Plague

- Plague is a vector-borne infectious disease caused by the bacteria known as *Yersinia pestis*.
- Plague has a high fatality rate, and people have described outbreaks of the bacterial infection for centuries.
- In the Middle Ages, plague was known as the "Black Death." It caused the death of 60% of the population of Europe during a pandemic (an epidemic of human disease that has spread through a large geographic area).
- Transmission occurs via fleas that feed on infected animals, typically wild rodents.
- There are three forms of plague in humans: bubonic plague, septicemic plague, and pneumonic plague.
- The signs and symptoms of plague generally develop between two and seven days after a person acquires the infection.

Symptoms and signs depend on the type of plague and include the following:

1. Bubonic plague symptoms and signs include painful and enlarged or swollen lymph nodes (an enlarged lymph node due to plague is called a bubo), chills, headache, fever, fatigue, and weakness.
2. Septicemic plague (Black Death or black plague) symptoms and signs include fever, weakness, abdominal pain, chills, and shock. Tissue bleeding and death may cause the dying tissues to appear black.
3. Pneumonic plague symptoms and signs include characteristic pneumonia symptoms like chest pain, shortness of breath, cough, fever, chills, nausea, vomiting, and diarrhea.

Causative Organism

- The bacterium *Yersinia pestis* causes plague. In the natural state, the bacteria infect wild rodents. The World Health Organization states that between 1,000-2,000 cases are reported each year worldwide, but there are estimated to be more cases that go unreported. Rat fleas (*Xenopsylla* species) that feed off of infected animals transmit the bacteria to other animals. Rats, ground squirrels, mice, prairie dogs, chipmunks, voles, and rabbits are examples of animals that may carry the plague bacteria. The bacteria are believed to persist at a low level in natural populations of these animals. When a large number of infected wild rodents die, fleas that have bitten these animals may bite humans and domestic animals. Cats that are bitten usually become ill, and they may cough infectious droplets into the surrounding air. While infected dogs may not appear ill, they may still carry infected fleas into the home.

- *Yersinia pestis* is a Gram-negative, a nonmotile, bipolar-staining coccobacillus, slow-growing, aerobic and facultative anaerobic organism classified in the family *Enterobacteriaceae* and is an obligate intracellular pathogen that must be contained within the blood to survive. It is also a fermentative that produces a thick anti-phagocytic slime layer in its path.

Biochemical Test of *Yersinia pestis*

CapsulePositive (+ve)CatalasePositive (+ve)CitrateNegative (-ve)FlagellaNon-FlagellatedGasNegative (-ve)Gelatin HydrolysisNegative (-ve)Gram StainingNegative (-ve)Growth in KCNNegative (-ve)H2SNegative (-ve)HemolysisNegative (-ve)IndoleNegative (-ve)MotilityNon-MotileMR (Methyl Red)Positive (+ve)Nitrate ReductionPositive (+ve)OF (Oxidative-Fermentative)Facultative anaerobesOxidaseNegative (-ve)PigmentNegative (-ve)ShapeRodsSporeNegative (-ve)UreaseNegative (-ve)VP (Voges Proskauer)Negative (-ve)

Fermentation of

AdonitolNegative (-ve)ArabinosePositive (+ve)CellobiosePositive (+ve)DNaseNegative (-ve)DulcitolNegative (-ve)GlucosePositive (+ve)GlycerolVariableLactoseNegative (-ve)MalonateNegative (-ve)MaltosePositive (+ve)MannitolPositive (+ve)MannosePositive (+ve)MelibioseVariableMucateNegative (-ve)MyoInositolNegative (-ve)RaffinoseNegative (-ve)RhamnoseNegative (-ve)SalicinVariableSorbitolVariableSorboseNegative (-ve)SucroseNegative (-ve)TartrateNegative (-ve)TrehalosePositive (+ve)XylosePositive (+ve)

Enzymatic Reactions

Acetate UtilizationNegative (-ve)Aesculine HydrolysisPositive (+ve)Arginine DehydrolaseNegative (-ve)LipaseNegative (-ve)LysineNegative (-ve)ONPG (β-galactosidase)Positive (+ve)Ornithine DecarboxylaseNegative (-ve)Phenylalanine DeaminaseNegative (-ve)

