Seminar 2: Good Bugs

We're not ALL BAD!!
Part 2

Viruses
Overview of Viral infections

- Encephalitis/meningitis
  - JC virus
  - Measles
  - LCM virus
  - Arbovirus
  - Rabies

- Common cold
  - Rhinoviruses
  - Parainfluenza virus
  - Respiratory syncytial virus

- Eye infections
  - Herpes simplex virus
  - Adenovirus
  - Cytomegalovirus

- Parotitis
  - Mumps virus

- Pharyngitis
  - Adenovirus
  - Epstein-Barr virus
  - Cytomegalovirus

- Gingivostomatitis
  - Herpes simplex type 1

- Cardiovascular
  - Coxsackie B virus

- Pneumonia
  - Influenza virus, Types A and B
  - Parainfluenza virus
  - Respiratory syncytial virus
  - Adenovirus
  - SARS coronavirus

- Hepatitis
  - Hepatitis virus, types A, B, C, D, E

- Myelitis
  - Poliovirus
  - HTLV-I

- Skin infections
  - Varicella zoster virus
  - Human herpesvirus 6
  - Smallpox
  - Molluscum contagiosum
  - Human papillomavirus
  - Parvovirus B19
  - Rubella
  - Measles
  - Coxsackie A virus

- Gastroenteritis
  - Adenovirus
  - Rotavirus
  - Norovirus
  - Astrovirus
  - Coronavirus

- Sexually transmitted diseases
  - Herpes simplex type 2
  - Human papillomavirus
  - HIV

- Pancreatitis
  - Coxsackie B virus
What is a virus?

• Microscopic particles that infect other organisms and can only replicate within a host cell

• Contain either contain DNA or RNA surrounded by a protective structural viral coat or capsid

• Hijacks the biochemical machinery of the host, alters cell physiology, morphology, biochemistry etc.

• Like bacteria, classification of viruses is difficult –
  – Use morphology, type of nucleic acid, mode of replication, hosts it can infect, and disease type
Viruses Are Tiny Little Things

How Small Is Small?

Water | Glucose | Antibody | Virus | Bacterium | Cancer cell | A period | Tennis ball

10⁻¹ | 1 | 10 | 10² | 10³ | 10⁴ | 10⁵ | 

Nanometers

20-900 nm

Diagram showing the sizes of different objects and viruses in comparison to each other.
Viral Morphology and Assembly

A highly-organized **capsid** comprised of proteins coded by the viral genome provides morphological distinction; **helical, icosahedral, or complex**

- subunits called **protomeres** assemble into **capsomeres** which spontaneously aggregate to form the capsid

- the **nucleocapsid** is the interface between viral DNA and capsid protein

- **spikes** are glycoproteins which forms long projections from the nucleocapsid or envelope, aid in viral attachment

- some viruses have an **envelope** outside the capsid. Presence/absence classifies a virus as naked or enveloped

- a fully assembled viral particle is known as a **virion**

Assembly differs for naked, enveloped, and complex viruses
Eukaryotic Microorganisms: Have been responsible for Malaria, tapeworm and roundworm infections, amoebic dysentery, Leishmanias, athlete’s foot, yeast infections etc.

Differ from prokaryotes in cell size, cellular structure, physiology, and genetic makeup

