Quinolones, macrolides, β-lactams, glycopeptides ... and a few others against resistant S. aureus

Paul M. Tulkens, MD, PhD
Françoise Van Bambeke, PharmD, PhD

Cellular and Molecular Pharmacology Unit
& Centre for Clinical Pharmacy

Catholic University of Louvain,
Brussels, Belgium

www.facm.ucl.ac.be
Why do we need **new** antistaphylococcal agents?

- rising resistance … reaching the limits of what we can give to patients …
- intrinsic PK/PD limitations of conventional glycopeptides towards *S. aureus* is severe infections
- difficulty in eradicating intracellular *S. aureus*… resulting in recurrences, relapses, and perhaps also favoring the selection/emergence of less susceptible organisms …
Intracellular infection and recurrence/relapses

In vivo importance assumed based on in vitro data

Intracellular Staphylococcus aureus. A mechanism for the indolence of osteomyelitis.
Ellington JK, Harris M, Webb L, Smith B, Smith T, Tan K, Hudson M.

Intracellular persistence of Staphylococcus aureus small-colony variants within keratinocytes: a cause for antibiotic treatment failure in a patient with Darier's disease.
von Eiff C, Becker K, Metze D, Lubritz G, Hockmann J, Schwarz T, Peters G.

Phagocytosis of Staphylococcus aureus by cultured bovine aortic endothelial cells: model for postadherence events in endovascular infections.
Hamill RJ, Vann JM, Proctor RA.
Intracellular infection and recurrence/relapses

Phagocytic and non-phagocytic cells in mastitis

Quinolones ...
Moxifloxacin is quite active against intracellular MSSA ...

2.5 log decrease at $C_{\text{max}}$ (4.5 mg/L)
Moxifloxacin is quite active against intracellular MSSA …

2.5 log decrease at $C_{\text{max}}$ (4.5 mg/L)

which is much better than oxacillin or gentamicin

Quinolones and MRSA...

Distribution of fluoroquinolone MICs for 100 MRSA isolated in 2002

<table>
<thead>
<tr>
<th>Drug</th>
<th>No. of strains with indicated MIC (mg/l)$^a$</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>≤0.25</td>
</tr>
<tr>
<td>NFLX</td>
<td>0</td>
</tr>
<tr>
<td>ENX</td>
<td>0</td>
</tr>
<tr>
<td>CPFX</td>
<td>1</td>
</tr>
<tr>
<td>TFLX</td>
<td>4</td>
</tr>
<tr>
<td>FLRX</td>
<td>0</td>
</tr>
<tr>
<td>SPFX</td>
<td>4</td>
</tr>
<tr>
<td>LVFX</td>
<td>3</td>
</tr>
<tr>
<td>GFLX</td>
<td>4</td>
</tr>
<tr>
<td>MFLX</td>
<td>4</td>
</tr>
</tbody>
</table>

Abbreviation: NFLX, norfloxacin; ENX, enoxacin; CPFX, ciprofloxacin; TFLX, tosufloxacin; FLRX, fleroxacin; SPFX, sparfloxacin; LVFX, levofloxacin; GFLX, gatifloxacin; MFLX, moxifloxacin.

$^a$ The positions of breakpoints for resistance interpreted by the NCCLS are underlined. Breakpoints of MFLX and TFLX have not been established by the NCCLS.

Moxifloxacin has the lowest MICs amongst currently available quinolones, but resistance does exist!
Yet, moxifloxacin may be quite active against intracellular HA-MRSA and VISA …
Quinolones under development: can they be better?

<table>
<thead>
<tr>
<th>quinolone</th>
<th>Range of MIC of MRSA</th>
<th>MIC 50</th>
<th>MIC 90</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moxifloxacin</td>
<td>0.03-32</td>
<td>1-2</td>
<td>4-16</td>
</tr>
<tr>
<td>WCK 771</td>
<td>0.015-4</td>
<td>0.5</td>
<td>1</td>
</tr>
<tr>
<td>DX 619</td>
<td>0.008-2</td>
<td>0.125</td>
<td>1</td>
</tr>
</tbody>
</table>

WCK 771, a new quinolone in clinical trials

DK 619, a new desquinolone in preclinical trials

β-lactams ...

