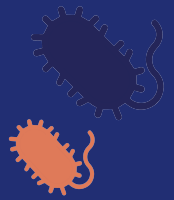
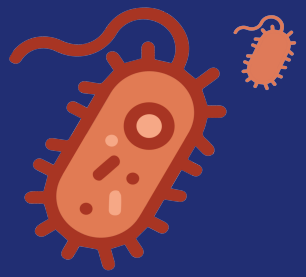
Host Immune Response

By: Sarika Verma

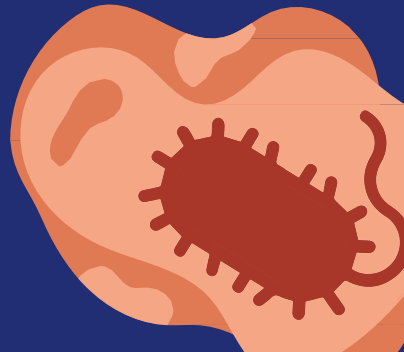


Case 1 Summary



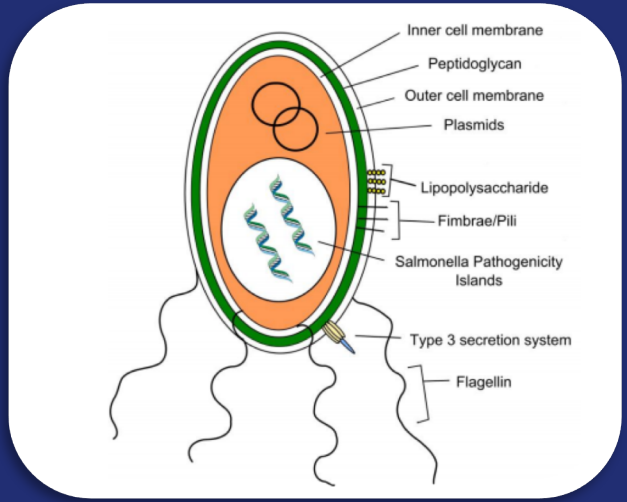
The Raw Food Diet

25-year-old Johnny has been eating raw eggs as part of his new body building diet. One week into his new diet, he develops a mild fever, severe abdominal cramps and watery diarrhea. After 4 days of diarrhea, he goes to a walk-in clinic where the doctor finds that Johnny is volume depleted and has some abdominal tenderness. She gives him a container to collect a stool sample to send to the Microbiology Laboratory and suggests that he stop eating the raw eggs. Johnny's stool sample grows ***Salmonella enteritidis.***



Salmonella enteritidis

- Gram-negative bacteria that causes gastroenteritis
- Transmitted via the fecal-oral route
- Thin peptidoglycan layer and an outer lipid membrane composed of lipopolysaccharides (LPS), proteins, and phospholipids
  - LPS functions as an essential virulence factor for *S. enteritidis*



Host Response

Elements of the innate and adaptive (humoral and cellular) immune response

01

02

Host Damage

Damage that ensues to the host from the immune response

Bacterial Evasion

Bacteria attempt to evade the host response elements

03

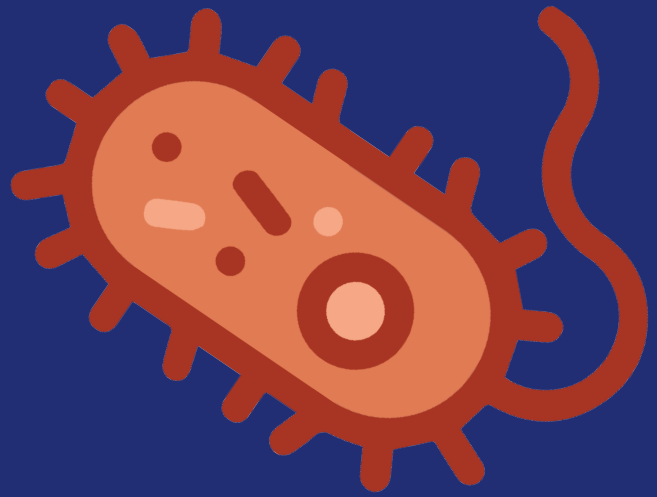
04

Outcome

Bacteria removal, patient recovery and immunity to future infections

Host Response

What elements of the innate and adaptive (humoral and cellular) immune response are involved in this infection?