- Five Major Branches of Eukaryotic Microorganisms
  - Fungi: yeasts, molds, or fleshy fungi
  - Algae: photosynthetic
  - Protozoans: single celled eukaryotes with animal characteristics
  - Slime Molds: can be single-celled or multicellular mold
  - Helminths (parasitic worms)
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<tr>
<th>Characteristic</th>
<th>Prokaryotic</th>
<th>Eukaryotic</th>
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<tr>
<td>Size of Cell</td>
<td>Typically 0.2–2.0 μm in diameter</td>
<td>Typically 10–100 μm in diameter</td>
</tr>
<tr>
<td>Nucleus</td>
<td>No nuclear membrane or nucleoli</td>
<td>True nucleus, consisting of nuclear membrane</td>
</tr>
<tr>
<td></td>
<td></td>
<td>and nucleoli</td>
</tr>
<tr>
<td>Membrane-Enclosed Organelles</td>
<td>Absent</td>
<td>Present; examples include lysosomes, Golgi</td>
</tr>
<tr>
<td></td>
<td></td>
<td>complex, endoplasmic reticulum, mitochondria,</td>
</tr>
<tr>
<td></td>
<td></td>
<td>and chloroplasts</td>
</tr>
<tr>
<td>Flagella</td>
<td>Consist of two protein building blocks</td>
<td>Complex; consist of multiple microtubules</td>
</tr>
<tr>
<td>Glycocalyx</td>
<td>Present as a capsule or slime layer</td>
<td>Present in some cells that lack a cell wall</td>
</tr>
<tr>
<td>Cell Wall</td>
<td>Usually present; chemically complex (typical</td>
<td>When present, chemically simple (includes</td>
</tr>
<tr>
<td></td>
<td>bacterial cell wall includes peptidoglycan)</td>
<td>cellulose and chitin)</td>
</tr>
<tr>
<td>Plasma Membrane</td>
<td>No carbohydrates and generally lacks sterols</td>
<td>Sterols and carbohydrates that serve as</td>
</tr>
<tr>
<td></td>
<td></td>
<td>receptors</td>
</tr>
<tr>
<td>Cytoplasm</td>
<td>No cytoskeleton or cytoplasmic streaming</td>
<td>Cytoskeleton; cytoplasmic streaming</td>
</tr>
<tr>
<td>Ribosomes</td>
<td>Smaller size (70S)</td>
<td>Larger size (80S); smaller size (70S) in</td>
</tr>
<tr>
<td></td>
<td></td>
<td>organelles</td>
</tr>
<tr>
<td>Chromosome (DNA)</td>
<td>Usually single circular chromosome; typically</td>
<td>Multiple linear chromosomes with histones</td>
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<tr>
<td></td>
<td>lacks histones</td>
<td></td>
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<tr>
<td>Cell Division</td>
<td>Binary fission</td>
<td>Involves mitosis</td>
</tr>
<tr>
<td>Sexual Recombination</td>
<td>None; transfer of DNA only</td>
<td>Involves meiosis</td>
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Fungi

- They appear as yeast, molds, fleshy fungi
- Study of fungi: Mycology
- Few fungi are pathogenic (<1%)
- Immune system usually fights fungal invasion
- Generally grow as filamentous, multicellular organisms
- They form filaments, called hyphae, or unicellular organisms
- Mycelium is a collective mass of hyphae
- Have cell wall composed of strong flexible polysaccharide called chitin
Fungi - Yeast

• Single cells that reproduce by budding

• Some may become multicellular by connecting budding cells

• May require oxygen or may grow in the absence of oxygen

• *Saccharomyces cerevisiae* is a model budding yeast. Most well known model species
• Rapidly growing
• In an appropriate environment, hyphae grow to form a mycelium
• Generally hyphae have cross walls called septa
  – Divide the hyphae into distinct uninuclear, cell-like units called septate hyphae
• Some have coenocytic hyphae
  – No septae
Dimorphic Fungi

- As mold with septate hyphae in their natural environment and at 25\(^\circ\) C in laboratory
- As yeast in tissues; in laboratory at 37\(^\circ\) C
- Some can cause systemic mycoses
Algae

• Photosynthetic organisms
• Widespread in fresh and marine waters, soil, plants etc
• Medical concern
  – Food poisoning caused by toxins of marine algae such as dinoflagellates
• Overgrowth of dinoflagellates causes the “red tide”
• Marine animals feed on dinoflagellates
  – Accumulate toxin given off by the algae
• Toxin can persist for several months
• Shellfish poisoning in humans
Protozoans

• Defined by three common characteristics
  – Eukaryotes
  – Unicellular
  – Lack a cell wall

• Many are free-living; others are potential parasites of humans and other animals

• With the exception of one subgroup, they are motile due to cilia, flagella, and/or pseudopodia
Paramecium

Macronucleus

Micronucleus
Giardia lamblia
Classification of Protozoans (Cont.)

