Chimica Farmaceutica e Tossicologica 2

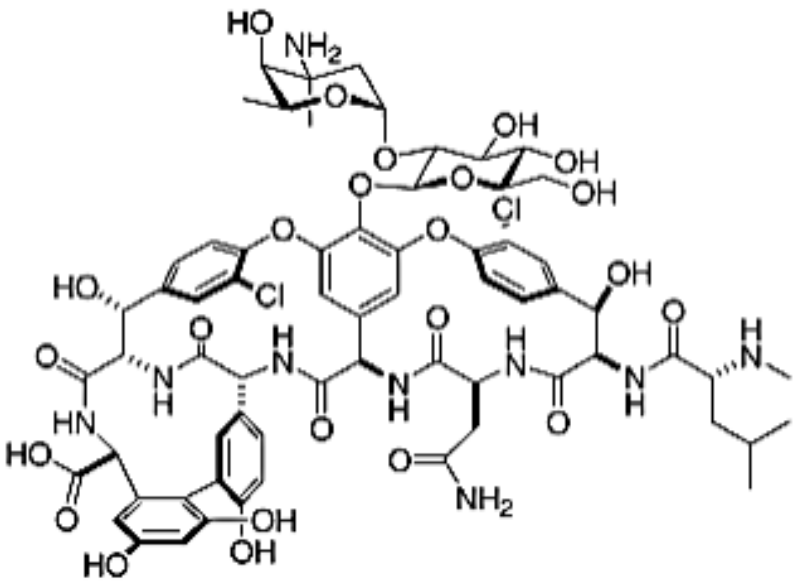
Antibiotici glicopeptidici

- Vancomicina
- Teicoplanina
- Oritavancina
- Telavancina
- Ramoplanina

Daptomicina

Table 2 | **Antibacterial compounds commonly used in systemic monotherapy**

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*



vancomicina

•**1952:** un missionario in Borneo (Indonesia) spedisce un campione di terra a E. C. Kornfield, chimico organico in Eli Lilly.
• un microorganismo isolato da quel campione (*Streptomyces orientalis*) produceva una sostanza (05865) biocida verso la maggior parte dei Gram-positivi, inclusi stafilococchi penicillino-resistenti, clostridia, Neisseria gonorrhea.

•*Geraci, J. E. et al. Some laboratory and clinical experiences with a new antibiotic, vancomycin. Mayo Clin. Proc. 1956, 31 , 564 – 582 .*
•*Levine, D. P. Vancomycin: a history. Clin. Infect. Dis. 2006 , 42 ,S5 – S12.*

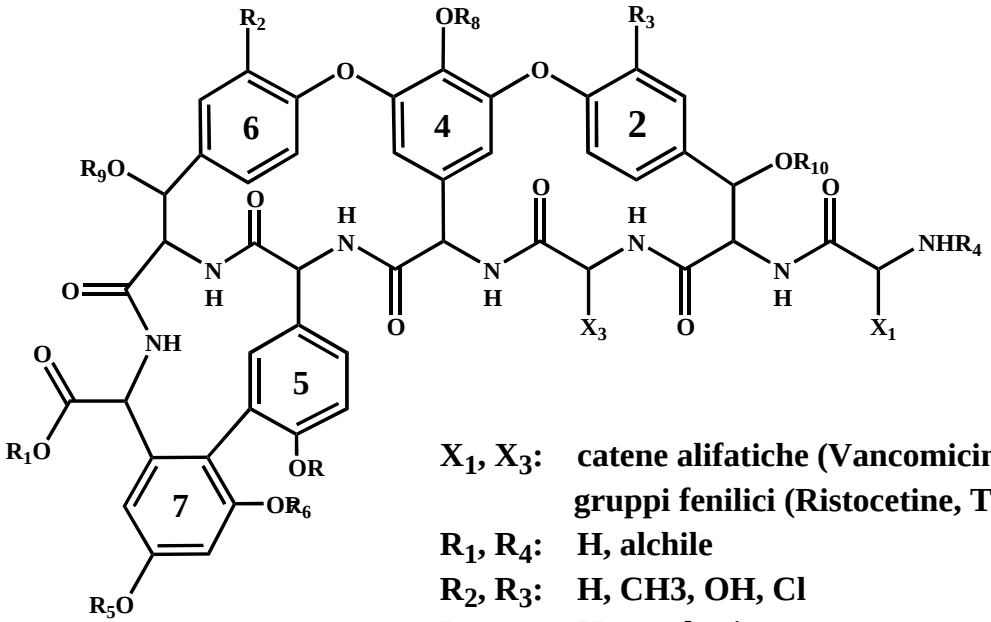
• sperimentazione in vitro: dopo 20 passaggi seriali di *stafilococchi*, la resistenza alla penicillina aumentava di 100.000-volte, a fronte di 4-8 volte con il composto 05865.
• la successiva sperimentazione in vivo (animale) confermò la bassa tossicità ed aprì all'uso in umani. Prima dei trials clinici, il composto " *Mississippi mud* " fu purificato e denominato "vancomicina" (*to vanquish*= vincere debellare).
• la vancomicina resta uno degli antibiotici più attivi nei confronti di infezioni sostenute da Gram-positivi anche meticillino-resistenti.
• FDA appr. 1958 (=meticillina)

ANTIBIOTICI GLICOPEPTIDICI *(lipoglicopeptidici)*

(FAMIGLIA DELLA VANCOMICINA-RISTOCETINA)

Nome alternativo proposto (*J. Antibiotics, 42, 1882 (1989)]:*
DALBAHEPTIDES (**D-A**lanyl-D-Alanine **B**inding **A**ntibiotics **Hep**tapep**tides**)

•ciclo peptidi legati (eterosidi) a glicidi e OH fenolici o benzilici;

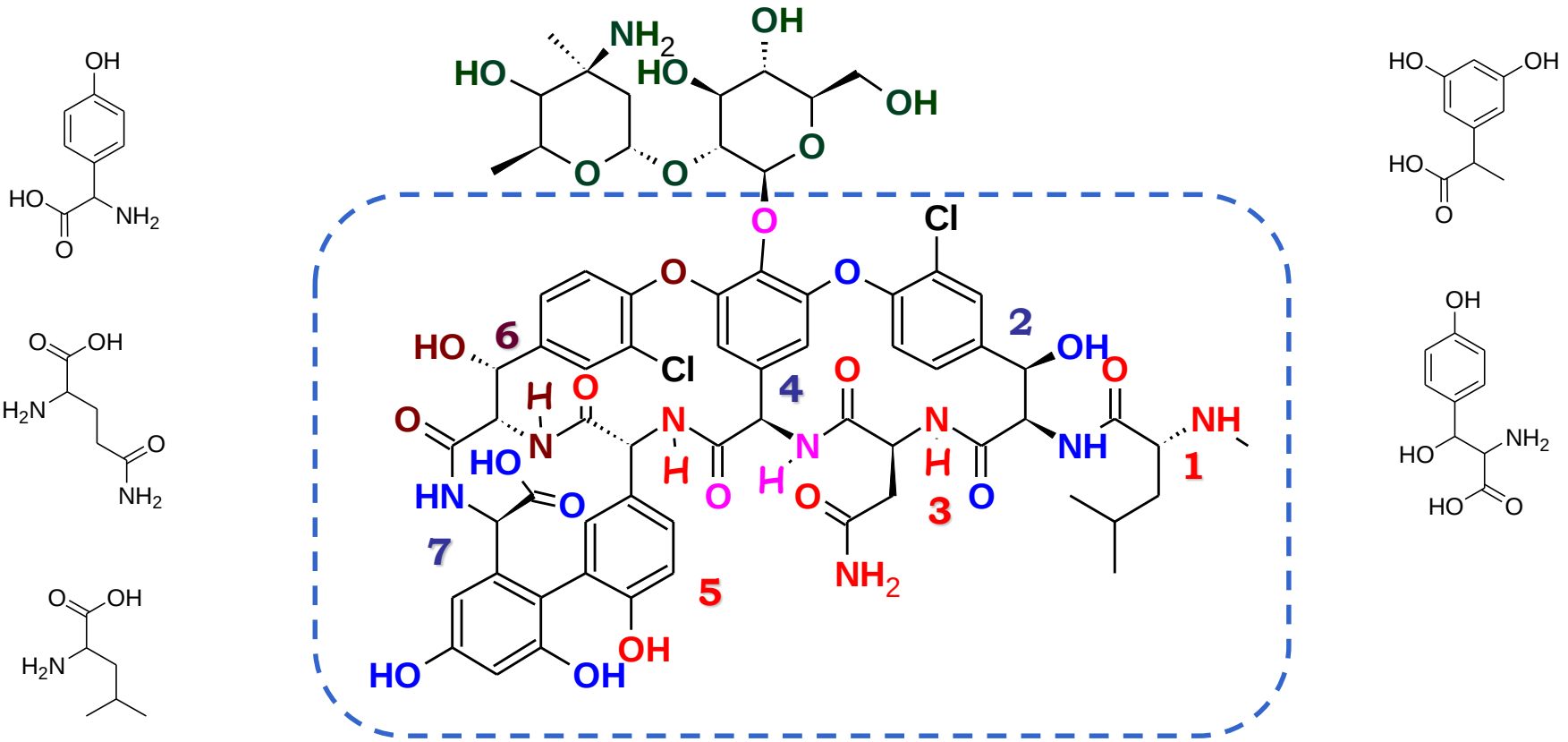


•**BIOGENESI**: condensazione di 7 a.a. seguita dalla formazione di legami (C-C, C-O-C) tra gruppi aromatici e successiva glicosilazione;

•**MECCANISMO D'AZIONE**: interagiscono con la porzione D-Ala-D-Ala del NAMA pentapeptide inibendo la sintesi del peptido-glicano;

