

Antimicrobial Drugs.

The scientific development of synthetic antimicrobial drugs began with Erlich in the 1890's, with the use of methylene blue for managing malaria, the organic arsenicals for trypanosomiasis (1904), and salvarsan 606 for syphilis (1909). 'Atebrin' was made in 1932 and used for prophylaxis of malaria, the first useful sulphonamide drug came about in 1935.

Antibiotic drug use began in the 1920's when Fleming observed the anti-staphylococcal activity of *Penicillium notatum*, and from this penicillin was developed for clinical use by Florey & Chain. Another important group of antibiotics was the aminoglycosides, when streptomycin was isolated from *Streptomyces griseus*, in 1944. The cephalosporins were developed from *Cephalosporium acremonium* in 1945.

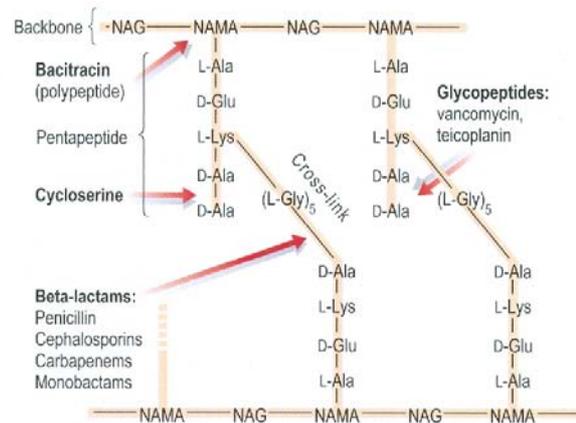
Classification.

Antimicrobials are classified by the pathogens targeted, e.g. as antibacterials or antifungals. This grouping may be subdivided as antibacterials also include urinary antiseptics and anti-mycobacterial drugs. Antimicrobials, especially antibacterials, are strictly classified into chemotherapeutic agents (synthetic chemicals), and antibiotics, produced from living organisms, usually fungi. However, 'antibiotic' is often used loosely to mean all antibacterials. Antibacterials can be further described by their:

- chemical structure (penicillins, cephalosporins)
- effect on bacterial growth (bacteriostatic or bactericidal)
- target site.

Target site classes

Cell wall synthesis inhibitors - The cell wall synthesis inhibitors are bactericidal because they block synthesis of different peptidoglycan components of the wall, so growing cells lyse and die.

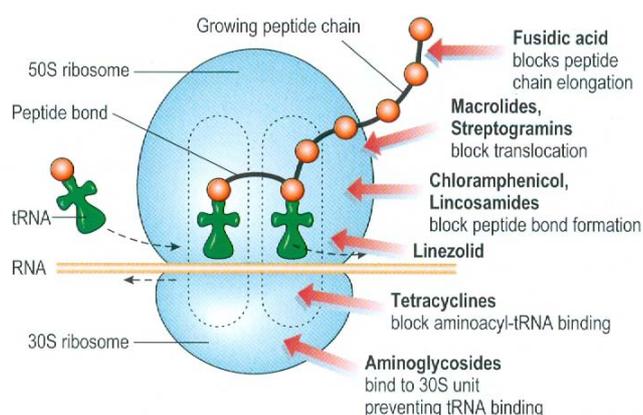


They do not affect eucaryotic cells, nor microbes that lack peptidoglycan.

These antibiotics are:

- beta-lactams: penicillins, cephalosporins, carbapenems, monobactams
- glycopeptides: vancomycin, teicoplanin
- polypeptide: bacitracin
- cycloserine

Protein synthesis inhibitors - Protein synthesis inhibitors act on varying stages of protein synthesis.



If these are unique to bacteria (e.g. affect the bacterial 70S ribosome rather than the eucaryotic 80S ribosome) they are selectively toxic. However, eucaryotic mitochondrial protein synthesis occurs on 70S ribosomes and can be affected. This group includes:

- aminoglycosides: streptomycin, gentamicin, tobramycin, netilmicin, amikacin, spectinomycin, neomycin
- tetracyclines: tetracycline, doxycycline, minocycline
- linezolid
- chloramphenicol
- lincosamides: clindamycin, lincomycin
- macrolides: erythromycin, roxithromycin, azithromycin, clarithromycin, spiramycin
- streptogramins
- fusidic acid.

Aminoglycosides cause ineffective proteins to form and so are bactericidal. All the others in this group have a reversible (bacteriostatic) action and so protein synthesis begins again when antibiotic levels decrease.

