In vitro susceptibility of Gram-positive pathogens to linezolid and teicoplanin and effect on outcome in critically ill patients

A. Peter R. Wilson1*, Jorge A. Cepeda1, Samantha Hayman1, Tony Whitehouse2, Mervyn Singer2 and Geoffrey Bellingan2

1Department of Clinical Microbiology, University College London Hospitals, 46 Cleveland Street, London W1T 4JF, UK; 2Bloomsbury Institute of Intensive Care Medicine, Department of Medicine, UCL Gower Street, London WC1E 6BT, UK

Received 17 March 2006; returned 14 April 2006; revised 5 May 2006; accepted 11 May 2006

Objectives: To determine the prevalence of teicoplanin and linezolid resistance amongst Gram-positive pathogens isolated in the intensive care unit (ICU) and the impact of any resistance on clinical outcome.

Methods: Gram-positive isolates were collected from two critical care units over 1 year. All patients were screened weekly for methicillin-resistant Staphylococcus aureus (MRSA). Susceptibility to teicoplanin and linezolid was tested by Etest. The length of hospital and critical care unit stay and the use of antibiotics in each patient were recorded.

Results: Reduced susceptibility to teicoplanin (MIC ≥ 16 mg/L) was found in 21 [3.3% (95% CI 2.0–5.0%)] of 643 strains of MRSA versus none of 374 methicillin-susceptible S. aureus (MSSA) [<0.3% (95% CI 0–0.9%)]. Of 49 enterococci 3 were teicoplanin-resistant. All Gram-positive isolates were susceptible to linezolid. The length of treatment with teicoplanin and outcome of patients infected with these strains were similar to that of susceptible strains. MRSA was a more common cause of infection than MSSA but a less frequent colonizer.

Conclusions: Resistance to teicoplanin remains at a comparatively low level and there was no clear relationship between susceptibility and outcome in this critically ill population. There was no resistance in Gram-positives to linezolid but this should be kept as a reserve antibiotic to maintain its activity.

Keywords: critical care, MRSA, glycopeptides

Introduction

Heterogeneous resistance of methicillin-resistant Staphylococcus aureus (MRSA) to teicoplanin is increasingly common, particularly in critical care, and isolates appear susceptible on disc testing.1 Etest susceptibility testing is the most reliable method of detecting these organisms but is expensive. Linezolid has good activity against glycopeptide-resistant strains including staphylococci, enterococci and streptococci. As oxazolidinones are chemically unrelated to any other available antibiotic, cross-resistance is not expected. The observed spontaneous mutation rate of staphylococci after exposure to linezolid at twice the MIC has been reported to be as low as <1 × 10−9.2 Nevertheless, the first cases of linezolid-resistant MRSA have been reported.2,3

The clinical importance of intermediate resistance to teicoplanin is unclear and the use of linezolid has been limited by adverse effects and cost. A randomized controlled trial of treatment of serious Gram-positive infection was undertaken comparing teicoplanin and linezolid over 1 year at University College London Hospitals.4 During the study all Gram-positive isolates from microbiological specimens or screening swabs were collected to determine the prevalence of resistance to each antibiotic and any relationship to outcome.

Materials and methods

From June 2000 to June 2001 Gram-positive organisms isolated from all patients admitted for more than 48 h to the general intensive care units (ICUs) at University College Hospital NHS Trusts were analysed. The study was in support of a randomized double-blind trial comparing linezolid and teicoplanin in the treatment of Gram-positive infections.4 Ethics approval, including for consent
Susceptibility of Gram-positive pathogens in ICU

