Antibiotic sensitivity test

Many types of bacteria ,fungi and viruses that cause diseases to human and animals show resistance to different antibiotics that used for treatment of pathological case leading to economical losses which represents :-

- Long-lasting treatment period
- Quantities of consumed drug
- Downfall of animal health

Advantage of uses this technique:

1. To choose more effective antibiotic which remove the causative agent?
2. To learn of non-effective antibiotics which are resistant to causative agent and changing it by more effective drug in order to decrease costs?

There are two main methods:

1- Dilution methods can done by

- **Tube dilution**
- **Agar dilution**

2- Disc diffusion method

1- Dilution tests

A. Tube dilution test

- Make two fold dilutions of antibiotics in broth media (Mueller Hinton broth) with standard bacterial inoculums.
- Prepare control tube (inoculums +broth) with out antibiotics
- Incubate tubes for 16-20 h , the result was determined by turbidity in tubes, and the tube that contain higher dilution of antibiotic that inhibit bacterial growth (no turbidity) was known as (MIC) Minimum Inhibitory Concentration .
- (MBC) Minimum Bactericidal Concentration could define as higher dilution of antibiotic that prevent bacterial growth (even after long incubation)

Tube dilution test differentiate between bacteriostatic an bactericidal antibiotic

B. Agar dilution test

Prepare antibiotic serial dilution in solid culture media ,each petri dish contain specific dilution this serial dilution is inoculated with one drop of bacterial inoculums using standard loop with out spreading some time replicating device is used

- Bacterial inoculation in this test is calculated according to McFarland's standard tube (0.5M) comparing with turbidity
- After incubation 12-16 h the result is MIC was determined by this way but MBC could not determined in this test

2- Disc diffusion test

- The test is widely used because is very simple and economic.
- Principle of this test depend on uses of standard antibiotic disk
- done by fixing the disk on the surface of agar which contain bacteria (Kirby Bauer method)
- There are two disc type:
  multi disc & uni disc

Procedure

- Saturated cotton swab with bacterial culture broth prepared by comparing with McFarland turbidity standard (0.5M).
- The surface of agar is screening by cotton swab contain bacteria at lest 3 time this done by round the plate in 120 ° time.
- let petri dish for 15 min to absorbed the inoculums
- Sterile forceps (alcohol and flaming) was used to put uni disc or multi disc on agar surface
- By using forceps press on disc to be sure that it adhesive to agar surface and let petri dish for 15 min.
- Incubate petri dish in 37 C ° for 18 h, in converted way

Determine zone of inhibition around the disc and according the diameter refers as resistance (R), sensitive (S), intermediate (M).

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