Clinical activity of anti-Gram-positive agents against methicillin-resistant Staphylococcus aureus

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Current concerns about multiresistance and a diminishing antibiotic pipeline are mainly addressed to Gram-negative bacteria. The greatest fear within the Gram-positive arena is vancomycin-resistant Staphylococcus aureus. Its epidemiology and clinical presentation give cause for concern, but so far its impact has been strictly limited. While this may change, the loss of glycopeptides as a treatment option may not, in fact, be all bad news.

Keywords: vancomycin-resistant staphylococci, linezolid, daptomycin

Introduction

Extreme, extensive and pan resistance are terms commonly used to describe the increasing problems of multiresistance in Gram-negative bacteria.1,2 What exactly is the situation in Gram-positive organisms? Do these terms have any applicability? Certainly in the 1980s and 1990s methicillin-resistant Staphylococcus aureus (MRSA) and vancomycin-resistant enterococci (VRE) seemed to be among the main resistance issues for clinicians. Arguably though, the development of new agents has ameliorated the situation, although a glance at the number of old agents available and still reasonably active for many Gram-positive species does question how bad a situation we were ever in. Now, with the development of plasmid-mediated resistance to aminoglycosides, quinolones, cephalosporins and carbapenems, often linked on integrons and pathogenicity islands, Gram-negative resistance is again firmly on the agenda, exacerbated by the lack of a new drug pipeline. Within Gram-positive bacteria, clearly the biggest concern is vancomycin resistance. This takes various forms. Of most concern is vancomycin-resistant S. aureus (VRSA), which can arise following the transfer of the vanA gene encoding high-level glycopeptide resistance from Enterococcus faecalis.

VRSA

There are only 11 well-characterized VRSA isolates reported in the English literature, all of them MRSA, and 9 of these are from the USA,3,4 with 7 of these coming from the state of Michigan. The first reported VRSA (USA1) was from Michigan (2002), with the second and third isolates being reported from Pennsylvania (2002) and New York (2003), respectively. The next five isolates were from Michigan (2005–7). Further isolates (2008) have been reported from India (Calcutta)5 and Iran.6 All 11 isolates had PCR-confirmed vanA. There have been several other reports of high-level vancomycin resistance in S. aureus from India, Iran and elsewhere, but none with vanA confirmed by PCR.7,8 All of the prior USA strains have been accompanied by detailed clinical and infection control data, but there is little such information for the Indian and Iranian isolates, which were from surveys of several hundred clinical isolates. It is known that the Indian strain was isolated from a skin lesion of an outpatient and, unusually, was ciprofloxacin susceptible. The Iranian strain was isolated from a post-operative wound of a diabetic cardiac surgery patient. In common with all the USA strains, it seems as though there was no systemic spread of the organism in these two cases and none of the 11 patients was severely ill with the organism.

Where information was available, most infections were characterized by short-term carriage of the organism, good response to treatment and no spread to other patients, albeit in the face of strict infection control. Published data on susceptibility of the nine USA strains suggested all were susceptible to linezolid, although the MIC for one of the strains was on the breakpoint (4 mg/L).7 Eight of the nine strains were susceptible to daptomycin, quinupristin/dalfopristin and rifampicin and two strains were resistant to tetracycline and trimethoprim/sulfamethoxazole.9 In addition, we know that the Indian strain was gentamicin susceptible and co-trimoxazole and rifampicin resistant and the Iranian strain was susceptible to linezolid, tetracycline and rifampicin. The most serious clinical presentations seem to have been cases of necrotizing fasciitis and osteomyelitis. Otherwise, infections were more minor, of skin soft tissue, and in one (the New York strain case), colonization of urine in a patient with a nephrostomy.

