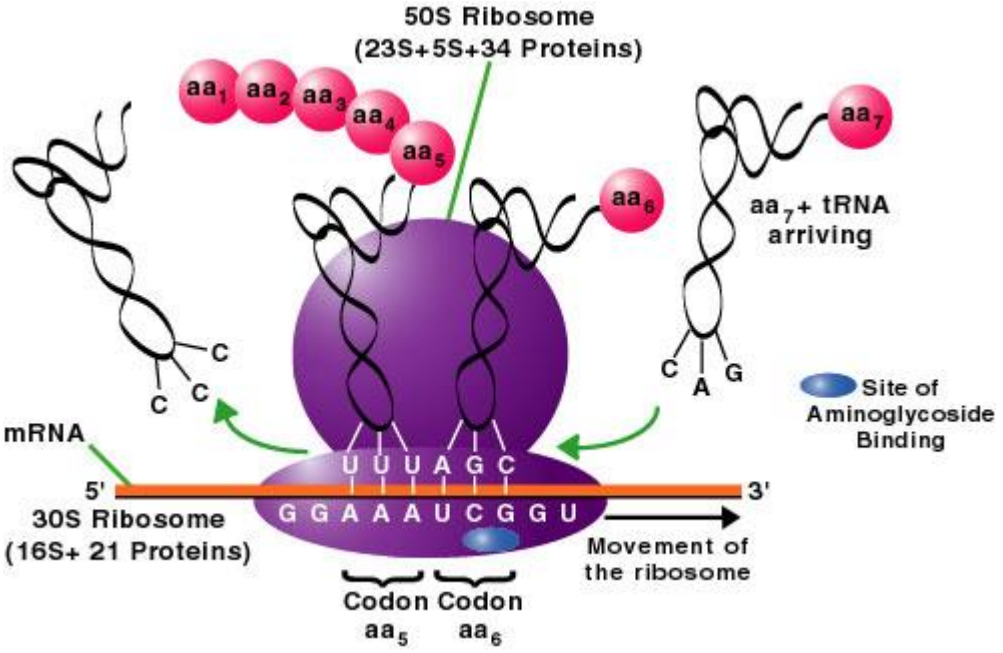
Chimica Farmaceutica e Tossicologica 2

- Ribosoma batterico;
- Amminoglicosidi;
- Macrolidi;
- Tetracicline;
- Streptogramine, Lincosammidi; Cloramfenicolo;
- Oxazolidinoni.

Class	Target	Mechanisms of high-level resistance that compromise therapy	
		Target related	Compound-chemistry related
β-lactams	Cell-wall synthesis: multiple penicillin-binding proteins (PBPs)	Horizontal transmission of resistant PBPs	Horizontal transmission of β-lactamase Upregulation of β-lactamase; permeability decrease, general efflux*
Vancomycin	Cell-wall synthesis: D-Ala-D-Ala of peptidoglycan substrate	Bypass pathway (VRE) Stepwise increase in wall thickness*	
Tetracyclines	Protein synthesis: 16S rRNA	Ribosome protection	Compound-specific efflux
Gentamicin	Protein synthesis: 16S rRNA		Inactivating enzymes
Macrolides	Protein synthesis: 23S rRNA	Ribosome protection Stepwise rRNA mutations* Low-frequency alterations in ribosomal proteins*	Compound-specific efflux
Lincosamides	Protein synthesis: 23S rRNA	Ribosome protection	
Chloramphenicol	Protein synthesis: 23S rRNA	Ribosome protection	Inactivating enzyme
Oxazolidinones	Protein synthesis: 23S rRNA	Stepwise rRNA mutations* Ribosome protection	
Fluoroquinolones	DNA replication: topoisomerases, gyrase and topo IV	Point mutations in both targets* Target protection	Compound-specific and general efflux*
Daptomycin	Bacterial membrane	Stepwise changes*	
Metronidazole	DNA alkylation		Loss of reductase
Nitrofurantoin†	DNA and protein alkylation		
Fosfomycin†	Cell-wall synthesis UDP-GlcNAc enolpyruvyl transferase	Inactivating enzymes	Loss of permease*

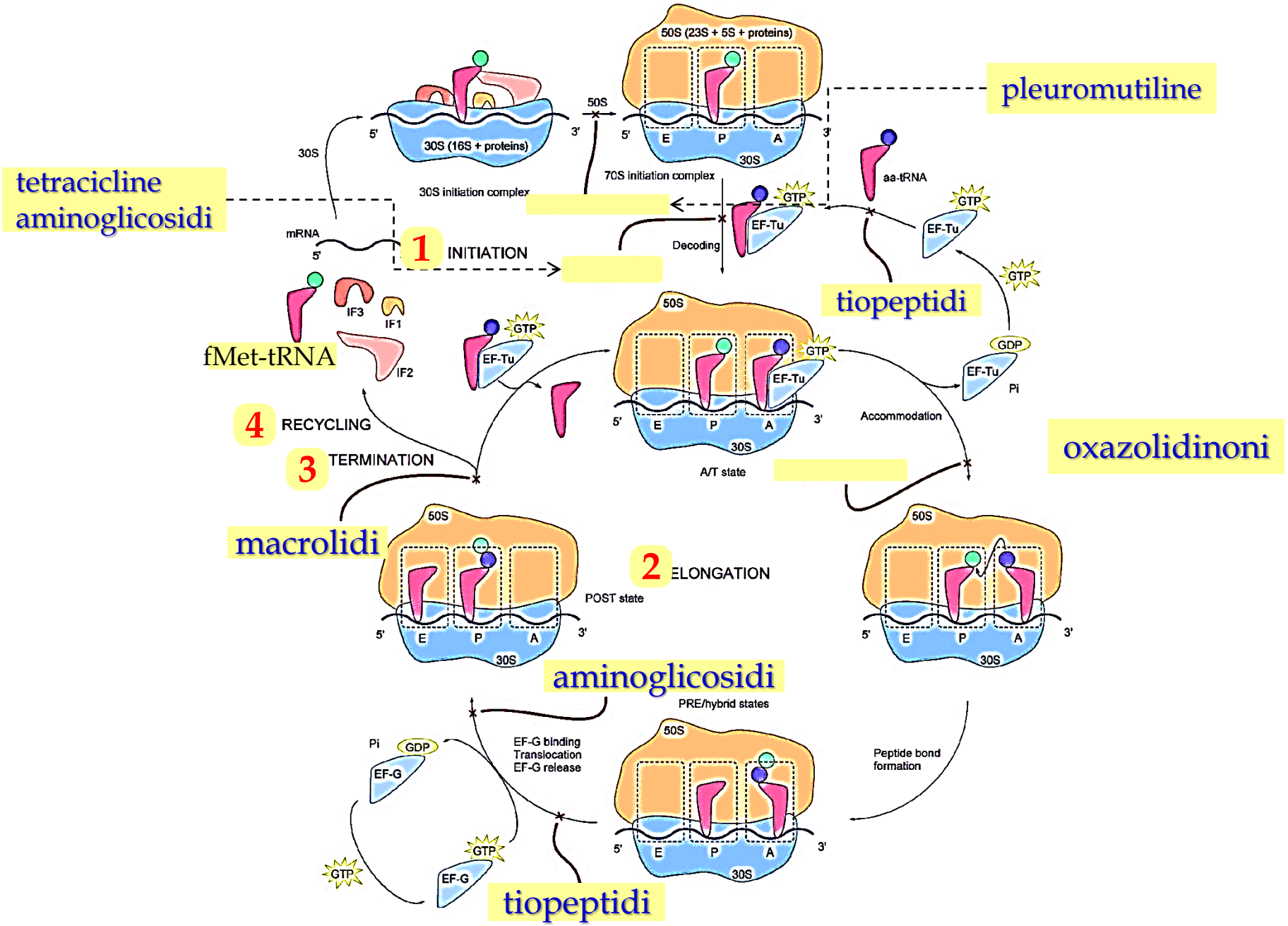
Antibiotics Targeting Ribosomes: Prof. Ada Yonath, Nobel Prize in Chemistry 2009

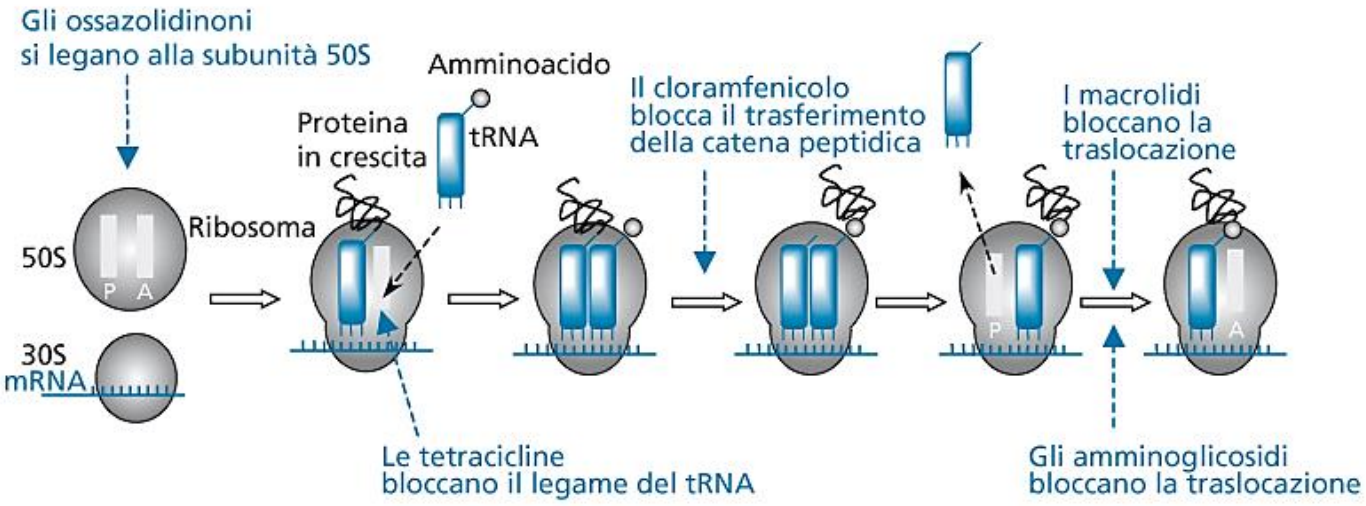
https://www.nobelprize.org/nobel_prizes/chemistry/laureates/2009/yonath-facts.html



2.7 ML Da
~ 55 proteine (6000-75000 D), domini globulari disposti sulla superficie ribosomiale.
Funzioni strutturali non ancora del tutto delucidate.

https://www.youtube.com/watch?v=Jml8CFBWcDs


pleuromutiline
tetracicline
aminoglicosidi
50S (23S + 5S + proteins)
50S
30S (16S + proteins)
30S
5'
3'
E
P
A
30S initiation complex
70S initiation complex
mRNA
1 INITIATION
Decoding
aa-tRNA
GTP
EF-Tu
tiopeptidi
IF3
IF1
IF2
fMet-tRNA
GDP
Pi
4 RECYCLING
3 TERMINATION
Accommodation
oxazolidinoni
A/T state
macrolidi
2 ELONGATION
POST state
aminoglicosidi
PRE/hybrid states
EF-G binding
Translocation
EF-G release
Peptide bond
formation
EF-G
tiopeptidi



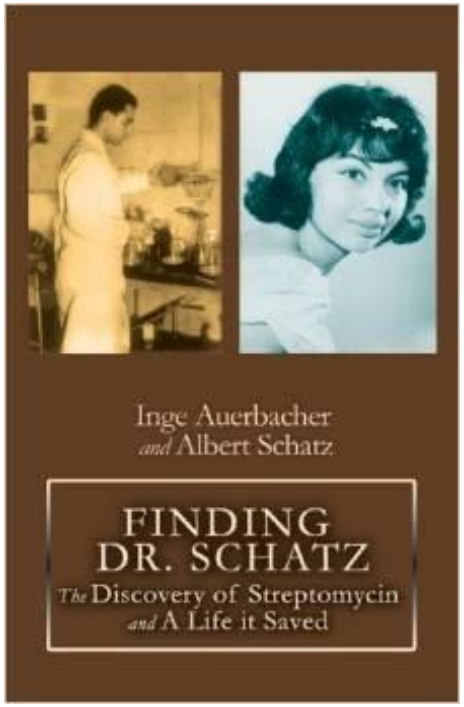
- Blocco del binding di *N*-formilmetionil-tRNA al ribosoma 70S
  - ossazolidinoni
- Alterazione trascrizione:
  - amminoglicosidi, paromomicina.
- Riduzione mobilità del ribosoma:
  - spectinomicina *(progressione mRNA).*
- Interferenza t-RNA sito di decodifica:
  - amminoglicosidi, paromomicina, tetracicline *(sito A subunità minore).*
- Interferenza con PTC (peptidyl-transferase center):
  - clindamicina, cloramfenicolo *(sito A subunità maggiore),*
  - streptogramina A.
- Blocco tunnel uscita:
  - macrolidi, streptogramina B

Antibiotici aminoglicosidici

Figure 1.1. The founders of the antibiotic era: Selman Waksman (discoverer of streptomycin) and Alexander Fleming (discoverer of penicillin).

The True Story of the Discovery of Streptomycin by Albert Schatz

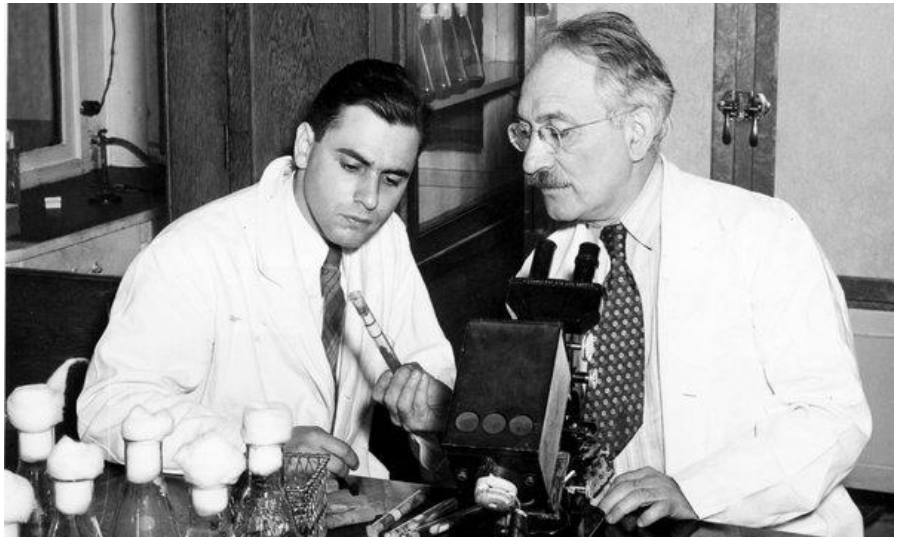
http://www.albertschatzphd.com/?cat=articles&subcat=streptomycin&itemnum=001



Albert Schatz

Selman Abraham Waksman (Nobel Medicina 1952)

(cfr Nobel penicillina 1945)



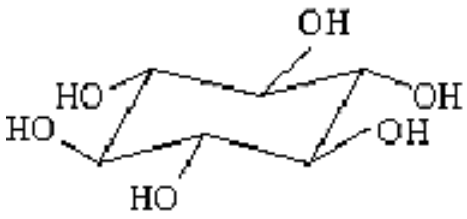
actinomycin, clavacin, streptothricin, streptomycin, grisein, neomycin, fradicin, candicidin, candidin

STREPTOMICINE (A, B, Diidro) *Streptomyces griseus*

primo antibiotici isolato da fonte batterica

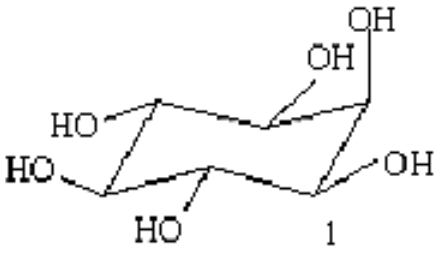
- **Spettro antibiotico**: attive su Gram - e micobatteri (TBC);
- Si sviluppano facilmente ceppi resistenti.
- **Meccanismo di azione**: Inibitori della sintesi proteica (traduzione); si legano alla subunità **30S** (binding dell'aa-t-RNA (sito A) nella porzione 16S della sub-unità 30S);
- mutazioni in proteine ribosomiali e rRNA, come pure metilazioni di specifiche basi dell'rRNA, conferiscono elevata resistenza agli AG:
- bloccano la sintesi proteica allo stadio di formazione del complesso di inizio; provocano una errata lettura dell'm-RNA (proteine non senso) e danni alla membrana cellulare
- **Tossicità**: soprattutto a carico del nervo acustico (ototossiche), con disturbi vestibolari (streptomicina) e sordità (diidrostreptomicina).
- **Farmacocinetica**: l'elevata idrofilia impedisce alle streptomicine di venire efficacemente assorbite dal tratto GI; se somministrate per os vengono recuperate intatte nelle feci.

Inositoli (ciclitoli, cicloesanesoli)



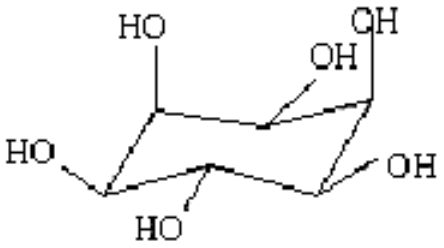
scyllo-Inositol (1)

(meso)



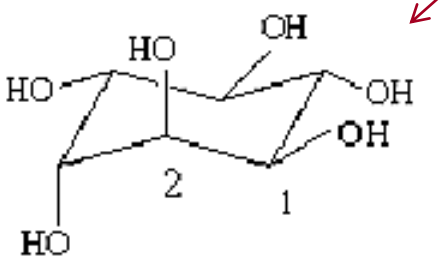
myo-Inositol (2)

(meso, B7)

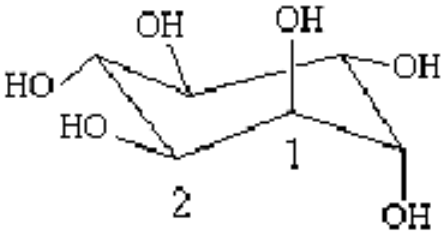


epi-Inositol (3)

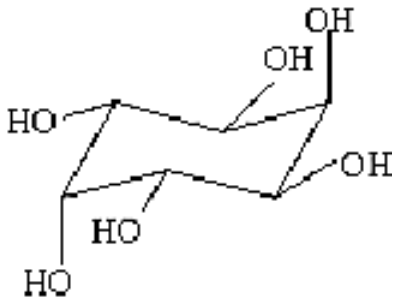
(meso)



D-*chiro*-Inositol (4)

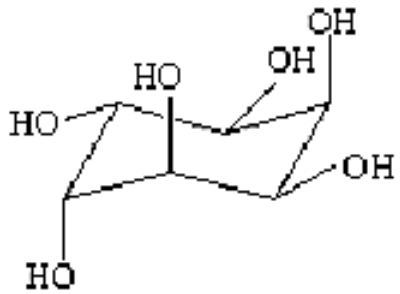


L-*chiro*-Inositol (5)

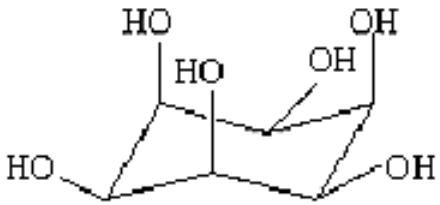


neo-Inositol (6)

(meso)

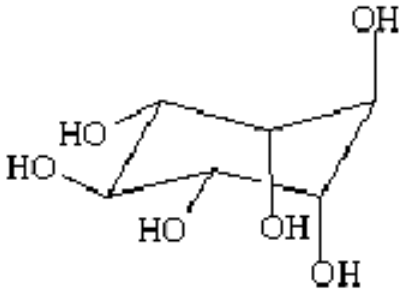


(chirale) *allo*-Inositol (7)



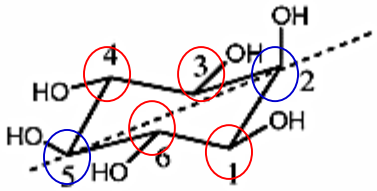
cis-Inositol (8)

(meso)



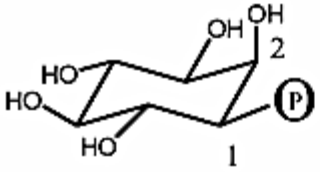
muco-Inositol (9)

prochirale



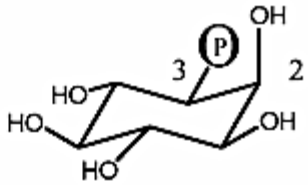
meso

myo-Inositol (13) ----- , plane of symmetry



1D-*myo*-inositol-1-monophosphate

(14)



1D-*myo*-inositol-3-monophosphate

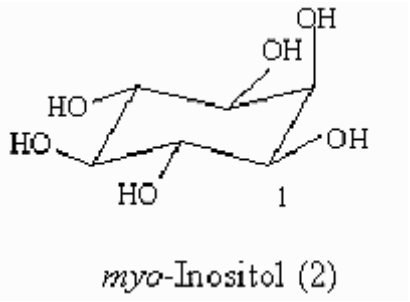
(15)

Ⓟ = Phosphate

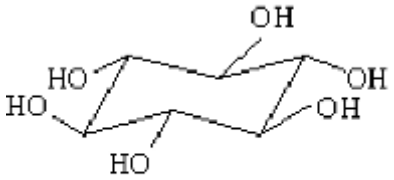
Figure 3. *myo*-Inositol and phosphorylated derivatives.

Pseudo-oligosaccaridi

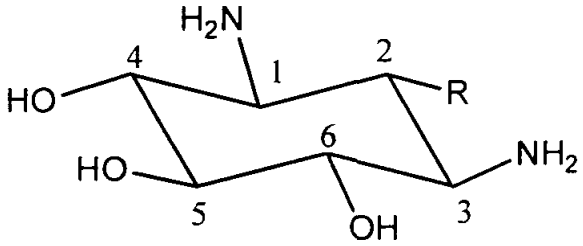
Farmacoforo derivato dall'1,3-diamminoscillitolo (streptamina/streptidina, 2-desossistreptamina, spectinamina), con le funzioni alcoliche legate, mediante legami glucosidici, ad amminozuccheri caratteristici



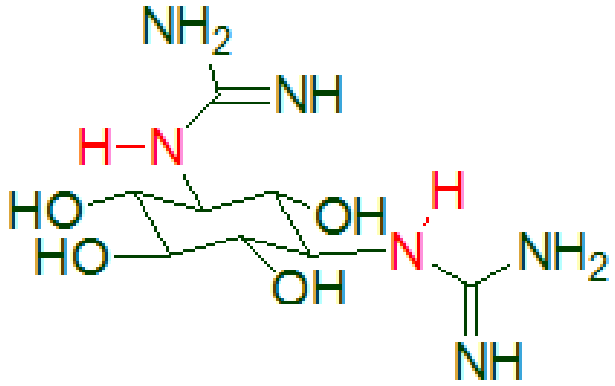
myo-Inositol (2)



scyllo-Inositol (1)



Streptamina



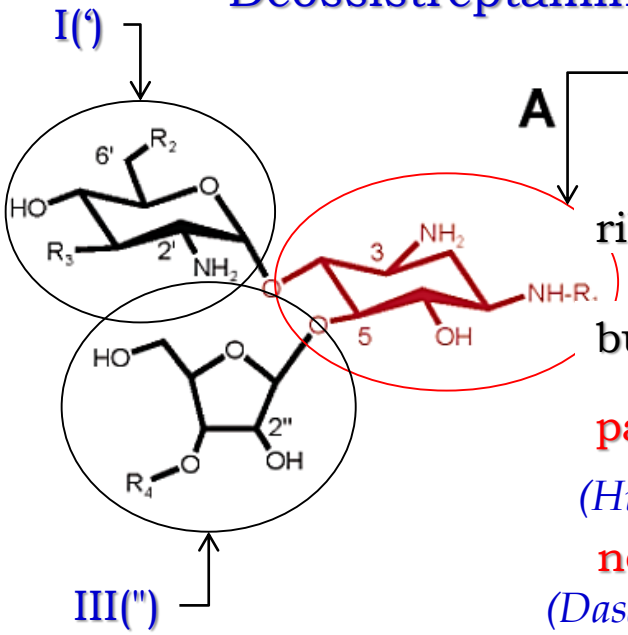
Streptidina

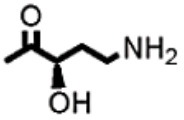
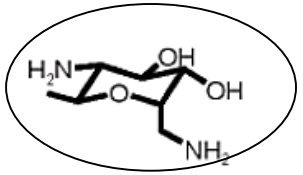
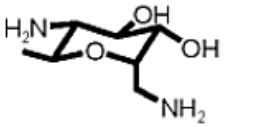
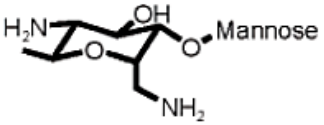
Gli aminoglicosidi letteralmente comprendono amminociclitoli ed uno zucchero a formare molecole di origine naturale prodotte generalmente da batteri del genere *Streptomyces* e *Micromonospora*.

Gli aminoglicosidi prodotti da *Micromonospora* seguono una nomenclatura che assegna il suffisso -micina (gentamicina) mentre quelli prodotti da Streptomyces hanno suffisso -mycina (streptomicina, neomicina, kanamicina, spectinomicina, paromomicina.

aminoglicosidi deossistreptamina 4,5-disostituiti

Deossistreptamina (aminociclitolo centrale II)

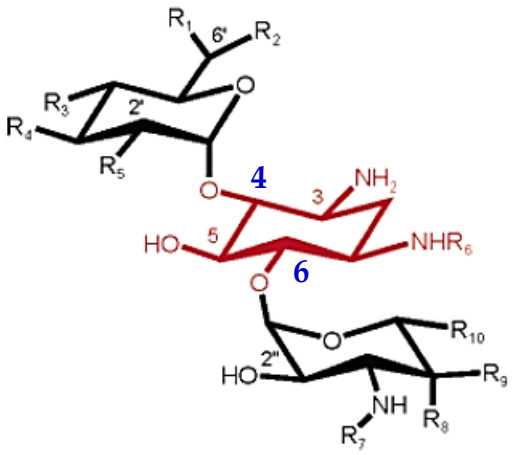


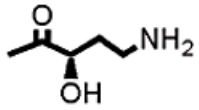
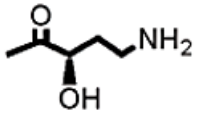
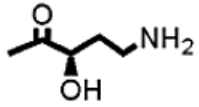
Antibiotic	R_1	R_2	R_3	R_4
ribostamicina	H	NH_2	OH	H
butirosina		NH_2	OH	H
paromomicina *(Humatin, Kaman)*	H	OH	OH	
neomicina B *(Dasamix[1] neomicina, Streptosil[2] nemocina)*	H	NH_2	OH	
lividomicina A	H	OH	H	

IV('")

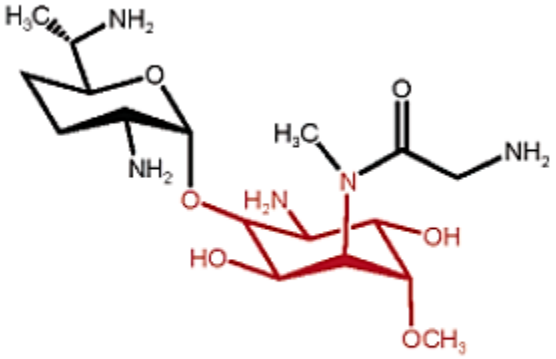
([1] +desametasone, [2] +sulfatiazolo, +alcinolide, bacitracina, beclometasone, costebol, flumetasone, fluocinolone, idrocortisone,)

aminoglicosidi deossistreptamina 4,6-disostituiti

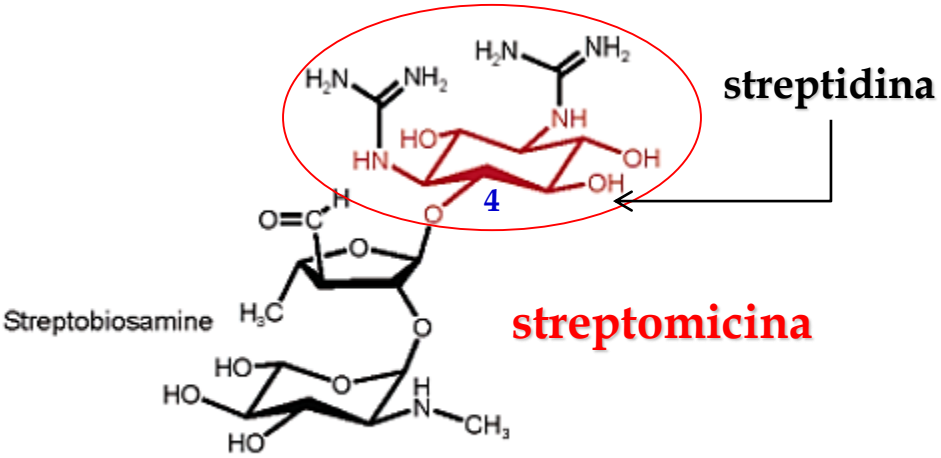


Antibiotic	R_1	R_2	R_3	R_4	R_5	R_6	R_7	R_8	R_9	R_{10}
Kanamycin A	H	NH_2	OH	OH	OH	H	H	H	OH	CH_2OH
Kanamycin B	H	NH_2	OH	OH	NH_2	H	H	H	OH	CH_2OH
Tobramycin	H	NH_2	OH	H	NH_2	H	H	H	OH	CH_2OH
Dibekacin	H	NH_2	H	H	NH_2	H	H	H	OH	CH_2OH
Gentamicin B	H	NH_2	OH	OH	OH	H	CH_3	OH	CH_3	H
Gentmicin C1	CH_3	$NHCH_3$	H	H	NH_2	H	CH_3	OH	CH_3	H
Gentmicin C1A	H	NH_2	H	H	NH_2	H	CH_3	OH	CH_3	H
Gentmicin C2	CH_3	NH_2	H	H	NH_2	H	CH_3	OH	CH_3	H
Sisomicin*	H	NH_2	H	H	NH_2	H	CH_3	OH	CH_3	H
Netilimicin*	H	NH_2	H	H	NH_2	CH_2CH_3	CH_3	OH	CH_3	H
Isepamicin	H	NH_2	OH	OH	OH		CH_3	OH	CH_3	H
Arbekacin	H	NH_2	H	H	NH_2		H	H	OH	CH_2OH
Amikacin	H	NH_2	OH	OH	OH		H	H	OH	CH_2OH

