

Haemophilus influenzae

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Haemophilus influenzae (formerly called Pfeiffer's bacillus or *Bacillus influenzae*) is a Gram-negative, coccobacillary, non-motile, facultatively anaerobic bacterium belonging to the *Pasteurellaceae* family. *H. influenzae* was first described in 1892 by Richard Pfeiffer during an influenza pandemic. The bacterium was mistakenly considered to be the cause of influenza until 1933 when the viral etiology of influenza became apparent, and is still known as 'bacterial influenza'. *H. influenzae* is responsible for a wide range of localized and invasive infections.

Domain:	Bacteria
Kingdom:	Eubacteria
Phylum:	Proteobacteria
Class:	Gammaproteobacteria
Order:	Pasteurellales
Family:	Pasteurellaceae
Genus:	<i>Haemophilus</i>
Species:	<i>H. influenzae</i>

Diseases

H.influenzae the leading cause of meningitis in young children ,it is still an important cause of upper respiratory tract infection (otitis media ,sinusitis ,and epiglottitis) ,causes sepsis in children also causes pneumonia in adults .

Important properties

-*H.influenzae* is small ,rod coccobacillus polysaccharide capsule

-Type b capsule is composed of polyribitol phosphate

-Growth of the organism on laboratory media requires the addition of two components hemin (factor X) and NAD (factor V) for adequate energy production

Pathogenesis

H.influenzae infect only humans , there is no animal reservoir ,it is enter the body through the upper respiratory tract ,the organism produce IgA protease that degrades secretory IgA thus facilitating attachment to the respiratory mucosa ,then enter the blood stream (bacteremia) and spread to the meninges ,pathogenesis involve antiphagocytic capsule and endotoxin ,no exotoxin is produced

Diagnosis

Gram stain

Direct Examination ,Perform Gram staining and microscopic observation: Gram negative pleomorphic thin rods or coccobacilli.

Culture

Inoculate samples onto chocolate agar media ,Incubate at 37°C in aerobic atmosphere containing 5-10% CO₂ for 24-48 hours.

Colony morphology

On chocolate agar: large flat, colorless to gray or opaque colonies. Colonies are 0.5 – 1mm circular, low convex, smooth, pale grey and transparent. No haemolysis or discolouration is seen.

Encapsulated strains appear more mucoid (watery) and non capsulated strains appear as compact grayish colonies.

Note: *H. influenzae* is a facultative anaerobe requiring growth factors X and V in its culture medium. The X factor is hemin, required by the bacteria to synthesize enzymes containing heme (cytochromes, catalase, oxidases). The X factor requirement is greatly reduced in anaerobic culturing. The V factor was identified as NAD or NADP. A standard blood agar plate does not contain sufficient free V factor. Some bacteria, in particular *Staphylococcus aureus*, produce excess NAD and even secrete this coenzyme into the medium. That is why *H. influenzae* can proliferate in the immediate vicinity of *S. aureus* colonies. This is known as the satellite phenomenon. The medium normally used to culture *H. influenzae* is chocolate agar containing sufficient amounts of the X and V factors.

Biochemical reactions for differentiation.

-oxidase test is positive,

-Confirmatory tests for X and V factor requirements. Inoculate a single suspected colony from chocolate agar onto Mueller Hinton agar plates. Place commercially available X, V, and XV factor discs/strips on streaked plates. Incubate plates at 37°C in 5-10% CO₂ atmosphere for 18-24 hours.

Observe growth around the discs and *H. influenzae* will only grow around the combined XV disc.

Note that if only X and V factor discs (without XV) are applied, place them at least 2 cm apart and *H. influenzae* will grow between the two discs.

-Latex particle agglutination

The latex particle agglutination test (LAT) is a more sensitive method to detect *H. influenzae* than culture. Because the method relies on antigen rather than viable bacteria,. It also has the added benefit of being much quicker than culture methods.

- Agglutinating antisera for serotypes “a” to “f” are available commercially. Such sera contain antibodies directed towards somatic antigens present in patient’s sera which result in agglutination.
- Apply one drop of normal saline on a slide and make a homogenous suspension with a single suspected colony of *H. influenzae*.
- Add one drop of specific antiserum and mix thoroughly.

Reading

- Observe for agglutination (visible clumping) within 1 minute.

Interpretation.

- A visible clumping within 1 minute is indicative of a positive reaction.

Molecular methods

Polymerase chain reaction (PCR) assays have been proven to be more sensitive than either LAT or culture tests, and highly specific. However, PCR assays have not yet become routine in clinical settings

Treatment

Haemophilus influenzae produces beta-lactamases, and it is also able to modify its penicillin-binding proteins, so it has gained resistance to the penicillin family of antibiotics. In severe cases, cefotaxime and ceftriaxone , and for the less severe cases, an association of ampicillin and sulbactam, cephalosporins of the second and third generation, or fluoroquinolones are preferred. (Fluoroquinolone-resistant *Haemophilus influenzae* has been observed.) Macrolide antibiotics (e.g., clarithromycin) may be used in patients with a history of allergy to beta-lactam antibiotics.

Prevention

Effective vaccines for *Haemophilus influenzae* Type B have been available since the early 1990 .The World Health Organization recommends a pentavalent vaccine, combining vaccines against diphtheria, tetanus, pertussis, hepatitis B and Hib.