

## Gastrointestinal tract infection (Gastroenteritis/Diarrhea)

### General Stool Exam (GSE):

Stool examination is carried out in laboratories for various diagnostic purposes. It is a specimen that is easily obtained but they may be a reluctance on the part of the patient to give the stool specimen due to its offensive nature and foul smell. Mostly a clean container, which does not contain any detergent or disinfectant, is sufficient for all types of stool examinations including stool culture.

A stool culture is ordered in the following cases:

- Severe or bloody diarrhea, with increased amounts of gas, bloating, loss of appetite, nausea, vomiting, fever and cramping pain.
- Food poisoning, inflammation of the large intestine (colitis), cholera and typhoid to identify the organisms that can cause these infections.
- Asymptomatic persons that can be as a carrier for bacteria and have the ability to spread infections to others patients.

### Pathogenic bacteria of GIT (Gastrointestinal tract)

*Campylobacter jejuni*, *E.coli*, *Shigella* spp., *Salmonella* spp., *Vibrio cholerae*, *Clostridium difficile*, *Staphylococcus aureus*

**Macroscopic examination:** by naked eye we can describe the following:

- **Consistency of the stool sample:** moisture in the stool, formed stool (which takes the shape of the colon), soft, liquid, mushy, loss, semi liquid, hard, very hard, watery.

- **Color of the stool:** brown (normal color), light brown, clay, green, blue, yellow. The type of food, medication, and during infection affects these colors.
- **Odor:** The fecal odor of stools may become offensive in conditions like intestinal amoebiasis. In cases of bacillary dysentery and cholera, the stools are not foul smelling due to the absence of fecal matter.
- **Blood:** thread of blood, frank of blood (in case of abscess), bright red blood on the surface of the formed stool is usually associated with bleeding and irritation.
- **Pus:** in case of infection.
- **Mucous:** In case of severe diarrhea, there will be shedding of the intestinal wall and in the stool, this will appear as the delicate cover of the egg.
- **Undigested food particles.**
- **Parasite:** adult worms or segment of them.
- **Epithelial cells:** in case of shedding in the intestine, the nucleus is large and the cell is globulated and sometimes part of the epithelium appears.
- **Crystals:** Charcot-leyding crystals: is a diamond in shape and it may occur in stool specimen and it is indicator for the parasitic infection. These crystals developed from the eosinophilic break down products, which indicate that an immune response has been taken place due to parasitic infection.

### **Stool culture:**

#### **Collecting a stool sample:**

1. Label the container with name, date of birth and the date.

2. Place something in the toilet to catch the stool, such as a potty or an empty plastic food container, or spread clean newspaper or plastic wrap over the rim of the toilet.
3. Make sure the sample doesn't touch the inside of the toilet.
4. Use the spoon or spatula that comes with the container to 5-place the sample in a clean screw-top container and screw the lid shut.
5. If you've been given a container, aim to fill around a third of it that's about the size of a walnut if you're using your own container.
6. Put anything you used to collect the sample in a plastic bag, tie it up and put it the bin.
7. Try not to collect urine or water from the toilet with the stool sample. If you need to urinate, do this first before collecting the stool sample.
8. Wash your hands thoroughly with soap and warm running water.

**Note:**

- A rectal swab is sometimes considered an easy, viable alternative to a stool sample.
- The stool collection process can be more difficult with infants in diapers or people with active diarrhea. If collecting a stool sample from baby, use a cotton swab to collect a sample from rectum.

**Storing a stool sample**

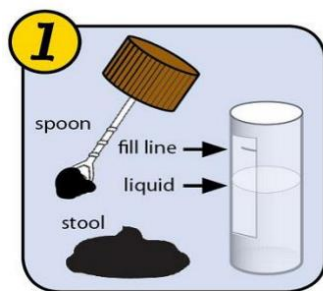
- Stool samples should be handed in as soon as possible, as some can't be analyzed if they've been refrigerated.
- If you can't hand the stool sample in immediately, you should store it in a fridge, but for no longer than 24 hours. Place the container in a sealed plastic bag first.
- Stool samples must be fresh – if they aren't, the bacteria in them can multiply. This means the levels of bacteria in the stool sample won't be

the same as the levels of bacteria in digestive system. If the levels of bacteria don't match, the test results may not be accurate.

