

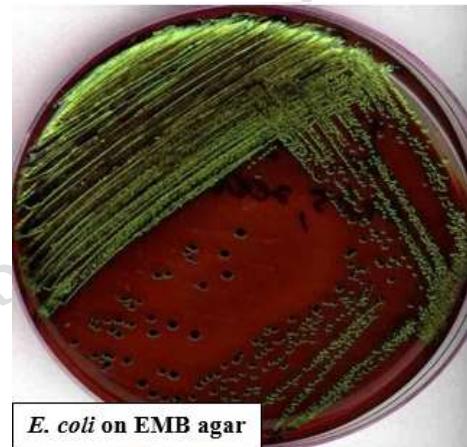


Lecture-15: Family of *Enterobacterceae*

Gram-negative rods related to the enteric tract include a large number of genera.

Diseases Caused by Members of the Enterobacteriaceae

Escherichia: It causes urinary tract infection, traveler's diarrhea, neonatal meningitis. Produces green metallic sheen colonies on Eosin Methylene blue (EMB).



E. coli on EMB agar

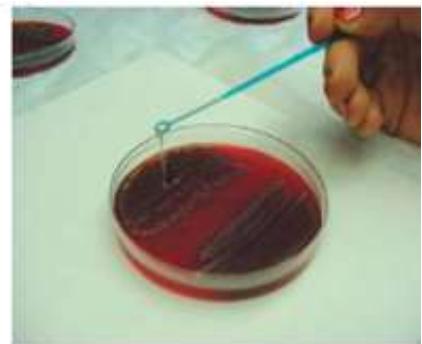
Shigella: The causative agent of **Dysentery**, non-motile. Lactose (-) on MacConkey.

Salmonella: The causative agent of **typhoid fever** and **enterocolitis**.

Klebsiella: Causes Pneumonia, urinary tract infection. **It produces heavy mucoid colonies.**

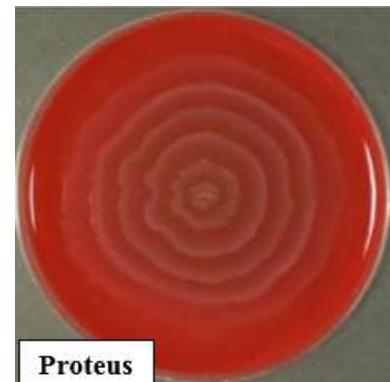


Klebsiella pneumoniae



Proteus: commonly causes urinary tract infection.
Forms Swarming growth on culture media.

Blood agar: Swarming effect over plate as Proteus is active in motility as shown in this figure



Serratia – causes pneumonia and urinary tract infection, forms red colonies on solid media:



Yersinia: The causative agent of **Plague, enterocolitis, mesenteric adenitis.**

Diagnosis:

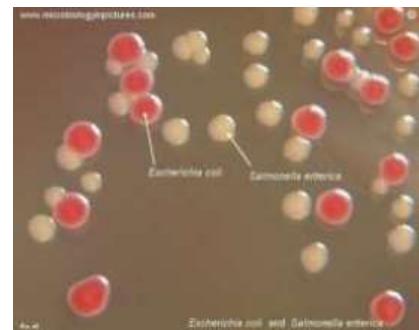
Culture Media

Specimens have suspended in broth and cultured on ordinary as well as differential media (**MacConkey agar**, EMB agar) to permit separation of non-lactose fermenting gram-negative rods from other enteric bacteria. If salmonella infection has suspected, the specimen has also placed in an enrichment medium (**selenite broth**) for 18 hours before has plated on differential media (**Hektoen enteric or Shigella- Salmonella agar, S-S agar**).

Identificatio

MacConkey agar is inoculated with tested organism using streak plate technique. Incubate the plate in incubator at 37 C for 24 hrs., then read the results as the following:

- Lactose fermenting (LF) organism appears as **pink colonies** (e.g. *E. coli* and *Klebsiella*)
- Non lactose fermenting (NLF) organism appears as **colorless colonies** (*Salmonella* and *Shigella*).



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Key Characteristics to differentiate some group of *Enterobacteriaceae*

<u>Bacteria</u>	<i>E. coli</i>	<i>Shigella sonnei</i>	<i>Salmonella typhi</i>	<i>Klebsiella pneumoniae</i>	<i>Klebsiella oxytoca</i>	<i>Proteus vulgaris</i>	<i>Proteus mirabilis</i>	<i>Morganella morganii</i>
<u>Test</u>								
Indole	+	-	-	-	-	+	-	+
Methyl Red (MR)	+	+	+	v	-(v)	+	+	+
VogesProskauer (VP)	-	-	-	+	+	-	v	-
Simmons' Citrate	-	-	-	+	+	-(v)	+(v)	-
Hydrogen Sulfide (H₂S)	-	-	+w	-	-	+	+	-
Urea	-	-	-	+	+	+	+	+
Motility	v	-	+	-	-	+	+	v
Gas from D- glucose	+	-	-	+	+	+	+	+
Lactose	+	-	-	+	+	-	-	-

Neisseria meningitidis: Family: *Neisseriaceae*: Genus: *Neisseria*.

