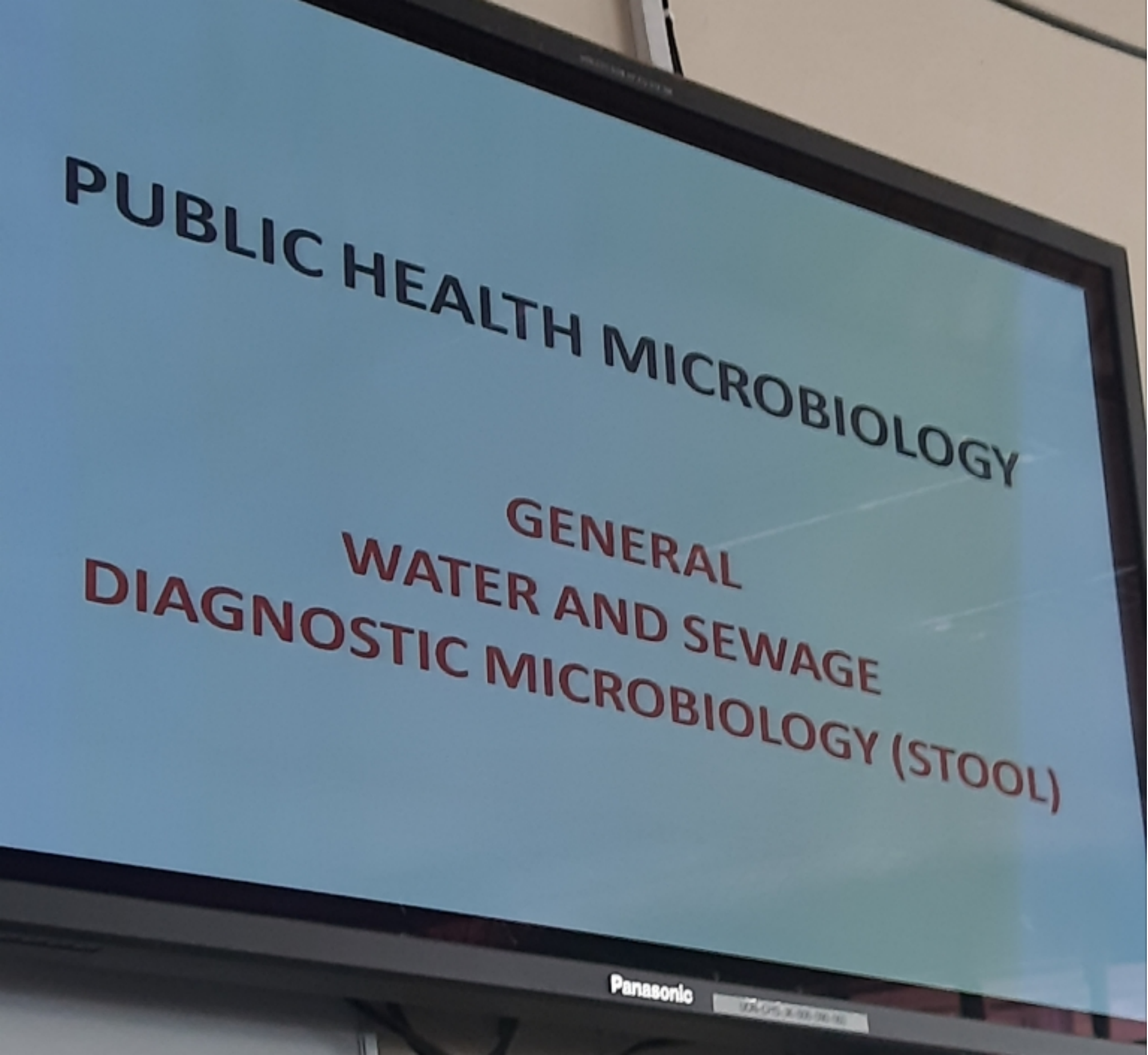
PUBLIC HEALTH MICROBIOLOGY
GENERAL
WATER AND SEWAGE
DIAGNOSTIC MICROBIOLOGY (STOOL)
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- *Public health microbiology is 'a cross-cutting area that spans the fields of human, animal, food, water and environmental microbiology, with a focus on human health and disease' (CDC)*

Public health related policy development..research the link between the microorganisms, the disease and its impact in the community