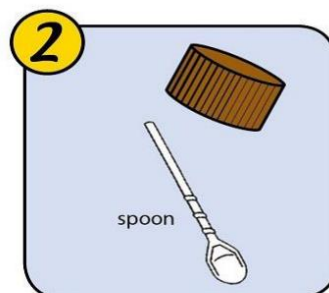


Container for stool sample collection

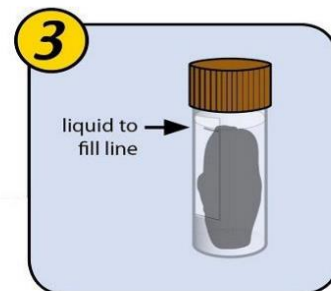
Swabs for stool sample collection



Collect on plastic wrap and transfer to vial until liquid reaches fill line.



Remove spoon from lid and discard.



Replace cap on vial tightly and shake for a minute. Place vial in refrigerator until ready to ship.

## Laboratory diagnosis

### 1-Specimen culture:

- a. Enrichment media:** Selenite F broth (isolation of *Salmonella* species), Alkaline peptone water (recommended as an enrichment broth for isolating *Vibrio cholerae*), Tetrathionate broth (isolation of *Salmonella* and *Shigella*), Campylobacter Enrichment broth (A selective enrichment broth for the isolation of *Campylobacter* spp.).

**b. Selective differential media:**

\*MacConkey agar.

\*Blood agar.

\***TCBS** agar (Thiosulfate Citrate bile salts sucrose) (for *Vibrio*)

\*XLD agar (Xylose Lysin Deoxycholate).

\*Abeyta-Hunt –Bark agar without antibiotics (for *Campylobacter*).

\* Heart infusion agar (HIA) Slant (for *Campylobacter*).

**2- Microscopy:** Gram stain.**3-Biochemical tests.**

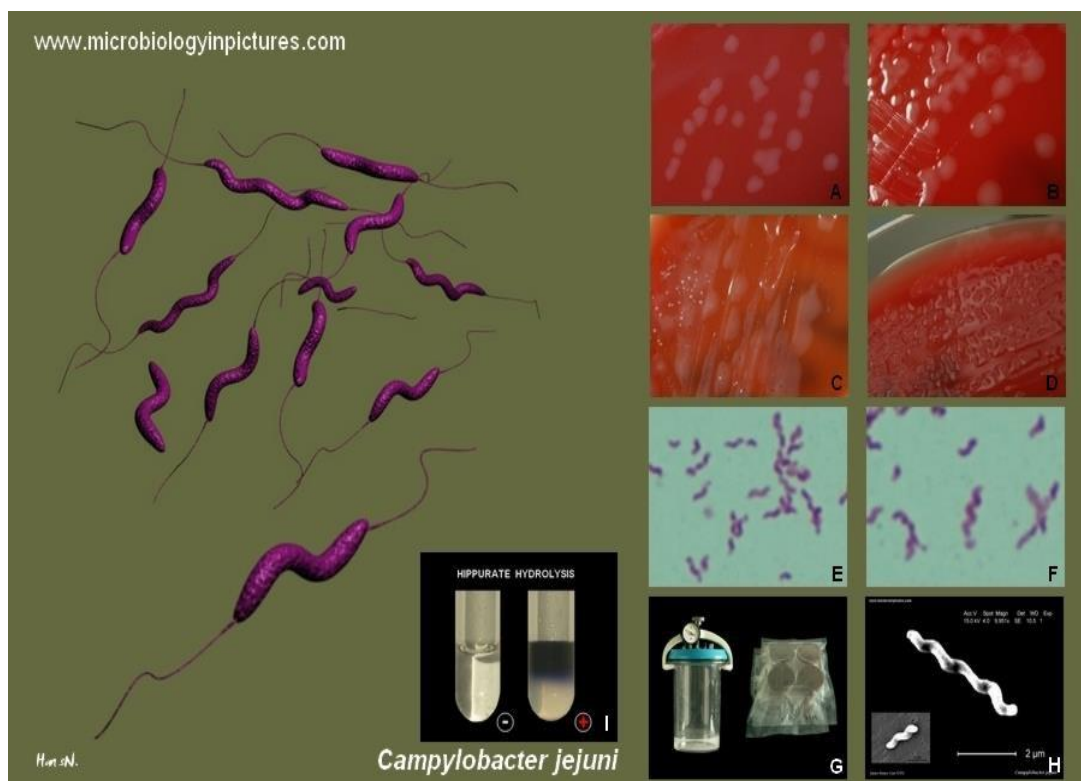
- Catalase Test.
- Motility test.
- Oxidase Test.
- Methyl Red / Voges-Proskauer (MR/VP).
- Kligler's Iron Agar (KIA).
- Nitrate Broth.
- Simmon's Citrate Agar.
- Urease test.
- Carbohydrates fermentation test.
- Sulfur Indole Motility Media (SIM).

