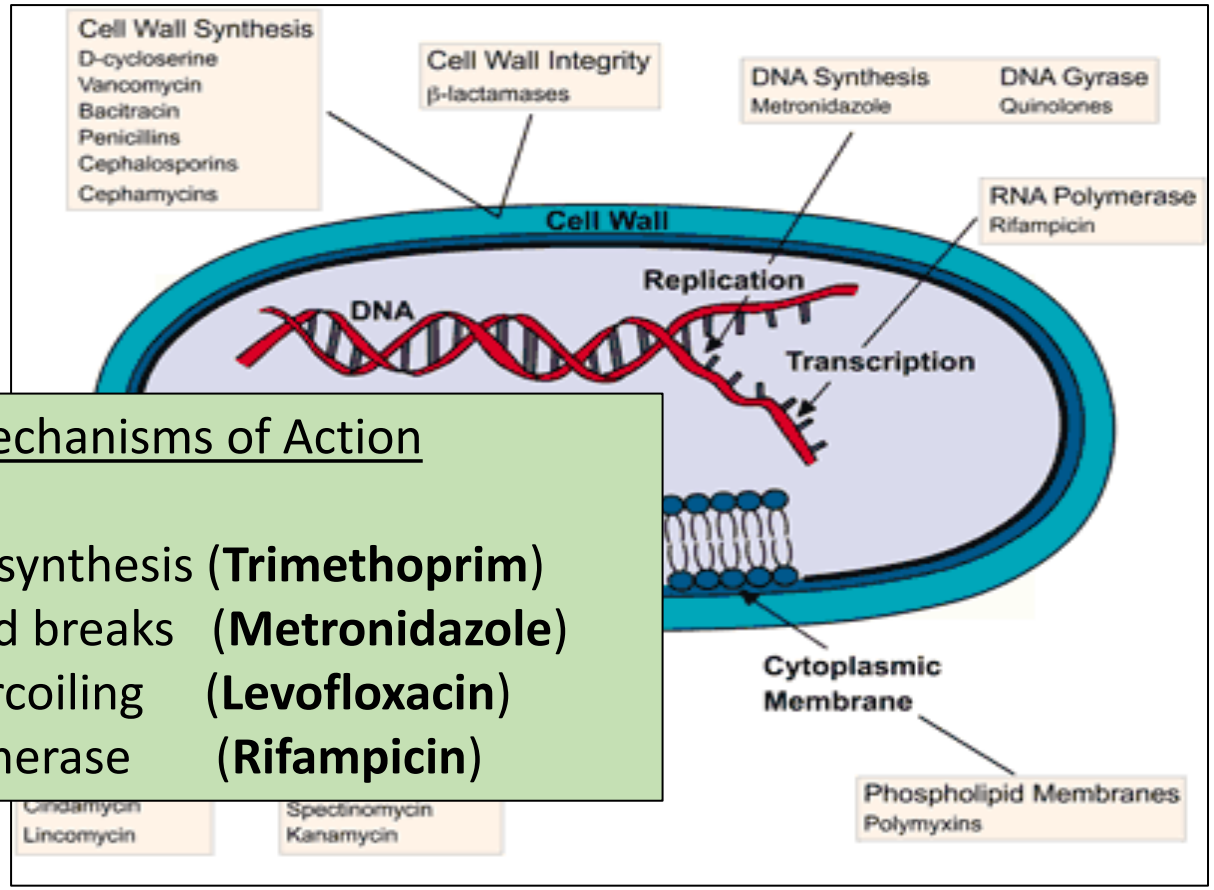
Antibiotics 2 : **DNA + Mycobacteria**

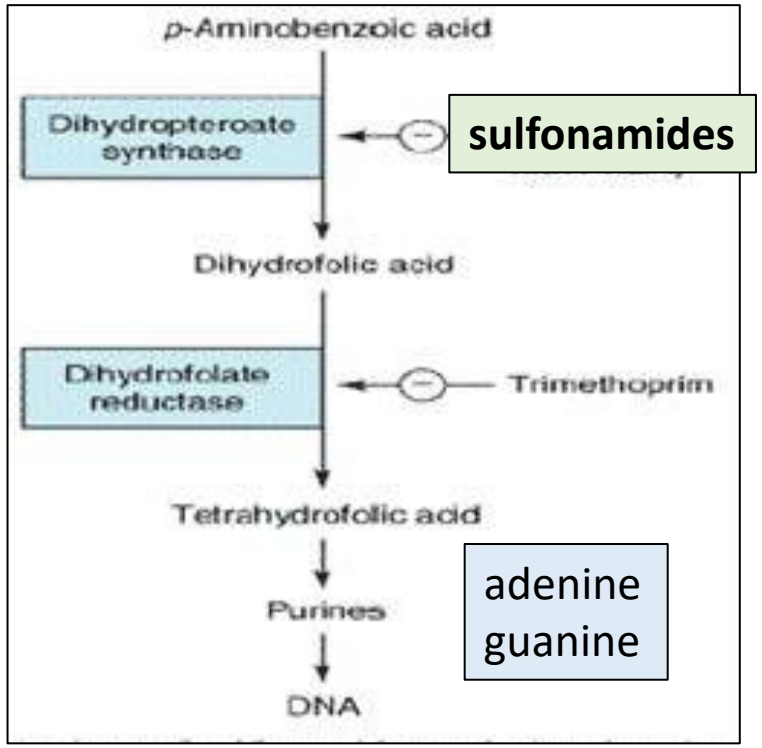


Mechanisms of Action

1. DNA base synthesis (**Trimethoprim**)
2. DNA strand breaks (**Metronidazole**)
3. DNA supercoiling (**Levofloxacin**)
4. RNA polymerase (**Rifampicin**)

The second lecture on antibiotics has been divided into 2 bitesize recordings dealing with antibiotics which firstly influence DNA or secondly inhibit protein synthesis in the ribosome. Antibiotics inhibit bacterial growth by influencing DNA in 4 major ways, inhibiting nucleotide base synthesis with the sulphonamides, preventing the unwinding of DNA supercoils with the fluoroquinolones, inducing DNA strand breaks with metronidazole and inhibiting RNA polymerase with rifampicin. The combination drug therapy required for the treatment of tuberculosis is included in this recording where drugs used in combination may also inhibit the mycobacterial cell wall or inhibit the ribosome.

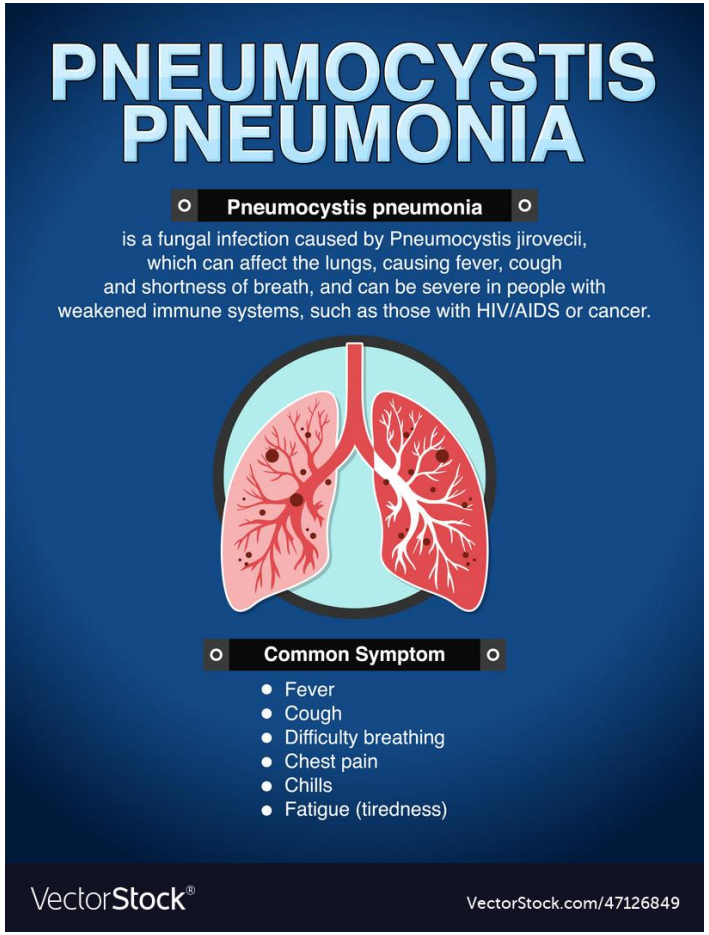
Sulfonamides : Inhibit Nucleotide Base Synthesis



Sulfanilamide (1935)
First systemic antibacterial agent

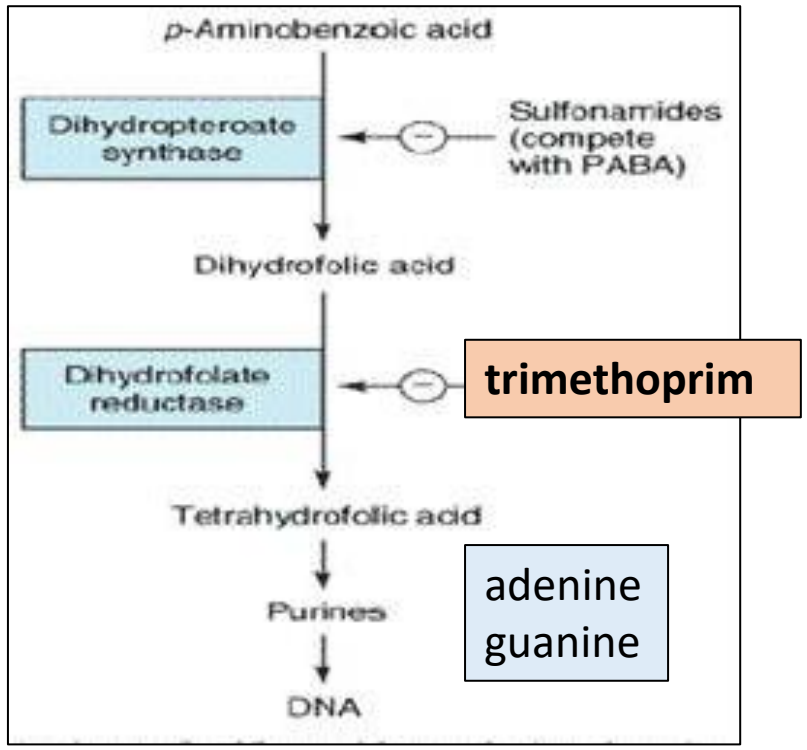
Selective Toxicity
Bacteria make their own folic acid. Humans can use folic acid from diet. Tetrahydrofolate is required for the synthesis of nucleotide bases, within the structure of DNA.

Co-Trimoxazole
(Sulfamethoxazole + trimethoprim)
Combination now reserved for pneumocystis pneumonia in immunosuppressed patients



The first systemic antibacterial agents, the sulfonamides, were developed from Paul Ehrlich's concept that drugs must 'bind to act' and were based on the dyes used to bind to and colour cloth. The sulfonamides are bacteriostatic agents which compete for the substrate binding site on an enzyme involved in the synthesis of folic acid. Folic acid is required for synthesis of the purine nucleotide bases present in DNA. A selective effect on bacteria is obtained since, bacteria have to synthesise folic acid, while man obtains folic acid from the diet. The mechanism of action of sulfonamides is to compete with the substrate p-aminobenzoic acid for the enzyme dihydropteroate synthase. The result is a lack of tetrahydrofolate required to form the ring structure of the purine bases present in the nucleotide structure of DNA. Sulfonamide powders were widely used with wound dressings in the second world war but are rarely used as sole agents today in the UK. In 1962, a second agent trimethoprim was introduced which inhibits the next enzyme downstream in the folic acid pathway, dihydrofolate reductase. A combination drug product was produced with the sulfonamide, sulfamethoxazole, called co-trimoxazole which was widely used as a broad-spectrum antibiotic. Today however, this combination is reserved to treat severe respiratory infections in immunosuppressed patients, in particular pneumocystis pneumonia, a fungal infection of the lung causing fever, cough and shortness of breath.

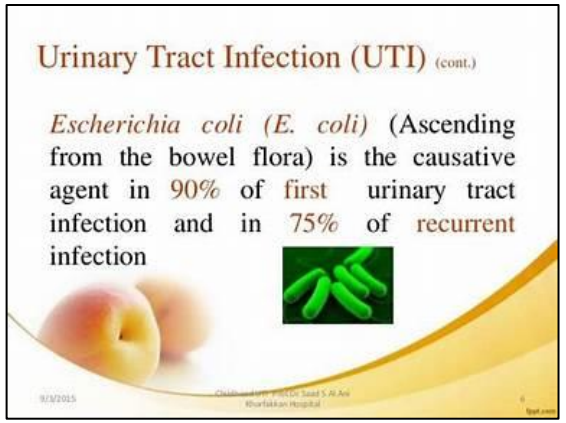
Trimethoprim : Inhibitor of Nucleotide Base Synthesis

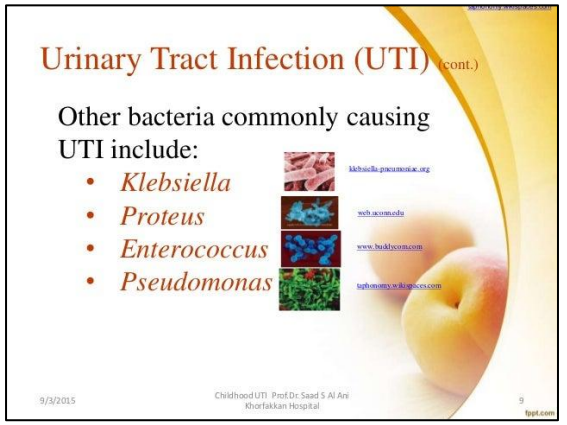


Trimethoprim (1962)

inhibits dihydrofolate reductase
bacteria (1,000x) human
Broad spectrum (not Pseudomonas A)
Aerobic gram-positive microorganisms
Staphylococcus species
(coagulase-negative strains, including *S. saprophyticus*)

Aerobic gram-negative microorganisms
Enterobacter species, Escherichia coli
Klebsiella pneumoniae
Proteus mirabilis





Trimethoprim is a selective inhibitor of the bacterial form of dihydrofolate reductase with a 1,000 higher affinity for the bacterial than the human form of the enzyme. Although trimethoprim has a broad spectrum of activity, it is excretion unchanged in the urine, and together with activity against both gram-positive and gram-negative bacteria including E.Coli, make it one of two drugs recommended by NICE (NG109) for the first line treatment of uncomplicated infections of the lower urinary tract.

