Low-level exposure of MRSA to octenidine dihydrochloride does not select for resistance

Z. Al-Doori1, P. Goroncy-Bermes2, C. G. Gemmell1,3 and D. Morrison1

1Scottish MRSA Reference Laboratory, Stobhill Hospital, Glasgow, UK; 2Schuelke and Mayr GmbH, Germany; 3Glasgow University Division of Immunology, Infection and Inflammation, Glasgow, Scotland, UK

Keywords: antiseptics, mutagenesis, biocides

Sir,

Methicillin-resistant Staphylococcus aureus (MRSA) is prevalent in hospitals and contributes significantly to morbidity and mortality in infected patients. Antiseptics are an important element in the prevention and treatment of MRSA infections.

Octenidine dihydrochloride {N, N’-(1,10 decanediyldi-1[4H]-pyridinyl-4-ylidene) bis-(1-octanamine)} is a new cationic antiseptic that belongs to the bispyridine class of chemicals. It has activity against Gram-positive and Gram-negative bacteria.1 It was effective in oral hygiene preventing plaque and gingivitis, as a whole body wash for MRSA decolonization2 and for skin disinfection of pre-mature newborn infants.3 The ability of bacteria, especially MRSA, to develop resistance to antimicrobials, in particular antibiotics, is well documented and, although not as common, resistance or ‘reduced susceptibility’ to biocides has also been reported.4,5 It is important that products should be tested for bacterial resistance arising from prolonged exposure. In this study, we determined the MIC of octenidine dihydrochloride for 100 S. aureus isolated in Scottish hospitals using an agar dilution method based on CLSI (formerly NCCLS) M7-A4.6 The 76 MRSA isolates included 31 different PFGE types, several clonal variants of EMRSA-15 and EMRSA-16 and representatives of community-associated MRSA clones. The 24 methicillin-susceptible S. aureus (MSSA) tested included 13 different PFGE types. The MIC of octenidine dihydrochloride for all isolates (MRSA and MSSA) was in the range of 2–4 mg/L with MIC50 and MIC90 = 4 mg/L. The MIC90 for MSSA (2 mg/L) was lower than that for MRSA (4 mg/L). To investigate whether prolonged exposure to low levels of octenidine dihydrochloride selects for resistance, representatives of five major international MRSA clones (clonal complex 5, CC8, CC22, CC30 and CC45) were tested using the method of the Gradel et al.7 method. A 100 µL inoculum of a 2 McFarland standard (≈4 × 108 cfu/mL) suspension of the ‘parent’ isolate was initially grown in 1 mg/L (sub-MIC concentration) octenidine dihydrochloride in brain heart infusion (BHI) broth and incubated at 35°C for up to 3 days; 100 µL was then transferred into fresh BHI broth containing 2 mg/L octenidine dihydrochloride. Concentrations were subsequently increased in 1 mg/L increments and broth cultures incubated at 35°C for 2–7 days.

This continued for a period of up to 3 months. Broths showing visual turbidity were checked for purity, and to confirm identity the ‘adapted’ and parent isolates were compared by PCR-based typing (16S–23S rRNA spacer length polymorphism). Small differences were observed in the highest concentration of octenidine dihydrochloride at which different clones were able to grow: CC5 (8 mg/L), CC8 (4 mg/L), CC22 (6 mg/L), CC30 (7 mg/L) and CC45 (8 mg/L). Adapted isolates were stored at −20°C in ‘non-selective’ medium (no octenidine dihydrochloride added) prior to MIC testing. Although growth occurred at concentrations higher than the MIC for the parent isolates, the MICs for the adapted and parent isolates were identical (4 mg/L). Adapted isolates were tested twice. The MIC data for the adapted isolates indicate that the reduced susceptibility observed during prolonged exposure to octenidine dihydrochloride in broth cultures was unstable. These data indicate that, under these experimental conditions, the five epidemic MRSA clones tested failed to acquire stable resistance following continuous exposure to low-level concentrations of octenidine dihydrochloride.

Acknowledgements

This work was supported by Schuelke and Mayr GmbH, Germany.

Transparency declarations

Z. A.-D. has nothing to declare. P. G.-B. is an employee of Schuelke and Mayr. C. G. G. has received financial payments from Schuelke–Mayr while acting as a consultant to the Company under the aegis of In vivo Simulations Ltd (C. G. G. is a director) and in relation to a presentation made at a symposium held at the meeting of 16th ECCMID in Nice, France in April 2006. D. M. has received financial payments from Schuelke–Mayr while acting as a consultant to the Company and in relation to a presentation made at a symposium held at the meeting of 16th ECCMID in Nice, France in April 2006.