aminoglicosidi atipici

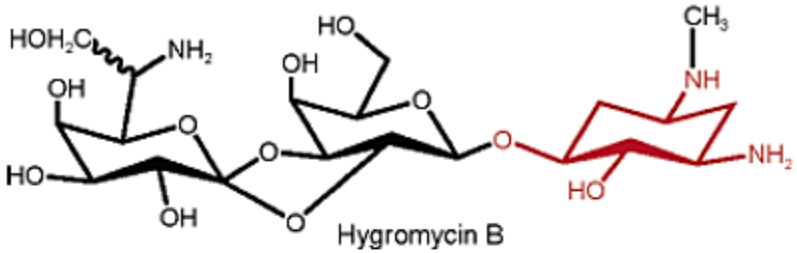


Fortimicin A

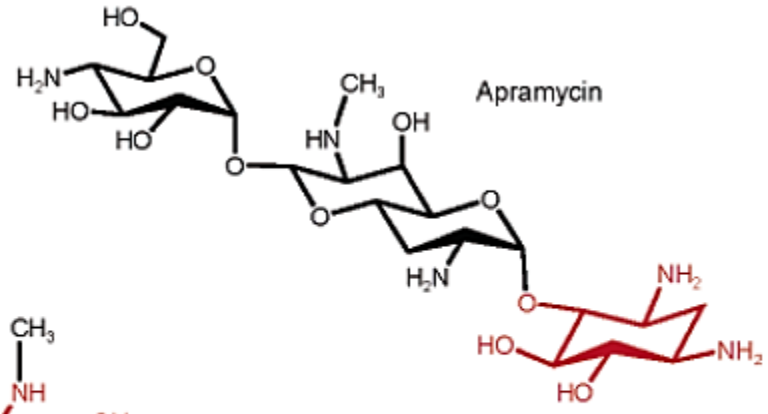


Streptobiosamine

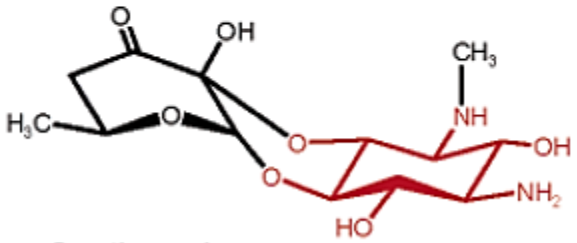
streptomicina



Hygromycin B



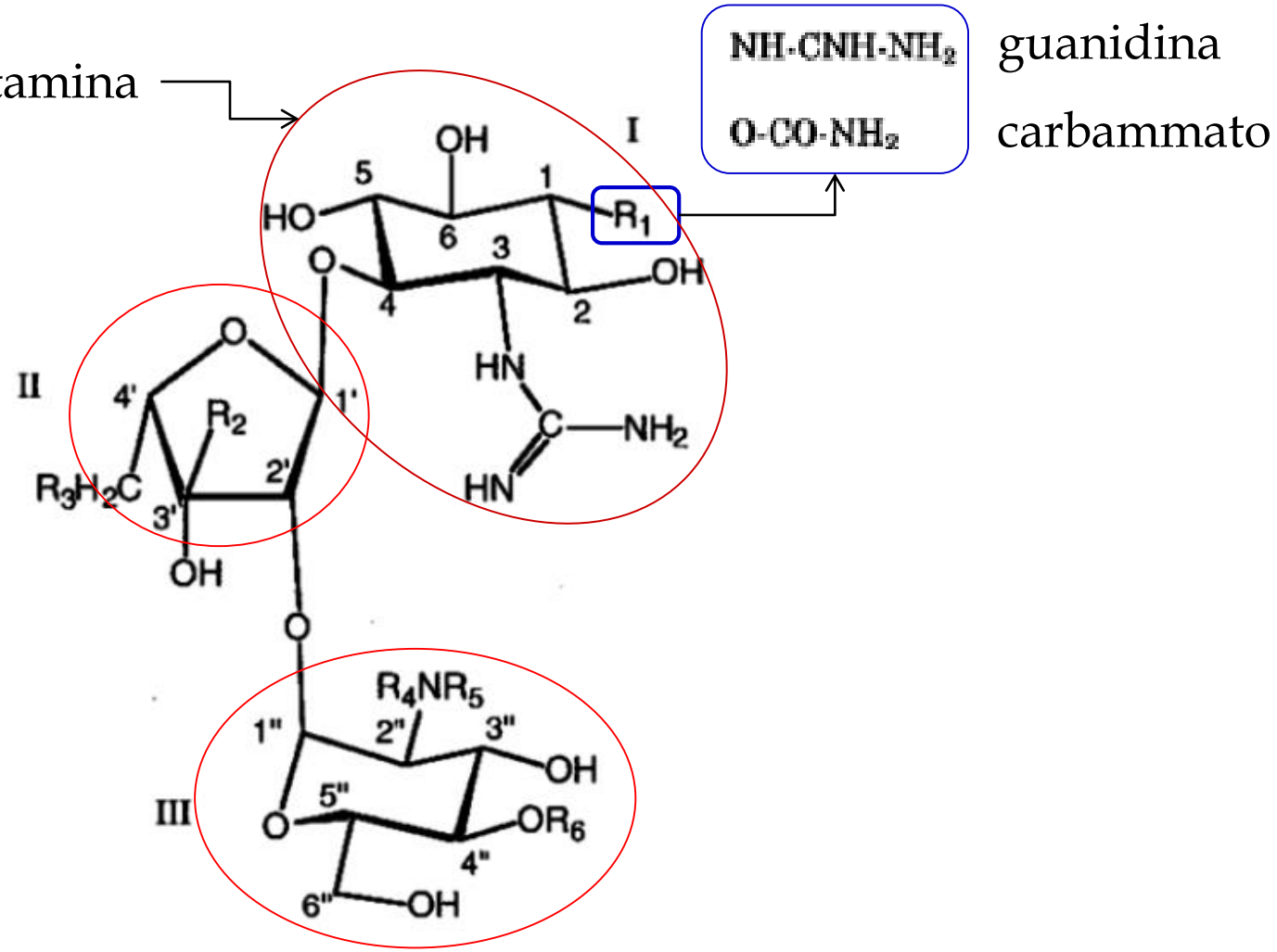
Apramycin



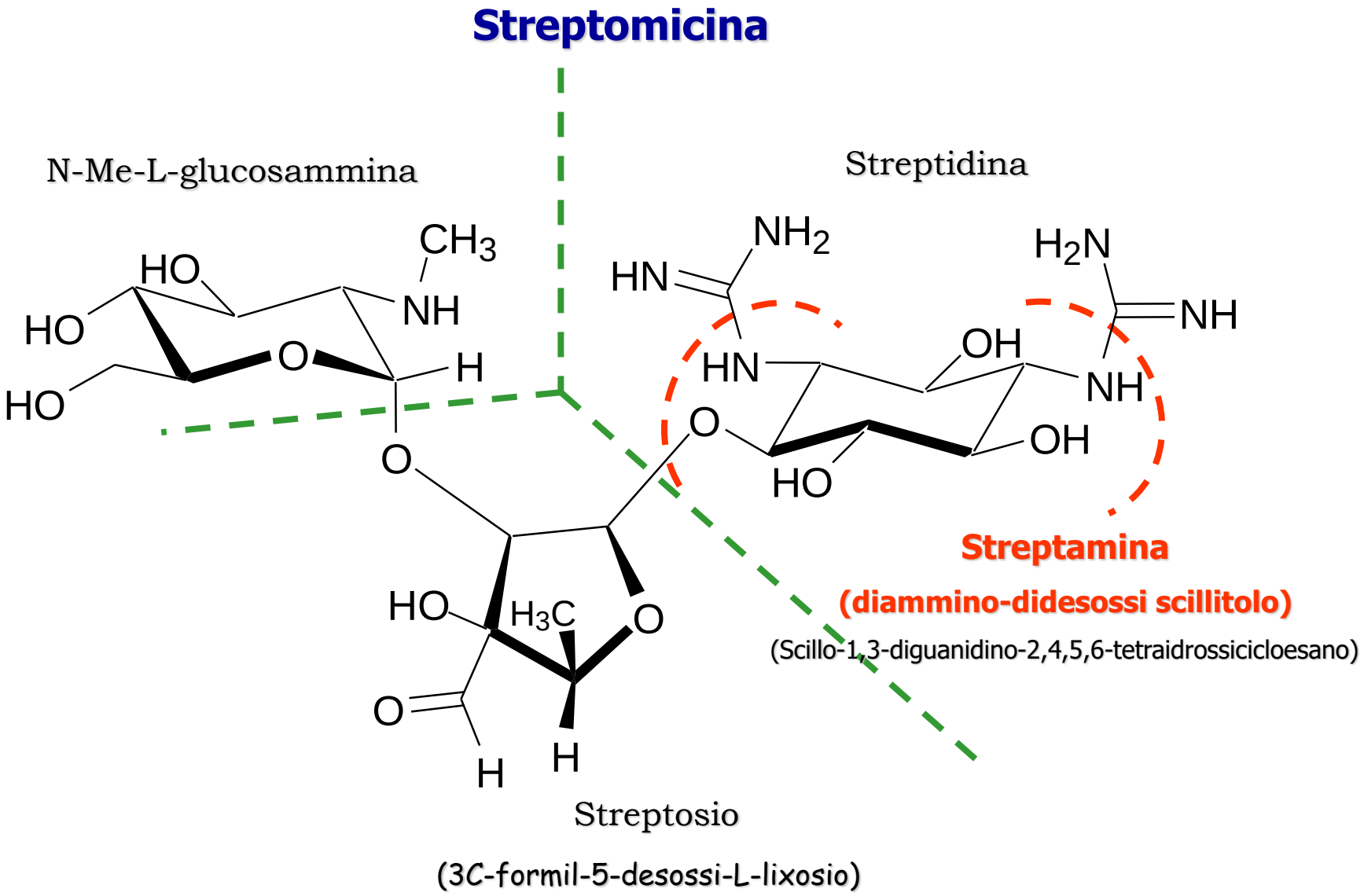
Spectinomycin

Streptomicina

ciclitolo-3-streptamina



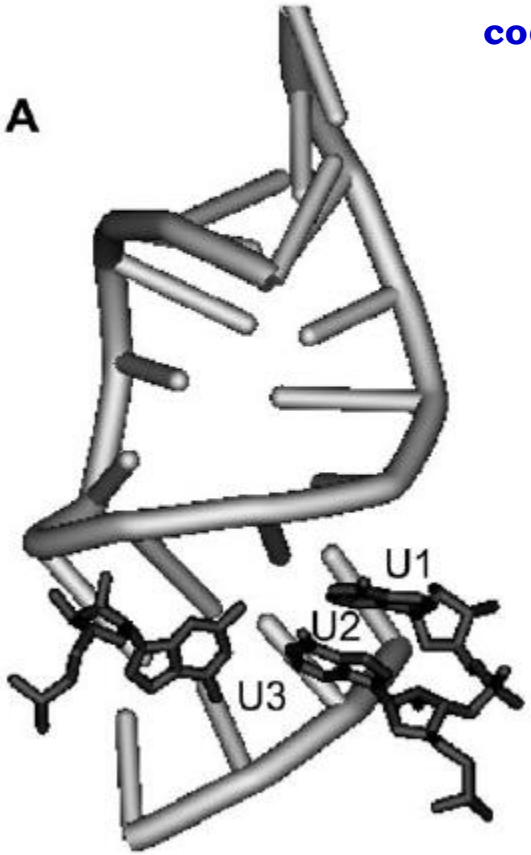
Biogenesi da glicidi

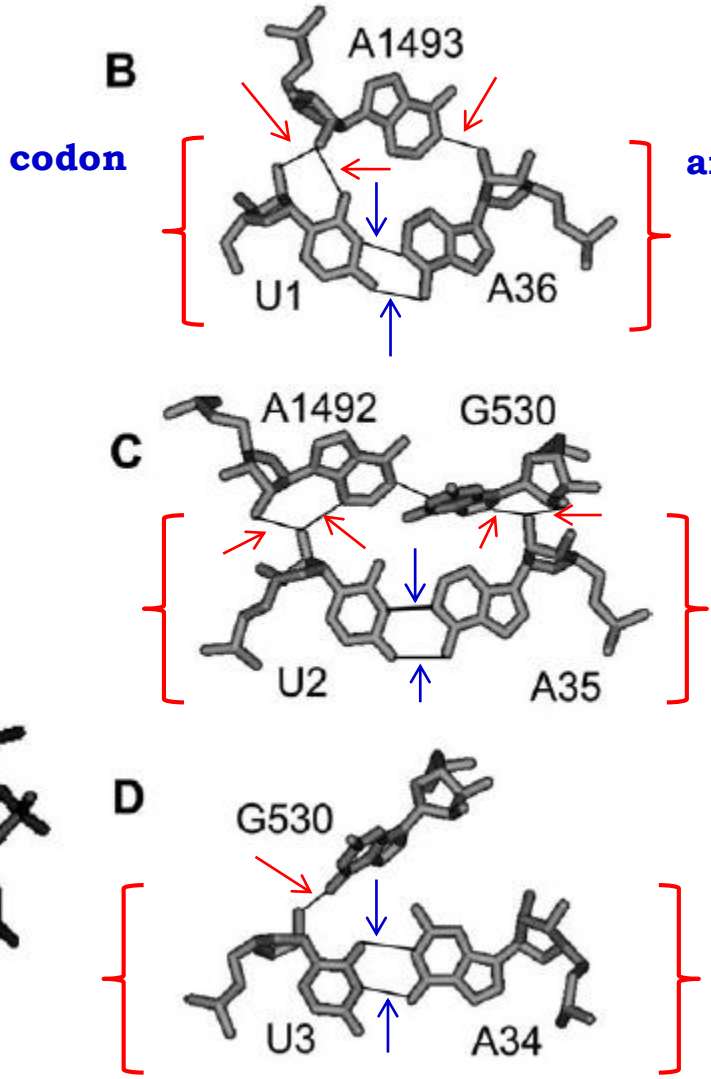


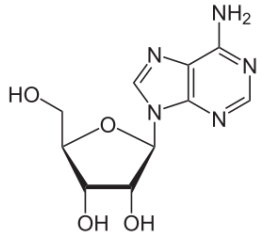
1948. blocco enzimatico in batteri sensibili;
1950-1960. studi genetici e biochimici identificano la sintesi proteica quale target primario dell'azione antibatterica della **S**. (Erdos e Ullmann, aminoacidi radioattivi la cui incorporazione è bloccata in *Mycobacterium tuberculosis* da **S**).
1961. Spotts e Stanier propongono il ribosoma quale target della **S**.
1962–1965. L'ipotesi di Spotts e Stainer è confermata da molti studi (binding della **S.** alla subunità 30S) con induzione di errori nella traduzione. Il ribosoma non appare più come supporto inerte nella formazione di peptidi ma sito di selezione tRNA aminoacilati da parte di mRNA ribosomiale.
Oggi I complessi 3D ribosomiali hanno ampiamente confermato il ruolo dinamico dei ribosomi nel meccanismo traslazionale che è perturbato dagli aminoglicosidi alla subunità 30S (*mistranslation*).
La capacità degli aminoglicosidi di causare *mistranslation* è stata recentemente applicata a una forma di terapia genica indiretta (gentamicina in emofilia o fibrosi cistica severa).
Gli aminoglicosidi, come altri antibiotici, inducono a concentrazioni sub-MIC un'alterazione transcrizionale nel 5% dei geni (batteri sensibili) e possono agire come elementi di trasduzione di segnali cellulari.
Diversamente dalla maggior parte degli antibiotici inibitori della sintesi proteica, gli aminoglicosidi, sono rapidamente battericidi (modello di Davis).

Codon recognition by tRNA

A1492, A1493 and G530 of 16S rRNA,





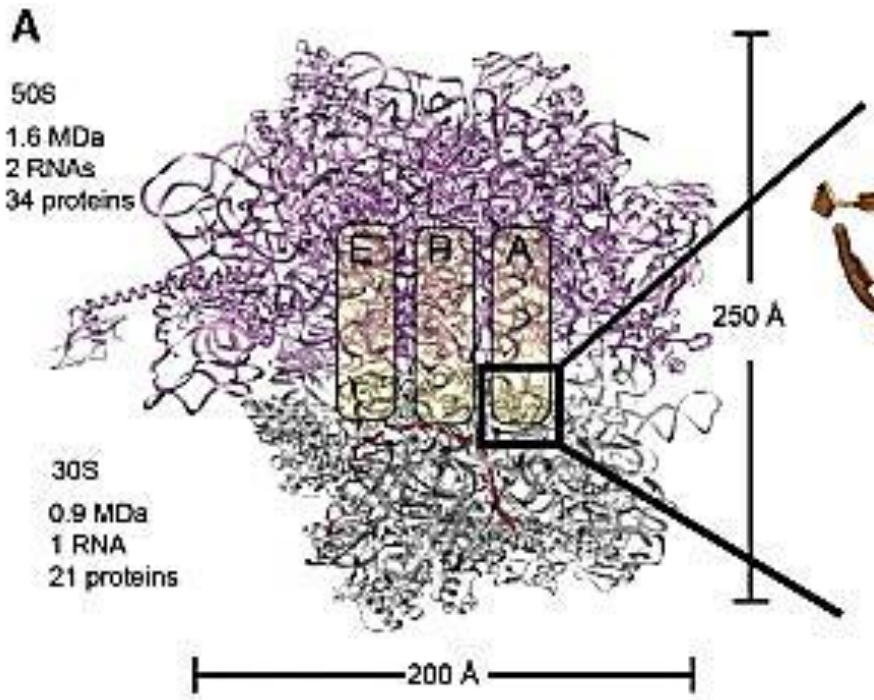


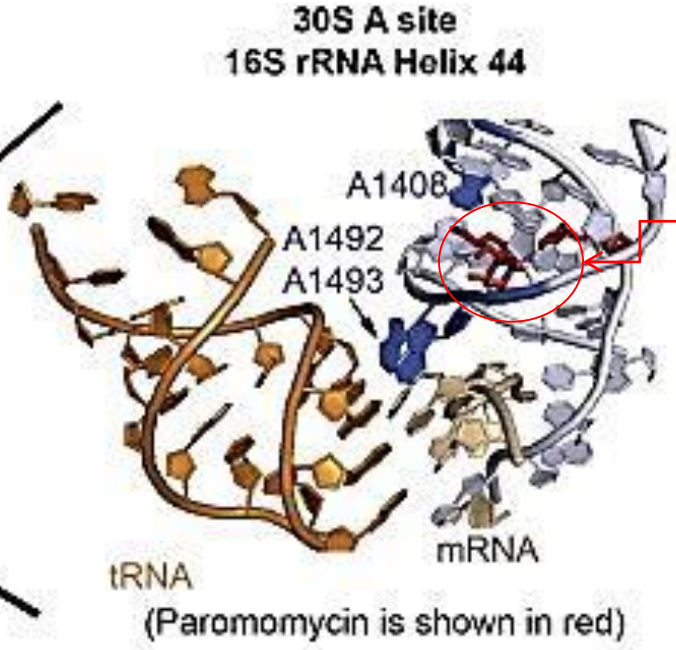
The first base pair of the codon-anticodon helix

The second base pair of the codon-anticodon helix

The third base pair of the codon-anticodon helix

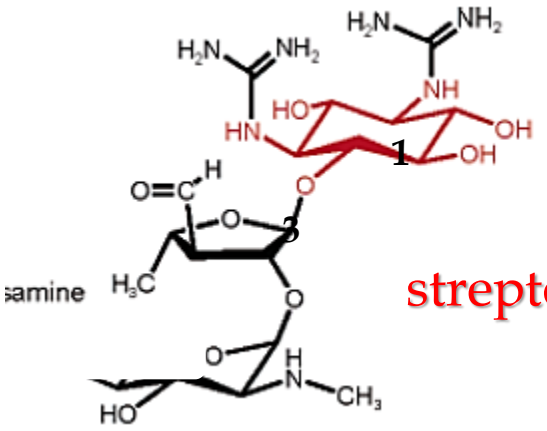
Mikolaj Olejniczak, BIOTECHNOLOGIA 1 (84) 59-64 2009.

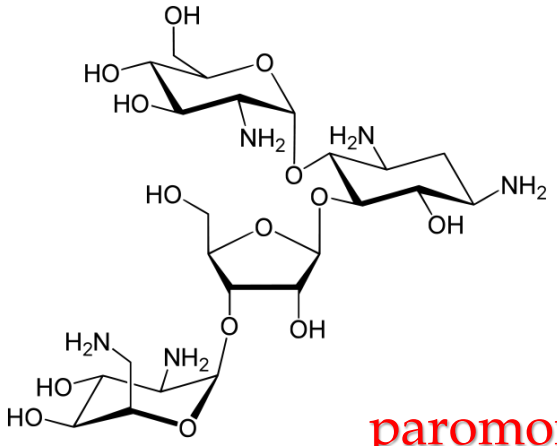




paromomicina

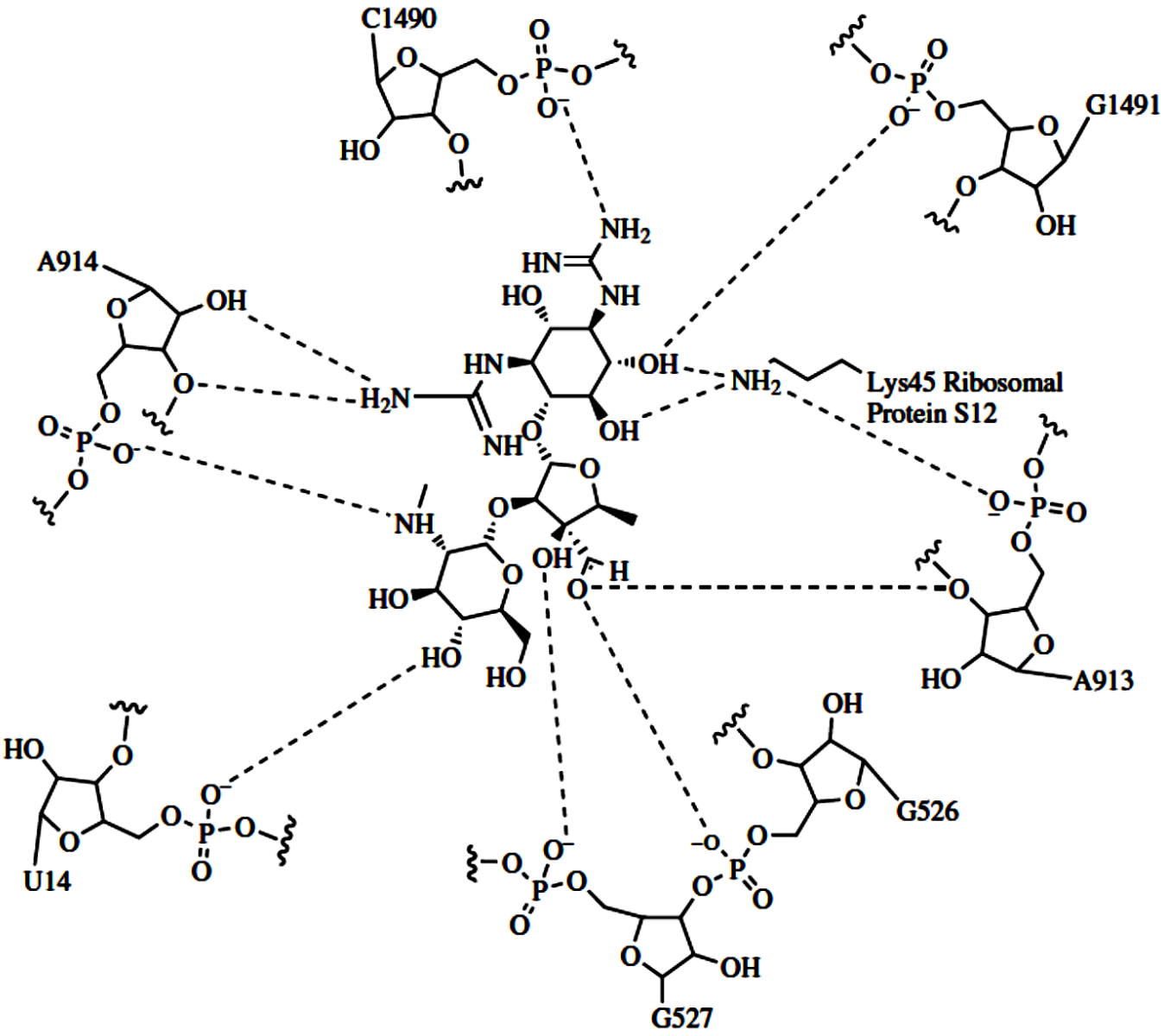
codon



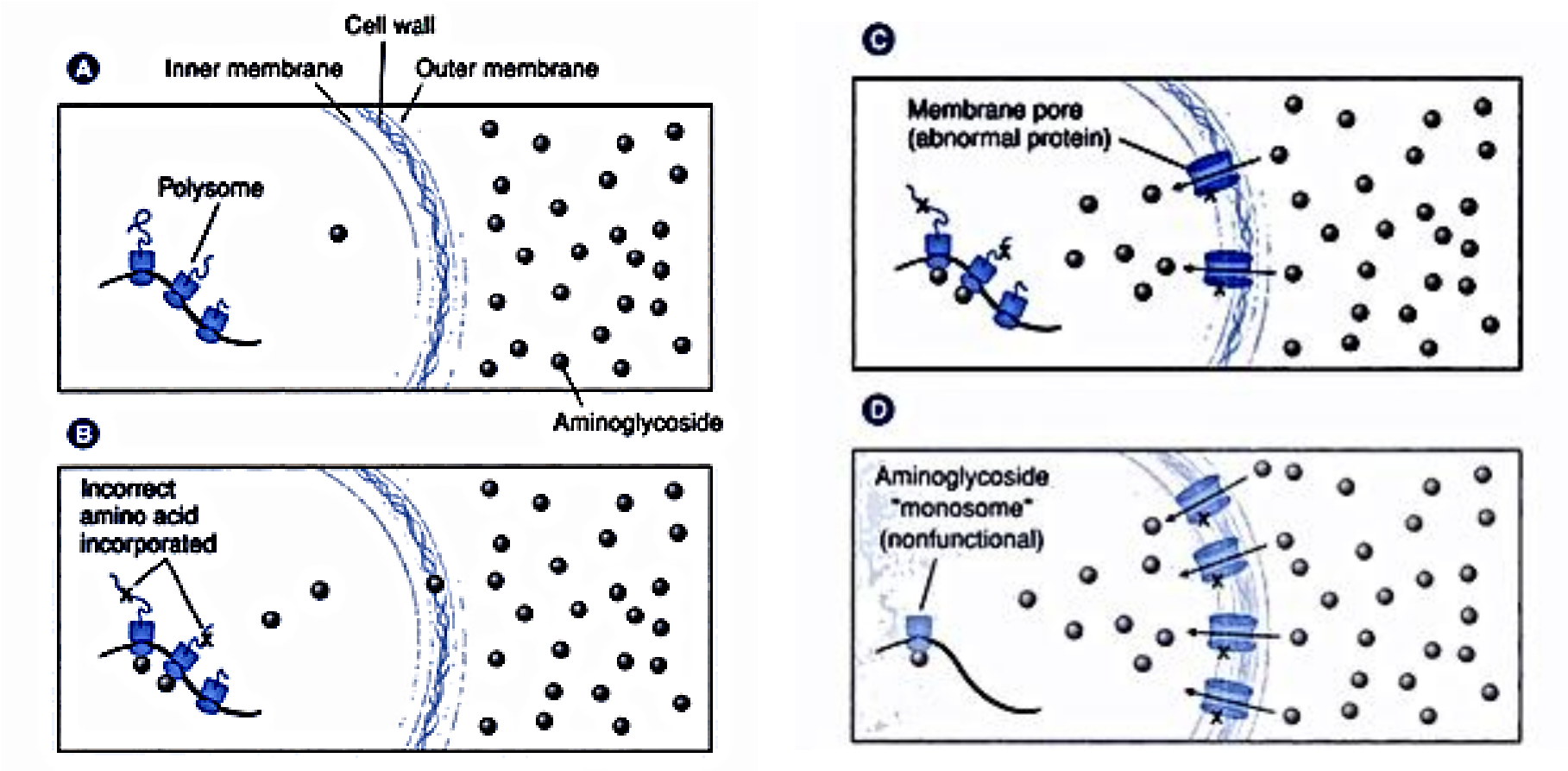


il solo aminoglicoside che interagisce anche con proteine ribosomiali (WHO Essential Medicines)

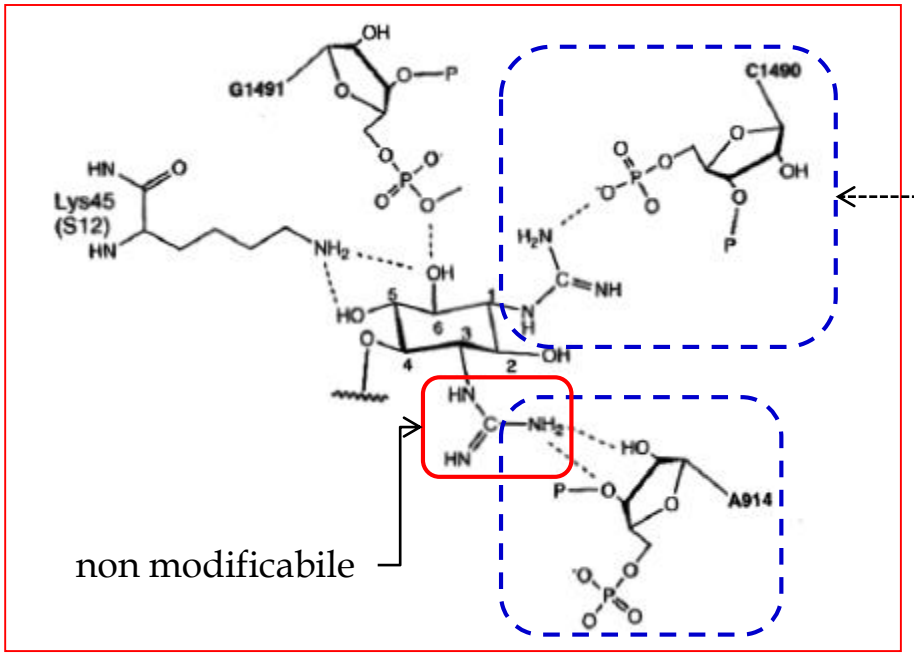
Interazioni di aminoglicosidi con sito accettore alla subunità 30S del ribosoma.
Streptomicina vs 16S rRNA e la proteina ribosomale S12



Ipotesi di Davis (Mechanism of Bactericidal Action of Aminoglycosides B.D. DAVIS. MICROBIOLOGICAL REVIEWS. Sept. 1987, p. 341-350)



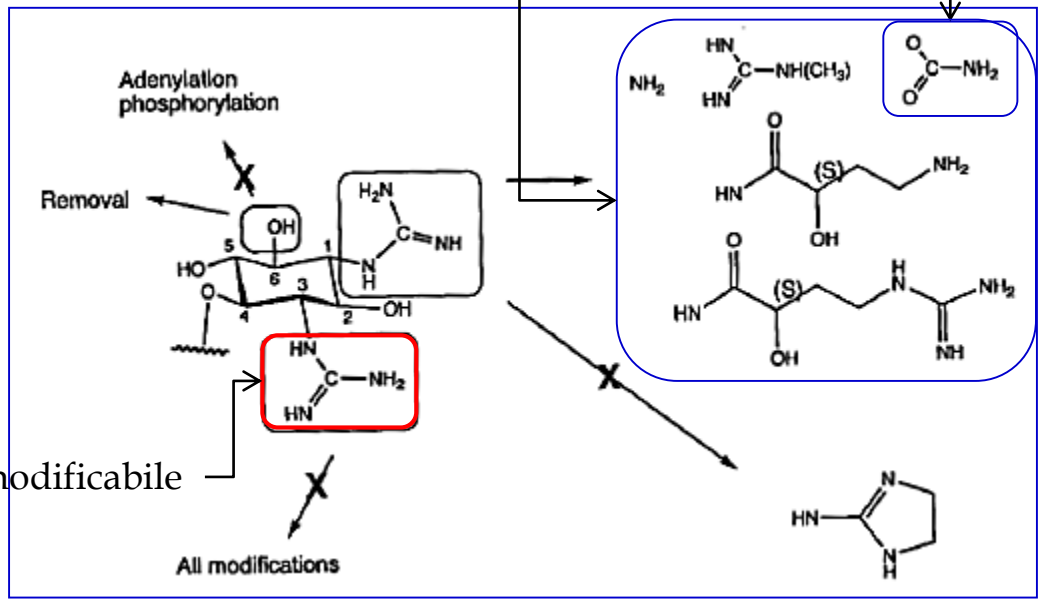
Principles of Pharmacology: The Pathophysiologic Basis of Drug Therapy.
David E. Golan. Wolters Kluwer Health 2008



binding mode

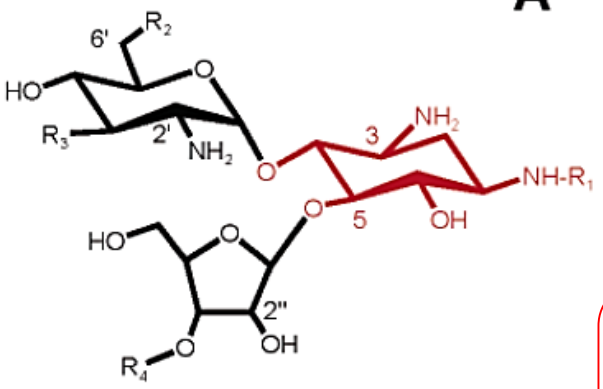
Attività generalmente inferiore vs streptomicina

bluensomicina (carbammato)

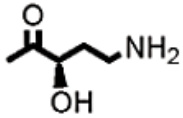
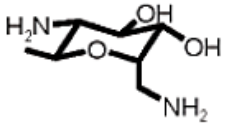
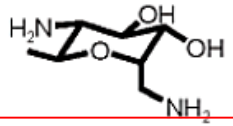
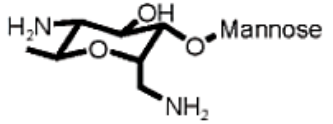


SAR (I)

Neomicine (*Streptomyces fradiae*), **Paromomicine** *Streptomyces rimosus* e *Streptomyces krestomyceticus* *(Humatin, Kaman)*

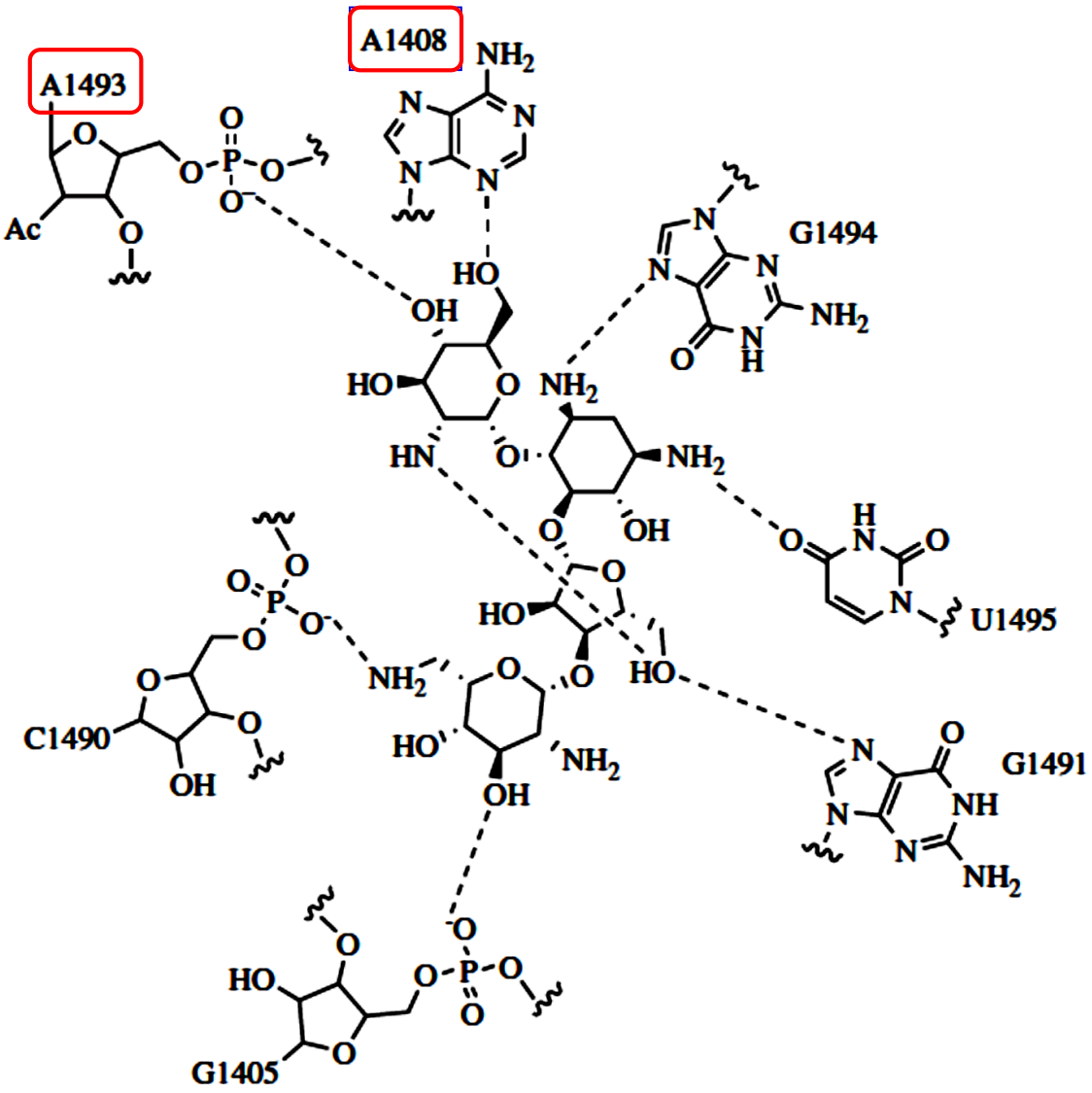


aminoglicosidi
deossistreptamina
4,5-disostituiti

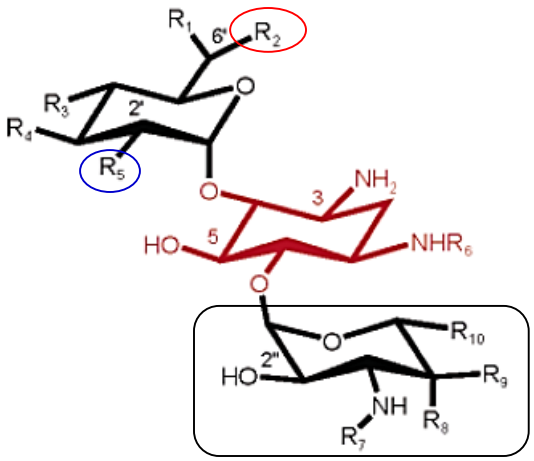
Antibiotic	R_1	R_2	R_3	R_4
Ribostamycin	H	NH_2	OH	H
Butirosin		NH_2	OH	H
paromomicina	H	OH	OH	
neomicina B	H	NH_2	OH	
Lividomycin A	H	OH	H	

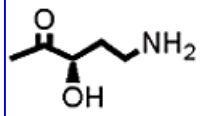
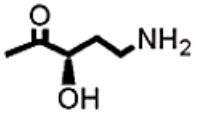
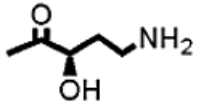
Attive contro Gram +, Gram - e micobatteri; Infezioni del tratto intestinale da germi sensibili (da *E. coli*, *Shigelle*, *Salmonelle*, escluso *S. Typhi*, ecc.). Sterilizzazione del contenuto intestinale nella preparazione ad interventi sull'intestino. Amebiasi intestinale (acuta e cronica). Nefrotossiche ed epatotossiche per via parenterale

Interazioni di aminoglicosidi con sito accettore alla subunità 30S del ribosoma.
Paromomicina vs 16S rRNA e la proteina ribosomale S12.



aminoglicosidi deossistreptamina 4,6-disostituiti



Antibiotic	R_1	R_2	R_3	R_4	R_5	R_6	R_7	R_8	R_9	R_{10}
Kanamycin A	H	NH_2	OH	OH	OH	H	H	H	OH	CH_2OH
Kanamycin B	H	NH_2	OH	OH	NH_2	H	H	H	OH	CH_2OH
Tobramycin	H	NH_2	OH	H	NH_2	H	H	H	OH	CH_2OH
Dibekacin	H	NH_2	H	H	NH_2	H	H	H	OH	CH_2OH
Gentamicin B	H	NH_2	OH	OH	OH	H	CH_3	OH	CH_3	H
Gentmicin C1	CH_3	$NHCH_3$	H	H	NH_2	H	CH_3	OH	CH_3	H
Gentmicin C1A	H	NH_2	H	H	NH_2	H	CH_3	OH	CH_3	H
Gentmicin C2	CH_3	NH_2	H	H	NH_2	H	CH_3	OH	CH_3	H
Sisomicin*	H	NH_2	H	H	NH_2	H	CH_3	OH	CH_3	H
Netilimicin*	H	NH_2	H	H	NH_2	CH_2CH_3	CH_3	OH	CH_3	H
Isepamicin	H	NH_2	OH	OH	OH		CH_3	OH	CH_3	H
Arbekacin	H	NH_2	H	H	NH_2		H	H	OH	CH_2OH
Amikacin	H	NH_2	OH	OH	OH		H	H	OH	CH_2OH

Kanamicine
(*Streptomyces kanamiceticus*)
Gentamicine
(*Micromonospora purpurea*)
Tobramicina
(*Streptomyces tenebrarius*)
Amikacina
(Amikan, Likacin, Migracin..)
Netilmicina
(Nettacin coll, Zetamicin im)

infezioni pleuro-polmonari, renali genito-urinarie, chirurgiche, ostetrico- ginecologiche, gastro-enteriche e biliari

Gentamicine *(Gentalyn, Nemalin, Ribomicin, Tacigen)* e Kanamicine:
Affezioni flogistiche della cute o del derma. Dermatiti varie accompagnate o con immediato rischio da sovrainfezioni

RESISTENZA AGLI AMMINOGLICOSIDI

Non enzimatica

(esclusivamente intrinseca!)