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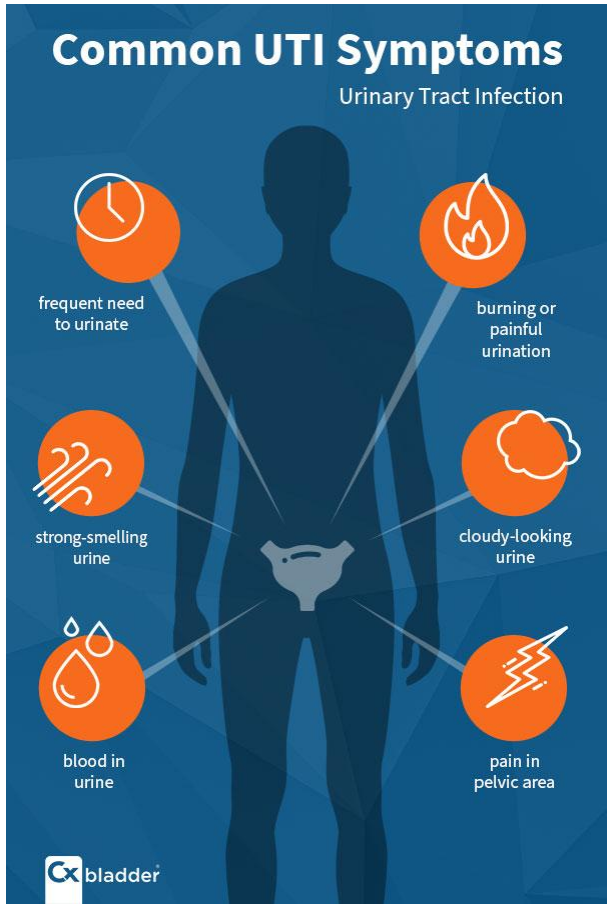
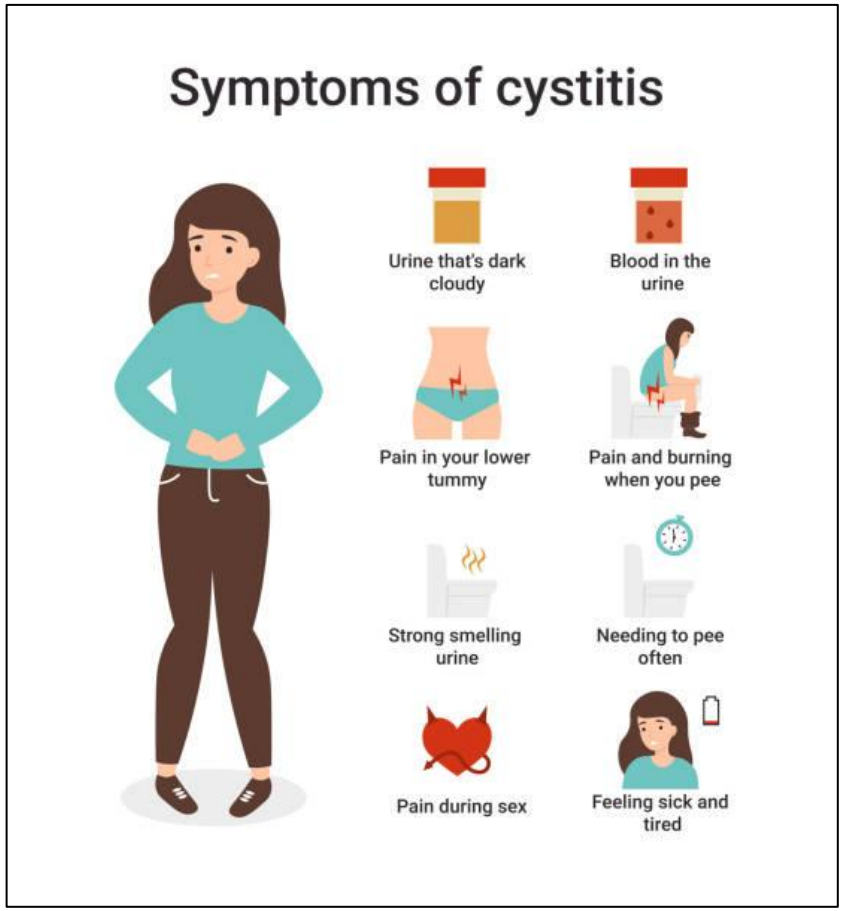
Trimethoprim



Indication :
Uncomplicated urinary tract infection.
Kinetics : Mainly excreted unchanged by kidneys higher concentration in urine than blood (also inhibits vaginal/faecal sources)
dose reduction in renal failure

Adverse Effects
GI upset
Skin rash - hypersensitivity
Haematological disorders
Hyperkalaemia
* CI 1st trimester of pregnancy



Trimethoprim and nitrofurantoin are the two antibiotics recommended for the first line treatment of uncomplicated lower urinary tract infections using a 3-day course of treatment. Common symptoms of a urinary tract infection include pain on urination and pain in the pelvic area, an increased urinary frequency and a change in the urine which may look cloudy, have a strong smell or more severely contain blood. However, resistance to trimethoprim can develop quickly and since folic acid is required for foetal development including the brain and spinal-chord, trimethoprim is contraindicated in the 1st trimester of pregnancy. Trimethoprim has structural similarities with some diuretics acting on the distal nephron and like such diuretics may induce hyperkalaemia as an adverse effect. Dosage adjustment is required in patients with reduced renal function

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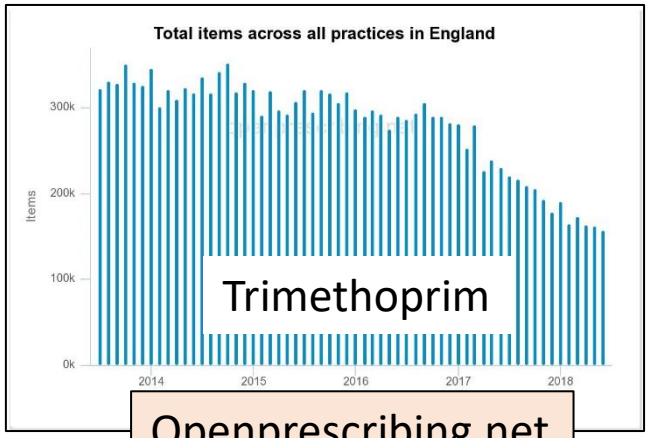
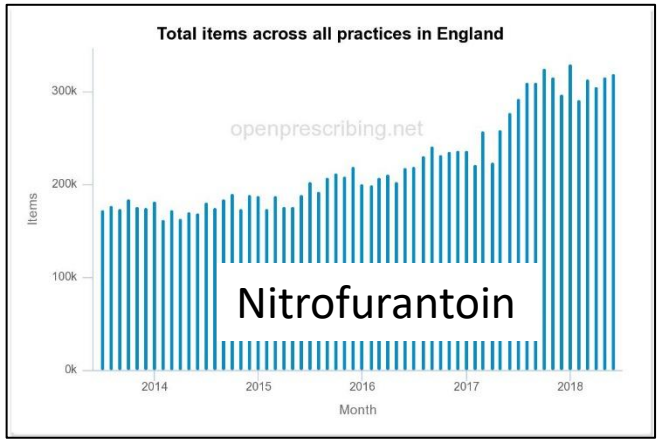
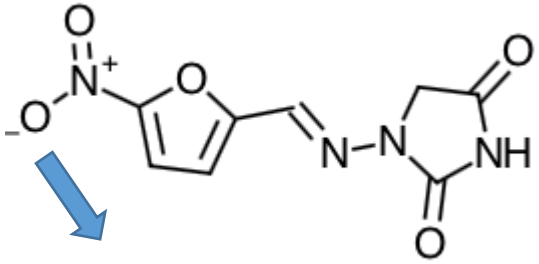
Nitrofurantoin



Openprescribing.net



Nitroso reductase



Free Radical

DNA damage
RNA damage
Protein Damage

Urinary tract infection
Multiple antibacterial mechanisms
(Low resistance)

Pharmacokinetics

75% 1st pass metabolism
25% Renal excretion unchanged
Poor tissue penetration
Low plasma concentration (1µg/ml)
High urinary concentration (200µg/ml)

Nitrofurantoin (1953) is activated by a nitroso reductase enzyme in bacterial cells to produce a free radical which can damage DNA, RNA and proteins. This is useful since potential activation in human cells might induce cancer or teratogenicity. Its multiple mechanisms of action give it a low level of resistance which probably explains the increase in its use over trimethoprim. Following extensive 1st pass degradation, 25% of an oral dose is excreted unchanged in the urine. This water solubility means poor tissue penetration, low plasma concentration but high concentration in the bladder. The plasma concentration is not high enough for nitrofurantoin to be used as a systemic antibiotic. Nitrofurantoin is not recommended in patients with a low eGFR (<45ml/min) where bladder concentrations will be reduced and plasma levels will increase. Nitrofurantoin is recommended for use in the 1st/2nd trimesters of pregnancy although avoided in the 3rd trimester due to possible neonatal haemolysis.

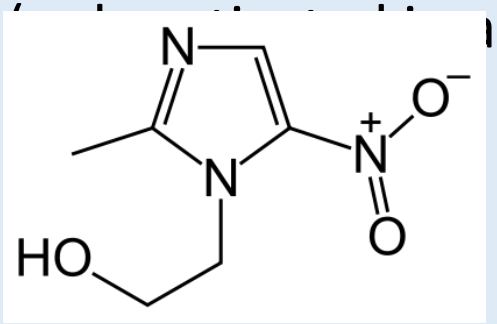
DNA : Strand Breaks

metronidazole (1960)

(initially antiprotozoal)

generates free radicals

(…naerobic cells)



Free Radical

antibacterial mechanism

strand break in DNA

Indications :

Oral infection/aspiration pneumonia

Surgical infection

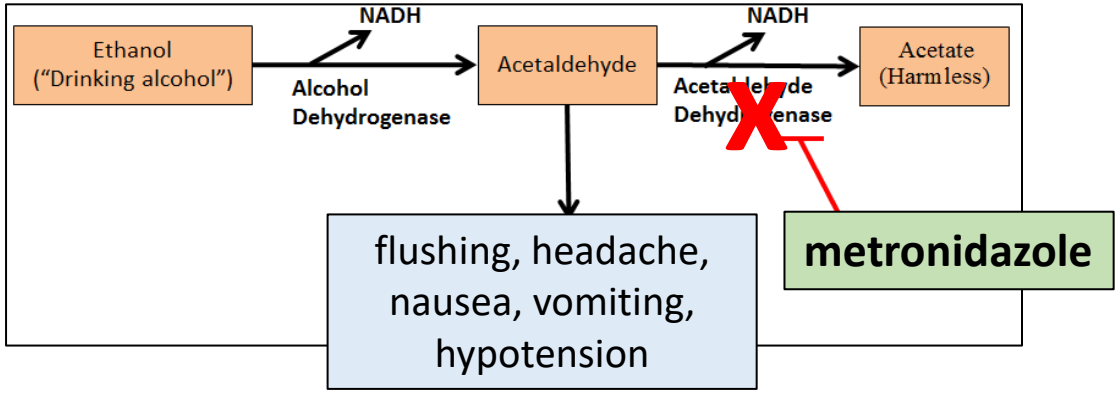
Drug of choice mild-to-moderate C difficile antibiotic associated colitis

Adverse Effects

Avoid alcohol

Drug interaction - inhibits CYP3A4

Long term – neurological effects



A second major mechanism to influence DNA is the induction of strand breaks with metronidazole. Metronidazole is activated in anaerobic cells to also generate free radicals which induce strand breaks in DNA. Originally introduced as Flagyl, an antiprotozoal agent, metronidazole is particularly effective at treating GI tract infections including abdominal sepsis and second-line n treating C. Difficile colitis caused by other oral antibiotic therapy. Important adverse reactions include an interaction with alcohol by inhibiting the enzyme acetaldehyde dehydrogenase, the second step in alcohol metabolism, inducing hypotension facial flushing and headache due to excess acetaldehyde concentrations.

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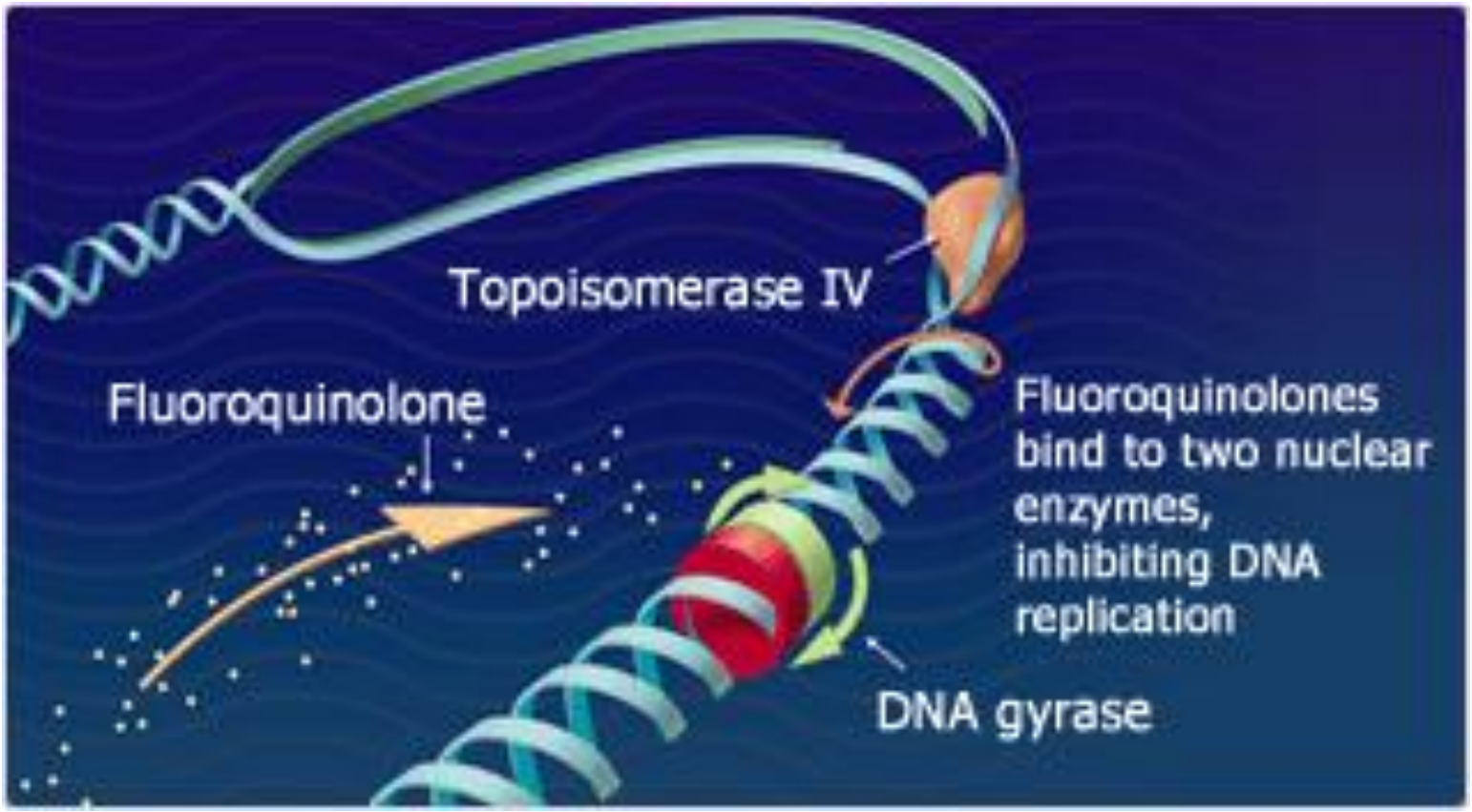
DNA : Inhibition of Unwinding DNA Supercoils



nalidixic acid
(1967)

ciprofloxacin
(Bayer 1987)
(active against gram –ve *Pseudomonas A*)

Respiratory Quinolones
(Active against *Strep. Pneumoniae*)

levofloxacin (1996)

moxifloxacin (1999)
(also active against MRSA)

A third mechanism for antibiotics to influence DNA is the ability to inhibit enzymes involved in the unwinding of DNA supercoils. In order to transfer its message, the DNA double helix must unwind to allow helicase enzymes to separate its base-pairs into a single strand. However, in doing so, supercoils are generated proximal to the site of separation, which are continually unwound by the cut/twist/reseal mechanisms of gyrase and topoisomerase enzymes. This mechanism was originally discovered in a bi-product of the synthesis of the anti-malarial agent chloroquine, called nalidixic acid which, because of its renal excretion, was used to treat urinary tract infections. The addition of a fluorine substituent produced ciprofloxacin, an oral broad-spectrum antibiotic, with efficacy against *Pseudomonas A*, responsible for serious gram-ve infections. Ciprofloxacin, introduced by Bayer in 1987, was by 2003 producing one third of the revenues for the company before the patent expired. Two further fluoroquinolones were introduced in the 1990's, termed respiratory quinolones, due to their activity against gram-positive Streptococcus pneumoniae. Activity against Pseudomonas A and MRSA, reduced with levofloxacin, was restored with the introduction of moxifloxacin in 1999.