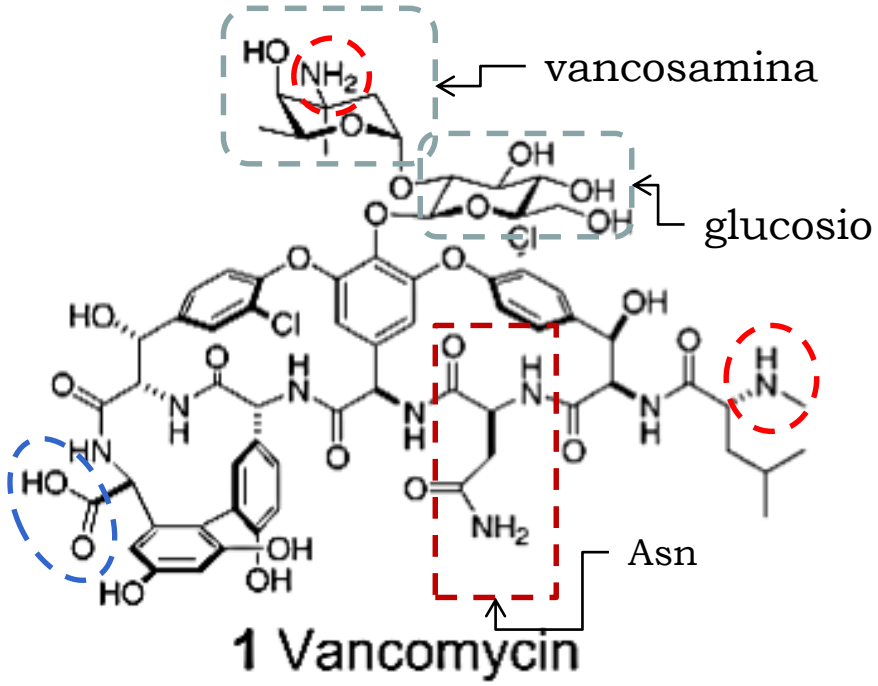
•**SPETTRO ANTIMICROBICO**: sono particolarmente attivi verso i GRAM+, anche su ceppi MRSA (resistenti a molti altri antibiotici)

TOSSICITA': oto- e nefrotossici

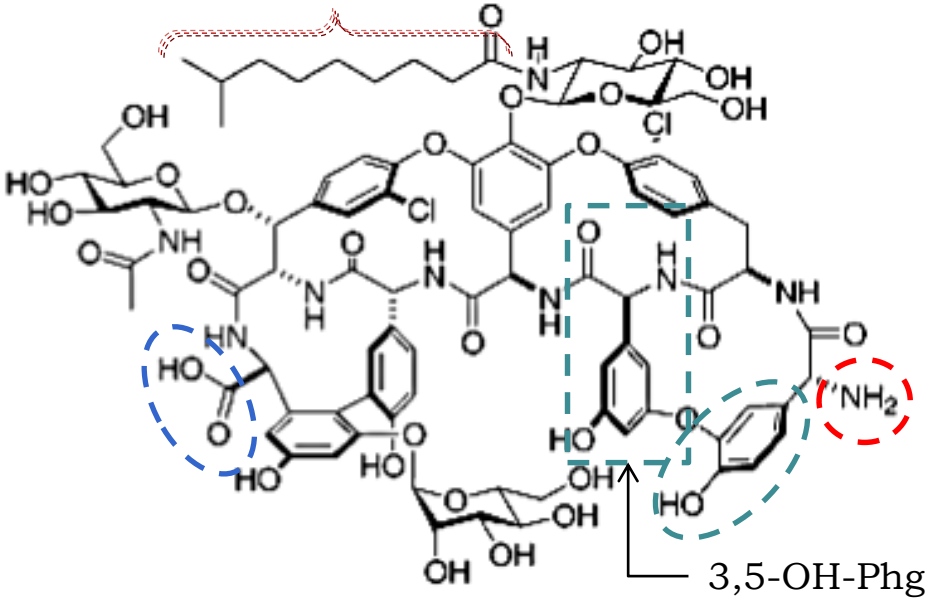


TIPO	AMMINOACIDO						
	COOH-X_7	X_6	X_5	X_4	X_3	X_2	X_1-NH_2
Vancomicina*	3,5-OH-Phg	β-OH-Tyr	p-OH-Phg	p-OH-Phg	Asn	β-OH-Tyr	Leu
Ristocetina**	3,5-OH-Phg	β-OH-Tyr	p-OH-Phg	p-OH-Phg	3,5-OH-Phg	β-OH-Tyr	p-OH-Phg
Teicoplanina	3,5-OH-Phg	β-OH-Tyr	p-OH-Phg	p-OH-Phg	3,5-OH-Phg	β-OH-Tyr	p-OH-Phg

3,5-OH-Phg = 3,5-diidrossifenilglicina β-OH-Tyr = β-idrossitirosina Asn = asparagina
Leu = leucina * Il gruppo amminico terminale è N-CH_3 ** Il gruppo carbossilico terminale è $COOCH_3$



1 Vancomycin



2 Teicoplanin

TIPO	AMMINOACIDO						
	COOH-X_7	X_6	X_5	X_4	X_3	X_2	X_1-NH_2
Vancomicina*	3,5-OH-Phg	β-OH-Tyr	p-OH-Phg	p-OH-Phg	Asn	β-OH-Tyr	Leu
Teicoplanina	3,5-OH-Phg	β-OH-Tyr	p-OH-Phg	p-OH-Phg	3,5-OH-Phg	β-OH-Tyr	p-OH-Phg

Gruppo Lepetit (Rifampicina e Teicoplanina);
Marion Merrel Dow Research Institute (’80);
Centro Ricerche di Gerenzano(’87);
Biosearch Italia s.p.a.(’96) (Ramoplanina);
Vicuron Pharmaceuticals (Dalbavancina);
Pfizer (*una delle più grandi collezioni al mondo di microrganismi produttori di principi attivi*);
FONDAZIONE ISTITUTO INSUBRICO
DI RICERCA PER LA VITA. http://www.ricercaperlavita.it/

1: $-(CH_2)_2CH{=}CH(CH_2)_4CH_3$ (9)

2: $-(CH_2)_6CH(CH_3)_2$ (9)

