# PRESENTATION TOPIC:
## INTRODUCTION TO DIAGNOSTIC BACTERIOLOGY

Subject: Systematic Diagnostic Bacteriology

Muhammad Abbas

BS MLT

F.sc MLT

Rehman College of Allied Health Sciences Peshawar

# INTRODUCTION TO DIAGNOSTIC BACTERIOLOGY

- The main function of all diagnostic bacteriology laboratories is the detection and identification of microorganisms in a variety of samples of human, animal, food, industrial, or environmental origin.

- in clinical laboratories, drug susceptibility testing of the isolates to allow correct treatment decisions is of major importance

# Diagnosis of Microbial Infection
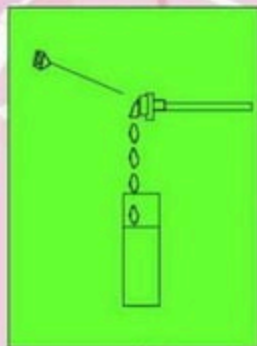
# Specimen processing

- Receiving
- Recording
- Culturing
- Staining
- Isolation
- Identification
- Sensitivity test

# Getting the specimen to the lab

- Problems in delay or inappropriate storage.
- Delay in diagnosis & treatment lead to:
  - ✓ pathogens die.
  - ✓ contaminants overgrow.
- Blood cultures directly into incubator not refrigerator!
- CSF straight to lab.
- Don't put an entire surgical specimen into formalin!
  But: Send a portion to microbiology in a sterile container.

# Collecting the specimen correctly

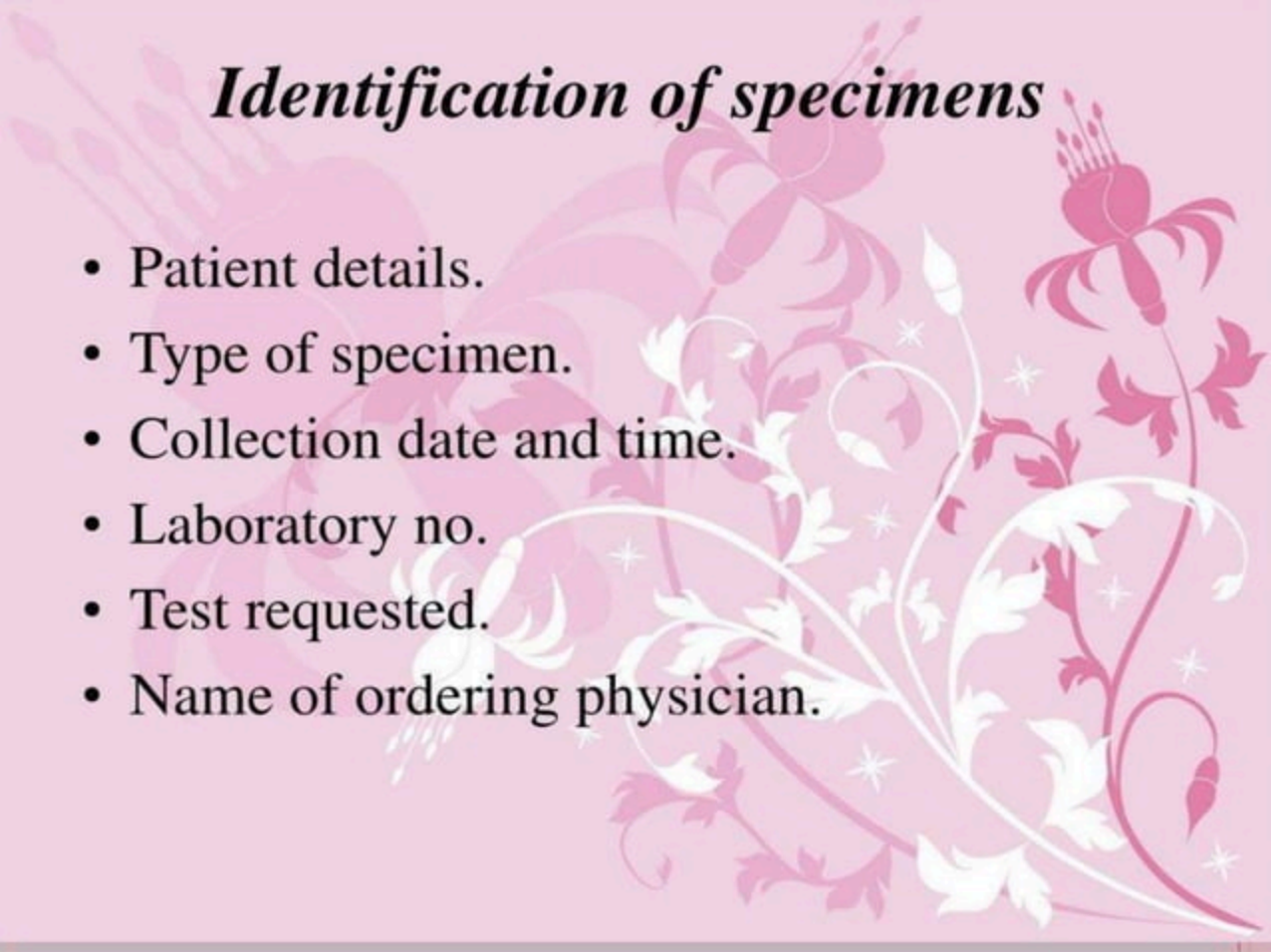- Take an mid-stream urine to:
  - ✓ avoids contamination with normal flora.