• Amoebozoa
  – Most are unicellular and common in soil and water
  – *Entamoeba histolytica*—amoebic dysentery
  – *Naegleria fowleri*—primary amoebic meningocencephalitis (PAM)
    • Rare but typically fatal brain infection
In the Lab

Clorox killed my friends

I am the 1%
Growing Bacteria

- **Escherichia coli**
- **Enterobacter aerogenes**
- **Proteus vulgaris**
- **Salmonella typhimurium**
- **Staphylococcus aureus**
- **MacConkey’s Agar**

**Left Panel:**
Lactose fermenting colonies are PINK.
Non-lactose fermenting colonies are COLORLESS.
E. coli

- *E. coli* is a hardy organism that is easy to culture and easy to manipulate in the lab.
- It is a **model organism** in biotechnology.
- **Model organisms** are extensively studied to understand biological phenomena and the information can be applied to other organisms.
- *E. coli* genome was one of the first to be sequenced in 1997
Advantages to budding yeast as a model organism

- Simple, eukaryotic cell (~10μm diameter)
- Compact genome (genome is sequenced)
- Cells grow on plates and in culture
- Short generation time (~90 minutes)
- Live happily as haploids and diploids
- Easy to manipulate genes (swap promoters, delete genes)
- Easy to conduct genetic screens
- Many yeast genes have evolutionarily conserved homologs in humans
The Yeast Knockout (YKO) Collection contains over 6,000 gene-disruption mutants as a unique tool for the functional analysis of the yeast genome.

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Model Organisms
Yeast
Good Bugs

We're not ALL BAD!!
Good Bugs
Your body has a well-entrenched army

**Microbiome in Numbers**

- **100 Trillion**
  - Symbiotic microbes live in and on every person and make up the human microbiota.

- **95%**
  - Of our microbiota is located in the GI tract.

- **150:1**
  - The genes in your microbiome outnumber the genes in our genome by about 150 to one.

- **>10,000**
  - Number of different microbial species that researchers have identified living in and on the human body.

- **1.3X**
  - You have 1.3 times more microbes than human cells.

- **2kg**
  - The gut microbiota can weigh up to 2Kg.

- **2.5**
  - The number of times your body’s microbes would circle the earth if positioned end to end.

- **Each individual has a unique gut microbiota, as personal as a fingerprint.**

- **5:1**
  - Viruses: Bacteria in the gut microbiota.

- **90%**
  - It is thought that 90% of disease can be linked in some way back to the gut and health of the microbiome.

- **The human body has more microbes than there are stars in the milky way.**

**The microbiome is more medically accessible and manipulable than the human genome.**
An infectious microbe has to out-compete the host microbiome

- Steal nutrients
- Establish a new colony in enemy territory
- Evade the immune response
Host-Microbe Relationship

- Under normal circumstances, humans are free of microbes in utero.
- During birth, a newborn will be exposed to microbes, which will start to colonize the infant’s intestinal tract and mucous membranes.
- Humans and microbes start to establish a symbiotic relationship.
Normal Flora

- Newborns’ first contact with microbes—birth canal

- Followed by breathing, then feeding

- Throughout life microbes will establish residency in mucous membranes open to the environment. This is your **normal flora**
  - **Resident flora:** Remain part of the normal flora throughout life of a person
  - **Transient flora:** Only remain for a few hours, days, or months before they vanish
Symbiosis
-a close relationship between two different types of organisms in a community

- **Mutualism**
  - Both members benefit from the interaction

- **Commensalism**
  - One organism benefits, and the other is neither harmed nor helped

- **Parasitism**
  - One organism benefits while the other is harmed or killed – most pathogens

- **Amensalism**
  - One organism can hamper or prevent the growth/survival of another without being affected by the other organism
Symbiosis - Examples

-a close relationship between two different types of organisms in a community

- Mutualism
  - i.e. normal gut flora such as *E. coli*

- Commensalism
  - i.e. bacteria that live in ear canal

- Parasitism
  - i.e. bacterial infection parasites capable of causing disease are pathogens

- Amensalism
  - i.e. *Pencicllium*
Opportunistic Pathogens

- Pathogen—a microorganism that is capable of causing disease

- When balance in the normal flora and human host is interrupted—microbes of the normal flora can become opportunistic pathogens

- Opportunistic pathogens do not cause disease in their normal habitat in a healthy person
Opportunistic Pathogens (Cont.)