Nucleic acid synthesis inhibitors - Nucleic acids are made by all cells so the possibility of selective drugs toxic only for microbes is limited. Some pathways have distinct features that can be targeted, or some enzymes are sufficiently different for a selective effect to occur:

- Folic acid synthesis: a precursor of purines and pyrimidines is folic acid which microbes can only synthesise; humans obtain folic acid in food. Sulphonamides and trimethoprim interfere with folic acid synthesis.
- RNA polymerase: inhibited by rifamycins (rifampicin, rifabutin).
- DNA structure: disrupted by nitroimidazoles (e.g. metronidazole).
- topoisomerase: blocked by quinolones (norfloxacin, ciprofloxacin and others).

Cell membrane function inhibitors- Drugs that destroy the selective permeability of membranes will kill both microbial and human cells. As a result, they will be relatively toxic if given systemically. Colistin acts like a detergent, disrupting the cell membrane phospholipid. The polyene antifungal drugs (e.g. amphotericin B and nystatin) act by damaging sterols in eucaryotic membranes; they are particularly toxic to fungi through their action on ergosterol but also affect human cells.

Uncertain targets - The target of some anti-mycobacterial drugs is uncertain: isoniazid may act on mycolic acid synthesis, which would explain its specific activity, while ethambutol may inhibit RNA synthesis.

Characteristics.

Physicochemical properties

These are important in relation to the effectiveness and mode of administration of a drug, particularly whether they are stable to gastric acid and are absorbed from the gut and so can be given orally; if unstable or not absorbed, they need injection. Other important factors are whether the drug will cross barriers within the body - into cells, into the brain across the blood-brain barrier, into other protected tissues like prostate, or into cysts.

Spectrum and type of activity

The spectrum of activity is the range of organisms against which an antimicrobial is usually active. The minimal inhibitory concentration (MIC) is the smallest concentration of antimicrobial which is bacteriostatic, reversibly inhibiting bacterial growth, so re-growth occurs if the antimicrobial is removed by excretion or inactivation. By contrast, the minimal bactericidal (or fungicidal) concentration (MBe, MFC) is the smallest concentration irreversibly killing the microbes, so they do not re-grow if the antimicrobial is removed.

Mechanisms of resistance

Some bacteria are innately resistant to certain antibiotics because they lack a target site or are impermeable to the antibiotic; other bacteria acquire resistance, by one of three mechanisms:

1. Altered target site - These may result in lower affinity for the antibiotic, or additional target enzymes may emerge unaffected by the drug.
2. Altered uptake - Effective drug concentration in the bacterial cell can be decreased either by decreasing permeability or by actively pumping the drug out of the bacterial cell.
3. Antibiotic-inactivating enzymes - These occur particularly against penicillins, cephalosporins and aminoglycosides.

Resistance spreads between bacteria in three genetic ways:

1. Chromosomal mutation, usually random, causes an altered protein, e.g. a ribosomal protein (streptomycin resistance) or altered enzyme (sulphonamides). Selection by the antibiotic after each cell division will result in a resistant population.
2. Transmissible plasmids are small circular DNA units replicating independently of the chromosome, and transmissible between cells. They have four advantages over chromosomal mutation: transfer between bacteria is more rapid than cell division; resistance to numerous *individual* antibiotics can be carried at once; resistance to several *classes* of antibiotic can be carried at once; and one class of plasmid can enter numerous genera, e.g. TEM-I beta-lactamase in enteric Gram-negative rods, in *N. sonorhoeae* and in *H. influenzae*.
3. Transposons, called 'jumping genes', move from the security of the chromosome to the mobility of a plasmid, and from one plasmid to another.

Resistance also spreads between bacteria in three physical ways:

1. Conjugation (by direct contact)
2. Transduction (by phages)
3. Transformation (uptake of free DNA).

Pharmacokinetics

The pharmacokinetics of a drug describe its behaviour in the body: absorption, distribution, protein binding, serum and tissue concentrations, serum half-life, metabolism and excretion. Important factors that will alter the effective half-life of the drug (and its toxicity) include the age of the patient, concurrent diseases particularly of organs which metabolise or excrete the drug (usually liver and kidney), genetic factors (slow and fast drug metabolism) and interaction with other drugs.

