Family: Vibrioaceae

Phylum: Proteobacteria

Class: Gammaproteobacteria

Order: Vibrionales

Family: Vibrioaceae

Genus: Vibrio

V. cholerae V. parahaemolyticus

General Characteristics : V. cholerae is a comma-shaped, curved rod. It is actively motile by means of a polar flagellum. On prolonged cultivation, vibrios may become straight rods that resemble the gram-negative enteric bacteria. Vibrios are among the most common bacteria in surface waters worldwide. They are curved aerobic rods and are motile, possessing a polar flagellum. V. cholerae serogroups O1 and O139 cause cholera in humans, while other vibrios may cause sepsis or enteritis. V. cholerae regularly ferments sucrose and mannose but not arabinose. Vibrios are oxidase-positive, which differentiates them from enteric gram-negative bacteria. Characteristically, vibrios grow at a very high pH (8.5-9.5) and are rapidly killed by acid. Most Vibrio species are halotolerant, and NaCl often stimulates their growth. Some vibrios are halophilic, requiring the presence of NaCl to grow.

Culture and Growth Characteristics: Growth is rapid in peptone agar, on blood agar with a pH near 9.0, or on TCBS agar, and typical colonies can be picked in 18 hours. For enrichment, a few drops of stool can be incubated for 6-8 hours in taurocholate-peptone broth (pH 8.0-9.0); organisms from this culture can be stained or subcultured. V. cholerae produces convex, smooth, round colonies that are opaque and granular in transmitted light. V. cholerae and most other vibrios grow well at 37 °C on many kinds of media, including defined media containing mineral salts and asparagine as sources of carbon and nitrogen. V. cholerae grows well on thiosulfatecitrate-bile-sucrose (TCBS) agar, on which it produces yellow colonies that are readily visible against the dark-green background of the agar.

Antigenic Structure & Biologic Classification:

V. cholerae has O lipopolysaccharides that confer serologic specificity. There are at least 139 O antigen groups. V. cholerae strains of O group 1 and O group 139 cause classic cholera; occasionally, non-O1/non-O139 V. cholerae causes cholera-like disease. The V. cholerae serogroup O1 antigen has determinants that make possible further typing Two biotypes of epidemic V. cholerae have been defined, classical and El Tor.

El Tor	Classical
Haemolytic	Non-Haemolytic
Resistant to polymyxin B.	Sensitive
Cause haemoagglutination of SRBCs	Dose not

Molecular techniques can also be used to type V. cholerae.

The Medically Important Vibrios.		
Organism	Human Disease	
V. cholerae serogroups O1 and O139	Epidemic and pandemic cholera	
V. cholerae serogroups non-O1/non-O139	Cholera-like diarrhea; mild diarrhea; rarely, extraintestinal infection	
V. parahaemolyticus	Gastroenteritis, perhaps extraintestinal infection	
Others V. mimicus, V. vulnificus, V. hollisae, V. fluvialis, V. damsela, V. anginolyticus, V. metschnikovii	Ear, wound, soft tissue, and other extraintestinal infections, all uncommon	

Pathogenesis:

Vibrio parahaemolyticus: is a halophilic bacterium that causes acute gastroenteritis following ingestion of contaminated seafood such as raw fish or shellfish. After an incubation period of 12-24 hours, nausea and vomiting, abdominal cramps, fever, and watery to bloody diarrhea occur. Fecal leukocytes are often observed. The enteritis tends to subside spontaneously in 1-4 days with no treatment other than restoration of water and electrolyte balance. The disease occurs worldwide, with highest incidence in areas where people eat raw seafood. V parahaemolyticus does grow well on blood agar. It also grows well on TCBS, where it yields green colonies.

Specimens: Specimens for culture consist of mucus flecks from stools.

Treatment: The most important part of therapy consists of water and electrolyte replacement to correct the severe dehydration and salt depletion. Many antimicrobial agents are effective against V cholerae. Oral tetracycline tends to reduce stool output in cholera and shortens the period of excretion of vibrios.

Laboratory diagnostic tests:

- 1. Gram stain (Gram-negative comma-shaped, curved rod).
- 2. Culture on blood agar
- 3. Culture on Selective and Differential media like TCBS agar.
- 4. V. cholerae organisms are identified by slide agglutination tests using anti-O group 1 or group 139 antisera
- 5. TSI
- 6. IMViC test
- 7. Nitrate reduction test
- 8. Gelatin liquefaction test.
- 9. Peptone water pH= 9, NaCl= 7%
- 10. Cholera Red Reaction (Nitroso endol test): to test the bacteria ability to produce deaminase enzyme

Tryptophane + H₂SO₄ (4 drops) Nitrosoindol (red color)

Deaminase

Tryptophane _____ NH₃

 $NH_3 + H_2SO_4 \longrightarrow Nitrosoindol (red color)$

14. String test: add 0.5% Na-deoxycholate solution to 1 drop of culture, the culture convert to thread like when draws by loop disappear after 45-60 sec.

