PHARMACOLOGY

بسم الله الرحمن الرحيم

FINAL – Lecture 11

Antifolate Antibiotics & Fluoroquinolones (Pt.1)

﴿ وَإِن تَتَوَلَّوْا يَسْتَبْدِلْ قَوْمًا غَيْرَكُمْ ثُمَّ لَا يَكُونُوا أَمْثَالَكُم ﴾

اللهم استعملنا ولا تستبدلنا

Written by:

Raya Al Weshah

Reviewed by:

Mays Aljundi

Quiz for previous lecture

CLICK HERE "QUIZ 10"

(اللهمَّ لا سهلَ إلا ما جعلتَه سهلًا، وأنت تجعلُ الحَزْنَ إذا شئتَ سهلًا)

Antifolate Antibiotics

Yacoub M. Irshaid, MD, PhD, ABCP

Department of Pharmacology

(إِنَّ الصَّلَاةَ كَانَتْ عَلَى الْمُؤْمِنِينَ كِتَابًا مَّوْقُوتًا)

لا تنسوا الصلاة والدعاء لأهلنا في غزة

بسم الله بدأنا وعلى الله توكلنا :)

Folic Acid in Bacteria

Bacteria synthesize **folic acid,** necessary for DNA synthesis, particularly purines and nucleic acids. To do this, bacteria use p-aminobenzoic acid (PABA) and dihydropteroate synthase (DHPS).

This enzyme's function is to incorporate PABA and dihydropteroic acid, a precursor of folic acid. Similarly, humans also utilize folic acid for DNA synthesis, but we don't synthesize folic acid de novo.

Instead, we obtain the needed folic acid from our diets, and then it undergoes reduction to become active. **Antifolate antibiotics**, like sulfonamides, inhibit DHPS in bacteria, thereby leading to less nucleic acid synthesis and less bacterial cell division, growth, and proliferation.

Since this process does not occur in human cells, our DNA synthesis remains unaffected by antifolate antibiotics.

(وجَزَاهُم بِمَا صَبَرُواْ جَنَّةً وَحَرِيرًا)

Sulfonamides

- **Similar chemically to *p*-aminobenzoic acid (PABA).**

PABA is the first step in the synthesis of folic acid. It is also the rate-limiting step. PABA is inhibited by sulfonamides, by the binding to DHPS (structurally similar), and preventing the binding of PABA. This inhibition disrupts folic acid synthesis in bacteria and impairs DNA synthesis, growth, and division.

Mechanism of Action:

- **They inhibit the enzyme dihydropteroate synthase which incorporate PABA into dihydropteroic acid and thus, folate production.**
- **This pathway is essential for production of purines and nucleic acid synthesis.**

Folic acid is the protonated form, and **folate** is its negatively-charged conjugate base.To counterbalance the negative charge, **folate** forms salts with positive ions.

(وَهُوَ مَعَكُمْ أَيْنَ مَا كُنْتُمْ)

Sulfonamides

Contain a very broad antimicrobial spectrum. However, they are used less frequently in clinical practice, due to the rapid development of bacterial resistance. Yet, when administered with other drugs, their effectiveness is enhanced.--> synergistic effect (1+1>2). Sulfonamides have many **adverse** effects.

Antibacterial Spectrum:

- **They inhibit both gram positive and gram-negative aerobic bacteria, Nocardia, *Chlamydia trachomatis*, and some protozoa.** Inhibit → bacteriostatic
- **Some enteric bacteria such as *E. coli, Klebsiella, Salmonella, Shigella* and *Enterobacter* are also inhibited.**
- **Active against, *Pneumocystis jiroveci*, *Toxoplasma*.**
- **Activity is poor against anaerobes.**

(وَمَنْ يُؤْمِنْ بِاللَّهِ يَهْدِ قَلْبَهُ)

Sulfonamides

Mechanism of Resistance:

- **Some bacteria lack dihydropteroate synthase and are not susceptible to sulfonamides.** These bacteria also obtain their folic acid supply from their environments.
- **Mutations that:** Acquired resistance

1. **Cause overproduction of PABA**

   PABA and sulfonamides are chemically similar, and they both bind to DHPS. Sulfonamide is a **competitive inhibitor,** therefore, increasing its [] will overcome the inhibition of sulfonamides.

2. **Cause production of an enzyme with low affinity to sulfonamides**
3. **Impair bacterial cell permeability to sulfonamides.**

   Drugs cannot act without reaching the site of action.