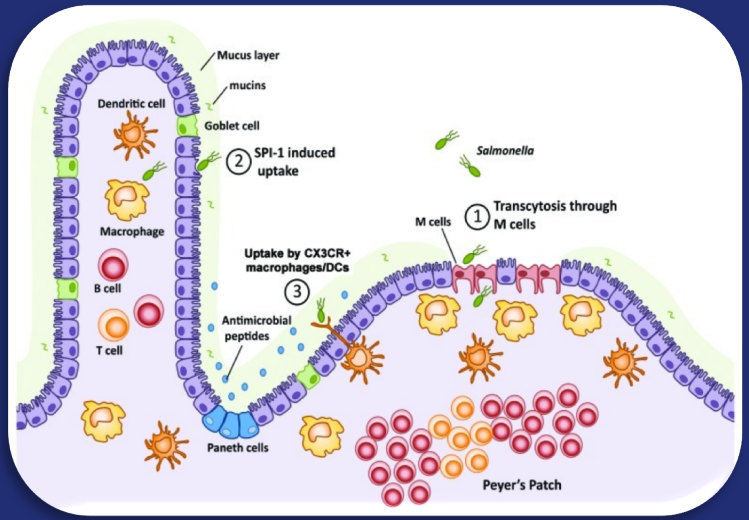


Non Specific Defenses

- Skin: serves as a physical barrier to help prevent *S. enteritidis* from entering the body
  - Surpasses this physical barrier once it is ingested orally
- Stomach pH: highly acidic (low pH) environment of the stomach can kill most ingested pathogens
  - Low amounts of stomach acid or less acidic stomach acid make individuals more susceptible to infection
- Normal Gut Microbiota: pathogen competes with normal microbiota for resources and nutrients

Innate Immune Response

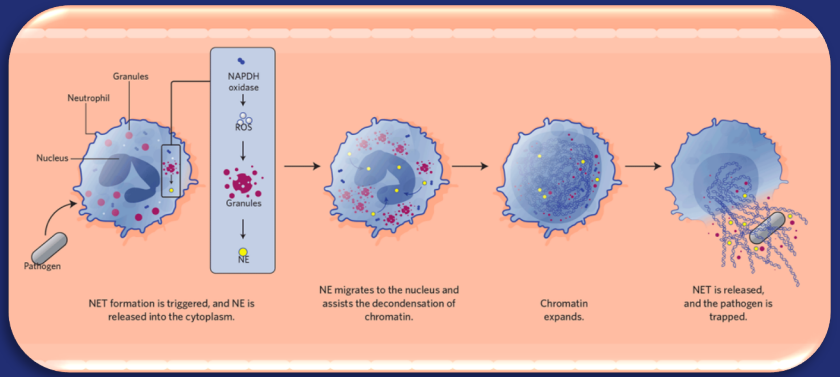
- *Salmonella enteritidis* crosses the small intestine mucosal epithelium at Microfold (M) cells found in mucosa-associated lymphoid tissues
    - *S. enteritidis* is then exposed to several destructive elements:
        i. Antimicrobial peptides produced by Paneth cells
        ii. Mucous produced by goblet cells
        iii. Enzymes
        iv. Bile containing secretory IgA



- In the small intestine, the bacterial LPS is recognized by Toll-like receptors 4 (TLR-4)
    - Initiates the release of cytokines and chemokines (ex. IL-1, IL-6, IL-8, and TNF-2) to attract neutrophils and macrophages

- Neutrophils: release reactive oxygen species (ROS), antimicrobial proteins, and neutrophil extracellular traps (NETs)