Con poche eccezioni (es. resistenza alla streptomicina nella TBC) non è clinicamente importante

L'ingresso di AG in cellula batterica richiede una catena di trasporto elettronico intatta. Batteri anaerobi (tetano, gangrena, enterocolite, ecc.) sono intrinsecamente resistenti agli AG.

Enzimatica

(generalmente, ma non esclusivamente, acquisita!)

- **Modificazione ribosomiale**
- **Modificazione AG**
- **Pompe efflusso**

Clinicamente importante!

RESISTENZA ENZIMATICA

Modificazione ribosomiale

Metilazione di basi specifiche rRNA (N7 di Guanina) nella sub-unità 16S produce elevata resistenza

Modificazione AG

- *O*-nucleotidil transfer (adenilazioni)
- *O*-fosforil transfer (fosforilazioni)
- *N*-acetil transfer (acetilazioni)

Gli AG modificati hanno ridotta affinità per i siti di binding nel ribosoma batterico

Enzimi inattivanti

(localizzazione intracellulare)

Classe

AAD = **A**g **AD**enil transferasi

ANT	**A**mminoglicoside **N**ucleotidil **T**ransferasi
APH	**A**mminoglicoside **PH**ospho transferasi
AAC	**A**mminoglicoside **AC**etil transferasi

Il numero in parentesi indica la **regiospecificità** del sito

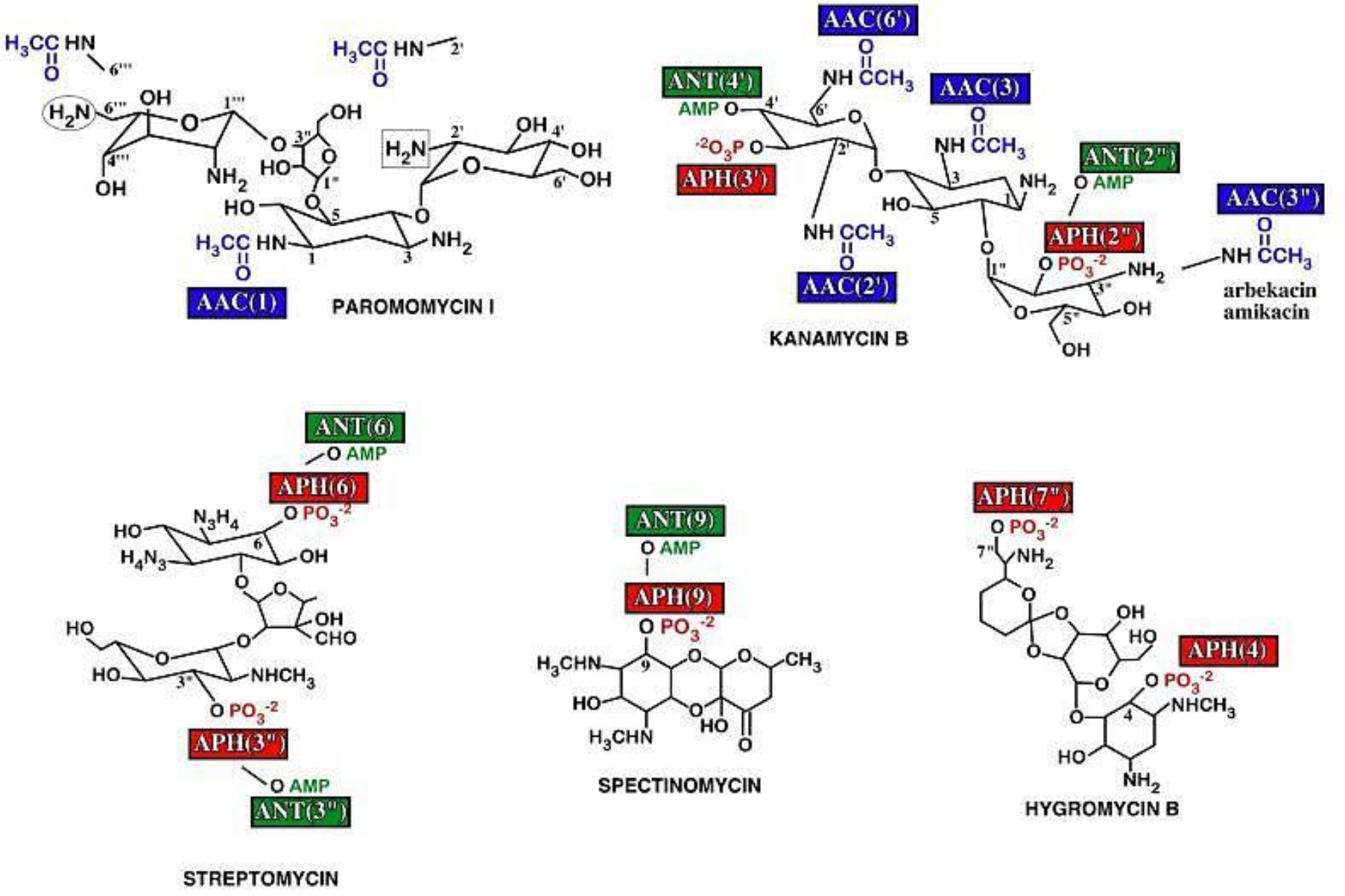
Es. **ANT(2″)**

Un numero romano indica l'unicità del **profilo di resistenza**

Es. **ANT(2″)-Ia**

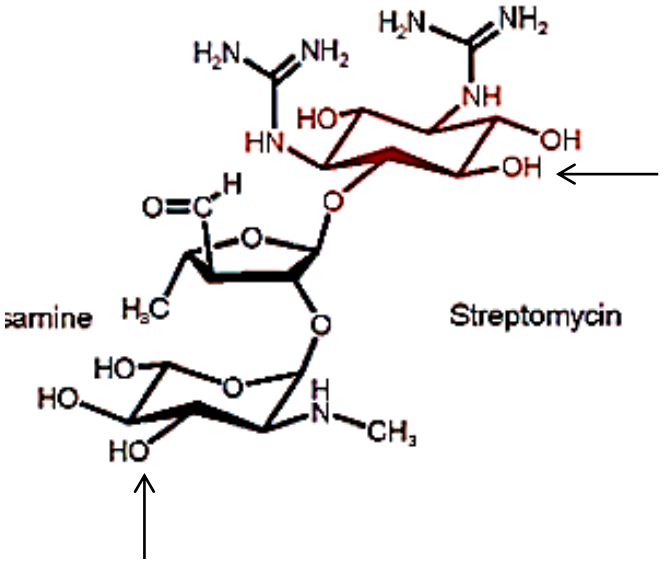
I **geni** che conferiscono resistenza vengono designati con lettere minuscole

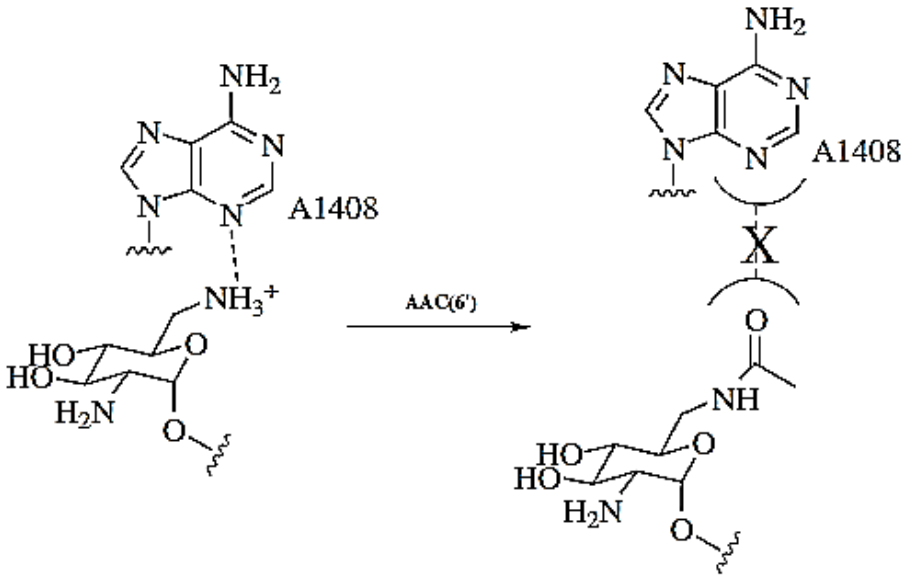
Es. **ant(6)-Ia** codifica una streptomicina NT che modifica la posizione 6 del farmaco



Aminoglycoside Modifying Enzymes. Drug Resist Updat. 2010 Dec; 13(6): 151–171.

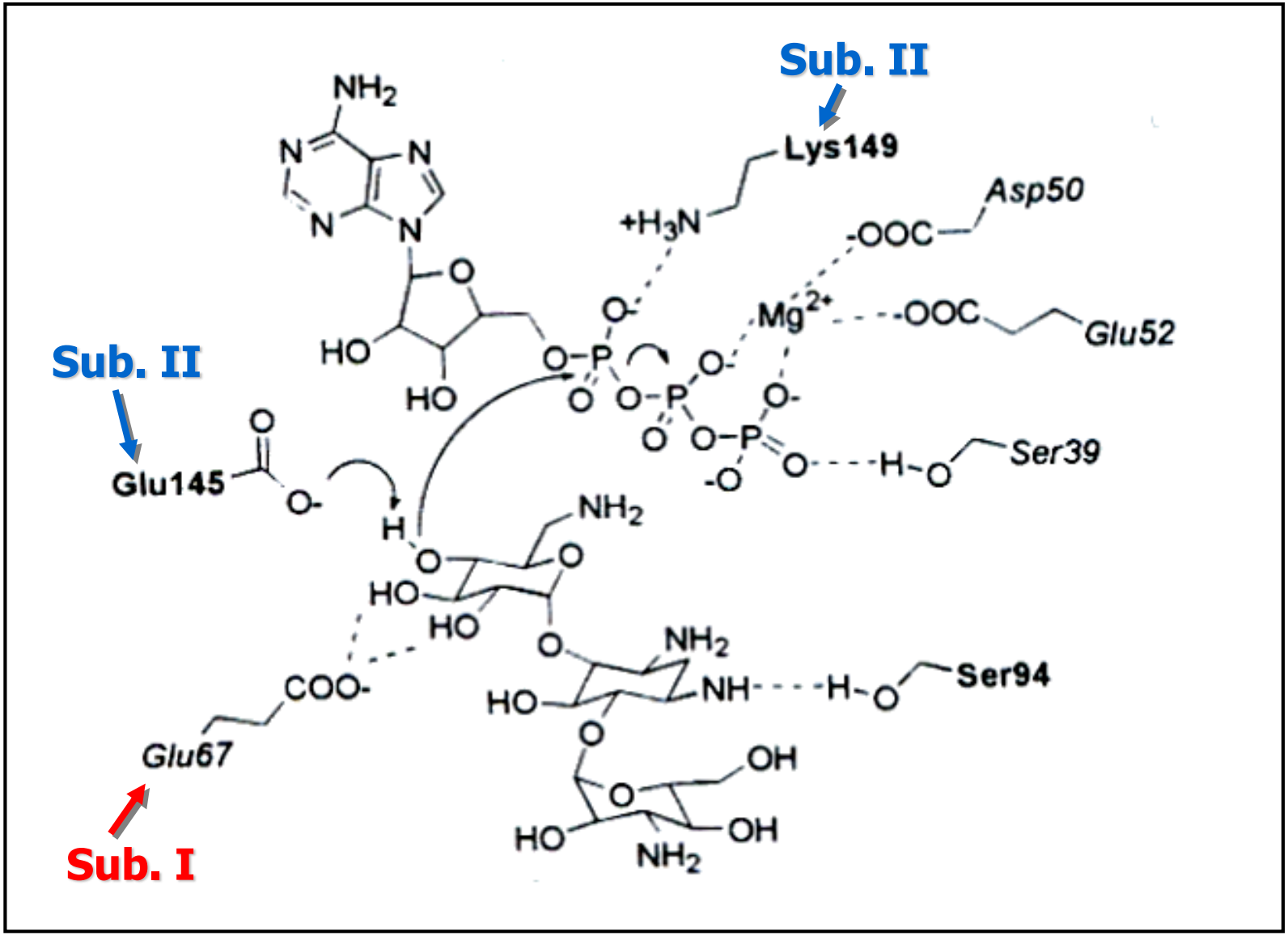
5-(2,4-diguanidino-3,5,6-trihydroxy-cyclohexoxy)- 4-[4,5-dihydroxy-6-(hydroxymethyl)-3-methylamino-tetrahydropyran-2-yl] oxy-3-hydroxy-2-methyl-tetrahydrofuran-3-carbaldehyde



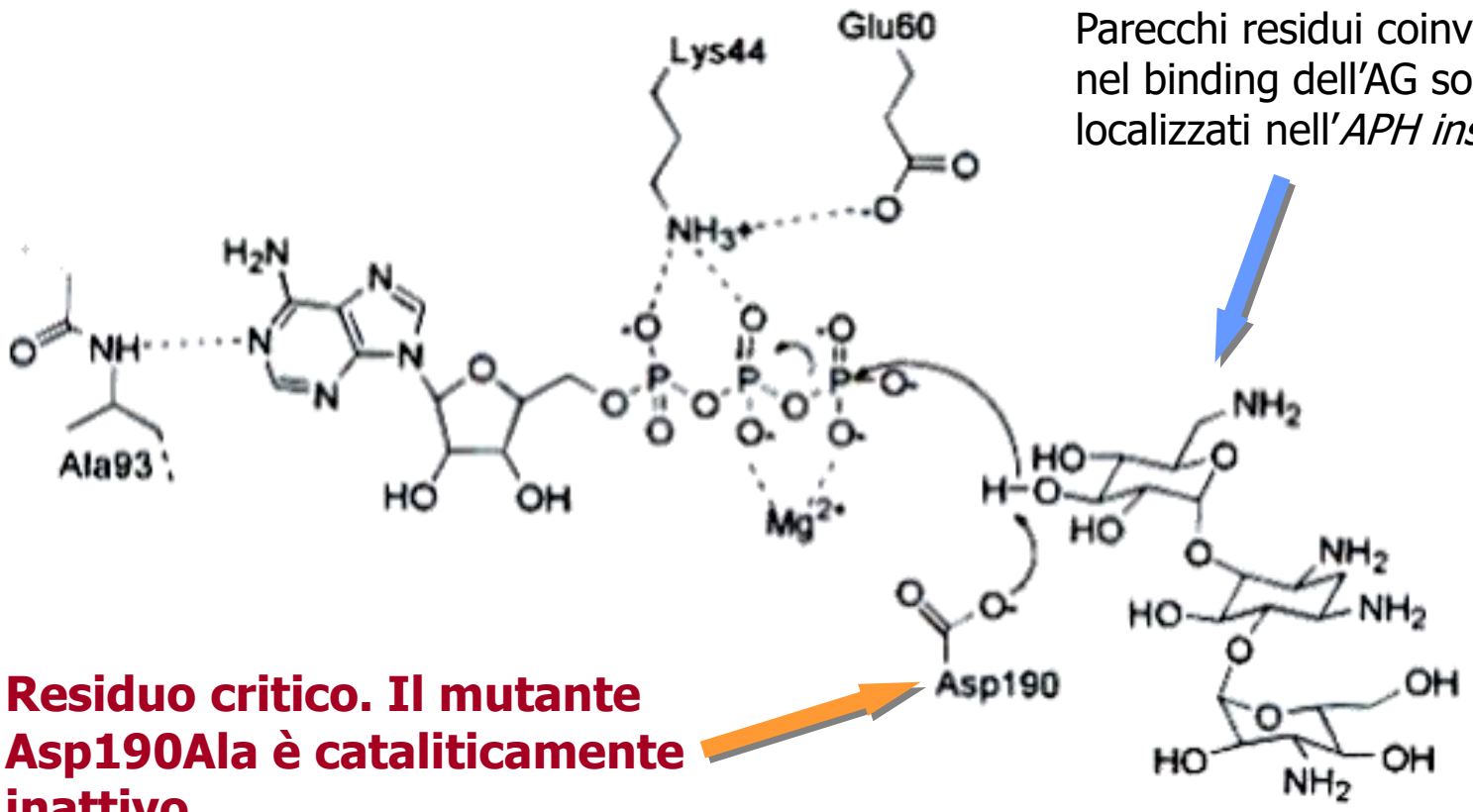


APH (Amminoglicoside PHospho transferasi)
AAD (Ag ADenil transferasi)
AAC – Amminoglucoside ACetil transferasi

Meccanismo di adenilazione da ANT(4')



Meccanismo di fosforilazione da APH(3')-IIIa



Parecchi residui coinvolti nel binding dell'AG sono localizzati nell'*APH insert*

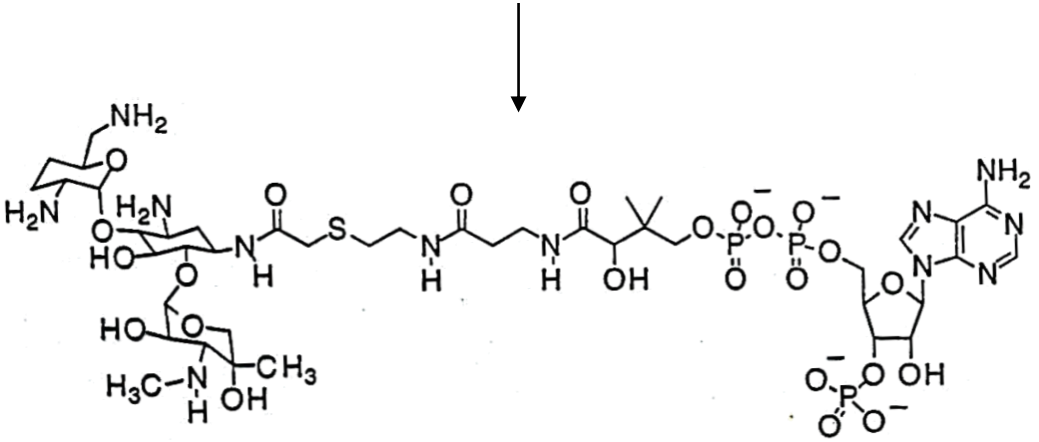
Residuo critico. Il mutante Asp190Ala è cataliticamente inattivo

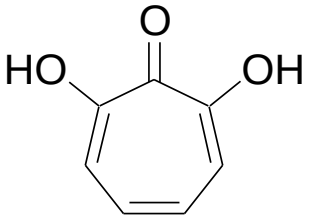
Inibitori degli enzimi inattivanti

Aminoglicosidici, non aminoglicosidici

(1) Inibitori AG

- **Tobramicina e dibekacina** (3'-desossi AG) sono inibitori competitivi della fosforilazione di kanamicina da parte di APH(3'): K_i < 1 μM
- **Paromomicina** (6'-OH AG) inibisce l'acetilazione in pos. 6' della kanamicina da parte di AAC(6')-Ia: K_i = 1.06 μM
- Inibitori bifunzionali di AAC(3)-I (bis-substrato)



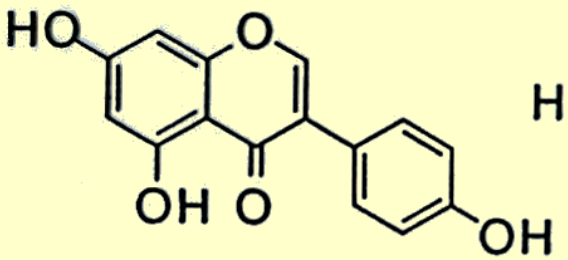


7-idrossitropolone

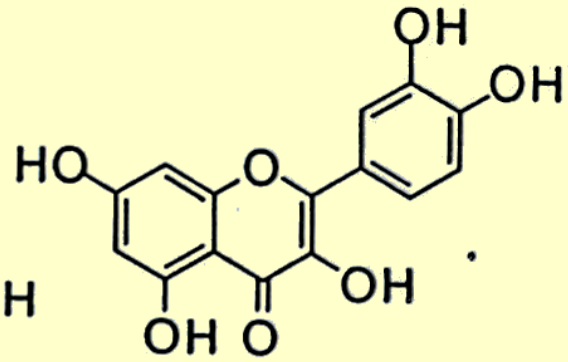
Antimicrob. Agents Chemother. **1982**, *22*, 824.

(2) Inibitori non-AG

Si tratta, per lo più, di inibitori di protein-chinasi (PK), che inibiscono APH



Genistein



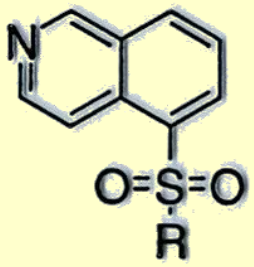
Quercetin

Flavonoidi

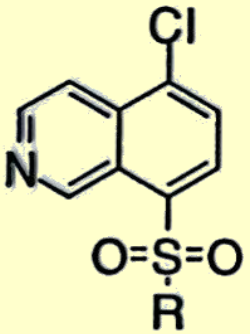
Inibitori di APH(3'), ma non di AAC(6') e APH2")

Derivati della isochinolinsolfonammide

- Potenti inibitori duali PK/APH
- K_i < 100 mM vs. APH(3')-IIIa, AAC(6')-APH(2")
- Inibizione competitiva vs. ATP e non-competitiva vs. canamicina



H series



CKI series

- Ribosoma batterico, processo di traslazione proteica;
- Amminoglicosidi;
- **Macrolidi;**
- Tetracicline;
- Streptogramine, Lincosammidi; Cloramfenicolo; Acido Fusidico; Oxazolidinoni.

ANTIBIOTICI MACROLIDICI

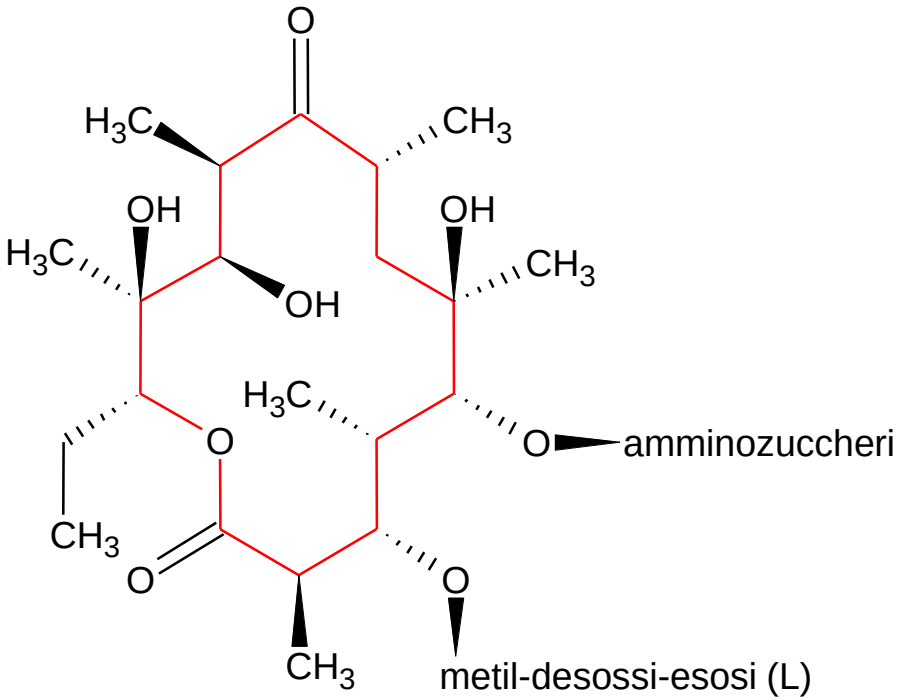
(MACROLIDI-*macro*lactone g*ly*cosi*de*)

Robert Burns Woodward (Nobel 1965) Angew. Chem. 1957, 69, 585.

Struttura eterosidica

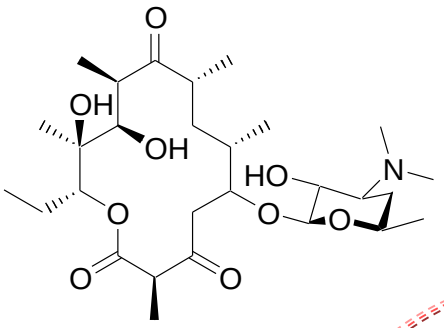
AGLICONE (polyketide)

- Lattone macrociclico, con uno scheletro saturo o polienico
- Macrolidi a 12, **14**, 16 termini (saturi) e a 26-38 termini (insaturi)



Origine biogenetica (metaboliti secondari da actinomiceti)

Abelardo Aguilar (1949)-Eli Lilly Co-Ilosone

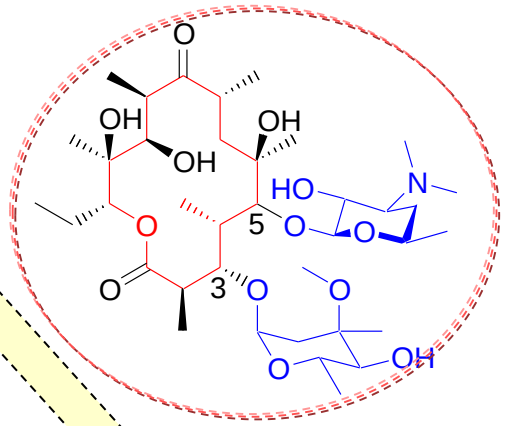


pikromycin

I generazione

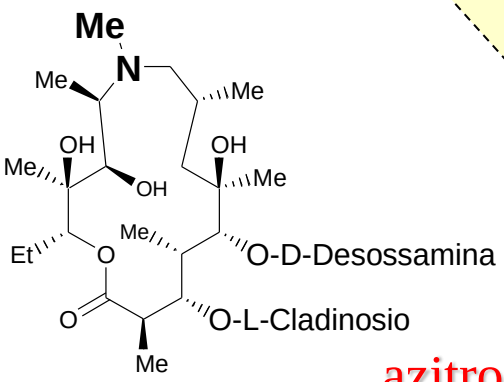
Macrolide 14 atomi;
Desosamina e Cladinosio (C5, C3);

1950



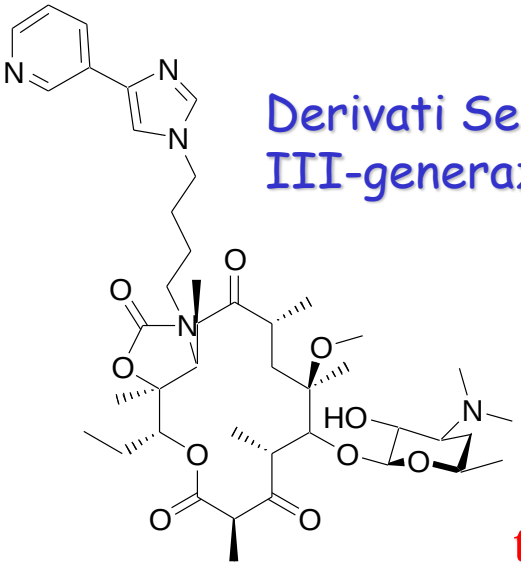
eritromicina

1955-60



azitromicina
(azalide)

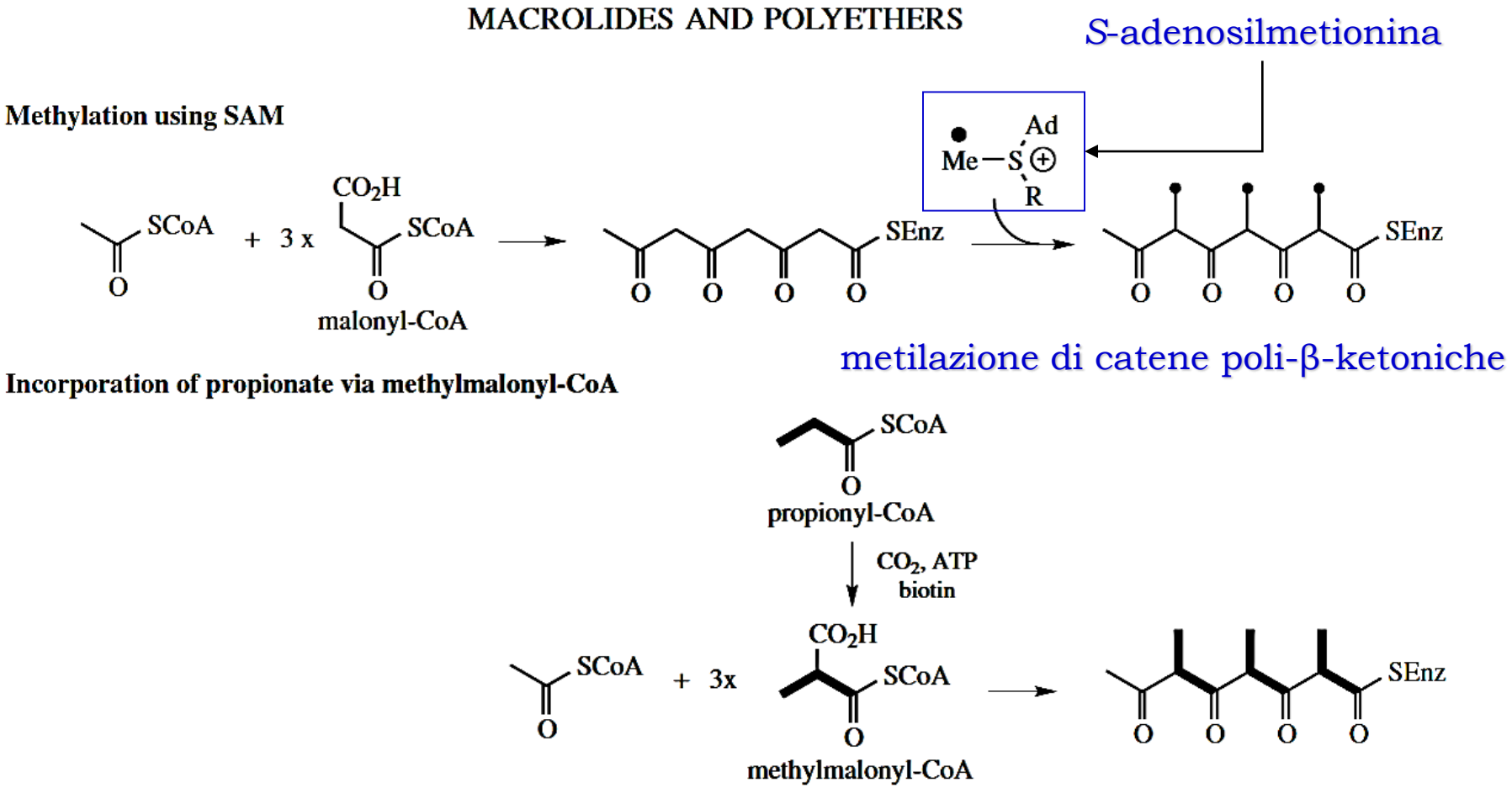
Derivati Semisintetici di
II-generazione (‘80-’90)



Derivati Semisintetici di
III-generazione 2000-4

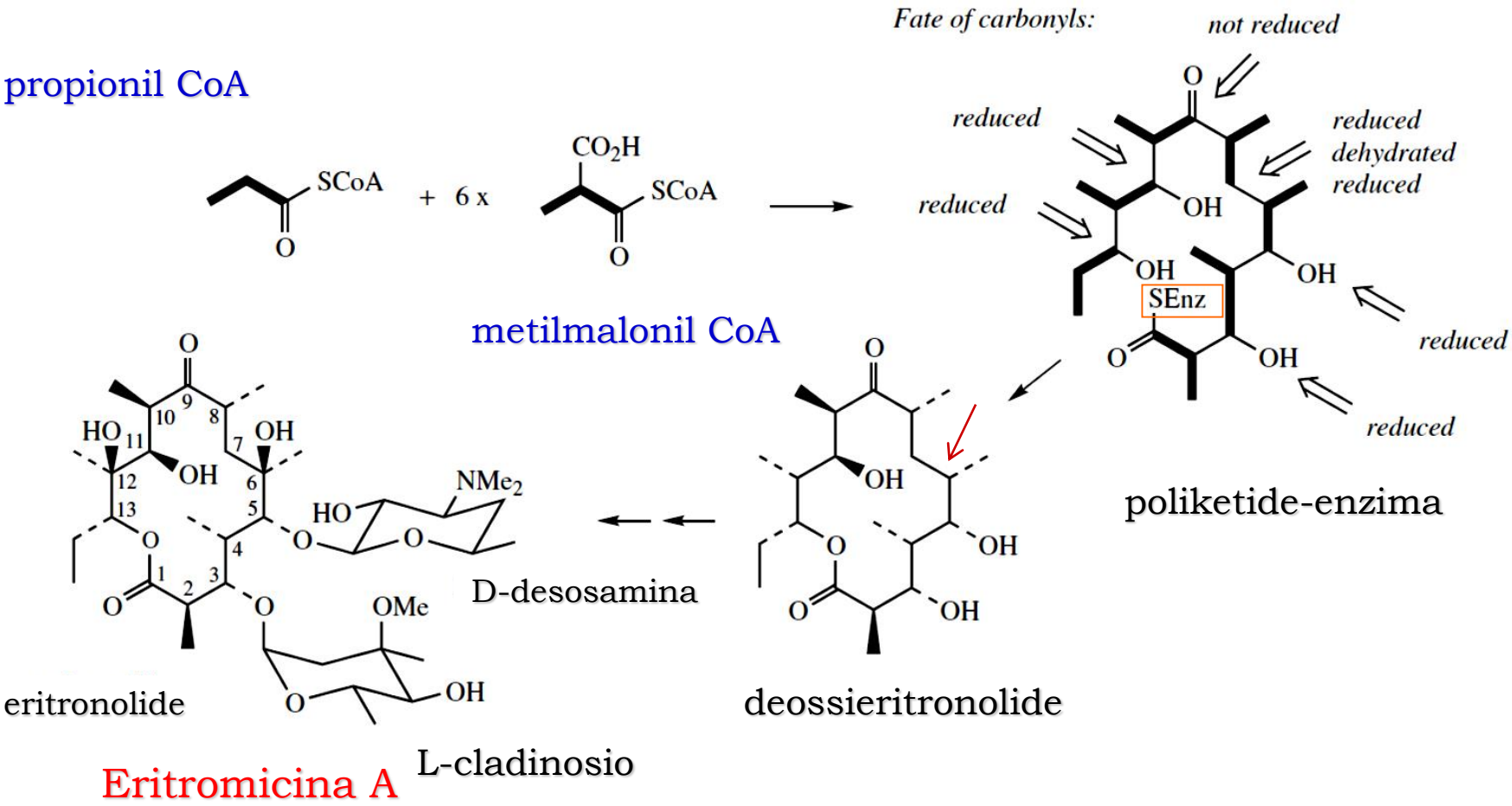
telitromicina

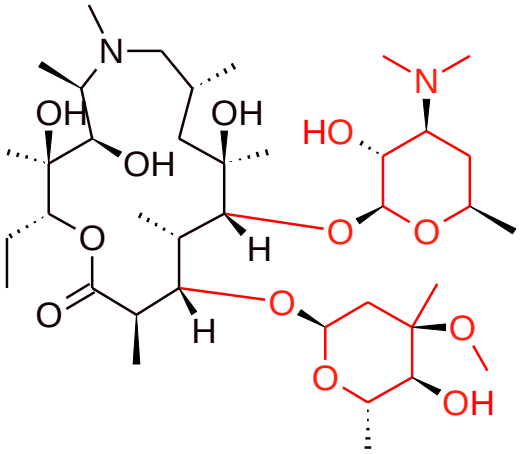
Biogenesi macrolidi 14 termini



Biosintesi di poliketide metilato

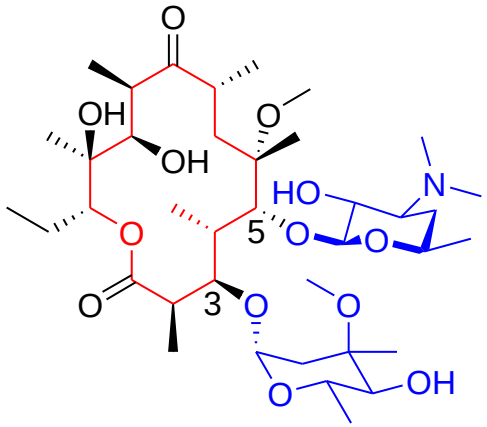
Biogenesi macrolidi 14 termini (es. *Saccharopolyspora (Streptomyces) erythraea*)