**4- Serological methods.****Pathogenic bacteria cause Gastroenteritis.**

- ***Campylobacter* spp.**

*Campylobacter* most common species is *C.jejuni*. (meaning "curved bacteria") **seagull wing shaped**) is the leading cause of bacterial enteritis in

the world. It is G-ve bacteria, typically appear comma or S-shaped and motile by unipolar or bipolar flagella. They generally survive in environments with low oxygen (Microaerophilic) 5% O<sub>2</sub> and 10% CO<sub>2</sub>. They are oxidase test (+ve) and catalase test(+ve), and nonfermentative. Campylobacters are best cultured at 42 °C .Survival at room temperature is poor, but Campylobacters can survive for a short time at refrigeration temperatures—up to 15 times longer at 2°C than at 20°C. The bacteria die out slowly at freezing temperatures and is heat sensitive, the cells are destroyed at temperatures above 48°C, (+ve) for hippurate hydrolysis.



- ***Salmonella* spp.**

*Salmonella* is a genus of rod-shaped (bacillus) gram-negative bacteria of the Enterobacteriaceae family. The two species of *Salmonella* are *Salmonella enterica* and *Salmonella bongori*. *Salmonella enterica* is the type species and is further divided into six subspecies that include over 2,500 serotypes. Strains of *Salmonella* cause illnesses such as typhoid

fever, paratyphoid fever, and food poisoning (salmonellosis). *Salmonella* species are nonspore-forming, predominantly motile enterobacteria. They are also facultative anaerobes, capable of surviving with or without oxygen. Most subspecies of *Salmonella* is H<sub>2</sub>S(+ve), which can readily be detected by growing them on media containing ferrous sulfate, such as is used in the triple sugar iron test, non-lactose fermenting.

## On Salmonella Shigella Agar

- is used for the isolation of *Salmonella spp.* and some strains of *Shigella spp.*
- inhibit gram-positive bacteria, most coliform bacteria, and inhibit swarming *Proteus spp.*, while allowing *Salmonella spp.* to grow.



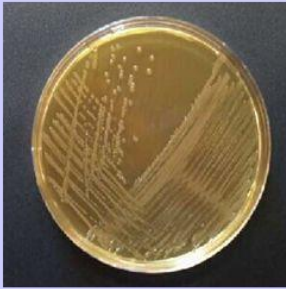
*Salmonella* will not ferment lactose, but do produce hydrogen sulfide gas. The resulting bacterial colonies will appear colorless with black centers.

- ***Shigella spp.***

*Shigella* is G<sup>-ve</sup>, facultative anaerobic, nonspore forming, non-motile, genetically closely related to *E.coli*. It is causative agent of shigellosis. It is classified by four serogroups: Serogroup A (*S. dysenteriae.*), Serogroup B (*S. flexneri.*), Serogroup C (*S. boydii.*) and Serogroup D (*S. sonnei.*). Lactose fermenting except *S. sonnei.*, H<sub>2</sub>S(-ve).

## *Shigella* spp Isolation

- *Shigella* organisms may be very difficult to distinguish biochemically from *Escherichia coli*.
- *Shigella* species are Gram-negative, facultatively anaerobic, nonsporulating, nonmotile rods in the family *Enterobacteriaceae*.
- They do not decarboxylate lysine or ferment lactose within 2 days.



