Microbiology

Doctor 2017 | Medicine | JU

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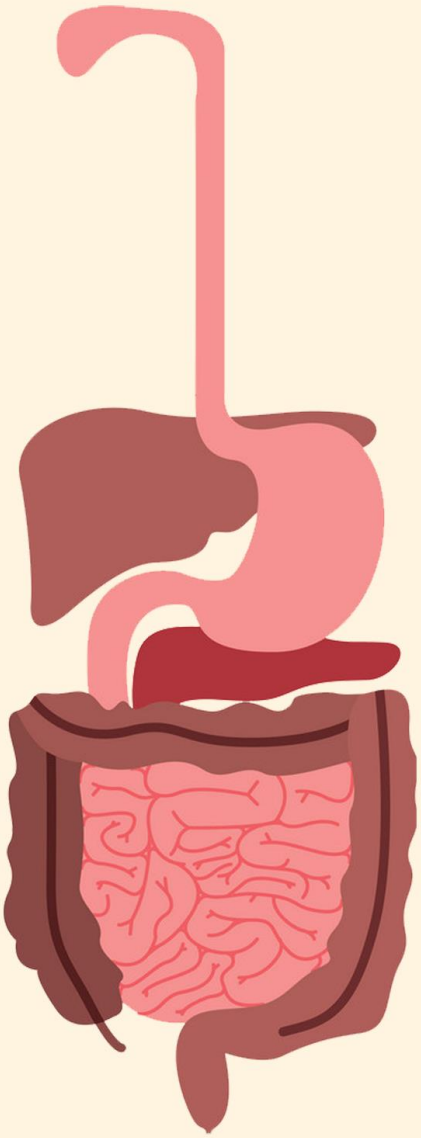


2nd system - GI

In this lecture we'll discuss the rest of the Enterobacteriaceae family in this course, Salmonella and Yersinia.

Salmonella

The bacteria of the Salmonella genus are motile (unlike shigella) non spore-forming G (-) bacilli, ubiquitous in nature but not normally part of our microbiome, and have a complicated taxonomy with various types of classification:

1. Family: Enterobacteriaceae → Genus: Salmonella → Species: Enterica → classified further into subspecies then serotypes.
   Salmonella Enterica is the species that causes human disease, the other species is called Salmonella Bongori and it is associated with other animals.
2. The second way of classification splits Salmonella into three main types:
   I. *S. Typhi*: the causative agent of typhoid.
   II. *S. Choleraesuis*: the causative agent of bacteremia and focal lesions.
   III. *S. Typhimurium*: the causative agent of gastroenteritis.
   There are 1500 different serotypes that cause bacteremia or gastroenteritis.
3. **The third way of classification is one of the simpler methods and this is the one we will use and be concerned with** (so just be familiar with the other two methods above, no need to memorize them)**, which is to classify them on one clinical aspect, whether they cause typhoid or not**:
   I. **Typhoid Salmonella**: includes all salmonella serotypes that cause typhoid/enteric fever, and they are four only: *S. Typhi*, *S. Paratyphi A*, *S. Paratyphi B*, & *S. Paratyphi C*.
   II. **Non-Typhoid Salmonella (NTS)**: all serotypes causing clinical presentation in humans other that typhoid, namely bacteremia with focal lesions, most commonly caused by *S. Choleraesuis*, or gastroenteritis commonly caused by *S. Typhimurium*.

   We have another group of infected individuals that concern us clinically, and those are the asymptomatic carriers. The salmonella in their bodies usually colonize their biliary tracts and those people are considered as reservoirs for typhoid disease.

Humans are the reservoirs for Typhoid Salmonella (and Shigella too), while animals are the reservoirs of other salmonella serotypes. Transmission can be by direct contact or by indirectly eating or drinking contaminated food/water, main examples are **poultry/dairy** contaminated by human/animal excreta. Birds and reptiles are usually inhabited by NTS.

Shigella, Salmonella, and Yersinia are not normally part of the human microbiome and are **always considered pathogenic** if found there.