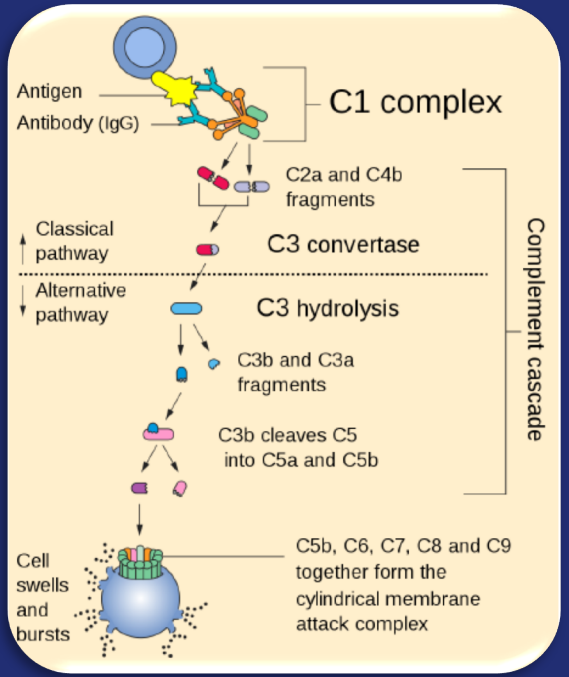
- Macrophages: produce nitric oxide (NO), which is a toxic defense molecule against *S. enteritidis*

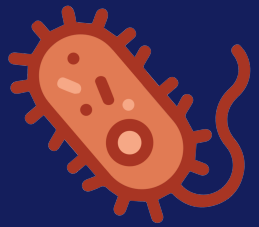


Both neutrophils and macrophages will also phagocytose the pathogen intracellularly to clear the infection

Complement System

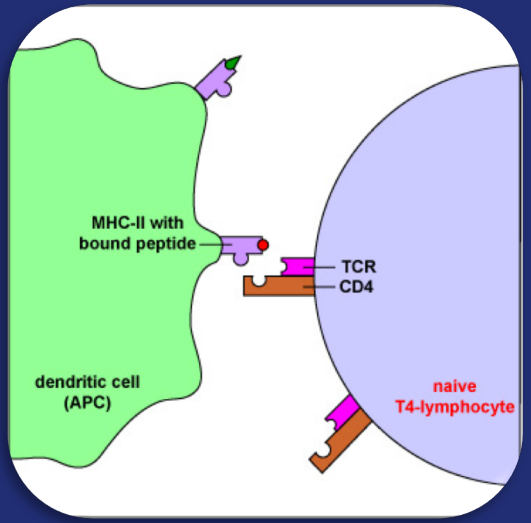
- When the complement system is activated, it triggers a series of protein cascades which lyse the bacterial cell wall via the MAC complex
    - MAC complex is formed when complement proteins bind to the bacterial surface and create a pore on the surface



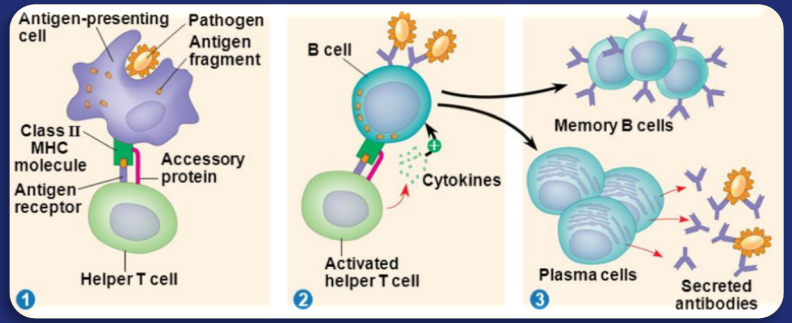


Adaptive Immune Response

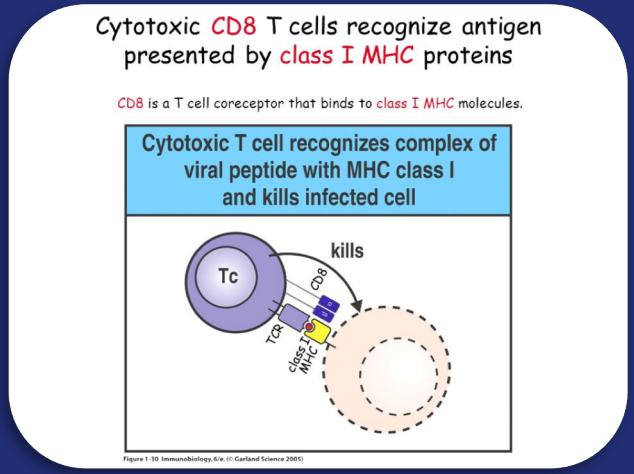
- Dendritic cells = antigen presenting cells (APCs)
    - Messengers between the innate and adaptive immune systems
    - Digest the pathogen and present its antigen material on Major Histocompatibility Complex Class I and II (MHC I and II) proteins
- CD4+ T helper cells become activated once they bind to MHC II on dendritic cell surfaces