- Blood cultures
  - ✓ Avoid contamination with skin organisms

- CSF
  - ✓ Avoid contamination.
  - ✓ Avoid bloody tap.

- Throat swab
  - ✓ Make the patient gag!

# _Patient Details_

- Name and age
- Hospital no
- Sex, for female: if she pregnant or lactating
- Address
- Suspected diagnosis
- Travel history
- Immunization

# Identification of specimens

- Patient details.
- Type of specimen.
- Collection date and time.
- Laboratory no.
- Test requested.
- Name of ordering physician.

# Normal microbiota

- All body surfaces possess a rich normal bacterial flora, especially the mouth, nose and skin.
    - This can be a nuisance in that
        - ✓ It can contaminate specimens.
        - ✓ It can cause disease.
    - This is beneficial in that
        - ✓ It can protect against infection by preventing pathogens colonising epithelial surfaces (*colonisation resistance*).

    *NOTE*:Removal of the normal flora with antibiotics can cause superinfection, usually with resistant microbes.

# Microbiota and humans

Disease can come about in several overlapping ways

1.  Some bacteria are entirely adapted to the pathogenic way of life in humans. They are never part of the normal flora but may cause subclinical infection, e.g. *M . Tuberculosis.*

2. Some bacteria which are part of the normal flora acquire extra virulence factors making them pathogenic, e.g. *E. coli.*

3. Some bacteria which are part of the normal flora can cause disease if they gain access to deep tissues by trauma, surgery, lines, e.g. *S. epidermidis.*

4. In immunocompromised patients many free-living bacteria and components of the normal flora can cause disease, especially if introduced into deep tissues, e.g. *Acinetobacter.*

# *Specimens & Infection Control*

- Please be considerate to lab staff!!
  - ✓ Label hazardous specimens

- Don't send specimens to the lab without proper packing.
  - ✓ Leaking or blood-stained specimens are not acceptable!!!

# Factors limiting usefulness of bacteriological investigations

- Wrong sample for example saliva mixed with sputum.
- Delay in transport / inappropriate storage e.g. CSF.
- Overgrowth by contaminants e.g. blood cultures.
- Insufficient sample / sampling error e.g. in mycobacterial diseases.

- Patient has received antibiotics.