Side effects, toxicity

Even safe effective antibiotics like penicillins fall short of the ideal of a 'magic bullet' which would not affect humans yet would eradicate germs by a single '*dosa sterilisa magna*' (great sterilising dose). Side effects and toxicity are often similar within a group of antibiotics, e.g. all aminoglycosides are ototoxic (ear) and nephrotoxic (kidney), but in varying degrees.

KEY LEARNING POINTS.



1. Antimicrobials are classified by the type of pathogen targeted, the disease for which they are effective, and the mechanism of action.
2. True antibiotics are the products of living organisms, although the term is used loosely to include synthetic and semi-synthetic chemicals.
3. Antibacterials can act on cell wall, protein and nucleic acid synthesis, on the cell membrane, or as anti-metabolites blocking metabolic pathways.
4. Antibiotic resistance may be innate because bacteria lack the target site or are impermeable to the antibiotic, or may be acquired. Bacteria acquire resistance by one of three mechanisms: an altered target site, altered uptake or by antibiotic-inactivating enzymes.
5. Resistance spreads between bacteria in three genetic ways: chromosomal mutation, transmissible plasmids or transposons.
6. Important characteristics of an antimicrobial include its physicochemical properties, mode of action, spectrum of activity and resistance, pharmacokinetic properties, dose and route, side effects and toxicity and clinical indications.

Specific anti-microbials.

Cell Wall Synthesis Inhibitors

Penicillins

The penicillins all have a similar structure, with different drugs being created using different side chains. The side chains of the natural product can be modified chemically to give a wider spectrum of activity. A common bacterial drug resistance is via betalactamases.

Pharmacokinetics - Some penicillins are stable to gastric acid and are absorbable (penicillin V, ampicillin/amoxicillin, flucloxacillin) and so can be given orally. Others must be given by injection: penicillin G, ticarcillin and piperacillin. All penetrate widely, except to CSF and brain tissue, and all have relatively short half-lives, are little metabolised and are excreted renally.

Toxicity - usually minimal, almost limited to hypersensitivity, chiefly rash or fever, and very rarely, anaphylaxis.

Cephalosporins

Structure of cephalosporins differs from the penicillins in having a six-member ring attached to the beta-lactam ring. The different side chains give the different cephalosporins, which are often classified into first, second, third and fourth 'generations' by their date of introduction; more logical classifications exist but are less used.

Pharmacokinetics - Cephalosporins behave like penicillins, although the later drugs have longer half-lives, especially ceftriaxone, and penetrate well to CSF and brain.

Spectrum- all are broad-spectrum, active against many Gram-positive, Gram-negative and anaerobic bacteria. Enterococci are resistant to all. In general first generation are best against Gram-positive bacteria, second

generation best against anaerobes, and third generation best against Gram-negative rods.

Toxicity - Low toxicity, similar to the penicillins.

Other β -lactams

Aztreonam is a mono-bactam, i.e. a single β -lactam ring. Its action and pharmacokinetics are like an injectable cephalosporin, but its spectrum is like the aminoglycoside gentamicin, i.e., solely Gram negative, including many pseudomonads. Its use is restricted by its high cost.

Imipenem, meropenem and ertapenem are carbapenems are structurally similar to penicillin. Their actions and pharmacokinetics are like an injectable cephalosporin, but their spectrum is very wide, although they are inactive against MRSA, *E. faecium*, and some Gram-negative rods. Their use is restricted by policy and cost to serious systemic infections before precise microbial diagnosis.

Vancomycin and Teicoplanin

Vancomycin and teicoplanin are glycopeptides and are bactericidal to Gram-positive bacteria.

Pharmacokinetics - Neither is absorbed from the gut, so vancomycin is given intravenously, by slow infusion to decrease the side effects of headache and flushing (the 'red man' syndrome). Teicoplanin is also given intramuscularly. Distribution of both is wide into most fluids and tissues, but suboptimal into CSF and brain. Their half-lives are long, hence 12 or 24-hourly dosing. Excretion is renal and, because of toxicity, serum levels are usually monitored, though safe levels have not yet been established.

Spectrum - their use is limited to severe Gram-positive (including MRSA) infections, and oral treatment of unresponsive *C. difficile*-associated colitis. Linezolid and streptogramins are active against most glycopeptide-resistant Gram-positive bacteria.

Toxicity - The main toxic effect is hearing loss, but phlebitis or neutropenia can also occur. Nephrotoxicity is uncommon with current preparations.