(ادْعُونِي أَسْتَجِبْ لَكُمْ)

Sulfonamides

Classification:

1. **Oral, absorbable agents: sulfamethoxazole and sulfadiazine.**

Absorbed from the GIT into the circulation and then distribute to target tissues. These are commonly used in combination with other antibiotics to treat UTIs caused by *E. coli*. **Sulfadiazine** can also be used to treat parasitic infections.

2. **Oral, nonabsorbable agents: Sulfasalazine.**

Sulfasalazine is a combination of two drugs: sulfonamide (antibiotic) and salicylic acid (anti-inflammatory). It's administered to patients suffering from inflammatory bowel disease (IBS). Since the drug targets the gut, its absorption its avoided (hence the nonabsorbable agent classification).

3. **Topical agents: Sulfacetamide and silver sulfadiazine.**

Sulfacetamide is administered as ointment or eyedrops to treat infections in the conjunctiva. **Silver sulfadiazine** is administered topically for patients with exposed burn wounds, which provide suitable environments for bacterial growth.

(وَنَحْنُ أَقْرَبُ إِلَيْهِ مِنْ حَبْلِ الْوَرِيدِ)

Sulfonamides

Very toxic drugs with major adverse effects.

Adverse Reactions:

Sensitive to sunlight.

1. **Exfoliative dermatitis, photosensitivity.**

An immune/allergic reaction. Patients who take sulfonamides are strictly advised to stay at home and avoid exposure to the sun due to UV rays which can cause inflammation in the skin. Patients are also advised to wear sunscreen especially on the exposed skin wounds. The initial skin reaction is due to exfoliative dermatitis; however, the second would be because of photosensitivity.

2. **Stevens-Johnson syndrome is a serious and potentially fatal type of skin and mucous membrane eruption.**

Mucous membranes are present in the nose, throat, mouth, bladder, gut, etc. Stevens-Johnson syndrome is like exfoliative dermatitis but for the mucous membranes as well as the skin. Peeling and blistering.

Sulfonamides Very toxic drugs with major adverse effects.

Adverse Reactions:

3. Stomatitis, conjunctivitis, arthritis, hepatitis, polyarteritis nodosa, allergic nephritis and psychosis.

Mucous membrane eruptions can occur without dermatitis. If these eruptions accompany dermatitis, then the patient is suffering from Stevens-Johnson syndrome. **Arthritis** is an inflammatory response in the joints. **Polyarteritis nodosa** is a very severe immune disease (inflammation in medium-sized arteries that causes organ damage). **Psychosis** is a condition where the patient suffers hallucinations, delusions, and overall loss of sense of reality. **Allergic nephritis** can also be known as **interstitial nephritis** which refers to inflammation of kidney interstitial tissue. While **cholestatic hepatitis** refers to inflammation in the liver due to impaired bile flow.

4. Crystalluria

Formation of crystals in urine, because of the precipitation of these drugs in urine. Crystals can damage renal tubules.

(وَمَنْ يَتَوَكَّلْ عَلَى اللَّهِ فَهُوَ حَسْبُهُ)

Very difficult to treat; almost irreversible.

Skin inflammation

Exfoliative Dermatitis

(لا تَدْرِي لَعَلَّ اللَّهَ يُحْدِثُ بَعْدَ ذَلِكَ أَمْرًا)

Sulfonamides

5. Hemolysis in patients with G6PD deficiency.

G6PD is one of the few enzymes important for [NADPH] restoration in the body. NADPH is important for the maintenance of glutathione in its reduced from in RBCs, as well as other NADPH-dependent pathways in the body including steroid hormone biosynthesis (metabolism).

6. Aplastic anemia, granulocytopenia, thrombocytopenia

Aplastic refers to no cells in the bone marrow; no WBCs, RBCs, platelets, or any combination. Low [WBCs] cause the patient to become more susceptible to infections, low [platelets] cause difficulty stopping bleeding, while low [RBCs] cause anemia with its accompanying symptoms-- weakness, fatigue, etc.

7. Contraindicated in pregnancy (teratogenic).

Interferes with folic acid metabolism, which is very crucial for fetal development. Pregnant women are recommended to take folic acid throughout their pregnancy to eliminate the risk of congenital malformations as much as possible.

(وَلَسَوْفَ يُعْطِيكَ رَبُّكَ فَتَرْضَى)

Trimethoprim

- **It inhibits bacterial dihydrofolate reductase which converts dihydrofolic acid to tetrahydrofolic acid (the active form of folic acid) which is needed for synthesis of purines and DNA.**

Deficiency in the formation of tetrahydrofolic acid leads to deficiency in folic acid functions.

- **Pyrimethamine is similar but inhibit protozoal dihydrofolate reductase.**

Similar mechanism of action, works on protozoa instead of bacteria.