3: $-n\text{-}C_9H_{19}$

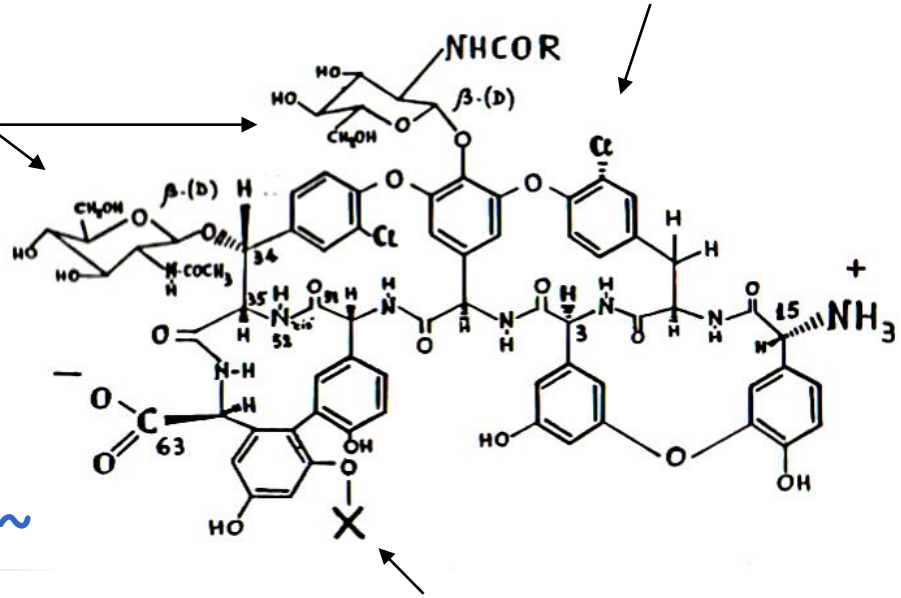
4: $-(CH_2)_6CH(CH_3)CH_2CH_3$

5: $-(CH_2)_7CH(CH_3)_2$

SAR Teicoplanina

De-cloro deriv. (aglicone)
~ Gram -⇧

aglicone ⇩



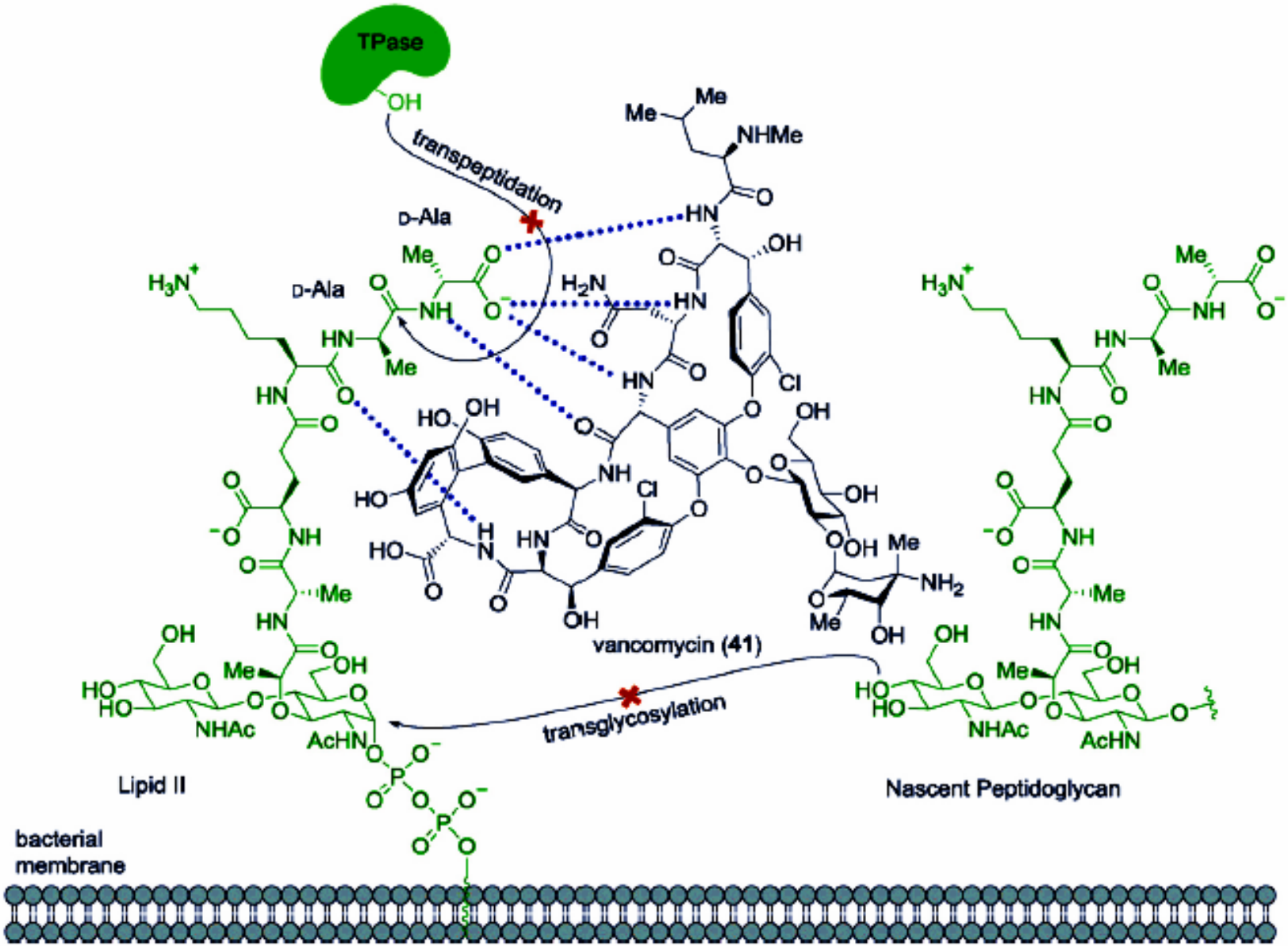
De-ammino ⇩

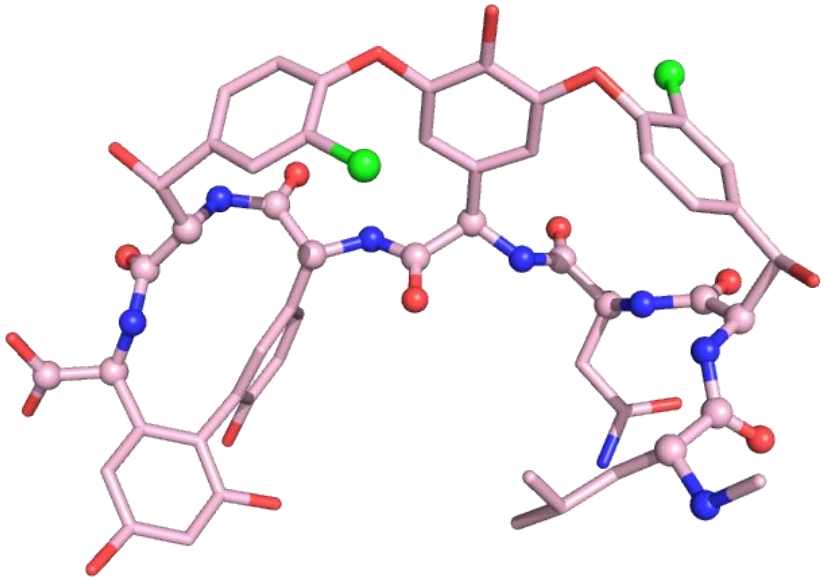
Carbossidrazidi ~

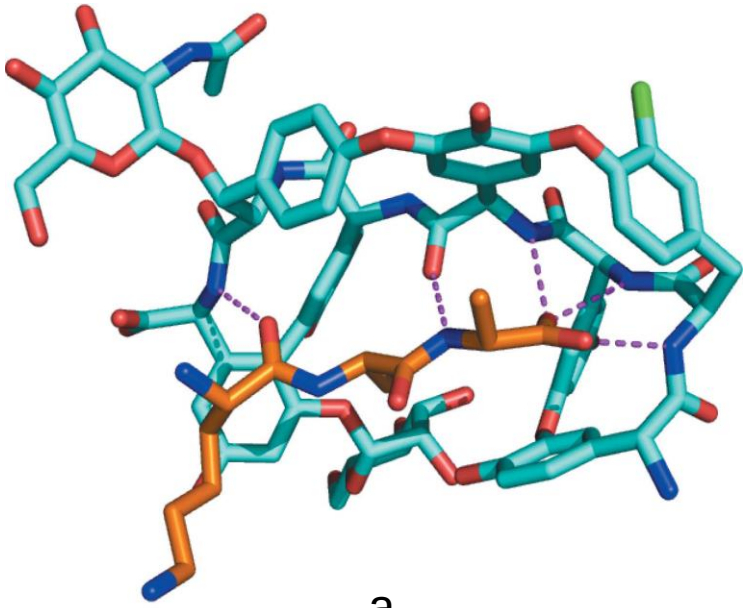
Esteri $_{aglicone}$ (Cl-Alchil) ~

Ammidi (>100cp) ~

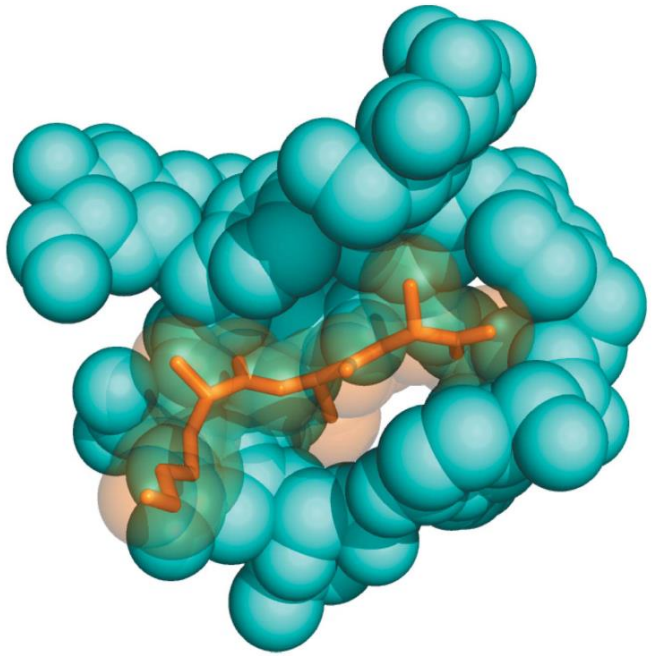
De-mannosil ~


TPase
OH
transpeptidation
D-Ala
D-Ala
vancomycin (41)
transglycosylation
Lipid II
Nascent Peptidoglycan
bacterial
membrane





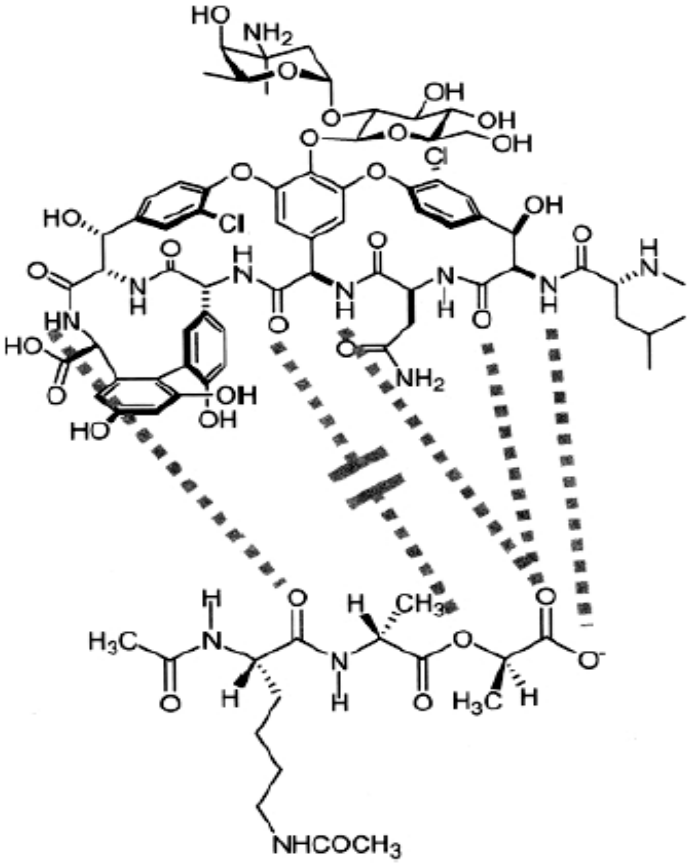
a

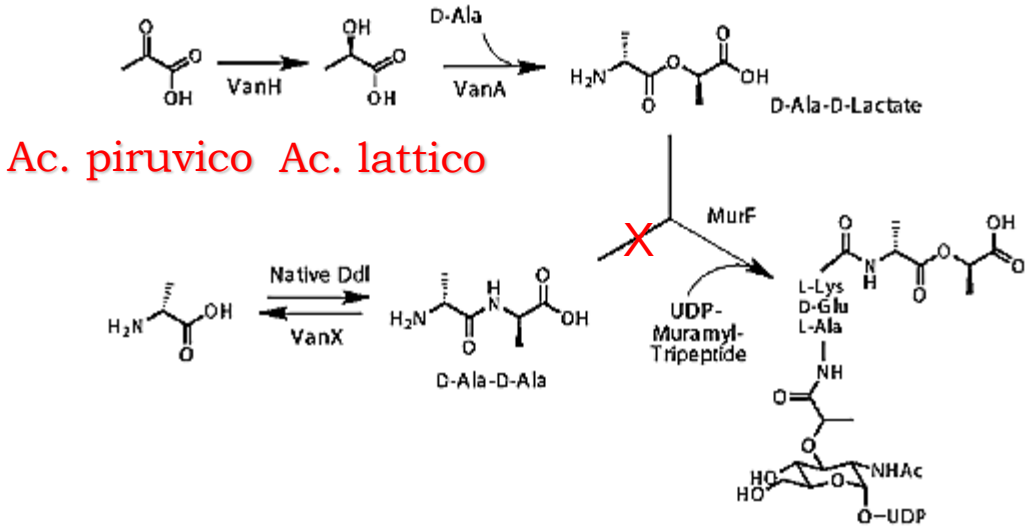


b

Interazioni della teicoplanina con il suo ligando.
(a) La teicoplanina riconosce il target per mezzo di 5 HB (tratteggio magenta) che legano il backbone peptidico dell'antibiotico al ligando. *Per semplificare l'immagine, l'N-acilglucosamina legata all' amino acido 4 è omessa.*
(b) La teicoplanina (sfere ciano) avvolge il proprio ligando peptidico (sticks arancio).

Basi molecolari della resistenza agli AB glicopeptidici





Ac. piruvico Ac. lattico

Vancomycin Resistant Enterococci (VRE-A,B)
Vancomycin-Resistant S. aureus (VRSA)
Vancomycin Intermediate Staphylococcus Aureus Infection (VISA)

1988: identificata una forma di resistenza alla vancomicina (Enterococchi, VRE).
Specie generalmente simbiotica ma patogena in caso di immunodeficienza per HIV o trapianti d'organo.
Geni di batteri resistenti possono essere trasferiti od altre specie come MRSA.
VRE: tipo A, B (D-Ala-D-Lact) e C.
Riduzione dell'attività di circa 1000 volte.