Protein synthesis inhibitors

All act on the 30S or 50S bacterial ribosome. Only aminoglycosides are bactericidal.

Aminoglycosides

The aminoglycosides contain streptamine or a streptidine-containing amino cyclitol, with side chains that are modified to produce the individual drugs. Gentamicin is actually a mixture of three related molecules.

Pharmacokinetics - Aminoglycosides are not absorbed from the gut, so must be injected for systemic use. They are not metabolised significantly and have a relatively long half-life, about 3 hours, so usually are given once- or twice-daily. Excretion is renal. Penetration is relatively poor into bone, lung and sputum, and non-existent into CSF and brain.

Spectrum - Bactericidal with a broad Gram-negative spectrum, however their uptake into cells is prevented by anaerobiosis so they are ineffective against anaerobes. They are mainly used against enteric Gram-negative rods; gentamicin, tobramycin and amikacin are also active against pseudomonads. Streptomycin was used for tuberculosis but is now used mainly in Gram-negative zoonoses including brucellosis, plague and tularaemia.

Toxicity - Aminoglycosides have both renal and ototoxicity, related mainly to total dose. This is reflected later in peak than in trough levels, which should be monitored carefully, especially in the old, the underweight and those with renal or auditory impairment.

Chloramphenicol

Chloramphenicol is a natural product that is now chemically synthesised. Action is on bacterial protein synthesis at the 50S ribosome, and is usually bacteriostatic only.

Pharmacokinetics - Chloramphenicol is lipophilic; it is orally absorbed and has wide penetration including into the interior of the eye, the CSF and brain. It is metabolised by the liver and excreted renally. It can also be given by injection.

Spectrum - Like its pharmacokinetics, the spectrum is extremely broad and includes most bacteria, chlamydiae, rickettsiae and mycoplasmata. It is also cheap.

Toxicity - The use of this antibiotic is limited by two toxicities. An unpredictable irreversible marrow aplasia causing aplastic anaemia occurs rarely (1 in 30000) and is fatal without marrow transplantation. If liver function is impaired, chloramphenicol levels rise above normal, and dose-related reversible marrow hypoplasia can occur; newborns who fail to metabolise chloramphenicol adequately die from toxic complications ('grey baby syndrome').

Lincosamides

Clindamycin and lincomycin are bacteriostatic. They are orally absorbed, but are also injectable, and widely distributed apart from CSF and brain. They are metabolised by the liver and excreted renally.

Spectrum - They are mainly used against anaerobes or staphylococci. Clindamycin is a reserve drug in toxoplasmosis.

Toxicity - Toxic effects include allergy, a metallic taste, and initiation of pseudomembranous enterocolitis associated with *C. difficile* overgrowth.

Macrolides

The macrolides have an unusual macrocyclic lactone ring with sugars attached. There are four macrolides in clinical use: erythromycin, roxithromycin, azithromycin and clarithromycin. Their action is bacteriostatic.

Pharmacokinetics - Erythromycin is inactivated by gastric acid so is protected by enteric-coating or given as a salt or ester. Intravenous use often causes thrombophlebitis. The half-life is about 2 hours, and excretion is hepatic; as a result, care is needed in liver failure but dosage is unaltered in renal impairment.

Spectrum - Macrolides are mainly used against Gram-positive and unusual bacteria: chlamydiae, legionellae, mycoplasmata and non-tuberculous mycobacteria.

Toxicity - These are very safe antibiotics with low toxicity.

Tetracyclines

The tetracyclines have a basic structure of four fused rings with side-chain changes to produce different drugs. They are bacteriostatic.

Pharmacokinetics - Tetracyclines are orally absorbed yet are also injectable; they are widely distributed including CSF and brain. Metabolism occurs in the liver, and excretion is renal.

Spectrum - The spectrum of tetracyclines is very broad, including most pathogenic bacterial genera (except *Pseudomonas*), plus chlamydiae, mycoplasmata and rickettsiae, but is not deep, with many resistant bacterial strains. The use of tetracyclines as growth promoters added to livestock feed increased the numbers of resistant strains. Use is, therefore, chiefly for unusual bacteria.

Toxicity - This is low, apart from deposition in immature bone and teeth (causing discoloration), and the usual allergy or gut intolerance.

Nucleic acid synthesis inhibitors

Nitroimidazoles

Metronidazole and tinidazole are nitroimidazoles, with a unique mode of bactericidal action, acting as electron acceptors and producing intermediate compounds toxic to bacterial DNA.

