Haemophilus spp.

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**Haemophilus influenzae** is a small, nonmotile Gram-negative bacterium in the family *Pasteurellaceae*. Encapsulated strains of *Haemophilus influenzae* isolated from cerebrospinal fluid are coccobacilli, 0.2 to 0.3 to 0.5 to 0.8 um, similar in morphology to *Bordetella pertussis*, the agent of whooping cough.

Non encapsulated organisms from sputum are pleomorphic and often exhibit long threads and filaments.
**Haemophilus influenzae**-

- Uncapsulated
  - normal flora of URT & noninvasive disease
- Capsulated
  - Polysaccharide capsule responsible for invasiveness
- There are six serotypes based on antigenicity of capsular polysaccharide.
  - These are a, b, c, d, e, f
- Of the 6 serotypes, only *H. influenzae* type b is responsible for most of the invasive diseases (Hib), and vaccine is available only against Hib.
• *H. influenzae* is highly adapted to its human host. It is present in the nasopharynx of approximately 75 percent of healthy children and adults.
• It is rarely encountered in the oral cavity and it has not been detected in any other animal species.
• It is usually the non encapsulated strains that are harbored as normal flora, but a minority of healthy individuals (3-7 percent) intermittently harbor *H. influenzae* type b (Hib) encapsulated strains in the upper respiratory tract.
• Pharyngeal carriage of Hib is important in the transmission of the bacterium. The success of current vaccination programs against Hib is due in part to the effect of vaccination on decreasing carriage of the organism.
What's in a name?

- *Haemophilus influenzae* is widespread in its distribution among the human population.
- It was first isolated by Pfeiffer during the influenza pandemic of 1890.
- It was mistakenly thought to be the cause of the disease influenza, and it was named accordingly. Probably, *H. influenzae* was an important secondary invader to the influenza virus in the 1890 pandemic, as it has been during many subsequent influenza epidemics.
Haemophilus "loves heme", more specifically it requires a precursor of heme in order to grow.

Nutritionally, *H. aemophilus influenzae* prefers a complex medium and requires preformed growth factors that are present in blood, specifically **X factor** (i.e., hemin) and **V factor** (NAD or NADP).
Cultural properties

• In the laboratory it is usually grown on chocolate blood agar which is prepared by adding blood to an agar base at 80 degrees. The heat releases X and V factors from the RBCs and turns the medium a chocolate brown color.

• The bacterium grows best at 35-37 degrees and has an optimal pH of 7.6. in the laboratory under slight CO2 tension (5% CO2) or under aerobic conditions.
In 1995, *Haemophilus influenzae* was the first free-living organism to have its entire chromosome sequenced, sneaking in just ahead of *Escherichia coli* in that race, mainly because its genome is smaller in size than *E. coli*'s. For a relatively obscure bacterium, there was already a good understanding of its genetic processes, especially transformation.
• Observations of genetic transformation in *Haemophilus* have included drug resistance and synthesis of specific capsular antigens. The latter is thought to be the main determinant of *H. influenzae*.

• Transformation in *Haemophilus influenzae* occurs by several different mechanisms and is more efficient than in enteric bacteria. When developing competence, the bacterium develops membranous "blebs" in the outer membrane that contain a specific DNA-binding protein. This outer membrane protein recognizes a specific 11-base pair sequence of DNA nucleotides that appears in *Haemophilus* DNA with much higher frequency than in other genera of bacteria. There is some evidence that *Haemophilus* is able to undergo both interspecies and intraspecies transformation in vivo (in host tissues). The restriction endonucleases from *Haemophilus*, e.g. *Hind III*, are widely used in biotechnology and in the analysis and cloning of DNA.
1. **Capsule:** The polyribosyl ribitol phosphate (PRP) capsule is the most important virulence factor because it renders type b *H. influenzae* resistant to phagocytosis by polymorphonuclear leukocytes.

2. **Fimbriae** increase the adherence of bacteria to human mucosal cells in vitro, and they are required for successful colonization of the nasopharynx.

3. **Neuraminidase** and an **IgA protease**, are produced by all virulent strains, though role in pathogenesis is unclear.

4. **Endotoxin** in meningitis or bacteremia is unclear,

5. outer membrane **lipooligosaccharide** is thought to play a role in inflammation associated with otitis media.
How do people get Hib disease?

- Hib disease is spread through contact with discharges or droplets from the nose or throat of an infected person. Hib disease can spread from person to person through sneezing, coughing, or speaking closely with an infected person. A person does not have to have symptoms to spread the bacterium.
What are the signs and symptoms of Hib disease?

- The most common and severe manifestation of Hib disease is meningitis (inflammation and swelling in the coverings of the brain and spinal cord). Symptoms of meningitis include fever, weakness, vomiting, and a stiff neck. Hib can also cause infection of the lungs, blood, joints, bones, throat, and covering of the heart. Symptoms depend on the part of the body affected.
The pathogenesis of *H. influenzae* infections is not completely understood,

The presence of the **type b polysaccharide capsule** is known to be the major factor in virulence.

Encapsulated organisms can penetrate the epithelium of the nasopharynx and invade the blood capillaries directly. Their capsule allows them to resist phagocytosis and complement-mediated lysis in the nonimmune host. Nontypable (non encapsulated) strains are less invasive, but they are apparently able to induce an inflammatory response that causes disease.

Outbreaks of *H. influenzae* type b infection may occur in nurseries and child care centers, and prophylactic administration of antibiotics is warranted.