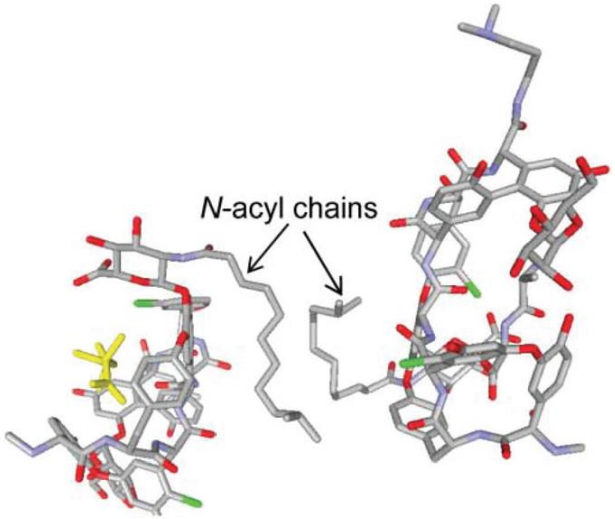
Table 1 Dimerisation thermodynamics of dalbavancin, vancomycin and ristocetin A in the absence or presence of ligand (Ac_2-Kaa) at 25 °C in 0.1 M NaOAc, pH 5.0

		(M^{-1})	(kJ mol^{-1})		
Antibiotic	Ligand	K_{dim} [a]	ΔH_{dim} [a]	$T\Delta S_{dim}$ [a]	ΔG_{dim} [a]
Dalbavancin	None	38 400 ± 8260	−45.0 ± 2.2	−18.9 ± 1.7	−26.1 ± 0.5
	Ac_2-Kaa	n/a[b]	n/a[b]	n/a[b]	n/a[b]
Vancomycin	None	750 ± 80	−11.5 ± 0.5	4.9 ± 0.7	−16.4 ± 0.3
	Ac_2-Kaa	1940 ± 170	−17.1 ± 0.4	1.6 ± 0.2	−18.7 ± 0.2
Ristocetin A	None	920 ± 120	−14.2 ± 0.5	2.7 ± 0.8	−16.9 ± 0.3
	Ac_2-Kaa	690 ± 140	−20.0 ± 1.3	−3.9 ± 1.8	−16.1 ± 0.5

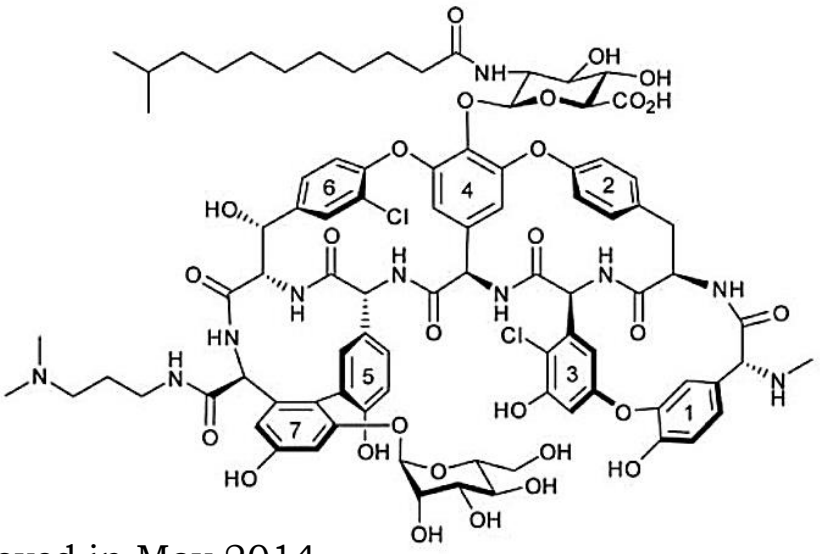
[a] Data are means ± SD for $n = 3$. [b] The thermodynamics of dalbavancin dimerisation in the presence of Ac_2-Kaa could not be determined due to the poor solubility of the complex.

Dalbavancin dimerisation and ligand binding are anti-cooperative

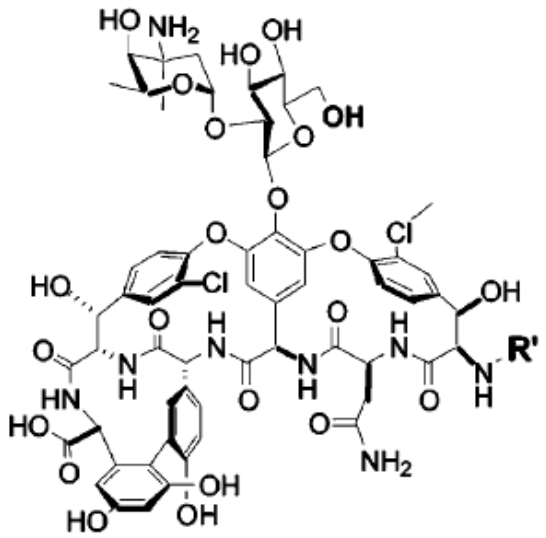
half-life of 170–210 h, which makes the once-weekly dosing optimal



c Dalbavancin

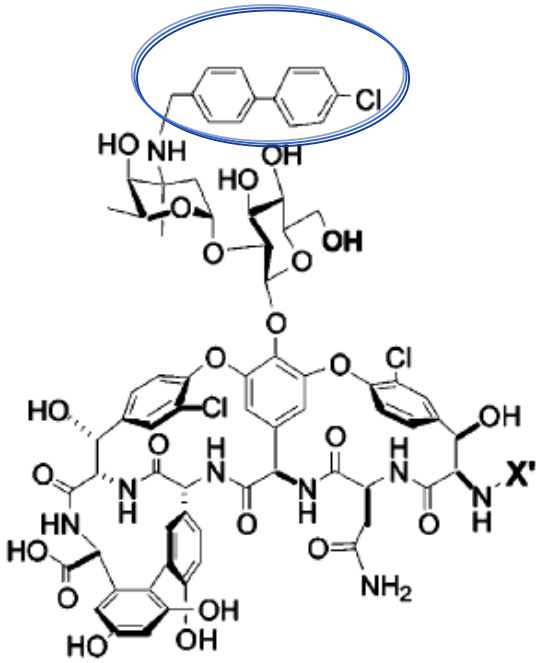


semisynthetic lipoglycopeptide approved in May 2014



Vancomycin
(R' = *N*-Me-D-Leu)

Damaged Vancomycin
(R' = H)

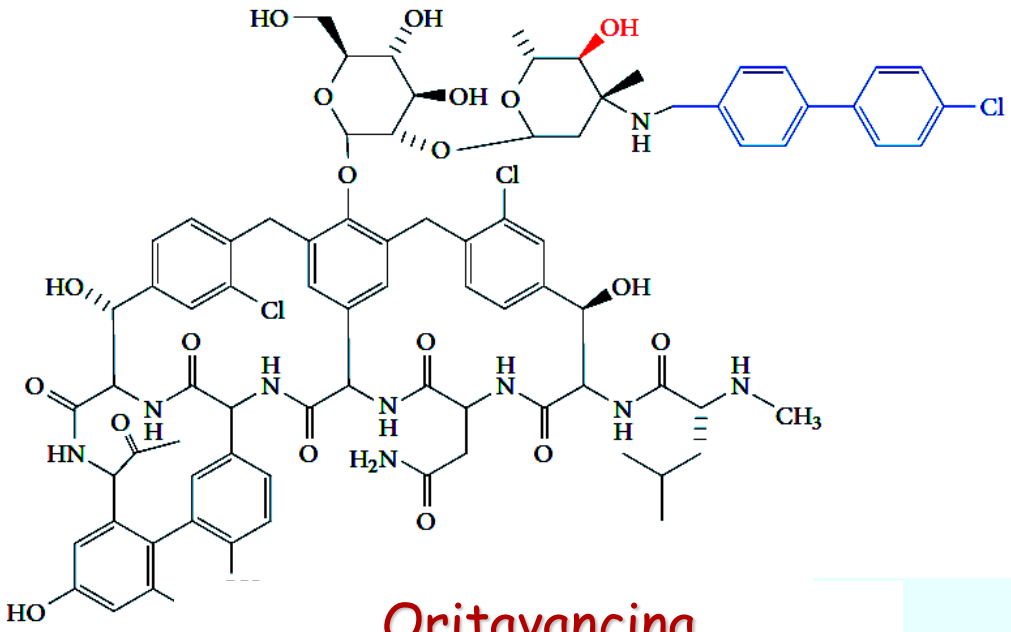


Chlorobiphenyl
Vancomycin
(X' = N-Me-D-Leu)

Damaged Chloro-
biphenly Vancomycin
(X' = H)

glycopeptide	MIC (μg/mL) sensitive *E. faecium*	MIC (μg/mL) resistant *E. faecium*	
vancomycin	1	2048	~2000
chlorobiphenyl vancomycin	0.03	16	~500
damaged vancomycin	no activity	no activity	
damaged chlorobiphenyl vancomycin	10	40	~4

Antibiotici multivalenti sperimentali



Oritavancina

(Orbactiv)

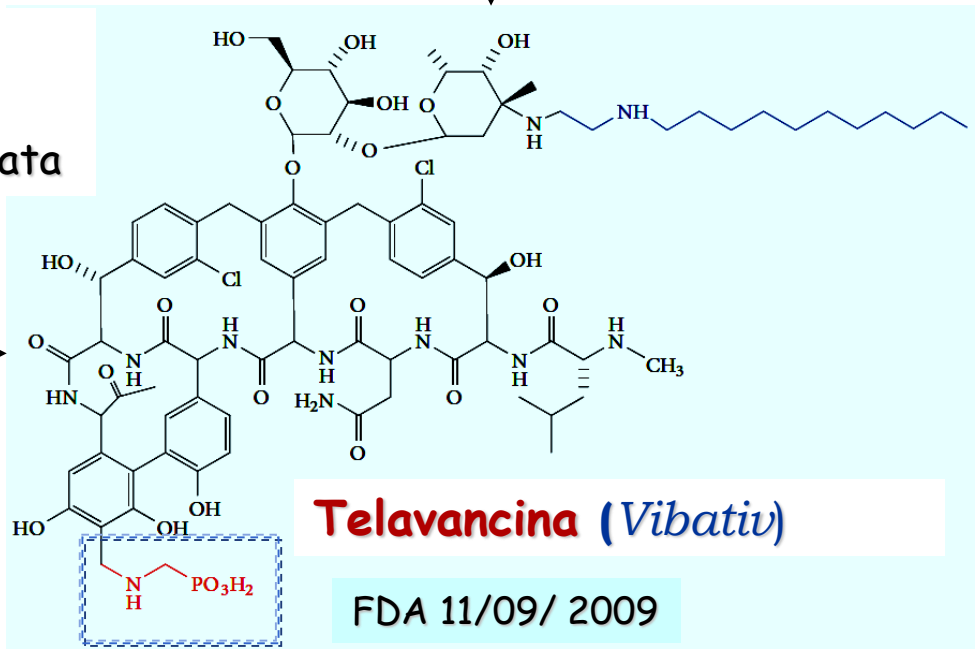
Polmonite da ventilazione forzata

Attivi vs MRSA e VRE

complicated skin and skin structure infections (cSSSI)

