Microbiology antimicrobial

List and explain 4 mechanism of antimicrobial resistance.

**1 Limiting drug uptake**

As already mentioned, there is a natural difference in the ability of bacteria to limit the uptake of antimicrobial agents. The structure and functions of the LPS layer in gram negative bacteria provides a barrier to certain types of molecules. This gives those bacteria innate resistance to certain groups of large antimicrobial agents . The mycobacteria have an outer membrane that has a high lipid content, and so hydrophobic drugs such as rifampicin and the fluoroquinolones have an easier access to the cell, but hydrophilic drugs have limited access .

Bacteria that lack a cell wall, such as *Mycoplasma* and related species, are therefore intrinsically resistant to all drugs that target the cell wall including β-lactams and glycopeptides . Gram positive bacteria do not possess an outer membrane, and restricting drug access is not as prevalent. In the enterococci, the fact that polar molecules have difficulty penetrating the cell wall gives intrinsic resistance to aminoglycosides. Another gram positive bacteria, *Staphylococcus aureus*, recently has developed resistance to vancomycin. Of the two mechanisms that *S. aureus* uses against vancomycin, a yet unexplained mechanism allows the bacteria to produce a thickened cell wall which makes it difficult for the drug to enter the cell, and provides an intermediate resistance to vancomycin. These strains are designated as VISA strains.

### 2 Modification of drug targets

There are multiple components in the bacterial cell that may be targets of antimicrobial agents; and there are just as many targets that may be modified by the bacteria to enable resistance to those drugs. One mechanism of resistance to the β-lactam drugs used almost exclusively by gram positive bacteria is via alterations in the structure and/or number of PBPs (penicillin-binding proteins). PBPs are transpeptidases involved in the construction of peptidoglycan in the cell wall. A change in the number (increase in PBPs that have a decrease in drug binding ability, or decrease in PBPs with normal drug binding) of PBPs impacts the amount of drug that can bind to that target. A change in structure (e.g. PBP2a in S. aureus by acquisition of the mecA gene) may decrease the ability of the drug to bind, or totally inhibit drug binding .

The glycopeptides (e.g. vancomycin) also work by inhibiting cell wall synthesis, and lipopeptides (e.g. daptomycin) work by depolarizing the cell membrane. Gram negative bacteria (thick LPS layer) have intrinsic resistance to these drugs . Resistance to vancomycin has become a major issue in the enterococci (VRE—vancomycin-resistant enterococci) and in Staphylococcus aureus (MRSA). Resistance is mediated through acquisition of van genes which results in changes in the structure of peptidoglycan precursors that cause a decrease in the binding ability of vancomycin . Daptomycin requires the presence of calcium for binding. Mutations in genes (e.g. mprF) change the charge of the cell membrane surface to positive, inhibiting the binding of calcium, and therefore, daptomycin .

### 3 Drug inactivation

There are two main ways in which bacteria inactivate drugs; by actual degradation of the drug, or by transfer of a chemical group to the drug. The β-lactamases are a very large group of drug hydrolyzing enzymes. Another drug that can be inactivated by hydrolyzation is tetracycline, via the tetX gene .

Drug inactivation by transfer of a chemical group to the drug most commonly uses transfer of acetyl, phosphoryl, and adenyl groups. There are a large number of transferases that have been identified. Acetylation is the most diversely used mechanism, and is known to be used against the aminoglycosides, chloramphenicol, the streptogramins, and the fluoroquinolones. Phosphorylation and adenylation are known to be used primarily against the aminoglycosides.

### β-lactamases

The most widely used group of antimicrobial agents are the β-lactam drugs. The members of this drug group all share a specific core structure which consists of a four-sided β-lactam ring. Resistance to the β-lactam drugs occurs through three general mechanisms:

 (1) preventing the interaction between the target PBP and the drug, usually by modifying the ability of the drug to bind to the PBP (this is mediated by alterations to existing PBPs or acquisition of other PBPs;

 (2) the presence of efflux pumps that can extrude β-lactam drugs;

 (3) hydrolysis of the drug by β-lactamase enzymes .

The β-lactamases (originally called penicillinases and cephalosporinases) inactivate β-lactam drugs by hydrolyzing a specific site in the β-lactam ring structure, causing the ring to open. The open-ring drugs are not able to bind to their target PBP proteins. The known β-lactamases are wide-spread, and the group contains enzymes that are able to inactivate any of the current β-lactam drugs. The production of β-lactamases is the most common resistance mechanism used by gram negative bacteria against β-lactam drugs, and the most important resistance mechanism against penicillin and cephalosporin drugs .

The β-lactamase enzymes are classified based on their molecular structure and/or functional characteristics. Structurally they are placed into four main categories (A, B, C, or D). There are three functional groupings based on the substrate specificity: the cephalosporinases, the serine β-lactamases, and the metallo (zinc-dependent) β-lactamases. These enzymes may also be commonly known by their enzyme family; for example: the TEM (named after the first patient) family, the SHV (sulphydryl variable) family, and the CTX (preferentially hydrolyze cefotaxime) family. Gram negative bacteria may produce β-lactamases from all four structural groups. The β-lactamases found in gram positive bacteria are mainly from group A, with some from group B .