Areas of importance include:

1. **Food microbiology**: meat inspection; milk inspection, public eating places licensing; food handlers licensing
2. **Water and Sanitation**: Treatment of water and Disposal of sewage
3. **Security**: Bioterrorism prevention/monitoring Bio-safety, Containment and biohazard response in outbreaks
4. **STD**: STD agents, diagnosis, prevention and public education

...areas of importance

5. **Emerging** and re-emerging pathogens
6. **Vector** borne and zoonotic diseases
7. **Blood transfusions**- Diseases transmitted through
8. **Migrant populations-** Diseases of concern to travellers, immigrants and refugees
9. **Vaccination**

Goals of public health microbiology

- Study the **course** or history of disease
- Determine the **frequency** of disease
- Identify **patterns** of disease **occurrence**
- Identify **risk factors** for potential causes of disease
- Evaluate the effectiveness of **preventative measures**

terms

- Carrier
  - Person or animal who harbors and spreads micro-organisms that cause disease
  - Person DOES NOT have clinical symptoms of disease
  - Example: chronic carriers of *S. enterica* typhi

...

- *Reservoir*
  - *Source of infection*
  - *Site where the pathogen can multiply or survive until its transferred to the host*
  - *Examples: people, animals, environmental elements eg water,soil*

- Endemic
  - Organism or disease is constantly present in a population
  - Examples: Cholera in third world countries
- Epidemic
  - Disease affects a significantly large number of people at the same time in a geographic area
  - Examples: West nile virus in 2002 in the U.S
- Pandemic
  - Worldwide epidemic
  - Example: Swine flu

- Index case
  - First case of a disease which serves as source of infection
- Morbidity Rate
  - Rate at which an illness occurs
- Mortality Rate
  - Number of deaths caused by a disease in a population
- Surveillance
  - Collection of data pertaining to disease occurrence

- Laboratory data on Prevalence/AST profiles of bacterial isolates
  - Influence the initial antibiotic choices and empirical therapy
  - Prevent the buildup of antibiotic resistance

- Water sources for domestic/industrial use: lakes, rivers, streams, wells, boreholes, piped water.

Contamination of water sources

- Surface and shallow ground water:

Show high degree of bacteriological and chemical pollution due to

- poor human excreta disposal, use of raw animal manure
- Industrial/agricultural activities..run off from fertilizer

- *Deep wells and springs - low bacterial content due to filtration through soil.*

- *Lakes and reservoirs: on-going process of sedimentation may decrease content significantly.*

❖*Boreholes and wells should be at least 30 meters from structures like pit latrines, septic tanks, refuse dumps: avoids seepage*

Water quality parameters

3: microbiological, physical and chemical

-Chemical parameters: industrial waste pollutants, agricultural chemicals run-off...

-Physical parameters: turbidity, colour, odour.

treatment should yield water free from coliform organisms.

Coliforms are Gram negative rod-like bacteria normally found in the GIT of man and other warm-blooded animals

-Water borne diseases
-Water hygiene-related diseases
-Vector-borne water habitat diseases.

Water borne Diseases

Enteric diseases (non-invasive diarrhea, dysentery, toxin related) due to pathogens. Examples

- Bacteria: Salmonella, Shigella, Escherichia, Vibrio, Staphylococcus,...
- Enteroviruses: Rotavirus, Polio virus, Norwalk...
- Protozoa: Entamoeba...
- Helminthes: Ancylostoma...

etc

-Inadequate/ improper use of water in personal cleanliness.

-Inadequate water for community needs.

-Use of polluted water sources eg rivers

- Use of contaminated water in recreation facilities

Diseases include

- Trachoma
- Legionnaire's

- *Snail vectors- transmission of Schistosomiasis*
- *Mosquito vectors- malaria, filariasis, arbovirus*
- *Fly vectors- onchocerciasis (river blindness), trypanosomiasis*

etc

- Intestinal bacterial pathogens known to contaminate drinking water include:

- *Salmonella*
- *Shigella*
- *E. coli*
- *Vibrio cholera*
- *Yersinia enterocolitica*
- *Campylobacter fetus*

etc

throat infections when present in large quantities in water used for drinking, bathing or medical purposes:

- *Pseudomonas*
- *Flavobacterium*
- *Acinetobacter*
- *Klebsiella*
- *Serratia*

- *Indicators of excreta pollution of water*

- *Tests check for presence of organisms normally present in stool of man and other warm blooded animals.*

- *Measures the efficacy of water treatment.*

- Naturally abundant in excreta
- easily isolated
- easily identified and enumerated
- more resistant to disinfectants such as chlorine.