Fluoroquinolones

Indications

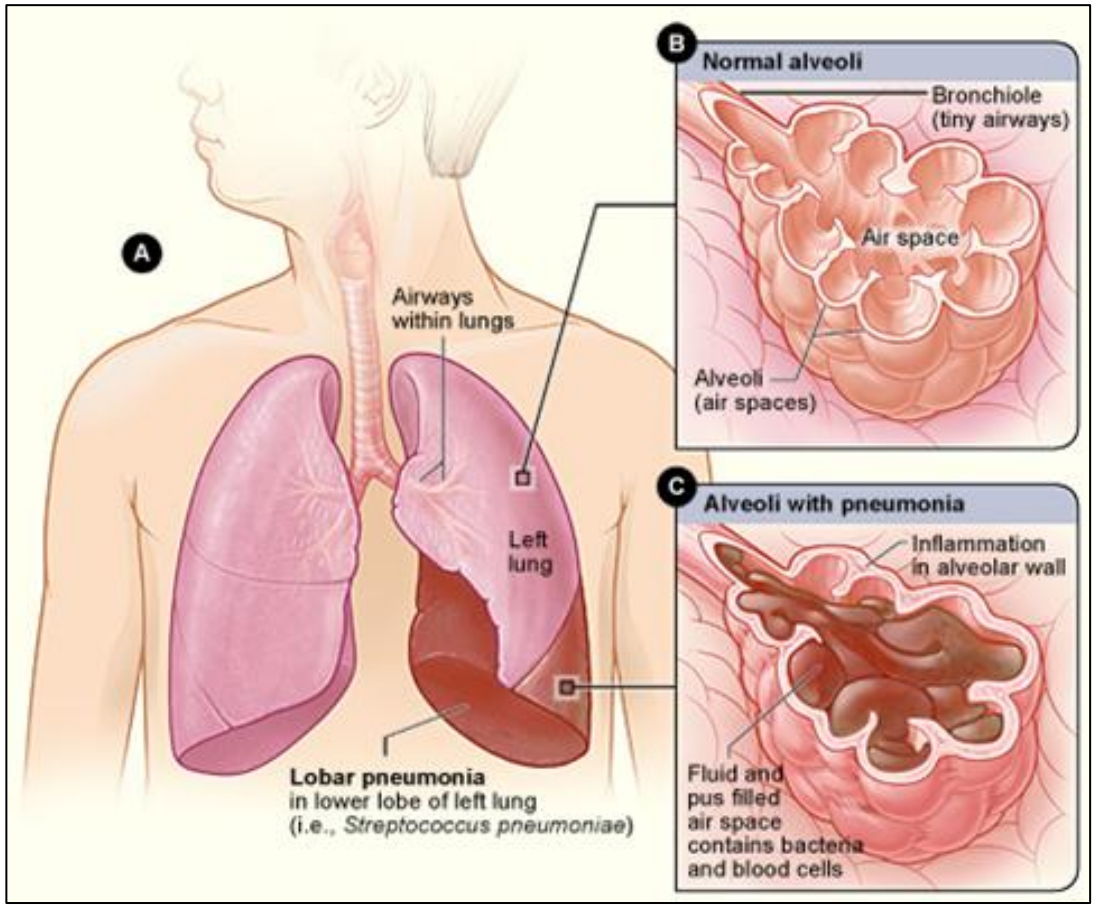
Ciprofloxacin is the only oral antibiotic in common use active against Pseudomonas Aeruginosa

- Effective against aerobic gram-ve bacteria - urinary tract infection/severe GI tract infection
- Rapid resistance : reserved for 2nd/3rd line use
  - Active against anthrax!!

levofloxacin & moxifloxacin

enhanced gram +ve activity (*S. pneumoniae*)

lower respiratory tract infection



Ciprofloxacin was stockpiled by the US government during the anthrax postal scares in the 1990s and is the only commonly used oral broad spectrum antibiotic which is effective against *Pseudomonas. A.* Ciprofloxacin is effective against aerobic gram-negative bacteria responsible for severe urinary tract and GI tract infections, but today tends to be reserved for 2/3rd line treatment due to the rapid development of resistance. Levofloxacin/moxifloxacin are called the respiratory quinolones with activity against Streptococcus pneumoniae responsible for the pneumonia of lower respiratory tract infections. The accumulation of fluid and pus in pneumonia fills the alveolar space in the lower respiratory tract reducing the surface area available for oxygen transport. Moxifloxacin also has activity against MRSA.

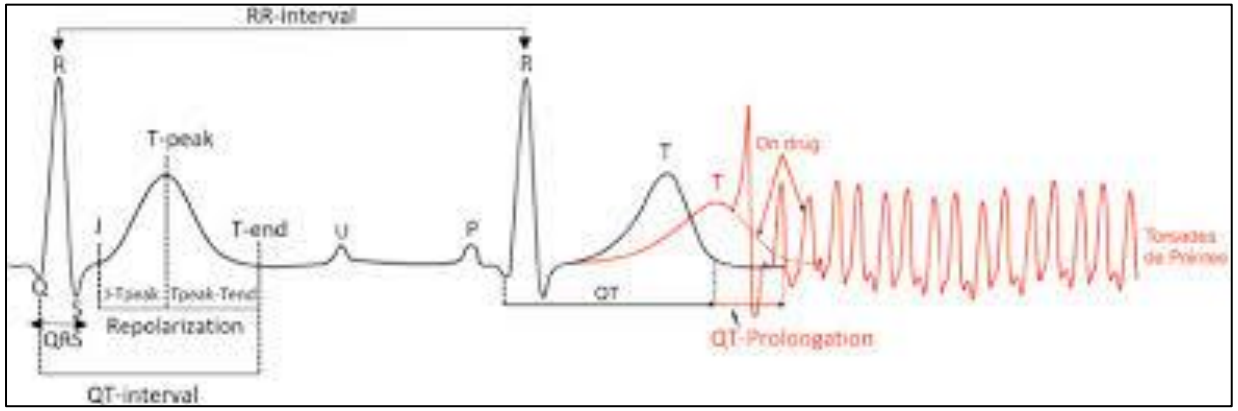
Fluoroquinolones : Adverse Effects

- Nausea/diarrhea; Promote C/Difficile colitis
- Increase arrhythmias (prolong QT interval)
- Lower seizure threshold
- Tendinitis leading to rupture of muscle tendons

Interactions

- Drugs/conditions that prolong the QT interval
- Inhibits CYP – theophylline
- NSAID – increase seizures

Prolongation of the QT Interval



Potential for Torsade de Pointes
(a form of ventricular tachycardia)

The adverse effects of the fluroquinolones include diarrhoea with an increased ability to induce C. Difficile overgrowth and colitis. The ability to delay repolarisation in the heart, prolongs the QT interval on the ECG, with the potential to produce Torsade de Pointes, a serious form of ventricular tachycardia, shown by the rapid, spiked wave-forms on the ECG in red on the right-hand side of the slide. This property is shared by the macrolide antibiotics such as clarithromycin. Other adverse effects include lowering the seizure threshold and inducing tendinitis, which can lead to tend rupture.

DNA : **RNA Polymerase**

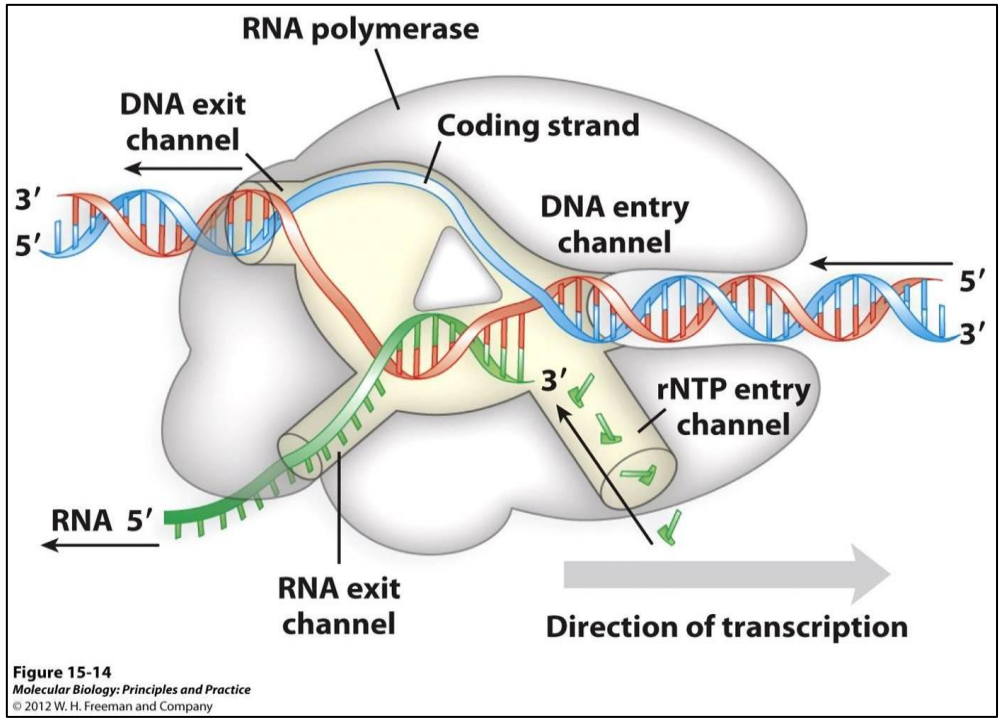


Figure 15-14
Molecular Biology: Principles and Practice

Rifampicin inhibits RNA polymerase
Prevents transcription of DNA to the RNA required for encoding protein synthesis

Rifampicin (1967)

Semi-synthetic broad spectrum antibiotic

Only inhibits RNA polymerase in prokaryotic cells

Orally active – enterohepatic cycling

Induces CYP enzymes

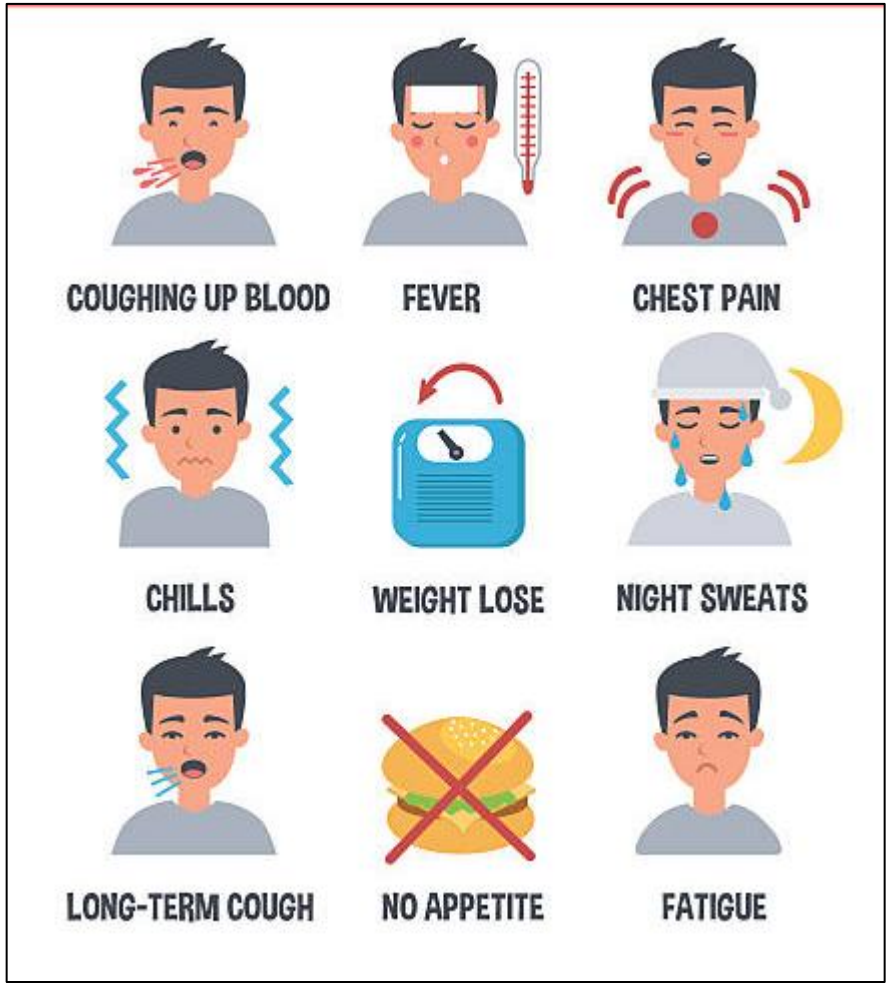
decreasing effect of liver metabolised drugs

Used in combination with isoniazid/ethambutol treat for 6-12 months

Also active against leprosy

RNA polymerase is the enzyme which transcribes the DNA message onto the RNA required to encode protein synthesis in the ribosome. Unlike some other bacterial infections, the tubercule bacillus is very slow growing requiring combination therapy which includes combination with the cell wall antibiotics, isoniazid and ethambutol, marketed as a Tritab, administered over a period of 6-12 months. Rifampicin has previously been discussed in pharmacokinetics as a drug which can induce drug-metabolising enzymes in the liver leading to a reduction in the blood levels and thereby the effectiveness of other liver-metabolised drugs.