Typhoid Fever: a severe *systemic* disease caused by *S. Typhi*, *S. Paratyphi A*, *B*, and *C*. Typhoid fever is *the most severe* presentation of salmonellosis. The characteristic feature of typhoid is that it is a *month-long fever*. Typhoid is a *major cause of morbidity/mortality worldwide*. Like shigellosis, it is *strictly a human disease* and must be traced back to a *human reservoir*. One of the problems with controlling typhoid is the *presence of asymptomatic carriers* (harbour the bacteria in their biliary tracts) since symptomatic clinical cases can be isolated to prevent spread of the infection. Imagine if the asymptomatic carrier was food handler!

- Pathogenesis: First, salmonella is ingested by *food/water contaminated by human excreta*. Fortunately, salmonella are ***sensitive to the stomach's high acidity*** and to cause disease, the infectious dose must be high enough (10^6 organisms are needed to establish the infection). If it is, the salmonella reach the small intestine and **invade the *M cells of the peyer's patches*** in the ileum via *vacuoles*. Till now this is similar to the pathogenesis of shigella. **But while shigella then escape their vacuoles and then spread laterally through the mucosa, salmonella stay in their vacuoles and invade further vertically till they reach the lamina propria → *enter lymphatic circulation* and reach lymph nodes→ spread throughout the body carried by macrophages → reach blood through thoracic duct → reach RES** (Reticule-Endothelial System) **→ endotoxin causes prolonged fever**. **Shigella** are spread by *PMNs instead of macrophages.*

  Note: the capsular antigen of salmonella is the Vi antigen not the K antigen like other Enterobacteriaceae members.

- Clinical Manifestations: the incubation period is from **1-2 weeks.**

  1st week: fever, malaise, increase in temp is gradual then plateaus (becomes constant).

  2nd week: high fever, fatigue, cough, rose spots and rash on chest/back. Abdominal symptoms not very prominent if present.

  3rd/4th week: If no complications arise, symptoms & signs gradually resolve on their own.

  In the pre-antibiotic era, the chief complications of enteric fever were intestinal hemorrhage and perforation, and the mortality rate was 10–15%. After abX, mortality decreases to less than 1%.

Gastroenteritis: is *the most common* presentation of salmonellosis (*typhoid is the most severe*) and is most commonly caused by *S. Typhimurium*. Transmission is from an animal reservoir typically poultry/dairy products (ex. **eggs/mayo**) but *S. Typhimurium can also contaminate vegetables and fruits.*

- Symptoms: start rapidly (within 48 hrs) and include fever, nausea, abdominal cramps, vomiting and bloody diarrhoea (diarrhoea can be not bloody). *Bloody diarrhoea is a main symptoms of salmonella infection (because salmonella invades enterocytes → bloody diarrhoea).*
  Bacteraemia is rare unless the patient is immunocompromised.
- Tx: supportive care. Symptoms resolve on their own within one week and *we don't normally give abX unless the patient is immunocompromised, elderly, or an infant.*

Bacteraemia w/ focal lesion: associated commonly with *S. Choleraesuis* but may be caused by any salmonella serotype. After oral infection, there is early invasion of the bloodstream. The most common site for focal lesions in this type of salmonellosis is in the meninges of the brain → **meningitis**, followed by the bone → **osteomyelitis**. **The group most affected by salmonella-caused osteomyelitis is sickle-cell anemia patients**.

Bacteremia → positive blood culture for salmonella.

Asymptomatic carriers: carrier state usually develops post-treatment where the salmonella reside in the biliary tracts even after symptoms disappear, especially if **gallstones** are present. **Shigella and salmonella can resist bile salts while other enteric bacteria don't.*

Diagnosis: same method for all clinical types; culture from a specimen then serology.

