الكلية: الطب

القسم او الفرع: الاحياء المجهرية

المرحلة: الثالثة

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اسم المادة باللغة العربية: احياء مجهرية

اسم المادة باللغة الإنكليزية: Microbiology

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اسم المحاضرة الرابعة باللغة الإنكليزية: Practical Lecture on Immunology Techniques

PRACTICAL LECTURE ON

IMMUNOLOGY TECHNIQUES

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Procedure:

1. PRECIPITATION REACTIONS

Precipitation occurs when a soluble antigen reacts with its specific antibody to form an insoluble complex visible as a precipitate

A. Ring Test:

B. Principle:

C. When antigen and antibody solutions are layered, a visible precipitate forms at their interface.

D. Materials:

E. Antigen solution

F. Antiserum (antibody)

G. Narrow test tubes or small tubes

H. Micropipettes

1-Place 1 mL of antiserum in a test tube.

2- Carefully overlay 1 mL of antigen solution without mixing.

3- Allow the tube to stand at room temperature undisturbed.

Observation:

A thin white ring of precipitate forms at the junction

if the antigen and antibody are specific.

A. Slide Agglutination

1- Precipitation Reactions

B. Single Radial Immunodiffusion (Mancini Method)

C- Double Immunodiffusion (Ouchterlony Test)

D. Immunoelectrophoresis

2. Agglutination Reactions

Agglutination occurs when insoluble antigens (cells, particles) react with specific antibodies, causing clumping.

Principle:

Particulate antigens (e.g., bacteria) clump when mixed with specific antibodies on a slide.

Materials:

- **Glass slides**
- **Bacterial suspension**
- **Antiserum**
- **Mixing sticks**

Procedure:

1. **Place a drop of antigen suspension on a clean slide.**
2. **Place a drop of antiserum next to it.**
3. **Mix gently with a stick.**
4. **Rock the slide and observe.**

Observation:

•Visible clumping within minutes indicates a positive result.

B. Tube Agglutination (Widal Test Example)

Principle:

Serial dilutions of serum are mixed with antigen suspensions in tubes to detect and measure antibody titers.

Materials:

•Test tubes

•Patient serum

•Known antigen suspensions

•Normal saline

Procedure:

1.Make serial two-fold dilutions of serum in tubes.

2.Add an equal volume of antigen suspension to each tube.

3.Incubate at 37°C for 2–4 hours or overnight.

Observation:

•Clumping/sedimentation in tubes indicates agglutination.

•Titer = highest dilution showing visible agglutination.

3- Complement Fixation Test (CFT)

Principle:

Complement is fixed if antigen-antibody reaction occurs. If complement remains free, it causes hemolysis of indicator red cells.

Materials:

•Complement source (e.g., guinea pig serum)

•Antigen

•Patient serum (antibody)

•Sensitized sheep RBCs (indicator system)

Procedure:

1.Incubate antigen with patient serum and complement.

2.Add sensitized sheep RBCs.

3.Observe hemolysis.

Interpretation:

•No hemolysis = Positive test (complement fixed, antigen-antibody present).

•Hemolysis = Negative test (no antigen-antibody reaction).

Thank you