There are a lot of questions that need answers, such as will strains emerge globally, will they cross-infect other patients, will the vanA gene spread, and finally, why the relative abundance of strains from Michigan? It is indeed fortunate that
none of the strains was particularly virulent. Expression of resist-
ance would seem to be associated with a high cost, but if not 
expressed, its biological cost may be minimal, so dissemination 
is a distinct possibility. Also, resistance has so far evolved 
through at least three different mechanisms, potentially with 
involvement of coagulase-negative staphylococci. Emergence 
of USA1 involved conjugation and transposition of Tn1546 vanA 
operon to an MRSA plasmid with self-replicatory ability. USA2 
had a plasmid with both staphylococcal and enterococcal 
sequences, while USA3–7 had enterococcal plasmids able to 
replicate in S. aureus. Typing data are available from just six 
USA strains. All were sequence type (ST) 5 (five USA100 and a 
single isolate of US800).11

If they were independent evolutionary occurrences, then why 
were so many of the isolates from Michigan? Michigan may have 
a peculiar propensity for producing these strains due to several 
factors, namely the high prevalence of an E. faecalis donor carry-
ing a broad host range Inc18 plasmid; a relatively high proportion 
of patients carrying MRSA, often with co-existent diabetes or 
renal failure, which can ‘enrich’ for MRSA carriage; and high van-
comycin use.10 It is likely, however, that similar conditions occur 
elsewhere. More worryingly, an ascertainment bias may have 
ocurred due to awareness and expertise. Current automated 
systems in widespread use are well able to detect known 
strains, so this may not be such an issue now.12 Recent guidance 
on cessation of agar disc diffusion for glycopeptide susceptibility 
testing should also help to improve detection capabilities.9

It seems improbable that more strains of VRSA will not 
emerge and spread, but perhaps neither event will happen 
quickly. So far neither MRSA nor S. aureus seem to have discov-
ered the correct ingredients for clonal spread of VRSA or horizon-
tal gene transfer of vanA, despite vancomycin being in clinical 
use for more than 50 years with sustained high, if not increasing, 
use for the past 20 years.

Such a supposed doomsday scenario is greatly feared and VRSA 
is a classic ‘alert’ organism. But are vancomycin, or teicoplanin for 
that matter, really such good, indispensable drugs? Actually this is 
probably not the case.13 Even in the few years after its introduction 
in 1955 vancomycin was deemed toxic, and from 1961 onwards, 
after the marketing of the first semisynthetic penicillins, it rapidly 
became a reserve drug. Obviously, with the worldwide surge in 
MRSA in the 1990s, vancomycin in a more purified form found 
extensive new uses, but this was still associated with significant 
nephrotoxicity, development of glycopeptide-intermediate S. 
aureus (GISA) and heterogeneous glycopeptide-intermediate S. 
aureus (hGISA) strains. In the last few years MIC creep or leap 
have resulted in many MRSA isolates having vancomycin MICs at 
or just below the breakpoint of 2 mg/L.14

Actually the situation is even worse than this. Although the 
breakpoint has recently been lowered from 4 to 2 mg/L,15 the 
clinical and pharmacokinetic–pharmacodynamic (PK–PD) evi-
dence is that the breakpoint should be 0.5 or 1 mg/L, thus clas-
sifying most MRSA [and methicillin-susceptible S. aureus (MSSA)] 
isolates as resistant in many published series.14,16 The clinical 
 evidence comes from at least six observational studies of serious 
MRSA infections, mainly bacteraemias, but also a signifi-
cant number of pneumonia cases. These studies demonstrate a 
clinical breakpoint of 0.5 or 1 mg/L.17 Studies that used Etest to 
determine the MIC had a clinical breakpoint of 1 mg/L and those 
that used reference method broth dilution had a lower 
breakpoint of 0.5 mg/L, perhaps because broth dilution 
methods, with a lower inoculum (10^4–10^5 cfu), are less likely to 
detect hGISA mutants than the higher inoculum of the Etest.18 This conclusion can only be tentative until we get 
better clinical data, but it is backed up by the available PK–PD 
data suggesting that only 60% of patients with normal renal 
function and infected with a strain with a vancomycin MIC 
value of 1 mg/L would achieve the target AUC:MIC ratio of 400, 
even with high-dose vancomycin (potentially nephrotoxic).19