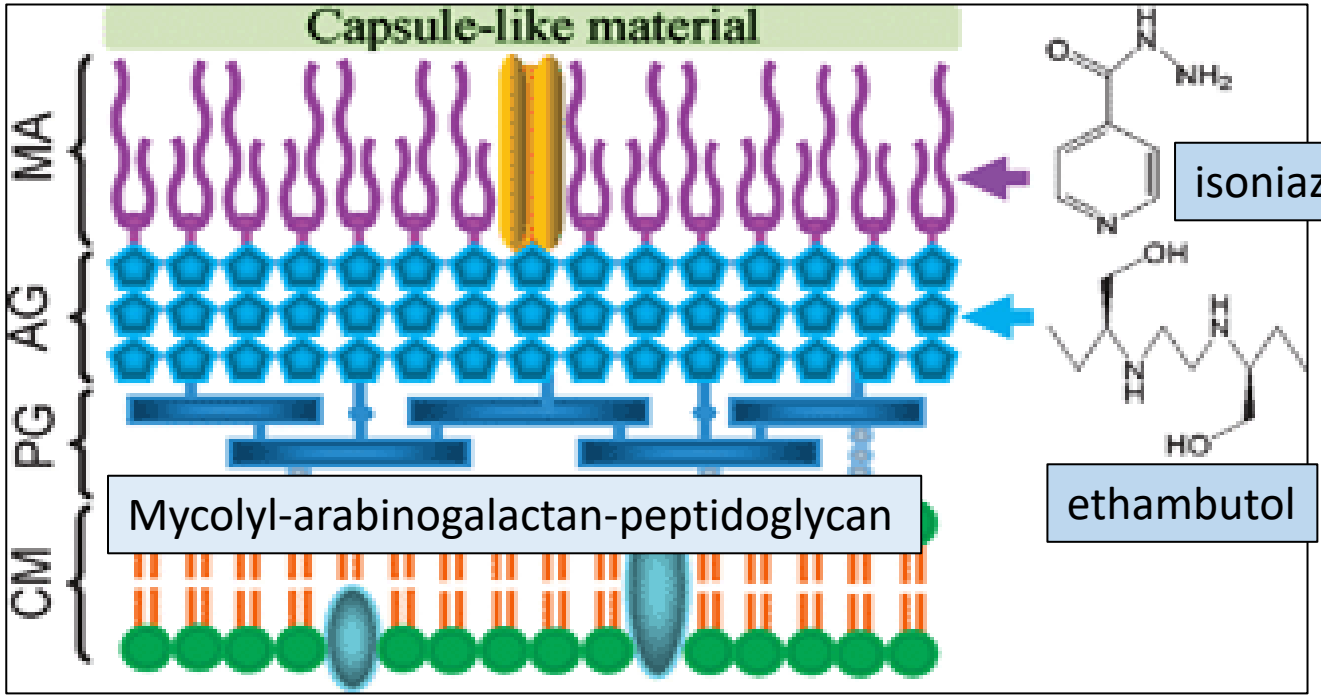
Mycobacteria



M. Tuberculosis

leading infectious cause of death worldwide

Cough, blood containing sputum, fever, night sweats, weight loss



Tuberculosis, previously known as consumption, is a leading infectious cause of death worldwide giving the symptoms of cough, blood-containing sputum and weight loss,. Both tuberculosis and leprosy are caused by slow growing mycobacteria which have a different cell wall structure to most gram +/-ve bacteria requiring multiple drug therapy. There are 3 major components to the cell wall of mycobacteria, an outer layer of mycolic fatty acids (shown in purple) over a central layer of arabinogalactan sugars (shown in light blue) with a peptidoglycan layer (shown in dark blue) lying next to the cell membrane. B-lactam antibiotics are however ineffective since the outer cell wall contains a phospholipid layer preventing their access. The major target sites for drugs acting on the cell wall of mycobacteria are the mycolic fatty acid and arabinogalactan layers.

Isoniazid

(eye-so-NYE-a-zid)

Isoniazid (1951) Prodrug

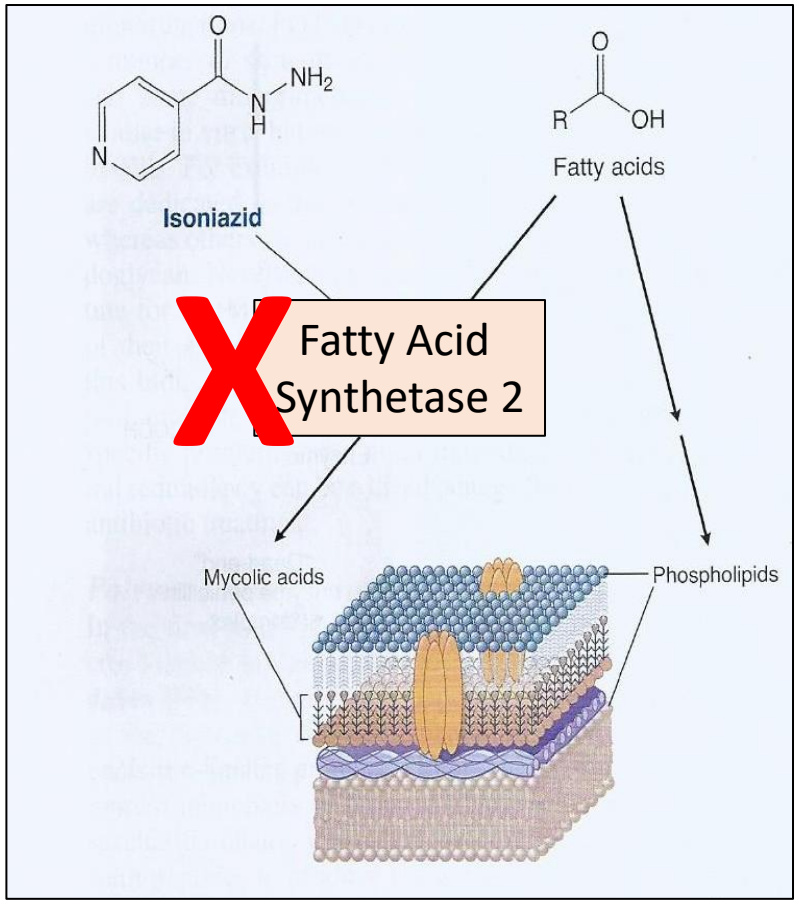
Good CSF penetration and into caseous granulomas
(necrotic tissue of tubercular lesions)

- Activated by bacterial peroxidase enzyme (KatG)
- Enzyme inhibitor of fatty acid synthetase 2 (FAS-2)
- inhibits polymerisation of fatty acids into long chain mycolic acid
- Metabolised by acetylation
- genetically determined into fast and slow metabolisers

Long term treatment (6-12 months) – slow growing bacteria
Multidrug therapy only – rapid resistance

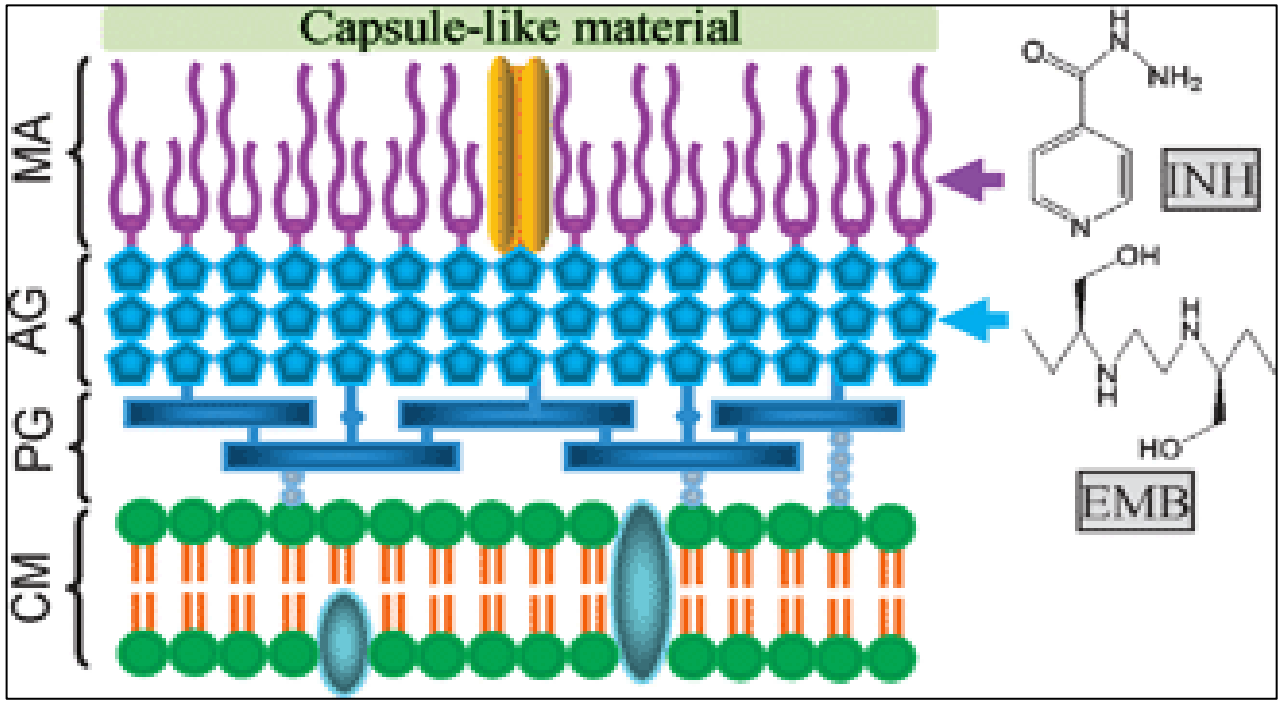
Adverse Effects

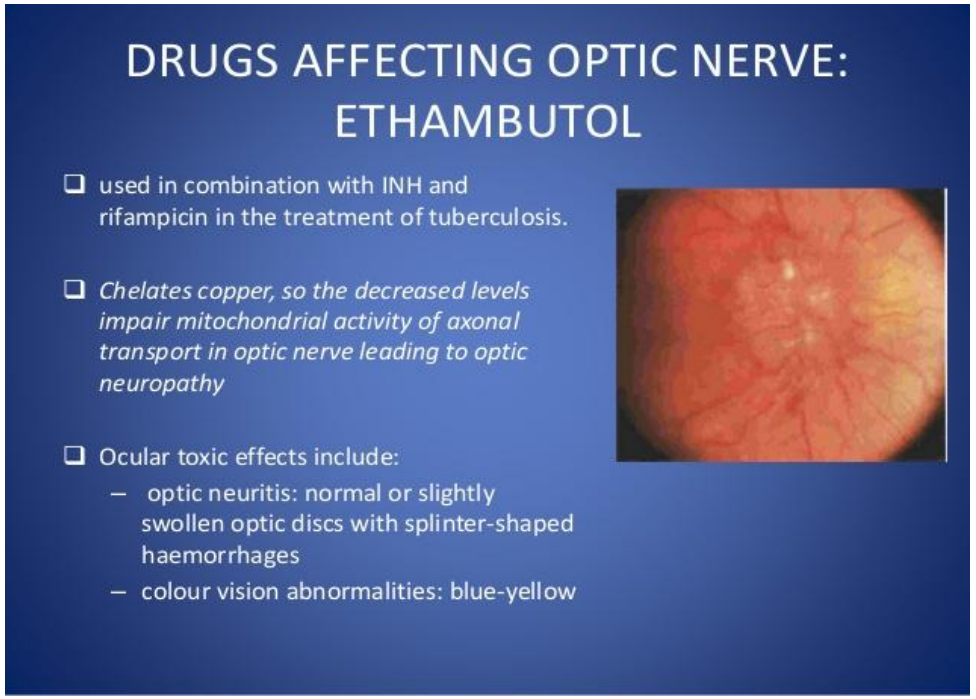
Hepatitis, peripheral neuropathy (prevented by vit B6)
Raised plasma levels of anti-epileptic drugs



The antibiotics used in the treatment of tuberculosis form a link between drugs which inhibit the bacterial cell wall and drugs which inhibit DNA or protein synthesis.. Probably the most well-known antitubercular drugs are isoniazid and rifampicin. Isoniazid, introduced in 1952, has good penetration into tubercular lesions and the CSF but important to its lack of toxicity, is that isoniazid requires activation by the tubercule bacillus itself. The activated product inhibits the enzyme FAS-2 which polymerises fatty acids into the long chain mycolic acid structure of the mycobacterial cell wall. The metabolism of isoniazid by acetylation is genetically determined and the Caucasian population can be divided into 50% fast and 50% slow metabolisers. Adverse effects of isoniazid include hepatitis and peripheral neuropathy (which can be treated by vitamin B6 supplementation). Pharmacokinetic drug interactions are possible, such as a reduction in the metabolism of anti-epileptic agents

Ethambutol





Ethambutol (1961)

Enzyme Inhibitor - inhibits arabinosyl transferase

- prevents attachment of arabinose to arabinogalactan chain
- prevents attachment of sugar chain to mycolic acids
- disrupts the mycolyl-arabinogalactan–peptidoglycan complex

Regular eye examination essential

Optic neuritis/ impaired visual acuity

Loss of colour discrimination

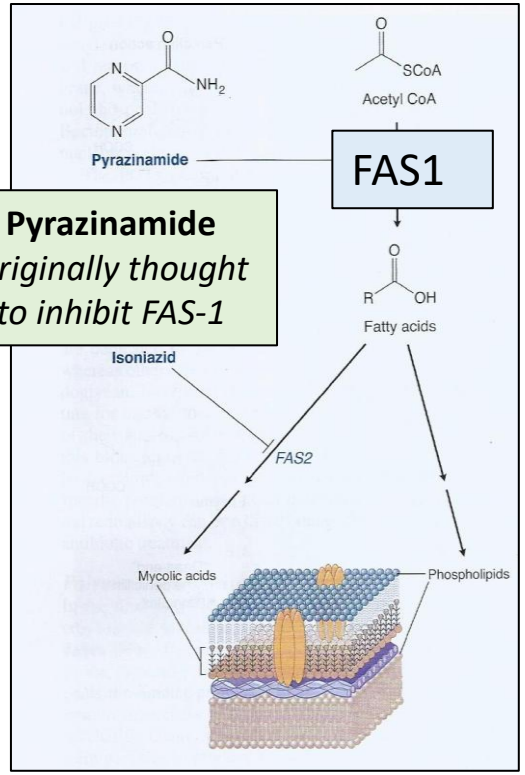
Constricted visual field

Usually occur after 1 month

(generally reversible)

A second antitubercular antibiotic which works on the mycobacterial cell wall, used together with isoniazid is ethambutol. Ethambutol, in contrast to isoniazid, inhibits the synthesis of a different layer of the cell wall, the sugar arabinogalactan layer, by inhibiting the enzyme arabinosyl transferase. The major adverse effect of ethambutol is on the eye, impairing visual acuity, constricting the visual field and losing colour discrimination. Such effects are however generally reversible on the cessation of therapy.

