Lecture 1 Introduction.

Individual organisms or groups that are obligate parasites, facultative parasites, or saprophytes, ie all viruses are obligate parasites.

- **Parasite:** an organism that depends on host for survival, growth, and replication
    - **Obligate parasite:** is physiologically dependent on host
        - All viruses
    - **Facultative parasite:** is not physiologically dependent on host
        - establishes symbiotic relationship with a host only if the opportunity presents itself (bacteria)
- **Saprophytes:** organisms which obtain nutrients from dead organic matter (Fungi)

Key Terms:

- **pathogen:** disease-causing microorganism // an infectious agent
- **Immuno-compromised:** those with impaired immune systems (burn victims, infected HIV)
- **Incubation time:** time between infection and appearance of clinical signs or symptoms
    - 6-12 hours (Norwalk virus) to 30-60 days (Hepatitis A Virus)
- **Vector:** describe insects that carry an infectious agent from one host to another
- **Vehicle:** Any item carrying an infectious agent from its source to a host (food, air)
- **Fomite**: Any inanimate object used in common by several hosts in a given setting that might increase the chance of transmission and infection
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Understand the idea of commensalism mutualism and parasitism and that this relationship can vary depending on the environment.

- commensalism: Living together with no benefit
- mutualism: Living together with benefits
- parasitism: Living together with a one-sided benefit

What is the difference in disease causing properties of a frank and an opportunistic pathogen?

- frank: capable of infecting healthy and immunocompromised persons
- opportunistic: capable of infecting immunocompromised host ONLY

Rank these 3 groups in order of abundance of individual organisms on the planet: worms, viruses, bacteria.

- Viruses > Bacteria > Worms

How can microbes influence the weather?

- microbes in upper atmosphere can freeze at fairly warm temperatures, so that the "biological ice nuclei" form condensation nuclei which trigger rain
- Kill photosynthetic plankton - sequesteration and mobilization of CO_2
- Involved in carbon movement, nitrogen fixation;
- Release huge amount of Carbon, impact Global warming,

What is the level of virus particles in the planktonic zone of sea water and in fresh water?

- sea water: 10^7 virus particles/ml
- fresh water: 10^4 virus particles/ml

What's the difference between pathogenicity and pathogenesis?

- **Pathogenicity** (Virulence) is the measure ability to cause disease
- **Pathogenesis** is the physical disease production process
- **Virulence factors:** microbial products that contribute to virulence (cell-surface proteins, toxins)

What is the difference between a microbe that can cause disease with a doses of $\geq$10 infectious units/particles and one that can cause an illness with $\geq$ 100 infectious units/particles?, What would explain this difference between these organism?

- Illness and Disease are the same thing; the first microbe is more toxic because it require less particles to induce an infection which will produce clinical symptons in the host. The second microbe requires a larger number of particles to induce disease and is therefore less virulent.

What are the stages in pathogenesis?

- Entry into host
- Primary Replication
- Spread (airway only)
- Tropism (targeting organs- lung, brain etc)
- Interaction with host defenses (avoid immunity)
- Tissue damage (ie extent of infection-- inflamation,)
- Clearance by host / Persistence (cannot clear)

One of the stages of pathogenesis in a host is tropism which can also be described as the niche in which a parasite lives, what dose this mean?

- A niche is an area which an organism inhabits; it refers to the relationship of a species with all the factors affecting it.
- Tropism refers to the way in which pathogens have eveloved to preferentially target specific host cell types. This is because of a beneficial relationship from the region of the particular organ.
- As such, tropism can be described as the niche in which the parasite thrives.

Name 3 stages in virus replication in an infected cell that are common to all virus infections.

- Binding/penetration
- Translation
- Assembly/Release

Give examples of an intermediate host and final host for 2 obligate parasites.

- **Intermediate:** Host in which the parasite develops but not to sexual maturity
- **Final:** Host in which the parasite develops to sexual maturity
- ***Giardia lamblia:***
    - **Intermediate: Cyst (I) in water, food, fomites, hand**
    - **Final: Trophozoites (D) in intestine**
- ***Plasmodium* spp.:**
    - **Intermediate: HUMAN: merozoite (liver cells and gametocytes (D) in RBC**
    - **Final: female mosquito(I) (gametes --> sporozoites)**

Malaria is caused by Plasmodium species. Name the vector and the final host. Which host produces the gametes and in which host do the gametes become fertilized? Name the diagnostic and infectious forms of this parasite.

