What is the importance of antibiotic production for the producing microorganism?
Recently, the following functions are proposed for the role of antibiotics in their producers:
1- Ecological role in nature
2- Role of antibiotics in regulation of metabolism
3- Role of antibiotics in regulation of differentiation and morphogenesis of the producing microorganisms.

Are antibiotics produced in nature or only in lab?
– Yes it is proved that antibiotics can be produced by micro-organisms in normal ecological conditions (out of lab conditions).

• Do antibiotics have a function when produced naturally?
– Some antibiotics may have a protective effect on the organism producing them and play an important role in the ecology of actinomycetes.
– Antibiotics are important for differentiation and maturation of microorganism.

• Is one function that of inhibiting or killing competing organism in nature?
– Yes some antibiotics producing microorganisms resist the deleterious effect of their antibiotics but in contrast, these antibiotics protects their producing microorganisms against the microbes or actinomycetes in vicinity.

• How do antibiotic-producing microorganism avoid suicide”:
- In some microorganisms, the sensitivity against antibiotic is high in the growth phase (trophophase) and decreases during antibiotic synthesis phase (idiophase).
- In other antibiotic producing microorganisms, this organisms lack the target structure for the antibiotic attack.
- Some other organisms protect itself by modifying the ribosome (target for antibiotic) by adding methyl group to one of its subunits (23S), hence the antibiotic cannot attack the ribosome.
- Some microorganisms modify the antib. itself to convert it into harmless compound.
- Other microorganism defend itself against its antibiotics by accumulating it away from the target molecule. For example it stores
antibiotic in the cell wall area which is impermeable, so the antibiotic cannot reach the site of protein synthesis in the cytoplasm.
- Finally, some microorganisms inactivates their antibiotics.

**Describe the different routes used for administration of antibiotics**
Antibiotics can be administered as:
1. Oral administration as capsules, tablets or syrup
2. Intravenous injection for rapid and more effective therapy
3. Local application as cream, eye drops or ointment

**Explain the serial dilution test of antibiotics in liquid medium and what is MIC?**
- A series of tubes of liquid media was prepared and supplied with decreasing concentrations of antibiotics.
- An equal amount of test bacteria are inoculated into the tubes and incubated for 12-16 hours at 37 °C with shaking.
- The tubes are examined either visually or turbidimetrically to measure the bacterial growth.
- Results are recorded as Minimum Inhibitory Concentrations (MIC).

**Explain the Plate diffusion test (Kirby Bauer disk diffusion test)**
- The surface of an agar nutrient medium is uniformly inoculated with the test bacterial culture.
- The tested antibiotics are added in the form of holes on the agar and filled with antibiotics or as circular discs of filter paper placed and impregnated with the antibiotic.
- The culture plates are incubated at 37 °C for 18-24 hours and inhibition zones are determined.
- The zone diameter (zone of inhibition) is measured and reference tables are used to determine if the bacteria are Sensitive (S), Intermediate (I) or Resistant (R) to the antimicrobial drugs.

**What is MIC and the CLSI interpretation guidelines?**
- The MIC is the lowest concentration (highest dilution) of antimicrobial drug that completely inhibits bacterial growth.
- The MIC value is reported as recommended by the Clinical and Laboratory Standards Institute (CLSI) with interpretation guidelines as follow:
  - Sensitive (S),
– Intermediate (I) or
– Resistant (R)

Name the different criteria used to classify antibiotics according to their mode of action?

1. *Antibiotics which interfere with cell-wall synthesis*
   1. beta-lactams, including penicillins and cephalosporins; mono-lactams, such as vancomycin, bacitracin

2. *Antibiotics which interfere with bacterial protein synthesis*
   3. *Antibiotics which bind to the 50S ribosomal unit*
      lincosamides/lincosides including clindamycin and lincomycin; chloramphenicol, macrolides

4. *Antibiotics which interfere the 30S ribosomal unit*
   tetracyclines; aminoglycosides including gentamicin

5. *drugs which interfere with DNA synthesis*
   rifampin, metronidazole, quinolines, novobiocin

6. *Drugs which interfere with cell membrane function*
   polymyxin B, gramicidin

Why streptozotocin damages Beta cells but not other cells and when it is used?
- Because it is similar enough to glucose to be transported into the cell by the glucose transport protein GLUT2 characteristic of beta cells, but is not recognized by the other glucose transporters present in others tissues.
- Streptozotocin is a naturally occurring chemical that is particularly toxic to the insulin-producing beta cells of the pancreas in mammals.
- It is used in medicine for treating certain cancers of the Islets of Langerhans (especially hypoglycemia due to excessive insulin secretion by insulinomas) and used in medical research to produce an animal model for Type 1 diabetes by damaging the DNA of beta cells.