Pyrazinamide



First line multidrug therapy of Tuberculosis

Isoniazid, Ethambutol, Rifampicin, Pyrazinamide

Pyrazinamide is a tissue prodrug converted by M. Tuberculosis to pyrazoic acid

Pyrazinamide is synergistic with rifampicin

Unique anti-TB drug

Primary activity on non-growing persisters

(Kills TB persisters not killed by other drugs)

reduces treatment duration from 12 to 6 months

Mechanism Proposed :

Inhibition ribosomal protein S1 which binds mRNA a possibility

(Activity increases with decreased metabolic activity at an acidic pH as seen in the earlier inflammatory phase (2 months) of infection)

The duration of combination therapy in tuberculosis was reduced to 6 months by the addition of a fourth drug, pyrazinamide. Pyrazinamide was originally thought to act, like isoniazid, inhibiting fatty acid synthesis, but at an earlier enzymatic step of FAS1 rather than FAS2. However, this has been questioned due to its lack of effect at inhibiting bacterial growth. Pyrazinamide is important in the treatment of TB. Its use in the first 2 months of therapy reduced the total treatment length with the Tritab from 12 to 6 months. More recent studies suggest pyrazinamide may kill tubercule bacteria which are not affected by other drugs, by a mechanism on the ribosome rather than the cell wall. This may require pyrazinamide to be re-classified under the group of antibiotic drugs which inhibit protein synthesis rather than inhibiting the cell wall or DNA.

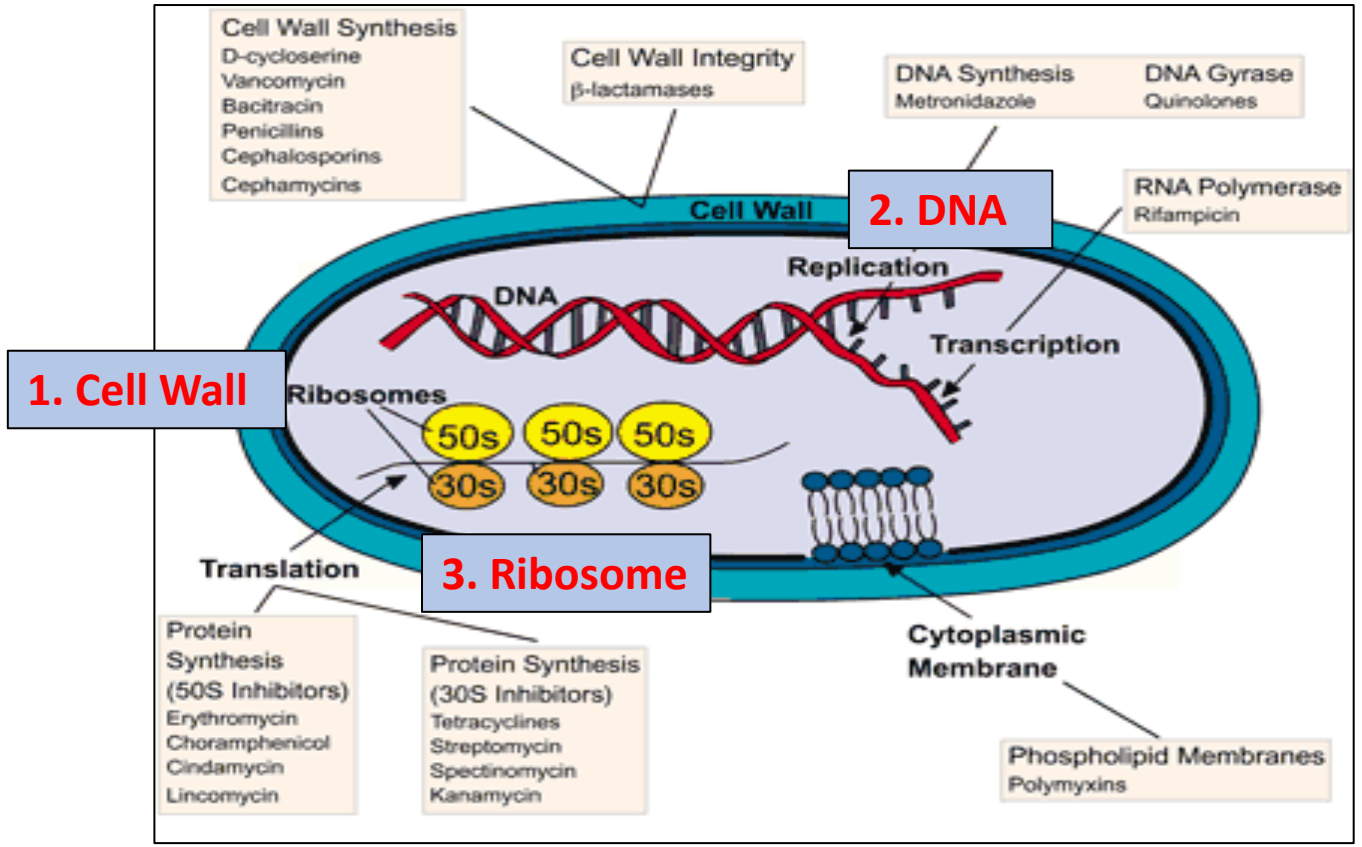
Antibiotics 2 : **DNA**

Important Antibiotics DNA

Trimethoprim
Nitrofurantoin
Metronidazole
Levofloxacin

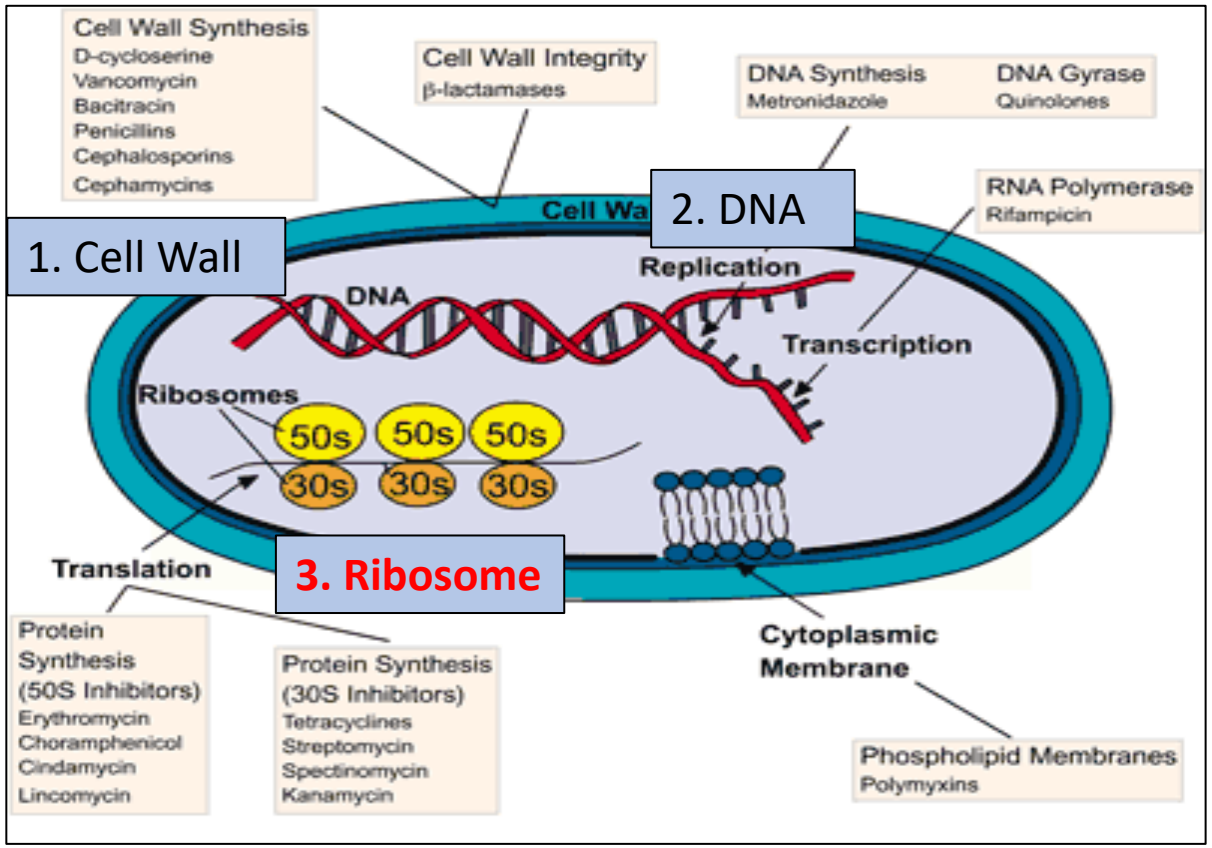
Mycobacteria

Rifampicin
Isoniazid
Ethambutol
Pyrazinamide



Important antibiotics to remember which interfere with bacterial DNA include trimethoprim, nitrofurantoin, metronidazole, levofloxacin. The consideration of pyrazinamide also links this bitesize session to the next which considers antibiotics which inhibit ribosomal protein synthesis.

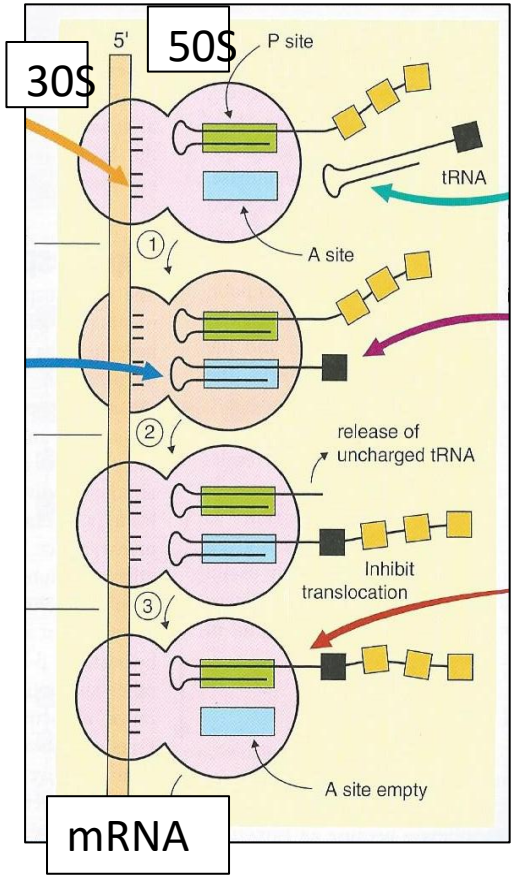
Antibiotics 2 : **Ribosome plus**



1. Aminoacyl-tRNA delivery

2. Transpeptidation

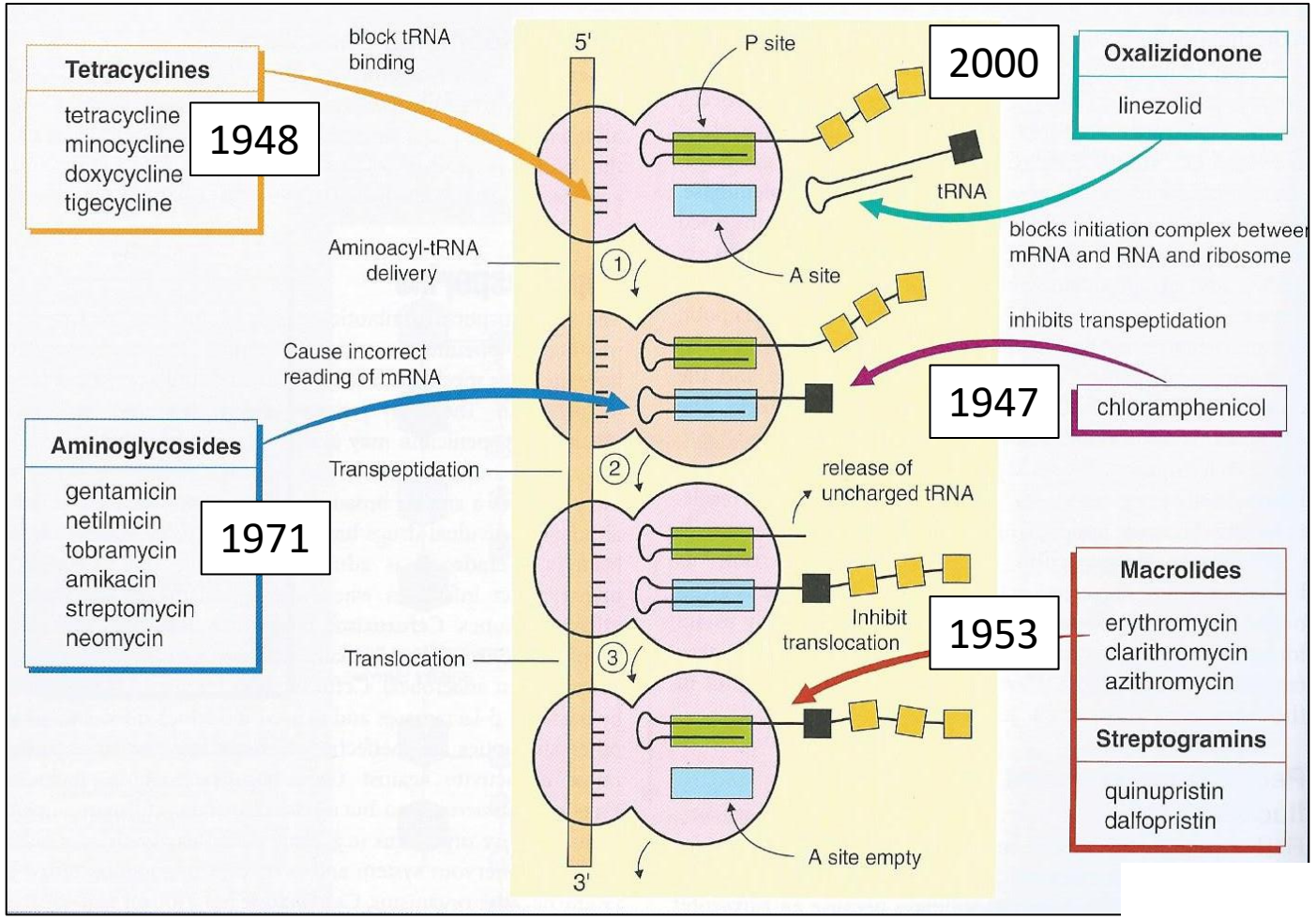
3. Translocation



Dr J Haylor, Medicine 2025

The third major target for antibiotics is the ribosome. The ribosome (shown in pink) contains two functional parts, a smaller 30S portion which reads the mRNA signal (codons on the buff-coloured bar) and a larger 50S portion which controls amino acid addition. Protein synthesis in the ribosome occurs in 3 steps to elongate the nascent amino acid chain (shown in yellow). The 50S portion has two tRNA binding sites, the A site (shown in blue) is for new amino acid addition (shown in black). The aminoacyl tRNA containing the next amino acid for addition to the A site is shown at the top of the slide. The second step of transpeptidation includes the removal of the nascent chain from the P site (shown in green) and its addition to the new amino acid on the A site. The third step is the release of the uncharged tRNA from the P site to allow the movement of the tRNA containing the extended amino acid chain to the P site, freeing up the A site for the next aminoacyl tRNA to be delivered.

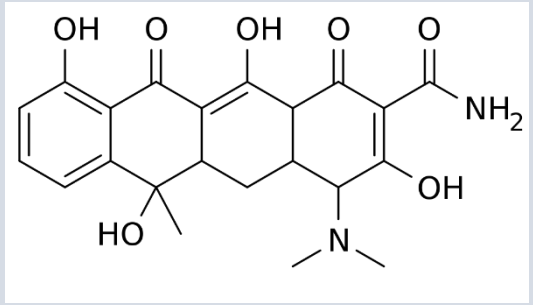
Antibacterial drugs which inhibit protein synthesis



Antibiotics influence the ribosome to inhibit protein synthesis at 5 different sites. The tetracyclines, macrolides and chloramphenicol were introduced in the early 1950's with more effective analogues being introduced in subsequent years. Although streptomycin was discovered in 1946, the major aminoglycosides weren't introduced until 1971 with gentamicin. The last new antibiotic mechanism to be introduced was suggested to be in 2000, some 20 years ago with linezolid.