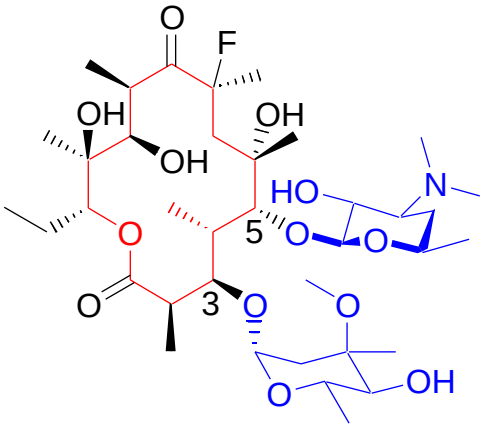


azitromicina

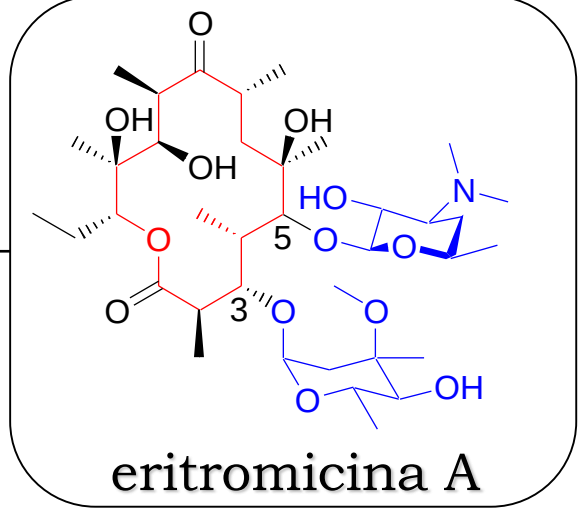
(azalide)



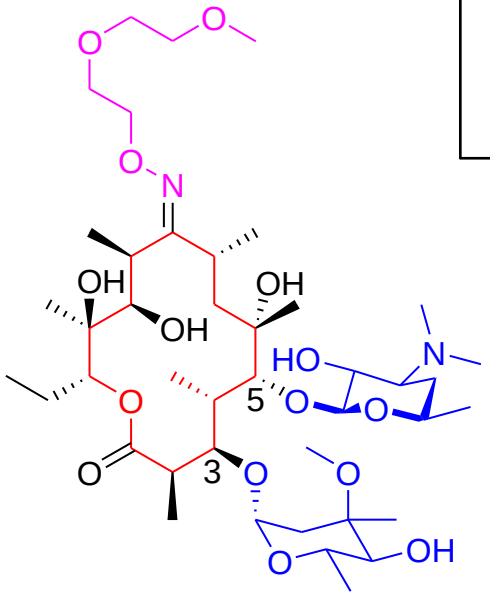
claritromicina



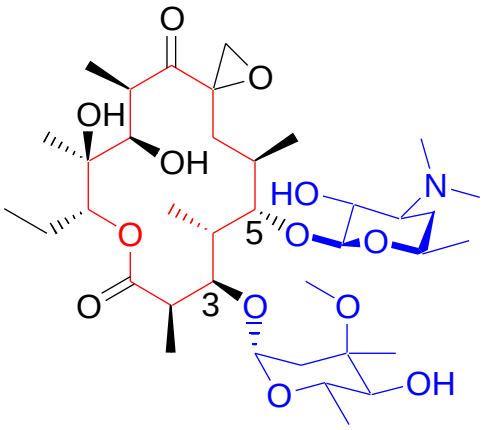
fluritromicina



eritromicina A

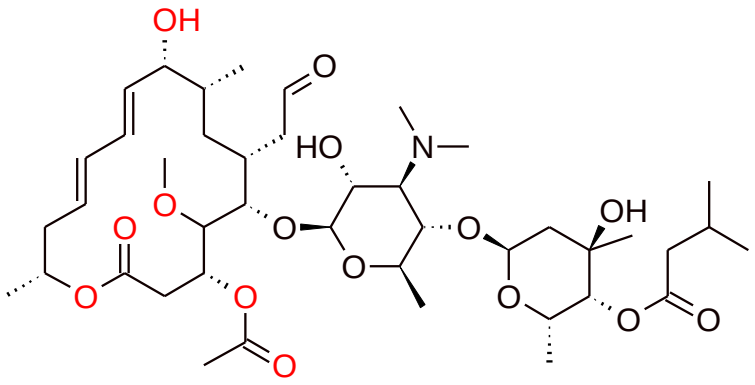


roxitromicina



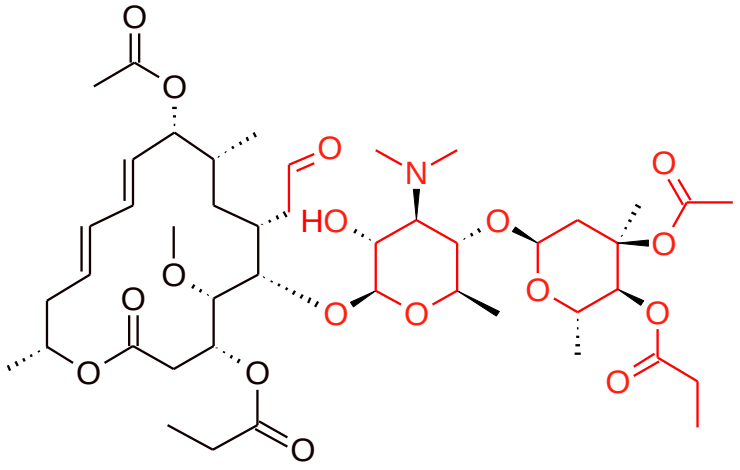
oleandomicina

Macrolidi a **16** termini



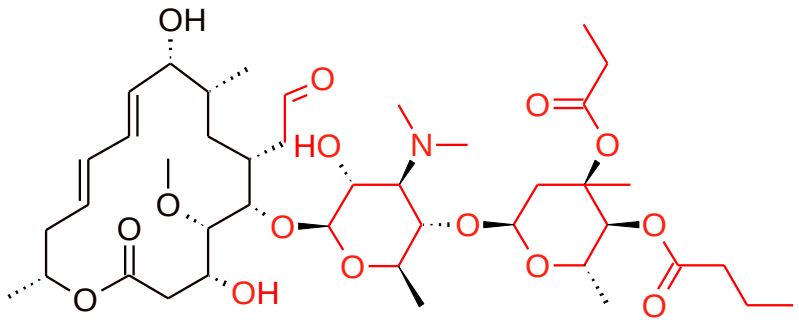
Josamicina *(Iosalide)*

G+ *Mycoplasma pneumoniae, Neisseria gonorrhea, Neisseria meningitidis, Bordetella pertussis*

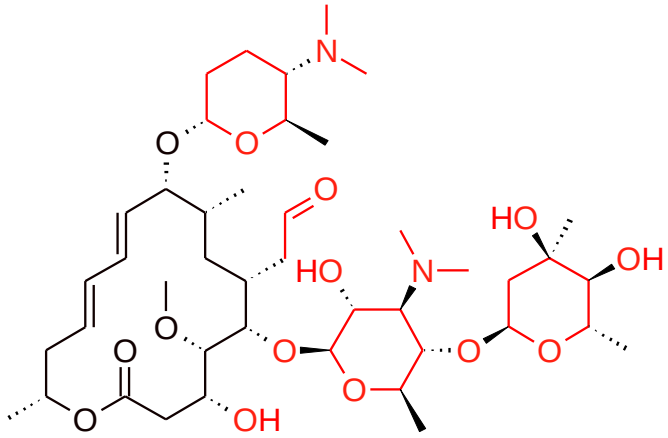


Miocamicina

(Macroral, Miocamen, Miokacin)



rokitamicina

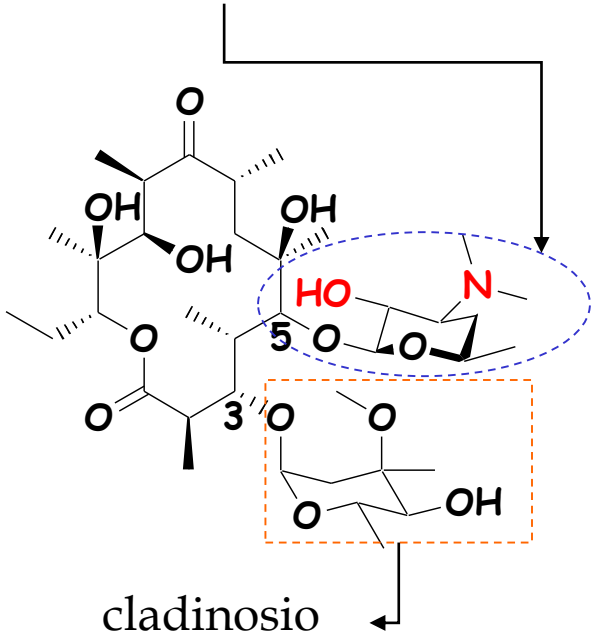


Spiramicina I

(Rovamicina, Spiromix, equiv)

ERITROMICINE

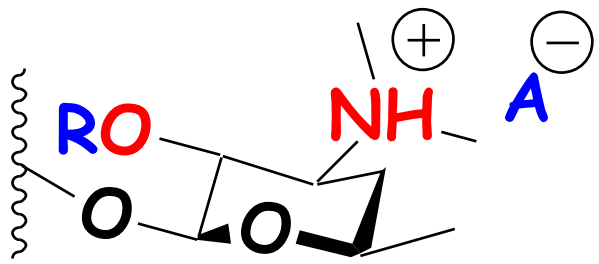
desosamina
(3-dimetilammino-3,4,6-tridesossi-D-glucosio)

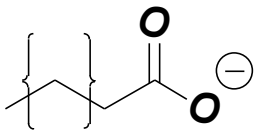
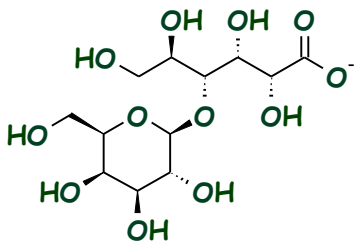
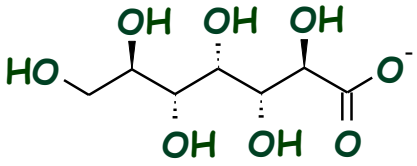
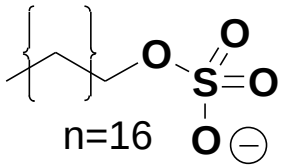
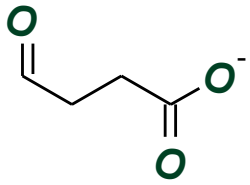


- Sostanze di natura basica
- Sali usati: cloridrato (os), stearato (os), glucoeptonato (ev), lattobionato (ev)
- Profarmaci (Esteri 2'-OH della desosammina): stearil solfato dell'estere propionico (os), emisuccinato (sospensione orale)

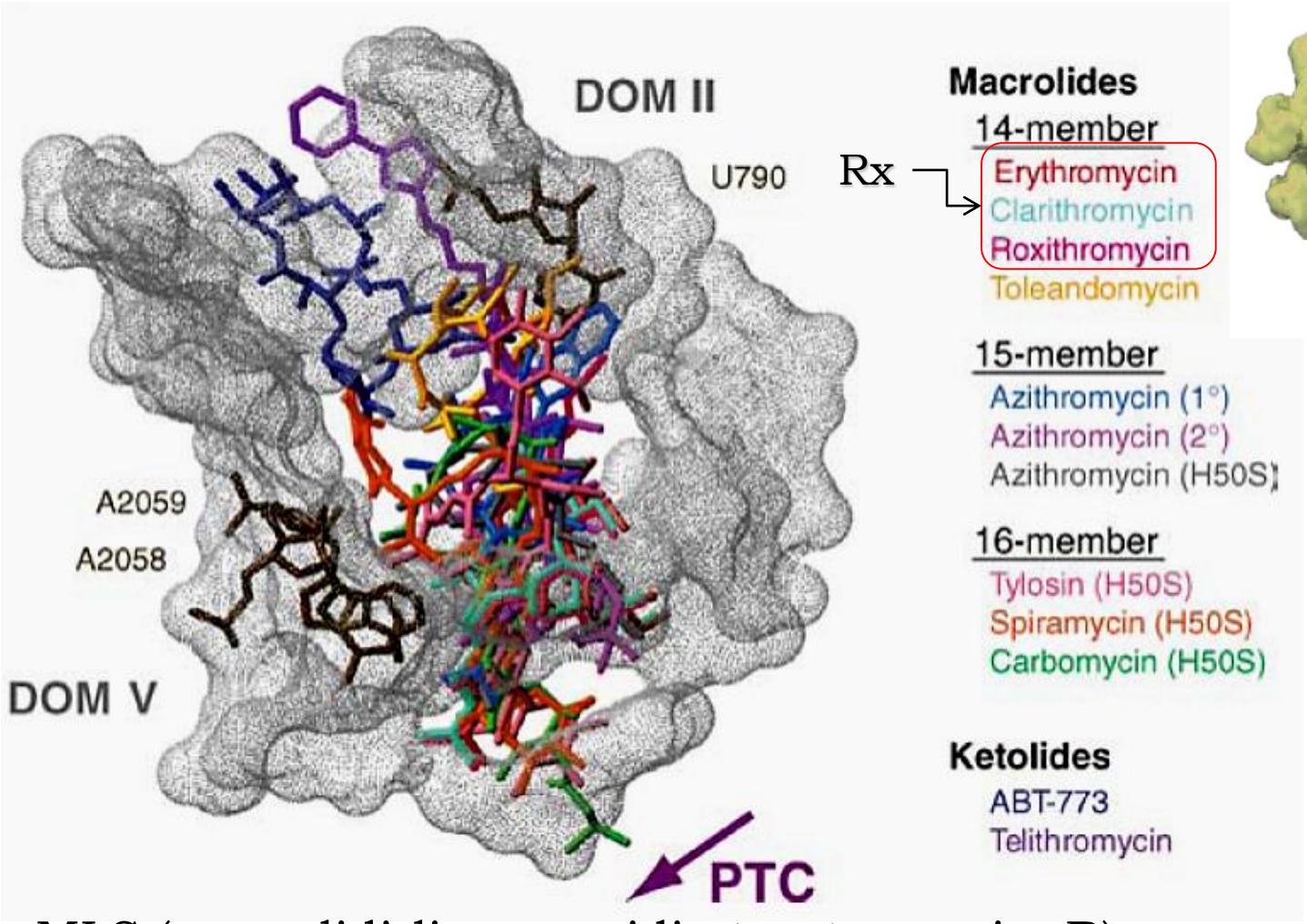
- **Spettro antibiotico**
  Relativamente ristretto, confrontabile con quello delle penicilline a medio spettro. Attive su Gram +, alcuni Gram - e protozoi.
- **Uso**
  Infezioni delle vie respiratorie, causate prevalentemente da Gram +; trovano impiego anche nelle malattie sessualmente trasmesse
- **Tossicità**
  Scarsa, bene assorbite per via orale, biodisponibilità modesta (3-4 somministrazioni giornaliere), breve emivita, crampi intestinali

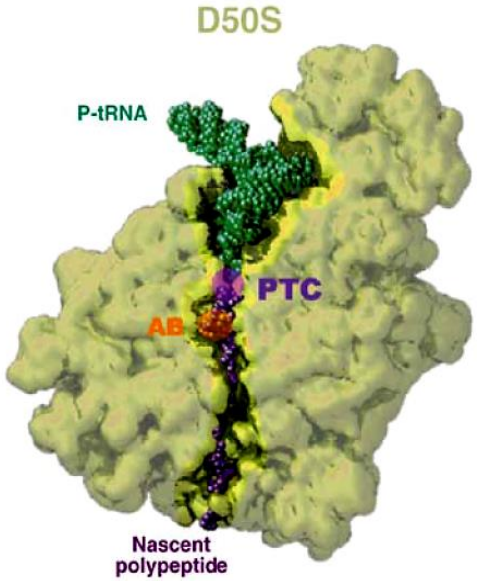
- Sali usati: cloridrato (os), stearato (os), glucoeptonato (ev), lattobionato (ev)
- Profarmaci (Esteri 2'-OH della desosammina): stearil solfato dell'estere propionico (os), emisuccinato (sospensione orale). < solubilità →Idrolisi intestinale



R	A^-
H	Cl
H	 n=16
H	
H	
CH_3CH_2CO	 n=16
	

Eritromicina, Claritromicina e Roxitromicina legate alla subunità ribosomiale 50S (sito P, tunnel di elongazione dei peptidi)



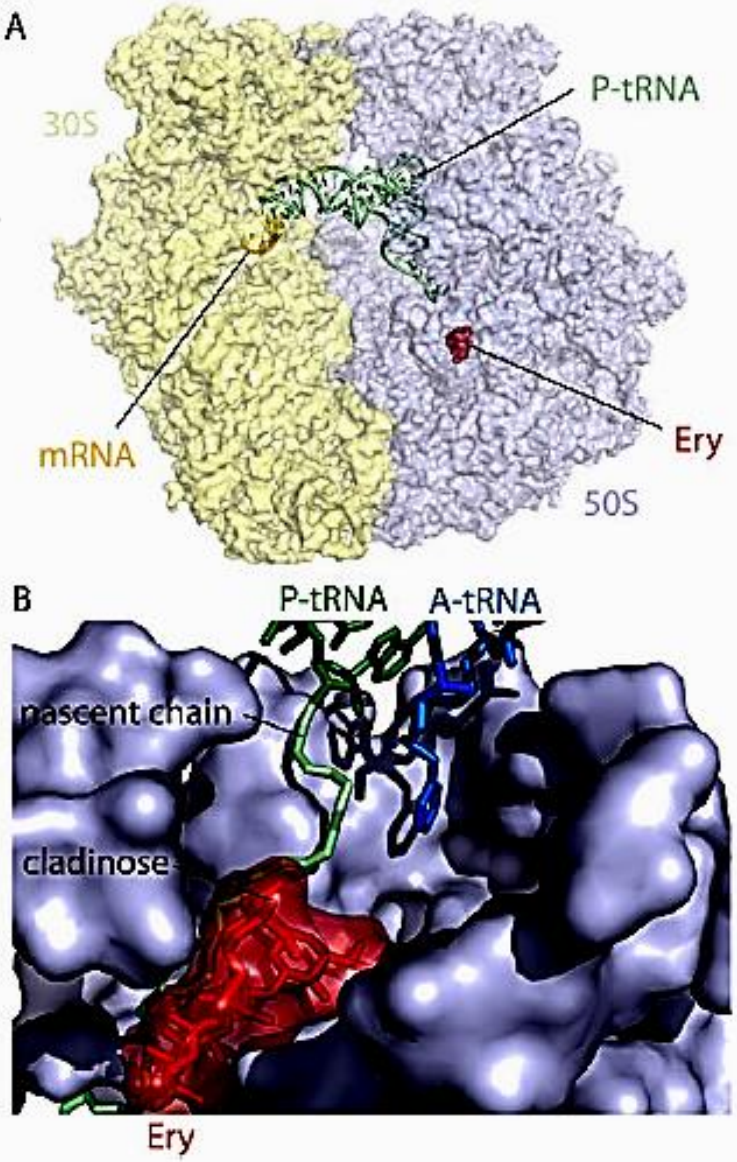


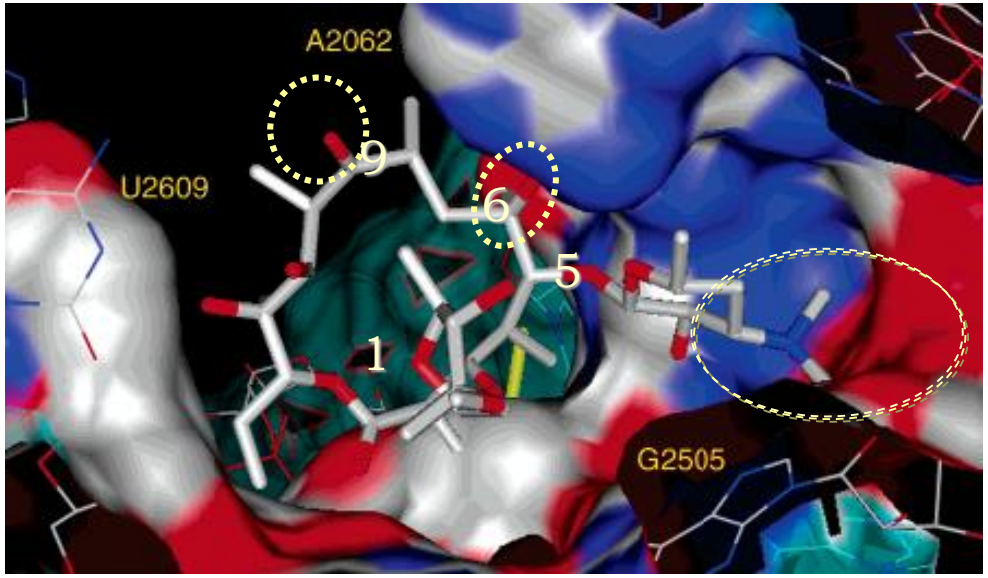
peptidyl-transferase center

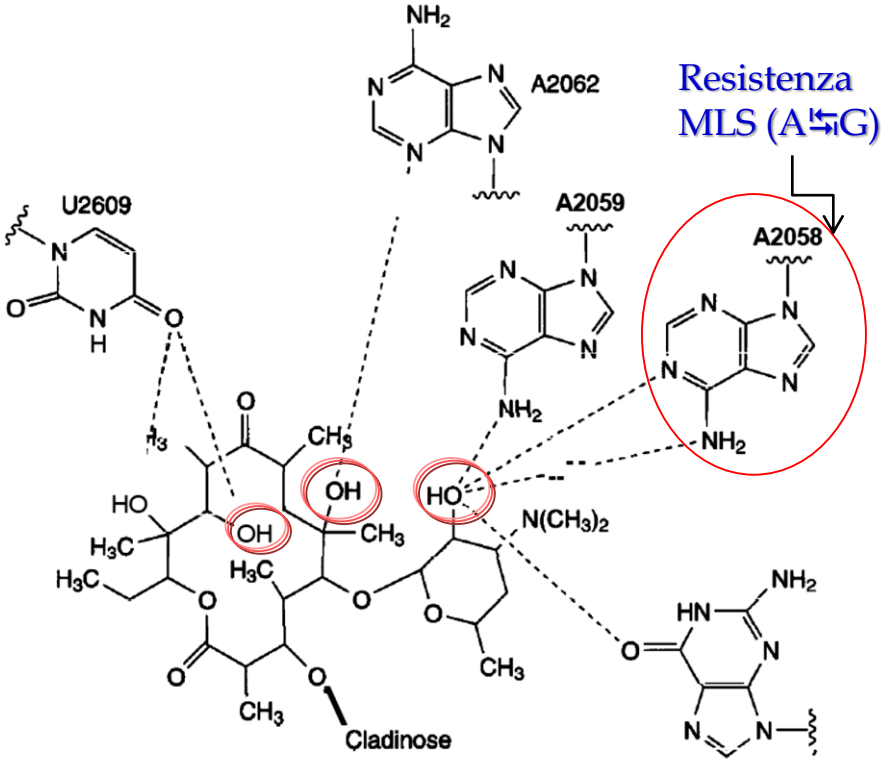
Annu. Rev. Biochem. 2005. 74:649-679.

MLS (macrolidi-lincosamidi-streptograminaB)
→meccanismo di resistenza (A⇆G); *erm*→adenina rRNA23S

70S *E. coli*


A
30S
P-tRNA
mRNA
Ery
50S
B
P-tRNA
A-tRNA
nascent chain
cladinose
Ery
C
cladinose
C2610
C2611
A2503
A2058/
A2059
Ery
U2609
lactone ring
D
Ery
C2610
H50S-Tel
A2058/
A2059
U2609
A752
T70S-Tel
D50S-Tel
A751





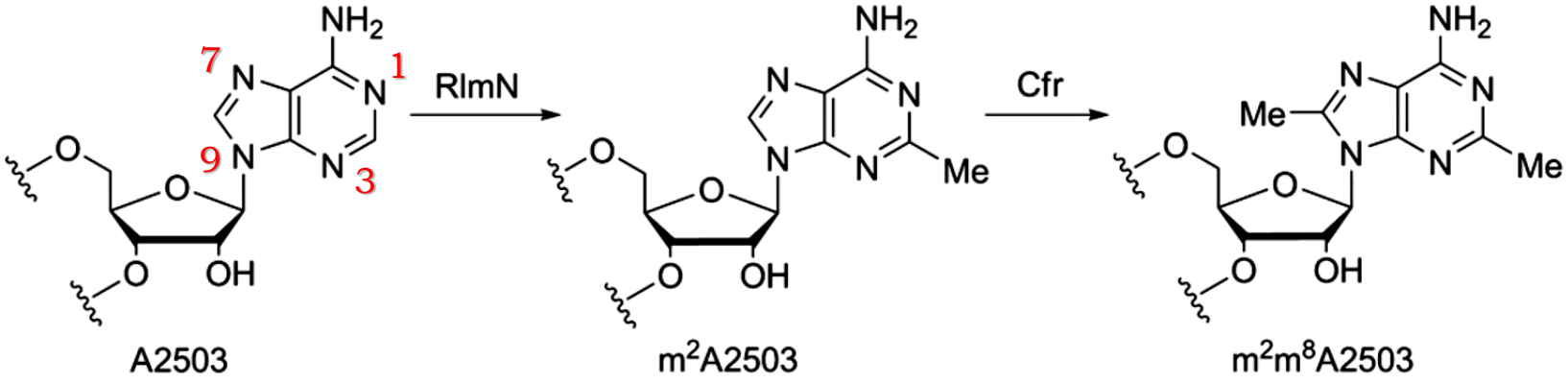
Binding dell'eritromicina alla subunità 50S mediato dai residui della desosammina.

Le regioni con elevata carica negativa sono in rosso; un'interazione importante si crea tra il gruppo fosfato di G2505 e l'ammina della desosammina.

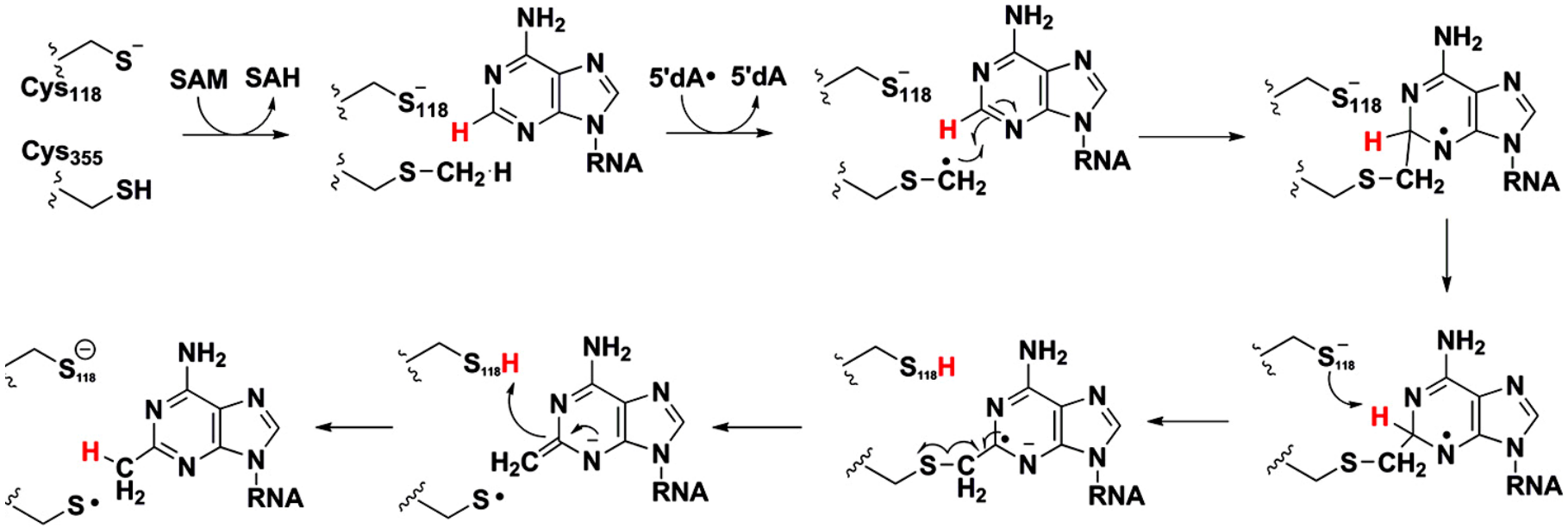
Resistenza ai Macrolidi (MLSB)

- **Proteine di efflusso.**
  - Mef: *antiporters*, scambi del macrolide legato con H^+; I geni *mefA* e *mefE* conferiscono resistenza ai M a 14- e 15-membri (ma non ai M a 16-membri), lincosamidi e streptogramine B
  - Msr: spiazzano I M dal ribosoma proteggendolo dal binding con gli antibiotici; *msr* geni espressi in *St. epidermidis, St. xylosus, St. aureus, Enterococcus, Streptococcus, Pseudomonas.*
- **Modificazioni rRNA 23S.**
  - Erm: enzimi transferasi (erythromycin resistance methylase) che catalizzano mono- (N) o di-metilazione (C/E) all'N6 dell'A2058. M a 14- o 15-membri con un carboidrato neutro (C3) sono generalmente induttori M a 16-membri e ketolidi a 14-membri non sono induttori. are not inducers. Forme inducibili e **costitutive.**
- **Mutazioni in rRNA.**
  - mutazione A2058G.
    resistenza ai M (<<ketolidi).
- **Mutazioni nelle proteine ribosomiali.**
- **Modificazioni di M a 14- e 15-membri da parte di specifiche esterasi**
  - ketolidi e 16-membri non sono substrati
- **Modificazioni da parte di kinasi o fosfotransferasi** (cfr aminoglicosidi)

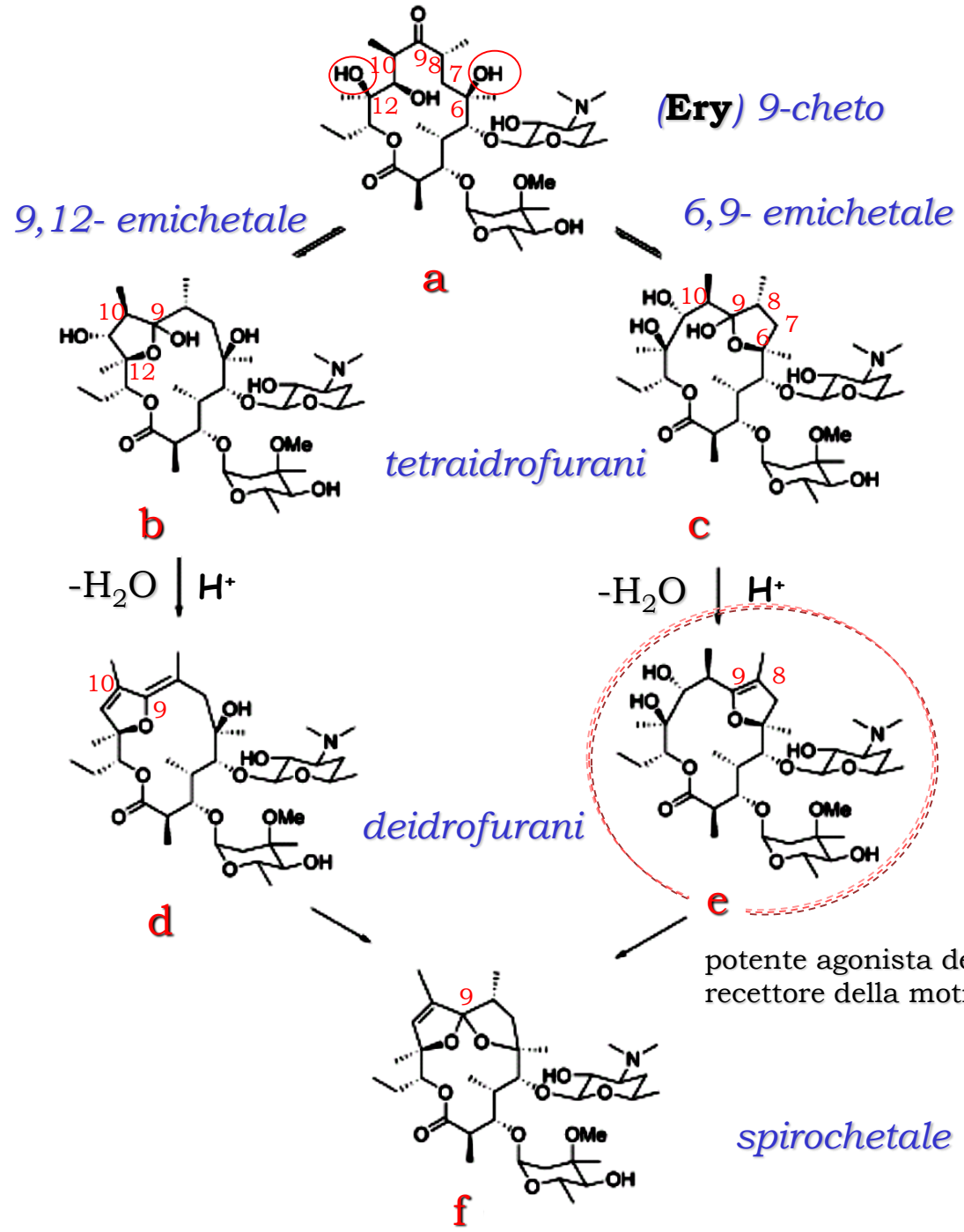
Resistenza *(MLSB-resistance phenotype)*



PNAS 2011, 108(10), 3930–3934



THE JOURNAL OF BIOLOGICAL CHEMISTRY VOL. 290, NO. 7, pp. 3995–4002, February 13, 2015



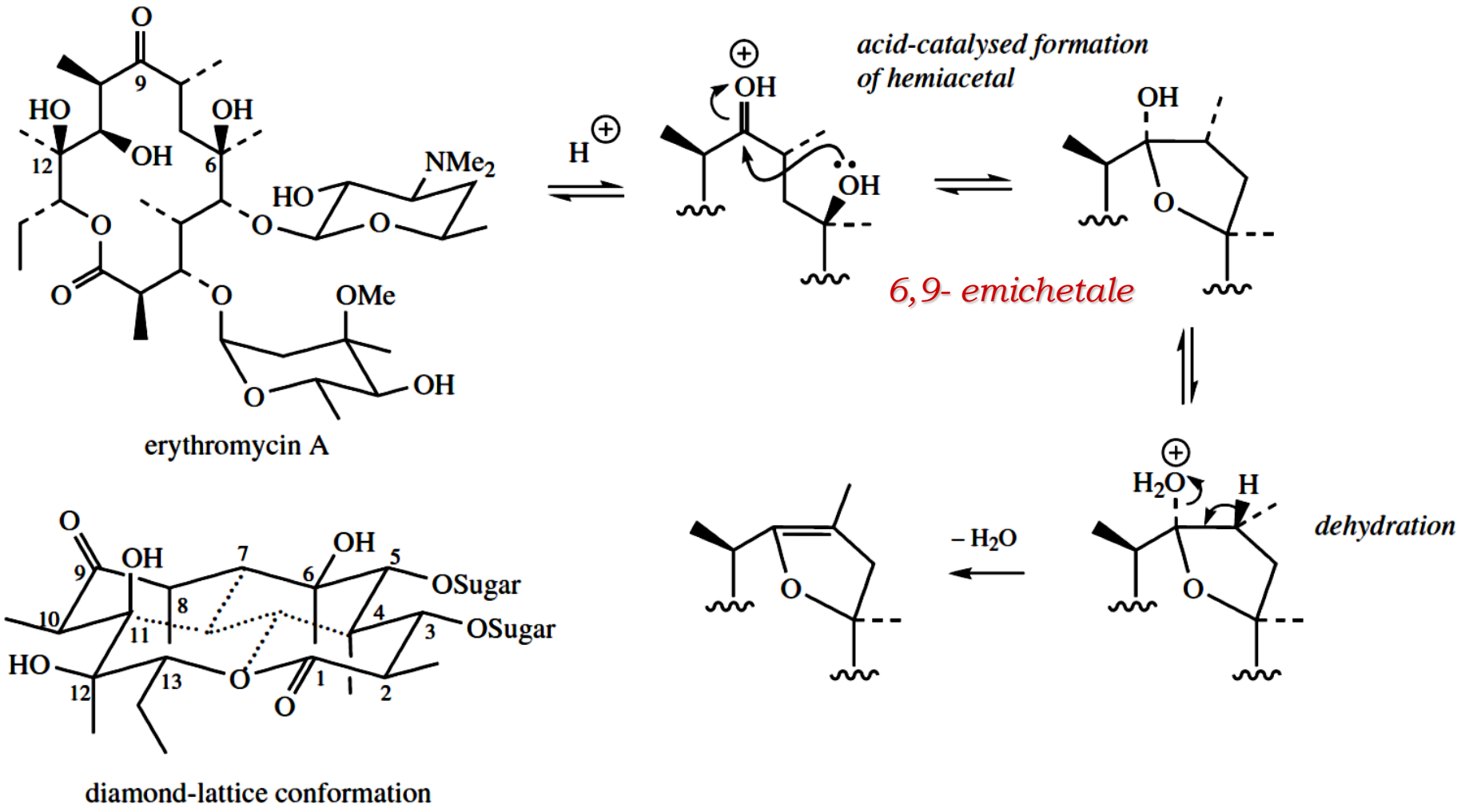
I gruppi OH in 6- e 12 sono la maggior fonte di instabilità.