The data are complicated, however, by a tendency for lower 
doses of vancomycin in the earlier studies, which were the 
to use reference antimicrobial susceptibility testing (AST) 
methods. Moreover, the PK–PD target of 400 seems to be a 
consensus based on just a couple of studies, one human and 
one animal, although it has to be said that the limited evidence 
could argue for even higher AUC:MIC ratios as a target if 
bactericidal activity is desired.17,19 This may well be desirable, 
as vancomycin and teicoplanin are renowned for very slow 
cidal activity and, at least in immunosuppressed patients and 
for the treatment of bacteraemia and endocarditis, cidalinity 
usally considered a desirable feature. Finally, and paradoxically, 
the most recent published clinical study in this area argues for a 
breakpoint of 1 mg/L but used reference broth dilution to estab-
lish MICs!20

Other anti-MRSA drugs

In light of all this, and the well-established inferiority of vanco-
mycin compared with β-lactams in the treatment of MSSA infec-
tion,13,14 it is pertinent to ask if the loss of vancomycin and 
teicoplanin, whether from the march of VRSA or (more subtly, 
but more likely) from MIC creep or leap, is such a doomsday 
scenario? With the development of several exciting new agents 
in this field and the retained activity of older agents such as 
co-trimoxazole and tetracycline against most strains of MRSA, 
there do seem to be good alternatives. Admittedly there is 
little published clinical data to support the use of these older 
agents for the treatment of serious staphylococcal infection,21,22 
but following from a retrospective cohort study that showed 
co-trimoxazole had a safety and efficacy profile similar to that 
of vancomycin,23 there is one ongoing trial comparing 
co-trimoxazole with vancomycin for MRSA bacteremia.24 Tige-
cycline (currently the only available intravenous tetracycline 
derivative in the UK) is a potentially useful drug for certain 
types of MRSA infection, although its low blood levels probably 
preclude its use for primary bacteremia.25,26 More data are 
needed, however, particularly on the optimum dosing schedules.

Newer anti-MRSA drugs

Despite a lot of activity in anti-Gram-positive drug development 
over the past decade, only linezolid, daptomycin and, very 
recently, telavancin and ceftaroline have successfully negotiated 
the regulatory hurdles. Linezolid and daptomycin have been in 
wide spread clinical use for several years, and some resistance 
mechanisms have been identified, although surveillance 
systems provide reassurance that they are not yet wide-
spread.27–32 Although telavancin is a glycopeptide, it is much 
more rapidly bactericidal than the older glycopeptides and
Measures of resistance to daptomycin and linezolid.35

- Sequential mutations lead to stepwise reduction in susceptibility
- 
- \textit{mprf} (membrane synthesis)—less binding of daptomycin through Ca
- 
- \textit{yyGC} (sensor histidine kinase)—may be another daptomycin target
- 
- \textit{rpoB}
- 
- \textit{rpoC} ? Alter transcription of key genes
- 
- \textit{dlt} operon

Linezolid resistance mechanisms28–32

<table>
<thead>
<tr>
<th>Mutations in domain V of 23SrRNA gene of 50S ribosomal subunit</th>
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<tbody>
<tr>
<td>e.g. G2576T</td>
</tr>
<tr>
<td>G2242A</td>
</tr>
<tr>
<td>G2603T</td>
</tr>
<tr>
<td>T2504A</td>
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<tr>
<td>T2500A etc</td>
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<tr>
<td>Cfr methyl transferase @ 2503 (transferable)</td>
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Figure 1. Described mechanisms of resistance to daptomycin and linezolid.35

seems to have useful activity against VRSA, GISA and hGISA. Time will tell how quickly resistance develops.33 Ceftraroline is a novel cephalosporin with broad-spectrum activity against Gram-positive pathogens, including MRSA. Its activity against MRSA is attributed to its ability to bind to penicillin-binding protein (PBP) 2a with high affinity and inhibit the biochemical activity of PBP2a more efficiently than other presently available β-lactams.34