- Vector: Female Mosquito
- Final Host: Female Mosquito: gametes are fertilized and form sporozoites (I)
- Intermediate Host: Human: sporozoites make Gametocytes (D) (gametes)

Name 3 parasites and how you would avoid becoming infected with them.

- ***Giardia lamblia:*** treat water before drinking in the outdoors
- ***Plasmodium* spp:** take medication, use mosquito repellent
- **Influenzavirus:** vaccination, frequent hand washing, covering coughs/sneeze

Lecture 2 Disease and Epidemiology

Understand positive and negative aspects of microbial activity.

Pro	Con
Maintain health	Food spoilage
Help cycle nutrients in enviroment	Disease
Bioremediation of toxic material (microbes metabolize pollutants)	Decomposition of wood
Industrial processes: yogurt, cheese, chocolate, wine, bread	
Biotechnology: hormones (insulin), clean oil spills	

The human microbiome has different components in different sites/niches- you should know some key organism at these sites.

Nose	Staphylococcus Aureus
Mouth	Streptococcus Species
Throat	Streptococcus Species, Mycoplasma species
Skin	Staphylococcus Epidermidis
Large Intestine	Escherichia coli
Vagina	Lactobacillus species
Urethra	Streptococcus species, mycobacterium species

How does the gut microbiome affect disease susceptibility?

- modulates the Immune system and contributes to auto immune diseases (chron's disease)

What are Pathogen-Associated Molecular Pattern receptors and give an example of one that senses bacteria and one that senses viruses?

- Toll-like receptors (TLRs) and pattern recognition receptors (PRRs)
- Recognize the small/repeated molecular sequences (PAMP) found on pathogens and activate the innate immune response
- TLR4 recognizes Bacterial LPS
- TLR3 recognizes viral RNA

How can the gut microbiome affect obesity?

- Leptin controls your appetite; increase leptin turns of your appetite
- Homozygous mice with [ob/ob] has an increase capactity for dietary energy harvest because they lack the leptin gene

What are the causative agents of small pox, the plague and tuberculosis.

- **Bubonic plague Caused by *Yersinia pestis***
- **Tuberculosis Caused by *Mycobacterium tuberculosis*** (primary lung infection, p2p)
- **Smallpox Caused by *Variola major* and *V. minor***

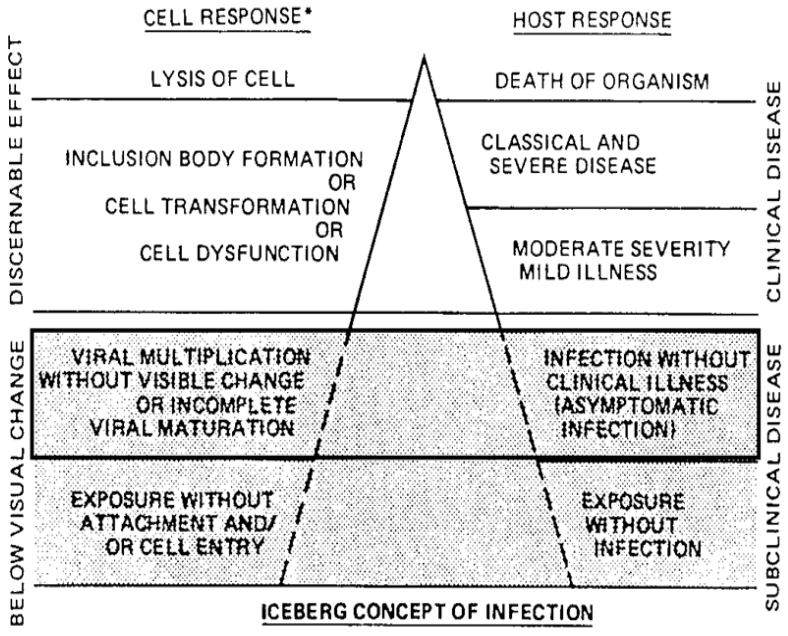
What was a major change in human culture that increased disease transmission and outbreaks and when did it occur?