# Specimen rejection criteria

- Mismatch information
- Improper container or temperature
- Insufficient specimen
- Leaking specimen
- Formalin specimen
- Dried out swap
- Late specimen

*Physician must be informed about rejection*
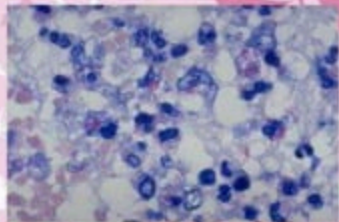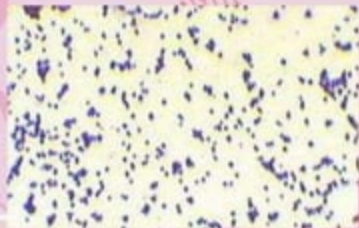
# METHODS OF IDENTIFICATION

There are different methods used in the diagnosis of diseases i-e

1. Microscopy
2. Culture of bacteria
3. Sensitivity tests
4. Serological tests
5. Molecular detection

# 1.Microscopy stained preparations

- Gram-stain.
- Acid-fast stain (Ziehl-Neelsen).
- Special stains.
- Fluorescence
  - Direct, e.g. auramine
  - Immunofluorescence

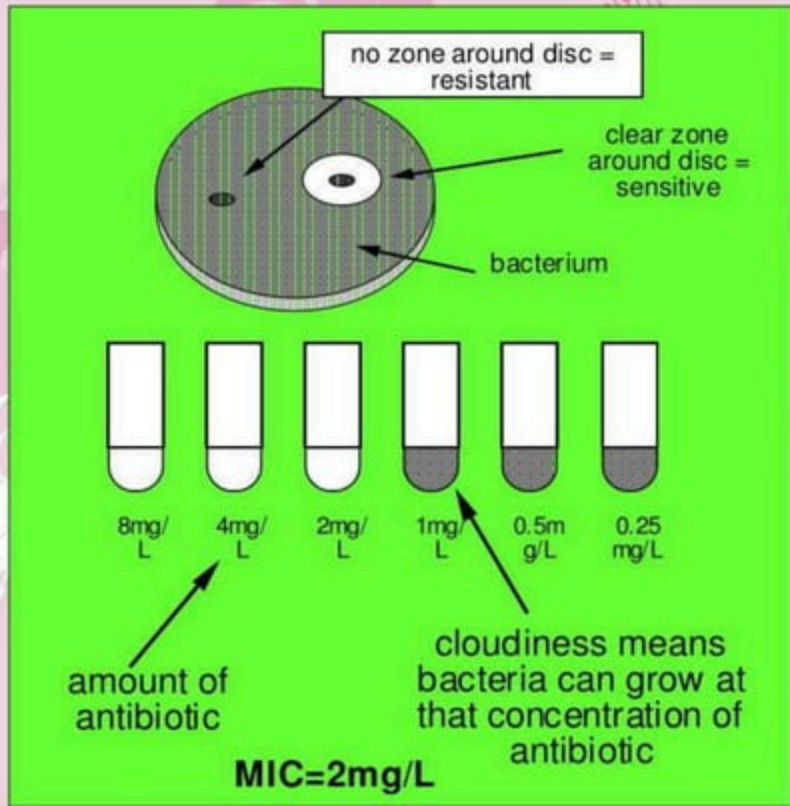# 2.Culture of Bacteria

- Solid media
  - ✓ Agar plates
    - ✓For identification
    - ✓For counting
  - ✓ Slant
    - ✓For safe long-term culture, e.g. Lowenstein-Jensen media for TB
- Liquid (broth) media
  - For enrichment or maximum sensitivity
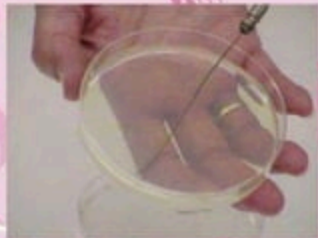
# 3. Sensitivity tests

