GLYCOPEPTIDES:

how can a structural modification bring to a new life an old family of antibiotics?

F. Van Bambeke
Pharmacologie cellulaire et moléculaire
Université catholique de Louvain – Brussels - Belgium
Glycopeptide story: from natural to semi-synthetic derivatives

~ 1950:
discovery of vancomycin in Mississippi mud

~ 1985:
large clinical use in USA
Gram(+) infections and digestive tract decontamination

Problems:
- toxicity of vancomycin due to impurities
  - better purification procedures (after 1970…)
- emergence of resistance ….

By the way …~ 1980:
discovery of teicoplanin, as a natural GP with improved PK
  - largely used in Europe
Glycopeptide story: from natural to semi-synthetic derivatives

~ 1990:
Launching of large scale research program for finding GP with optimized properties → hemi-synthetic compounds

Malabarba and Nicas

- vancomycin
- teicoplanin
- oritavancin (1996)
- telavancin (2001)
- dalbavancin (1999)

- oritavancin (1996)
- telavancin (2001)
- dalbavancin (1999)
Peptidoglycan synthesis

transpeptidase

transglycosylase

d-ala

ddl

UDP

precursor synthesis

cytosol

reticulation

cell wall

precursor synthesis

cytosol
Glycopeptide mechanism of action

binding to D-Ala-D-Ala prevents the reticulation of peptidoglycan precursors
But resistance came in ...


**Vancomycin-resistant enterococci.**

Uttley AH, Collins CH, Naidoo J, George RC.


**Plasmid-mediated resistance to vancomycin and teicoplanin in Enterococcus faecium.**

Leclercq R, Derlot E, Duval J, Courvalin P.

Service de Bacteriologie, Virologie Hygiene, Hopital Henri Mondor, Universite Paris XII, France.


**Characterization of vancomycin resistance in Enterococcus faecium and Enterococcus faecalis.**

Nicas Tl, Wu CY, Hobbs JN Jr, Preston DA, Allen NE.

Lilly Research Laboratories, Eli Lilly & Co., Indianapolis, Indiana 46285-0438.
Resistance in enterococci

large clinical use in USA started in ~ 1985

Resistance in enterococci

P. Courvalin et al’s work
Resistance in enterococci

from susceptible ... ... to resistant

1 hydrogen bound is missing!

### Methicillin-resistant *Staphylococcus aureus* clinical strain with reduced vancomycin susceptibility


K. Hiramatsu\textsuperscript{a*}, H. Hanaki\textsuperscript{a}, T. Ino\textsuperscript{b}, K. Yabuta\textsuperscript{b}, T. Oguri\textsuperscript{c} and F. C. Tenover\textsuperscript{d}

\textsuperscript{a}Department of Bacteriology; \textsuperscript{b}Department of Pediatrics, Juntendo University, Tokyo; \textsuperscript{c}Clinical Laboratory, Juntendo Hospital, Tokyo, Japan; \textsuperscript{d}Nosocomial Pathogens Laboratory, Centers for Disease Control and Prevention, Atlanta, GA, USA

<table>
<thead>
<tr>
<th>AB</th>
<th>MIC</th>
</tr>
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<tbody>
<tr>
<td>AMP</td>
<td>64</td>
</tr>
<tr>
<td>VAN</td>
<td>8</td>
</tr>
<tr>
<td>GEN</td>
<td>128</td>
</tr>
<tr>
<td>RIF</td>
<td>2048</td>
</tr>
<tr>
<td>LVX</td>
<td>8</td>
</tr>
<tr>
<td>TET</td>
<td>128</td>
</tr>
<tr>
<td>SMX</td>
<td>0.125</td>
</tr>
<tr>
<td>Q-D</td>
<td>0.5</td>
</tr>
<tr>
<td>LZD</td>
<td>2</td>
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</table>
Resistance in staphylococci (GISA)

**Tickened Cell Wall**

**Multiplication of the Target!**


Infection with Vancomycin-Resistant Staphylococcus aureus Containing the vanA Resistance Gene

Soju Chang, M.D., M.P.H., Dawn M. Sievert, M.S., Jeffrey C. Hageman, M.H.S., Matthew L. Bouldin, M.D., Fred C. Tenover, Ph.D., M.P.H., Frances Pouch Downes, Dr.F.H., Sandip Shah, M.S., James T. Rudrik, Ph.D., Guy R. Pupp, D.P.M., William J. Brown, Ph.D., Denise Cardo, M.D., Scott K. Fridkin, M.D., for the Vancomycin-Resistant Staphylococcus aureus Investigative Team

Resistance in staphylococci (GRSA)
## Current glycopeptides: what do they offer to us?

