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المضادات الحيوية (BCH 476)

Antibiotics

Lecture 19-20: Mechanism of action of antibiotics

Antibiotics affecting microbial protein synthesis

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Lecture No.	Topics
19-20	-Protein biosynthesis-inhibition by antibiotics a- Inhibitors of the initiation stage (e.g. streptomycin & tetracycline). b- Inhibitors of the elongation stage (e.g. chloramphenicol, cycloheximide & erythromycin). c- Inhibitors of the termination stage (e.g. puromycin).

Why antimicrobial drugs can inhibit protein synthesis in bacteria ribosomes without having a major effect on mammalian ribosome?

Because the ribosomes in both organisms are different with respect of:

1- Bacteria have 70S ribosomes, whereas mammalian cells have 80S ribosomes.

2- The subunits of each type of ribosome, their chemical composition, and their functional specificities are sufficiently different to explain.

In normal microbial protein synthesis, the mRNA message is simultaneously "read" by several ribosomes that are strung out along the mRNA strand. These are called polysomes.

Loading

Inhibitors of the initiation stage of protein synthesis (Aminoglycosides)

- Aminoglycosides are the most important candidate of the inhibitors of the initiation stage of protein synthesis.
- **Streptomycin**: The mode of action of streptomycin has been studied far more intensively than that of other aminoglycosides (Kanamycin, neomycin, gentamicin, tobramycin, amikancin, etc….), but probably all act similarly.

I- Inhibitors of the initiation stage of protein synthesis (Aminoglycosides)

The proposed mechanism of protein synthesis inhibition by aminoglycosides:

- The attachment of the aminoglycoside to a specific receptor protein (P 12 in the case of streptomycin) on the 30S subunit of the microbial ribosome.
- The aminoglycoside blocks the normal activity of the “initiation complex” of peptide formation (mRNA+formyl methionine + tRNA).
- The mRNA message is misread on the “recognition” of the ribosome; consequently, the wrong amino acid is inserted into the peptide, resulting in a nonfunctional protein.
- Aminoglycoside attachment results in the breakup of polysomes and their separation into monosomes incapable of protein synthesis.

Resistance against aminoglycosides

Three modes of resistance against aminoglycosides can be notices:

1- Chromosomal resistance, the lack of a specific protein receptor on the 30S subunit of the ribosome.

2- Plasmid-dependent resistance to aminoglycosides depends on the production of adenylating, phosphorylating, or acetylating enzymes by the micro-organism that destroy the drugs.

3- Resistance depends on "permeability defect" an outer membrane change that reduces active transport of the aminoglycoside into the cell so that the drug cannot reach the ribosome. Often this is "Plasmid-Mediated".

Tetracyclines

- Tetracyclines bind to the 30S subunit of microbial ribosomes.
- They inhibit protein synthesis by blocking the attachment of charged aminoacyl-tRNA, thus, they prevent introduction of new amino acids to the nascent peptide chain.
- The action is usually inhibitory and reversible upon withdrawal of the drug.

Resistance against tetracyclines

- Resistance to tetracyclines results from changes in permeability of the microbial cell envelope.
  - **In susceptible cells:** the drug is concentrated from the environment and does not readily leave the cell.
  - **In resistance cells:** the drug is not actively transported into the cell or leaves it so rapidly that inhibitory concentrations are not maintained.
- Mammalian cells do not actively concentrate tetracyclines.

II- Inhibitors of the elongation stage
Chloramphenicol

- Chloramphenicol binds to A2451 and A2452 residues in the 23S rRNA of the **ribosome** and inhibits peptide bond formation.
- Chloramphenicol binds to the 50S subunit of the ribosome.
- It interferes with the binding of new amino acids to the nascent peptide chain, largely because chloramphenicol inhibits peptidyl transferase.
- Chloramphenicol is mainly bacteriostatic and growth of micro-organisms resumes (ie, drug action is reversible) when the drug is withdrawn.

Resistance against chloramphenicol

- Micro-organisms resistant to chloramphenicol produce the enzyme chloramphenicol acetyltransferase, which destroys drug activity.
- The production of this enzyme is usually under control of a plasmid.

Macrolides, Azalides (Erythromycin, Azithromycin, Clarithromycin, Dirithromycin)

- These drugs bind to the 50S subunit of the ribosome, and the binding site is 23S rRNA.
- They may interfere with the formation of the initiation complexes for peptide chain synthesis or may interfere with aminoacyl translocation reactions.
- Some macrolide-resistant bacteria lack the proper receptor on the ribosome (through methylation of rRNA). This may be under plasmid or chromosomal control.

Lincomycins (Clindamycin)

- Clindamycin binds to the 50S subunit of the microbial ribosome and resembles macrolides in binding site, antibacterial activity, and more of action.
- Chromosomal mutants are resistant because they lack the proper binding site on the 50S subunit.

III- Inhibitors of the termination stage (e.g. puromycin).

- Puromycin has a structure similar to the tyrosinyl aminoacyl-tRNA.
- Thus, it binds to the ribosomal A site, transfer to the growing chain, causing premature chain release, and participates in peptide bond formation, producing peptidyl-puromycin.
- However, it does not engage in translocation and quickly dissociates from the ribosome causing a premature termination of polypeptide synthesis.
- So, it causes premature chain termination during translation taking place in the ribosome.