

Bacterial Pathogenesis

Alice Prince

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How do bacteria cause disease?

The purpose of this lecture is to provide some basic concepts regarding the host-pathogen interaction. Several organisms will be mentioned as examples, but the details of their biology and pathogenesis of specific disease entities will be covered in later lectures and course material.

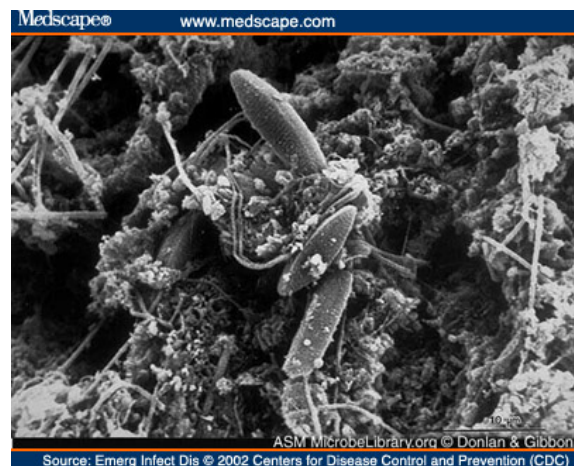
1 – Which bacteria cause disease?

Commensal flora versus pathogenic organisms – Whether specific bacteria are part of the normal flora or are pathogens (i.e.- cause disease) depends upon the host (infant, adult, immunocompromised patient, patient with intravascular catheter, CNS shunt, prosthetic hips or knees etc.); where the organisms are: in a normally sterile site like the blood, lower lung, brain or on a mucosal surface, and the specific virulence properties of the organism.

For example – *Staphylococcus epidermidis*, one of the coagulase – negative staphylococci, is a normal resident of the skin – but a pathogen when it contaminates a heart valve post-operatively and causes endocarditis. *Neisseria gonorrhoea* is a pathogen and should not be considered "normal flora" when cultured from a mucosal surface. *Escherichia coli* is part of the commensal flora of the GI tract, but is a pathogen if there is a major breach in the mucosal barrier that enables the organisms to enter the blood stream. Alternatively, some *E. coli* acquire specific virulence genes – such as the toxin that causes hemolytic uremic syndrome (*E. coli* O157:H7). These *E. coli* cause disease by colonizing the gut and secreting toxin that targets the micro vasculature of the kidneys and platelets.

Staph Infection - Scanning EM

2- **Bacterial life styles** – Bacteria have preferred lifestyles; some grow best in the presence of oxygen, others are strictly anaerobic, and some are facultative, in that they can adapt to different amounts of oxygen. Some bacteria are EXTRAcellular; others prefer to replicate within the host cells such as *M. tuberculosis* and *Listeria*.



Many pathogens are able to sense the environment and rapidly adapt through the activity of two-component regulatory systems. A sensor, that responds to divalent cations (or another factor) activates a kinase that phosphorylates specific transcription factors, which then activate (or repress) a group of genes. This "coordinate regulation" enables the organism to express an entire cascade of genes in response to the conditions sensed in the host.

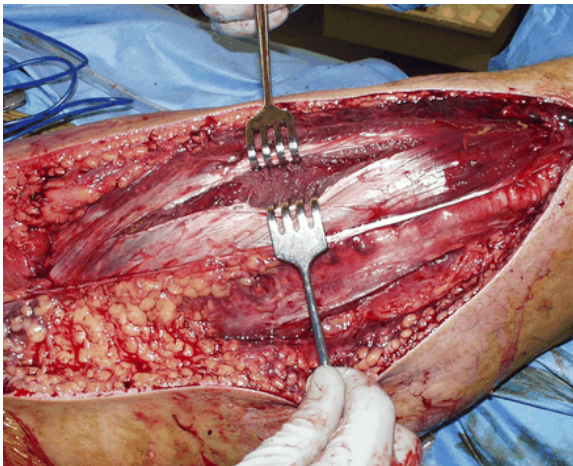
For example: when you ingest *Salmonella* contaminated food at the local salad bar, the organisms that survive the acidity of the stomach (not much since you are taking antacids to cope with the stress of medical school); may be ingested by macrophages. However, the salmonella sense the intracellular environment in the macrophage and activate genes to facilitate survival within the phagolysosome as well as to change the structure of their surface lipopolysaccharide to make them more resistant to the anti-microbial peptides that are present in the gut. To deal with this *intracellular* pathogen, the host must have T cells available to recognize the infected macrophages and eliminate them. Thus, patients with defective T cell function (or number)--infants, HIV infected patients--will be more susceptible to *Salmonella* infection. Antibiotics that only kill *extracellular* *Salmonella*, may not be sufficient to kill the intracellular organisms, which will be handled by the host immune response. A recent outbreak of *Salmonella* has been associated with tomatoes.