EMEA 19/03/2015

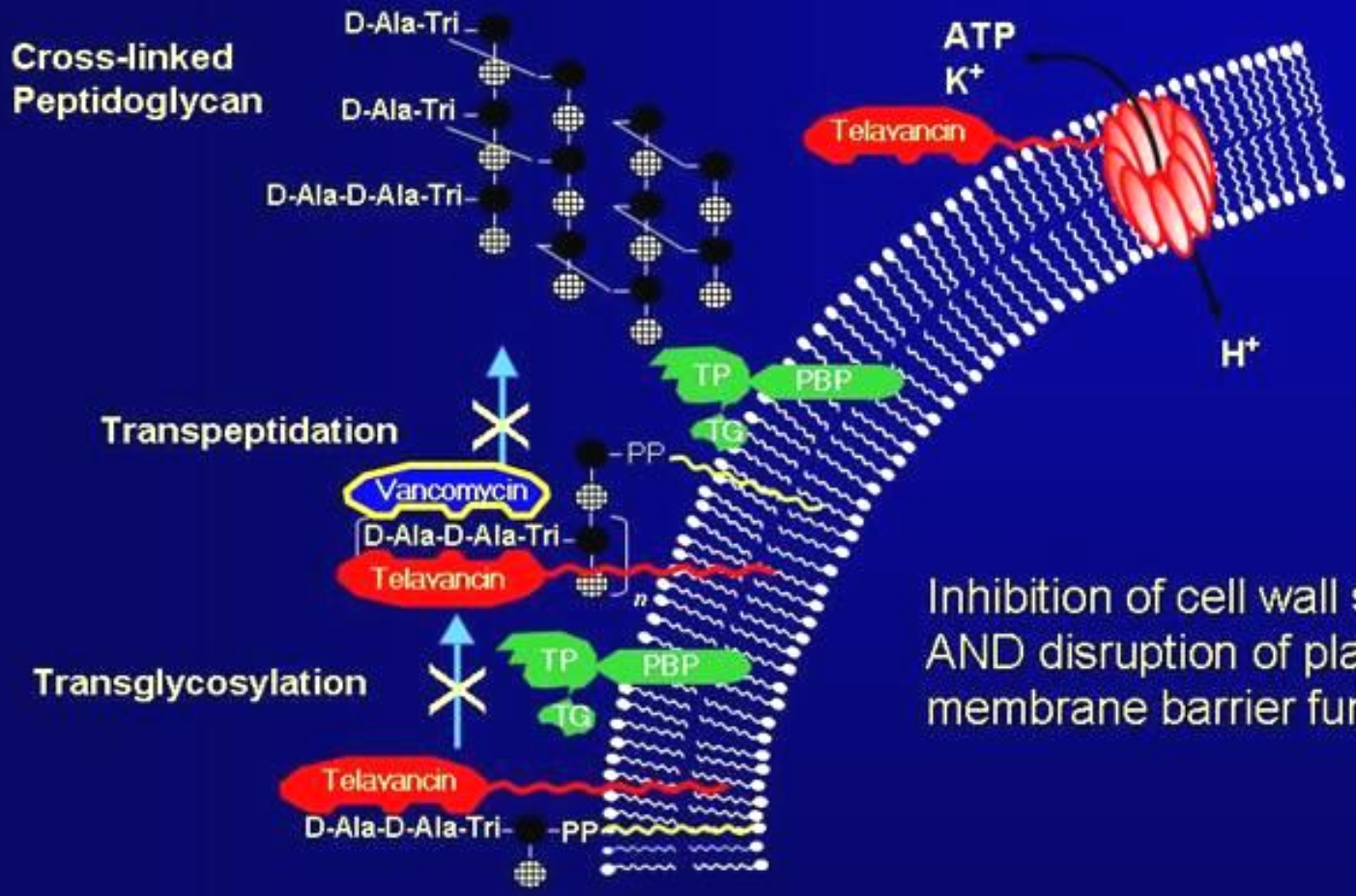
Blocco D-Ala-D-Ala + permeabilizzazione-depolarizzazione di membrana



Telavancina (*Vibativ*)

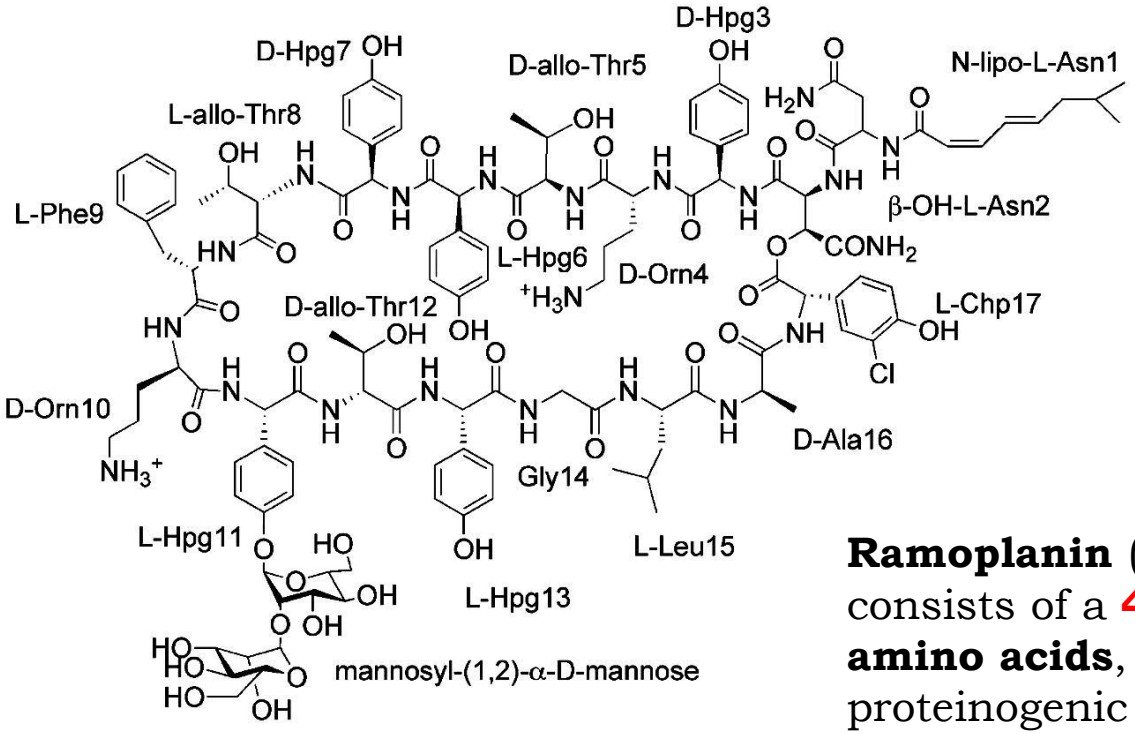
FDA 11/09/ 2009

Unique Multifunctional Mechanism of Action



Inhibition of cell wall synthesis AND disruption of plasma membrane barrier functions

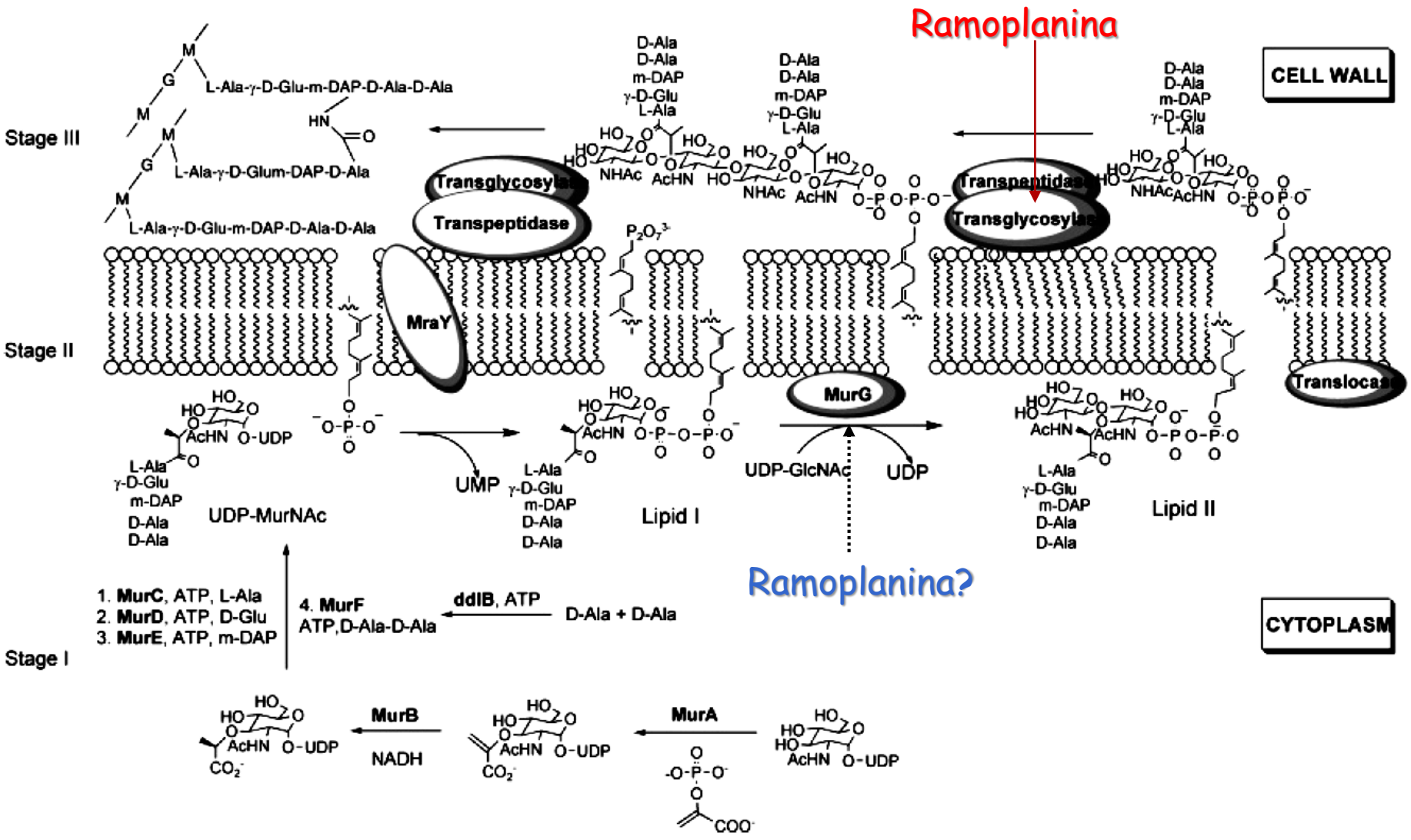
Ramoplanina (1984 Biosearch Italia, formerly Gruppo Lepetit SPA, Italy) antibiotico lipoglicopeptidico, approvato da FDA; instabile nel circolo ematico nel quale degrada in sottoprodotti inattivi; **somministrabile solo o.s.** (MW 2554)