- The activated CD4+ T helper cells then:
  - Facilitate a TH1-type immune response, characterized by release of pro-inflammatory molecules (IFN-gamma, TNF-alpha, IL-6, & IL-12)
  - Increase phagocytic activity of macrophages
  - Activate B cells to produce and secrete antibodies specific against *Salmonella enteritidis*
    - IgA antibodies bind to the surface proteins of *Salmonella* and prevent its adherence to the intestinal mucosa
    - IgG antibodies destroy *Salmonella* via opsonisation or activation of classical complement pathway

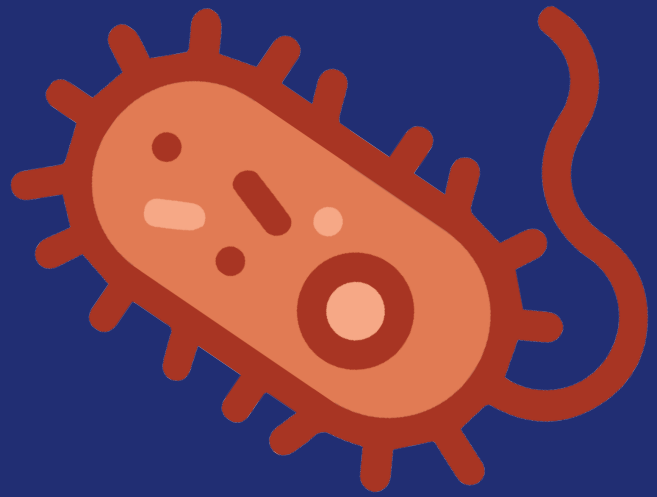


- CD8+ cytotoxic T cells (activated by CD4+ T helper cells) bind to MHC I molecules
    - CD8+ T cells detect *S. enteritidis* antigens and then release perforin and granzyme into the infected cells to destroy them via cell lysis



Host Damage

What damage ensues to the host from the immune response?

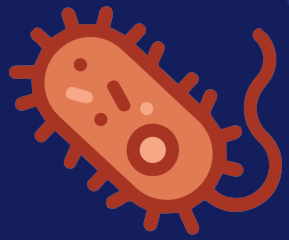


Oxidative Damage

- Activated neutrophils and macrophages release reactive oxygen and nitrogen species that cause inflammation and damage host tissue epithelium
    - Can cause DNA and RNA damage, potentially leading to cell-mediated apoptosis and death
- Inflammatory cytokines produced by host epithelial cells can also result in intestinal damage, fever, and abdominal pain

Secretion of Fluids

- Neutrophils also activate the release of prostaglandins
  - Prostaglandins increase the secretion of fluid and electrolytes
  - Causes diarrhea and leads to dehydration and electrolyte imbalance
- Cytokines (ex. TNF-alpha, IL-1, and IFN-gamma) released by macrophages and T lymphocytes during the infection can lead to fever