N. meningitidis is aerobic, gram-negative cocci typically arranged in **pairs (diplococci)** with adjacent sides flattened together (**resembling coffee beans**).

Specimens collection

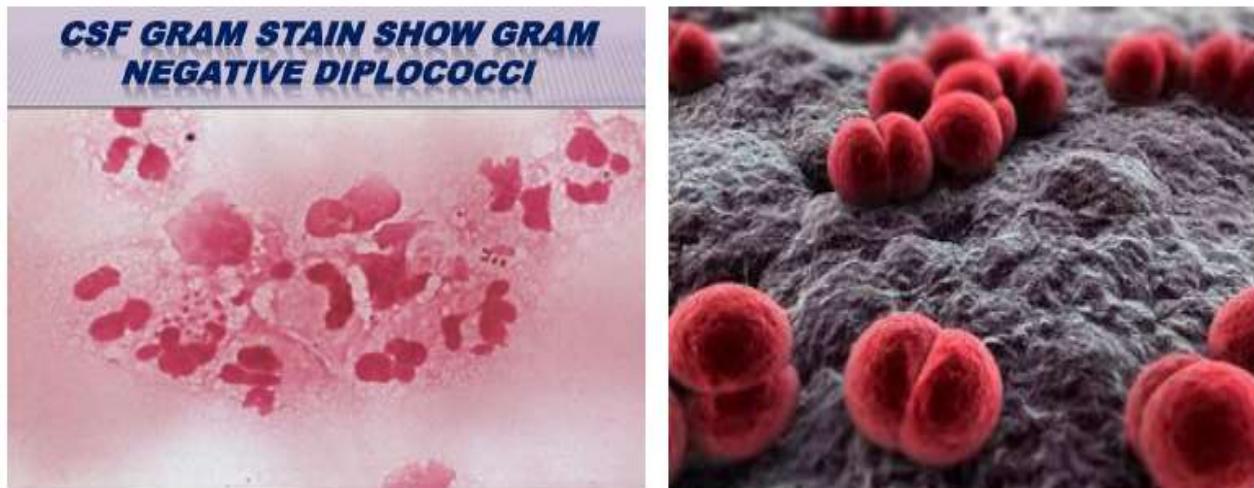
- Nasopharyngeal swabs; body fluids (**joint** fluid or **CSF**) should be stored at 37°C because it was sensitive to cold.
- Any volume (greater than 1 ml) of clear body fluid should be centrifuged at room temperature at 1500xg for 15 min. the sediment should be vortexed and inoculated onto appropriate media.

Diagnosis:

a) Direct detection methods

1- By Gram stain

As indicated above, *N. meningitidis* is **Gram negative diplococci** with adjacent sides flattened. They are often referred to as (**Kidney bean**) shaped diplococci.



N. meningitidis by electron microscope

2- Antigen detection

The detection of *Neisseria meningitidis* **capsular polysaccharides antigen** in body fluids is no longer recommended.

b) Cultivation

The culture media used for *Neisseria* **5% sheep blood agar** and **chocolate agars**. Colonies of *N. meningitidis* are grey and unpigmented on a blood agar and appear round, smooth, moist, shiny, and convex, with a clearly defined edge. *N. meningitidis* appear as large, colorless-to-grey, opaque colonies on a chocolate agar.

N. gonorrhoeae, *N. meningitidis*, and *M. catarrhalis* grow best under conditions of increased CO₂ (3% to 7%).

Colonial appearance (morphology)

N. meningitidis colonies are **medium, smooth, round, moist, gray to white**; **encapsulated strains are mucoid**; may be greenish cast in agar underneath colonies.

Biochemical identification

Test	Growth on			Rapid fermentation		Nitrate reductions	Gas from Nitrate reduction
	Modified Thayer-Martin	Nutrient Agar at 35°C	Blood or Chocolate Agar at 25°C	sugars			
Organism				Glucose	Maltose	Lactose	
<i>N. gonorrhoeae</i>	+	-	-	+	-	-	-
<i>N. meningitidis</i>	+	-	-	+	+	-	-