arrangements, was obtained from the UCLH Committee. All anti-
biotic treatment, duration and doses were recorded. The incidence 
of staphylococcal colonization and infection were determined from (i) screening samples (nose and groin swabs) taken on ad-
mission, weekly thereafter and at discharge and (ii) samples taken as clinically indicated, e.g. sputum, wound and blood cultures. Colon-
ization with MRSA was defined by the presence of MRSA in nose, 
groin, sputum, wound or other sites that did not require treatment 
with an appropriate antibiotic. Infection was described as the pres-
ence of the pathogen in any clinical isolate coinciding (within 5 days) 
with treatment with an appropriate antibiotic (e.g. for MRSA, 
glycopeptide, linezolid or rifampicin/trimethoprim). S. aureus 
was detected using nutrient broth with salt (2.5% with aztreonam 
75 mg/L) incubated at 37°C overnight. After overnight incubation 
at 37°C, the salt broth was subcultured onto mannitol salt agar 
(without oxacillin). Suspect colonies on the original plate at 24 
and 48 h were identified and subcultured on a blood agar plate 
with oxacillin disc and incubated at 30°C overnight. The salt 
broth subculture was incubated for a further 24 h and re-examined 
on day 4. Patients staying more than 48 h in the ICU had swabs taken 
on discharge from ICU to identify MRSA acquisition in the ICU. 

All Gram-positive isolates from clinical samples and screens were 
tested for teicoplanin and linezolid susceptibility. Initially suscep-
tibility was determined on all isolates by the standard disc test (line-
zolid disc 10 µg, teicoplanin 30 µg) and then Etest (AB Biodisk, 
Solna, Sweden). In accordance with the manufacturer’s instructions, 
a 0.5 McFarland inoculum was used on Mueller–Hinton agar for linezolid, reading to the hazy zone edge at 80% inhibition at 
16–18 h. For teicoplanin, a 2.0 McFarland inoculum was used on brain–heart infusion agar, reading to the point of complete inhibition 
at 48 h. The breakpoints for linezolid and teicoplanin were 4 mg/L \( ^{3} \) and 8 mg/L, respectively. The Antibiotic Resistance Monitoring 
and Reference Laboratory confirmed teicoplanin resistance in MRSA 
isolates by agar dilution (Dr D. Livermore, Health Protection 
Agency, Centre for Infections, Colindale, UK). 

Coagulase-negative staphylococci or enterococci were judged 
clinically significant by standard criteria, i.e. patient requiring anti-
biotic treatment.

Results

Antibiotic susceptibility

A total of 2569 specimens (including 179 blood, 110 sputum/ 
tracheal aspirate and 117 wound specimens) were tested from 
917 patients. S. aureus was isolated from 1017 specimens. Of 
2023 nose/groin screening swabs, 689 (34.0%) produced a growth of 
S. aureus. The MICs of linezolid were consistently below the 
breakpoint for all isolates collected [MRSA \( n = 643, \text{MIC}_{90} \) 
2.0 mg/L, range 0.4–4.0; methicillin-susceptible S. aureus 
(MSSA) \( n = 374, \text{MIC}_{90} 1.5 \text{mg/L, range 0.8–3.0} \)] in accordance with the manufacturer’s instructions, 
S. aureus (MIC\( _{90} \) 24 mg/L, range 0.8–128), 
compared with only 12 isolates shown to be resistant on disc 
testing. Linezolid was active against all strains (MIC\( _{90} \) 1.0 mg/L, 
range 0.2–3.0). Three (6%) of 49 isolates (2 patients) of 
Enterococcus spp. were resistant to teicoplanin on both disc 
testing and Etest (MIC 96–256 mg/L) but were susceptible to 
linezolid (MIC 0.8–2.0 mg/L).

Treatment and outcome

There were 1601 daily defined doses (DDD) of antibiotics with 
Gram-positive activity (median course 4 days, 0–16 days), 
including 147 courses of teicoplanin (median 7 days, 
0–24 days), 63 courses of linezolid (median 8 days, 1–24 days) 
and 29 courses of vancomycin (median 5 days, 0–30 days). 
Staphylococci (all coagulase negative) resistant to teicoplanin 
(MIC \( \geq 32 \text{ mg/L by Etest} \) were nevertheless treated with 
teicoplanin in three cases. One of these patients failed treatment, 
one improved and one was cured (as defined in an earlier paper). \( ^{4} \) 
Of a total of 27 patients infected with staphylococci with inter-
mediate susceptibility to teicoplanin (MIC \( 16 \text{ mg/L by Etest} \) were 
nevertheless treated with teicoplanin in three cases. One of these patients failed treatment, 
one improved and one was cured (as defined in an earlier paper). \( ^{4} \)