3. Bacterial virulence factors – What is a virulence factor?

Bacteria can cause pathology in a number of ways: i. they may express specific gene products that *facilitate colonization* – such as adhesions that enable them to attach to host tissues. ii. They may express genes that specifically enable them to *thwart the immune response* – many different mechanisms are possible as we shall see.... iii – bacteria may express gene products that *directly damage host tissues* – this is the easiest to understand – bacterial products that destroy tissues, phospholipases, collagenases, proteases or toxins.

For example: *Streptococcus pyogenes* – or Group A streptococci attaches to the skin or pharynx via lipoteichoic acid in the bacterial cell wall – binding to fibronectin; and M protein (a major virulence factor) attaching to skin keratinocytes. The M protein also interferes with phagocytosis. Once attached, these streptococci produce DNases (4 separate enzymes); hyaluronidase – degrades the hyaluronic acid linkages in connective tissues; streptokinase – dissolves clots, and a C5a peptidase – which cleaves complement (which you need to opsonize the organisms). These are all secreted products that enable the organism to first colonize the host then locally destroy tissue and spread through tissue planes. In addition, the organisms produce superantigens (SPE A, B, C)– proteins

that indiscriminately (antigen non-specific) activate T cells – causing systemic reactions. Images of group A streptococcal infection.



The host response to these organisms can also result in disease, such as rheumatic fever. There is strong epidemiological

data linking group A streptococcal pharyngitis to the development of rheumatic heart disease. This is thought to be due to the development of cross-reactive antibodies to the M protein that also recognize the host myocardium and other tissues.

Bacterial toxins – Bacteria can also cause important diseases through toxin production. While the organisms themselves do not destroy tissue or elicit much of an immune response, organisms can secrete toxins that target critical host tissues.

For example: *Clostridium botulinum* are ubiquitous Gram positive bacilli that form spores and are commonly found in the soil. These spores commonly contaminate honey. While not normally a problem, infants who are fed contaminated honey develop "*infant botulism*". This disease is characterized by the gradual onset of paralysis: first constipation, then droopy eyes and bulbar palsies, poor suck, flaccid extremities, and eventually respiratory compromise. Botulinum toxin binds irreversibly to the receptors on the presynaptic cholinergic and adrenergic nerve endings. While the infants are alert – they can't move. Ingested spores germinate and infants for reasons that are unclear, are highly susceptible to the small amounts of toxin that are released from the bacteria.



4. Pathology caused by the host response to bacteria or their components

Perhaps the most common cause of disease elicited by bacteria is that evoked by an "excessive" host response to the organisms. Bacterial components are sensed by many components of the immune system; "professional" immune cells of hematopoietic origin as well as by the mucosal immune system, mucosal epithelial cells, and recruited phagocytic cells, including dendritic cells. Both pro-inflammatory responses (PMNs, macrophages) and anti-inflammatory responses to regulate the recruitment and activation of PMNs are involved in the host response to bacterial infection. Excessive inflammation, as in the lung, causes respiratory compromise – inability to exchange oxygen and respiratory failure. Excessive inflammation in the CNS causes increased pressure and death. Excessive immune activation systemically causes the sepsis syndrome that is often fatal.

How are bacteria sensed? The innate immune system recognizes bacterial and viral "PAMPs" – pathogen associated molecular patterns and in response to these ligands activates a signaling system designed to recruit and activate PMNs to the site of infection. The best studied, and most complex PAMP is LPS or lipopolysaccharide, a component of the cell wall of Gram negative bacteria.

LPS consists of a lipid A core – which is the immunogenic portion of the molecule. It can be variably acylated (penta, hexa etc) which affects its immunogenicity. Attached to the lipid A core are the O-side chains, carbohydrates of various composition and length that are also immunogenic – eliciting Ab production.

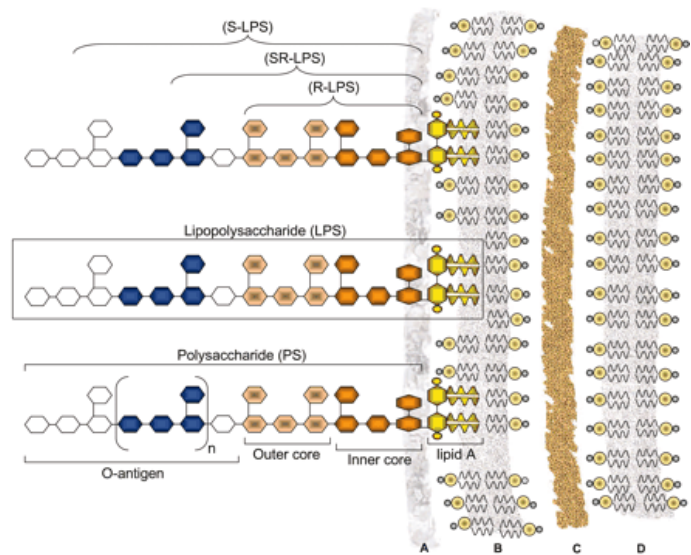
Organisms can vary the chemical composition of their LPS and thus be more or less immunogenic.