1. Choosing a specimen: specimens can be taken from blood, stool, or bone marrow. Those usually give a positive result if taken they were taken in the **2nd** week of infection in typhoid fevers or bacteraemia. Enterogastritis *stool* samples however can give a positive result from the first week.
   A positive culture of duodenal contents indicates the presence of salmonellae in the biliary tract in carriers.
2. Culture: many sequential cultures are used to isolate the salmonella from the rest of any enteric bacteria that may be present in the specimen.
   I. First, **enrichment cultures** are used: the specimen (usually stool) is put into selenite F or tetrathionate broth, both of which **inhibit replication of normal intestinal bacteria and permit multiplication of salmonellae**.

II. Next, **differential and selective** medium cultures for salmonella are used: EMB (eosin methylene blue), MacConkey, or deoxycholate medium agar is used because they differentiate between lactose and non-lactose fermenters. Salmonella (and shigella) are non-fermenters.
Salmonella-shigella (SS) agar, Hektoen enteric agar and xylose-lysine decarboxylase (XLD) agar, all isolate salmonella and shigella only.

MacConkey agar: Complex medium used to isolate G (-) rods that typically reside in the intestine. Selective because bile salts and dyes inhibit G (+) organisms and G (-) cocci. Differential because the pH indicator turns pink-red when the sugar in the medium, lactose, is fermented.

III. Finally, we use methods that differentiate between shigella and salmonella. This method can be structural (*salmonella are motile while shigella are not*), biochemical (*salmonella produce gas & acid in culture while shigella don't*), or by serology.

3. Serology: based on antigen/antibody rxns. There are two main types of serologic tests:
   I. Agglutination test: particularly useful for rapid preliminary identification of cultures. In this test, the *unknown culture is mixed with lab-prepared **antibodies** for salmonella O antigens on a slide*. Clumping, if the culture contained salmonella antigens, can be observed within a few minutes.
   II. Tube dilution agglutination test (Widal test):
   Instead of having lab-prepared antibodies, the Widal test uses lab-prepared **antigens** and depends in its diagnosis on *binding to antibodies -in the specimen- against salmonella*. You can easily see now why this test might give a lot of false positives, since *antibodies against an antigen can stay in the body for years after an infection*. Therefore, this test was long abandoned in the developed world. It is still in use in developing countries because it is cheaper that the other test.

Immunity: although the Dr mentioned that immunity won't be included in our material, he did go over a few points here so I will write them here for anyone interested or with enough time.

NTS usually don't confer any level of immunity while typhoid salmonella do. Two types of immunity are observed:

- Secretory IgA antibodies may prevent attachment of salmonellae to intestinal epithelium.
- Cell-mediated immunity since it is an intracellular infection.
- Also important to remember is that persons with **sickle cell disease** are exceedingly susceptible to salmonella infections, particularly **osteomyelitis**.

Treatment:

Gastroenteritis is treated only with electrolyte and fluid replacement therapy. Again, symptoms resolve on their own within one week unless the patient is immunocompromised and we don't normally give abX unless the patient is immunocompromised, elderly, or an infant.

In the case of typhoid salmonella however, abX must be administered. We usually use the newest generations of cephalosporins (ex. *Cefotaxime*) as well as fluoroquinolones (ex. *Ciprofloxacin*). Remember that the use of antimicrobials lowered the mortality rate from 30% to less than 1%.

Some chronic carriers have been cured by ampicillin alone, but in most cases cholecystectomy (removal of the gallbladder) must be combined with drug treatment.

Prevention and Control:

- Thoroughly cook food.
- Treat carriers as mentioned above.
- A **vaccine** is present for typhoid salmonella, it is either a live-attenuated oral vaccine or Vi capsular antigens (IM), however its **effectiveness is only 50-70%** and **protection is limited to a few years only.** It's recommended for travellers to endemic regions

Yersinia

Are *G(-) non-spore forming bacilli*. There are three main types: *Y. pestis*, the causative agent of the plague, *Y. enterocolitica*, which causes yersinosis gastroenteritis, and *Y. pseudotuberculosis*. Spread of infection is by an animal reservoir.