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# Glycopeptides: how can we improve?

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<td>Spectrum</td>
<td>Gram (+) &amp; MRSA but VRE – GI/RSA</td>
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Glycopeptides: how can we improve?
Glycopeptides: how can we improve?
Glycopeptide updated mechanism of action

dimerization and / or membrane anchoring ➔ activity
Glycopeptide updated mechanism of action in VRE

efficient binding still possible!
Glycopeptides: how can we improve?

binding to D-Ala-D-Ala
Glycopeptides: how can we improve?
Glycopeptides: how can we improve?

- Membrane anchoring, transglycosylase inhibition
- Dimerization
- Binding to D-Ala-D-Ala
Molecules in clinical development:

DESIGN
From vancomycin to oritavancin
From vancomycin to oritavancin

epi-vancosamine
From vancomycin to oritavancin

4-epi-vancosamine
▼ self-association capacity

LY264626
From vancomycin to oritavancin

lipophilic side chain
ém activity
including against resistant enterococci

Cooper et al (1996)
J Antibiot 49: 575-581
From vancomycin to telavancin
From vancomycin to telavancin

chain length offering balanced activity against MRSA & resistant enterococci
From vancomycin to telavancin

compensates the effect on the lipophilic side chain on half-life

J Antibiot 57: 326-336
From teicoplanin to dalbavancin
From teicoplanin to dalbavancin
removal of N-acetylglucosamine activity (against resistant enterococci)
From teicoplanin to dalbavancin

Malabarba & Ciabatti (2001)
Curr Med Chem 8: 1759-1773
New glycopeptides are more cationic and amphiphilic than vancomycin.
New glycopeptides are more cationic and amphiphilic than vancomycin

New chemical entities

- New mode of action and new pharmacodynamic properties
- New pharmacokinetic profile
- But also .... new potential side effects
New glycopeptides:

new mode of action and pharmacodynamic properties
## Spectrum of activity

<table>
<thead>
<tr>
<th>strain</th>
<th>resist.</th>
<th>vanco</th>
<th>orita</th>
<th>tela</th>
<th>teico</th>
<th>dalba</th>
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<tr>
<td><strong>enterococci</strong></td>
<td>susc.</td>
<td>1-2</td>
<td>0.06-0.25</td>
<td>0.5</td>
<td>0.13-0.5</td>
<td>0.125</td>
</tr>
<tr>
<td></td>
<td>VanA</td>
<td>&gt;128</td>
<td>1-4</td>
<td>4-8</td>
<td>64-&gt;128</td>
<td>0.5-&gt;128</td>
</tr>
<tr>
<td></td>
<td>VanB</td>
<td>8-128</td>
<td>0.125</td>
<td>128</td>
<td>0.125-8</td>
<td>1</td>
</tr>
<tr>
<td><strong>S. aureus</strong></td>
<td>Methi-S</td>
<td>1-2</td>
<td>1</td>
<td>0.5</td>
<td>1-8</td>
<td>&lt; 0.5</td>
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<td></td>
<td>Methi-R</td>
<td>1-4</td>
<td>1-2</td>
<td>0.5-1</td>
<td>1-8</td>
<td>0.06-1</td>
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<tr>
<td></td>
<td>GISA</td>
<td>8</td>
<td>1-8</td>
<td>2</td>
<td>8-32</td>
<td>2</td>
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<td>GRSA</td>
<td>&gt; 128</td>
<td>0.5</td>
<td>2</td>
<td>4</td>
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### Spectrum of Activity

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Activity against strains resistant to vancomycin.
Pharmacodynamic properties

slowly cidal effect

conc. dependent, bactericidal effect

Barcia-Macay et al. Unpublished
Altered membrane integrity

Higgins et al., ICAAC (2004)
New glycopeptides:

new pharmacokinetic profile
## Pharmacokinetic properties in humans

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Vanco (15 mg/kg)</th>
<th>Orita (3 mg/kg)</th>
<th>Tela (7.5 mg/kg)</th>
<th>Teico (6 mg/kg)</th>
<th>Dalba (15 mg/kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peak (mg/L)</td>
<td>20-50</td>
<td>31</td>
<td>89</td>
<td>43</td>
<td>300</td>
</tr>
<tr>
<td>Through (mg/L)</td>
<td>5-12 (24 h)</td>
<td>1.7 (24 h)</td>
<td>&lt;5 (24 h)</td>
<td>40 (168 h)</td>
<td></td>
</tr>
<tr>
<td>Protein binding</td>
<td>10-55 %</td>
<td>90 %</td>
<td>90-93%</td>
<td>90 %</td>
<td>98 %</td>
</tr>
<tr>
<td>Terminal t½ (h)</td>
<td>4-8</td>
<td>≤ 360</td>
<td>7</td>
<td>83-168</td>
<td>257 h</td>
</tr>
</tbody>
</table>