Vaccination with type b polysaccharide (in the form of **Hib conjugate vaccines**) is effective in preventing infection, and several vaccines are now available for routine use.
Diseases

**Type b**
- Meningitis
  - CSF 50%-95% culture positive
  - Blood 50%-95% culture positive
- Conjunctivitis
  - Eye 50%-75% culture positive
  - Blood < 10% culture positive
- Cellulitis
  - Skin 75% - 90% culture positive
  - Blood 50% - 75% culture positive
- Epiglottitis
  - Blood 90% - 95% culture positive
  - Epiglottitis culture contraindicated
- Arthritis
  - Synovial fluid
    - 70% - 90% culture positive
  - Blood
    - 50% - 80% culture positive

**Unencapsulated**
- Otitis Media
  - Tympanocentesis
    - 50% - 70% culture positive
- Sinusitis
  - Sinus aspirate
    - 50% - 75% culture positive
- Pneumonia, bronchitis
  - Sputum
    - 20% - 75% culture positive
  - Blood
    - 10% - 30% culture positive

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• The age incidence of *H. influenzae* meningitis is inversely proportional to the titer of bactericidal antibody in the blood, whether passively acquired from the mother or actively formed (see Figure 5 below).

• Without artificial immunization, in children aged 2 months to 3 years, antibody levels are minimal; thereafter antibody levels increase and the disease becomes much less common.
Figure 5. Relation of the age incidence of bacterial meningitis caused by *Haemophilus influenzae* to bactericidal antibody titers in the blood (data pre 1985)

From this curve, it is obvious that artificial active immunization should begin at 2 months of age, when nearly all passive immunity has waned, and the child enters a vulnerable non-immune period of life.
Who is at risk for Hib disease?

- Unvaccinated children under age 5 are at risk for Hib disease. Hib disease occurs most often in children ages 3 months to 2 years. As children grow older, they are less likely to develop Hib disease. Very few cases occur in persons over age 5.
Treatment

• Virtually all patients treated early in the course of *H. influenzae* meningitis are cured. The mortality rate of treated infections is less than 10 percent, but nearly 30 percent of the children who recover have residual neurologic effects.

• Third-generation cephalosporin (ceftiraxone or cefotaxime, are effective against *H. influenzae* and penetrate the meninges well.

• Amoxicillin+ clavulanic acid for treatment of other URTI
• Vaccination against Hib
  – There are several types of Hib conjugate vaccines available for use. All of the vaccines are approved for use in children 15 months of age and older and some are approved for use in children beginning at 2 months of age. All of the vaccines are considered effective. The vaccines are given by injections. More than 90% of infants obtain long term immunity with 2-3 doses of the vaccine.
  • All children should have a vaccine approved for infants beginning at 2 months.
  • All unvaccinated children 15 - 59 months old should receive a single dose of conjugate vaccine.
  • Children 60 months of age or older and adults normally do not need to be immunized.
Sputum
chains of *Haemophilus influenzae*
Acute Epiglottitis

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Prevalence of bacterial causes of common infections

<table>
<thead>
<tr>
<th>Bacteria</th>
<th>Pneumoniae</th>
<th>Otitis media</th>
<th>Sinusitis</th>
<th>Meningitis</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Streptococcus pneumoniae</em></td>
<td>8.4%</td>
<td>40%</td>
<td>30%</td>
<td>35%</td>
</tr>
<tr>
<td><em>Legionella</em> species</td>
<td>6%</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td><em>Haemophilus influenzae</em></td>
<td>5.3%</td>
<td>25%</td>
<td>20%</td>
<td>10%</td>
</tr>
<tr>
<td><em>Mycoplasma pneumoniae</em></td>
<td>3.6%</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td><em>Moraxella (Branhamella)</em></td>
<td>--</td>
<td>10%</td>
<td>20%</td>
<td>--</td>
</tr>
<tr>
<td><em>Catarrhalis</em></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Neisseria meningitidis</em></td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>35%</td>
</tr>
<tr>
<td><em>Mycobacterium tuberculosis</em></td>
<td>1.5%</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>Unknown</td>
<td>43%</td>
<td>25%</td>
<td>30%</td>
<td>--</td>
</tr>
</tbody>
</table>

*Percentages do not add up to 100% in some cases because not all species are included.*
Relation of the age incidence of *H. influenzae* meningitis to bactericidal antibody titers in the blood (similar in meningococcal meningitis)
<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Trade Name</th>
<th>Manufacturer</th>
<th>Protein Carrier</th>
</tr>
</thead>
<tbody>
<tr>
<td>HbOC</td>
<td>HibTITER</td>
<td>Wyeth</td>
<td>CRM$_{197}$ (a nontoxic mutant of diphtheria toxin)</td>
</tr>
<tr>
<td>PRP - OMP *</td>
<td>PedvaxHIB</td>
<td>Merck</td>
<td>OMP (an outer membrane protein complex of <em>Neisseria meningitidis</em>)</td>
</tr>
<tr>
<td>PRP - T †</td>
<td>ActHIB</td>
<td>Aventis Pasteur</td>
<td>Tetanus toxoid</td>
</tr>
<tr>
<td></td>
<td>OmniHIB</td>
<td>Aventis Pasteur ‡</td>
<td>Tetanus toxoid</td>
</tr>
</tbody>
</table>

* PRP-OMP is also available as a combination vaccine with hepatitis B vaccine (Comvax). This should not be used for hepatitis B immunization at birth.
† PRP-T can be reconstituted with Connaught DTaP vaccine (Tripedia), to produce a combination marketed as TriHIBit, which is acceptable only for the booster (4th) dose in infants ≥ 15 mo of age.
‡ Marketed by GlaxoSmithKline in the United States.
“Do not walk proudly on the earth; your feet cannot tear apart the earth nor are you as tall as the mountains.”

- *Quran* (17:37)

*Muslim Speakers*