![Graph showing antibiotic effectiveness against extracellular and intracellular bacteria](image)

Lemaire et al., JAC 55:897-904, 2005
Anti-MRSA cephalosporins: ceftobiprole...

- Highly resistant to beta-lactamases
- High affinity for PBP2a

- Originally discovered by Roche in the late 90's
- Activity against PBP2a is related to the hydrophobic side chain in C3 (conformational change)
- Poor solubility necessitated the design of a pro-drug form

Anti-MRSA cephalosporins (ceftobiprole)...

- MIC range: 0.25-0.5 mg/L for MSSA
- 0.25-2 mg/L for MRSA
- 0.5-2 for SCV
- bactericidal
- synergistic with aminoglycosides

- FDA fast track designation for
  - the treatment of complicated skin and skin structure infections due to MRSA
  - a second indication in the treatment of hospital-acquired (nosocomial) pneumonia, including ventilator-associated pneumonia due to suspected or proven MRSA

- Excellent tissue penetration and powerful activity in models of
  - osteomyelitis
  - foreign-body infection
  - aortic valve endocarditis

- No available data on intracellular activity...
New carbapenems active on MRSA

MIC range:
- MSSA: 0.06-2 mg/L
- MRSA: 0.25-32 mg/L

*PZ-601 is also known as SM-216601 and SMP-601

MIC range for MRSA:
- PZ-601: 0.03-4 mg/L
- IMI: 0.25-32 mg/L
- OXA: 4-128 mg/L

→ ongoing Phase I trials
New glycopeptides (oritavancin, telavancin, dalbavancin)...

Hemi-synthetic derivatives derived from

- **vancomycin**
  - Oritavancin
  - Phase III, Theravance → Astellas

- **teicoplanin**
  - Telavancin
  - Phase III, Theravance → Astellas
  - Dalbavancin
  - Awaiting FDA approval, Vicuron → Pfizer

Telavancin (and oritavancin) new modes of action …

- Possibility of dimerization
  - potential increase of intrinsic activity against D-Ala-D-Ala displaying organisms (MSSA, MRSA, VISA)

- Membrane destabilization effects…
  - strong concentration-dependent bactericidal effect (all strains ...) *

Beauregard et al., AAC 1995; 39:781-85
Telavancin …


this causes increase in bacterial membrane permeability
Time-kill of telavancin vs. vancomycin against MSSA and MRSA

vancomycin  telavancin

<table>
<thead>
<tr>
<th>MSSA</th>
<th>MSSA</th>
</tr>
</thead>
<tbody>
<tr>
<td>ATCC25923</td>
<td>ATCC25923</td>
</tr>
<tr>
<td>ATCC29213</td>
<td>ATCC29213</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>MRSA</th>
<th>MRSA</th>
</tr>
</thead>
<tbody>
<tr>
<td>ATCC33591</td>
<td>ATCC33591</td>
</tr>
<tr>
<td>ATCC43300</td>
<td>ATCC43300</td>
</tr>
</tbody>
</table>

Barcia-Macay et al., JAC, in the press
Time-kill of telavancin vs. vancomycin against MSSA and MRSA

**vancomycin**  
**telavancin**

### MSSA

<table>
<thead>
<tr>
<th>ATCC25923</th>
<th>ATCC25923</th>
<th>ATCC29213</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image1" alt="Graph" /></td>
<td><img src="image2" alt="Graph" /></td>
<td><img src="image3" alt="Graph" /></td>
</tr>
</tbody>
</table>

### MRSA

<table>
<thead>
<tr>
<th>ATCC33591</th>
<th>ATCC33591</th>
<th>ATCC43300</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image4" alt="Graph" /></td>
<td><img src="image5" alt="Graph" /></td>
<td><img src="image6" alt="Graph" /></td>
</tr>
</tbody>
</table>