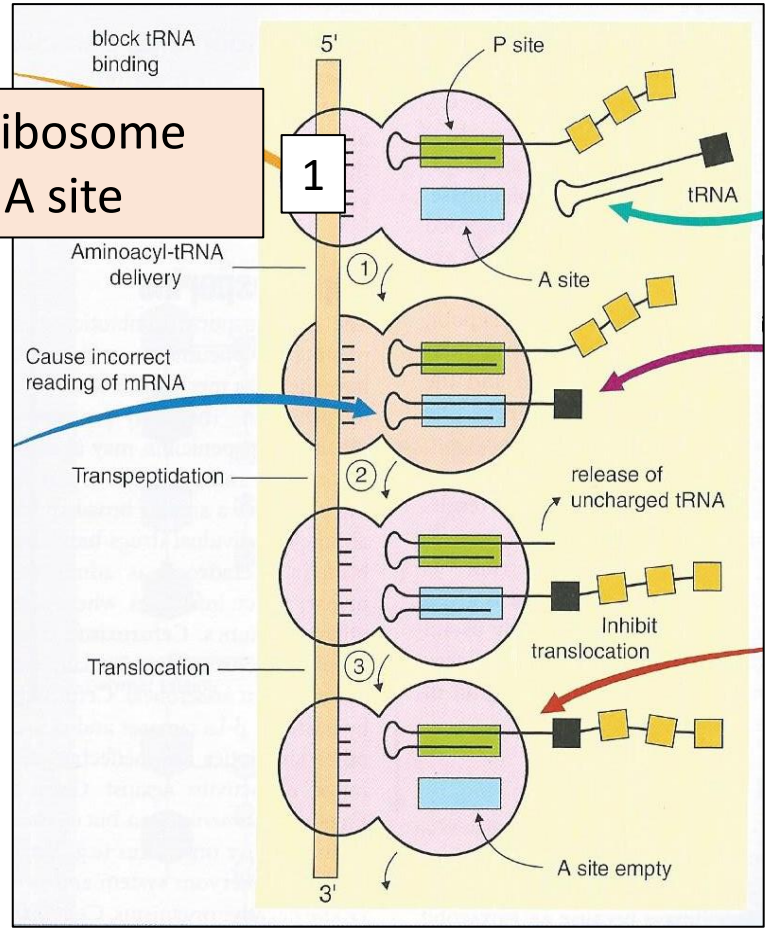
Ribosome 1. Tetracyclines

Tetracyclines 1948



- Origin - soil bacteria Streptomyces
- Name derived from chemical structure
- Bacteriostatic, broad spectrum (oral)
- Early agents - chlortetracycline, oxytetracycline (Lederele/Pfizer)
- Tetracycline – first semi-synthetic antibiotic

Binds to 30s segment of ribosome prevents tRNA binding at A site



The tetracyclines, introduced in 1948, were like many antibiotics originally derived from soil bacteria. The name tetracycline is derived from their chemical structure which contains 4 linked 6 membered rings. Chemical terms are frequently used for antibiotic drug groups which is unhelpful when discussing their mechanisms of action and therapeutic uses. Tetracyclines inhibit protein synthesis by binding to the 30S segment of the ribosome, preventing aminoacyl tRNA (shown in black) from binding to the A site on the 50S segment. Tetracyclines are oral active broad-spectrum antibiotics with a bacteriostatic effect.

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Doxycycline

Common Indications

- Acne vulgaris
- Lower respiratory tract, including exacerbations of COPD and pneumonia
- Chlamydia infection – pelvic inflammatory disease
- Malaria/anthrax/Lyme Disease

Resistance

efflux pumps prevent cellular accumulation

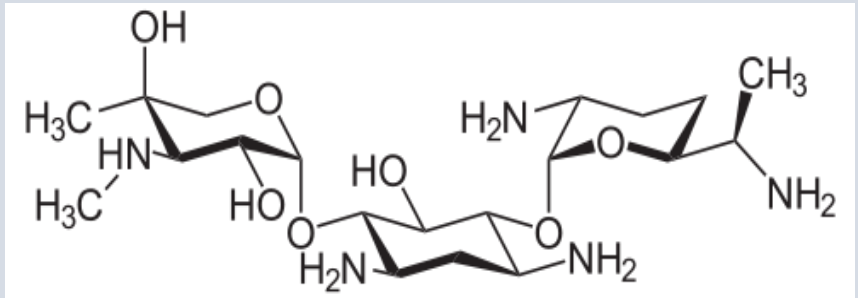
Adverse Effects

- Nausea, vomiting, diarrhoea – take with food (not dairy)
- Least likely antibiotic to cause C.Difficile colitis
- Oesphageal irritation/ulceration
- Photosensitivity (exaggerated sunburn)
- Avoid in pregnancy/breastfeeding binds to teeth and bones
- Children under 5, teeth discolouration

Lymecycline (1963) – highly water soluble absorbed by active carbohydrate mechanisms allows lower dose
Doxycycline (1967) – absorption less affected by ion complexes and food, less nephrotoxic than tetracycline

Tetracyclines are oral broad-spectrum antibiotics useful in treating lower respiratory tract infection, acne, chlamydia and malaria with probably the least effect, of all broad-spectrum antibiotics, on the GI tract biome and less likely to cause diarrhoea or lead to C. Difficile colitis. The most well described adverse effect of tetracyclines is their ability to chelate metals including chelating calcium. This means that tetracyclines should be avoided in pregnancy and in children under 5 where bone development may be delayed and teeth discoloured. The commonest tetracycline in current use is doxycycline. Compared to the older tetracycline compounds, doxycycline has a reduced ability to chelate calcium while lymecycline has improved absorption from the GI tract. Resistance to tetracyclines is due to the presence of efflux pumps removing the drug from the bacterial cell.

Ribosome **2. Aminoglycosides**

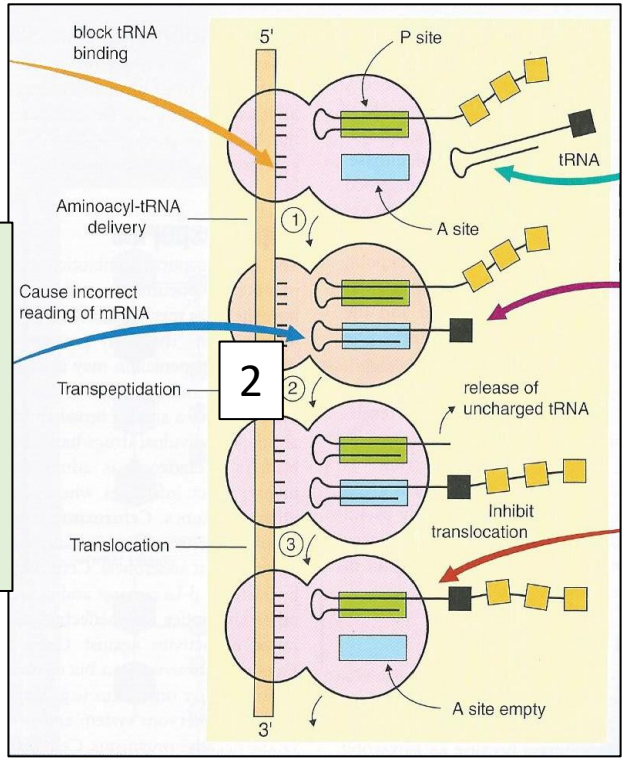


Gentamicin

- Bactericidal – water soluble enter cells via O2-dependent pathway (absent in anaerobic bacteria/streptococci)
- IV/IM route, narrow therapeutic index.
- Synthesised from *Micromonospora Purpurea*
- Mixture of at least 4 compounds

* First aminoglycoside streptomycin 1944

Binds irreversibly to 30s, misreading of codon giving abnormal or short protein



The aminoglycosides, first introduced with streptomycin in 1944 followed by gentamicin in 1971, were originally obtained from a soil bacteria *micromonospora purpurea*. Their aminoglycoside name is also derived from their chemical structure which, as shown for gentamicin, contains three linked sugars with amino groups attached. The aminoglycosides bind irreversibly to the 30S portion of the ribosome but, as a result the mRNA codon messages are misread generating short or abnormal non-functional proteins. In addition, gentamicin only enters cells via an oxygen-dependent pathway resulting in a lack of effect on anaerobic organisms. In comparison to the tetracyclinesg. Gentamicin is given by injection for more severe infections and has a narrow therapeutic index with serious adverse effects.

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Gentamicin

Common Indications

- Severe sepsis,
- Complicated urinary tract infection
- Intra-abdominal sepsis
- Endocarditis

Works well with penicillins which weaken cell wall to enhance aminoglycoside uptake

Ineffective in anaerobic bacteria

Adverse Effects

Concentrated in ear and kidney

Ototoxicity (1/4) : Killing of inner ear auditory hair cells, loss of high frequency sound leading to permanent deafness, vestibular hair cells (balance) may recover. Seen after treatment complete.

Nephrotoxicity (1/4) : podocyte effacement. Water soluble renal excretion. Elevated blood levels.

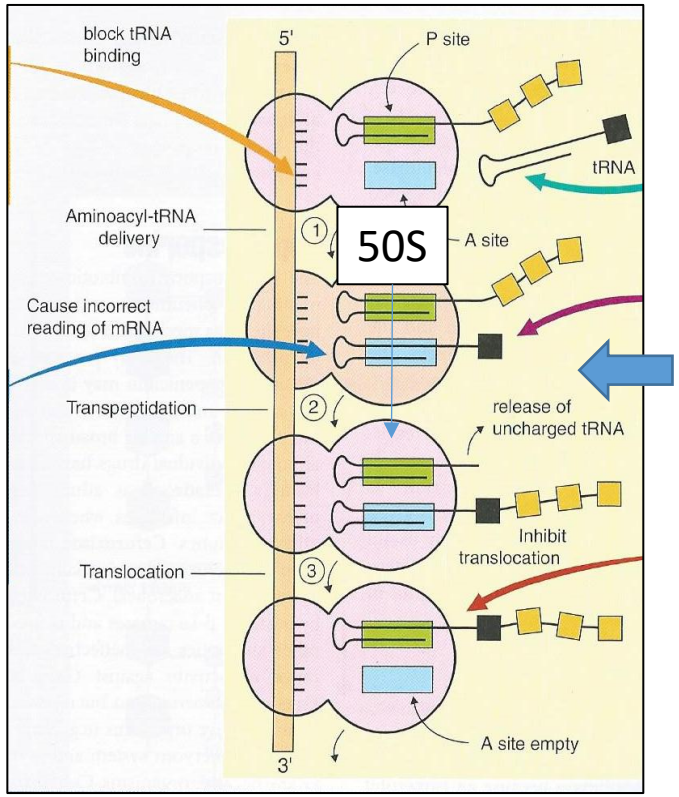
Also impairment of neuromuscular transmission.

IMPORTANT : Therapeutic drug monitoring required. Dose to ideal body weight for height

Tobramycin (1975) – gram –ve bacteria esp pseudomonas. IV but also by inhalation in cystic fibrosis
Amikacin (1976) – reduced development of resistance unlike gentamicin/tobramycin not a substrate for some 9 bacterial enzymes which inactivate aminoglycosides

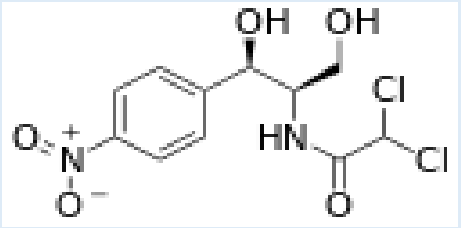
Gentamicin is reserved to treat severe infections including sepsis, and infections of the heart and urinary tract. Its works well in combination with penicillins which, by weakening the cell wall, enhance its cellular uptake. Gentamicin has a narrow therapeutic index which requires monitoring gentamicin blood levels to avoid toxicity. Gentamicin can induce a high incidence (up to 1 in 4) of ototoxicity and nephrotoxicity. Damage to auditory hair cells may lead to a permanent loss of hearing although effects on the loss of balance control tends to be reversible. Kidney damage results from a loss of podocytes damaging the glomerular filtration barrier. Other aminoglycoside analogues include tobramycin, which has a broader antibacterial spectrum, including the inhibition of *Pseudomonas A.* infection and can be given to cystic fibrosis patients by inhalation. Amikacin is less susceptible to degradation by bacterial enzymes, and thereby less likely to acquire resistance.

Ribosome **3. Chloramphenicol**



Inhibition of transpeptidation by interfering with substrate binding to enzyme

Chloramphenicol (1947)

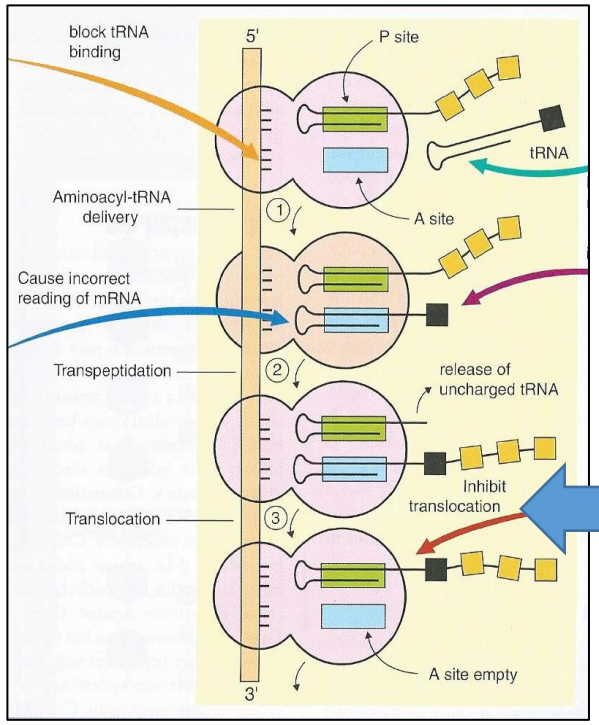


Binds to 50s portion of ribosome
Inhibits formation of peptide bond (peptidyl transferase)

- Streptomyces origin – broad spectrum (not Pseudomonas)
- Eye Ointment
- Lipid soluble – CNS penetration
- Bone marrow suppression (panocytopenia)
- Inhibits cytochrome CYP3A4

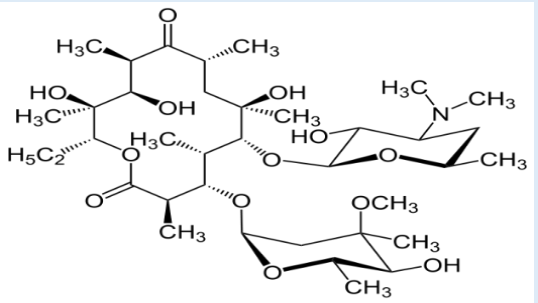
Chloramphenicol, also originally derived from a *Streptomyces* bacteria was the first antibiotic to be totally chemically synthesised and inhibits transpeptidation. Chloramphenicol binds to the 50S segment of the ribosome, inhibiting the activity of peptidyl transferase enzymes by interfering with substrate binding, responsible for the transfer of the nascent amino acid chain to the new amino acid substituent. Chloramphenicol is lipid soluble enough to cross the blood brain barrier, but when given systemically can induce bone marrow depression. Chloramphenicol is perhaps most widely used today in its formulation as an eye ointment.