*Intermune, Inc, data on file*

*Barriere et al, ICAAC 2003*

Cellular pharmacokinetics

J774 macrophages; extracellular concentration: 25 mg/L; 24 h

Comparison with other antibiotics

J774 macrophages; extracellular concentration: 25 mg/L; 24 h
Mechanism of cellular accumulation

chloroquine
azithromycin

HRP latex beads
Mechanism of cellular accumulation

- Uptake linear over 4 h as for markers of endocytosis
- Rate of uptake similar to that of a marker of adsorptive endocytosis

Subcellular localization

MARKERS OF:
- lysosomes
- mitochondria
- membranes

![Graphs showing subcellular localization of NAGase, cytochrome c-oxidase, inosine diphosphatase, and HRP in control and treated cells.](image)
New glycopeptides:
cellular pharmacokinetics

↓

cellular pharmacodynamics
PK/PD properties of oritavancin in a model of S. aureus infected macrophages

Seral et al AAC (2003) 47 : 2283-2292
PK/PD properties of oritavancin in a model of *S. aureus* infected macrophages

Barcia-Macay et al. Unpublished
Oritavancin is the most active antibiotic against intracellular *S. aureus* among those tested so far!
Intracellular bactericidal activity is visible!
New glycopeptides:

IN VIVO DATA
animal models

Combined extra-and intracellular activity

recurrent infections, including by resistant organisms
efficacy shown in ....

Combined extra-and intracellular activity

recurrent infections, including by resistant organisms
New glycopeptides:

IN VIVO DATA
clinical studies

Combined extra-and intracellular activity

recurrent infections, including by resistant organisms
Clinical experience

complicated skin and skin structure infection caused by Gram (+) including MRSA
(phase II/III; double blind, randomized)
517 pts

Vancomycin 15 mg/kg bid
3-7 days
followed by oral cephalexin
10-14 days

Oritavancin 1.5-3 mg/kg qd
3-7 days

SUCCESS:

bacteriological
76 %

clinical
80 %
with MRSA
80 %

Wasilewski et al. ICAAC (2001)
Clinical experience

complicated skin and skin structure infection caused by Gram (+) including MRSA (phase II; double blind, randomized)
167 pts

vancomycin / penicillin

SUCCESS:

bacteriological
vancomycin / penicillin 74 %
telavancin 84 %

clinical
vancomycin / penicillin 77 % =
telavancin 80 %

with MRSA
vancomycin / penicillin 69 % <
telavancin 82 %

Stryjewski et al. IDSA meeting (2004)
Clinical experience

complicated skin and skin structure infection caused by Gram (+) including MRSA
(phase II; controlled, randomized)
62 pts

Vancomycin, ceftriaxone, cefazolin or clindamycin for 7-21 days

Dalbavancin
15 mg/kg day 1
+ 7.5 mg/kg day 8

SUCCESS:
bacteriological 64 % 73 %
clinical 76 % 94 %

New glycopeptides: cellular pharmacokinetics → cellular toxicity ?
Microscopic examination of oritavancin treated cells

Van Bambeke et al. submitted
Microscopic examination of oritavancin treated cells

fibroblasts
Lipid accumulation: concentration-effect relationships
Lipid accumulation: time effect and reversibility
Lipid accumulation: correlations

![Graphs showing correlations between oritavancin cellular concentration and lipid accumulation.](image-url)
Safety profile in humans

No major side effect in clinical trials,

BUT

• small number of patients
• appropriate techniques should be used to detect cellular alterations ....
how can a structural modification bring to a new life an old family of antibiotics?

- **new mode of action**
  - activity against resistant strains
  - bactericidal, conc.-dependent effect
    - ideal PD profile

- **new pharmacokinetic profile**
  - prolonged half-life/high prot. binding
  - high cell accumulation/intracell. activity
    - infrequent administrations
    - recurrent infections

- **new toxic effects in vitro**
  - thesaurismosis
    - safety issues
Glycopeptides: have we improved?

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Note: The red mark ( ún k) indicates an improvement.
Thanks to …

Maritza Barcia-Macay
Stéphane Carryn
Hugues Chanteux
Cristina Seral
Donatienne Tyteca
Marie-Paule Mingeot-Leclercq
Paul M. Tulkens

Francine,
Marie-Claire,
Nancy