Test	V. cholera	V. parahaemolyticus
Catalase	+	+
Oxidase	+	+
NO ₃ reduction	+	+
Indole	+	+
MR	+ weak	
VP	-	
Citrate	+/-	+/-
Peptone water + 7% NaCl	-	+
Peptone water + 0% NaCl	+	
TSI	A/A	K/A
Motility	+	+
Cholera red	+	
Mannitol	+ weak	+ weak
String test	+	+
OF medium	Oxidation-fermentation	Oxidation-fermentation
TCBS medium	Yellow colonies	Green colonies



Vibrio cholerae



The String Test



TCBS medium



Vibrio parahaemolyticus on TCBS medium





Vibrio cholerae on TCBS medium

Family: Neisseriaceae

Phylum: Proteobacteria

Class: Betaproteobacteria

Order: Neisseriales

Family: Neisseriaceae

Genus I: Neisseria Genus II: Kingella Genus III: Eikenella GenusIV: Simonsiella GenusV: Alysiella

Neisseria

General Characteristics: is a gram-negative, nonmotile diplococcus, Individual cocci are kidney-shaped; when the organisms occur in pairs, the flat or concave sides are adjacent. Neisseria gonorrhoeae (gonococci) and Neisseria meningitidis (meningococci) are pathogenic for humans and typically are found associated with or inside polymorphonuclear cells. Some neisseriae are normal inhabitants of the human respiratory tract, rarely if ever cause disease, Meningococci have polysaccharide capsules, whereas gonococci do not, and meningococci rarely have plasmids whereas most gonococci do. The neisseriae produce oxidase and give positive oxidase reactions; the oxidase test is a key test for identifying them.

Culture and Growth Characteristics: In 48 hours on enriched media (eg, Mueller-Hinton, modified Thayer-Martin, "chocolate" agar), gonococci and meningococci form convex, glistening, elevated, mucoid colonies 1–5 mm in diameter. Colonies are transparent or opaque, nonpigmented, and nonhemolytic. The neisseriae grow best under aerobic conditions, but some will grow in an anaerobic environment. They have complex growth requirements. Most neisseriae ferment carbohydrates, producing acid but not gas, and their carbohydrate fermentation patterns are a means of distinguishing them. Meningococci and gonococci grow best on media containing complex organic substances such as heated blood, hemin, and animal proteins and in an atmosphere containing 5% CO₂ (e.g, candle jar). To avoid overgrowth by contaminants, the selective medium contains antimicrobial drugs (eg, vancomycin, colistin, amphotericin B, and trimethoprim). A modified Thayer-Martin medium with antibiotics (vancomycin, colistin, amphotericin) favors the growth of neisseriae, inhibits many other bacteria, and is used for nasopharyngeal cultures. The organisms are rapidly killed by drying, sunlight, moist heat, and many disinfectants. They produce autolytic enzymes that result in rapid swelling and lysis in vitro at 25 °C and at an alkaline pH. Gonococci ferment only glucose and differ antigenically from the other neisseriae. Gonococci usually produce smaller colonies than those of the other neisseriae

Pathogenesis:

Neisseria meningitidis (Meningococci) typically are found in the upper respiratory tract and cause meningitis,

while *Neisseria gonorrhoeae* (gonococci) cause genital infections (gonorrhoeal disease, sexual transmitted disease

Specimens: Pus and secretions are taken from the urethra, cervix, rectum, conjunctiva, throat, and spinal fluid for culture and smear. Blood culture is necessary in systemic illness.

Laboratory diagnostic tests:

- 1. Gram stain (G-ve diplococci).
- **2. Enriched media** (eg, Mueller-Hinton, modified Thayer-Martin, "chocolate" agar).
- 3. Oxidase test (+ve)
- 4. Carbohydrate fermentation.
- 5. Nitrate reduction test.



Neisseria gram-negative cocci

Test	Neisseria gonorrhoeae	Neisseria meningitidis
Glucose	+	+
Maltose		+
Fructose	-	_
Lactose		-
Pigments	Greyish white	Greyish white
CO ₂ requirement	Necessary	Necessary
Growth at 22°C		_
Growth at 35°C	+	+
NO ₃ reduction		+

Pathogenic bacteria	Biology department
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