In solventi protici **Ery** esiste come miscela di

1. 9-keto (**a**),
2. 9,12-emiketale (**b**)
3. 6,9-emiketale (**c**).

In ambiente acido gli emiketali deidratano rispettivamente ai derivati enol eteri (**d**) e (**e**), che a loro volta degradano a derivato spiroketale (**f**).

La forma keto **a** è la sola con attività antibatterica, mentre l'enol etere (**e**) è un potente agonista del recettore della motilina (spasmi gastro-intestinali).



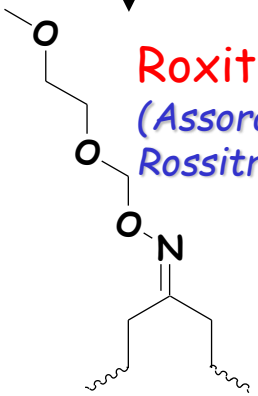
Ery adotta una conformazione tipo diamante-lattice (a sedia). Il C6 riduce l'interazione 1,3-diassiale con C4 discostandosi da questa conformazione avvicinandosi al carbonile C9 e reagendo (OH) con formazione di emichetale (analoga reazione ha luogo tra OH (C12) e carbonile C9.

SAR

chetone C9:

• Riduzione→9-diidroeritromicina (più stabile, meno attiva);
• ossime (E>Z) e idrazoni in genere conservano l'attività (minore biodisponibilità)

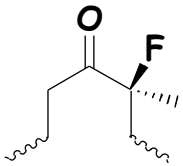
cfr cefalosporine III gen. (oximino)



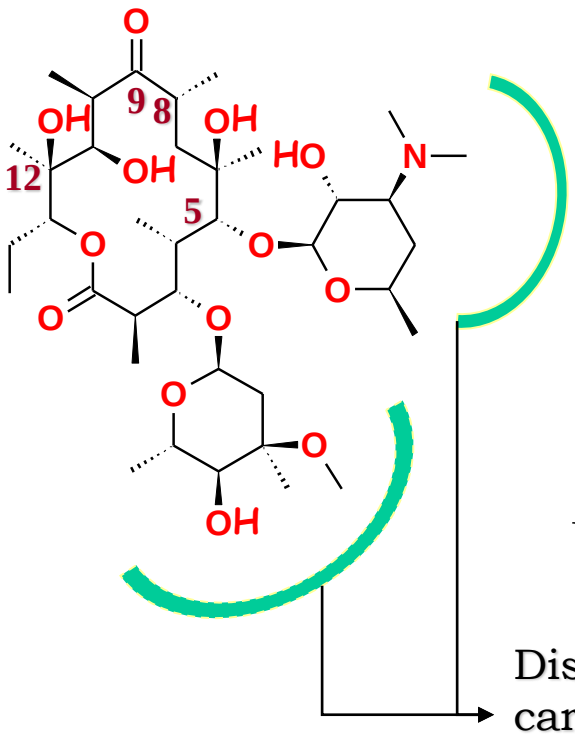
Roxitromicina
(Assoral, Overal, Rossitrol, Rulid)

(metossietossietere ossima) spettro antibatterico simile all'eritromicina ma più efficace verso Gram- (*Legionella pneumophila*)

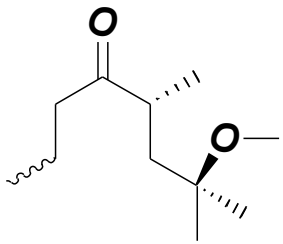
C8: F: previene formazione anidro-emichetale (no H), attività antistafilococcica (strep. simile); + potente anaerobi



Fluritromicina
(Flurizic, Mizar, Ritro)

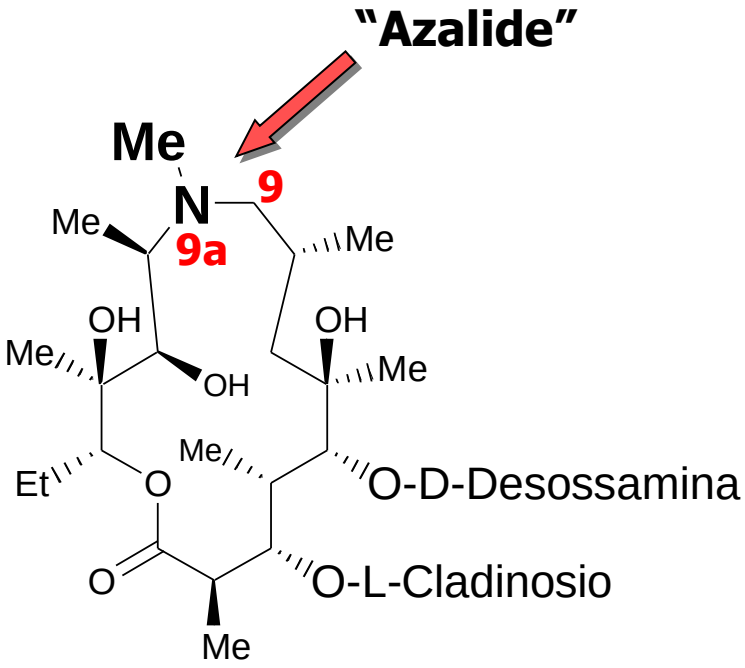


C6: ingegneria genetica (inattivazione *eryF* che codifica una C6 idrolasi specifica);
• 6-deossieritromicine generalmente meno potenti;
• 6-O-alchileritromicine + stabili in ambiente acido

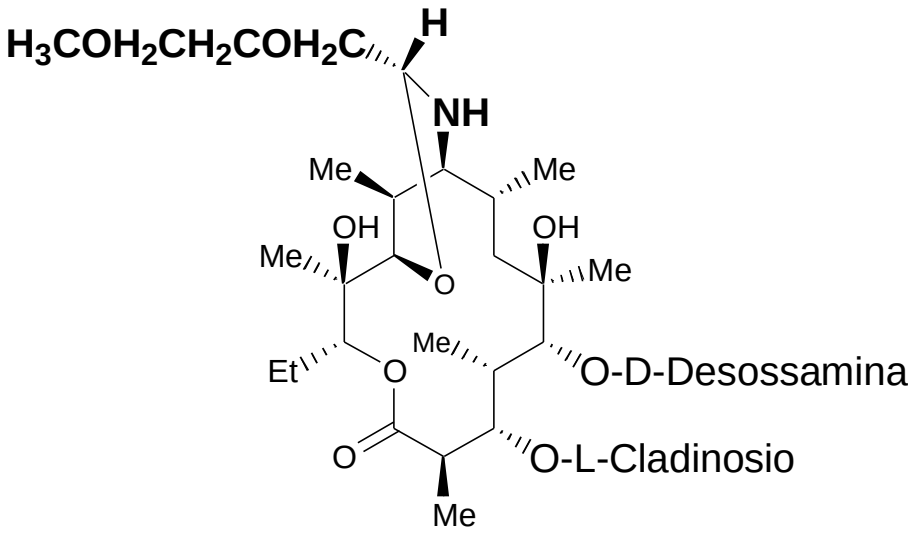


Claritromicina
(Klacid, Macladin, Veclam)

Disposizione relativa carboidrato-aglicone importante per l'attività



Azitromicina
(Azitrocin, Zitromax)

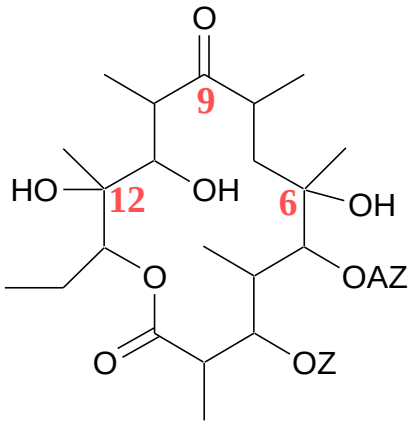


Diritromicina

Più stabile di Eritromicina in ambiente acido. EV più lunga (~ 60hr).
Singole somministrazioni giornaliere (a stomaco vuoto).

Dati farmacocinetici

Macrolidi a 14 termini

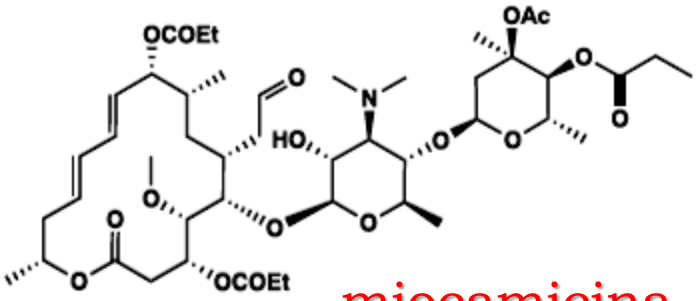


Eritromicina A

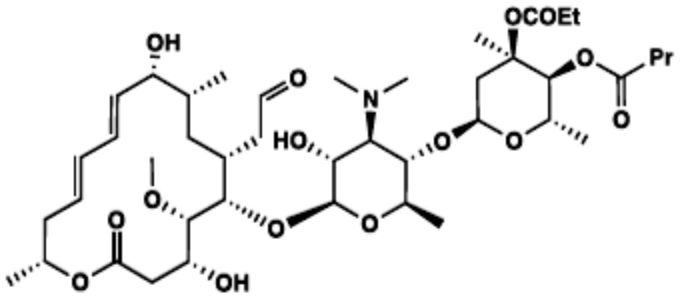
Z = Desossizucchero (cladinosio)

AZ = Ammino desossizucchero (Desossamina)

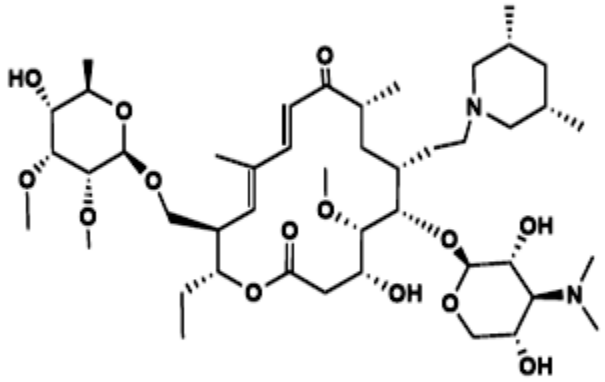
	Variazione strutturale	Esteri (sali)	EV (h)	LP (%)
Eritromicina		(solfato)	**2**	**70**
"		(lattobionato)		
"		(stearato)		
" **(estolato)**		Propionico (AZ) (laurilsolfato)		**98**
Claritromicina	6-OMe		**4**	**65**
Roxitromicina	Ossima sost.		**10**	**90**
Azitromicina	CO(9) → NCH_3		**15-60**	**50**
Oleandomicina	Desossi (6, 12) + Epossido (8)		**2-3**	
"	"	Triacetico (11, Z, AZ)		**50-60**



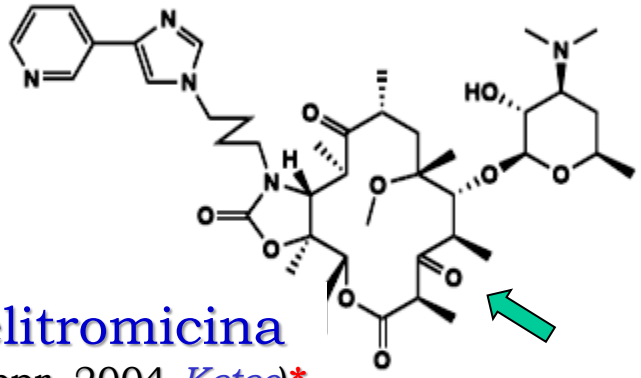
miocamicina



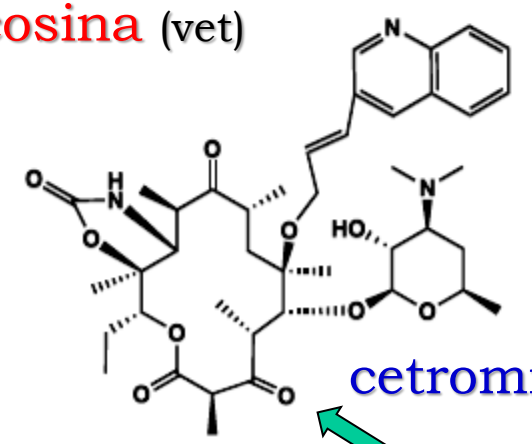
rokitamicina



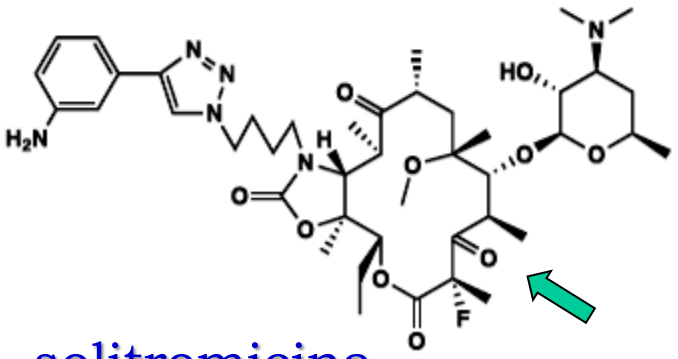
tilmicosina (vet)



telitromicina
(appr. 2004-*Ketec*)*



cetromicina



solitromicina

ketolidi

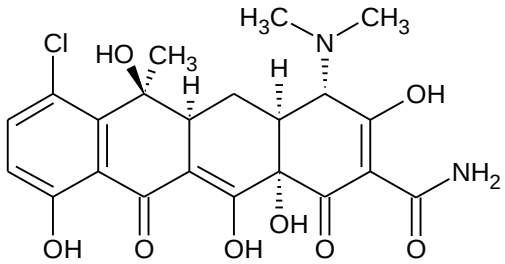
* tossicità-FDA (2006)

- Ribosoma batterico, processo di traslazione proteica;
- Amminoglicosidi;
- Macrolidi;
- **Tetracicline;**
- Streptogramine, Lincosammidi; Cloramfenicolo; Oxazolidinoni.

Toby Hockett (5Y) uno dei primi pazienti trattati con Aureomicina (Johns Hopkins Children's Hospital in Washington, DC, 1948)

Benjamin M. Duggar (consulente Cyanamide) e la sua ultramuffa prodotta da *Streptomyces aureofaciens* (Aureomicina, Ann. N.Y. Acad. Sci. 1948, appr FDA)

Ricercatori Pfizer→ struttura tetraciclina: Pilgrim, Conover (tetraciclina), Brunings, Gordon, Stephens (deoxiciclina)

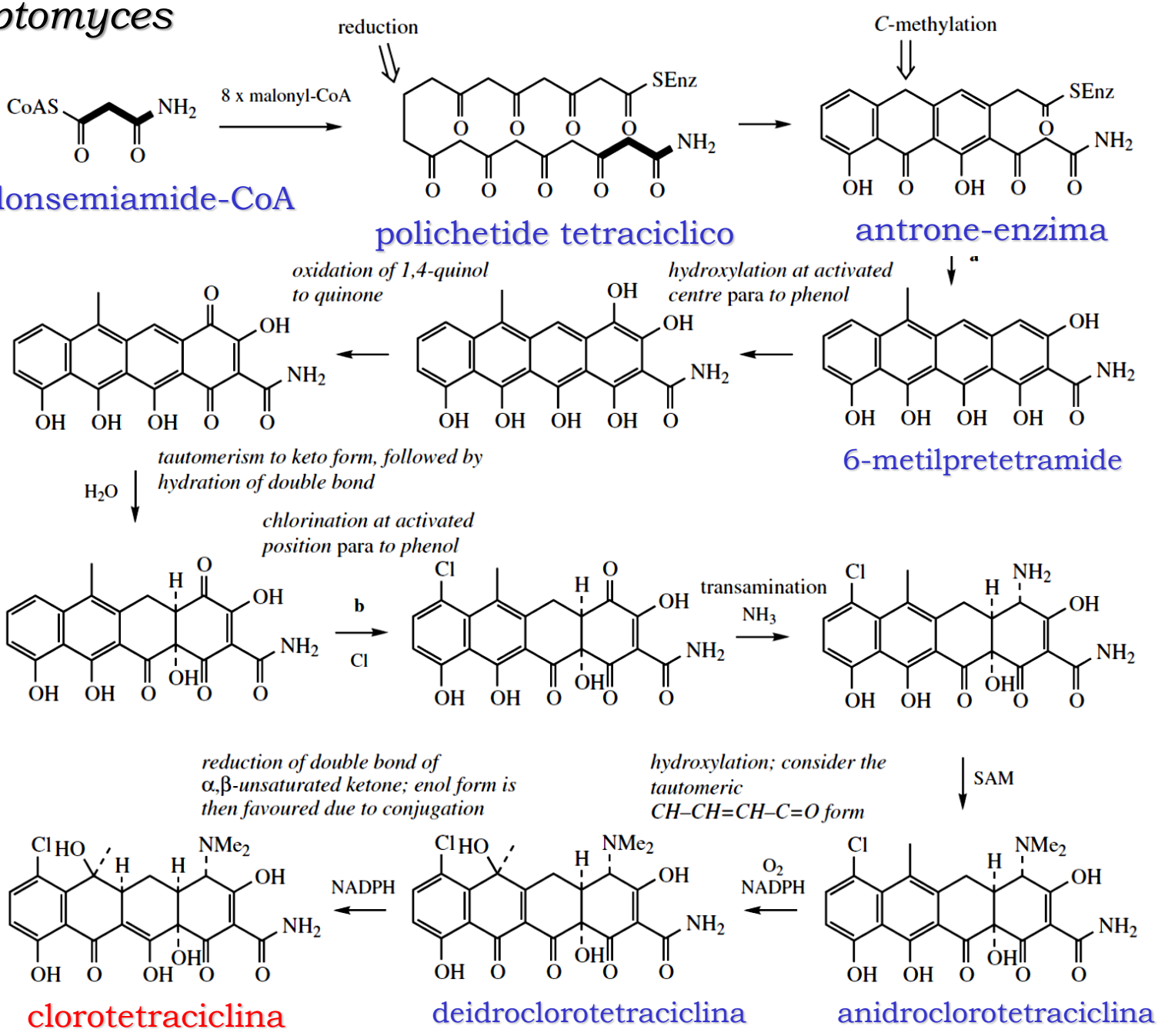


ANNALS OF THE NEW YORK ACADEMY OF SCIENCES Issue: Antimicrobial Therapeutics Reviews

The history of the tetracyclines

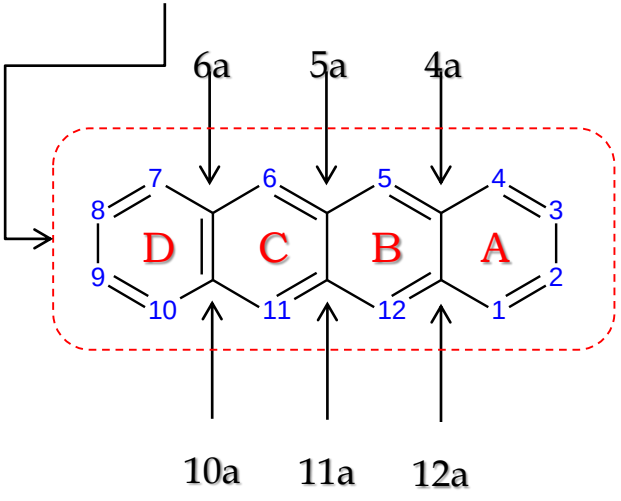
Ann. N.Y. Acad. Sci. 1241 (2011) 17–32 c 2011 New York Academy of Sciences.

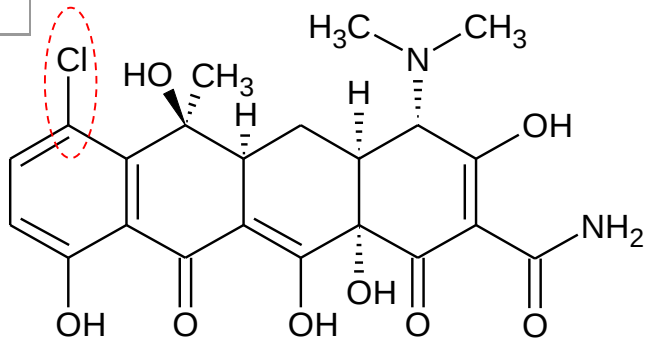
Streptomyces



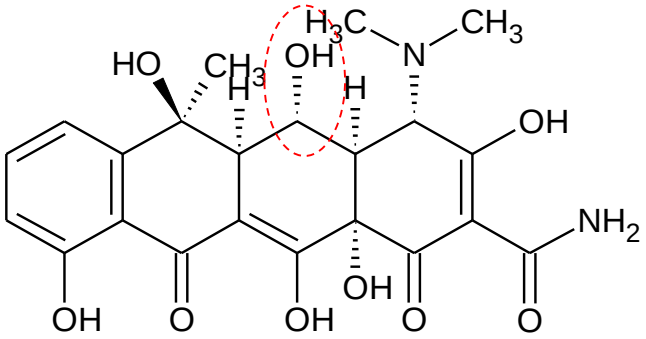
I^ generazione (o.s.)

naftacene
(tetracene)



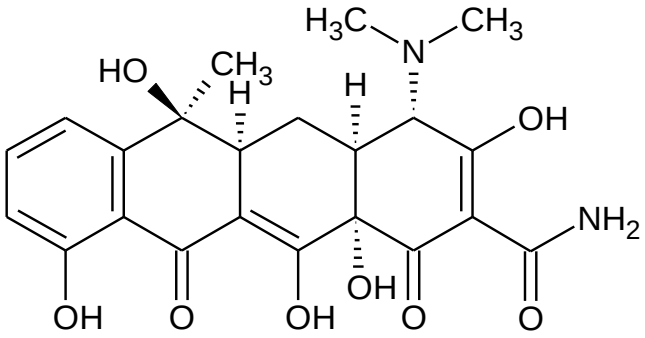


Clortetraciclina
(aureomicina, 1948)
Streptomyces aureofaciens



Oxytetraciclina
(terramicina, 1951)

Aureomicina
Primo antibiotico ad ampio spettro; anche gli estratti grezzi erano attivi vs patogeni responsabili di infezioni allora letali (tifo, richettsiosi, clamidiosi, micoplasmosi, bru cellosi, colera..)

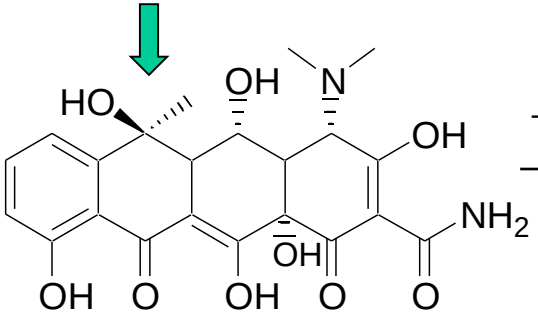


Tetraciclina
(teracina, desdecloro derivato aureomicina, 1953)

mutante blocco clorurazione

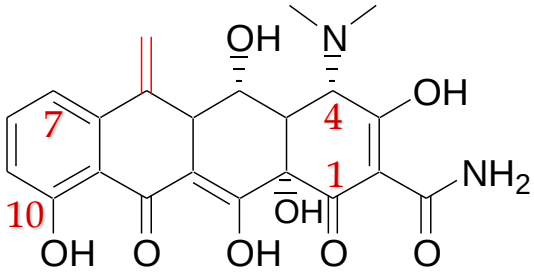
II^ generazione

processo Pfizer (chimico)

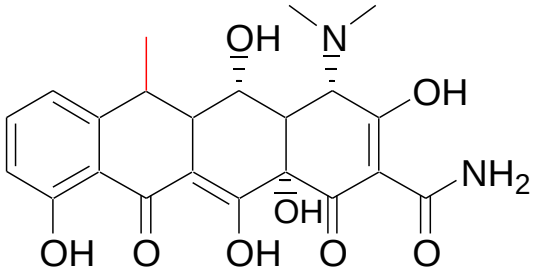


Oxytetraciclina

$-H_2O$

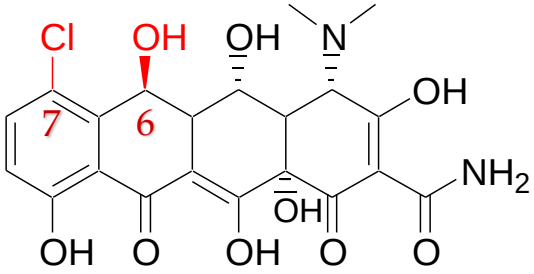


metaciclina
(*rondomicina*)

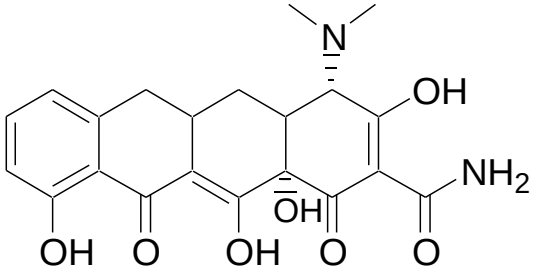


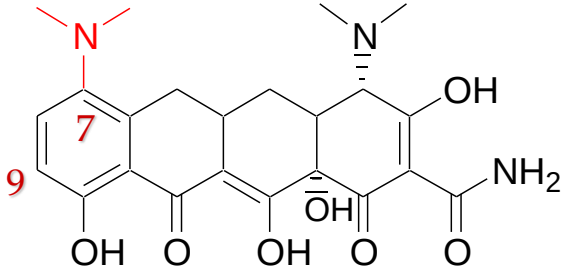
doxiciclina
(*vibramicina*,1967)
Miraclin (iclato)
B.anthracis, P.falciparum

processo Lederle (biotecnologico)



demeclociclina



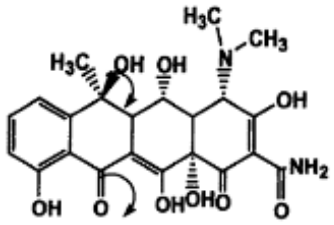


sanciclina
struttura
farmacoforica
minima

minociclina
(Minocin, 1971)

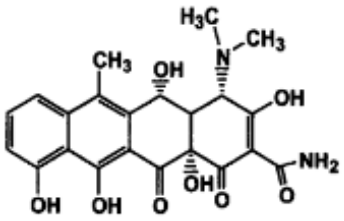
perturbazione membrane,
→ β-galattosidasi/LDH

oxytetraciclina



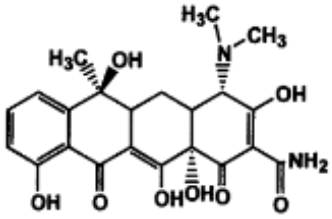
H+

$-H_2O$



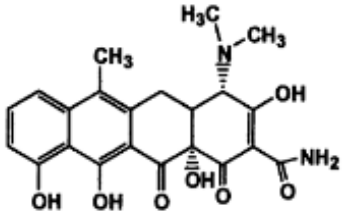
I

tetraciclina



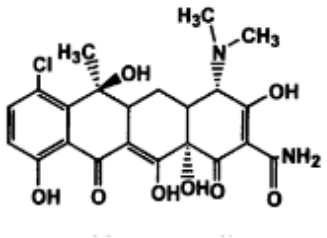
H+

$-H_2O$



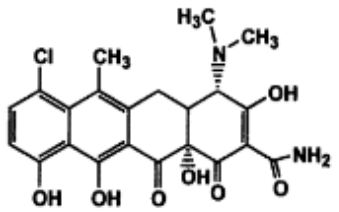
< attività
fototossicità,
epatotossicità,
induttori anemie

clorotetraciclina



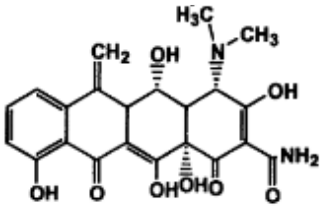
H+

$-H_2O$



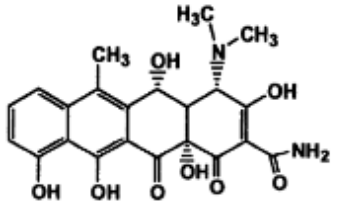
III

metaciclina

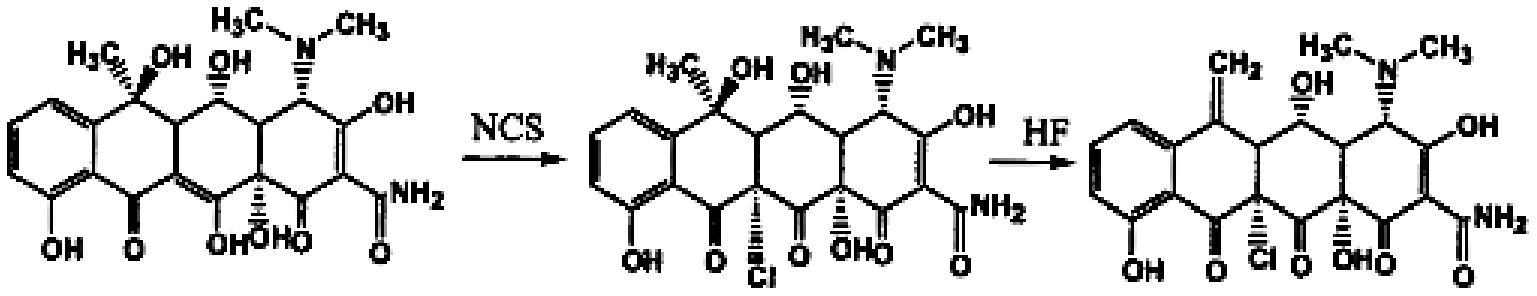


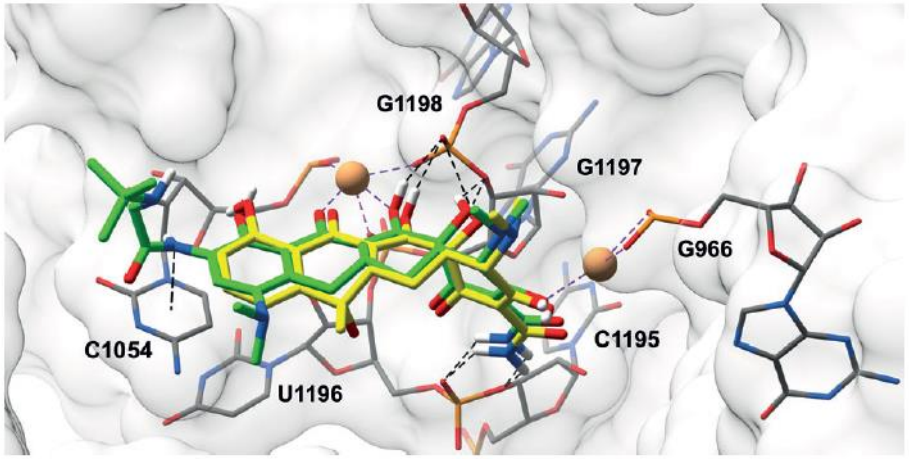
H+

$-H_2O$



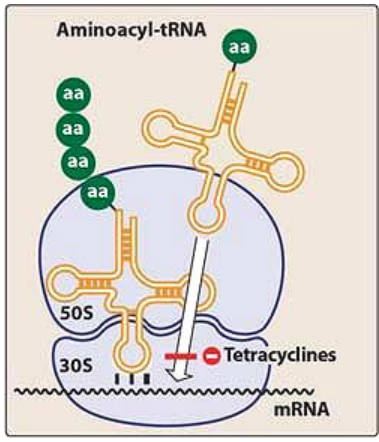
IV


NCS
HF
oxytetraciclina
I
II
11a
NaHSO3
metaciclina

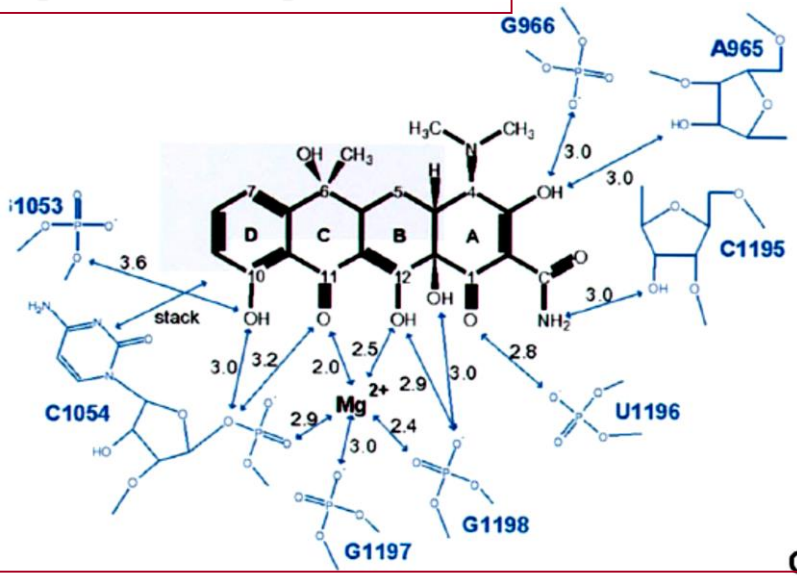


Overlay of tetracycline (yellow) and tigecycline (green) bound to the 30S ribosomal subunit from Thermus thermophilus. Tigecycline makes additional stacking interaction with C1054 compared to tetracycline.