Ribosome **4. Macrolides**



Inhibits translocation
Prevents transfer of tRNA containing amino acid sequence from A to P site

Erythromycin
(1952)



Chemically called macrolide due to large size of molecule. Contains 4 compounds (A-D) isolated from Philipino soil sample *Streptomyces Erythreus* (Ell Lilley)

- Uptake by phagocytes – delivered to infection site
- Acid unstable – enteric coated/stable esters
- CYP3A4 inhibitor

The macrolides, again soil bacteria derived, are also named by elements of their chemical structure which contains a large ring structure with attached sugar residues. The macrolides can be taken up by phagocytes and thereby delivered to a site of infection. Erythromycin introduced in 1952 inhibits translocation by binding to the 50S portion of the ribosome, preventing the tRNA containing the modifed amino acid chain moving from the A site to the P site, thus stopping its elongation.

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Clarithromycin

Common Indications

- Bacteriostatic
- Broad spectrum gram +ve and some gram –ve, (similar to penicillins)
- Respiratory and skin/soft tissue infections (alternative in penicillin allergy)
- H.Pylori triple therapy for GI ulcers
- Severe pneumonia (Legionaires Disease)

Adverse Effects

- Oral irritant (thrombophlebitis IV)
- Hepatic elimination – caution in liver impairment
- Released during phagocytosis
- Antibiotic associated colitis
- Prolongation of QT interval (arrhythmia)
- Inhibits CYP enzymes – increases levels of drugs metabolised by liver

Synthetic Macrolide Analogues

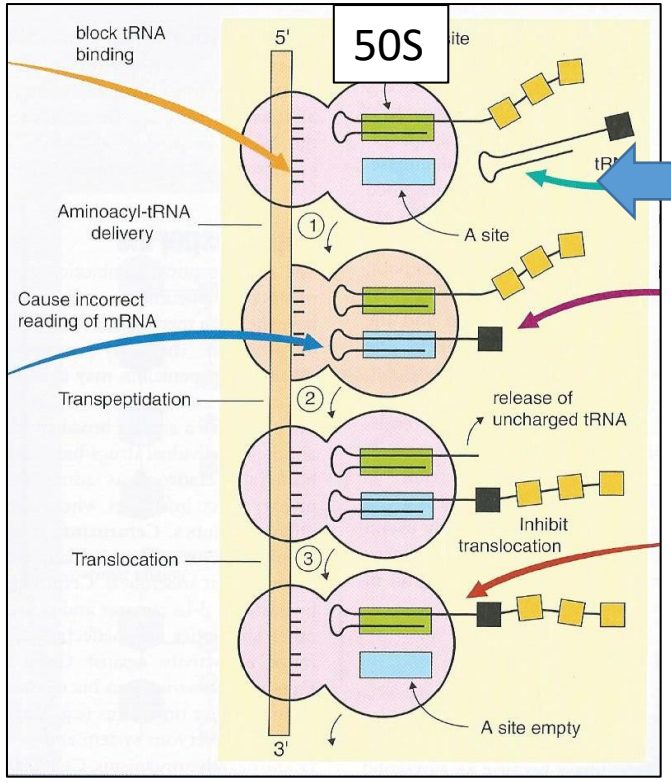
Clarithromycin (1980) – Japanese discovery (Abbott). Commonest UK oral macrolide, acid stable good absorption - reduced GI side effects. Greater gram –ve effects (H.Influenzae)

Azithromycin (1980) – Croatian discovery (Pfizer) – similar but does not inhibit CYP

* **Clindamycin** – not a macrolide, but similar mechanism - inhibits translocation.
Broad spectrum antibiotic, however can produce C. Difficile colitis, used for severe anaerobic and topical infections.

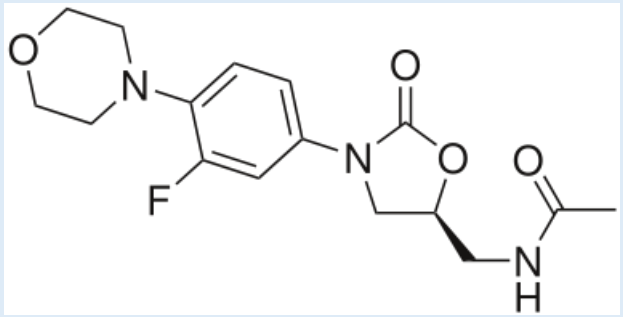
Erythromycin, the first macrolide antibiotic, has a bacteriostatic action but with a broad spectrum against both gram-positive and gram-negative bacteria similar to that of amoxicillin and therefore can be a useful alternative in penicillin allergy. Its effective in both soft tissue and respiratory infections. The commonest oral macrolide prescribed in the UK is clarithromycin which is acid stable with good oral absorption and enhanced activity against gram –ve bacteria including H. Influenzae. Clarithromycin, like erythromycin, has important adverse effects of prolonging the QT interval in the heart predisposing to arrhythmia. It is also an important inhibitor of cytochrome enzymes in the liver, elevating the plasma concentration of other drugs subject to hepatic metabolism.. A further analogue, azithromycin has a similar antibacterial spectrum but, does not inhibit drug metabolising enzyme activity in the liver.

Ribosome **5. Linezolid**



Binds to 50s A site preventing tRNA from initiating translation. Inhibits the initiation NOT the elongation of protein synthesis

Linezolid (2000)



LAST NEW ANTIBIOTIC

- chemical synthesis (not a bacterial product)
- Developed from monoamine oxidase inhibitors (MAOI) used to treat depression

Linezolid is said to be the last antibiotic to be introduced with a new molecular mechanism of action. Its origin however is very different from any other antibiotic being derived from drug programs to develop a treatment for depression by inhibiting the enzyme monoamine oxidase to enhance monoamine transmitter levels in the brain. Antimicrobial activity was initially used to control infection on tomatoes. Further analogues showed antibacterial activity against gram +ve bacteria with Upjohn eventually producing linezolid with good oral bioavailability. As with the tetracyclines, linezolid prevents the binding of aminoacyl t-RNA to the A site but inhibits the initiation of transcription rather than elongation of the nascent amino acid chain. However, linezolid binds to the 50S portion (not the 30S portion) of the ribosome.

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Linezolid

Indications

- Gram + ve bacteria including Streptococci (skin/pneumonia)
- Gram +ve resistant bacteria including MRSA and vancomycin resistant organisms

Kinetics

- Well absorbed, 100% bioavailable
- No dose adjustment IV to oral
- No interaction with liver CYP

Adverse Effects

1. Blood disorders – weekly blood/platelet count
2. Peripheral neuropathy, optic neuropathy, lactic acidosis (28 days or longer)
3. MAO inhibitor – interaction with MAO **uptake** inhibitors

* No cross resistance with other protein synthesis inhibitors

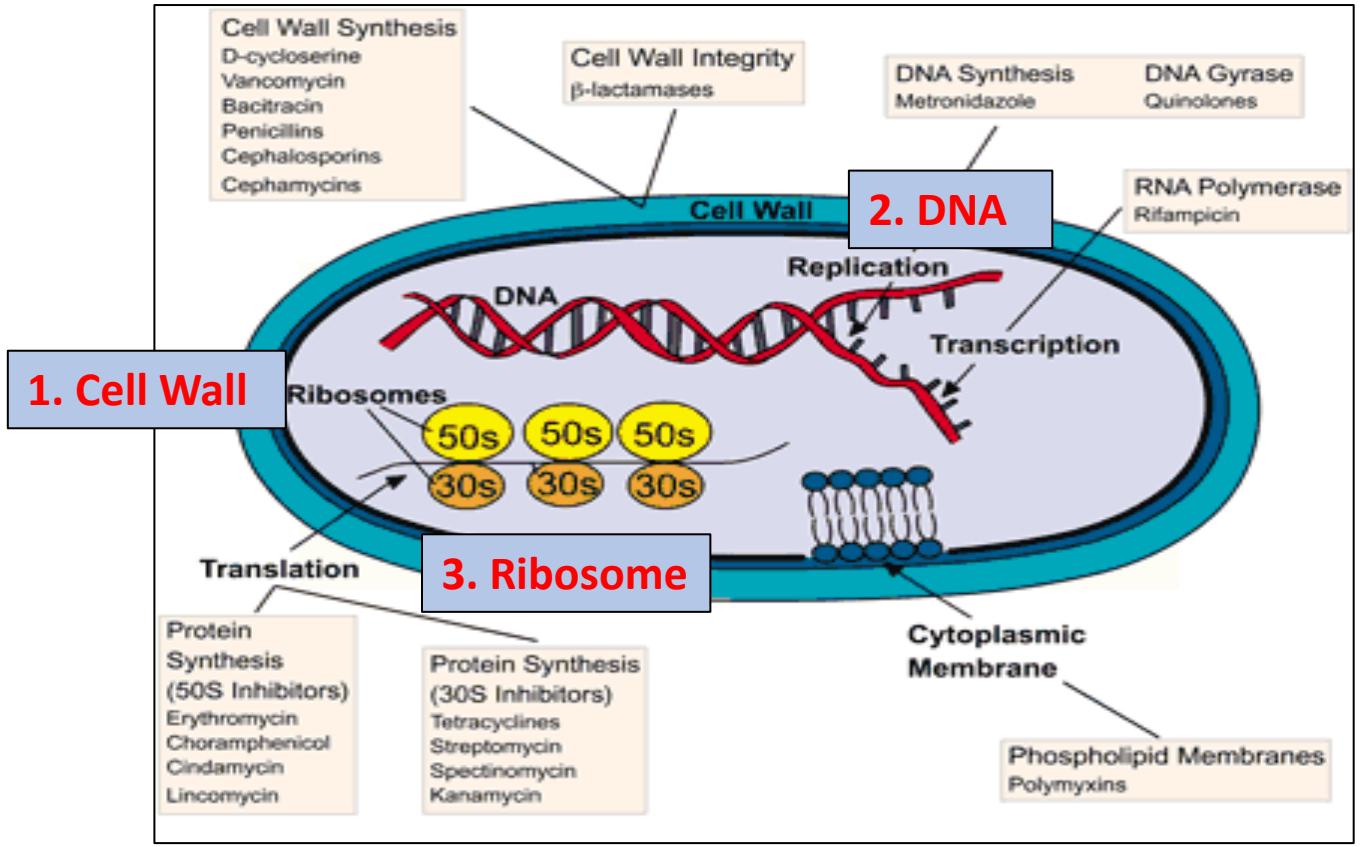
Linezolid should be reserved as an alternative agent for the treatment of multiple drug resistant strains.
It should not be used when other agents are likely to be effective even though it may have the indication.
(ie community acquired pneumonia)
Indiscriminate use/overuse will hasten the selection of resistant organisms and eventual loss of this valuable agent

Linezolid has a broad spectrum of activity including effects on MRSA and vancomycin resistant organisms. Its adverse effects include blood disorders, peripheral neuropathy and lactic acidosis. It also retains elements of monoamine uptake inhibitory activity. However, the major issue with linezolid is that even though it may be effective, it is important to reserve the use of linezolid for infections which are resistant to the effects of other antibiotics. Indiscriminate or overuse will hasten the selection of resistant organisms and eventual loss of this valuable agent.

Antibiotics 2 : **Ribosome plus**

Important Antibiotics and the Ribosome

Doxycycline
Gentamicin
Chloramphenicol
Clarithromycin
Clindamycin
Linezolid



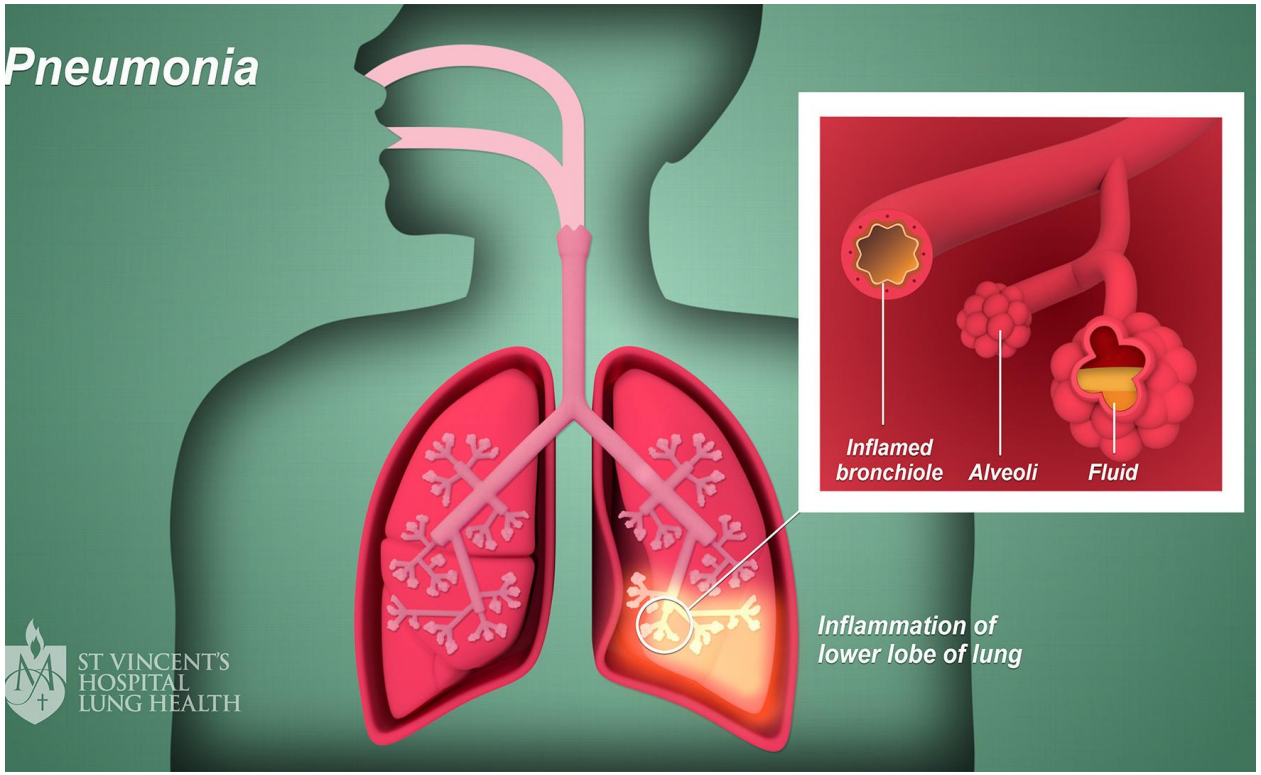
Important antibiotics to remember which interfere with protein synthesis in the bacterial ribosome include doxycycline, gentamicin, chloramphenicol, clarithromycin, clindamycin and linezolid.

