of giant unilamellar vesicles (GUVs). These constitute different lipid compositions. Chen *et al.* provided evidence that the interaction of daptomycin with the cell membrane results in a marked alteration of phospholipid content (so-called lipid extracting effect). In the presence of low concentrations of daptomycin, an initial expansion of the GUV occurs owing to binding of the daptomycin to the surface. At higher daptomycin concentrations, this initial expansion is followed by a decrease in outer surface area of the GUVs (i.e. lipids are removed from the lipid bilayer). These processes were observed when phosphatidyl glycerol (PG) was present in the GUV and calcium was added to the solution (Chen *et al.*, 2014).

The specific mechanisms leading to bacterial cell death remain, however, unclear. The changes in cell membrane homeostasis lead to leakage of ions and loss of cell membrane potential (Pogliano *et al.*, 2012). A correlation between dissipation of membrane potential and the bactericidal activity of daptomycin was demonstrated by Silverman *et al.* (2003). Membrane depolarization was measured by both fluorimetric and flow cytometric assays. Adding daptomycin (5 mg/l) to *S. aureus* gradually dissipated membrane potential. In both assays, cell viability was reduced by > 99% and membrane potential was reduced by > 90% within 30 minutes of adding daptomycin. Cell viability decreased in parallel with changes in membrane potential, demonstrating a temporal correlation between bactericidal activity and membrane depolarization. Decreases in viability and potential also showed a dose-dependent correlation. Depolarization is indicative of ion movement across the cytoplasmic membrane. Fluorescent probes were used to demonstrate Ca^{2+}-dependent, daptomycin-triggered potassium release from *S. aureus*. Potassium release was also correlated with bactericidal activity (Silverman *et al.*, 2003).

IMMUNE MODULATION

The mode of action of daptomycin endows it with a broad spectrum of bactericidal activity against Gram-positive bacteria without being bacteriolytic. The ability of daptomycin to produce bactericidal activity against *S. aureus* while causing negligible cell lysis has been demonstrated using electron microscopy and the membrane integrity probes calcein and ToPro3. The formation of aberrant septa on the cell wall, suggestive of impairment of the cell division machinery, was also observed (Cotroneo *et al.*, 2008), a feature that contributes to reducing overstimulation of the immune response by bacteria and prolongation of inflammation. A reduced macrophage inflammatory response to *S. aureus* isolates was noted *in vitro* in the presence of daptomycin, relative to vancomycin or oxacillin. Exposure of any of six clinical isolates of *S. aureus* to daptomycin alone or in combination with vancomycin or oxacillin (compared with vancomycin or oxacillin alone) led to a dampened macrophage inflammatory response with diminished tumor necrosis factor secretion and reduced accumulation of inducible nitric oxide synthase protein (English *et al.*, 2006). It is postulated that fewer proinflammatory bacterial fragments were released by the bactericidal activity of daptomycin than by the other agents. A study that supports this postulate compared therapy with daptomycin with ceftriaxone in experimental pneumococcal meningitis. The treatments were evaluated for their effects on inflammation and brain injury. Daptomycin cleared the bacteria more efficiently from the CSF than ceftriaxone within 2 hours of the initiation of therapy, reduced the inflammatory host reaction, and prevented the development of cortical injury (Grandgirard *et al.*, 2007). Daptomycin may have minimal effects on cytokine production and may have synergistic immunomodulatory effects in combination with other immunomodulators (Kelesidis, 2014). Although clinical evidence is limited, daptomycin has immunomodulatory properties, resulting in the suppression of cytokine expression after host immune response stimulation by MRSA. Experimental studies showed an improved efficacy of daptomycin in combination with administration of vitamin E (an immune enhancer) before infecting wounds by MRSA (Tirilomis, 2014). Further clinical studies are needed to confirm these findings and determine the effect of a reduced inflammatory response.

4. MODE OF DRUG ADMINISTRATION AND DOSAGE

Daptomycin is available for i.v. use only as a concentrated lyophilized powder for dissolution for infusion. Freshly reconstituted solutions of daptomycin range in color from pale yellow to light brown.

In addition to i.v. administration, there has been some off-label use of daptomycin given intraventricularly and intraperitoneally (see section 5b, Drug distribution).

STABILITY

Chemical and physical in-use stability of the reconstituted solution in the vial has been demonstrated for 12 hours at 25°C and up to 48 hours at 2– 8°C. Chemical and physical stability of the diluted solution in infusion bags is established as 12 hours at 25°C or 24 hours at 2–8°C (Summary of Product Characteristics, 2016). The stability of an admixture containing reconstituted daptomycin and heparin in lactated Ringer's injection was also evaluated. The admixture of daptomycin (5 mg/ml) and heparin sodium (100 USP units/ml diluted in lactated Ringer's injection) was stable when stored in polypropylene syringes for up to 14 days at 4°C and –20°C (Ortega *et al.*, 2014).

Daptomycin has been reported to degrade in 5% glucose solutions at a rate of 15–20% per 24 hours at room temperature (Parra *et al.*, 2013). This might be important for the use of daptomycin in peritoneal dialysis solutions. Over 24 hours, daptomycin concentrations declined similarly in Physioneal 2.27% glucose solution held at 25°C (92%) and 37°C (98%); in Glucosada Grifols 5% and Viaflo Glucosa 10% solutions, they declined 60% below the level chosen to denote stability. The observed difference in daptomycin recovery can be explained only by the different glucose concentrations of the evaluated solutions (Parra *et al.*, 2013). In both cases stability of daptomycin declines by more than 10% after 6 hours at