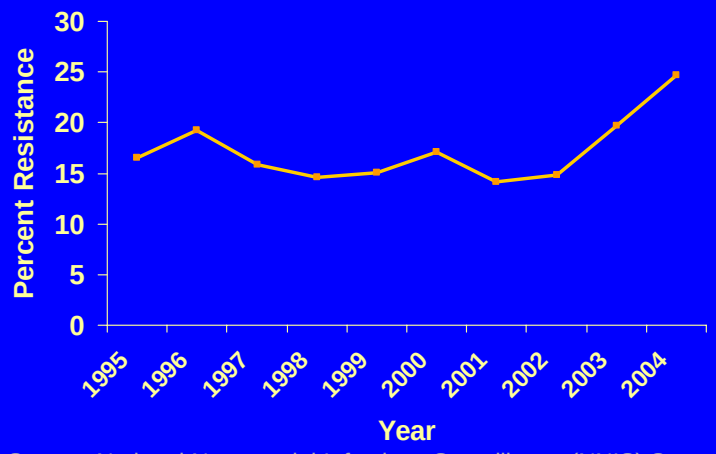
Clavulanic acid -
Sulbactam
Tazobactam

Expands spectrum of activity
- Anaerobes

NOT effective against the beta-lactamases of the SPACE organisms



3rd generation cephalosporin-resistant *Kliebsiella pneumoniae* Among ICU Patients,1995-2004



Source: National Nosocomial Infections Surveillance (NNIS) System

Pharmacology of the penicillins

Absorption - Amoxicillin - acid stable
dosing - give more - longer intervals
Augmentin - amox + clav - diarrhea

Metabolism - minor

Excretion - Renal - tubular secretion
Increase serum levels with probenecid
Biliary - only ureido penicillins
Nafcillin

Distribution - Anions - charged - extracellular space
CSF - with inflammation
Concentrated in urine