Antibiotic Therapy
Five Common Bacterial Infections in Primary Care

1. Community acquired pneumonia
2. Otitis media
3. Acute Sore Throat
4. Cellulitis
5. Urinary tract infection

The following 5 slides contain information from the current NICE guidelines for the use of antibiotics in the treatment of 5 common bacterial infections treated in primary care including community-acquired pneumonia, otitis meadia, acute sore throat, cellulitis and urinary tract infections.

1. Pneumonia : Community Acquired (NG138)



Low Severity :

Amoxicillin 500mg, 3 times a day

(Pen Allergy – use Doxycycline or Clarithromycin)

Moderate Severity :

Amoxicillin 500mg, 3 times a day

(+ Clarithromycin if atypical pathogen suspected)

High Severity :

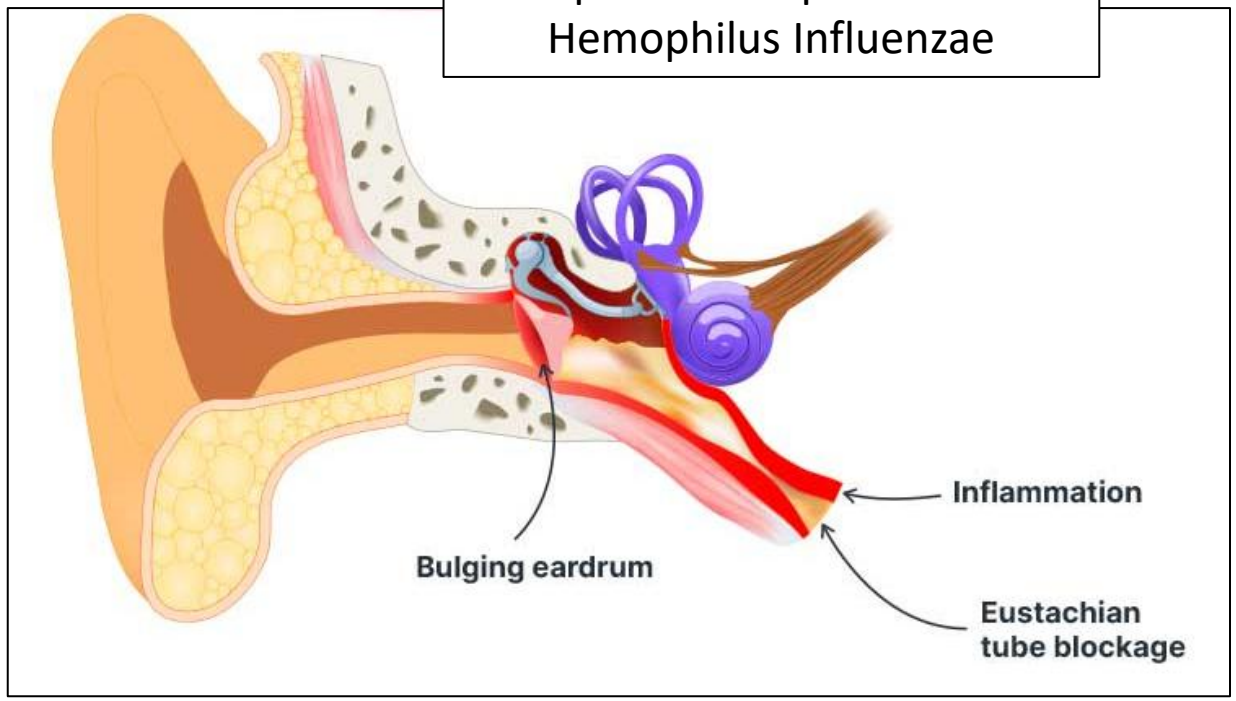
Co-amoxiclav + Clarithromycin

(alternative levofloxacin)

Streptoccoccus pneumoniae is the commonest bacterial cause of community-acquired pneumonia, fluid accumulation due to inflammation in the lower lobe of the lung. In conditions of low/moderate severity, first line treatment is the oral broad-spectrum, amoxicillin. For penicillin allergy, the other main oral broad spectrum antibiotics doxycycline or clarithromycin are available. If atypical pathogens such *as Chlamydophila pneumoniae or Mycoplasma pneumoniae* are suspected, clarithromycin is the preferred option. In conditions of high severity, broader cover is required using a combination of co-amoxiclav and clarithromycin.

2. Otitis Media (NG 91)

Streptoccoccus pneumoniae
Hemophilus Influenzae



Common in children 5-15 years

Phenazone/lidocaine ear drops

First line

Oral **amoxicillin**

Penicillin allergy – clarithromycin/doxycycline (erythromycin in pregnancy)

Second line

Co-amoxiclav

Otitis media is an inflammation of the middle ear causing the build-up of fluid behind the eardrum which is commonest in children 5-15 years old. Bacterial causes include *Streptoccoccus pneumoniae* and *Hemophilus influenzae*. Analgesic/anaesthetic ear drops may be useful prior to antibiotic therapy if the eardrum is intact.(phenazone/lidocaine). The first-choice antibiotic is oral amoxicillin, with clarithromycin in penicillin allergy and erythromycin in pregnancy. Co-amoxiclav would be the most appropriate second line choice.

3. Acute Sore Throat (NG 84)

Phenoxymethylpenicillin (Pen V)

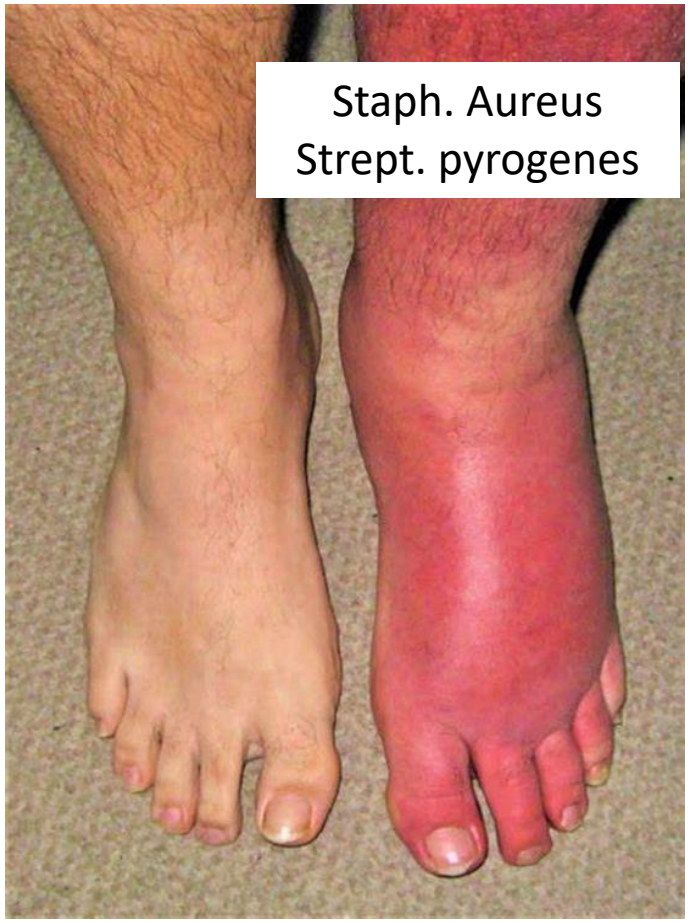
Acid stable, orally active but absorption unpredictable

Narrow spectrum - Equally effective for gram +ve (less so for gram -ve)

Antibiotic [1]	Dosage and course length for adults aged 18 and over
First choice	
Phenoxymethylpenicillin	500 mg four times a day or 1000 mg twice a day for 5 to 10 days Five days of phenoxymethylpenicillin may be enough for symptomatic cure, but a 10-day course may increase the chance of microbiological cure
Alternative first choice for penicillin allergy or intolerance (for people who are not pregnant)	
Clarithromycin	250 mg to 500 mg twice a day for 5 days
Alternative first choice for penicillin allergy in pregnancy	
Erythromycin	250 mg to 500 mg four times a day, or 500 mg to 1000 mg twice a day for 5 days Erythromycin is preferred if a macrolide is needed in pregnancy, for example, if there is true penicillin allergy and the benefits of antibiotic treatment outweigh the harms. See the Medicines and Healthcare products Regulatory Agency (MHRA) Public Assessment Report on the safety of macrolide antibiotics in pregnancy

Oral phenoxymethylpenicillin (Pen V) for 5-10 days is first line treatment for Strep. throat (NG84). Strep. throat is an acute sore throat caused by group A *Streptoccocci.* Symptoms include an enlarged soft palate and uvula, enlarged tonsils with white patches and red haemorrhages on the soft palate. Phenoxymethylpencillin has a narrow spectrum of action, compared to amoxicillin with reduced sensitivity to gram-negative bacteria. This results in a lower potential to inhibit the gut microbiome and thereby reduced adverse effects on the GI tract compared to the broad-spectrum amoxicillin. Clarithromycin is a suitable alternative in penicillin allergy with erythromycin being the preferred macrolide in pregnancy.

4. Cellulitis (NG 141)



Flucloxacillin

narrow spectrum penicillinase resistant

Penicillin Allergy
Clarithromycin/doxycycline

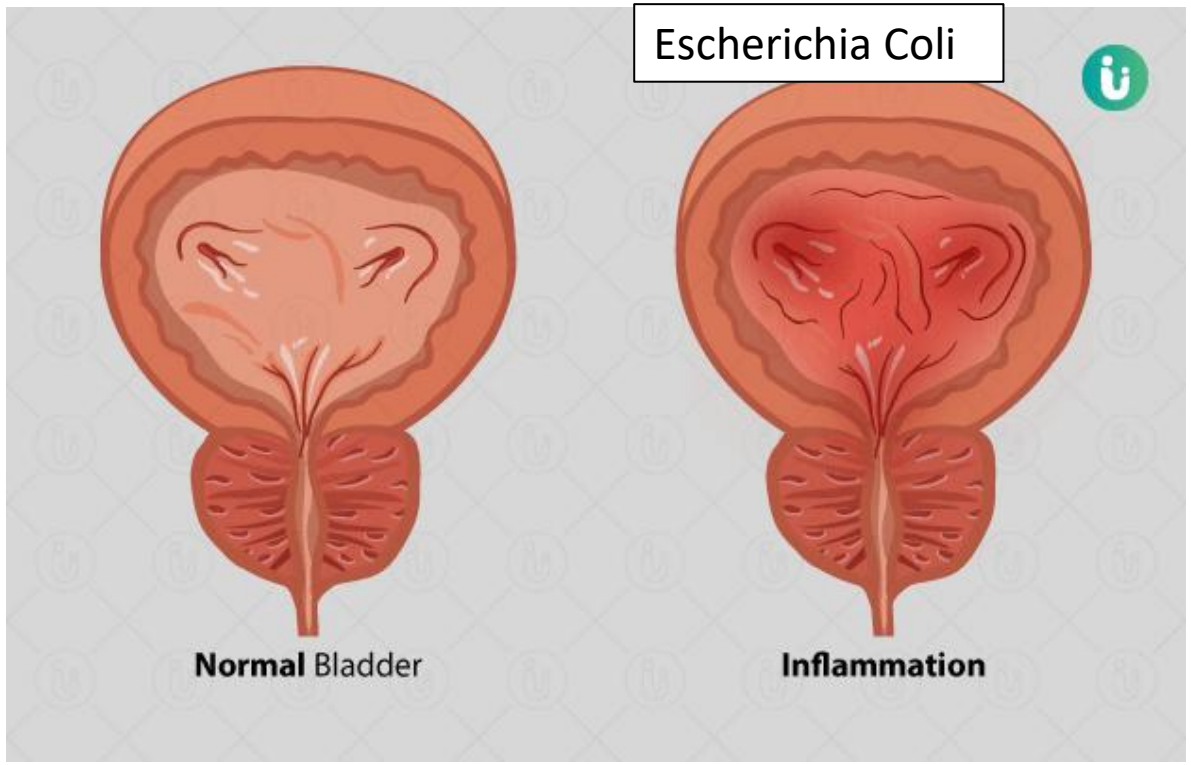
Facial cellulitis – co-amoxiclav

MRSA

IV vancomycin

Cellulitis is an inflammatory condition associated with redness, pain, swelling and warming of the skin caused by a bacterial infection in the deeper skin layers (left leg in diagram). Causative organisms are *Staph. Aureus*, a penicillinase-producing bacteria, and *Streptoccoccius pyrogenes*. Flucloxacillin, a narrow-spectrum penicillinase-resistant penicillin, is the first line treatment. Clarithromycin or doxycycline are available in penicillin allergy with erythromycin preferred during pregnancy. Co-amoxiclav is an alternative if the infection is near the eyes/nose where a broader spectrum antibiotic is required. *Methicillin-resistant Staph Aureus* (MRSA) is treated with IV vancomycin. Combination therapies with the narrow-spectrum flucloxacillin, to treat *Staph Aureus* and PenV/amoxicillin to treat *Streptoccocci* are a possible option.

5. Urinary Tract Infection (NG 109)



First Line

Nitrofurantoin (>45ml/min)

OR trimethoprim

Second Line

Nitrofurantoin (>45ml/min)

AND trimethoprim

Alternative

Pivmecillinam or fosfomycin

Lower urinary tract infections are most commonly caused by *Escherichia Coli* possibly originating from the GI tract. Symptoms of cystitis result from inflammation of the bladder and include pain or burning on urination, frequent urination or lower abdominal or back pain. *E. Coli* Is a gram-negative organism and first-line treatment is for 3 days with either nitrofurantoin (which causes free radical damage) or trimethoprim (which inhibits DNA synthesis). Nitrofurantoin is restricted to patients with an eGFR of > 45mL/min. Reduced renal function may elevate plasma concentrations increasing the risk of adverse effects on the GI tract and reduce renal tubular secretion and bladder concentrations resulting in treatment failure. Second line treatment uses both trimethoprim and nitrofurantoin in combination with pivmecillinam or fosfomycin as alternatives. Pivmecillinam is a penicillin liver prodrug, more effective against gram-negative bacteria than amoxicillin. While fosfomycin inhibits the formation of the building blocks for the peptidoglycan layer of the cell wall from inside the cell in an earlier step than bacitracin and can be given as a single dose. Trimethoprim should be NOT taken during the 1st trimester of pregnancy while nitrofurantoin, considered safe to use in the 1st/2nd trimesters should be avoided in the 3rd trimester due to the possibility of neonatal haemolysis. Nitrofurantoin is less effective in men when there is prostate involvement.

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Some added resources

Active Learning in Pharmacology

ANTIBIOTICS

Selection games to aid learning

Play as many times as you like

(please let me know if you find this helpful)

download
'GP antibiotics'

From NHS Greater Glasgow and Clyde

Antibiotics are a large drug group with many mechanisms of action, used to treat a very wide variety of infectious diseases. There are two additional sources of information which may help you learn and understand this area of pharmacology. Firstly, active learning exercises have been formulated using articulate rise and added to this area of Blackboard to help with learning this important subject area, based on simple selection games. Secondly, there is a very good concise App entitled 'GP Antibiotics' to download onto your phone which would be particularly useful in your clinical years.