- **Both, pyrimethamine & trimethoprim, in combination with a sulfonamide block sequential steps in folate synthesis → synergism of activity of both drugs. The combination is bactericidal.** Trimethoprim and sulfonamide.

This combo is very commonly used. All the previously discussed sulfonamide adverse effects may occur with different combinations as well. **Cotrimoxazole** (trimethoprim + sulfonamide) are administered as a single pill.

(وَبَشِّرِ الصَّابِرِينَ)

Trimethoprim

Mechanisms of Resistance:

1. **Reduced cell permeability.**

   Reduced bacterial cell permeability impedes entry of trimethoprim.

2. **Overproduction of dihydrofolate reductase.**

   To overcome trimethoprim's mechanism of action.

3. **Altered dihydrofolate reductase with low binding to drug (most important clinically).**

   This alteration initiates in the DNA; mRNA → protein synthesis → protein modification → production of dihydrofolate reductase resistant to trimethoprim.

(إِنَّ مَعَ العُسْرِ يُسْراً)

Trimethoprim

- **Excreted in urine partially as metabolites. Dose should be reduced in renal failure.**

Can be used to treat UTIs since it is excreted in urine if the bacteria is **susceptible**.
Renal failure causes low drug excretion, and thus, a prolonging of its half-life in the body and causes dose-dependent adverse effects. Patients with renal failure and UTIs can not be administered trimethoprim to treat the UTIs due to the drug's inability to reach the site of action.

- **It concentrates in prostatic and vaginal fluids, which are more acidic than plasma.**

Used for treatment of prostatic and vaginal infections.

Trimethoprim

Antibacterial spectrum:

1. **E. coli (Acute UTI) either alone or in combination with sulfamethoxazole (Co-trimoxazole).**
2. **Salmonella**
3. **Shigella**
4. ***Pneumocystis jiroveci* (IV infusion).**

P. jiroveci is a fungus that causes pneumonia in immunosuppressed patients & is susceptible cotrimoxazole.

(وَلَكِنَّ الْبِرَّ مَنِ اتَّقَى)

Pyrimethamine

Pyrimethamine is antiparasitic.

1. **+ sulfadiazine Leishmania and Toxoplasma.**
2. **+ sulfadoxine active against malaria.**

(كُلُّ مَنْ عَلَيْهَا فَانٍ)

Trimethoprim

- **Adverse Effects:**

1. **Megaloplastic anemia, leukopenia and granulocytopenia.**

Remember, trimethoprim interferes with folic acid metabolism in the human body, but not as strongly nor specifically as with bacterial folic acid synthesis. It can cause partial inhibition of the human folic acid enzymes (DHFR), which can lead to folic acid deficiency in already borderline patients. **Leukopenia** is the reduction of leukocytes, which also require cell division (need for folic acid). **Granulocytopenia** is low granulocytes-- WBCs involved in the body's immune response (neutrophils, basophils, eosinophils).

(وَاذْكُرِ اسْمَ رَبِّكَ وَتَبَتَّلْ إِلَيْهِ تَبْتِيلًا)

Trimethoprim

- **Adverse Effects:**

2. Diarrhea

This is due to the broad antimicrobial spectrum of trimethoprim, which can also eradicate microbial flora. Microbial flora are responsible for the digestion of the food we intake. For example, cellulose is a polysaccharide that our bodies **cannot** metabolize (beta-linkages). Instead, microbial flora is responsible for its digestion, therefore, eliminated microbial flora leads to no digestion which can cause bloating. Diarrhea can also be a manifestation of superinfection, just like *C. dificile.*

(قَدْ أَفْلَحَ مَنْ تَزَكَّى وَذَكَرَ اسْمَ رَبِّهِ فَصَلَّى)

Trimethoprim

- **Adverse Effects:**

3. Hyperkalemia and hyponatremia.

Hyperkalemia affects potassium in renal tubules, causes cardiac dysrhythmia (hypokalemia can also cause this), as well as cardiac arrest. Lethal injections used for inmates on death row are high doses of potassium, which cause instant cardiac arrest and death. While trimethoprim itself doesn't cause cardiac arrest, prescribing an already hyperkalemic patient with trimethoprim increases hyperkalemia and thus, their chance of suffering cardiac arrest.

Fluoroquinolones

Yacoub M. Irshaid, MD, PhD, ABCP

Department of Pharmacology

(إِنَّ أَكْرَمَكُمْ عِنْدَ اللَّهِ أَتْقَاكُمْ إِنَّ اللَّهَ عَلِيمٌ خَبِيرٌ)

Fluoroquinolones

As the name suggests, fluoroquinolones contain fluorine, which is a halogen. Halogens, particularly fluorine, are very lipid soluble. The attachment of fluorine to a quinolone increases its lipophilicity and allows the resulting fluoroquinolone to cross through membranes and have wide tissue distribution. Fluoroquinolones can be used for UTIs.