Virulence Factors

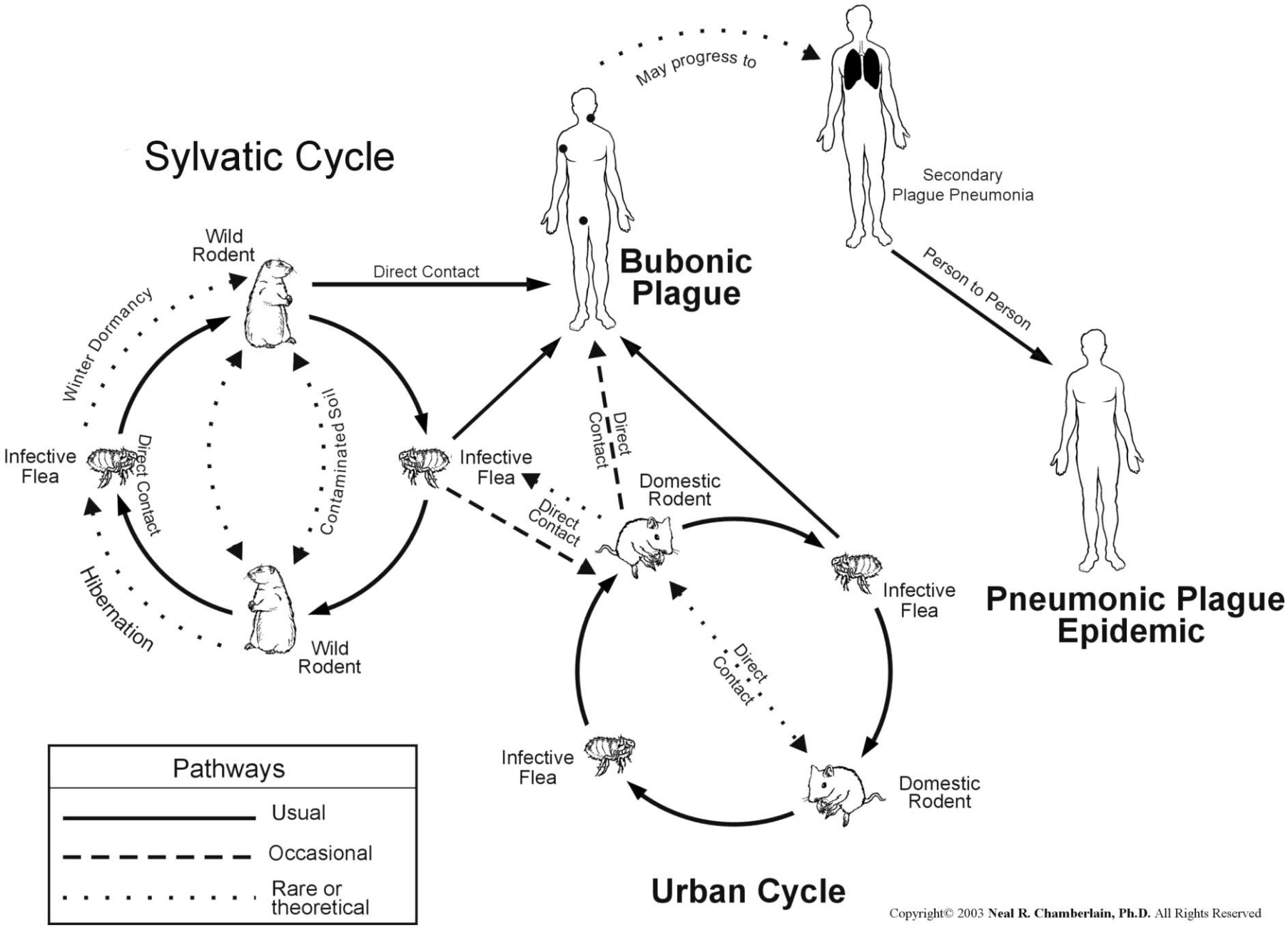
- *Yersinia pestis* encodes two antigenic molecules:
- Fraction 1 (F1) capsular antigen,
- VW antigen.
- Both of these molecules are needed for pathogenicity, and are not expressed at temperatures lower than 37°C. This requirement is the main reason why *Yersinia* is not virulent in fleas, since their body temperature normally levels around 25°C. *Yersinia* is a model for studying Type III Secretion Systems (TTSS) that inject bacterial proteins into a host cell. In *Y. pestis*, it is the translocation of Yersinia outer proteins (Yop's) that blocks the host cell's ability to communicate with immune system cells and down-regulates the response of phagocytic host cells to infection. Through the TTSS, YopH and *Yersinia* protein kinase A (YpkA) are delivered by YopB and YopD into the host cell, where they subvert signal transduction and inhibit oxidative bursts. Also, the rough/short lipopolysaccharide (LPS) chains on the outer membrane of *Yersinia* mediate antibody resistance by causing abnormal attachment of membrane attack complexes (MACs).