Yersinia are able to grow at low temperatures (it is motile at low temperatures, grow best at 25°C and are motile at 25°C, but non-motile at body temperature), which is one of their differential characteristics from the very similar shigella and salmonella, and a method used in their diagnosis. Another defining feature is that they present bipolar staining, where the stain concentrates at the poles of the bacillus while the middle is left faint-coloured.

A slight note on the above paragraph is that yersinosis is commonly associated with blood transfusions, since their growth is not inhibited by low temperatures like most other disease-causing bacteria.

We'll concentrate now on both *Y. enterocolitica* and *Y. pseudotuberculosis*. **Both** can cause ***mesenteric lymphadenitis and terminal ileitis***, which cause right ileac fossa pain, mimicking appendicitis pain → **pseudoappendicitis (refer to sheet Patho 6 page 10)**. The liver and spleen can also be involved after oral infection.

A small extra word on *Y. enterocolitica*: the most common human disease-causing serotypes are O:3, O:5, and O:9.

Pathogenesis: almost the same invasion process as shigella and salmonella, in that Yersinia invades M cells of peyer's patches of the ileum, but the whole process is yet to be completely understood. The most important virulence factors are:

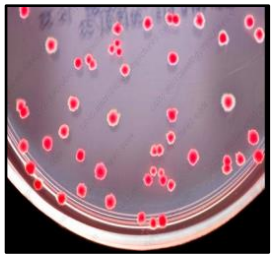
- Enterotoxin,
- YOPs (Yersinia Outer-membrane Proteins), which are secreted by
- Type III secretion systems, directly into the cytoplasm of host cells., and
- Pathogenicity island (PAI) that encodes for an iron scavenging siderophore.

Yersinia causes microabscesses and ulcerations → bloody diarrhoea.

Clinical manifestations: we've already discussed pseudoappendicitis. Gastroenteritis presents with bloody diarrhoea, abdominal cramps, nausea, and vomiting. *Diarrhoea is self-limiting.* Gastrointestinal complications include granulomatous appendicitis.

Diagnosis:

1. Specimen: may be stool, blood, or material obtained at surgical exploration.
2. Culture: the number of Yersinia in stool may be small and can be increased by "cold enrichment" (use enrichment agar and incubate at low temperatures → only Yersinia grow in number).
    a. Cultures can be on *MacConkey agar since Yersinia are non-lactose fermenters.*
    b. Alternatively, most labs use a more Yersinia selective agar such as *cefsulodin-Irgasan-novobiocin* agar (CIN: a combination of **three antimicrobial that inhibit the growth of all bacteria but Yersinia**) incubated at room temperature for several days →Y enterocolitica colonies give pink/red bull's eye appearance.



3. Serology: **NOT** reliable because of cross-reactions between Yersinia and other organisms like vibrios, salmonellae, and brucellae which give false positives.

Treatment: the role of abX in yersinosis is controversial, especially in yersinosis gastroenteritis. However, abX are administered in cases involving *Y. pseudotuberculosis*, abX such as ampicillin, cephalosporins, chloramphenicol, and tetracyclines are administered since *Y. pseudotuberculosis* can be susceptible to these drugs, unlike *Y. enterocolitis* because they produce β lactamases which provide them with drug-resistance. This can be used in cultures to **differentiate** between *Y. pseudotuberculosis* and *Y. enterocolitis*.

Systemic infections with bacteraemia or focal infections outside the gastrointestinal tract generally require antimicrobial therapy. Fluoroquinolone therapy is effective for bacteraemia in adults; for example, ciprofloxacin, a third-generation cephalosporin is an alternative.

Prevention and Control:

- Proper hygiene
- Proper and thorough cooking
- Avoid raw meats.

Remember: Shigella, Salmonella, and Yersinia all have the differential diagnosis of bloody diarrhoea due to invasion, also considered triggers for Reiter's syndrome (reactive arthritis).

The manifestations of reactive arthritis include the following triad of symptoms: an inflammatory arthritis of large joints, inflammation of the eyes in the form of conjunctivitis or uveitis, and urethritis in men or cervicitis in women.