- Gram + (*Staphylococcus, Enterococcus*, *Bacillus, Clostridium difficile*)
- inattiva Gram (-)
- (non penetra la membrana)

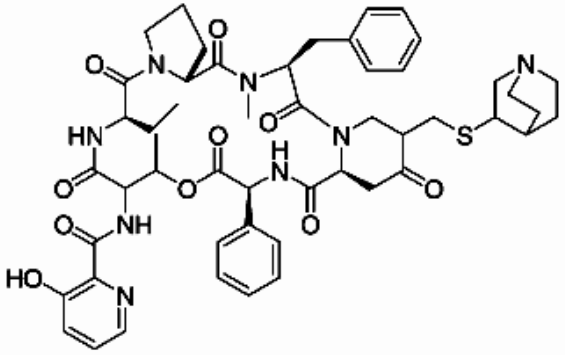
Ramoplanin ($C_{119}H_{154}ClN2_1O_{40}$/2554.07 Da) consists of a **49-member ring** containing **17 amino acids**, including several non proteinogenic amino acids

Una infezione CDAD (Clostridium difficile-associated diarrhea) può causare diarrea, colite fulminante con possibile esito fatale. Ramoplanina può essere usata per curare CDAD con la sua azione battericida in vitro vs C. difficile ed altri batteri simili.

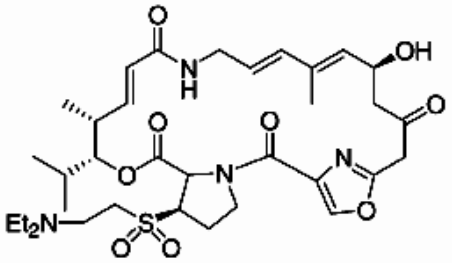


Non compete con D-Ala-D-Ala. Sequestro lipide II

Nuove classi di antibiotici multivalenti approvati per il trattamento delle infezioni MRSA e VRE

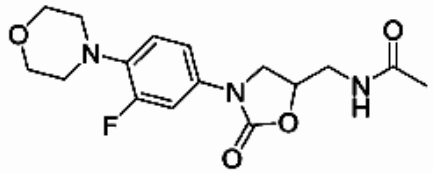


Quinupristin

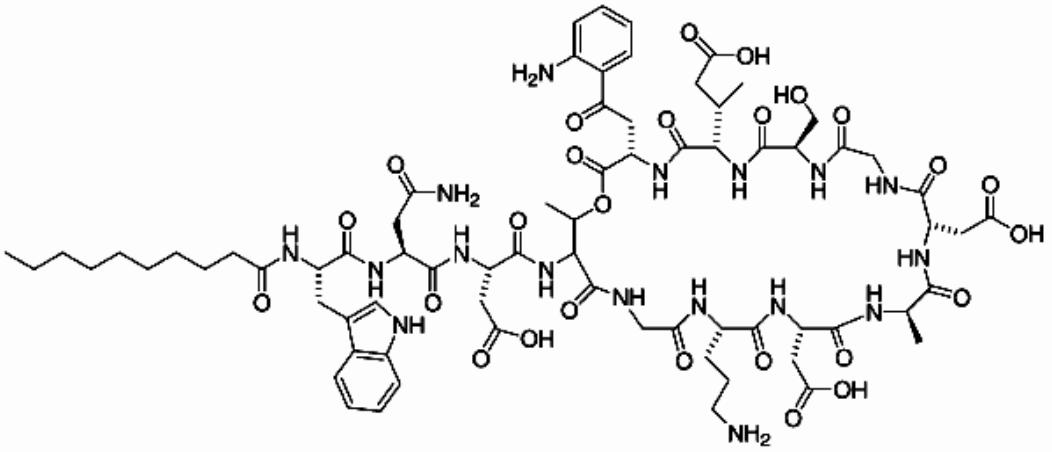


Dalfopristin

Streptogramine semisintetiche (antibiotici ribosomiali)