Pharmacokinetics - They are well absorbed, penetrate widely, are metabolised by the liver, and excreted by the kidney.

Spectrum - Their spectrum includes almost all pathogenic anaerobes (except some cocci), microaerophilic bacteria and some parasites. Their principal use is in prophylaxis and treatment of anaerobic bacterial infections, and in amoebiasis, giardiasis and trichomoniasis.

Quinolones

Structurally, the quinolones (e.g. norfloxacin and ciprofloxacin) are fluoroquinolone carboxylic acid derivatives with two six-member rings, and distinctive side chains. Action is bactericidal by inhibiting bacterial DNA gyrase, hence preventing supercoiling of DNA.

Pharmacokinetics - Quinolones are well absorbed and distributed, but serum concentrations are low. Their half-lives are relatively long, about 4 hours, and excretion is renal.

Spectrum - Older drugs mainly kill Gram-negative bacteria including pseudomonads, with poor activity against Gram-positive and anaerobic bacteria, now improved with broad-spectrum quinolones. Norfloxacin is used mainly in urinary and gut infections, while ciprofloxacin is used chiefly in serious systemic Gram-negative infections. Numerous other quinolones are available in different countries.

Toxicity can affect the CNS with headache, mood changes and fits.

Trimethoprim and sulphonamides

Sulphonamides are derived from sulphanilamide, a single ring compound structurally similar to, and competing with, an intermediate in folic acid synthesis, para-aminobenzoic acid. They are commonly used in combination with trimethoprim, which inhibits the next step to tetrahydrofolic acid in folic acid synthesis. Co-trimoxazole is sulphamethoxazole plus trimethoprim.

Pharmacokinetics - All have good absorption, wide distribution, long half-lives (to 10 hours) and renal excretion.

Spectrum - Chiefly Gram-negative rods were sensitive, but resistance is now common. Uses are mild urinary or respiratory infections, and unusual infections including *P. jirovecii* pneumonia, nocardiosis, chancroid and typhoid fever.

Toxicity - This is mainly allergy with rash and fever. Megaloblastic anaemia is uncommon and reversed by folinic acid.

Rifamycins

These are red-coloured derivatives of a natural product, rifamycin B.

Pharmacokinetics - All are well absorbed orally, penetrate widely including the CNS, and are cleared by liver metabolism and excretion mainly in bile.

Spectrum - Because rifamycins enter cells they are used against intracellular organisms such as mycobacteria. Rifampicin is a first line drug for TB and leprosy, and also used for resistant (especially MRSA) staphylococcal infections (with fusidic acid) and as prophylaxis in contacts of meningococcal and *Haemophilus meningitis*. Rifabutin is used in combination with other drugs (e.g. ethambutol, clarithromycin) in the treatment and prophylaxis of atypical mycobacterial infections in AIDS.

Toxicity - Rifampicin has few toxic side effects but numerous drug interactions. It colours body fluids blood-red.

The 'ABC' of antibiotics (antibiotic - spectrum - clinical use).