Penetrazione parete G-neg diffusione passiva (porine OmpF (Outer membrane proteins) e OmpC) come complessi M^{++}. Periplasma → liberazione TC penetrazione membrana interna trasporto attivo energia-dipendente. Citoplasma ricostituzione complessi M^{++} →specie attive



Molecola capace di produrre interazioni ione-ione, idrofobiche e stacking con diversi gruppi funzionali presenti.
Due siti di binding nella sub-unità ribosomiale 30S: **1**) sito primario vicino a quello accettore per tRNA aminoacilato (sito A); **2**) sito secondario, interfaccia tra tre domini RNA nel corpo principale e subunità ribosomiali.
Resistenza: > 40 *tet/otr* → enzimi modificanti tet, proteine efflusso, proteine protezione ribosomiale

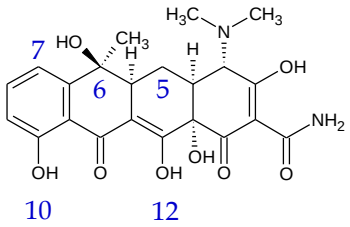


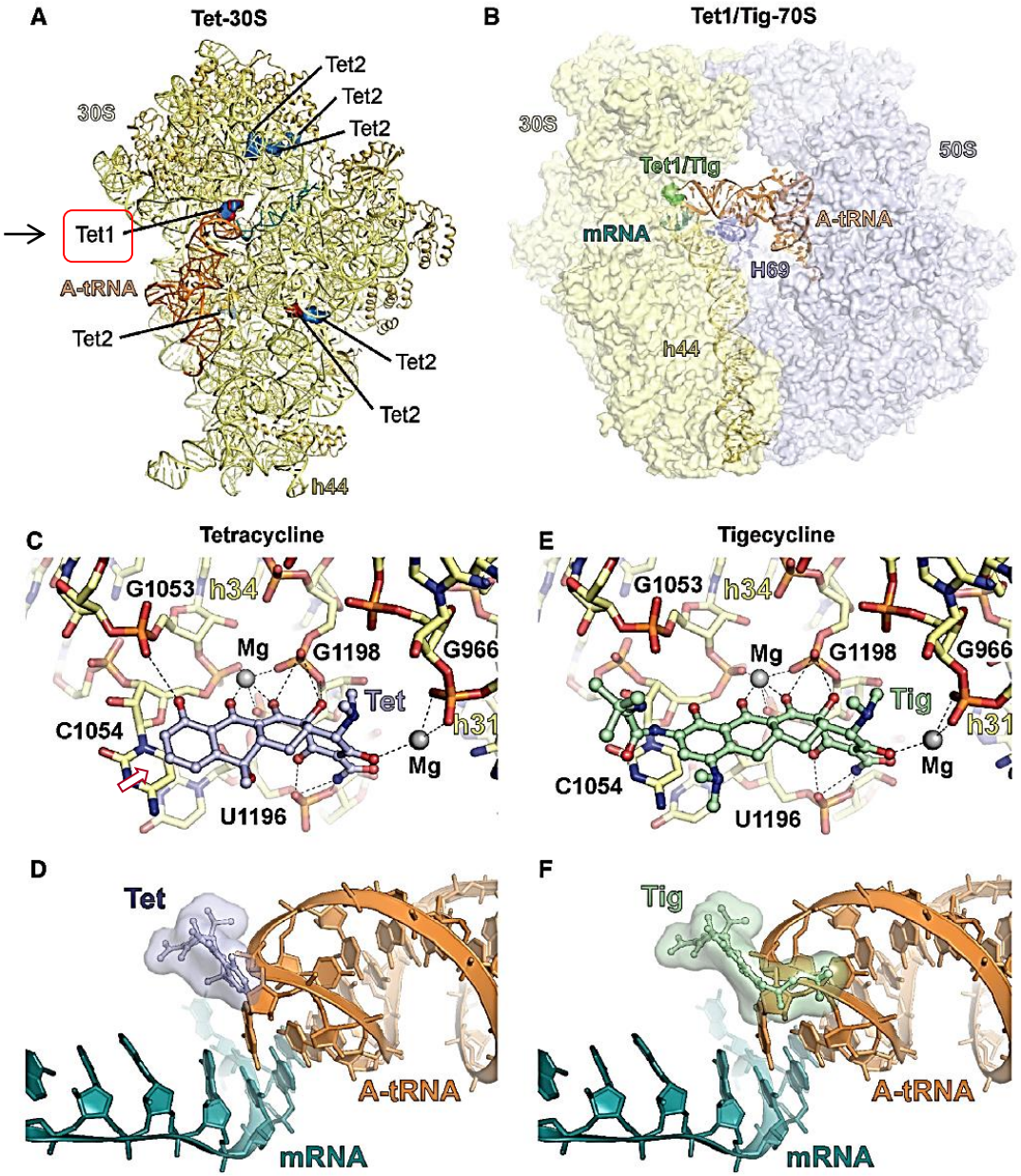
Il meccanismo di alcune tetracicline (minociclina) è ribosoma-indipendente ma ancora associato all'inibizione sintesi proteica

Proc. Natl. Acad. Sci. USA 2013, 110, 3812 – 3816. Cell 2000, 103, 1143-1154.

X-ray structures of tetracycline in complex with the *Thermus thermophilus* 30S subunit

primary binding site →





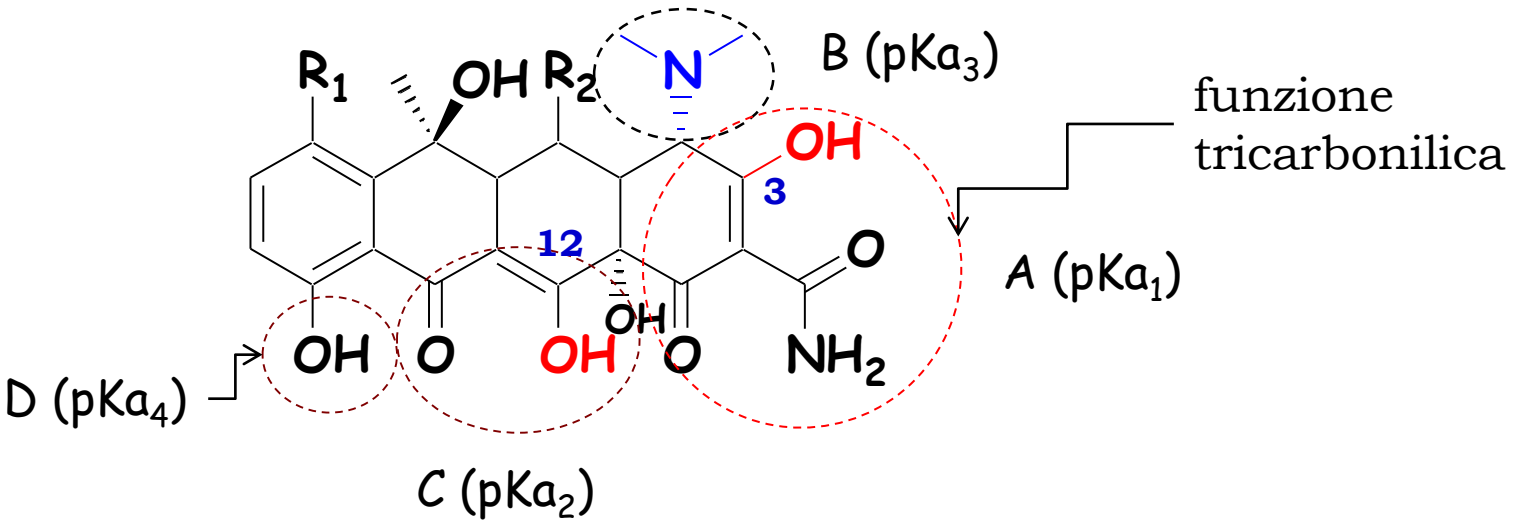
Tetracycline antibiotics and resistance mechanisms. Biol. Chem. 2014; 395(5): 559–575

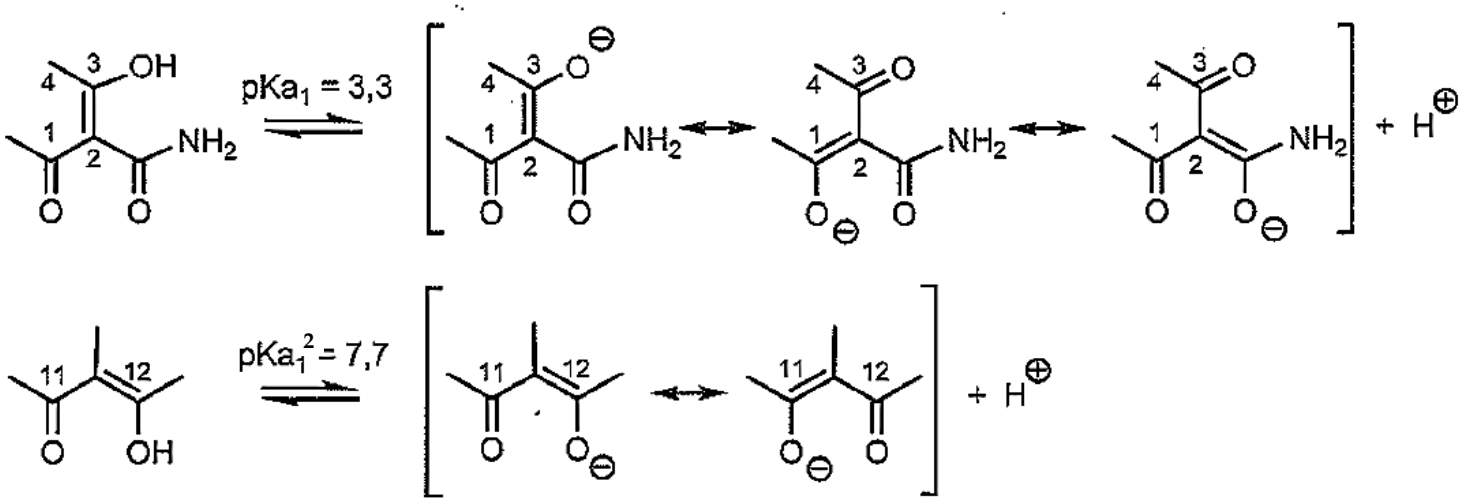
Patologie infettive sostenute da

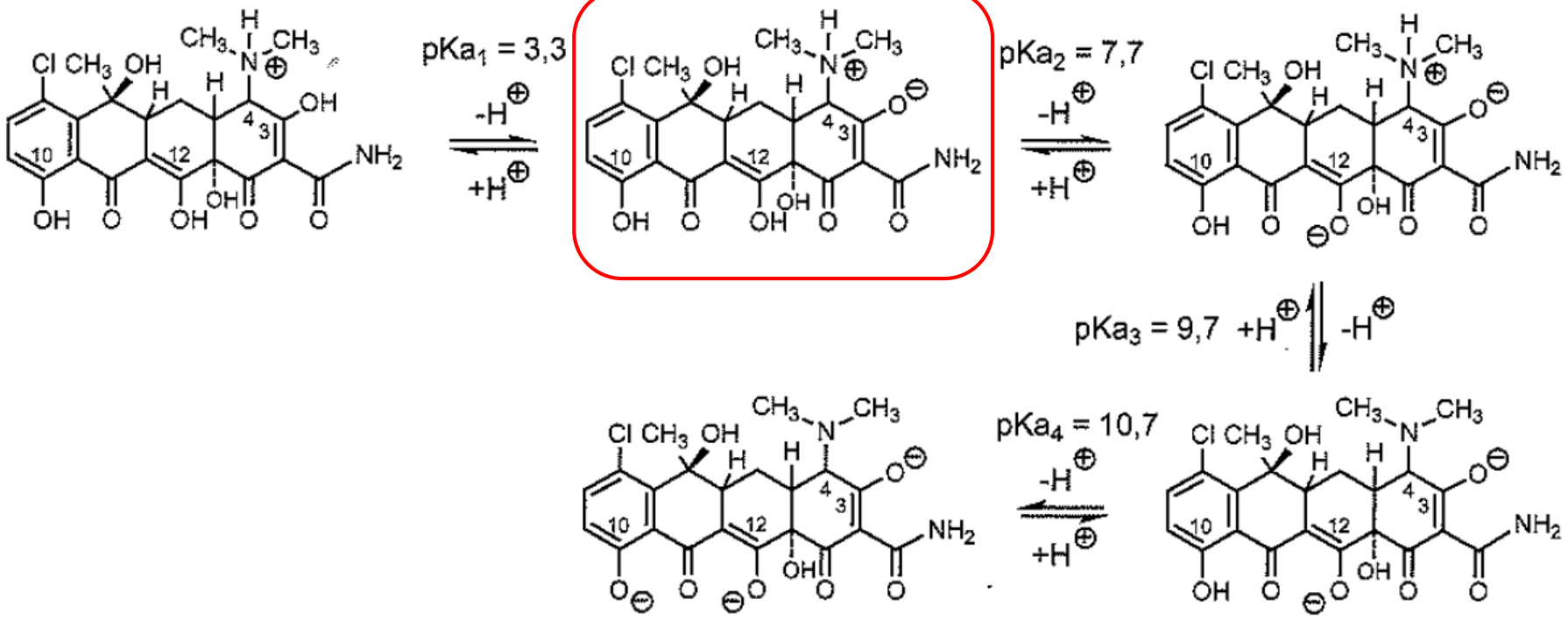
- *Rickettsiae;*
- *Mycoplasma pneumoniae;*
- *Borrelia recurrentis;*
- *Haemophilus ducreyi;*
- *Yersinia pestis;*
- *Bacterioides;*
- *Vibrio cholerae;*
- *Neisseria;*
- *Treponema;*
- *Clostridium;*
- *Bacillus anthracis;*
- *Chlamydia;*
- profilassi malaria

Streptococcus
amebiasi
acne severa
E. coli
Enterobacter aerogenes
Shigella
Klebsiella

Gram neg, Gram pos;
Alta frequenza di resistenza; a) proteine codificate da geni *tet* (tetraciclina) e *otr* (ossitetraciclina) che modificano chimicamente le TC; b) geni che codificano per proteine di efflusso; c) geni che codificano per proteine citoplasmatiche di protezione del ribosoma (distruzione allosterica del sito di binding delle TC) tossicità scarsa: nausea, alterazioni flora intestinale (micosi, avitaminosi), fotosensibilizzazione, colorazione gialla denti


R1
OH
R2
N
B (pKa3)
funzione
tricarbonilica
OH
3
12
O
A (pKa1)
OH
O
OH
O
OH
NH2
D (pKa4)
C (pKa2)


pKa1 = 3,3
pKa1 2 = 7,7
NH2
OH
+ H⊕


pKa1 = 3,3
-H⊕
+H⊕
pKa2 = 7,7
-H⊕
+H⊕
pKa3 = 9,7 +H⊕
-H⊕
pKa4 = 10,7
-H⊕
+H⊕

Proton and metal ion binding of tetracyclines

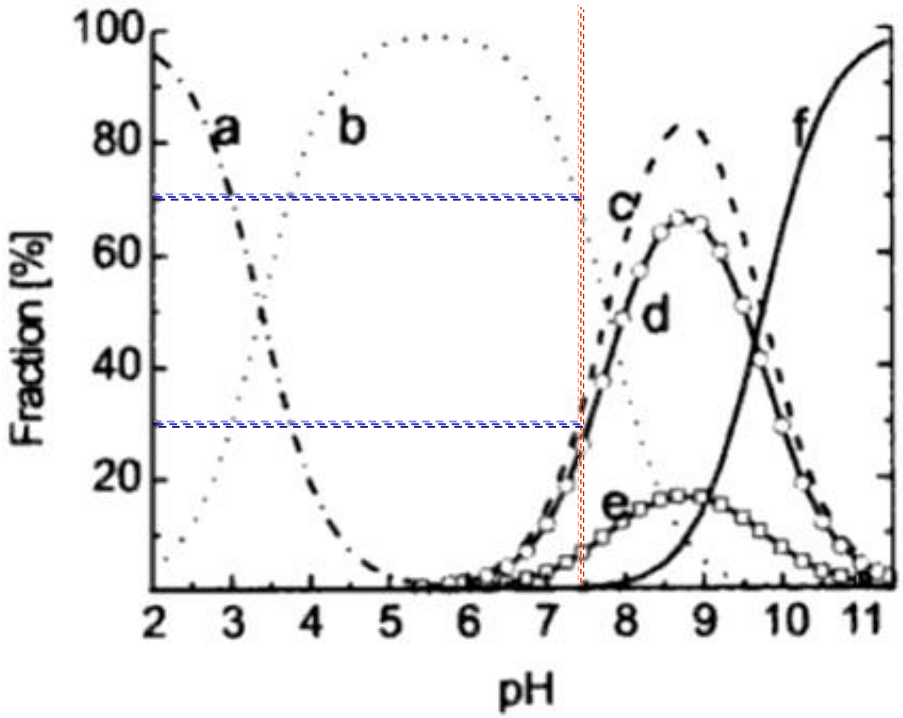
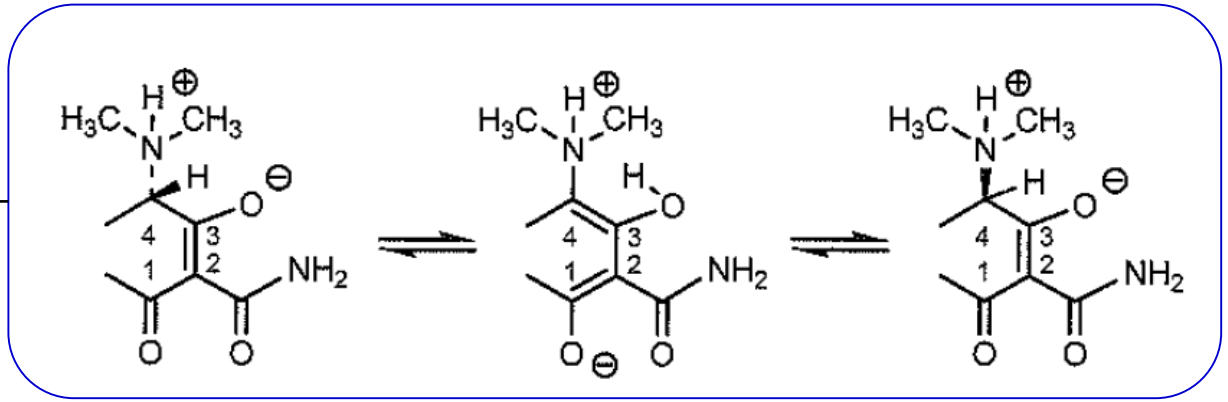
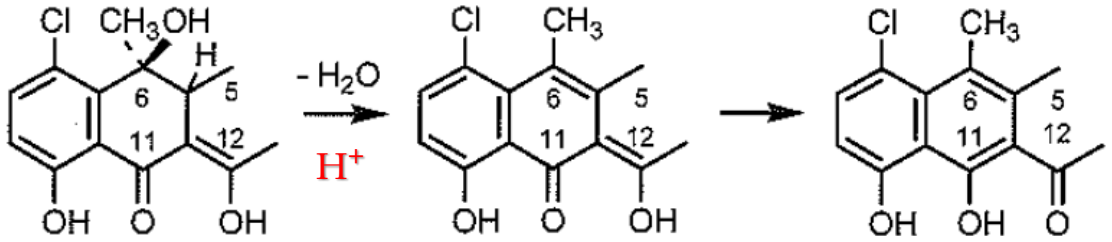


Figure 5. Relative amount F of the different species found in an aqueous solution of variable pH: LH_3^+ (a), $LH_2^{\pm}$ (b), LH^- (c), $LH^{=}$ (f); LH^- can be present as $a^-b^+c^-$ (trace d) or a^-bc (trace e). The calculation is based on the microscopic acidity constants given in reference [3].

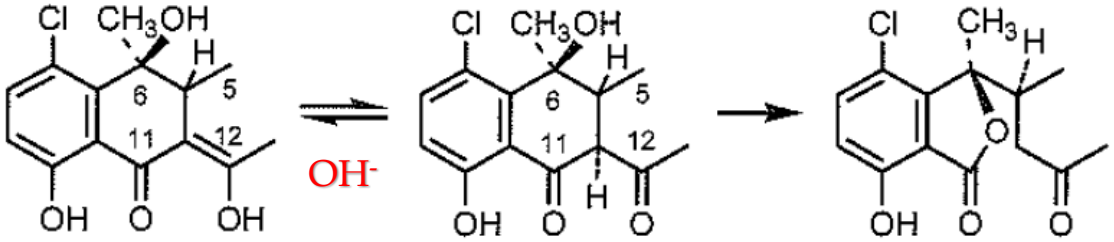
a) $^{75}LH_3^+$
b) LH_2^*
c) LH^-



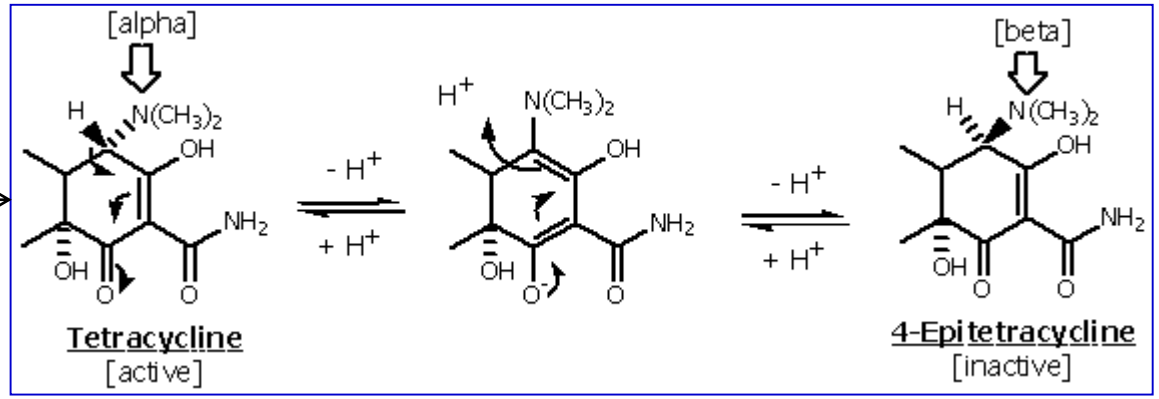
epitetraciclina
(inattiva)
reazione fotochimica
rapida



anidro tetraciclina
(inattiva)



isotetraciclina
(inattiva)

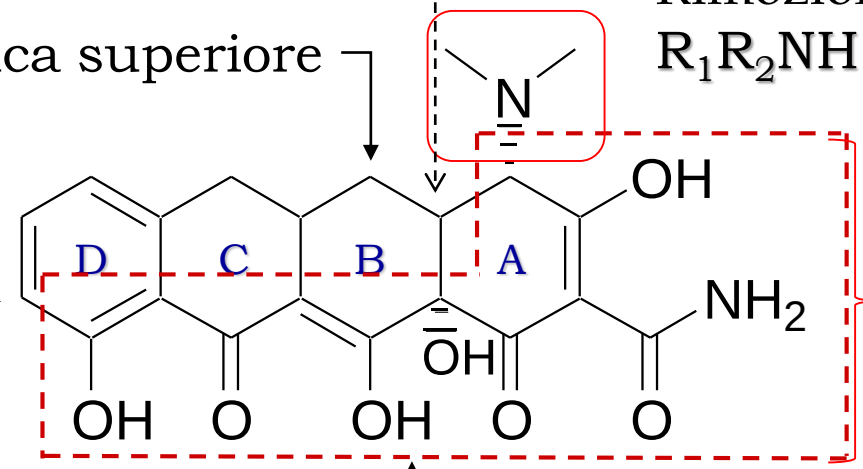


SAR

Stereochimica α importante

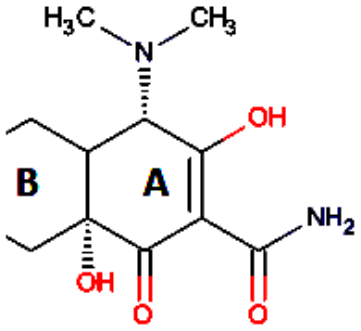
Rimozione ↓;
R_1R_2NH ↓

regione periferica superiore

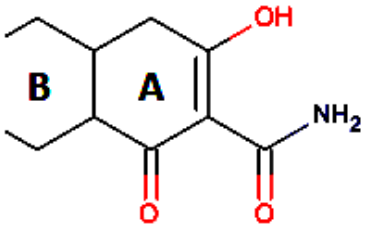


RCHO/RCN ↓;
Pt/Pd ↓

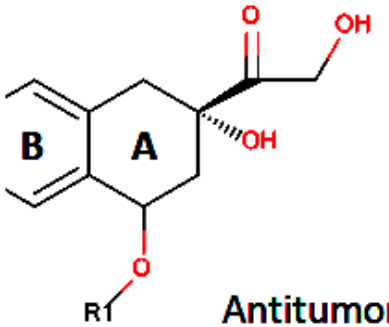
Substituents and conformation on ring A are the most important to determine the use in therapy:



Antibacterial



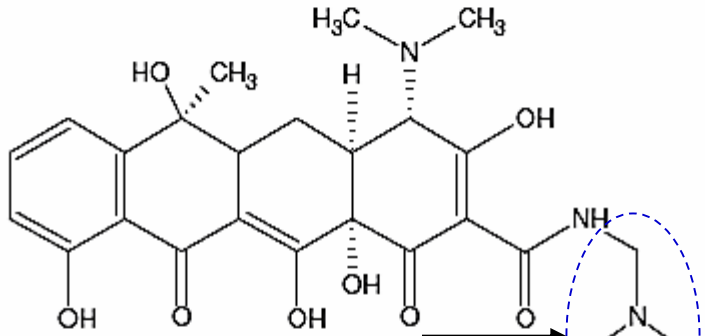
Antifungal



Antitumor

Tetracicline semisintetiche
DERIVATI idrosolubili

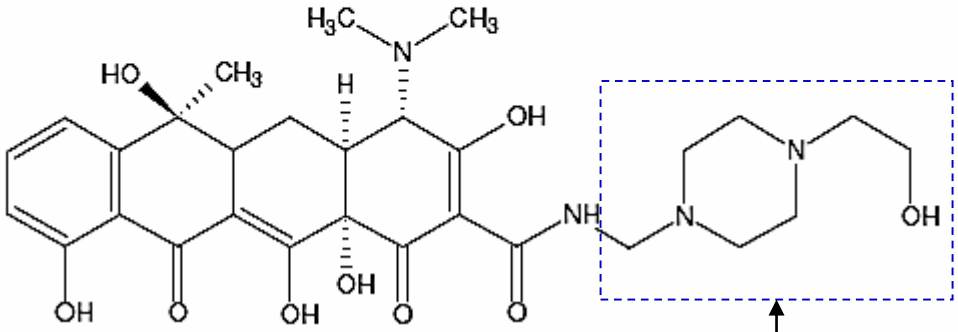
Basi di Mannich all'azoto



metilen-pirrolidina

Rolitetraciclina
idrosolubile, somministrazione parenterale

Colbiocin (+cloramfenicolo, tetraciclina)

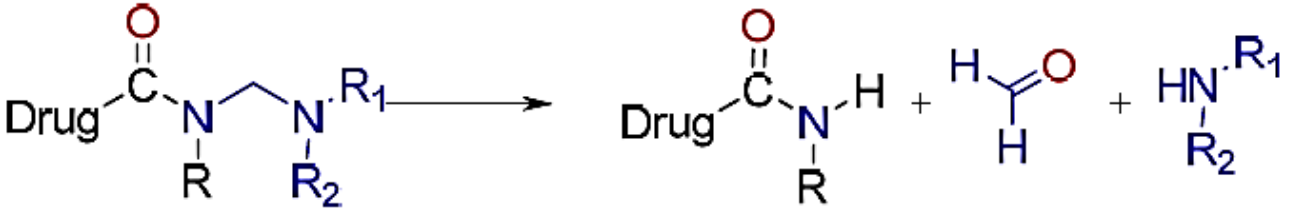


2-(4-metilen-piperazin-1-yl)-etanolo

Mepiciclina
sale con fenossimetilpenicillina

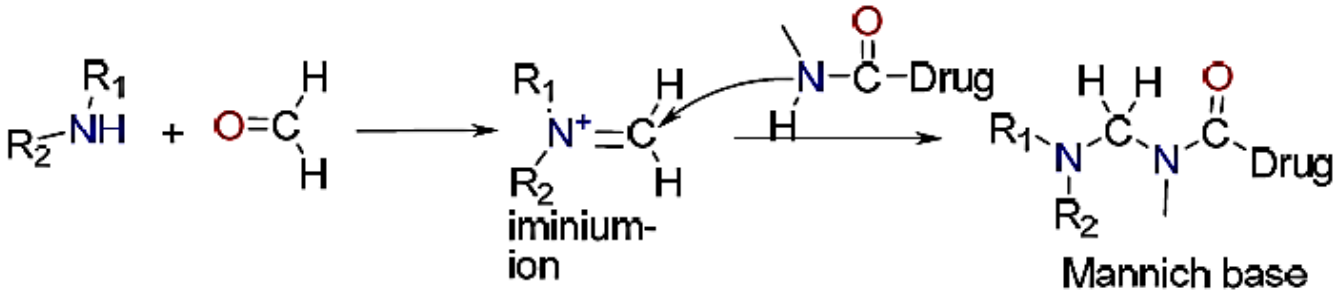
Mannich bases as Prodrugs

- N-Mannich base prodrugs can be applied to both -NH acids (e.g. amides) and amines, and undergo bioconversion to release the parent -NH acid or amine and an aldehyde by chemical hydrolysis in aqueous, alkaline, and slightly acidic solutions.



Mannich base

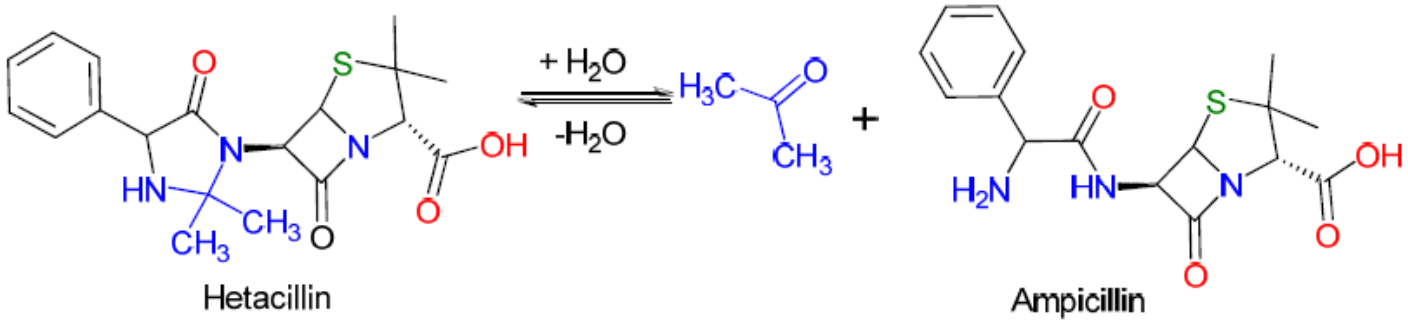
- Mannich reaction is nucleophilic addition reaction of an aldehyde and any primary or secondary amine to produce resonance stabilized schiff base (iminium ion). The addition of a carbanion from a CH acidic compound (enolizable carbonyl compound, amide, carbamate, hydantoin or urea) to the schiff base gives another base called the Mannich base.



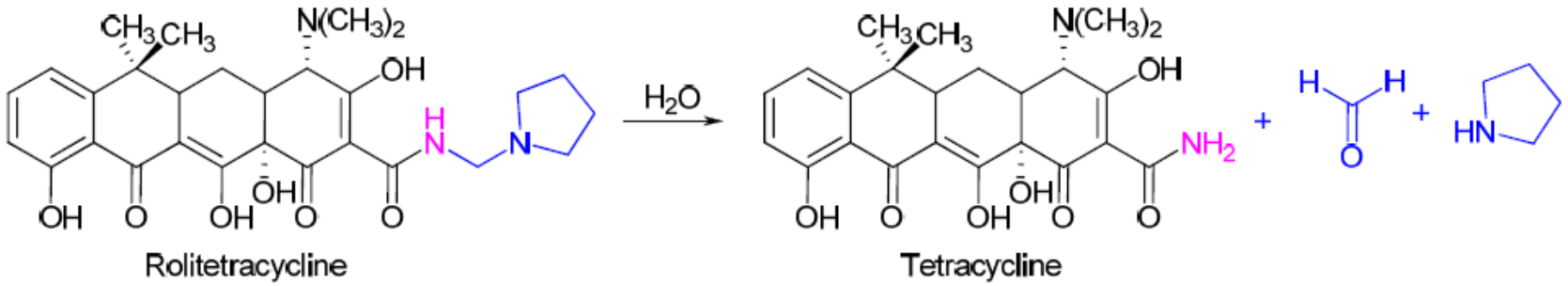
Mannich base

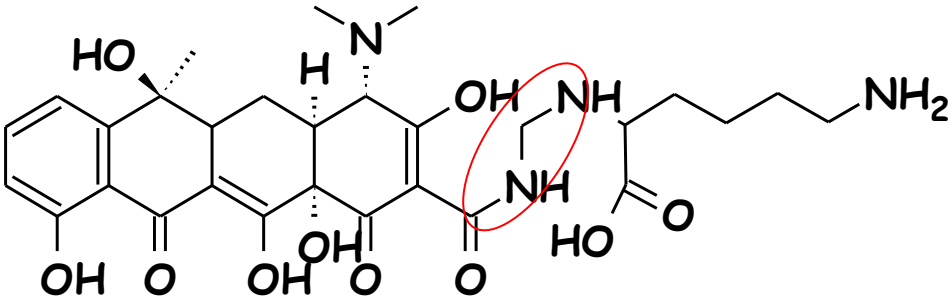
Mannich Base Prodrugs

- Hetacillin is rapidly hydrolyzed to ampicillin in aqueous solutions and in vivo (U. Klixbull and H. Bundgaard, *Int. J. Pharm.* **23** (1985), pp. 163–173)



- Rolitetracycline liberates tetracycline in vivo



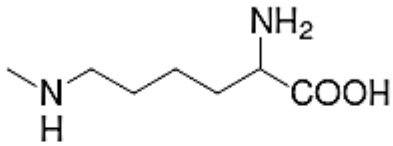


Limeciclina

(Tetralysal)

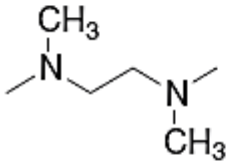
Acne infiammatoria da moderata a severa, infiammazione nell'acne mista.