- **control**
- **1x MIC**
- **10 x MIC**
- **Cmax**

3 log decr.:
- **vanco**: ~ 15h
- **TLV**: 2-10h

*Barcia-Macay et al., JAC, in the press*
Time-kill of telavancin vs. vancomycin against VISA and VRSA,

vancomycin

VISA

<table>
<thead>
<tr>
<th>Graph 1</th>
<th>Graph 2</th>
<th>Graph 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>NRS23</td>
<td>NRS23</td>
<td>NRS52</td>
</tr>
<tr>
<td>Δ log CFU from time 0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>time (h)</td>
<td>0</td>
<td>6</td>
</tr>
<tr>
<td>0</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>-1</td>
<td>-2</td>
<td>-3</td>
</tr>
<tr>
<td>-4</td>
<td>-5</td>
<td></td>
</tr>
</tbody>
</table>

telavancin

VRSA

<table>
<thead>
<tr>
<th>Graph 4</th>
<th>Graph 5</th>
</tr>
</thead>
<tbody>
<tr>
<td>VRS1</td>
<td>VRS2</td>
</tr>
<tr>
<td>Δ log CFU from time 0</td>
<td></td>
</tr>
<tr>
<td>time (h)</td>
<td>0</td>
</tr>
<tr>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>-1</td>
<td>-2</td>
</tr>
<tr>
<td>-4</td>
<td>-5</td>
</tr>
</tbody>
</table>

Barcia-Macay et al., JAC, in the press
Time-kill of telavancin vs. vancomycin against VISA and VRSA,

<table>
<thead>
<tr>
<th>vancomycin</th>
<th>telavancin</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>VISA</strong></td>
<td></td>
</tr>
<tr>
<td><a href="#">Graphs showing bacterial growth inhibition</a></td>
<td><a href="#">Graphs showing bacterial growth inhibition</a></td>
</tr>
</tbody>
</table>

3 log decreas:
- vanco: ~ 15h
- TLV: 2-10h

Barcia-Macay et al., JAC, in the press
Televancin
dual mode of action?

3 h kill curves

Niagara Falls, Canada, 11-10-06

New anti-staphylococcal agents

Barcia-Macay et al., JAC, in the press
Telavancin intracellular accumulation and subcellular disposition

- **Uptake**
  - Rate: $0.86 \pm 0.05 \mu g/mg prot/h$

- **Efflux**
  - Rate: $-0.15 \pm 0.03 \mu g/mg prot/h$

**Graphs:**
1. Cellular concentration (\(\mu g/mg\ prot\)) vs. time (h)
2. Cellular concentration (\(\mu g/mg\ prot\)) vs. extracellular concentration (mg/L)
3. Percentage of recovered constituent vs. density (g/cm$^3$)

*Van Bambeke et al., unpublished*
Intracellular activity of telavancin vs. vancomycin:
- MSSA
- MRSA

24h CFU at $C_{max}$:
- vanco: $\sim 0.5$ log
- TLV: $\sim 2$ log
New anti-staphylococcal agents

Niagara Falls, Canada, 11-10-06

Barcia-Macay et al., JAC, in the press

vancomycin  telavancin

VISA

\[ \Delta \log \text{CFU from time 0} \]

\[ \text{log concentration (X MIC)} \]

- vanco: static
- TLV: \( \sim 1.2 \log \)

Telavancin vs. vancomycin:
- VISA
- VRSA

VRSA

24h CFU \( \Rightarrow \) at \( C_{max} \):
- vanco: static
- TLV: \( \sim 1.2 \log \)
Telavancin intracellular activity in comparison with other drugs…
Microbiology, pharmacokinetics and clinical indications under investigation for the new glycopeptides

<table>
<thead>
<tr>
<th></th>
<th>telavancin</th>
<th>oritavancin</th>
<th>dalbavancin</th>
</tr>
</thead>
<tbody>
<tr>
<td>MIC MRSA</td>
<td>0.125-1</td>
<td>0.125-4</td>
<td>0.06-1</td>
</tr>
<tr>
<td>VISA</td>
<td>0.5-4</td>
<td>1-8</td>
<td>2</td>
</tr>
<tr>
<td>VRSA</td>
<td>2</td>
<td>0.5</td>
<td>inactive</td>
</tr>
<tr>
<td>Half-life</td>
<td>7 h</td>
<td>18 h ($\beta$)</td>
<td>140-300h $\rightarrow$ once-a-week !!</td>
</tr>
<tr>
<td></td>
<td>$\rightarrow$ once-a-day</td>
<td>360 h ($\gamma$)</td>
<td></td>
</tr>
<tr>
<td>Tissular and cellular distribution</td>
<td>yes</td>
<td>yes</td>
<td>yes</td>
</tr>
<tr>
<td>Clinical efficacy</td>
<td>skin &amp; soft tissue</td>
<td>skin &amp; soft tissue</td>
<td>skin and soft tissue catheter-related bloodstream infections</td>
</tr>
<tr>
<td></td>
<td>HAP</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Activity in animal models</td>
<td>endocarditis</td>
<td>endocarditis</td>
<td>endocarditis</td>
</tr>
<tr>
<td></td>
<td>meningitis</td>
<td>meningitis</td>
<td>pneumonia</td>
</tr>
<tr>
<td></td>
<td></td>
<td>catheter infections</td>
<td>disseminated infections</td>
</tr>
</tbody>
</table>
Macrolides ...

