(9 MRSA, 1 MSSA) from patients with bacteremia in one center in the USA (2004–2006), pre- and postdaptomycin therapy MICs could be compared. Most patients had failed vancomycin therapy prior to starting daptomycin. The preexposure MIC was known in seven cases (0.125–0.5 mg/l). Postexposure MIC was elevated in 6 of 10 cases (MICs 2–4 mg/l). An MIC increase was noted after 5–10 days of exposure. Pulsed-field gel electrophoresis band pattern of isolates with increased MIC revealed one to three band differences, implying genetic relatedness. All patients with nonsusceptible isolates relapsed or failed therapy (Sharma *et al*., 2008).

An elderly patient with MRSA bacteremia was treated sequentially with vancomycin plus rifampicin, then daptomycin plus gentamicin. The MRSA strain developed diminished susceptibility to vancomycin (MIC increase and tolerance), daptomycin, and gentamicin and resistance to rifampicin during therapy (Bennett *et al*., 2008). Huang *et al*. (2008) described the development of nonsusceptibility to daptomycin and vancomycin during treatment for MRSA bacteremia associated with infective endocarditis and probable septic thrombophlebitis in a uremic patient from Taiwan. VISA MRSA bacteremia persisted during glycopeptide treatment and subsequent daptomycin treatment but cleared after 5 days' treatment with linezolid and fusidic acid.

A patient with native valve endocarditis caused by a vancomycin "heteroresistant" strain of *E. faecium* experienced failure of daptomycin monotherapy without evidence of daptomycin resistance. The infecting organism exhibited *in vivo* emergence of a vancomycin-susceptible subpopulation lacking the *vanA* gene cluster. Treatment with a combination of high-dose daptomycin, gentamicin, and high-dose ampicillin cleared the infection (Arias *et al*., 2007). Failures in clinical treatment of *S. aureus* infection with daptomycin appear to be associated with alterations in surface charge, membrane phospholipid asymmetry, and drug binding (Jones *et al*., 2008).

7c. Osteoarticular infections

EXPERIMENTAL STUDIES USING BIOFILM MODELS

Experimental studies using a biofilm model have been performed for *S. epidermidis* and *S. aureus*. One study showed the effect of daptomycin alone or combined with azithromycin on *S. epidermidis* biofilm in the static phase by detection of changes in the optical density of the treated vs. the untreated biofilm. Daptomycin alone at concentrations of 2 mg/l and 5 mg/l had no effect on the biofilm, and the addition of azithromycin also had no effect (Presterl *et al*., 2009). In the other study, Olson *et al.* (2010) studied *S. epidermidis* biofilm using flow cell and guinea pig tissue cage models and found that by using both models, the viable cell count after treatment with daptomycin–rifampicin was significantly lower ($p < 0.05$) than after treatment with vancomycin, vancomycin–rifampicin, daptomycin, or rifampicin alone. They also found that daptomycin alone had no effect on the *S. epidermidis* biofilm. This lack of effect occurs despite the good penetration of daptomycin in *S. epidermidis* biofilms (Stewart *et al*., 2009).

In contrast to the results for *S. epidermidis* biofilm models, which show no activity of daptomycin monotherapy, studies for *S. aureus* biofilm models do show activity for daptomycin monotherapy. In a study comparing the activity of various antibiotics in a MRSA biofilm model, daptomycin and delafloxacin were the most potent, reducing viability by more than 50% at clinically achievable concentrations against both strains as well as reducing biofilm depth, as observed in confocal microscopy. Rifampin, tigecycline, and moxifloxacin were effective against mature MRSA biofilms, whereas oxacillin demonstrated activity against MSSA. Fusidic acid, vancomycin, and linezolid were less potent overall. Antibiotic activity depends on biofilm maturity and bacterial strain (Bauer *et al*., 2013).

However, studies with combinations of antibiotics indicate that the activity of daptomycin on *S. aureus* biofilms can be increased by combination therapy. The combination of daptomycin with clarithromycin significantly increased the activity against both biofilm-embedded MRSA and planktonic MRSA ($p < 0.01$) (Parra-Ruiz *et al.*, 2010). The combination of linezolid plus daptomycin has been shown to be more effective than both drugs alone in a biofilm MRSA reactor model (Parra-Ruiz *et al*., 2012a). Time-kill studies indicate that daptomycin plus rifampicin was superior over vancomycin plus rifampicin for *E. faecium* (Holmberg *et al*., 2014).

EXPERIMENTAL FOREIGN-BODY MODELS: COMBINATION THERAPY

A MRSA foreign-body model was used to evaluate the efficacy of daptomycin at usual and high doses (equivalent to 6 and 10 mg/kg/day in humans, respectively) in combination with cloxacillin in a rat tissue cage MRSA infection model and to compare its efficacy to that of daptomycin–rifampin. Daptomycin–rifampin was the best therapy ($p < 0.05$). The low dosage of daptomycin was the least effective treatment and did not protect against the emergence of resistant strains. There were no differences between the two dosages of daptomycin plus cloxacillin in any situation, and both protected against resistance. The overall effect of the addition of cloxacillin to daptomycin was a significantly greater cure rate (against adhered bacteria) than that for daptomycin alone. In conclusion, daptomycin–cloxacillin enhanced modestly the *in vivo* efficacy of daptomycin alone against foreign-body infection by MRSA and was less effective than daptomycin plus rifampin (Garrigós *et al.*, 2012).

A beneficial effect of rifampicin was also found in two other studies. In a foreign-body model in guinea pigs infected with MRSA, it has been shown that daptomycin alone (doses equal to 6 mg/kg/day in humans) was not able to cure any of the cage-associated infections. When combined with rifampicin, however, the cure rate was 67% and resulted in a reduction of > 6 $\log_{10}$ CFU/ml. This daptomycin dose also prevented the emergence of resistance (John *et al*., 2009). In rats infected with MRSA, this combination was also studied. Daptomycin doses that were equal to human doses of 6 mg/kg and 10 mg/kg were used. Daptomycin monotherapy