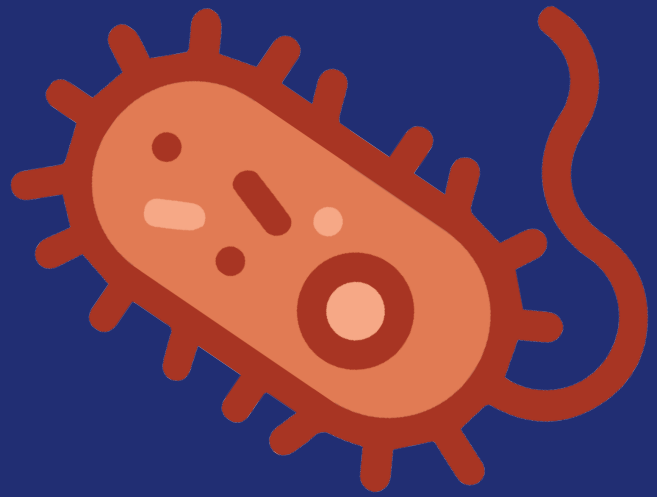


Extracellular Damage from CD8+ T cells

- CD8+ cytotoxic T cells target and destroy infected cells
  - This mechanism is highly controlled, but sometimes neighbouring tissues can be harmed in the process
  - Occurs via leakage of granzymes into the extracellular space, but is rare

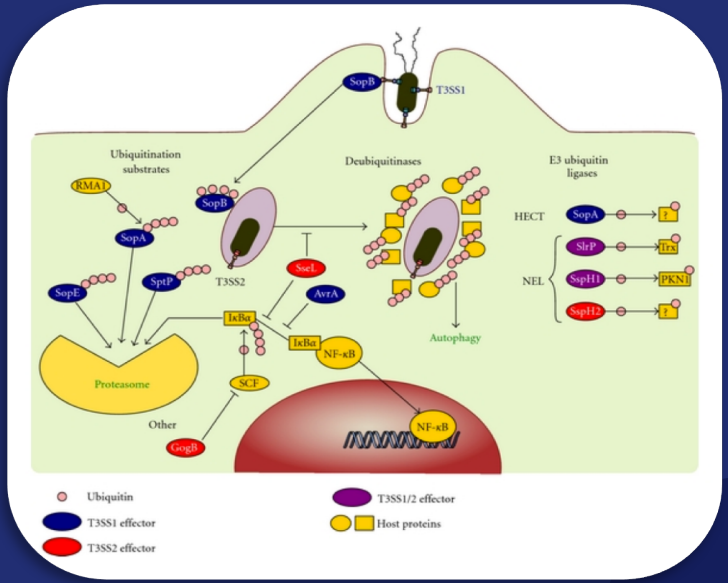
Bacterial Evasion

How does the bacteria attempt to evade these host response elements?



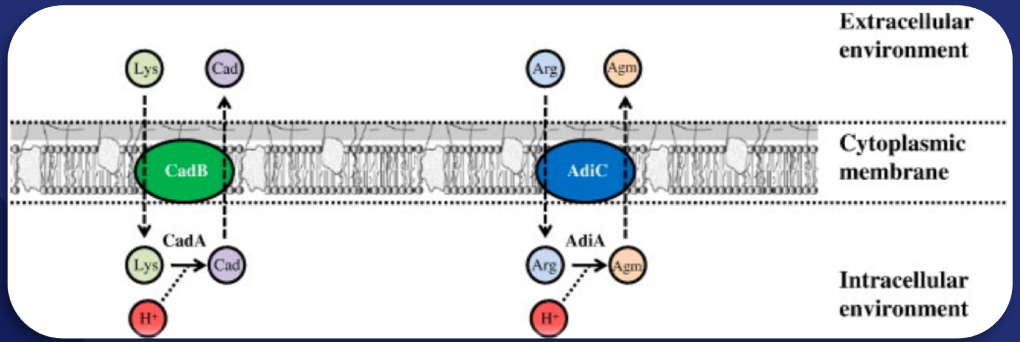
Salmonella enteritidis have virulence genes clustered in Salmonella Pathogenicity Islands (SPIs), to evade host defences

- SPIs use type III secretion systems to inject effectors (bacterial proteins) into host cells
- Effectors interfere with host cellular functions and thus promote pathogen growth



Innate Immunity Evasion

- In the low pH of the stomach acid, *Salmonella enteritidis* enacts an Acid Tolerance Response (ATR)
    - Uses acid shock proteins to shield bacterial proteins from damage and repair damaged proteins
    - Excess protons can be pumped out from *Salmonella* organisms via antiporters to maintain their intracellular pH



Innate Immunity Evasion

Salmonella also has ways of subverting the intestinal innate defenses in order to invade host cells and become an intracellular pathogen:

1. Has a bacterial cell envelope that prevents bile salts from entering
    - If bile does enter, *Salmonella* uses efflux pumps (ex. AcrAB-TolC efflux system) to export the bile out of their cytoplasm
2. Has glycosyl hydrolases to degrade glycan layers found within the mucus barrier
    - Uses glycan-degrading enzymes: NanH and MalS

Innate Immunity Evasion Cont'd.