Citrobacter; Enterobacter; Aeromonas; Klebsiella.

meet the above criteria.

- Other supplementary indicators include:

- Faecal Streptococci
- Sulfite reducing Clostridia
- Bacteroides
- Bifidobacteria

- *An indicator of treatment efficiency or post treatment contamination*
- *Used to simultaneously detect all coliform bacteria in a drinking sample.*
- *They should not be detectable in* ***treated water*** *supplies: no. of coliforms should* ***be zero***

- *Coliform organisms able to ferment lactose at 44^0c or 44.5^oc.*

- *Comprise of E. coli*, and <u>to a lesser extent</u> *Enterobacter*, Citrobacter and Klebsiella.

- Avoid contamination.
- Collect adequate amount 200ml in sterile bottle.
- Wide opened mouth with sterile glass stopper
- Cap released at point of collection.

- Deliver sample to the lab ASAP, 2-6 hours; or refrigerate.
- Neutralize the chloride ions with 3% sodium thiosulphate.
- Mix the sample and pick at different water levels.

Determining Bacterial Counts:

1. Quantitative Analysis

- non-differential test
- for counts of all viable bacterial
- pour onto media plates in duplicates
- eliminates the probability of error and is reproducible.

2. Qualitative Analysis: Eijkman test

- differential test.
- Specific for coliform eg *E.coli*.
- Uses MacConkey media (look for Lactose fermenters) and Durham's tube.

- Incubated at 44°c (thermotolerant *E. coli*)
- if gas and acid is produced and the media turns yellow at 44°c, it is faecal coliform *E.coli*.

- For sampling larger volumes of water.
- Filter a known amount through a pore filter which you will incubate
- Bacteria grows and forms colonies on the filter.
- Identify the bacteria.
- Quantity can be calculated using colony counts and volume filtered.

Water Disinfection

-Destroy pathogens in water

-Prevent entry of pathogens to water system (residual effect)

-Suppress bacterial growth in pipe environment (residual effect)

...water disinfectants

- must kill the organisms and also have a non-toxic residual effect...
- Can be physical or chemical

...examples of chemical disinfectants

- Chlorine....(ratio of 4 parts per million)
- Bromine
- Metals eg copper
- Hydrogen peroxide

...examples of physical disinfectants

- UV light
- Electronic radiation
- Gamma rays

Sewage Treatment

- Sewage treatment is a controlled intensification of natural self-purification processes
- Involves primary, secondary and tertiary treatments.

Primary	- Mainly physical processes - Removing insoluble particulate matter with coagulation agents- alum
Secondary	- Biological removal of dissolved organic matter; anaerobic digesters, extended aeration systems
Tertiary	- Biological removal of inorganic matter - Chemical removal of inorganic matter - Virus removal/inactivation - Trace chemical removal

- *In laboratory diagnosis of diarrhoeal disease, examination of stool specimen is key for identification of causative agent(s)*

OUTLINE OF INVESTIGATION of INFECTIOUS DIARRHOEA

- Stool
  - Collection and Transport of specimen
  - Macroscopic appearance
  - Microscopy
  - Culture
  - Biochemical Tests and Serological tests
- Others investigations

- *Provide patient with suitable container:*
  (bedpan or wide mouthed container.)

  -Clean and dry
  -Disinfectant free
  -Leak-proof

- *Avoid contamination with urine*

- *Transport labelled specimen to lab within* **1 hour.**

- Otherwise use transport media

Cary- Blair:

-Cotton swab of specimen/ Rectal swabs

-Salmonella, Shigella, Vibrio, Yersinia: viable for 48 hours

-Campylobacter: viable for 6 hours

Alkaline Peptone Water:**enrichment

- Used when cholera is suspected.
- 1ml specimen in 10ml of media:
- Maintains viability for 8 hours.