Recall: what is the antifolate antibiotic that is overcome in bacteria by overproduction of PABA?

(ادْعُوا رَبَّكُمْ تَضَرُّعًا وَخُفْيَةً إِنَّهُ لَا يُحِبُّ الْمُعْتَدِينَ)

Fluoroquinolones

Mechanism of Action:

- **They block bacterial DNA synthesis by inhibiting bacterial topoisomerase II (DNA gyrase) and topoisomerase IV.**
- **Inhibition of DNA gyrase prevents the relaxation of positively supercoiled DNA that is required for transcription and replication.**

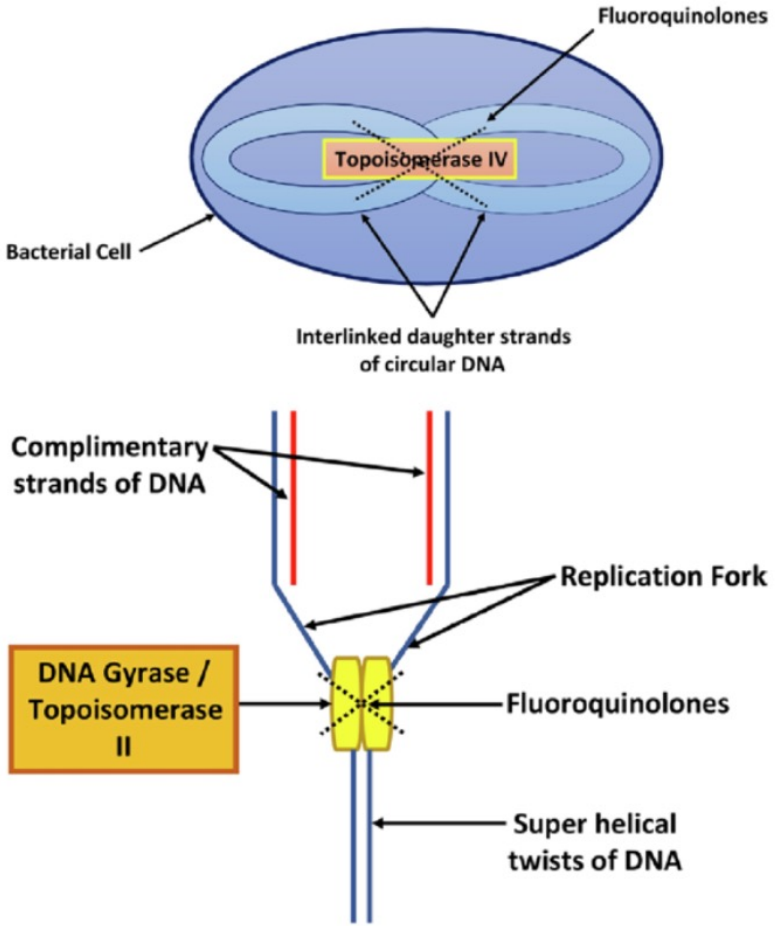
When the DNA is relaxed, promoters and required enzymes can bind to it and initiate both translation and transcription. Inhibition of bacterial topoisomerase II prevents the relaxation of stiffly supercoiled DNA necessary for DNA replication and transcription. Thus, the bacteria will not proliferate. In a similar fashion, mRNA translation also requires for the relaxation of supercoiled DNA indirectly..

(وَإِنِّي لَغَفَّارٌ لِّمَن تَابَ وَآمَنَ وَعَمِلَ صَالِحًا ثُمَّ اهْتَدَى)

Fluoroquinolones

- **Inhibition of topoisomerase IV interferes with separation of replicated chromosomal DNA into daughter cells during cell division.**

Topoisomerase IV is an enzyme that assists bacteria in the detangling of DNA following replication. This detangling in crucial to ensure proper chromosomal segregation into the two daughter cells. Inhibition of topoisomerase IV prevents the separation of DNA into daughter cells, due to DNA entanglement resulting in the formation of a single, large cell with duplicated DNA, instead of two daughter cells.



Not a required picture, used to give some context.

(وَٱصْبِرْ لِحُكْمِ رَبِّكَ فَإِنَّكَ بِأَعْيُنِنَا)

Fluoroquinolones

Mechanisms of Resistance:

1. **One or more point mutations in the quinolone binding region of the target enzyme. Confer high level resistance.**

Inhibit fluoroquinolone action. Again, no access to site of action, no therapeutic effect.