- 8000-10000 years ago; permanent settlements, agricultures, trade, domestication, agriculture increased exposure and contact
- prior to this they were hunter/gatherer so limited outside contact; low exposure or spread

What are the tests for proving that a given microorganism causes disease and mention some difficulties the proving this.

- Robert Koch's: specific organisms were linked to specific diseases → **Koch's postulates**
- microorganism is present in all hosts who exhibit the disease, but absent from those without disease
    - Limitations: subclinical infections
- The microorganism must be capable of being grown outside the diseased host as a pure culture
    - Limitations: pathogen is unculturable and difficult to grow (example: syphilis)
- When administering the pure culture to a new host, the symptoms of the disease must develop
    - Limitations: ethics of human/animal models, some human pathogens might not cause disease in animals
- Re-isolation of the identical microorganism from the experimentally infected host must be possible

Understand the iceberg concept of disease and an example.



What is the difference between infection and disease.

- **Infection** is the penetration and multiplication of an agent within a host. It is governed by exposure conditions host susceptibility
- **Disease** is the host's response to infection when it is severe enough to evoke clinical symptoms. It is governed by type of pathogen, portal of entry, host susceptibility

- **Minimum Infective Dose (MID):** the minimum number of pathogens that are necessary to enter the host to establish an infection

If hamburger is contaminated with *Listeria moncytogenes* that can cause fatal infections with a MID of 10^7 bacteria for normal individuals, and 10^5 for high-risk people what is the risk for infection from eating a rare hamburger that started with 10^8 bacteria versus a well-done hamburger, is the risk of infection versus disease the same for a 65 yr old versus a 20 year old and explain why? Discuss the risk of subclinical infection versus disease.

- Higher risk from eating rare burger because it exceeds MID
- higher risk for 65y-o b/c their immune system is more fragile and more compromised

Know the host factors that would contribute to a change in pathogenesis to increase pathogenicity.

- **Pathogen**
  - Variability in genetic expression
  - Interaction of pathogen with transmitting vehicle
  - Interaction with other organisms in ecosystem
- **Host**
  - Age
  - General health
  - Pregnancy
  - Occupation
  - Nutritional status
  - Alcoholism
  - Metabolic disorders
  - Immune competence
  - Genetic disturbances

How can gene expression a microbe affect its ability to cause dieses?

- **Gene expression:** process by which information from a gene is used in the synthesis of a functional gene product → proteins or functional RNA.
- M. Tb: exposed to nutrient depletion and environmental stress → non-replicative state with up-regulation of β-oxidation and virulence genes; re-entry into cell division → similar to latent infection in TB patients (1/3 infected)

How can interaction with other bacteria affect the ability of bacteria o survey in the environment?

- **Internalization:** protozoa are more resistant than bacteria and can internalize bacteria offereing better environments for replication
- **Biofilm formation:** protection, access to nutrients, communication

What are the four patterns of disease spread in a population, how would you characterize the 20 Legionnaire pneumonia deaths in Quebec City this summer given that they had none last year?

- **Sporadic:** Few isolated cases occurring in an unpredictable manner (Rabies)
- **Endemic:** A relatively constant but low number of cases in a particular location (Giardiasis)
- **Epidemic:** An unusual high number of cases of a disease in a relatively short period in a particular region (HIV or Influenza)
- **Pandemic:** A world-wide epidemic (Influenza)
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- Quebec: Sporadic

Definitions

- **Microbial contamination**: presence of living organisms in or on a host
- **Communicable or infectious disease**: spread from one person to another with relative ease (example: common cold)
- **Clinical case**: a host with obvious signs and symptoms of infection
- **Subclinical or inapparent infection**: case with no obvious signs or symptoms
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- **Zoonosis**: an infection that humans can acquire from animals (ex. Rabies)
- **Anthroponosis**: an infection that humans can acquire directly from other humans (ex. HIV)
- **Sapronosis**: an infection that humans can acquire from saprophytes (dead matter eater, nonliving often decaying nutrient sources (ex. *Cryptococcus* spp.)

- **Prevalence =**

$$\frac{\text{total number of cases in the population}}{\text{number of individuals in the population}}$$

- **Incidence=**

$$\frac{\text{number of new cases in population}}{\text{time period}}$$

Lecture 3 Transmission

Know the 7 means of horizontal transmission.