The immunogenicity of the core – lipid A also is species dependent.

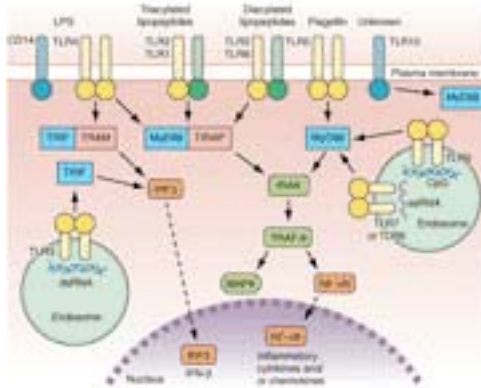
For example the LPS (actually LOS) of *Neisseria meningitidis*, a virulent pathogen that causes sepsis and meningitis is highly toxic, whereas that of *Pseudomonas aeruginosa*, an opportunistic pathogen that usually is only a problem in immunocompromised patients, is much less so.

LPS is sensed by the toll like receptor – TLR4 on the host immune and other cells (endothelial and epithelial cells). This is a transmembrane receptor that is activated by LPS, which is presented

bound to an adaptor protein MD2 and LPS binding protein. LPS- TLR4 at the surface of an immune cell rapidly activates the transcription factor



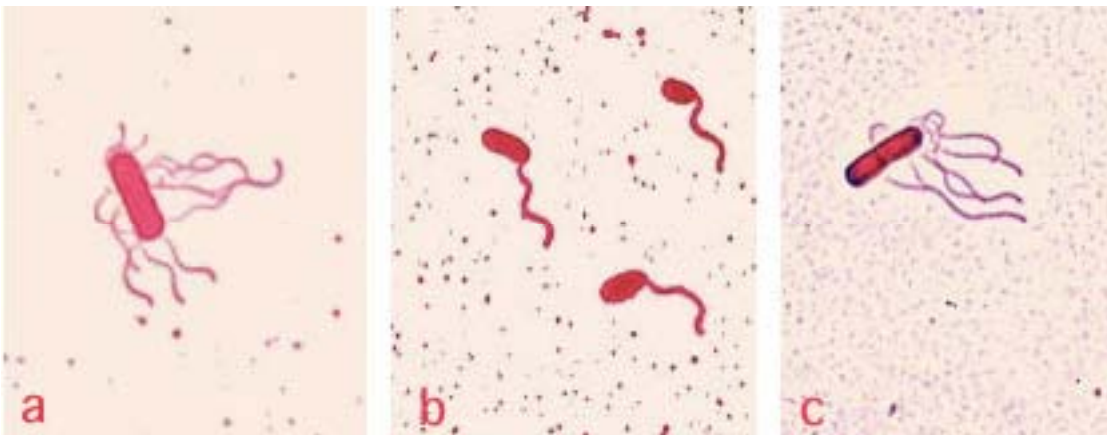
NF- κ B which evokes a proinflammatory immune response, chemokine and cytokine production locally with subsequent systemic consequences.



Flagella – Flagella are bacterial appendages that provide a motility function for the organisms enabling them to chemotax in the direction of a desired carbon source. These critical bacterial components are sensed by TLR5 on the surface of host immune and mucosal cells as well as by a

separate class of intracellular receptors called the NOD proteins. Thus the flagella of intracellular pathogens are efficiently sensed within the host cells. Flagella are highly immunostimulatory and many bacteria once they have established infection, stop producing flagella to decrease the immune response to the organisms.

Cell wall components, lipoteichoic acid are also PAMPs that activate through a similar signaling system. Double stranded RNA, cGp DNA sequences and other PAMPs similarly have their own receptors, thus the innate immune system is highly redundant with several discrete receptors that sense components of the same organism.



5- Bacterial interference with host immune function – While bacterial components are recognized by receptors of the innate immune system that function specifically in host defense, they have also evolved many mechanisms to enable them to evade or even activate normal immune functions. *Staphylococcus aureus*, a common and important human pathogen has multiple mechanisms to thwart immunological clearance mechanisms.

For example: In addition to expressing many surface proteins that specifically bind collagen, fibronectin, platelets – *S. aureus* express protein A. Protein A has an IgG binding domain that prevents the Fc portion of immunoglobulin from binding the PMN – thus blocking opsonization. This same IgG binding domain also activates von Willebrand factor – causing platelet agglutination, and it binds and activates the TNF receptor – initiating TNF α -like signaling. Thus the organism elicits a large PMN response, but then is protected from opsonization and ingestion.