Important virulence factors include proteins encoded by three different plasmids:

- When placed at 37^{o} C, low-Ca^{2+} concentrations and in a nutrient rich environment a plasmid (70-kbps called **pYV** or pCD1) encodes the Yop (Yersinia Outer membrane Proteins) virulon and a type III secretion apparatus called Ysc or Yersinia Secretion. There are 29 different Ysc proteins which assemble to form a pore in the inner and outer membrane of the bacteria. Once the bacterium makes contact with a eukaryotic cell certain translocator Yops will form a pore in the eukaryotic cell. Effector Yops then go across the channel formed through the bacterial and eukaryotic membranes and obtain access to the eukaryotic cell's cytoplasm. There are at least 6 different effector Yops which when transported into the eukaryotic cells inhibit phagocytosis, inflammation, and induce apoptosis of macrophages. This plasmid also encodes the V antigen that appears to also be involved in the type III secretion apparatus. The V antigen also appears to have immunosuppressive effects on the host's immune system.
- Virulence is enhanced by another 9.5-kb plasmid (**pPst** or pPCP1) that encodes the outer membrane protein plasminogen activator (Pla). Pla is a protease that interferes with blood coagulation and complement activation pathways.
- Yet another 100-kb plasmid (**pFra** or pMT1) also enhances virulence. It contains the genes for the capsular protein (fraction 1) and a murine toxin. Some believe the capsule enhances resistance to phagocytosis by monocytes.

Transmission of plague

- The disease is caused by the gram-negative bacterium *Yersinia pestis*. It is transmitted from rodent to human by the bite of an infected flea, direct contact with infected animals or their products, or inhalation of contaminated airborne droplets . Once in the human body, the bacteria multiply in the blood and lymph. An important factor in the virulence of *Y. pestis* is its ability to survive and proliferate inside phagocytic cells rather than be killed by them. One of the ways this is accomplished is by the YOPS (yersinal plasmid-encoded outer membrane proteins) that are secreted by the bacterium and act as antiphagocytic proteins to counteract natural defense mechanisms and help the bacteria multiply and disseminate in the host



Pathogenesis

Symptoms and signs of plague usually develop between two and seven days after acquiring the *Yersinia pestis* infection, although they may appear after only one day in cases of exposure to pneumonic plague.

The signs and symptoms of plague may take three forms:

- **1. Bubonic plague**

  In this form of the infection, bacteria infiltrate the lymph nodes, causing enlarged, painful, tender lymph nodes called buboes. Accompanying flu-like symptoms include fever, chills, headaches, and weakness. If not treated, the infection can spread to other areas of the body.In 50-70% of the untreated cases, death follows in 3 to 5 days from toxic conditions caused by the large number of bacilli in the blood.

- **2. Septicemic plague**

  This form of plague is a result of plague bacteria entering the bloodstream. It can occur on its own or it may develop from bubonic plague. Symptoms include fever, chills, weakness, abdominal pain, and shock. There can be bleeding and tissue death, especially of the fingers and toes. These dying tissues may appear black, hence the name Black Death. . The mortality rate for this kind of plague is almost 100% .

- **3. Pneumonic plague**

  In the pneumonic form of the illness, symptoms of other types of plague can be present, but the characteristic clinical picture of pneumonia is present. The plague bacteria spread to the lungs or infect the lungs directly when infected droplets in the air are inhaled. This is the only form of plague that can be transmitted from person to person. Shortness of breath, chest pain, fever, and cough with watery or bloody mucus production are symptoms of pneumonic plague. The mortality rate for this kind of plague is almost 100% if it is not recognized within 12 to 24 hours. Obviously great care must be taken to prevent the spread of airborne infections to personnel taking care of pneumonic plague patients.

Laboratory diagnosis of plague

1. D**irect microscopic examination,culture of the bacterium**,

- Bipolar staining rod (Wright-Giemsa) on direct smear
- Pinpoint colony at 24 h on SBA- *Y. pestis* grows slowly in culture, with only pinpoint, gray-white translucent colonies on SBA after 24 hours, with little or no hemolysis. After 48 to72 hours of incubation, colonies have a raised, irregular "fried egg" appearance under magnification.
- Non-lactose fermenter, may not be visible on MAC or EMB at 24 h
- Oxidase and urease negative
- Catalase positive
- Growth often better at 28^{o}C

2.serological tests

3.PCR for detection of bacteria in infected fleas,

4.phage testing.

Treatment

Treatment is with streptomycin, chloramphenicol, or tetracycline (especially doxycycline), and the fluoroquinolone ciprofloxacin and recovery from the disease gives a good immunity.

Vaccine

- A killed whole cell plague vaccine has been used in the past, but recent studies in animals have shown that this vaccine offers poor protection against pneumonic disease. A live attenuated vaccine is also available. While this vaccine is effective, it retains some virulence and in most countries it is not considered to be suitable for use in humans. Now new vaccine develop improved sub-unit and live attenuated vaccines against plague. A sub-unit vaccine based on the F1- and V-antigens is highly effective against both bubonic and pneumonic plague, when tested in animal models of disease. This vaccine has been used to explore the utility of different intranasal and oral delivery systems.