- Direct contact transmissiond
    - This requires close contact
        - touching an infected individual
        - sexual contactsexual contact
        - contact with oral secretions
        - contact with body lesions
    - These pathogens do not survive well in the external environment
- Indirect transmission-(fomites)
    - transmission occurs through fomites
    - ex: head lice via clothing, toys
    - these pathogens can survive well in the external environment
- Vehicle-based transmission
    - transmission through vehicles such as: water, food, air, soil
    - frequently linked to fecal-oral contamination
    -
    - Ascaris lumbricoides
    - Hookworms: Ancylostoma duodenale
- Vector-borne transmission
    - through flies, mosquitos, mites, fleas, ticks, rats
    - increase transmission range of pathogen
    - spread either through biting or fecal-oral route of a vector
- Droplet
- Airborne transmission
- Iatrogenic transmission

Know the differences in organism for direct and indirect contact.

Examples of vector born transmission.

Droplet versus airborne transmission.

Types of vertical transmission.

Differences between the times of shedding of infectious organism and the timeline of disease.

Epidemiology concepts and Factors Influencing Disease Transmission.

Lecture 4 Water

What are the 4 major routes of entry of microbes into the human body with an example microbe for each?

- Skin: HepB
- Mouth: Salmonella
- Genital: HIV
- Body fluid (Blood): Rubella
- Droplet (respiratory tract): Legionella

Know the major control strategies for waterborne infections.

- Personal hygiene
- water sanitation (drinking vs washing)
- Sewage treatment
- proper cooking of food
- food inspections
- vector control

How dos *Vibreo cholera* cause diarrhea?

- *V. cholerae* secretes cholera exotoxin (A and B) into the extracellular space
- The A/B complex binds to the GM1 ganglioside receptor and is endocytosed
- Subunit A activates adenylate cyclase: → [cAMP] ↑: → ↑ salt secretion
- Disrupts cellular osmotic balance resulting in diarrhea

Endotoxin vs. Exotoxin?

Exotoxin	**Endotoxin**
Produced by Gram (+) and Gram(-) bacteria	Only by Gram (-) bacteria
Released from cell	Integral part of cell wall
Protein	Lipid A or lipopolysccahride
Many types based on function and structure	Only one type
Heat labile	Heat stable
Specific receptors on host target cell	Diverse range of cells and systems affected
Specific effects	Diverse range of effects

Key features of *Legionella pneumophilia.*

- Gram (-)
- Causes legionellosis
    - Severe pneumonia
    - Pontiac Fever (non-pneumonic, mild,)
- Forms biofilms (detach in chunks; more likely to deliver an infectious dose)
- Abundant in aquatic environments, stagnant waters (ditches, soil, swamp)

- survive from 0-63C and a pH of 5.0-8.5 (due to symbioses)
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Must be able to differentiate the different ways that pathogens can be transmitted in water and their classification ie water-borne, water-washed, etc; including parasites and intermediate vectors.

- water-borne: Caused by ingestion of pathogen-contaminated water
  - Cholera (*Vibrio cholerae*)
  - Legionellosis (*Legionella* spp.)
  - Dysentery *(Salmonella enterica)*
- Water-washed: caused by poor personal hygiene and skin and eye contact with contaminated water
  - Trachoma (*Chlamydia trachomatis*)
  - Typhus (*Rickettsia* spp.)
- Water-related: Transmitted by insect vectors (mosquitoes) that breed or feed near contaminated water)
  - Malaria (*Plasmodium spp.*)
  - Elephantiasis (*Wuchereria bancrofti)*
- Water-based: Caused by parasites found in intermediate organisms living in contaminated water
  - Snail fever (*Schistosoma mansoni)*
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Understand the basis for the UECS grouping of water-related diseases.