1. Describe the appearance (macroscopic)

APPEARANCE	POSSIBLE CAUSE
Unformed, Pus, Mucous, Blood	Shigellosis EIEC Dysentry Campylobacter spp
Bloody Diarrhoea- without pus cells	EHEC O157 (Hemmorhagic colitis)
Watery Stools	ETEC, EPEC Diarhhoea
Rice Water Stools with mucous flakes	Cholera
Unformed/ watery sometimes with blood, mucous,	Salmonella

2. **Examine the specimen microscopically**

- Saline Wet preparation

To detect Red Blood Cells in specimen.

- Methylene blue preparations:

To examine for leucocytes Mononuclear cells, PMNs, red blood cells

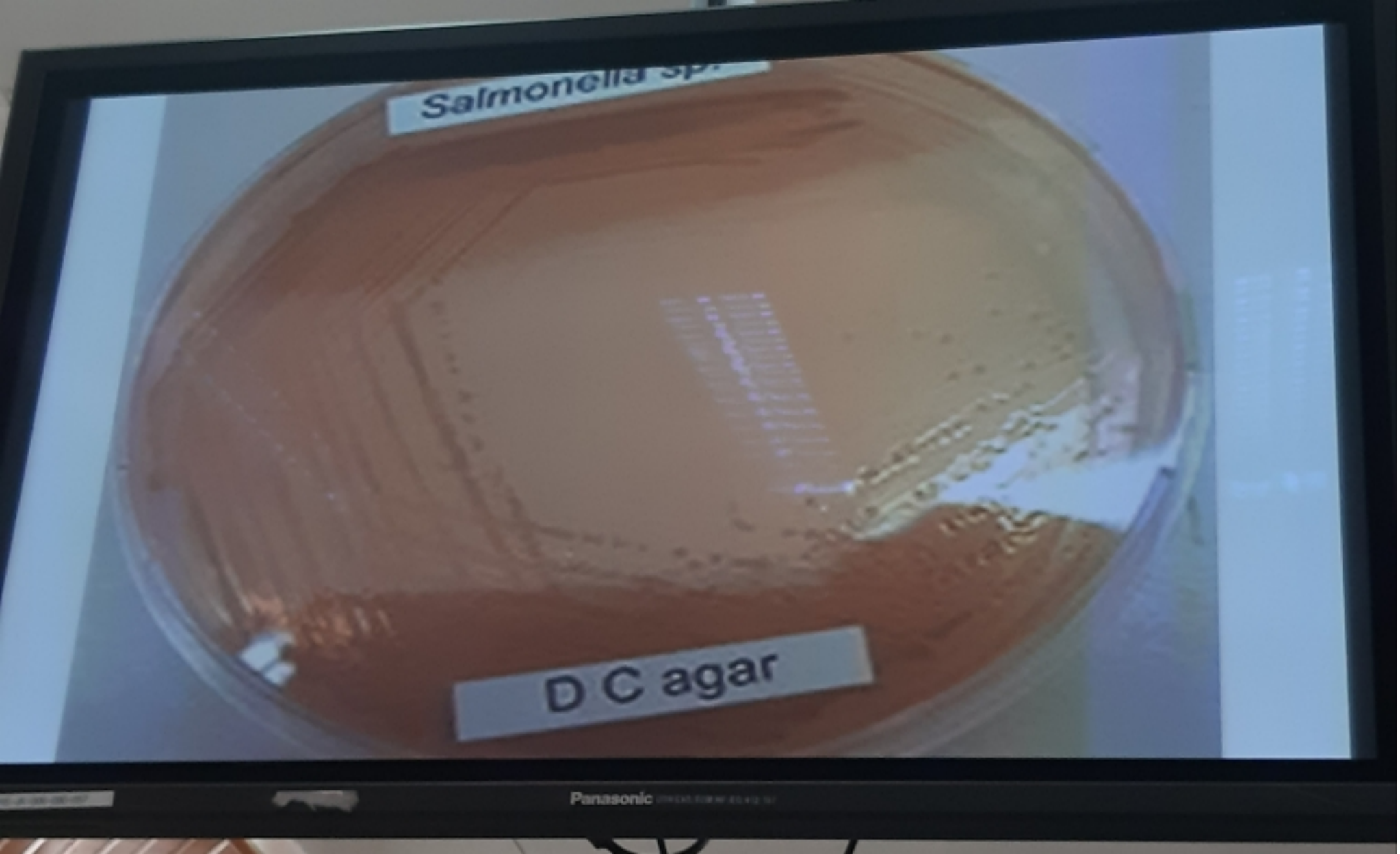
3. **Culture the Specimen**

Enrichment media

- Alkaline Peptone Water: Vibrio
- Selenite F Broth: Salmonella, Shigella

Selective Media

- Xylose Lysine Deoxycholate Agar: Salmonella, Shigella
- Salmonella Shigella Agar: ”
- Deoxycholate Agar: ”
- TCBS: Vibrio
- Skirrows: Campylobacter