3. Contains PhoQ (a histidine kinase), which phosphorylates PhoP
    - PhoP activation prevents dephosphorylation of PmrA, which is the protein that controls gene expression for LPS modifications that are used to resist antimicrobial peptides

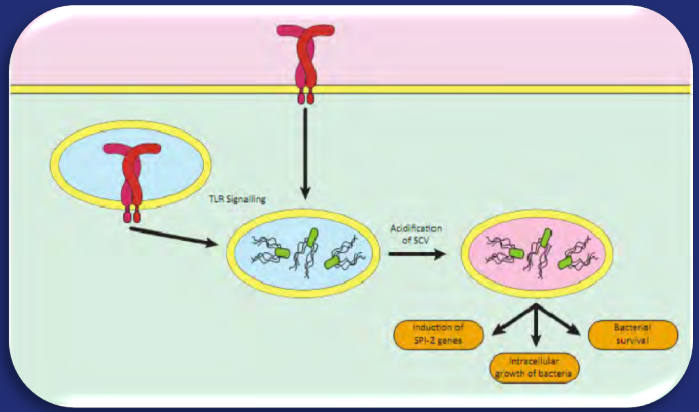
4. Can breach the tight junctions between mucosal epithelial cells by injecting T3SS-1 into the cells
    - This induces actin rearrangement and junction disconnection so *Salmonella* can enter intracellularly

- Host inflammatory immune responses can create conditions that allow *Salmonella* to have a growth advantage against normal gut flora

- If engulfed by phagocytic cells, *Salmonella* can reside within a phagosome, called a Salmonella-containing Vacuole (SCV)

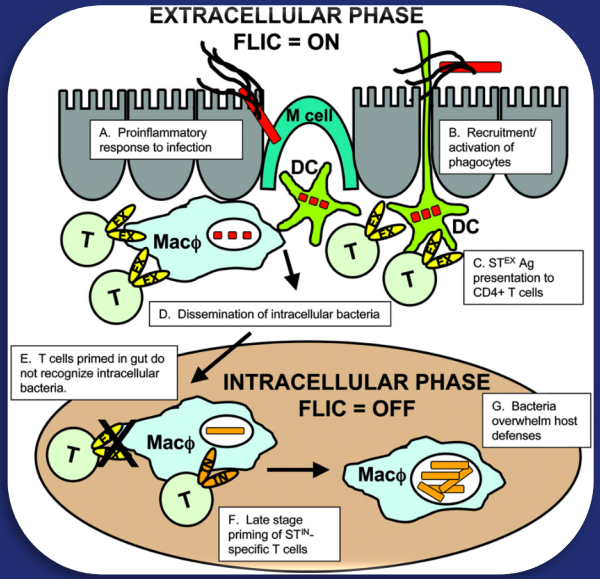
    - Helps it survive and proliferate due the absence of host defences

    - SCV is developed via effector protein activity, to prevent fusion and degradation by a lysosome



