In vitro anti-methicillin-resistant Staphylococcus aureus activity of 2,4-diacetylphloroglucinol produced by Pseudomonas sp. AMSN isolated from a marine alga

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Sir, Methicillin-resistant Staphylococcus aureus (MRSA) causes serious public health problems throughout the world. Moreover, the susceptibility of MRSA to vancomycin, the ‘last resort’ antibiotic, has recently been shown to be reduced, necessitating the development of alternative post-vancomycin antibiotics and other chemotherapeutic agents.

In a previous study to identify new types of antibacterial compounds active against MRSA, we screened various species of marine algae and identified several algal species that showed anti-MRSA activity. One of these, a red alga (Ceratodyction spongiosum) showed strong activity against all MRSA strains tested, and eventually the Pseudomonas sp. strain AMSN, isolated from the surface of this red alga, proved to be producing an anti-MRSA substance. We therefore attempted to isolate and purify this anti-MRSA compound from a culture of Pseudomonas sp. AMSN, isolated from the surface of this red alga, and determined that it was 2,4-diacetylphloroglucinol (DAPG) by 

Although DAPG is known to be produced by Pseudomonas and to be an antifungal substance, there has been no report on its anti-MRSA activity. We evaluated the antibacterial activity of DAPG against 10 clinical isolates of MRSA (MICs against MRSA > 100 mg/L) in comparison with 16 commercial antibiotics, and its antibacterial action was examined on the basis of bactericidal and bacteriolytic activity.

In the present study, the anti-MRSA activity of DAPG was demonstrated for the first time, showing high activity at approximated MICs ranging from 0.25 to 1 mg/L. A comparison of DAPG and commercial antibiotics demonstrated that the anti-MRSA activity of DAPG was higher than that of all commercial antibiotics tested, with the exceptions of vancomycin and clindamycin, which were more active against eight and four MRSA strains, respectively. The approximated MICs of the commercial antibiotics josamycin, tobramycin, gentamicin, cefotaxime, chloramphenicol, doxycycline, clindamycin, penicillin-G, ampicillin, amoxycllin, cefazolin, erythromycin, kanamycin, streptomycin and vancomycin against the MRSA strains tested were 1–32, 1–32, 2–32, 2–8, 2–32, 0.5–32, 0.25–32, 0.78–100, 0.78–100, 0.78–100, 0.78–100, >100, >100, 3.13–100 and 0.16–0.78 mg/L, respectively. Furthermore, with the exception of vancomycin, all of these commercial antibiotics were ineffective against at least one of the MRSA strains tested.

Figure. Bactericidal activities of 2,4-diacetylphloroglucinol (a) and cefotaxime (b) against MRSA (strain E 31224) at various concentrations. Symbols: ●, control; ○, 1 × MIC; ▲, 2 × MIC; ×, 4 × MIC; ○, 8 × MIC.
strains. All MRSA strains used in this study also demonstrated multidrug-resistant properties. In contrast, the MRSA strains tested were all susceptible to DAPG. When the anti-MRSA activity of phloroglucinol was evaluated, this compound showed no effect on MRSA strains, indicating that the two acetyl residues of DAPG might contribute to the antibacterial activity.

The bactericidal activity of DAPG against MRSA (strain E 31224) was greater than that of cefotaxime, since DAPG killed MRSA at 4 mg/L, whereas cefotaxime required a higher concentration of 32 mg/L. DAPG failed to inhibit MRSA growth at the MIC of 1 mg/L, but reduced the MRSA cell counts slightly at a concentration of 2 × MIC after 6 h incubation. At 4 × and 8 × MIC of DAPG, cell numbers were decreased significantly after 2 h incubation and drastically after 6 h incubation.

Cefotaxime slightly reduced the MRSA cell count at the MIC of 4 mg/L and 2 × MIC after 2 h incubation, but it increased again after 12 h incubation at the MIC. Higher concentrations of cefotaxime at 4 × and 8 × MIC significantly reduced the MRSA cell count after 2 h incubation, but the MRSA cell number remained relatively constant during the subsequent 18 h incubation period at 4 × MIC.

Not only is MRSA spreading to many hospitals throughout the world, but vancomycin- and teicoplanin-resistant S. aureus and Enterococcus spp. strains have been found in the USA, Canada, Japan and European countries. With respect to post-vancomycin treatment, DAPG or its derivatives could be possible candidates in the development of new types of anti-MRSA compounds.

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References