Salmonella sp.
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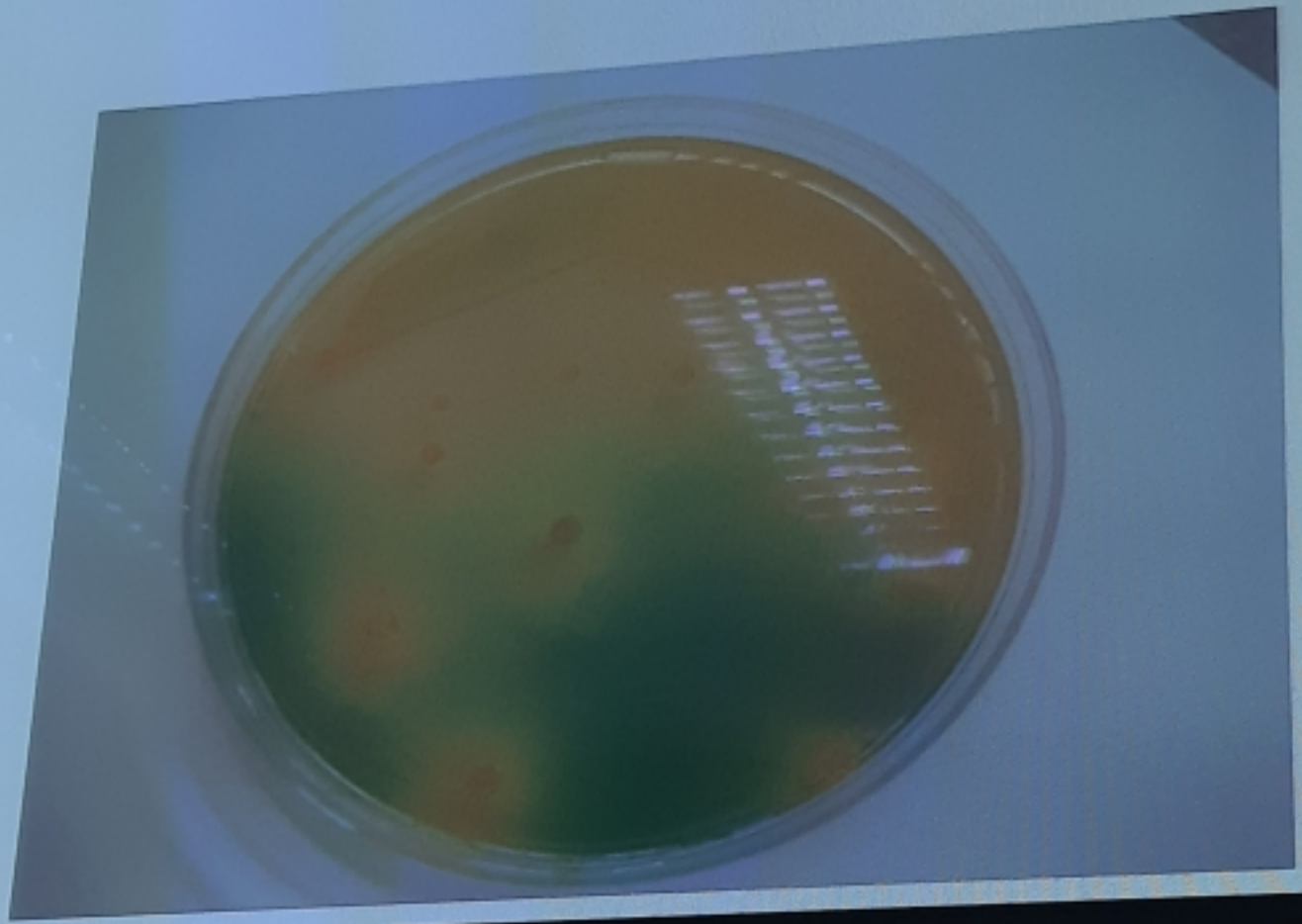