- Erythromycin, clarithromycin, azithromycin, ... Are no longer real options...
- Telithromycin, disregarding liver toxicity, is active only against inducible MLSB-resistant strains...
- Extracellular and intracellular activities are essentially static...

---

MSSA ATCC25923

![Graph showing extracellular and intracellular activities of different antibiotics against MSSA ATCC25923.](image)
SYNERCID® = quinupristin + dalfopristin

SA blocks peptide bound formation

SB blocks the path of the nascent peptide
SYNERCID®

- originally discovered and developed by Rhône-Poulenc (France)
- European (mutual recognition) and FDA approval in the late 90's for:
  - complicated skin and soft tissues infections by MSSA/strepto
  - bacteremia due to VR *E. faecium* (fast track at FDA)
  - efficacy also demonstrated in nosocomial pneumonia (= vanco; lower success if MRSA in both groups)
- abandoned in early 2000's because of
  - side effects (rash; infusion-site inflammation; pain and edema; thrombophlebitis …) and inhibition of cytochrome P450 3A4
  - difficulties of production in large quantities
  - loss of interest after the merge of Rhône-Poulenc with Hoechst-Marion-Roussel to form AVENTIS…

- presently commercialized at a low scale by
  - Nordic Pharma in Europe
  - King Pharmaceuticals in the US
SYNERCID® does not behave too badly for intracellular S. aureus...
Tigecycline...

- truly made to resist efflux-mediated resistance in Gram(-) bacteria
- broad spectrum including MRSA (MIC < 2 mg/L) and VISA
- tet(M) [ribosomal protection] or tet(K) [efflux] have no discernible effect on MICs (AAC 2006 Feb;50(2):505-10).
- large tissue accumulation (Vd=7-9L/kg) ➔ low C\textsubscript{max} (1.5 mg/L; 70-80% protein-bound).
- approved by the FDA in June 2005 (and by the EME in April 2006) for
  - complicated skin infections, skin-structure infections (complicated skin and soft tissue infections);
  - intra-abdominal infections (complicated intra-abdominal infections)
  - both at 100 mg IV (initial) followed by 50 mg/12h IV
  - bkpt for \textit{S. aureus} (FDA & EUCAST): \textit{S} \leq 0.5 mg/L
Tigecycline... why a breakpoint $\leq 0.5$ mg/L?

Distributions of MIC as submitted to EUCAST

Probability of target attainment of a suitable AUC/MIC ratio ($\geq 7$) for the recommended dosage

http://217.70.33.99/Eucast2/SearchController/regShow.jsp?id=7563

Wyeth: data on file
Tigecycline… why a breakpoint $\leq 0.5$ mg/L?

Putting all together ….

$\Rightarrow$ you will be effective as long as the MIC remain $\leq 0.5$ mg/L
Tigecycline and intracellular *S. aureus*…

accumulation in PMN: about 20-30 fold

activity in PMN: about $1 \log_{10}$ at 1 mg/L for *S. aureus* ATCC 29213 (MIC = 0.25 mg/L)

Daptomycin …

- very bactericidal towards Gram (+) organisms through membrane destabilization (no need of proteinaceous receptor!)
- BUT intrinsically inactive against Gram(-) due to LPS protection
- spare mammalian cells because they lack phosphatidylglycerol (critical for binding to Gram(+) membranes)

- got a fast track registration in the US because of activity against vancomycin-resistant enterococci (VRE)
Setting Daptomycin breakpoint for \textit{S. aureus}...