What is the mode of action of streptomycin and why it kills bacteria but does not affect human and when it is used?
- Because humans ribosomes are structurally different from bacteria, thereby allowing the selectivity of this antibiotic for bacteria.
- It kills microbes by inhibiting protein synthesis, it binds to the 16S rRNA of the bacterial ribosome, interfering with the binding of formyl-methionyl-tRNA to the 30S subunit. This prevents initiation of protein synthesis and leads to death of bacteria.

Why Nystatin kills fungus but not affect human?
Because its target is ergosterol which is fairly unique to fungi, and not exist in animals. So it does not have such catastrophic effects on animals.
Nystatin binds to ergosterol, a major component of the fungal cell membrane. When present in sufficient concentrations, it forms pores in the membrane that lead to K⁺ leakage and death of the fungus.

**Why gramicidin is used topically and cannot be introduced by intravenous injection?**
- Its therapeutic use is limited to topical application as it induces hemolysis in lower concentrations than bacteria cell death thus cannot be administered internally. The exterior epidermis is composed of dead cells, thus applying it to the surface of the skin will not cause harm.
- Gramicidin's bactericidal activity is a result of increasing the permeability of the bacterial cell wall allowing inorganic monovalent cations (e.g. H⁺) to travel through unrestricted, thereby destroying the ion gradient between the cytoplasm and the extracellular environment.

**Name the six steps of bacterial cell wall synthesis.**
- N-acetylmuramic acid is manufactured from N-acetylglucosamine by the addition of lactic acid derived from phosphoenolpyruvate.
- Three amino acids are then added to N-acetylmuramic acid to form a muramic acid tripeptide.
- Meanwhile, two D-alanine residues, produced from L-alanine by an enzyme called alanine racemase, are joined together by another enzyme, D-alanine synthetase. These D-ala-D-ala, is added to the tripeptide to form pentapeptide of muramic acid.
- The pentapeptide of muramic acid is joined to other N-acetylglucosamine molecule and passed to a lipid carrier in the cell membrane.
- The whole building block of the newly synthesized soluble peptidoglycan is transported across the cell membrane and added to the end of the existing cell wall insoluble crosslinked peptidoglycan.
- Finally, adjacent units are cross-linked to give more strong crosslinked insoluble peptidoglycan (the cell wall).

**How Fosfomycin and Cycloserine antibiotics inhibit cell wall synthesis?**
- **Fosfomycin:** is a naturally occurring antibiotic originally obtained from a species of Streptomyces. It inhibits the pyruvyl transferase enzyme that brings about the condensation of phosphoenolpyruvate and N-acetylglucos-amine in the formation of N-acetylmuramic acid.
- **Cycloserine:** has broad-spectrum, antibacterial activity. It is used against multiresistant Mycobacterium tuberculosis. The drug bears a structural
resemblance to the D-isomer of alanine and inhibits alanine racemase. It also blocks the synthetase enzyme that links two D-ala molecules together before they are inserted into the cell wall.

**How penicillin, cephalosporin and other b–lactam antibiotics inhibit cell wall synthesis?**
b-lactam antibiotics (like penicillin) are structural analogs of D-Ala-D-Ala termini of the immature peptidoglycan. The cell wall enzyme binding sites can bind b-lactam ring of the antibiotic as the natural substrate (D-Ala-D-Ala-). These enzymes are called Penicillin Binding Proteins (PBPs). The b-lactams (Like Penicillin) **IRRIVERSIBLY** bind the PBPs in the enzyme active site and thus prevent the access of D-alanyl-D-alanine residues to the enzyme and so prevent transpeptidation and peptidoglycan maturation.

**Is it better to take high concentration of antibiotic or lower concentration?**
An efficient antibiotic must be given in a concentration that affect few targets or only one target, to be safe for normal cells. By lowering antibiotic concentration, it will be harmful to only one target and safe for others.

**Antibiotics are characterized by selective toxicity, explain?**
An antibiotic to be marketed it must exhibit selective toxicity. This means that the drug is harmful to the microbe and safe to the host. The term selective toxicity is relative rather than absolute; this means that a drug in a concentration tolerated by host may damage an infecting microorganism. The selective toxicity may be due to the existence of specific antibiotic receptor in the microbe that is not present in the host cells.

**Antibiotics are considered as .....**
1- Primary metabolites   - secondary metabolites

**Antibiotics are produced during.....**
1- trophphase   - idophase

Choose the structure corresponding to polyene antibiotic (Nystatin) :
Choose the structure corresponding to penicillin or b-lactam antibiotics:

Choose the structure corresponding to **Cyclosporine**

Choose the structure corresponding to **Azacytidine**

Choose the structure corresponding to **Chloramphenicol**
Choose the structure corresponding to **Fosfomycin**