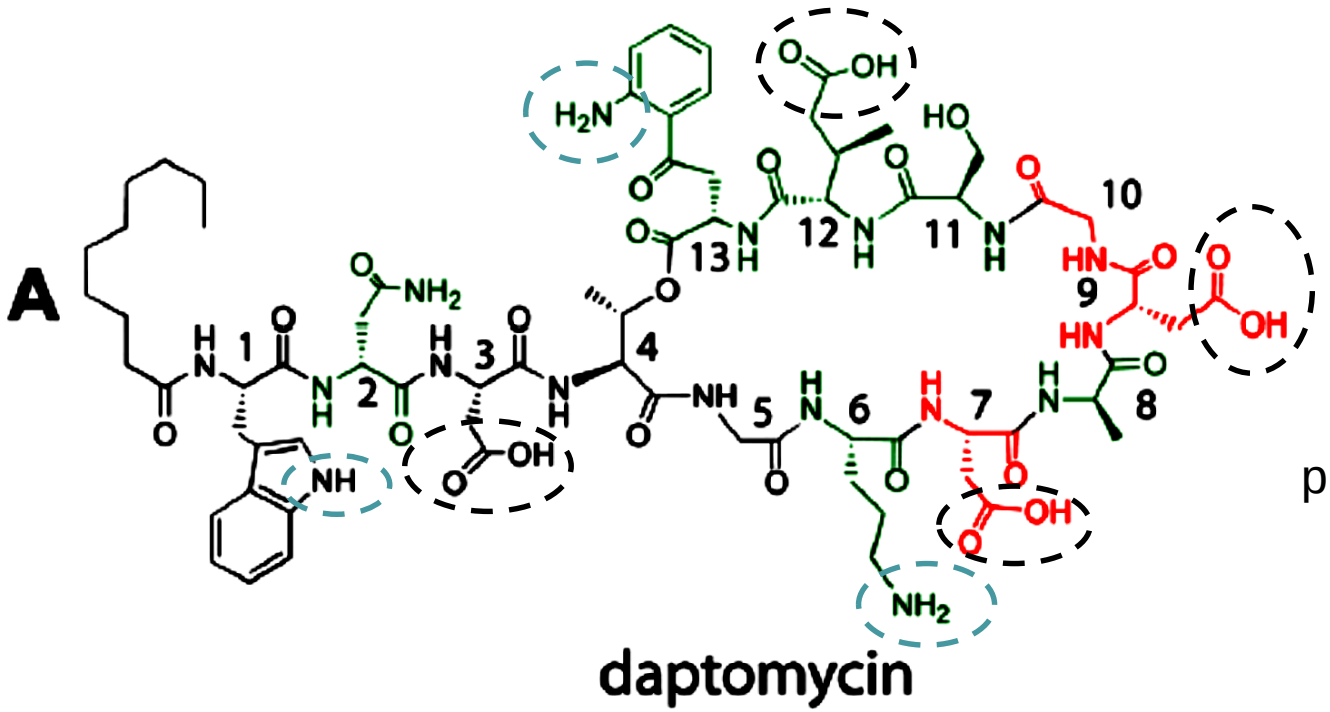


12 Linezolid



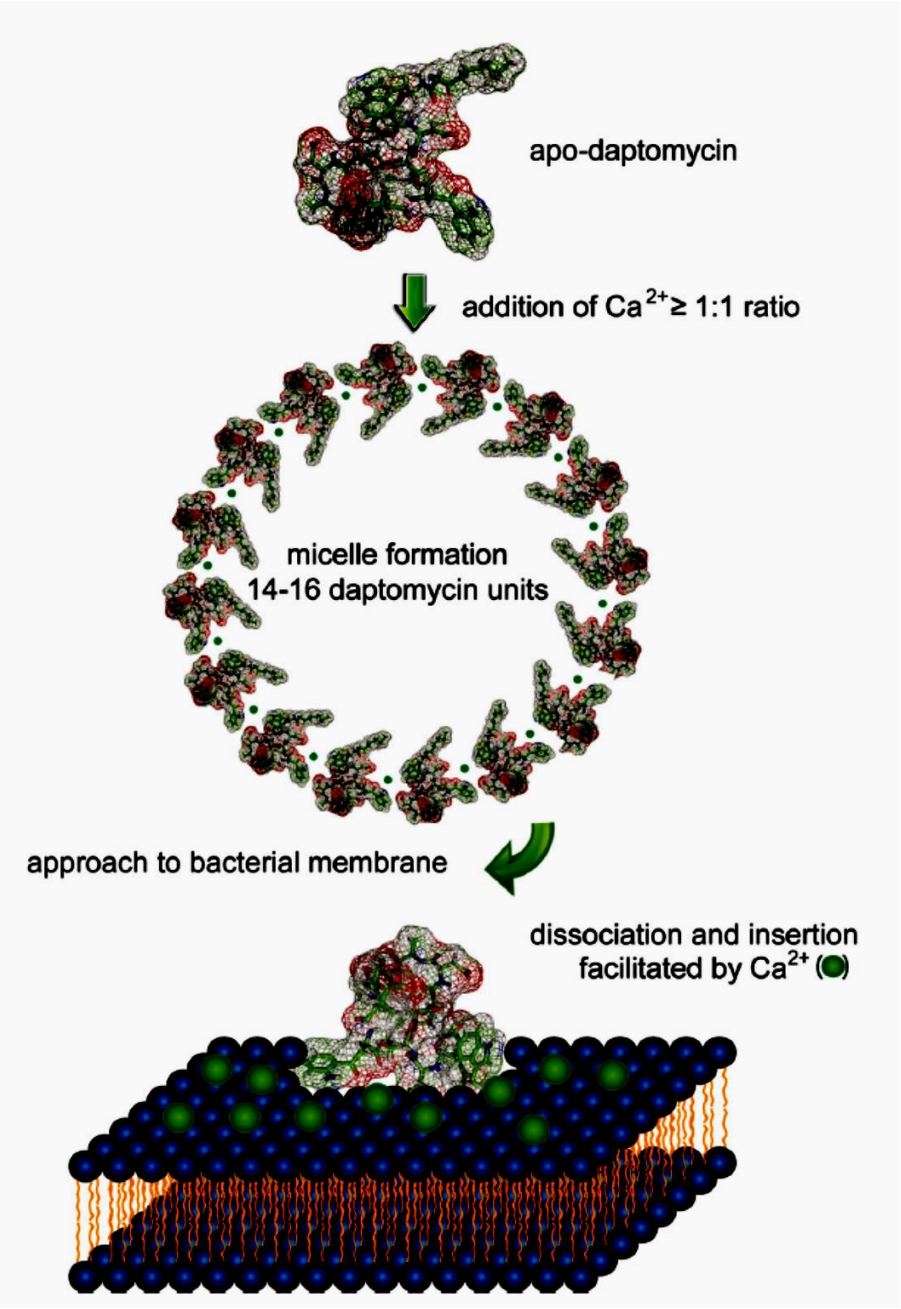
13 Daptomycin

Figure 24. Three nonglycopeptide classes of antibiotics recently approved for treatment of VRE infections: Synercid (**11**) combination of Quinupristin and Dalfopristin blocks protein synthesis; the oxazolidinone Linezolid (**12**) blocks the first peptide-bond-forming step at bacterial ribosomes; the lipopeptide daptomycin (**13**) damages membranes and causes ion leaks in bacteria.



pI 5.9 to 6.2

Daptomicina: *Streptomyces roseosporus*, antibiotico lipopeptidico ciclico di 13aa approvato nel 2003 (FDA) per il trattamento di dermatiti da Gram-positivi ed endocarditi (*S. aureus* e MRSA).



La daptomicina amfifilica (*Cubicin*) forma aggregati di 14–16 membri dopo aggiunta di Ca^{++} (1:1).
Il complesso dissocia in prossimità del bilayer lipidico formando inserti ed oligomeri.
La formazione di pori destabilizza la membrana per efflusso di K^{+} fino al lisi cellulare. t½ 7 days

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ARTICLE

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doi:10.1038/nature14098

A new antibiotic kills pathogens without detectable resistance

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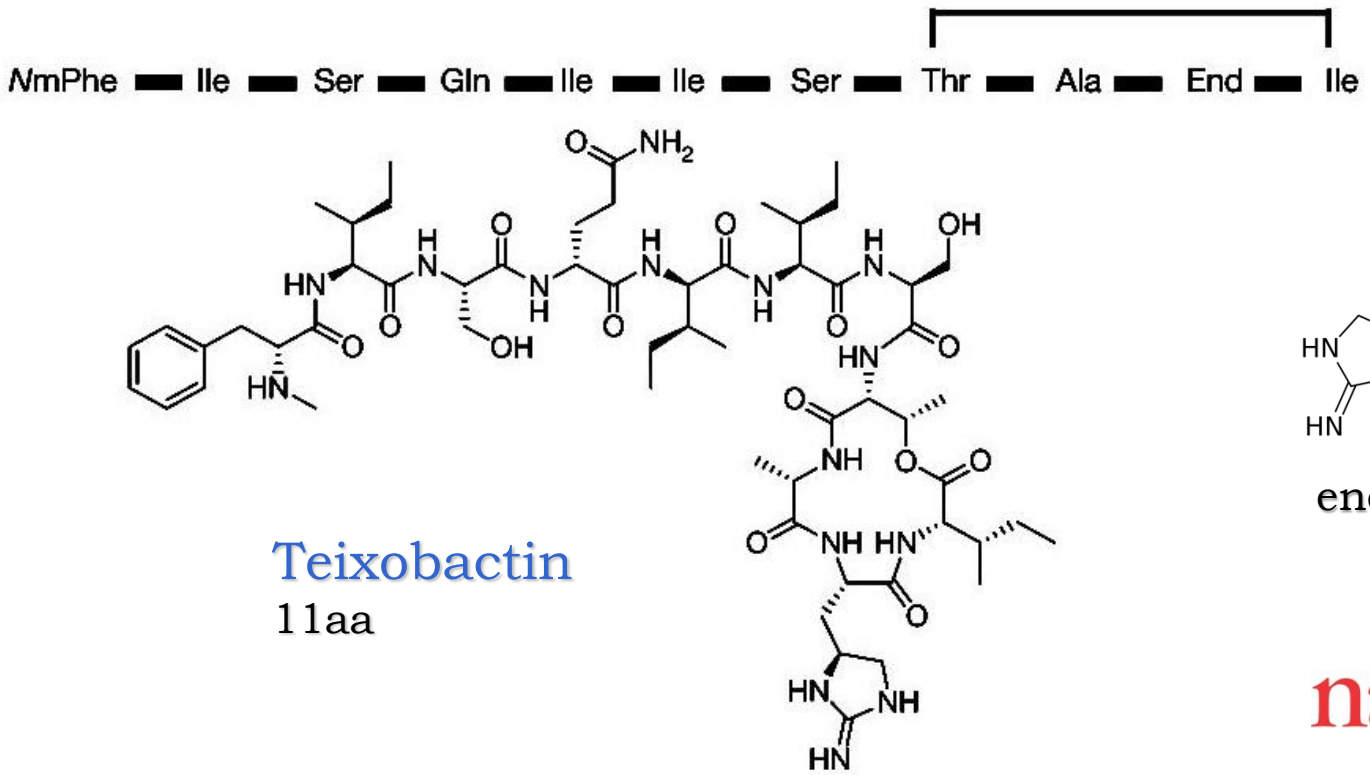
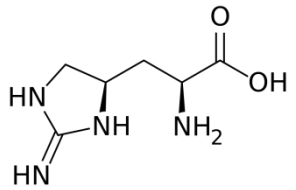


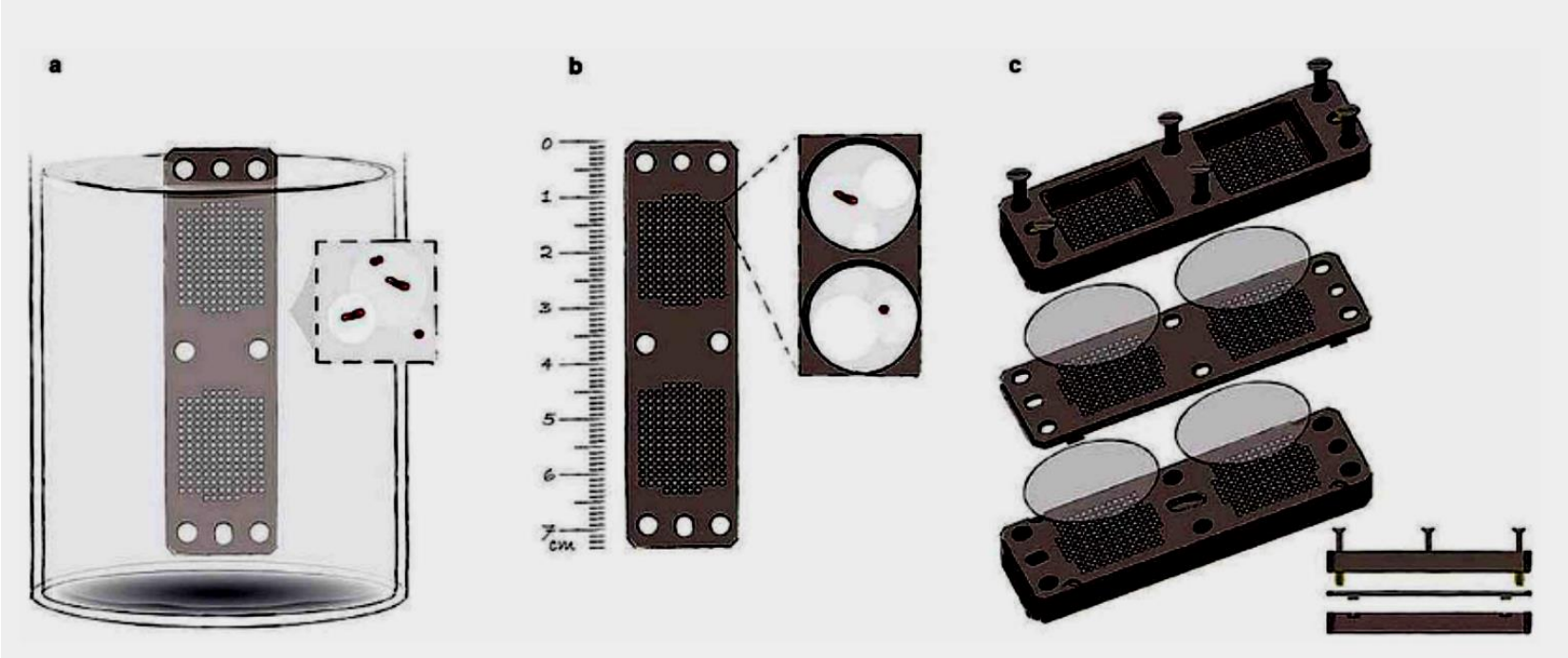
Teixobactin

11aa



enduracididine

nature



Extracts from 10,000 isolates obtained by growth in iChips were screened for antimicrobial activity on plates overlaid with *S. aureus*. An extract froma new species of β-proteobacteria provisionally named *Eleftheria terrae* showed good activity. The genome of E. terrae was sequenced