- the unitary environmental classification system (UECS) is based on

  a) The seven resulting disease categories:

  1) feco-oral waterborne and water-washed diseases
  2) non feco-oral water-washed diseases
  3) geohelminthiases (parasitic nematodes without intermediate hosts or vectors)
  4) taeniases (tapeworms)
  5) water-based diseases
  6) insect-vector diseases
  7) rodent-vector diseases

  b) Five transmission features: latency, intermediate hosts, infectivity, persistence, ability to multiply outside the host

Disease	Latency	Intermediate hosts	Infectivity	Persistence	Multiply
Feco-oral waterborne and washed	No	No	Medium to low	Med to high or Low to med	Able
Non feco-oral water washed	No	No	High	Med to high	No
Taeniases (Tapeworms)	Yes	Yes (cows/pigs)	Very high	Yes	Yes
Water-based (Schistosoma)	Yes	Yes (aquatic)	High	Yes	Yes

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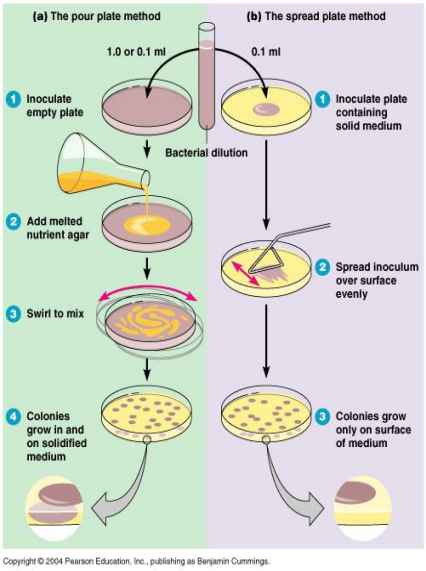

Should know some examples of major water- and excreta- related pathogens (ie a virus, bacterial and parasite).

Know water culturing methods and what is a MPN?

- Spread plates/Pour plates
  - **Spread Plate:** sample is diluted in a series of tubes containing sterile liquid. An Aliquot from diluted sample is spread evenly over the surface.
  - **Pour plate:** Sample is diluted in the melted agar medium
- Membrane filtration
  - samples is passed over sterile 47 mm membrane filters containing 0.45/0.2 µm pores
- MPN methods
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Lecture 5 Food.

Understand good and bad aspect of microbes in food ie fermented foods versus rotten foods or pathogen contaminated foods.

- **Digestive tract:** Breakdown nutrients and role in immunity
- **Food industry:** enhance or change food properties (fermentation)
    - Yeast for bread
    - Wine and Beer
    - Yogurt and cheese
    - Lactobacilli and streptococci for pickling
- **Chemical industry:** production of amino acids, organic chemicals and enantiomerically pure compounds
- **Research:** Model organism
- **Pharmaceutics:** Insulin, Growth Hormone, Factor IX
- **Medicine:** Immunizations
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- **Food spoilage: U**nwanted changes in food from undesirable metabolic reactions, growth of a pathogen or presence of unwanted organisms. Leads to loss of nutritive value, appearance, taste and illness.
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Nutrition that microbes require to grow.

- Water / moisture
- A source of carbon (*e.g.*, organic material, CO_2, CO, CH_3).
- A source of energy (*e.g.*, protein, fat, sugar, lactic acid, light)
- A source of **P, O, N**, S, K, Mg, Ca, Fe, Na.
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- Depends on pH, Temp, Oxygen, Salt/Osmotic Pressure

Aspects of food borne bacteria that cause disease.

- Toxic chemicals secreted by a microorganism
    - Endotoxin (LPS)
    - Exotoxin (botulinum toxin, aflatoxin,)
- **not always inactivated by cooking**
    -

Understand intrinsic food spoilage factors- preservation versus spoilage factors; know some high risk and low risk of spoilage foods and why.

- **Nutritional composition of food**
    - Determines which microbes are present and whether or not they will grow (e*xample: protein versus starch)*
    - Presence of natural antimicrobial agents (*benzoic acid in cranberries)*
    - Fortified foods may lead to the growth of microorganisms → good growth medium
- **Water activity of food**
    - unbound or free water

    - Pure water activity is 1. Microbes require at least 0.9 (*colonization of meat versus dried legumes*)
    - Food processors reduce water activity by drying or addition of salts and sugars *(Jam)*
- **Food acidity**
    - a natural quality *(citrus fruits)* or the result of fermentation *(pickles)*
    - pH < 5: reduced microbial growth
    - pH ~ 7: supports microbial growth
- **Physical structure of food**
    - rinds of fruits and vegetables or shells on eggs
    - coverings are dry and nutritionally poor
    - grinding meats creates air pockets and more surfaces to harbor microorganisms, unlike uncut meats
- **Microbial competition**
    - In fermented foods- large number of fermentative bacteria but few pathogens due to competition

Factor	High risk food	Low risk food
Nutritional composition	Chemically rich or fortified food (yogurt, bread, steak)	Chemically limited foods (grains, cereals, flour)
Water activity	Moist food (meat, milk)	Dry food (pasta, jam)
pH	Neutral (bread)	Low (orange juice)
Physical structure	Food without shells, rinds, ground meat	Intact food with rinds, shells, skins, intact
Microbial competition	Food that lack resident microbial population (meat)	Food that have large microbial population (pickles)

Preservation by Pasteurization and some findings of Pasteur (ie "germ theory").