Table 1. Clinical outcome of patients according to susceptibility of the infecting pathogen to teicoplanin

<table>
<thead>
<tr>
<th>Isolate:</th>
<th>Coagulase-negative staphylococci 177 isolates (75 patients)</th>
<th>MRSA 643 isolates (143 patients)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Etest MIC:</td>
<td>Susceptible ≤8 mg/L</td>
<td>Intermediate 16 mg/L</td>
</tr>
<tr>
<td>Total isolates</td>
<td>100</td>
<td>61</td>
</tr>
<tr>
<td>Total patients</td>
<td>35</td>
<td>23</td>
</tr>
<tr>
<td>Median length of ICU stay, days (quartiles)</td>
<td>14 (9, 24)</td>
<td>22 (12, 33)</td>
</tr>
<tr>
<td>Length of hospital stay, days (quartiles)</td>
<td>40 (28, 74)</td>
<td>40 (20, 71)</td>
</tr>
<tr>
<td>Patients died*</td>
<td>12</td>
<td>11</td>
</tr>
</tbody>
</table>

*Where more than one isolate per patient, the highest MIC was used.
In the ICU population, MSSA was more common in screening specimens than MRSA (Table 2). However, MRSA was significantly more frequent than MSSA in blood, catheter tip, respiratory and wound infections. The susceptibility of S. aureus strains to linezolid or teicoplanin was not significantly affected by the site of isolation. However the range of susceptibilities of MRSA to teicoplanin was greater than that of MSSA.

Discussion

The present study demonstrates 3–6% resistance to teicoplanin in Gram-positive isolates in the ICU. Teicoplanin-intermediate susceptible S. aureus has been reported from this unit.1 No resistance to linezolid was found. However, the teicoplanin resistance did not translate into any clear clinical consequences either in the present study or in the accompanying double-blind prospective trial that showed teicoplanin to have similar efficacy to linezolid.4 The level of resistance to teicoplanin is modest given that it has been the main antibiotic used for treatment of Gram-positive infection in these ICUs since 1990.

As expected, there was a wider range of susceptibility to teicoplanin among staphylococci than to linezolid. Increasing resistance of coagulase-negative staphylococci, the mortality rate in the 20 patients with resistant strains was not significantly different from controls (25% versus 18%).5 Resistance was associated with previous use of glycopeptides but not the amount of use of glycopeptides on the ward.

The predominance of MRSA in blood isolates, but not on the skin, suggests that there are differences in the invasiveness of MRSA and MSSA. A higher rate of bacteraemia in carriers of MRSA (38%) than in carriers of MSSA (9.5%) has been observed in an MRSA outbreak in 147 ICU patients.9

The emergence of glycopeptide intermediate-resistant and now fully resistant S. aureus demonstrates the importance of keeping an effective reserve antibiotic for the treatment of Gram-positive infection in critically ill.10 Some strains of staphylococci with reduced susceptibility to teicoplanin remain susceptible to vancomycin. However, linezolid is currently active against most staphylococci, irrespective of their susceptibility to glycopeptides. Therefore it can be recommended in units where teicoplanin resistance is problematic. Despite 15 years of use, teicoplanin resistance in our ICUs remains uncommon and resistance appeared to have few clinical consequences. The use of linezolid therefore should be restricted to ensure that its activity is preserved.

Acknowledgements

An unrestricted educational grant was provided by Pfizer Ltd, Tadworth, Surrey, UK. The study was initiated and conducted independently by the named authors.

Transparency declarations

There were no conflicts of interest for any of the authors in the study.

References

Susceptibility of Gram-positive pathogens in ICU