Limeciclina (tetraciclina metilen-lisina)



Etamociclina

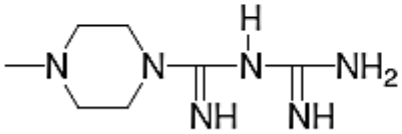
(N,N'-dimetiletilendiammino-ditetraciclina)



(tra 2 nuclei di tetraciclina)

Guameciclina

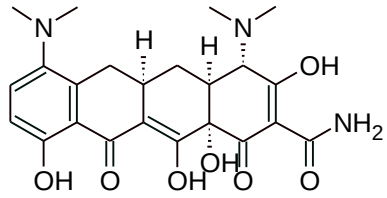
(derivato biguanidico)



Tigeciclina (*Tygacil*)

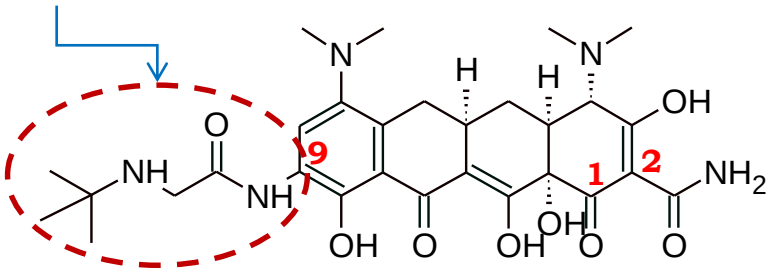
III^ generazione
glicilcicline

- MRSA: *S. aureus resist. Meticillina*
- VRSA (VISA): *S. aureus resist. Vancomicina*
- ORSA: *St. aureus resist. Oxacillina*
- CREC*: E. coli resist. Ciprofloxacina*
- CRKP: *Klebsiella spp. resist. Cftazidime*
- *patogeni produttori di KPCs e MBLs (resist. ac.clavulanico e tazobactam)*



minociclina

N-ter-butil-glicinammide



appr. FDA 2005 infez. cutanee severe ed intra-addominali;
appr. FDA 2009 polmoniti

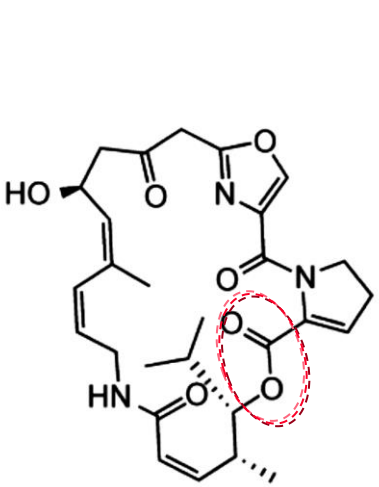
➢Interazione reversibile alla subunità 30S (cfr TRC, blocco ingresso aminoacil-t-RNA al sito di binding A ribosomiale);
➢geni *tet* (resist. TRC) e *otr* (resist. oxiTRC) →proteine→ a) dissociaz. TRC dai siti ribosomiali, b) trasporto attivo (efflusso dalla cellula batterica)
➢La TGC si lega 5 volte più tenacemente delle TRC ai ribosomi ed è insensibile ai sistemi di efflusso (ingombro sterico N-t-butil-glicinammide ?)

Clinical Therapeutics/Volume 28, Number 8, 2006

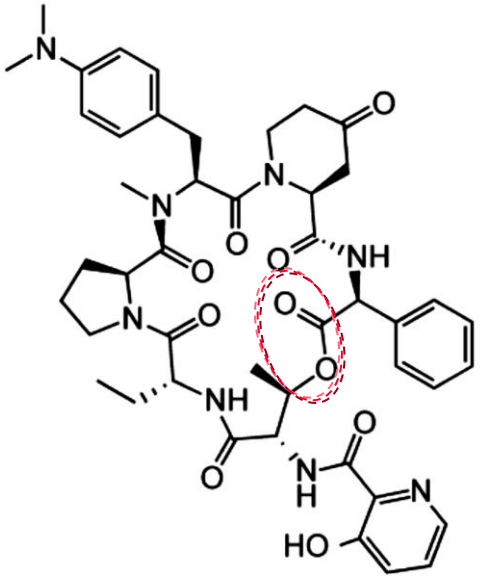
- Ribosoma batterico, processo di traslazione proteica;
- Amminoglicosidi;
- Macrolidi;
- Tetracicline;
- **Streptogramine; Cloramfenicolo;**
- Oxazolidinoni.

Streptogramine (Streptomyces genus)

- caratterizzate dalla coesistenza di due componenti (A e B) strutturalmente non correlati.
- gruppo A (~70%) sono macrolattoni polinsaturi, con struttura ibrida peptide-poliketide, (pristinamicina II A).
- gruppo B (~30%) sono macrolattoni peptidici ciclici. (pristinamicina I A).



Pristinamycin IIa
Streptogramin A



Pristinamycin Ia
Streptogramin B

- sinergistine (16volte).
- gram-positivi, poco usate, anche perché non era disponibile una formulazione parenterale da utilizzare nelle infezioni gravi.
- utilizzati in passato solo due farmaci orali: la virginiamicina e la pristinamicina.
- sviluppato derivato semisintetico iniettabile e.v.: *Synercid*
  - quinupristina, derivato della pristinamicina IA (30 parti)
  - dalfopristina, derivato della pristinamicina IIA (70 parti) (uso ospedaliero).

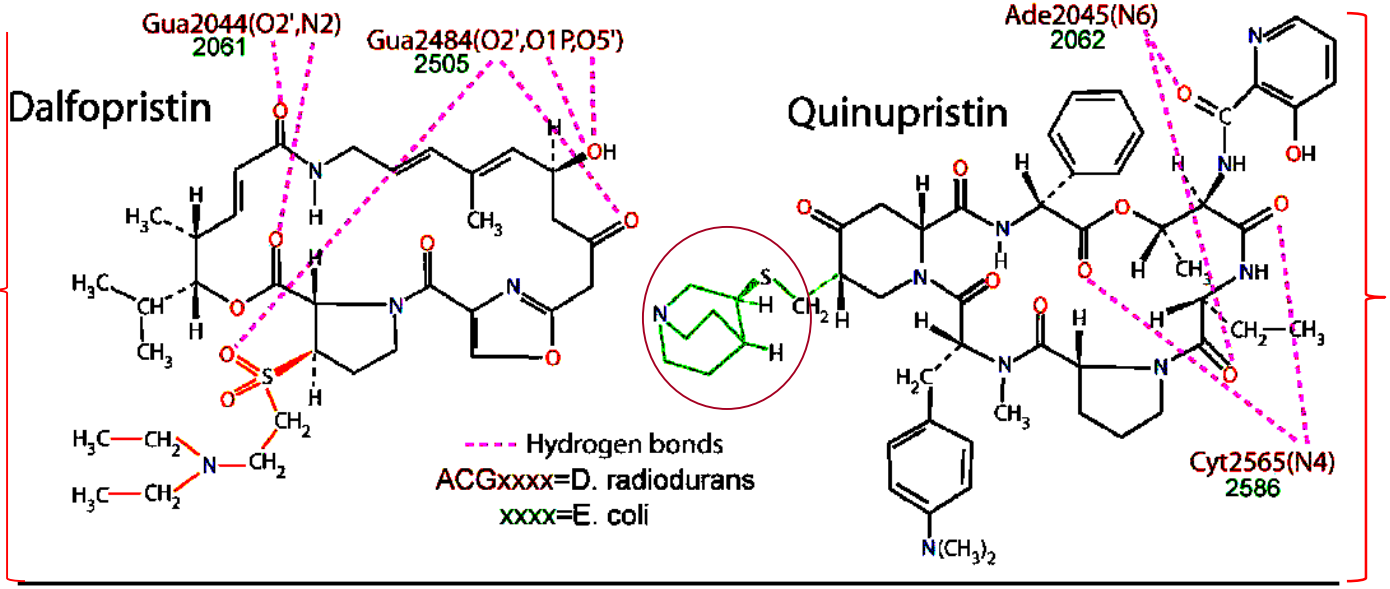
cicloesadepsipeptidi

Streptogramine tipo A

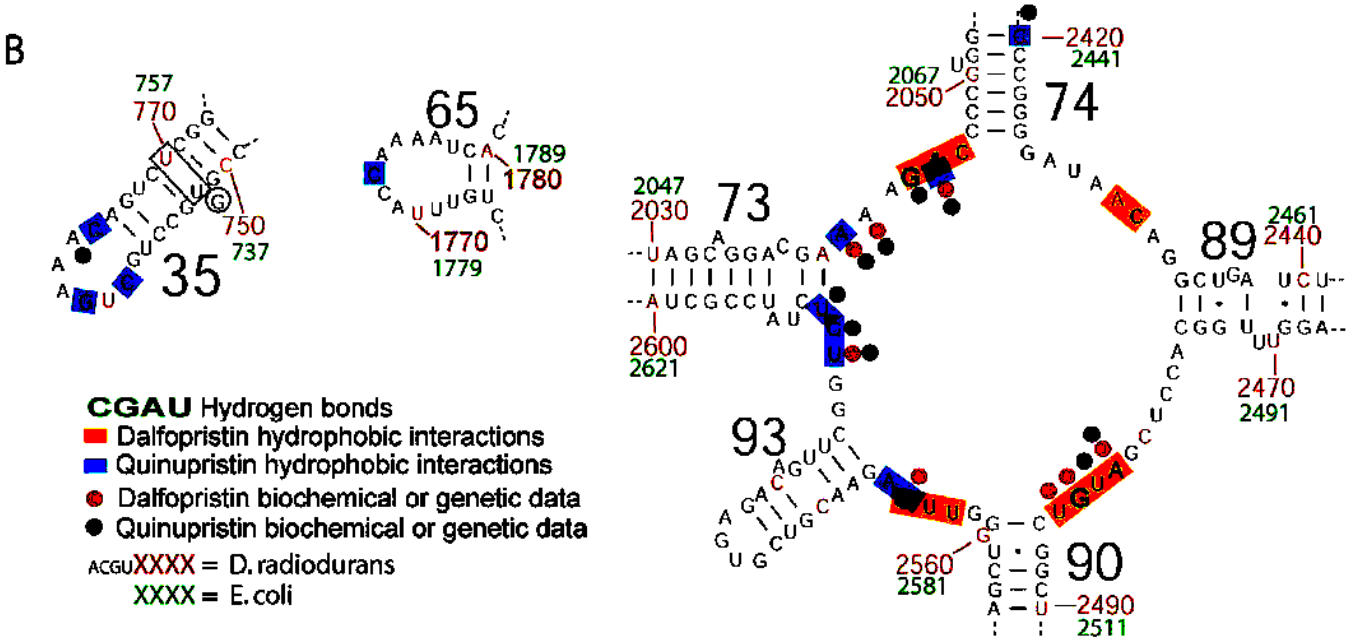
Streptogramine tipo B

Inibizione diretta PTC. (A, P) competiz. substrato

Attività batteriostatica prolungata per perturbazione stabile PTC

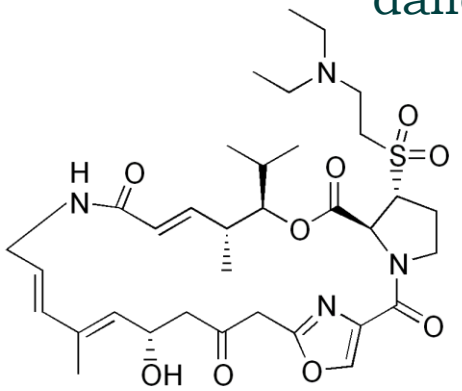


50S, tunnel elongazione (cfr. macrolidi, resistenza MLSB, A2058)

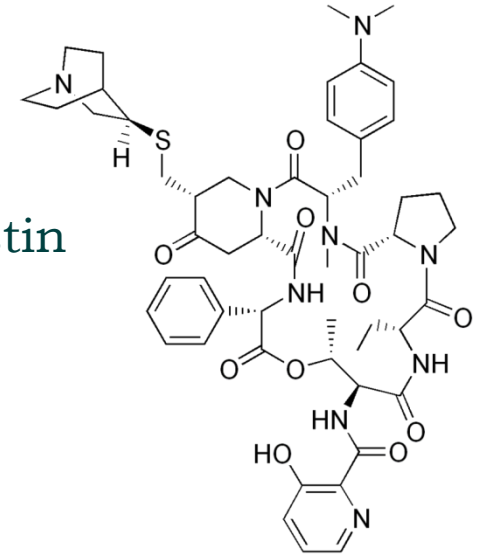


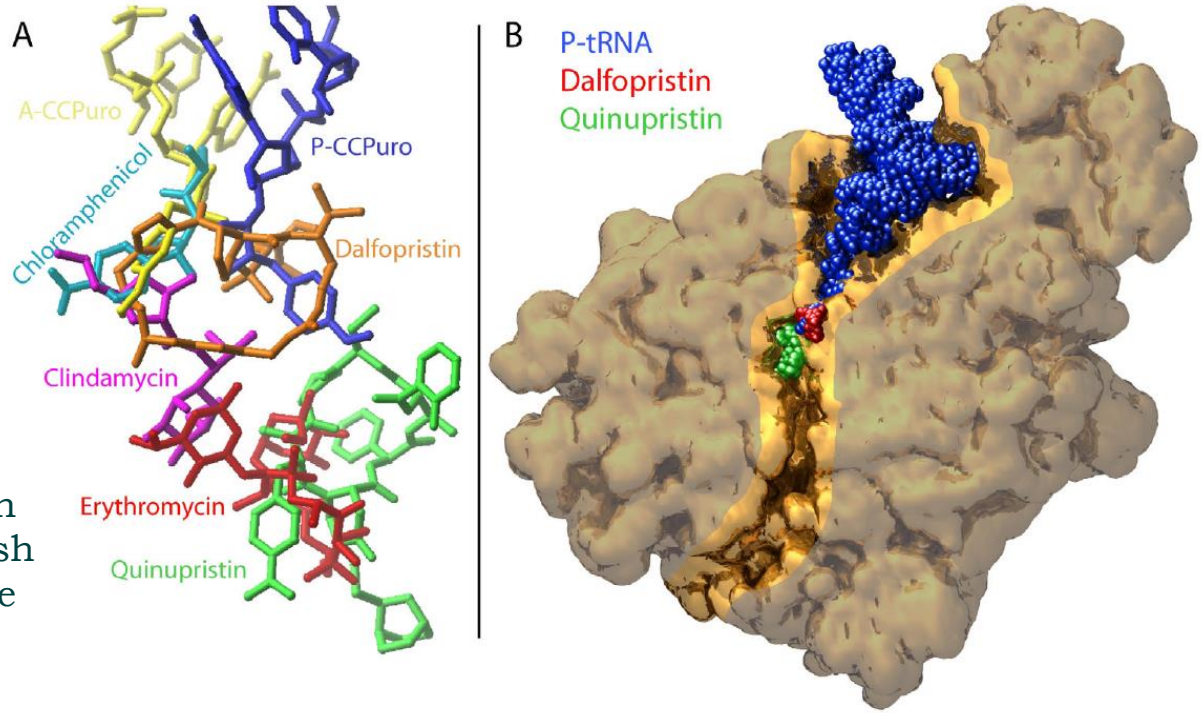
23S rRNA

dalfopristin



quinupristin





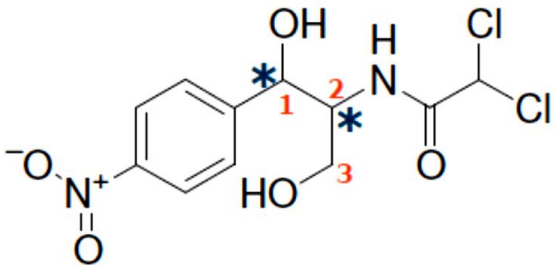
erythromycin can completely abolish SB binding to the ribosome,

Utilizzazione delle streptogramine

- La combinazione quinupristin-dalfopristin è attiva soprattutto contro i batteri gram-positivi quali:
  - *Staphylococcus aureus* (sia sensibile che resistente alla meticillina)
  - stafilococchi coagulasi-negativi (anche MRSA)
  - streptococchi (compreso *Streptococcus pneumoniae* resistente a penicillina e macrolidi)
  - enterococchi (anche VRE), *E. faecium*).
- Quinupristin-dalfopristin è attivo anche contro alcuni batteri gram-negativi e intracellulari che spesso rappresentano patogeni respiratori: *Neisseria meningitidis*, *Haemophilus influenzae*, *Moraxella catarrhalis*, *Mycoplasma pneumoniae*, *Legionella* spp. e *Chlamydia* spp.
- Quinupristin-dalfopristin infine mostra ottima attività in vitro contro anaerobi sia gram-positivi che gram-negativi: *Bacteroides*, *Prevotella*, *Fusobacterium*, *Clostridium*, *Actinomyces*, *Peptostreptococcus*…

CLORAMFENICOLO

Prodotto da *Str. venezuelae, Str. lavendulae* ecc.



- ✓ Biogenesi da fenilalanina
- ✓ Sintesi totale
- ✓ Sostanza neutra, poco solubile in acqua

1-*p*-nitrofenil-2-dicloroacetammido-1,3-propandiolo

Primo AB ad ampio spettro usato negli USA (1947), per un lungo periodo ha avuto largo impiego. Gravi discrasie ematiche ne hanno ridotto l'uso, facendone un AB di seconda scelta.

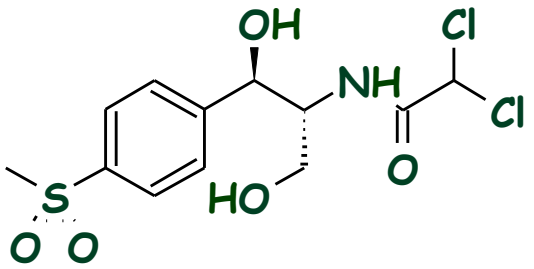
Spettro AB

Ampio, soprattutto su gram- (Tifo, Coli, pertosse, proteus ecc.); rickettsie, clamidie (psitaccosi, linfogranuloma)

Meccanismo d'azione

Si lega alla peptidil tranferasi (subunità 50 S) del ribosoma batterico, inibisce il trasferimento della catena peptidica in crescita al sito accettore dell'aa-tRNA

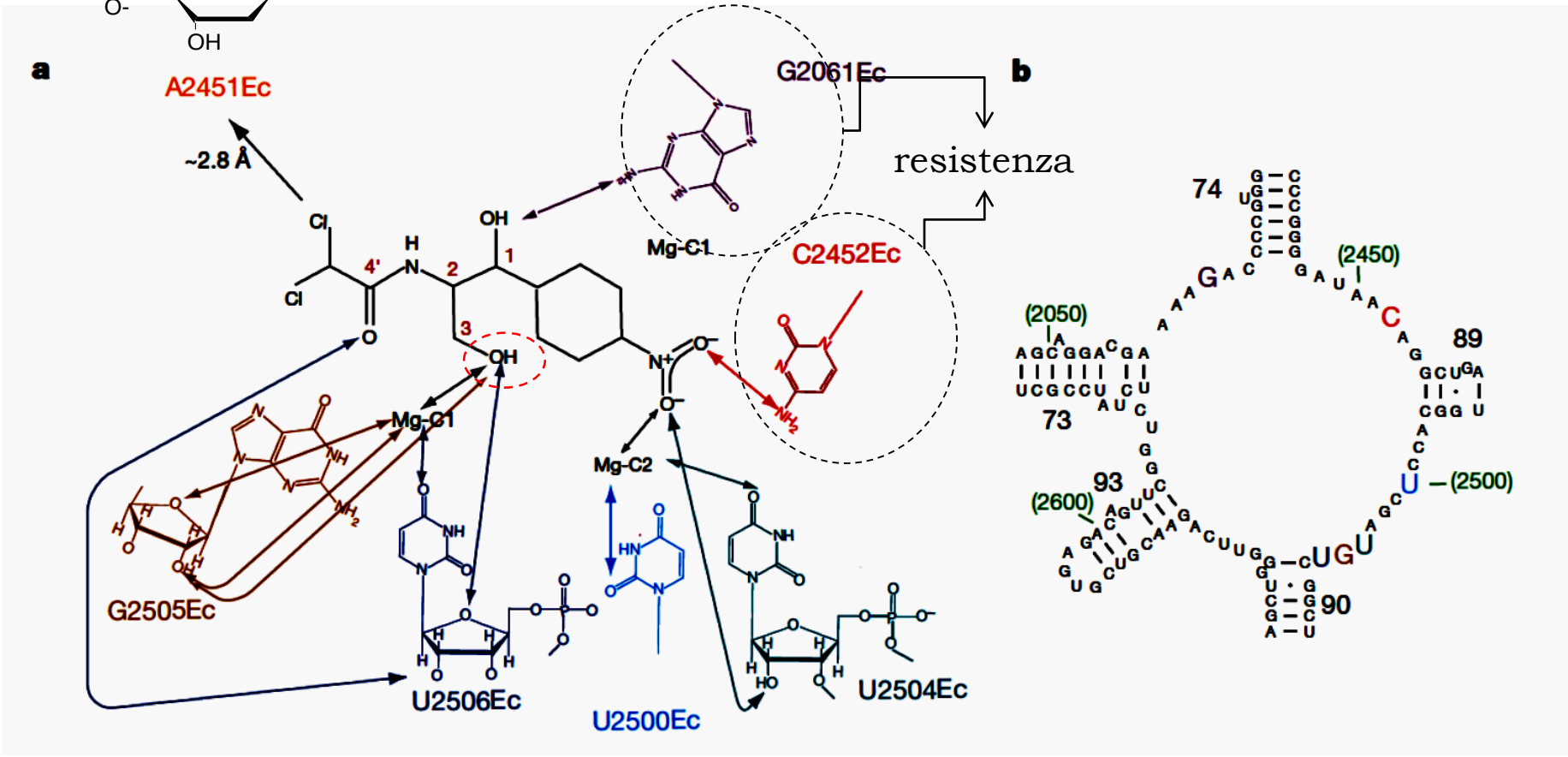
TIAMFENICOLO (*Fluimicil*)



D-d-treo-2-dicloroacetamido-1-(4-metilsulfonilfenil)-1,3-propandiolo;

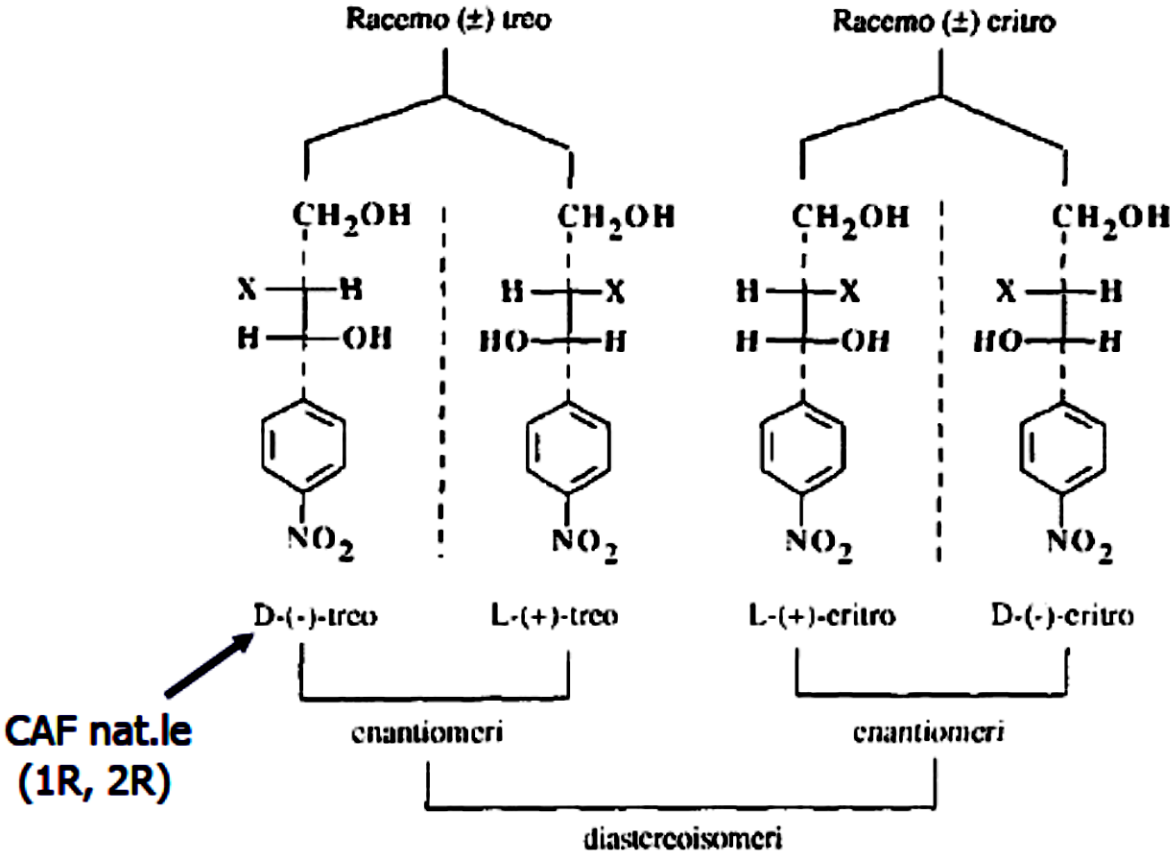
Tiamfenicolo è escreto essenzialmente immodificato nelle urine; indicato nelle infezioni batteriche respiratorie da germi sensibili al tiamfenicolo, nelle quali la presenza di mucostasi complica o rallenta l'evoluzione clinica. Bronchiti acute e croniche. Broncopolmoniti e polmoniti a lenta risoluzione. Ascessi polmonari. Enfisema ostruttivo.

Cloramfenicolo blocca l'attività peptidiltransferasica (PTC) tramite binding al sito A (tRNA).



The Ribosomal Peptidyl Transferase Center: Structure, Function, Evolution, Inhibition. Critical Reviews in Biochemistry and Molecular Biology, 40:285–311, 2005

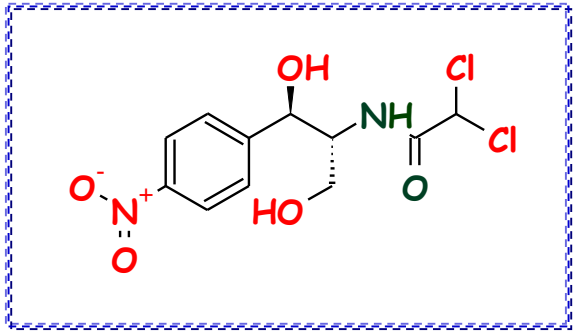
Diastereoisomeri del CAF



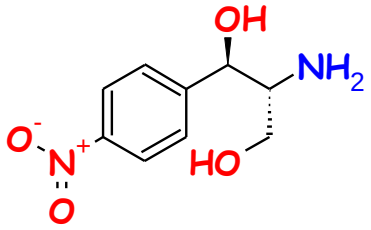
Coppia	Forma e configurazione relativa (Fenilalanina)	Configurazione assoluta	Attività antibatterica
Eritro	D (-)	1S, 2R	0
	L (+)	1R, 2S	2
Treo	**D (-): CAF naturale**	**1R, 2R**	**100**
	L (+)	1S, 2S	0

Cloramfenicolo

(hl: 1.6-4.6hrs, >neonati)



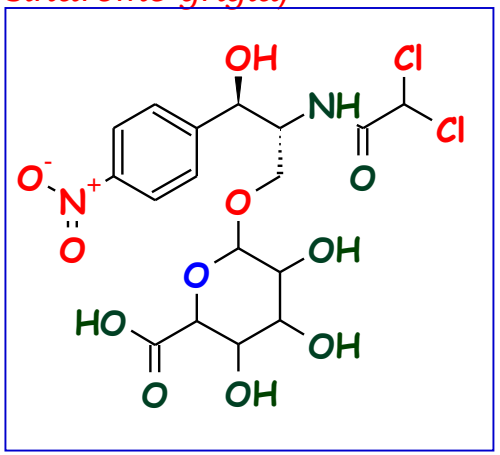
Cloramfenicolo base (26%)



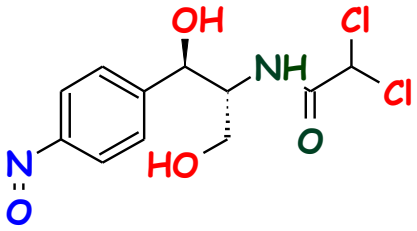
Cloramfenicolo glucuronide (> metab)

Glucuronosiltransferasi

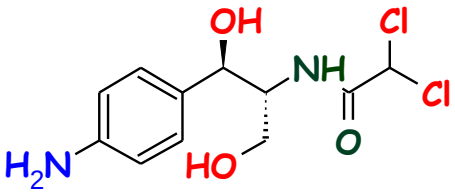
(sindrome grigia)



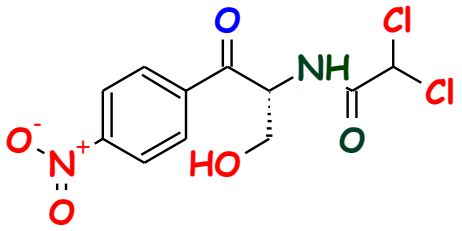
Nitroso C.



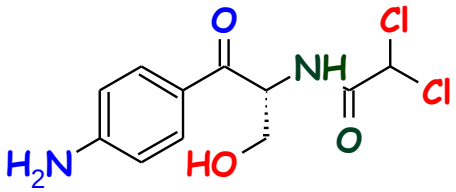
Ammino (intestino, 20% N-acetil deriv.)



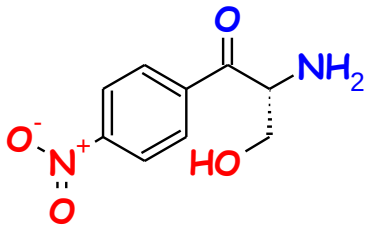
Deidro C.



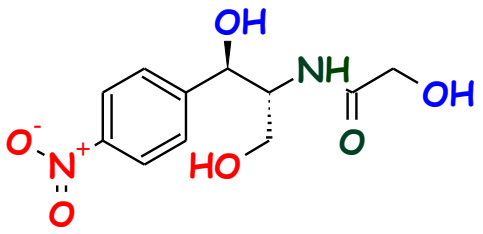
Amminodeidro C.



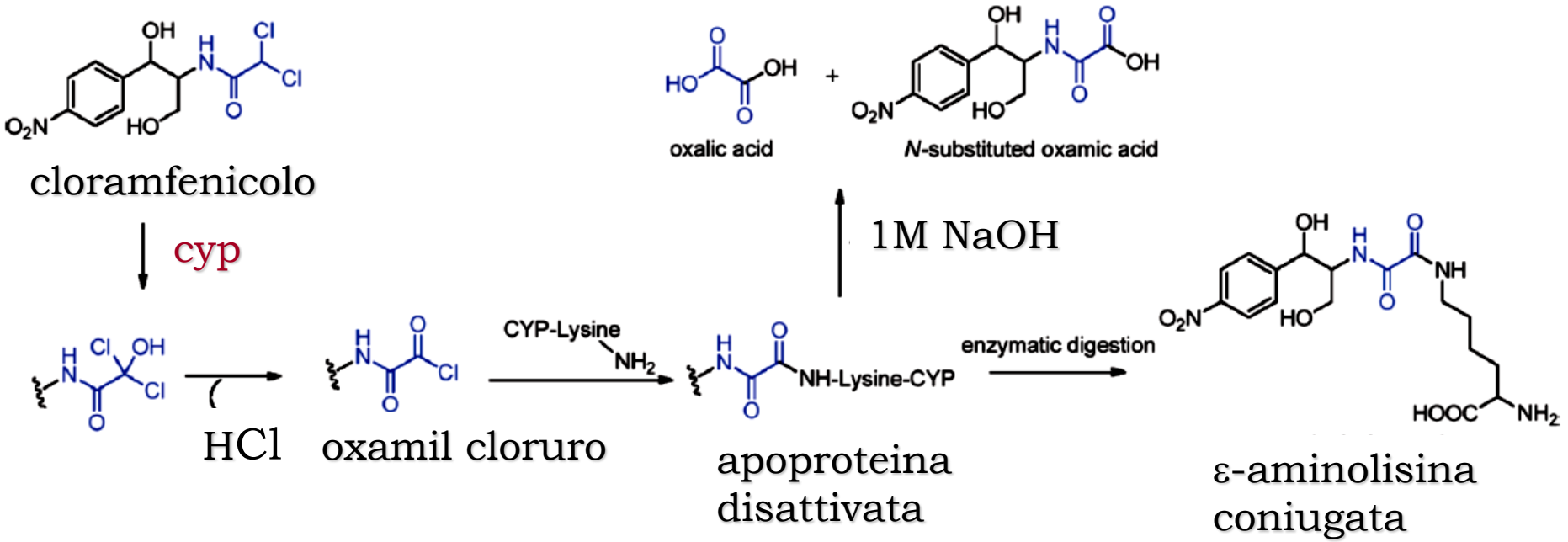
Deidro C. base



Alcol C.



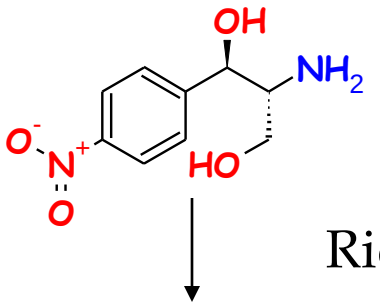
e 3. CYP Apoprotein Acylation by the Broad-Spectrum Antibiotic Chloramphenicol



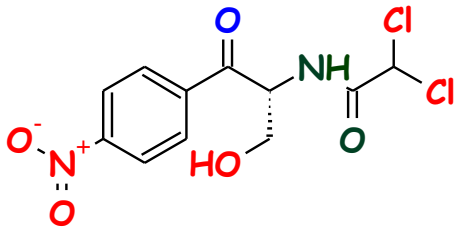
Il Cloramfenicolo è stato uno dei primi inattivatori CYP scoperti (1982) capace di modificare irreversibilmente apoproteine (digestione proteolitica di enzima inattivato da [^{14}C]-cloramfenicolo → singolo aminoacido ^{14}C-modificato.
Il rilascio di lisina e derivati del cloramfenicolo (acidi ossalico ed N-oxamico) a seguito di idrolisi alcalina dell'aminoacido modificato e l'inattività (CYP) del difluorometil derivato, ha suggerito la declorinazione ossidativa a oxamil cloruro che acila la lisina e si idroliza ad acido oxamico.

Cloramfenicolo base

nitrophenylaminopropane



Deidro Cloramfenicolo



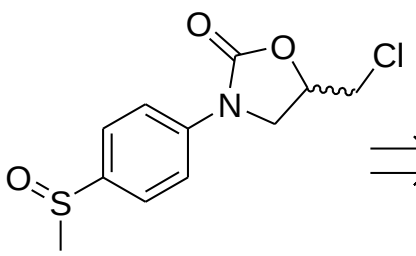
Riduzione (tessuto osseo)

Metaboliti sufficientemente stabili da raggiungere il midollo osseo in cui sono substrati per una nitroriduzione. I nitroso-metaboliti prodotti dal fegato sono ciotossici (DNA) in vitro ma poco stabili in vivo e per questo incapaci di raggiungere il midollo osseo. La presenza di nitroso-intermedi nel midolo osseo può interferire con la produzione di fattori di crescita ematopoietica ed indurre citotossicità (anemia aplastica e leucemia).