Figure 1: Daptomycin MIC distribution and probability of target attainment for \textit{S. aureus}

- **Daptomycin / Staphylococcus aureus**
- Antimicrobial wild type distributions of microorganisms – reference database
- EUCAST

358 is the minimal value for efficacy based on animal and human data.
Is there a (real) place for daptomycin?

- PK/PD-based breakpoint (as per EUCAST): 1 mg/L

- registered in USA/Europe for complicated skin and soft tissue infections
  (4 mg/kg administered once every 24 hours for 7-14 days)

- New registration in USA for bacteremia

- potential issues:
  - no clinical evidence of superiority to vancomycin for vancomycin-susceptible strains;
  - VISA strains tend to have MIC > 1 mg/L
  - poorly efficient in pneumonia (inactivated by surfactant)
  - safety concerns with higher dosages (myopathy);
  - price (about 3-4 x vancomycin …)
MX-2401: a (close) cousin of daptomycin?

MX-2401: semi-synthetic derivative of amphomycin...

**In vitro** demonstration of efficacy in models of infections (including pneumonia) by *S. aureus* and *S. pneumoniae*

Target indications: cSSTI pneumonia

MIC for MRSA: 0.25 mg/L

Dab-9 Amine

Figure 1: Amphomycin lipopeptide core.

NO surfactant effect?

MIGENIX has an agreement with the Government of Canada under the Technology Partnership’s Canada program which is funding 26% of eligible costs (up to $9.3 million) for the development of MX-2401.
Another membrane-active agent ...

CSA-13

MIC range for MRSA: 1 mg/L
VISA: 1 mg/L
VRSA: 1 mg/L

Highly bactericidal, but as toxic for eucaryotic as for procaryotic membrane ....

Also active in biofilms; will be further developed for topical applications

Lemaire et al., ICAAC 2006 A 0633
New diaminopyridines active on MRSA

Iclaprim (AR-100): hospital use – cSSTI (Phase II/III)

MIC for MRSA:
- Iclaprim: 0.06 mg/L
- AR-709: 0.25-1 mg/L
- TMP: 1- >16 mg/L
active on TMP-resistant strains

AR-709: community use?

Table 4: Binding affinities of AR-709 and TMP

<table>
<thead>
<tr>
<th>DHFR Enzyme</th>
<th>Ka (x10^7 M^-1)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>AR-709</td>
</tr>
<tr>
<td>Binary complex with NADPH</td>
<td></td>
</tr>
<tr>
<td>S. pneumoniae ATCC 49619 wild-type, TMP_R</td>
<td>&gt;684</td>
</tr>
<tr>
<td>S. pneumoniae I100L, TMP_R</td>
<td>92</td>
</tr>
<tr>
<td>S. aureus NCTC 8325, wild-type, TMP_R</td>
<td>109</td>
</tr>
<tr>
<td>S. aureus F98Y, TMP_R</td>
<td>2.5</td>
</tr>
</tbody>
</table>

ICAAC 2006 F1 1959
Revamping older drugs (and rediscovering targets...)

Retapamulin

MIC for MRSA: 0.12 mg/L

Topic application for cSSTI; accepted for review by the US Food and Drug Administration in February 2006

Retapamulin binds to the bacterial ribosome with high affinity, inhibits ribosomal peptidyl transferase activity, and partially inhibits the binding of the initiator tRNA substrate to the ribosomal P-site. Taken together, these data distinguish the retapamulin mode of action from that of other classes of antibiotics. This unique mode of action may explain the lack of clinically relevant, target specific cross-resistance of retapamulin with antibacterials in current use.
New target: FabI

- FabI (enoyl-ACP reductase) catalyzes the final step in FASII chain elongation cycle
- Different than the mammalian system (FASI)
- FabI is essential for bacterial growth and survival

- Primary mechanism of action of API-1252 is via inhibition of lipid biosynthesis
- Selective for inhibition of acetate incorporation
  - 52% inhibition at 20 minutes and 75% inhibition at 60 minutes
New target: FabI

API-1252

MIC range for MRSA: < 0.002-0.016 mg/L
VISA: 0.03-0.06 mg/L
VRSA: < 0.008-0.25 mg/L

Time- and concentration-dependent in
in vitro pharmacodynamic models

AUC-dependent in vivo

Relationship between AUC\textsubscript{free}/MIC and change in bacterial
density at 24 hours following treatment with API-1252.
Will this be successful?