- **Pasteurization**
    - Commonly used with dairy (68^{o}C for 30 min, etc), *(*destroys over 90% germs*)*
    - Foods are heated just enough to kill mesophilic, non-endospore-forming bacteria
    - Some bacteria survive and these foods spoil without refrigeration
- 1862: disproves ***Spontaneous Generation***
- 1885: germ theory (germs cause disease)
    - weakened disease causing germs can protect against more virulent strains

- Designed the first vaccines for rabies and anthrax
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Know some major food borne pathogens; what are the most common what are the most serious and which foods do they come from?

Agent	Source	Comments
Campylobacter jejuni	Raw or undercooked meats, raw milk	Immunosuppressant exotoxin
Clostridium botulinum	Home prepared foods (canning)	Neurotoxin
Yersinia enterocolitica	Pork, dairy and produce	Diarrhea and cramping (mimic appendicitis) and grows at low temp
***E. coli* O157:H7**	Raw milk and meat	Enterotoxin (released in the intestine)
Listeria monocytogenes	Dairy products, undercooked meats, seafood and produce	Very common in soil and water, contamination quite easy, grows at refrigerator temp
Norovirus (Norwalk virus)	Uncooked seafood	Spread by food handlers. Extremely infectious. Destroyed by chlorine and cooking.

Agent	Source	Comments
***Shigella* spp.**	Salads, milk, and dairy products	Colonization of the gut; causes dysentery; enterotoxin
Staphylococcus aureus	Cooked high protein foods	Produces a potent exotoxin not destroyed by cooking
Toxoplasma gondii	Meats (pork in particular); cat litter	Protozoan parasite; intracellular
Vibrio vulnificus	Raw, undercooked seafood	Causes septicemia
***Salmonella* spp.**	Raw and undercooked eggs, meat, dairy products, fruits and vegetables	Destructive intestinal inflammation (LPS)

Know the 5 forms of toxic *E. coli*.

- ***Enterohemmorhagic E. coli (EHEC)*** ***E. coli* O157:H7**
  - Have fimbriae (attachment appendages)
  - Produces shiga-toxin
    - Elicit massive inflammatory response
    - Moderately invasive
    - Causes bloody diarrhea, hemolytic uremic syndrome, kidney failure
- **Enterotoxigenic *E. coli* (ETEC)**
  - Have fimbriae (attachment appendages)
  - Produces 2 enterotoxins (LT, ST)
  - Non-invasive and doesn't leave lumen
  - Leading cause of diarrhea in children and travellers
- **Enteropathogenic *E. coli* (EPEC)**
  - No fimbriae--Instead produces intimin toxin that mediates attachment
  - No enterotoxins--Instead produces shigella-like toxins
  - Moderately invasive
  - Results in massive inflammatory responses
- **Enteroinvasive *E. coli* (EIEC)**
  - Humans are the only reservoirs
  - Similar symptoms to shigellosis
    - Transmitted through, fecal-oral route, contaminated food
    - Dysentery, fever, vomiting
- **Enteroaggregative *E. coli* (EAEC)**

- Humans are the only reservoirs
- Have fimbriae (attachment appendages)
- Non-invasive
- Produces hemolysin and ST toxin
    - Causes watery diarrhea
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Lecture 6 Pathogen Spread Through Animate And Inanimate Surfaces

Types of environmental surfaces and their characteristics that promote spread.

What's the big deal about Methicillin-Resistant *Staph. aureus* (MRSA) and *Clostridium difficile*?

Pathogen spread by medical devices; why a problem, and what are the major microbes via this route.

Why is contamination of hands so important for spreading disease, from which sources, how do you disinfect.

What pathogens are spred via hands?