- Compromised immune system
  - acute and chronic disease (primarily immune disease)
  - malnutrition
  - stress
  - age
  - radiation/chemotherapy
  - immunosuppressive drugs

- Changes in the normal flora
  - Normal flora is usually protective
  - Competition no longer exists

- Entrance of the normal flora into areas of the body where it is not present under normal conditions
Normal Flora vs Infection vs Disease

- Normal Flora: non-invasive presence of microorganisms at specific anatomical sites

- Infection: an invasion and sustained colonization of an infectious agent in the host. The microorganism has to gain entry into the host and its tissue

- Infectious Disease: when an infectious agent causes pathophysiological changes and disrupts normal physiological function

IMPORTANT TAKE AWAY – You can have infection without disease
## Microflora of the oral cavity

<table>
<thead>
<tr>
<th>Habitat</th>
<th>Comment</th>
</tr>
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| Lips, cheek, palate    | • Biomass limited by desquamation  
                        | • Some surfaces have specialised host cell types                        |
| Tongue                 | • Highly papillated surface  
                        | • Acts as a reservoir for obligate anaerobes                           |
| Teeth                  | Non-shedding surface enabling large masses of microbes to accumulate (dental plaque biofilm)  
                        | Teeth have distinct surfaces for microbial colonisation (e.g. Smooth surfaces, pits & fissures etc) will support distinct micro flora due to their biological properties |
| Gingival crevice / pocket | Health (aerobic), disease (anaerobic)                                  |
Microflora of the oral cavity

**Gram Positive organisms:**

Bulk of oral bacteria
Rods (bacilli), cocci or irregular shape (pleomorphic)
Oxygen tolerance varies from aerobes to strict anaerobes
Most are fermentative
Cell wall has thick peptidoglycan layer

- Strep mutans (caries, bacterial endocarditis)
- Strep anginosus (maxillofacial infections, brain, liver)
- Strep mitis (bacterial endocarditis)
Microflora of the oral cavity

Gram Negative organisms:

Found mostly in established/subgingival plaque
Strict or facultative anaerobes
Some fermentative, others produce enzymes which break down tissue
Cell wall different to Gram positive with a thin peptidoglycan layer
Has B-lactamase, LPS/endotoxin

P. gingivalis (major periodontal pathogen)
A. actinomycetemcomitans (aggressive periodontitis)
Treponema (acute periodontal conditions)
Plaque Hypotheses

- **Specific plaque hypothesis:** Certain bacteria in the mouth are responsible for disease

- Non-specific plaque hypothesis: It is the combined effect of all of the bacteria in your mouth that cause disease – some are more susceptible due to genetics, behaviour etc.

- Ecological plaque hypothesis: A shift in the ecology (the type of bacteria dominant) of the mouth causes disease
PERIODONTAL PROBE

The periodontal probe is used routinely to examine the space below the gum tissue alongside your tooth. This space is similar to the space between your fingernail and your finger. When your gum tissue becomes inflamed or diseased, dentists refer to this space as a pocket. Periodontal probes have calibrations in millimeters to measure the changes that occur during the progression of periodontal disease. The higher the number of millimeters below the gum, the more loss of attachment to the tooth has occurred.

- **1-3 mm** is normal space below the gum tissue
- **3-5 mm** is early or mild periodontitis
- **5-7 mm** is moderate periodontitis
- **7-10 mm** is advanced periodontitis
- Plaque build up leads to inflammation, which changes the environment, which leads to new bacteria building up below the gums

- Gram negative, obligate anaerobes (dislike oxygen) begin to dominate and cause disease
Ecological Plaque Hypothesis

- Plaque build up leads to inflammation, which changes the environment, which leads to new bacteria building up below the gums