References

Effect of linezolid and teicoplanin on skin staphylococci

Samantha Hayman¹, A. Peter R. Wilson¹*, Mervyn Singer² and Geoffrey Bellingan²

¹Department of Clinical Microbiology, University College London Hospitals, 46 Cleveland Street, London WIT 4JF, UK; ²Bloomsbury Institute of Intensive Care Medicine, Department of Medicine and Wolfson Institute of Biomedical Research, University College London, Gower Street, London WC1E 6BT, UK

Keywords: glycopeptides, oxazolidinones, MRSA

*Corresponding author. Tel: +44-207-380-9516; Fax: +44-207-636-6482; E-mail: peter.wilson@uclh.nhs.uk

Sir,

Linezolid is an oxazolidinone antibiotic with excellent activity against methicillin-resistant Staphylococcus aureus (MRSA) including those with reduced susceptibility to teicoplanin or vancomycin. A randomized comparative study of the use of linezolid and teicoplanin in critically ill patients showed no difference in efficacy.² A significant difference was, however, noted in the clearance of MRSA from the skin.¹ At the start of treatment, 45 linezolid- and 43 teicoplanin-treated patients were colonized with MRSA. By the end of treatment, 23 (51%) patients receiving linezolid and 8 (19%) patients receiving teicoplanin had cleared MRSA carriage (P = 0.002). Although re-colonization occurred in a few cases, the use of linezolid reduced the risk of transmission of MRSA within the intensive care unit for 1–2 weeks after completion of treatment. To determine the effect of linezolid and teicoplanin on the detection of staphylococci in skin flora, a study was conducted using contact plates applied to the patient’s skin during treatment.

The study was approved by the University College London Hospitals Ethics Committee (00/0029). Mannitol salt agar (without oxacillin) contact plates (6 cm diameter, ~28.25 cm²) were used to monitor MRSA, methicillin-susceptible S. aureus (MSSA) and coagulase-negative staphylococci on the skin of patients receiving either teicoplanin or linezolid. The antibiotics were given as part of the main trial published previously¹ in a blinded fashion: teicoplanin 6 mg/kg daily intravenous (iv) or linezolid 600 mg twice daily iv. Sampling was performed at the same time each day before administering the antibiotic on days 0, 1, 2, 3, 5 and 7. After first reaching room temperature, agar was pressed to the inner, fleshy part of the patient’s knees (separate plates). Plates were incubated at 37°C for 48 h, at which time the numbers of each type of staphylococcus were categorized as none, light (1–50 cfu), medium (51–500 cfu) and heavy (>500 cfu). Five colonies of each type were tested by standard methods to distinguish S. aureus and coagulase-negative staphylococci.

Growth of MRSA was in the same category (none, light, medium or heavy) for at least 4 of the sampling days in 44 (85%) of 52 samples in 26 patients treated with teicoplanin and 37 (88%) of 42 samples in 21 patients treated with linezolid. Of the 24 MRSA carriers, none of the 9 patients in the linezolid group was positive for MRSA from day 5, compared with 9 of 15 carriers in the teicoplanin group (Fisher’s exact test P = 0.007). A reduction in category for at least 2 consecutive days was recorded in six (12%) samples in four patients given teicoplanin and in eight (19%) samples in five patients given linezolid. A rise in category was recorded in two patients treated with teicoplanin. MSSA was only isolated from the skin of two patients.

Coagulase-negative staphylococci were isolated from all patients. The average category in the first and last 3 days of sampling fell in 27 (52%) of 52 samples in the teicoplanin group versus 31 (76%) of 41 in the linezolid group (χ² test P < 0.05). Sample growth was in the same category for at least 4 sampling days in 24 (46%) cases in the teicoplanin group and 20 cases (49%) in the linezolid group. Sampling the skin daily for 5 days in two cases not given antibiotics showed no significant trend over time in the numbers of coagulase-negative staphylococci retrieved.

After 3 days of treatment, linezolid was more effective than teicoplanin in eradicating MRSA from the skin and in reducing the load of coagulase-negative staphylococci. Unlike teicoplanin, linezolid is highly concentrated in the skin and soft tissues, sufficient to exceed the MIC for most Gram-positive bacteria.ª,² Therefore, not only is linezolid capable of clearing MRSA carriage from some patients as demonstrated in the main trial,¹ but also it could reduce the rate of transmission of MRSA and potentially reduce rates of wound and line infection by coagulase-negative staphylococci. Others have found linezolid effective in eradicating carriage in critical care patients, but used it in combination with topical agents.ª Linezolid is expensive and MRSA can develop resistance,³ but this property might usefully be further investigated when treating infections in wards with a high level of endemic MRSA or potentially to decontaminate high-risk patients when topical treatment has failed, e.g. prior to major surgery.

Acknowledgements

This work was supported by an unrestricted educational grant from Pfizer UK.

Transparency declarations

M. S. and A. P. R. W. were members of a linezolid advisory panel for Pfizer.

References