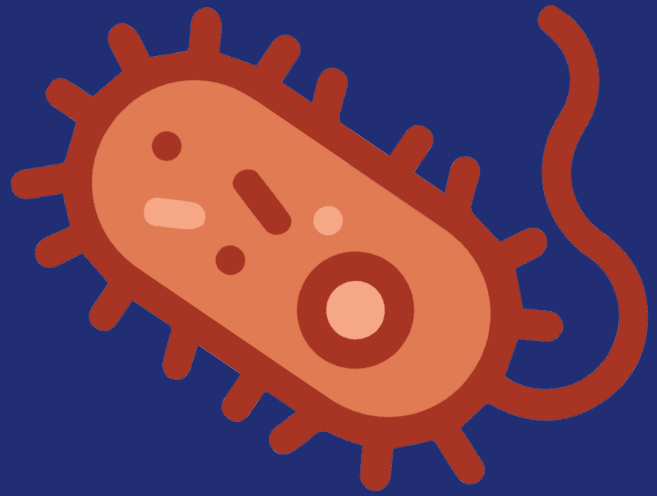
Adaptive Immunity Evasion

- S. *enteritidis* can subvert APC function by interfering with the presentation of their antigens on the MHC class I and II complexes
    - Can reduce the availability of its antigen FliC by decreasing FliC expression below the T cell activation threshold
    - This inhibits the activation of the T cells, and ultimately the adaptive immune response
- SteD (*Salmonella* protein) can supress B lymphopoiesis by reducing MHC class II molecules on CD4+ T helper cells
    - As a result, B cells cannot be activated and differentiated



Outcome

Is the bacteria completely removed, does the patient recover fully and is there immunity to future infections from this particular bacteria?



Recovery

- Most of the *Salmonella* enteric infections result in localized, acute gastroenteritis and can be controlled and eliminated by the host immune system
    - If needed, therapy through oral or IV fluids can be given to prevent dehydration and replenish electrolytes
- Healthy populations usually recover from symptoms within 2-7 days after the infection

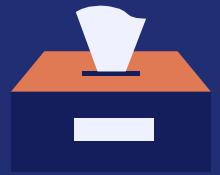
In vulnerable populations (the young, elderly, and immunocompromised), *Salmonella* can enter the bloodstream, resulting in septicemia and potential death if not treated

Treatment

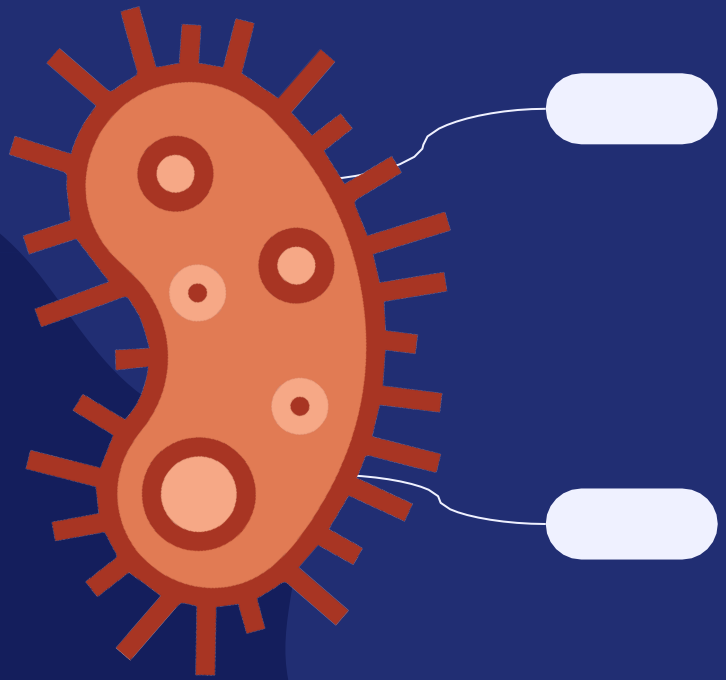
- Antibiotics are not usually prescribed in uncomplicated *S. enteritidis* infections because they:
    - Do not shorten the illness or resolve symptoms
    - Can increase development of antibiotic resistant stains
    - Can prolong periods of transmission
- Antibiotics can also alter host microbiota compositions and increase the susceptibility of *Salmonella* colonization in the intestine

Future Implications

- *Salmonella* can continue to shed in the stool of the infected individuals for extended periods of time, until the normal gut microbiota is restored
    - Until the normal gut microbiota is fully restored, there is reduced competition for colonization of the bacteria which increases the probability of reinfection
- Reinfection of *Salmonella enteritidis* via the fecal-oral route is less than 1%
    - Secondary infections by *Salmonella* are largely clonal



If a secondary infection of Salmonellosis occurred in an individual:



Memory T cells can rapidly release cytokines and activate macrophages in the intestine

B cells can present antigens to Salmonella-specific TH1 cells, which produce specific inflammatory cytokines and activate macrophages

Thank You