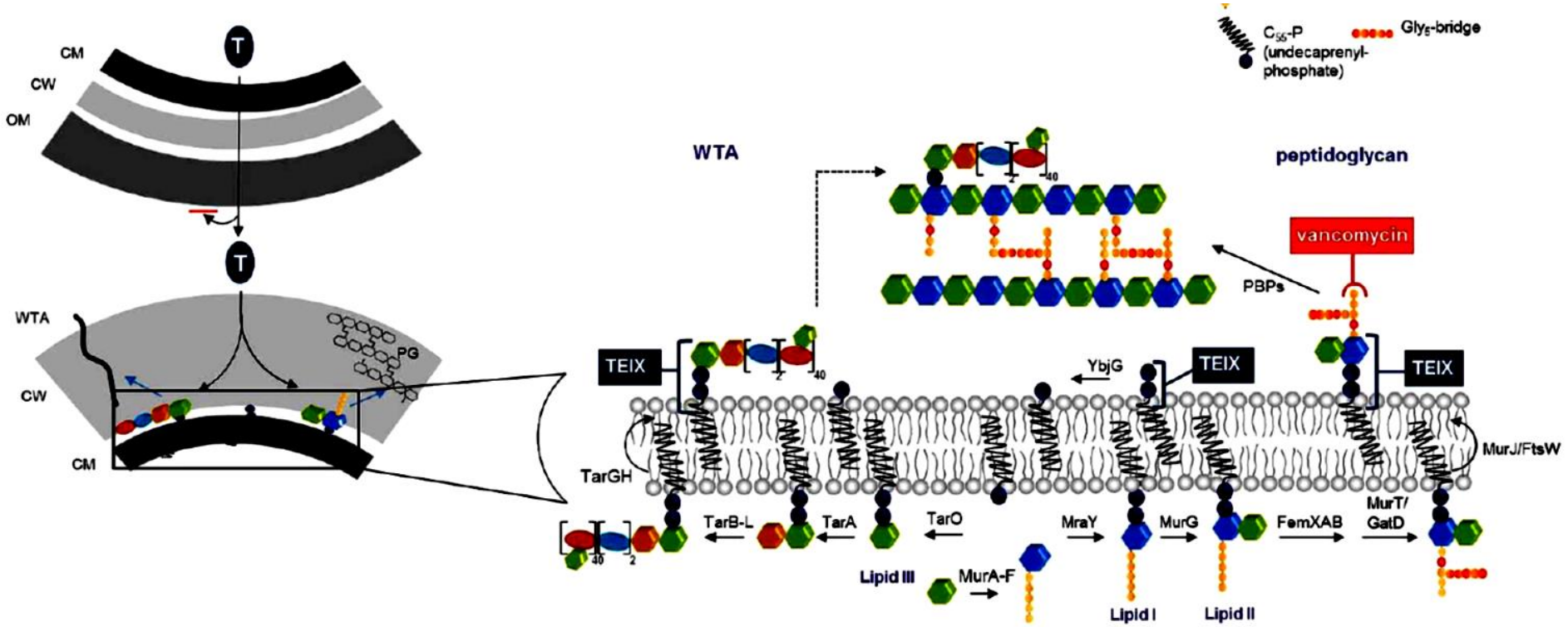
Table 1 | Activity of teixobactin against pathogenic microorganisms

Organism and genotype	Teixobactin MIC ($\mu g\,ml^{-1}$)
S. aureus (MSSA)	0.25
S. aureus + 10% serum	0.25
S. aureus (MRSA)	0.25
Enterococcus faecalis (VRE)	0.5
Enterococcus faecium (VRE)	0.5
Streptococcus pneumoniae (penicillin^R)	≤0.03
Streptococcus pyogenes	0.06
Streptococcus agalactiae	0.12
Viridans group streptococci	0.12
B. anthracis	≤0.06
Clostridium difficile	0.005
Propionibacterium acnes	0.08
M. tuberculosis H37Rv	0.125
Haemophilus influenzae	4
Moraxella catarrhalis	2
Escherichia coli	25
Escherichia coli (*asmB1*)	2.5
Pseudomonas aeruginosa	>32
Klebsiella pneumoniae	>32

nature

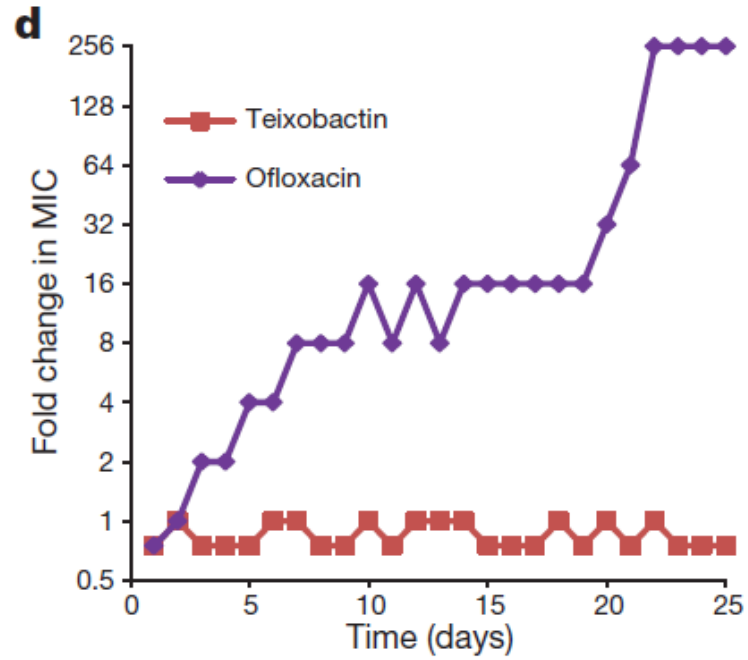
Teixobactina è inattiva vs Gram neg ma mostra buona attività vs *E. coli (asmB1)* con membrana esterna con scarsa funzione di barriera permeabile.

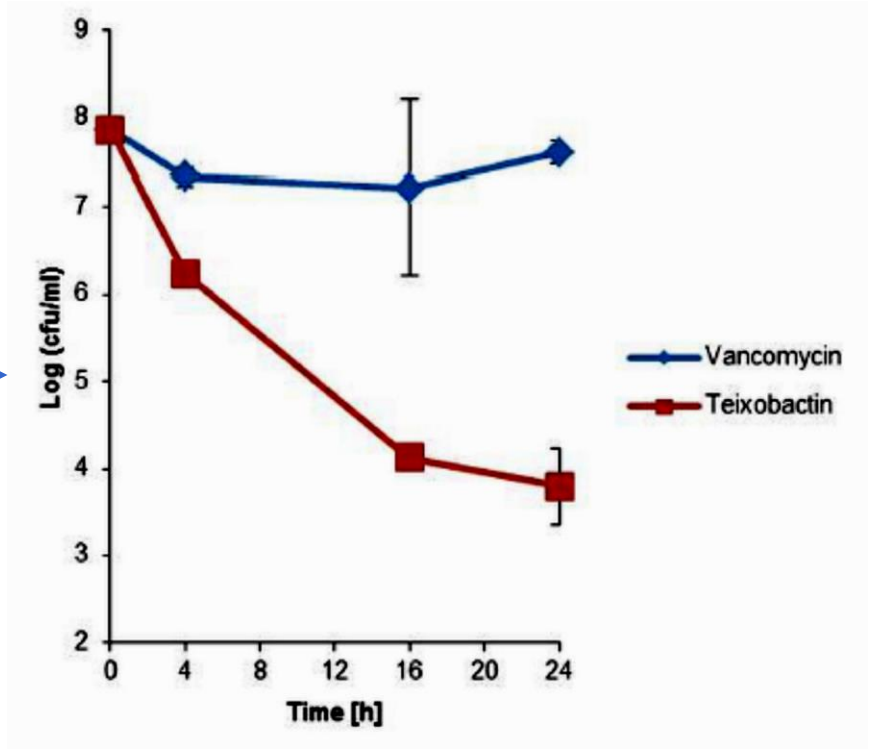
nature



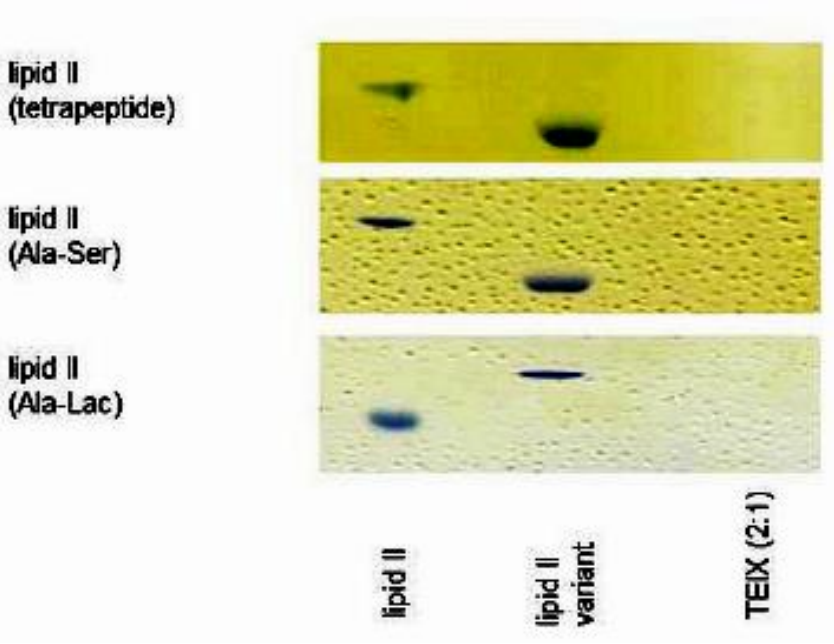
Teixobactina (T) è prodotta da Gram neg che la estrudono (inattiva) attraverso la membrana esterna (OM). I Gram-pos sono privi della OM e la teixobactina accede rapidamente allo spazio peiplasmatico legando I precursori del peptidoglicano (PG) e dell'acido teicoico della parete (WTA). (CM, membrana citoplasmatica).

T inibisce fortemente la sintesi del peptidoglicano, ma è priva di effetto sull'incorporazione di DNA, RNA e proteine. Il trattamento di S. aureus con **T** produce un accumulo di (UDP-MurNAc-pentapeptide) come nel controllo trattato con Vancomicina. Lipide I, lipide II e undecaprenilpirofosfato sono substrati di **T**.





Vancomycin intermediate S. aureus (VISA)



Formazione di complessi tra teixobactina e percursori della parete cellulare comprese alcune varianti associate alla resistenza ai glicopeptidi.

nature