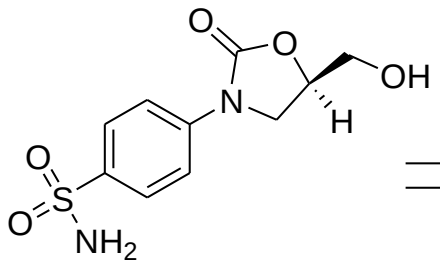
- Ribosoma batterico, processo di traslazione proteica;
- Amminoglicosidi;
- Macrolidi;
- Tetracicline;
- Streptogramine; Cloramfenicolo;
- **Oxazolidinoni.**

Oxazolidinoni

1978



4



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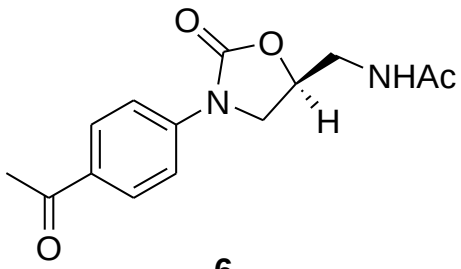
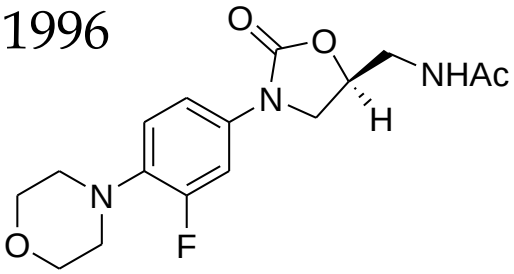


6

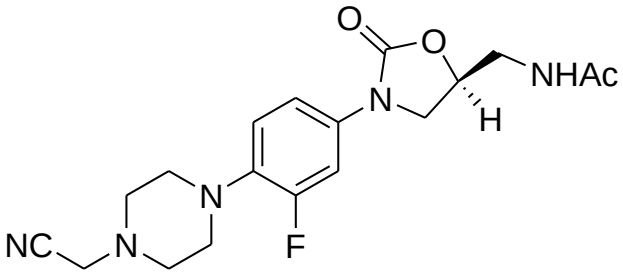
1987

Linezolid
(appr. 2000)

1996



1



7

Nuova classe strutturale di antibiotici con nuovo (unico) meccanismo d'azione). Fino al 2000 assenza di ceppi resistenti da isolati clinici (G+) [*Multicenter evaluation of linezolid antimicrobial activity in North America,2000*].

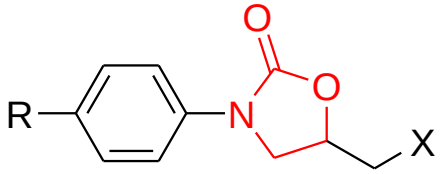
Linezolid è un inibitore della sintesi proteica che blocca il binding di N-formilmetionil-tRNA al ribosoma 70S.

Linezolid, formulato os e parenterale, è attivo verso MRSA e VRE incluse patologie infettive gravi dell'epidermide, osteomieliti, polmonite.

DuPont

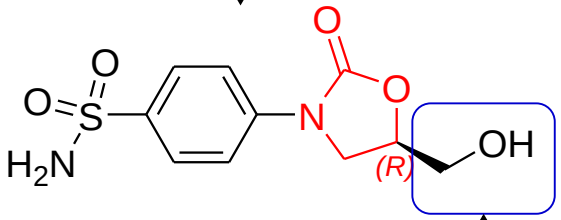
5-halomethyl-3-phenyloxazolidinones

only modest in vitro activity against several Gram**+/-** ; good in vivo efficacy against *E.Coli* in a mouse lethal infection model



R = CH_3SO^- ; X = Cl (**8**)
R = $CH_3SO_2^-$; X = Cl (**9**)
R = $CH_3SO_2^-$; X = Br (**10**)

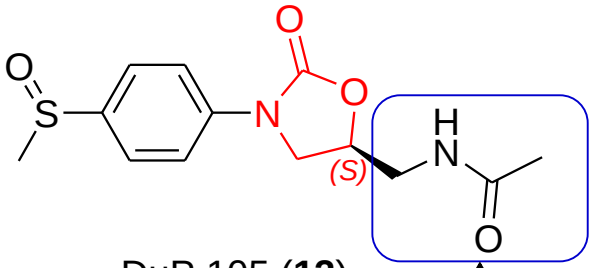
opt



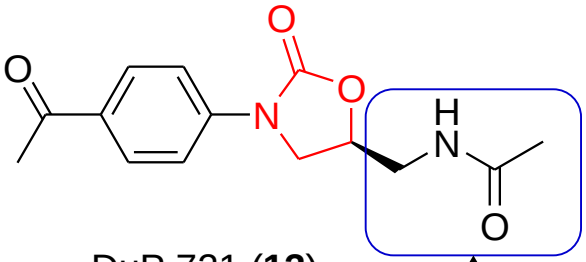
S-6123 (**11**)

hydroxymethyl

plant bacterial fungal diseases (8)
Staphylococcus pathogens in in vitro and in vivo animal models (9,10)



DuP 105 (**12**)



DuP 721 (**13**)

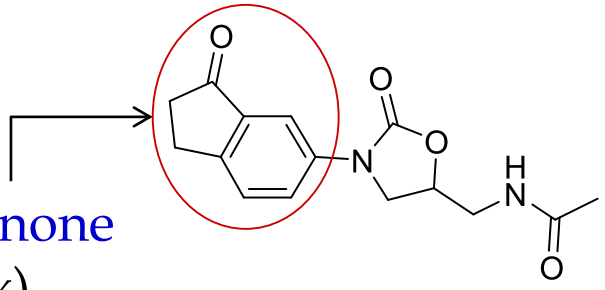
>>Staph., Enter. Strept. in in vitro and in vivo animal models; do not generate resistant mutants in vitro.

acetamidomethyl

Phase I → animal toxicity → lethal toxicity in rats when dosed at 100 mg/kg → oxazolidinone program at DuPont was terminated with the failure of the two drug candidates

Upjohn company (Pharmacia, now Pfizer)

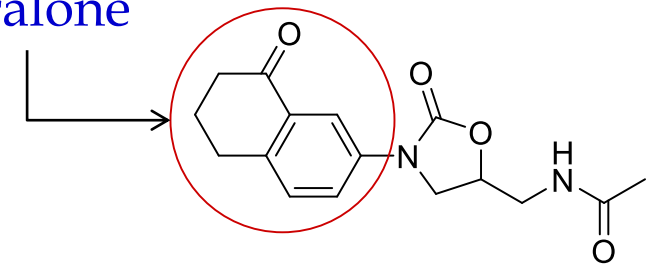
rac→ SAR/STR



Indanone
(<tox)

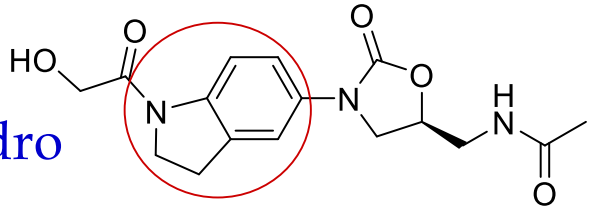
PNU-82965 (**14**)

tetralone



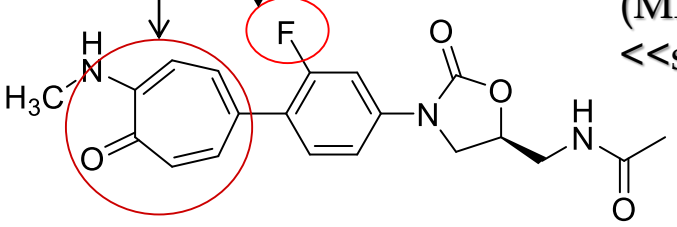
PNU-85055 (**15**)

troponyl
(>>activity (MRSA), <<solub)



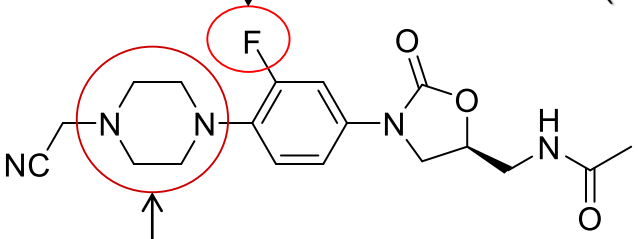
oxodihydro indenyl
(<tox, >activity)

PNU-97456 (**16**)

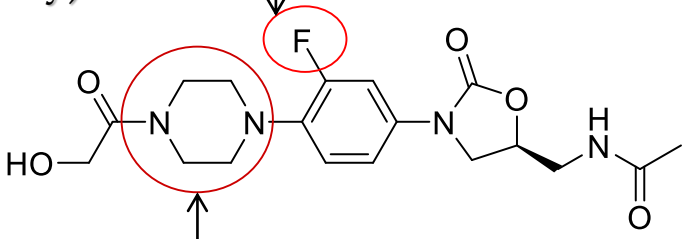


PNU-97786 (**17**)

(>>activity)



PNU-97665 (**18**)

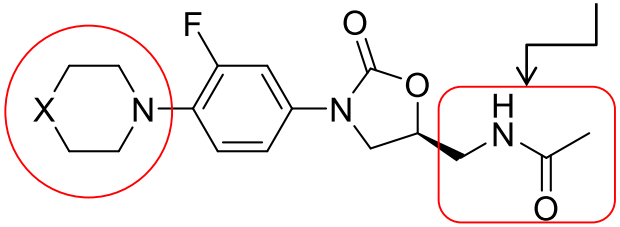


eperezolid (**19**)

piperazine (>>activity, >> good water solubility and pharmacokinetic properties)

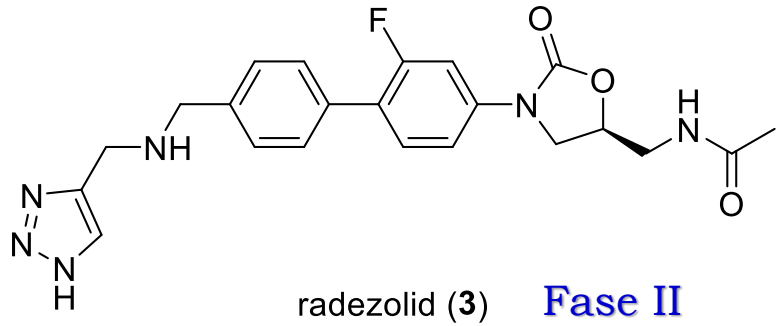
linezolid

acetamidomethyl

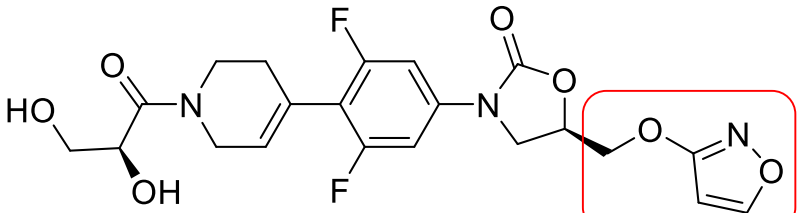


linezolid; X = O (**1**)

Fase I/II (TBC) sutezolid; X = S (**2**)

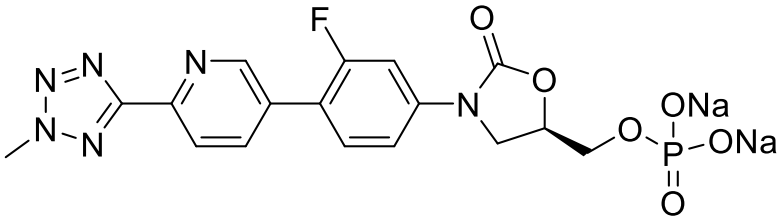


radezolid (**3**) Fase II



Fase II (TBC) posizolid (**4**)

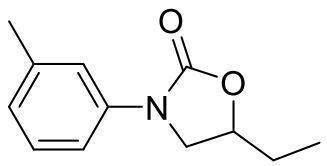
O-linked isoxazole



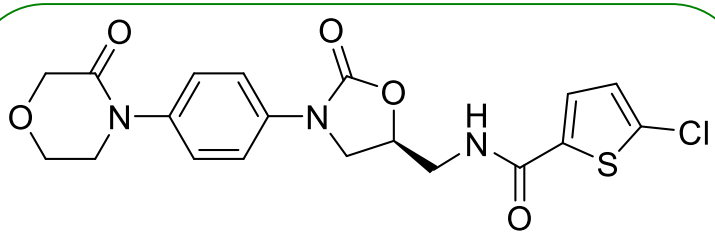
tedizolid phsophate (**5**)

Sivextro (EMEA 19/11/2015)

Inibitore MAO
(antidepressivo)

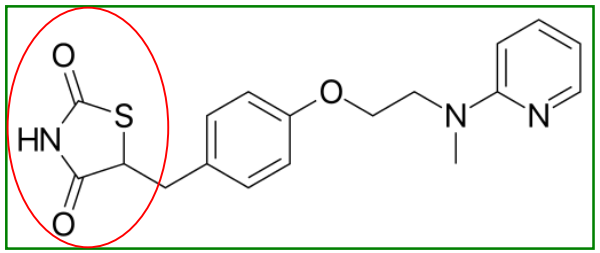


toloxatone (**6**)



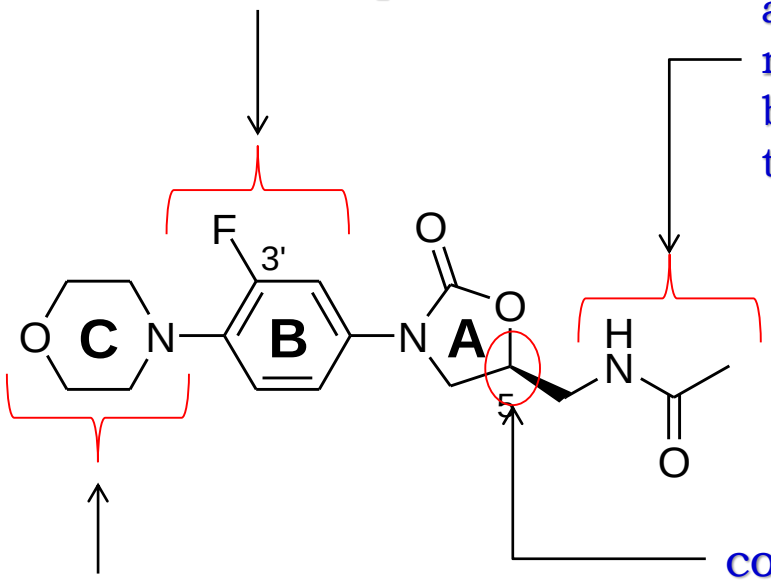
rivaroxaban (**7**)

Inibitore fattore Xa
(profilassi tromboflebiti venose)



Agonista PPAR γ
(ipolipidemico)

N-arile necessario; 3'-F migliora biodisponibilità e aumenta la potenza



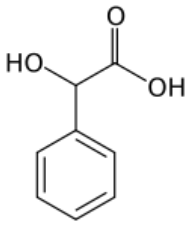
Essenziale come gruppo accettore/donatore di HB al sito ribosomiale.; sostituzione bioisosterica con eteroarili a 5 termini (vedi posizolid)

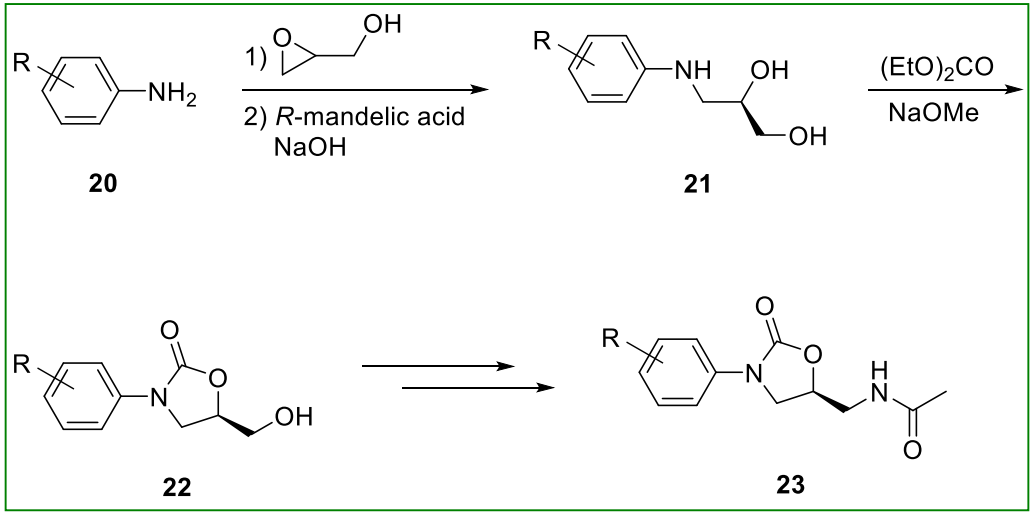
configurazione 5-(S)

Il gruppo morfolino aumenta la solubilità in acqua e assicura un buon profilo tossicologico; sostituzione bioisosterica con eteroarili (vedi tedizolid) aumenta la potenza.

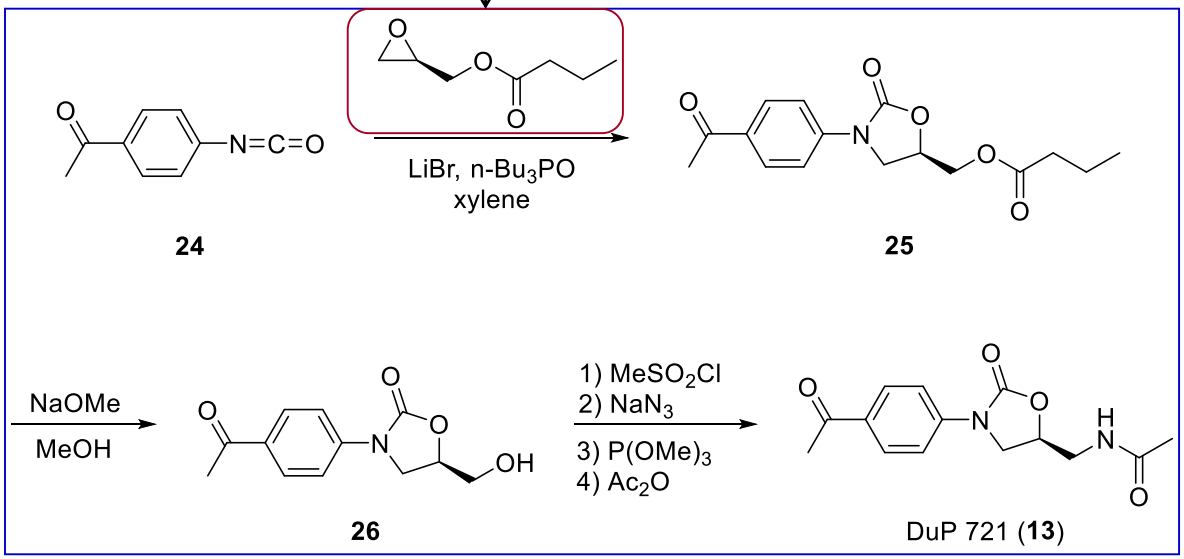
DuPont synthesis

glycidol



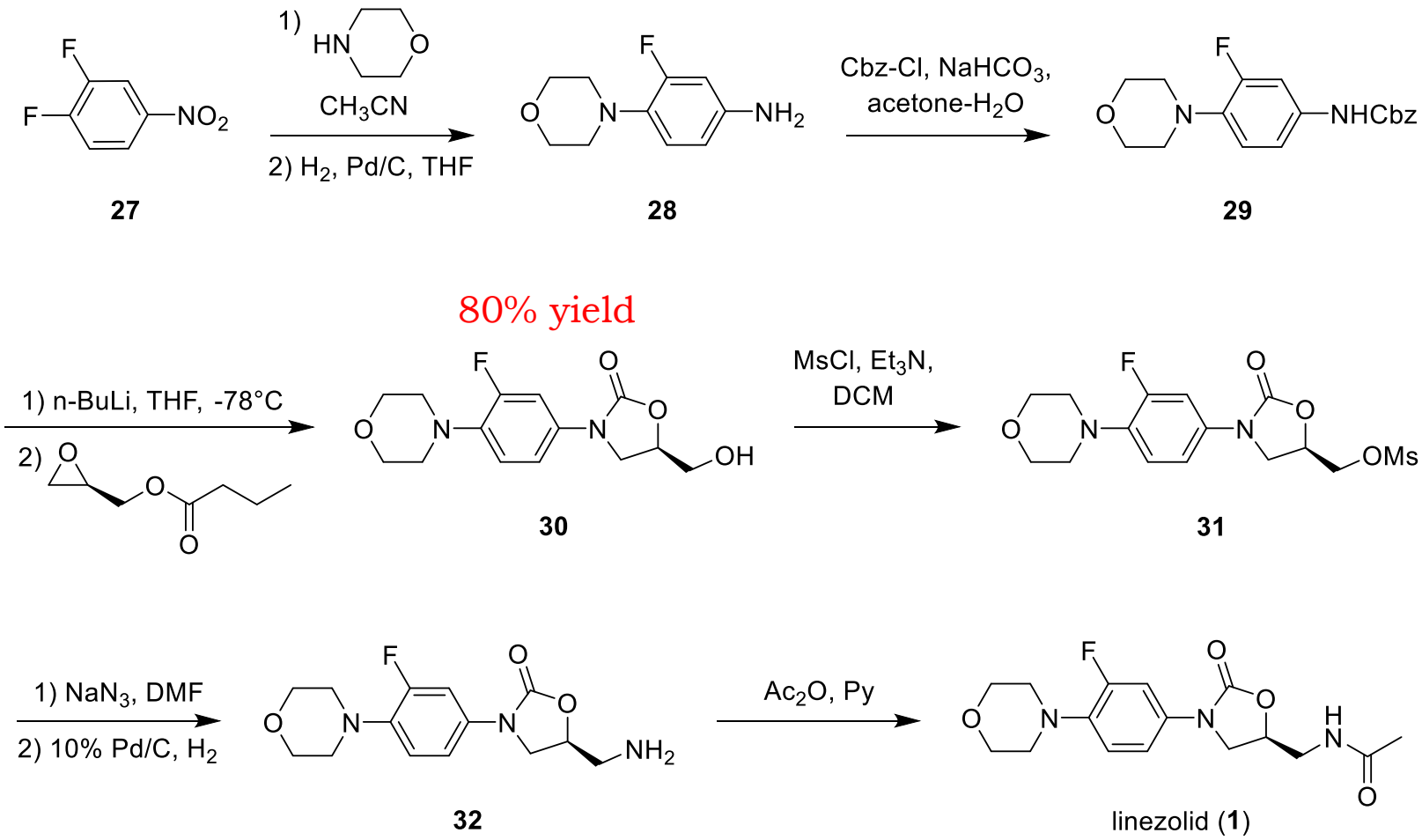


(*R*)-glycidyl butyrate

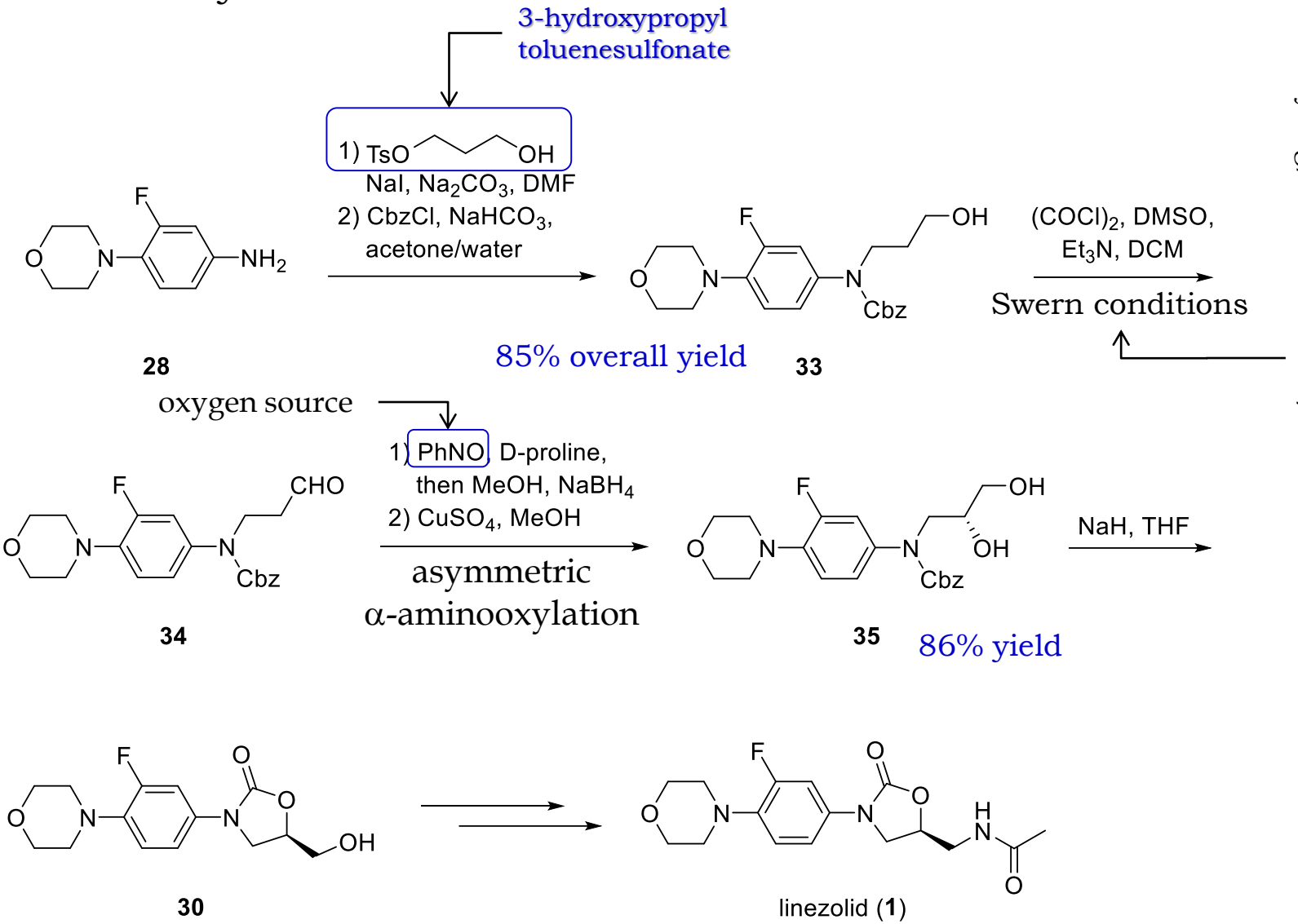


Trimethyl phosphite

Upjohn synthesis

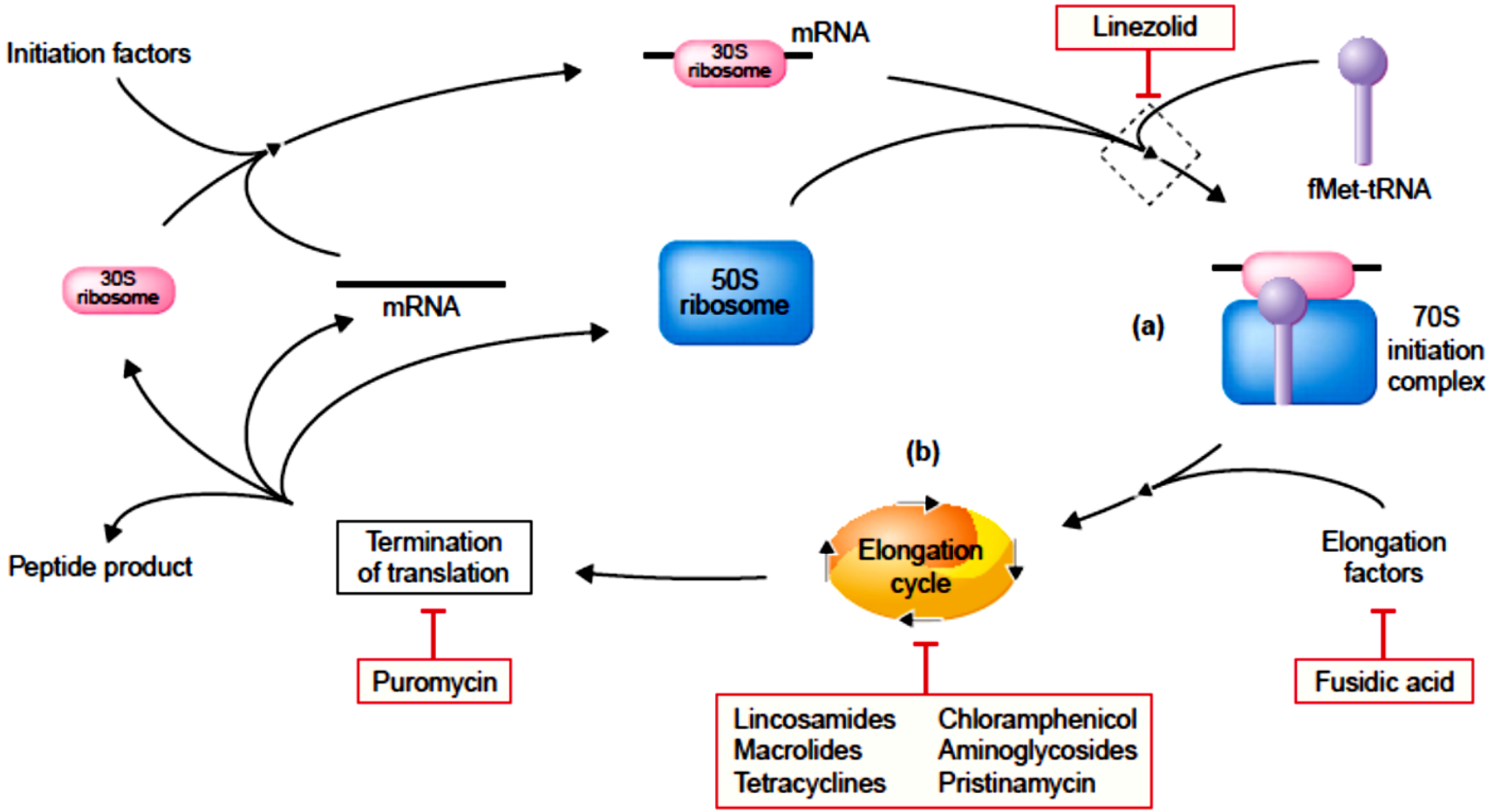


enantioselective synthesis



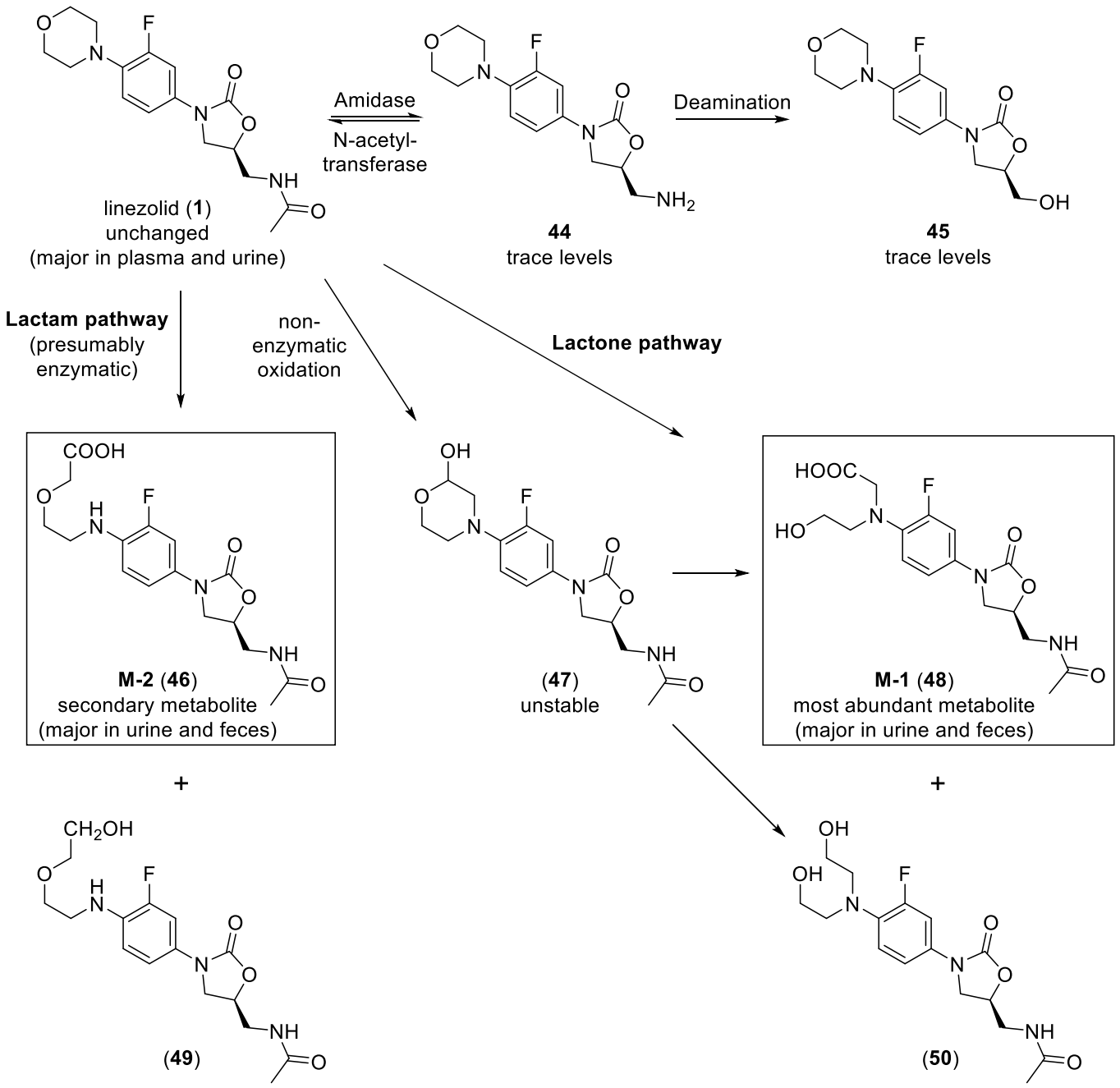
Direct proline catalyzed asymmetric α-aminooxylation of aldehydes. Tetrahedron Letters 44 (2003) 8293–8296

http://www.organic-chemistry.org/namedreactions/swern-oxidation.shtm



Current Opinion in Pharmacology

(a) La sintesi proteica ha inizio con la combinazione della sub-unità ribosomiale 30S, mRNA, fMet-tRNA, 50S.
Linezolid si lega alla sub-unità 50S impedendo l'accesso a fMet-tRNA (N-formilmetionil transfer RNA) e l'inizio della sintesi proteica. (b) La maggior parte degli antibiotici ribosomiali inibisce l'ultimo stadio di elongazione e crescita della catena peptidica. fMet-tRNA, N-formylmethionyl transfer RNA.


linezolid (1)
unchanged
(major in plasma and urine)
Amidase
N-acetyl-
transferase
44
trace levels
Deamination
45
trace levels
Lactam pathway
(presumably
enzymatic)
non-
enzymatic
oxidation
Lactone pathway
M-2 (46)
secondary metabolite
(major in urine and feces)
(47)
unstable
M-1 (48)
most abundant metabolite
(major in urine and feces)
+
+
(49)
(50)