NON-LACTOSE  
FERMENTERS:  
COLOURLESS  
COLONIES



MacConkey Agar  
LACTOSE FERMENTERS: RED/PINK COLONIES

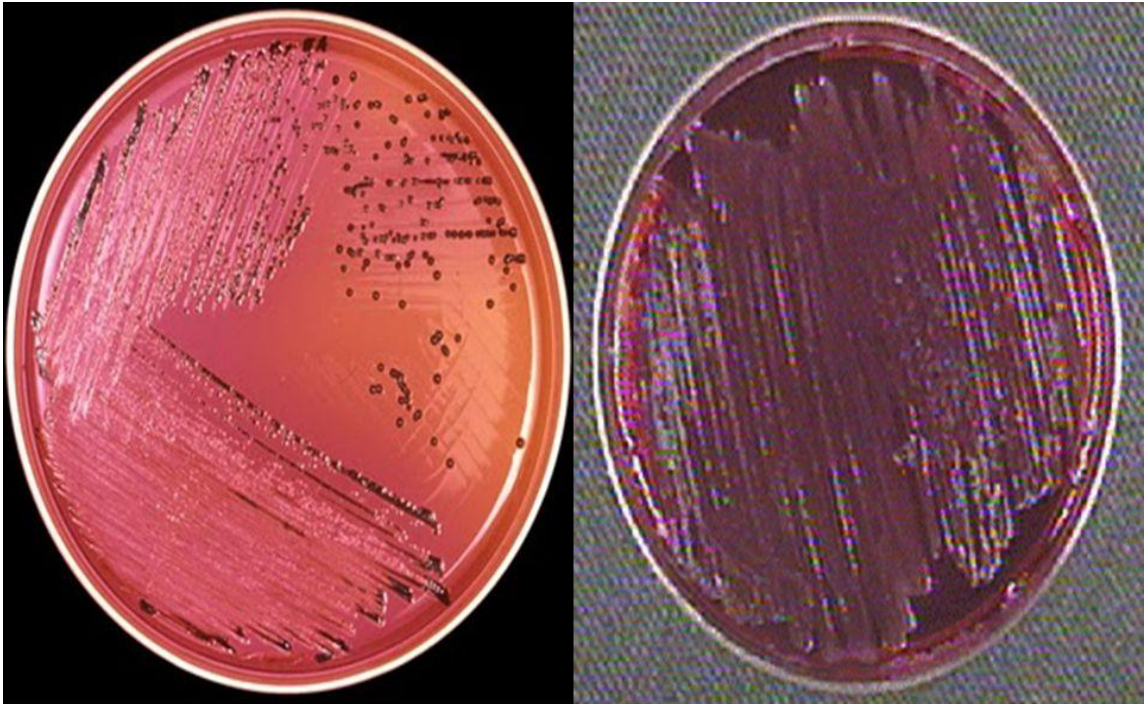


*Salmonella* on SS Agar



*Shigella* on SS Agar





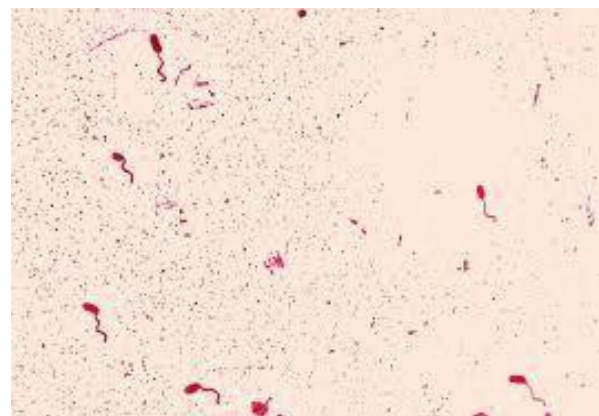
Salmonella on XLD agar

Shigella on XLD agar

- ***Vibrio* spp.**

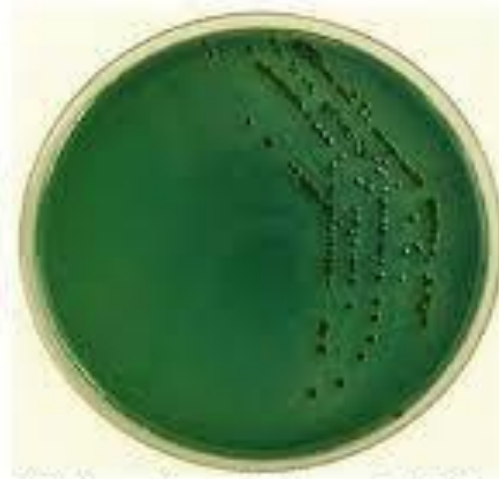
*Vibrio* is a G-ve bacteria, appear as curved rod (comma-shape), facultative anaerobic. Typically found in salt water, and transmitted by contaminated water and cause cholera . Non-spore former, motile, oxidase (+ve)

This genus includes: *V.cholerae.*, *V.paraahaemolyticus.*, and *V.vulnificus.*





*Vibrio cholerae* on TCBS Agar



*Vibrio parahaemolyticus* on TCBS Agar