Antibiotic	Bacterial spectrum							Clinical uses
	AnO	Sta	Str	Enc	GNC	GNR	Ps	
Penicillins								
Flucloxacillin	++	3+	++	+	-	-	-	Staphylococcal infections (not MRSA)
Penicillin V, G	++	+	3+	++	++	+	-	Erysipelas, gas gangrene, endocarditis, meningitis, pharyngitis
Ampi/amoxicillin (improved by BLI)	++	+	++	3+	++	+	-	Bronchitis/sinusitis/otitis media; combined with gentamicin in abdominal and urinary infections
Ticar/piperacillin (improved by BLI)	++	+	++	++	++	+	3+	Pseudomonal infections, often combined with aminoglycoside
Cephalosporins								
First: cephalothin, cefazolin, cephalexin	++	3+	++	0	+	++	-	Surgical chemoprophylaxis, skin/soft tissue infections, also combined with aminoglycosides
Second: cefuroxime, cefoxitin, cefotetan	3+	++	++	0	++	++	-	Mild-moderate mixed anaerobic-aerobic skin/soft tissue, respiratory and abdominal infections
Third: cefotaxime, ceftriaxone	++	+	++	0	3+	3+	+	Severe Gram-negative infections including meningitis
3/4: ceftazidime, cefepime	++	+	+	0	3+	3+	3+	Pseudomonal and other Gram-negative rod infections
Other beta-lactams								
Aztreonam	-	-	-	-	3+	3+	3+	Severe systemic Gram-negative infections
Imipenem, meropenem	3+	3+	3+	++	3+	3+	3+	Severe systemic infections (not MRSA)
Vancomycin, fusidic acid, linezolid, streptogramins								
	-	MRSA	++	3+	-	-	-	Serious staphylococcal infections, especially MRSA; enterococcal infections
Aminoglycosides								
Streptomycin	-	-	-	-	-	3+	-	Tuberculosis, Gram-negative zoonoses (brucellosis, plague, tularaemia)
Gentamicin	-	-	-	-	++	3+	3+	Severe systemic Gram-negative infections
Tobramycin	-	-	-	-	++	++	3+	Pseudomonal infections
Amikacin	-	-	-	-	++	3+	3+	Otherwise-resistant Gram-negative rod infections
Chloramphenicol	3+	MRSA	3+	++	3+	++	-	MRSA, rickettsial and rare infections
Lincosamides								
Clindamycin, lincomycin	3+	++	++	-	-	-	-	Anaerobic infection, toxoplasmosis
Macrolides								
Erythromycin	++	++	++	+	++	-	-	Chancroid, legionellosis, pertussis, Campylobacter infection
Roxithromycin	++	++	++	+	++	-	-	Erythromycin substitute
Azithromycin	3+	+	+	-	3+	-	-	<i>Chlamydia</i> , STDs, unusual infections
Clarithromycin	++	3+	3+	+	3+	-	-	<i>Chlamydia</i> , MAC, leprosy
Tetracyclines	+	+	+	+	+	+	-	Unusual bacteria: <i>Borrelia</i> spp., brucellae, mycoplasmata, rickettsiae, vibrios
Nitroimidazoles								
Metronidazole, tinidazole	3+	-	-	-	-	-	-	Anaerobic infections, amoebiasis, giardiasis
Quinolones								
Norfloxacin	-	-	-	-	-	++	-	Urinary infections (2nd line)
Ciprofloxacin	-	+	+	-	3+	3+	++	Serious Gram-negative infections (MRSA)
Moxi/gatifloxacin	++	++	++	++	3+	3+	++	Difficult atypical pneumonia
Rifamycins								
Rifampicin (rifabutin p. 231)	++	MRSA	3+	3+	3+	++	+	TB, MRSA, leprosy, prophylaxis in meningococcal and haemophilus meningitis
Trimethoprim and sulphonamides	-	+	+	-	+	++	-	Mild urinary and respiratory infections, nocardial infection, toxoplasmosis, PCP

Key :

Efficacy: 3+, usually effective (> 80% strains usually sensitive); ++, moderate efficacy (50–80% strains usually sensitive); +, poor efficacy (25–50% of strains usually sensitive); –, minimal or no efficacy; 0, no efficacy. Hint to students: highlight every 3+.

AnO, anaerobes (e.g. *Clostridia*, *Bacteroides* spp.); Sta, staphylococci; Str, streptococci; Enc, enterococci; GNC, Gram-negative cocci (e.g. *Neisseria*, *Haemophilus* spp.); GNR, Gram-negative rods, especially enteric (e.g. *E. coli*); Ps, pseudomonads; MAC, *Mycobacterium avium* complex; MRSA, methicillin- (and multi-) resistant *S. aureus*; PCP, *Pneumocystis jirovecii* pneumonia; STD, sexually transmitted diseases; BLI, beta lactamase inhibitor.

Taken from : 'Clinical Microbiology and Infectious Diseases'. Spicer, 2008.

KEY LEARNING POINTS.



1. Spectrum determines use, which is modified by toxicity and cost.
2. Penicillins, cephalosporins and other beta-lactams are safe, effective bactericidal antibiotics of great use in a wide range of infections, limited by developing resistance.
3. Aminoglycosides, particularly gentamicin, are very effective against many Gram-negative infections but need careful dosing and monitoring because of toxicity. Resistance is a lesser problem.
4. Metronidazole is very useful against most anaerobes, while clindamycin is less reliable and more costly.
5. Older broad-spectrum bacteriostatic antibiotics including chloramphenicol, co-trimoxazole and tetracyclines are now mainly used for unusual bacteria, chlamydiae, mycoplasmata or rickettsiae.
6. Quinolones are costly and used mainly in special infections.
7. Vancomycin, teicoplanin and linezolid are reserved for serious resistant Gram-positive infections.