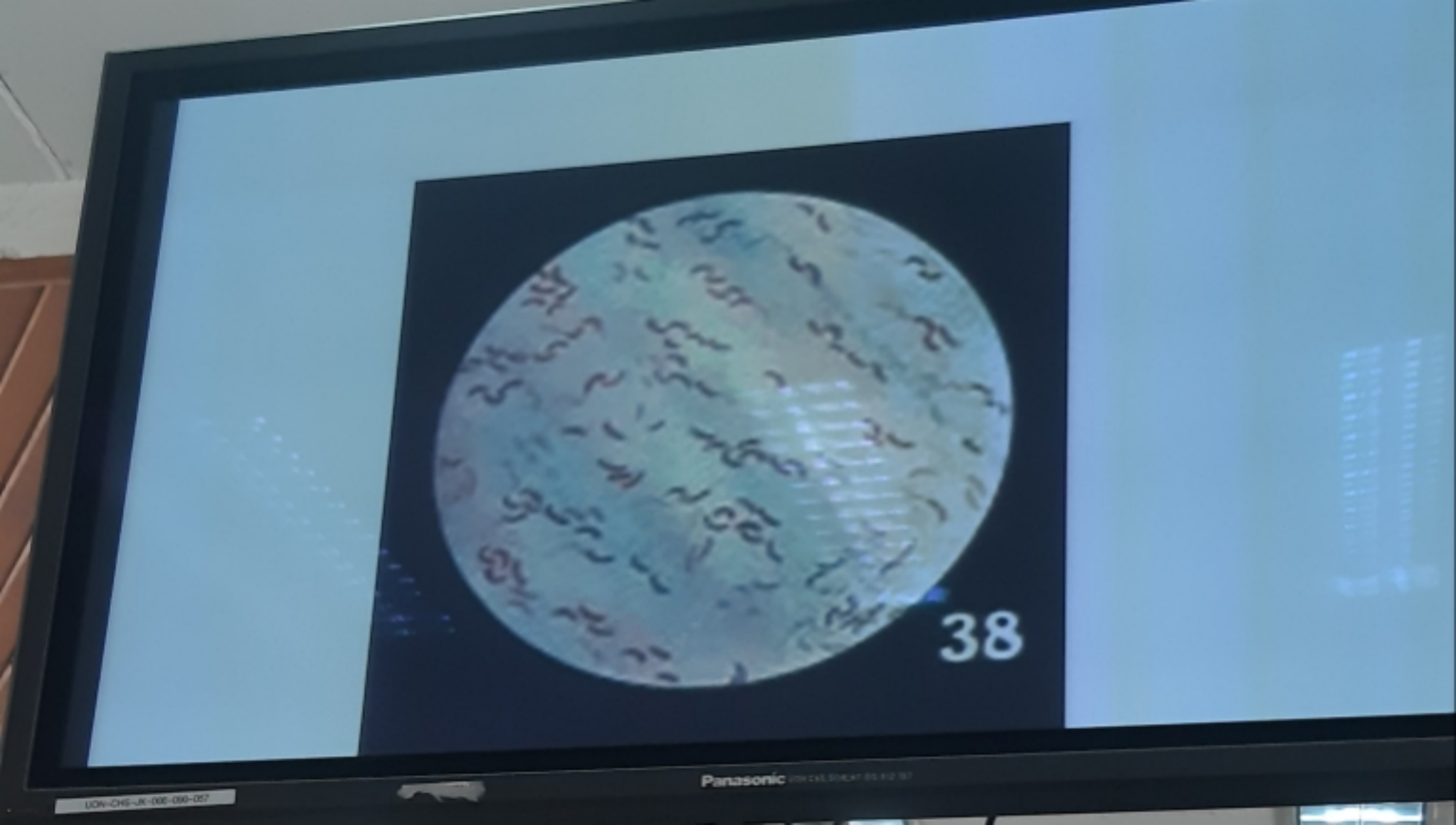
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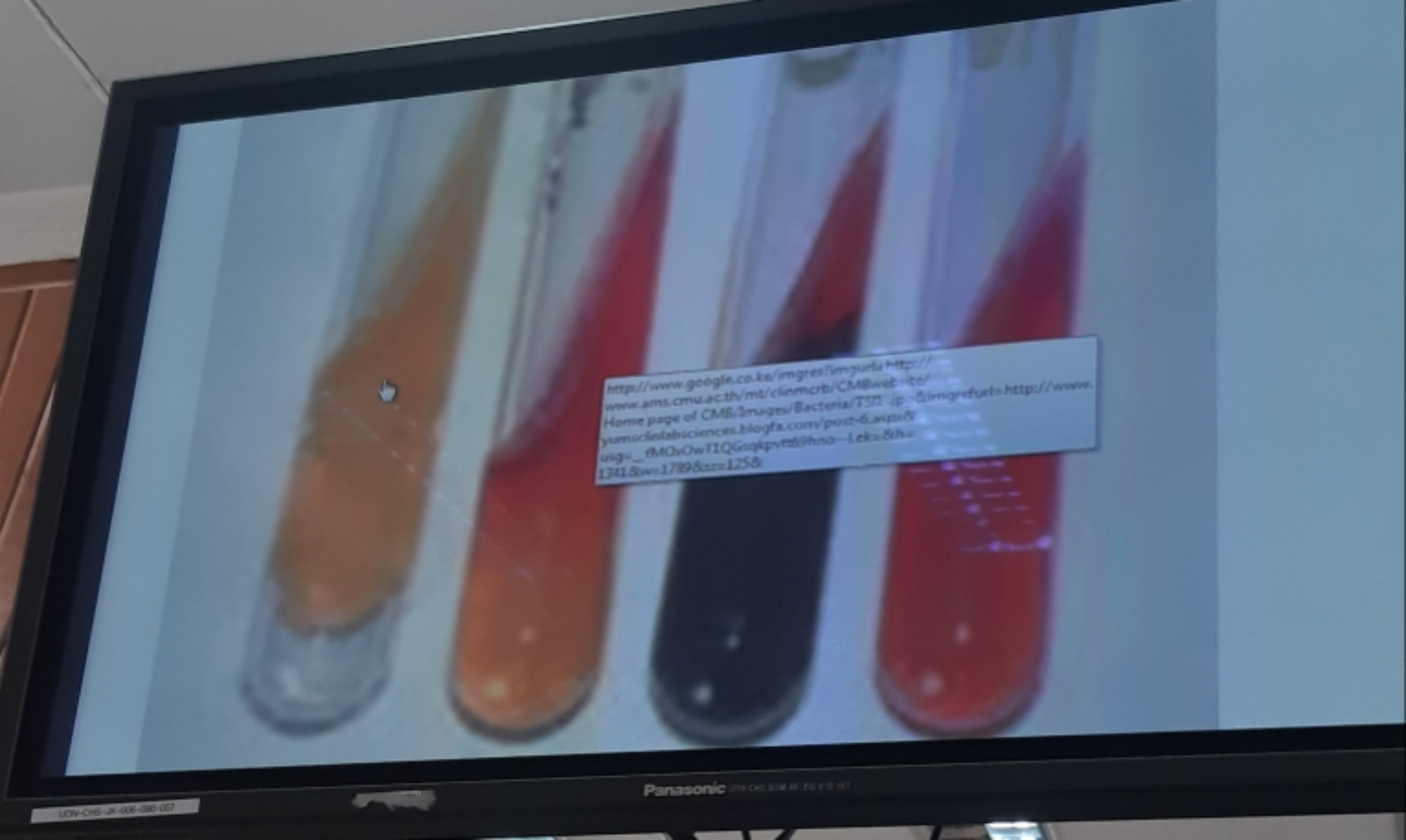
5. **Biochemical Tests**

- On XLD, SS, DCA, MAC plates:
  - Exclude Proteus using urease
  - Set Indole, Motility, TSI/ KIA tests
  - Identify serologically

- On TCBS
  - Gram stain colonies
  - Subculture to NA
  - Oxidase Test
  - Identify serologically

- On Sorbitol MacConkey
  - O157 Latex agglutination Tests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6. **Serotyping**

- This is done using commercially prepared antisera for identification of specific bacteria serotypes from pure cultures.

- The tests are based on antibody-antigen agglutination reactions.

Besides lab examination of stool specimen,other investigations can include:

- Blood tests
- Sigmoidscopy: view colonic mucosa
- Biopsies
- Abdominal CT scans

- Mild cases: oral rehydartion
- Severe dehydration: IV replacement of fluids, elctrolytes
- Antibiotics: Specific to pathogen