17.3.2 Cell Wall/Membrane Proteins

Many antibiotics (β-lactam antibiotics, glycopeptides, etc.) used today interfere with cell wall synthesis and concentrations of intracellular antibiotics in bacteria depend on cell wall permeability and transport. Also, several proteins in the cell wall are important to bacterial antibiotic resistance, e.g. penicillin-binding proteins (Nikolaidis et al. 2014). Different approaches that interact with cell wall integrity such as destabilization of the bacterial membrane, increase of cell wall permeability, and modification of cell wall proteins could be promising ways to sensitize bacteria to antibiotics.

Some novel strategies have been proposed that can disturb the cell wall structure and improve the sensitivity to antibiotics. For example, Taylor et al., in a screen of 30 000 small molecules, found an inhibitor of MreB, a regulator of actin filament homologues in bacteria (Taylor et al. 2012). This inhibition made *E. coli* sensitive to novobiocin, an antibiotic that usually has poor effect on Gram-negative bacteria. Ernst et al., examined MrpF, a membrane protein in *S. aureus* that induced resistance to daptomycin through modification of phosphatidylglycerol (PG) (Ernst et al. 2015). They discovered that MrpF harbors a flippase activity that translocates the modified PG from the cytoplasm to the outer membrane, changing the electrical charge of the membrane to become more positive on the outside. The increased surface charge leads to repulsion of daptomycin, causing resistance. Although the data are preliminary, the authors see an opportunity to target MrpF and other flippases as a new way of sensitizing bacteria to antibiotics in the future.

Chiosis and Boneca have described a way to overcome vancomycin resistance in enterococci due to structural changes in the cell wall peptidoglycan (Chiosis and Boneca 2001). Peptidoglycan precursors in the membrane of resistant strains contain D-Ala-D-lactate instead of D-Ala-D-Ala, which lowers the affinity of vancomycin by a factor of over a thousand. To combat this, Chiosis and Boneca found small molecules with cleaving activity specifically of the D-Ala-D-lactate moiety, which could increase the proportion of D-Ala-D-Ala in the cell wall, thus re-sensitizing the bacteria to vancomycin. Another study has shown that vancomycin-resistant enterococci (VRE) could be re-sensitized to vancomycin by treatment with flavonoids (Liu et al. 2001). The mechanism by which this sensitization works might be through the ability of flavonoids to inhibit D-Ala-D-Lac peptide production during cell wall formation.

Another interesting study that sensitized bacteria to antibiotics that target the cell wall involved the ClpXP protease in bacteria (McGillivray et al. 2012). This protease performs controlled proteolysis that regulate protein turnover in many bacterial species. Inhibition of this protease with a compound they called F2, showed an increased susceptibility to the antimicrobial peptide (AMP) cathelicidin and to antibiotics in both *S. aureus* and *Bacillus anthracis*, but did