2. **Change in the permeability of bacterial cell.**
3. **Plasmid-mediated resistance:** Resistance gene obtained from the environment or bacteriophages in incorporated in the bacterial DNA.

a) **One protects DNA gyrase from flouroquinolones**

Related to the function of topoisomerase II

(ثُمَّ أَنْزَلَ اللَّهُ سَكِينَتَهُ عَلَى رَسُولِهِ وَعَلَى الْمُؤْمِنِينَ)

Fluoroquinolones

b) Another is a variant of aminoglycoside acetyltransferase which modifies ciprofloxacin.

The other plasmid-mediated mechanism of resistance is the enhanced formation of an enzyme, **acetyltransferase**, which acetylates or modifies the structure of fluoroquinolones and prevents their action.

Both (a & b) produce low level resistance but facilitate point mutations. Point mutations confer a high level of resistance.

- **Resistance to one of these drugs confer resistance to the others (cross-resistance).**

Resistance of ciprofloxacin would include resistance to all fluoroquinolones.

(قَالَ رَبِّ إِنِّي ظَلَمْتُ نَفْسِي فَاغْفِرْ لِي فَغَفَرَ لَهُ ۚ إِنَّهُ هُوَ الْغَفُورُ الرَّحِيمُ)

Fluoroquinolones

Antibacterial Spectrum:

1. **Ciprofloxacin, levofloxacin, and ofloxacin have excellent activity against gram negative aerobic bacteria (*Enterobacter sp, Pseudomonas aeruginosa, Neisseria meningitidis, Haemophilus sp, and Campylobacter jejuni*), and moderate to good activity against gram positive bacteria. They are active against staphylococci but not methicillin-resistant strains.**

Ciprofloxacin is the prototype drug. The treatment of *P. aeruginosa* can be tricky, and may require two antibiotics, depending on the severity of infection. ***N. meningitidis*** causes meningitis. ***Haemophilus*** causes otitis media and pneumonia. As per discussed previously, aminoglycosides and aztreonam, a beta-lactam, are both effective against gram negative bacteria.

(ادْعُوا رَبَّكُمْ تَضَرُّعًا وَخُفْيَةً إِنَّهُ لَا يُحِبُّ الْمُعْتَدِينَ)

Fluoroquinolones

- **Streptococci and enterococci are less susceptible than staphylococci.** But **not** against MRSA.
- **Ciprofloxacin is the most active against *Pseudomonas aeruginosa*.**

(وَمَنْ يَتَّقِ اللَّهَ يَجْعَلْ لَهُ مَخْرَجًا، وَيَرْزُقْهُ مِنْ حَيْثُ لَا يَحْتَسِب)

Fluoroquinolones

2. **Gemifloxacin and Moxifloxacin have improved activity against gram positive bacteria, particularly *Streptococcus pneumoniae* and some staphylococci.**

- **Levofloxacin has superior activity against *Streptococcus pneumoniae*.**

More effective than the first group (ciprofloxacin, lovafloxacin, ofloxacin) against gram positive bacteria. Therefore, a member of the second group (gemifloxacin, moxifloxacin, levofloxacin) is chosen as treatment when dealing with gram positive bacteria. These drugs are called **respiratory fluoroquinolones** and can be used to treat pneumonia if it were caused by a gram-positive infection, which is more commonly the case.

(اللَّهُ وَلِيُّ الَّذِينَ آمَنُوا يُخْرِجُهُمْ مِنَ الظُّلُمَاتِ إِلَى النُّورِ)

Fluoroquinolones

- **Fluoroquinolones are also active against agents of atypical pneumonia (*Mycoplasma* and *Chlamydia*) and against intracellular pathogens such as *Legionella* and *Mycobacteria*.**

Most of the time, community-acquired pneumonia is caused by gram + or atypical bacteria. **Mycobacteria** causes *M. tuberculosis*. Atypical bacteria are cell wall-lacking bacteria.

For any feedback, scan the code or click on it.

Corrections from previous versions:

Versions	Slide # and Place of Error	Before Correction	After Correction
V0 → V1			
V1 → V2			

Additional Resources:

رسالة من الفريق العلمي:

Extra References for the Reader to Use:

[Quinolone- Mechanism of Action]

إذا ضاقَ الزَمانُ عَلَيكَ فَاصبِر
وَلا تَيأَس مِنَ الفَرَجِ القَريبِ
وَطِب نَفساً بِما تَلِد اللَيالي
عَسى تَأتيكَ بِالوَلَدِ النَجيبِ

- علي بن أبي طالب -