- on solid media
  - ✓ disc diffusion technique
- in liquid media
  - ✓ minimum inhibitory concentration (MIC) test
- E-test



no zone around disc = resistant

clear zone around disc = sensitive

bacterium

8mg/L  4mg/L  2mg/L  1mg/L  0.5mg/L  0.25mg/L

amount of antibiotic

cloudiness means bacteria can grow at that concentration of antibiotic

MIC=2mg/L

# 4.Serology tests

- Antigen detection
  - ✓ e.g. latex agglutination
- Antibody detection
  - ✓ e. g. agglutination tests, complement fixation tests, indirect immunofluorescence

# 5.Molecular methods

✓ Polymerase Chain Reaction (PCR)

# Overview of
# Bacterial infections

**Bacterial meningitis**
- *Streptococcus pneumoniae*
- *Neisseria meningitidis*
- *Haemophilus influenzae*
- *Streptococcus agalactiae*
- *Listeria monocytogenes*

**Otitis media**
- *Streptococcus pneumoniae*

**Pneumonia**
Community-acquired:
- *Streptococcus pneumoniae*
- *Haemophilus influenzae*
- *Staphylococcus aureus*
Atypical:
- *Mycoplasma pneumoniae*
- *Chlamydia pneumoniae*
- *Legionella pneumophila*
Tuberculosis
- *Mycobacterium tuberculosis*

**Skin infections**
- *Staphylococcus aureus*
- *Streptococcus pyogenes*
- *Pseudomonas aeruginosa*

**Eye infections**
- *Staphylococcus aureus*
- *Neisseria gonorrhoeae*
- *Chlamydia trachomatis*

**Sinusitis**
- *Streptococcus pneumoniae*
- *Haemophilus influenzae*

**Upper respiratory tract infection**
- *Streptococcus pyogenes*
- *Haemophilus influenzae*

**Gastritis**
- *Helicobacter pylori*

**Food poisoning**
- *Campylobacter jejuni*
- *Salmonella*
- *Shigella*
- *Clostridium*
- *Staphylococcus aureus*
- *Escherichia coli*

**Sexually transmitted diseases**
- *Chlamydia trachomatis*
- *Neisseria gonorrhoeae*
- *Treponema pallidum*
- *Ureaplasma urealyticum*
- *Haemophilus ducreyi*

**Urinary tract infections**
- *Escherichia coli*
- *Other Enterobacteriaceae*
- *Staphylococcus saprophyticus*
- *Pseudomonas aeruginosa*
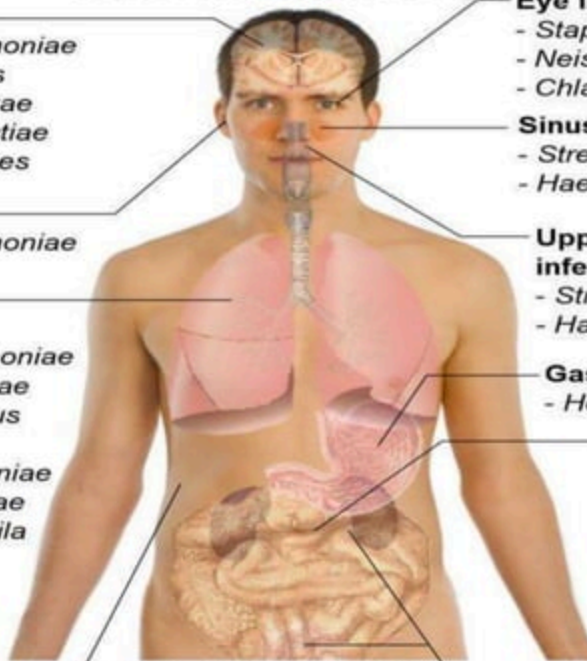
Thank You so much.....

Presentation by Muhammad Abbas