Figure 1 lists the daptomycin and linezolid resistance mechanisms that have been described. For daptomycin, these are poorly understood, but can involve trapping of the drug in a thickened cell wall and loss of target binding affinity. There is some cross-resistance with glycopeptides due to this cell wall trapping, so it is important to check the daptomycin MIC if the strain being treated has had prior exposure to glycopeptides. Emergence of resistance during treatment is also of concern if there is a high inoculum infection or abscess with no possible drainage. There were a particularly alarming number of such cases in the daptomycin registration study of bacteraemia and endocarditis.35 Subsequent cases have been described, although probably with less frequency than might have been anticipated following this original study.36 Although the strains were technically resistant to daptomycin by breakpoint criteria, the drug usually retained its bactericidal activity. Doses higher than the original 6 mg/kg used in the above study may well reduce the emergence of resistance, and in surveillance studies, significant levels of daptomycin resistance are not recorded.

Linezolid resistance can be due to sequential mutations, like daptomycin resistance, but of most concern is potential transferable resistance through the \textit{cfr} gene, which may be linked to pleuromutilin resistance.37 Outbreaks of MRSA infection in intensive care units (ICUs) owing to high linezolid use and mediated by the \textit{cfr} gene have been described, but they seem to have been controlled by a combination of reduced linezolid use and good infection control practices.37 Of more concern perhaps is the carriage of this gene by coagulase-negative staphylococci with the potential for transfer to MRSA. Nevertheless, current surveillance systems continue to describe very little linezolid resistance. It remains to be seen whether newer oxazolidinones in development will have clinically significant improved activity against MRSA carrying the \textit{cfr} gene.38 Consecutive isolates of linezolid-resistant (VanB) Enterococcus faecium from one patient demonstrated the dynamic process of linezolid resistance due to G/T mutation at position 2576 in the genes coding for 23S rRNA. Here there was complete reversion of resistant alleles back to wild type (susceptible) in consecutive isolates.31 Newer oxazolidinones (ranbezolid and radezolid) that can overcome the ribosomal binding issues of linezolid-resistant Gram-positive pathogens, particularly enterococci and pneumococci, are in development.32

Tigecycline, a glycylcycline derivative of tetracycline, is very broad spectrum, including most MRSA. It is a useful agent for skin and soft tissue infection (SSTI) and intra-abdominal infections where multiresistant bacteria are involved, and can be used as a second-line anti-MRSA agent in these situations. Resistance in MRSA has rarely been described thus far.32,39,40

Finally, topical and systemic antibiotics are often used to decolonize MRSA carriers. With the advent of high-level plasmid-mediated mupirocin resistance, this will become a bigger issue. Currently in Glasgow, 15% of MRSA bloodstream isolates are mupirocin resistant (G. Edwards, MRSA Reference Laboratory, Glasgow, personal communication). Trimethoprim is sometimes used for MRSA decolonization, but this can lead to high levels of resistance (Hunt AC, Edwards B, Girvan EK, Cosgrove B, Edwards GFS, Gould IM, manuscript in preparation). Similarly, fusidic acid and rifampicin are often used in this setting, but high levels of resistance mutations dictate they always be used as part of a combination.

Drugs in development

Dalbavancin40 and oritavancin41 have both failed FDA approval. Dalbavancin, a glycopeptide with a very prolonged half-life, has less than 5 years of patent left, so may not have further trials. Oritavancin is being restudied to include more MRSA patients. It looks to be the most active glycopeptide against strains containing vanA. Ceftrarople, another cephalosporin with significant
activity against MRSA due to altered PBP2 affinity, is delayed in its regulatory approval because of issues of trial quality. Finally, ica-prim, a folate antagonist, did not achieve non-inferiority in an SSTI trial when compared with linezolid.\textsuperscript{40}

**Conclusions**

In conclusion, the demise of vancomycin and teicoplanin is well heralded, but perhaps not quite accomplished yet, although surely it must be soon. However, this will not be the doomsday event that it once would have been, with existing and potential new drugs, both less toxic and potentially more efficacious, likely to replace them. Nevertheless, if MRSA is not controlled around the world, we will have to use these new drugs wisely or resistance is likely to become a clinical problem rather than just a curiosity.

**Transparency declarations**

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