CHAPTER 31

Approach to Diarrhea in Returned Travelers

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Gastrointestinal infections are the most common illnesses in travelers, occurring in 34% of all travelers, typically those traveling from high-income countries to low and middle-income countries. Traveler's diarrhea (TD) is defined as three or more unformed stools per day and at least one additional gastrointestinal symptom, such as nausea, vomiting, and abdominal pain, and possibly systemic findings including fever and malaise. The majority of TD is commonly attributed to enterotoxigenic strains of *Escherichia coli* (ETEC) and occurs within the first 2 weeks of travel with a mean duration of 4-5 days. Since most patients travel for at least 1 week, a medical provider will seldom see a case of new-onset diarrhea in a returned traveler. Instead, the majority of cases of travel-acquired diarrhea seen by the medical establishment will be protracted or recurrent. Approximately 10% of patients with TD will experience symptoms for more than 1 week, and in 5-10%, symptoms will last for 2 weeks or longer. This chapter will focus on the evaluation of TD that is likely to present to the healthcare system.

EXPOSURE HISTORY AND RISK FACTORS

Many factors influence the acquisition of TD, including geographic location, type of travel, and host factors. Of these factors, travel destination and duration are the most important determinants of attack rate. Highest risk of TD is associated with travel for more than 2 weeks to South and Southeast Asia, sub-Saharan Africa, the Middle East, and Latin America. Gathering information regarding the patient's travel arrangements is important in determining if the patient is suffering from TD and may narrow the list of potential pathogens. While ETEC and enteroaggregative *E. coli* (EAEC) are the most common causes of TD, especially in Africa and Latin America, the invasive pathogens *Campylobacter*, *Vibrio*, and *Salmonella* are just as common in South and Southeast Asia. High rates of TD have been associated with those visiting family and relatives, trekkers and campers, and travelers on adventure tours or cruise ships.

Information about consumption of street food and local water sources increases the risk of TD but is unhelpful in ascertaining the TD etiology. A history of shellfish or seafood consumption may be useful, as they are common sources of *Vibrio*. In addition, hepatitis A virus and noroviruses may be acquired through contaminated shellfish. Acute hepatitis A viral infection is associated with an average of 30 days of missed work and 1% mortality in adults.

Direct contact with animals should raise the suspicion for *Campylobacter*, shiga toxin–producing *E. coli* (STEC), and *Giardia* infections. Treatment with antibiotics for TD adds the risk of antibiotic-associated diarrhea caused by *Clostridium difficile*. If antibiotics were administered and an initial response obtained, the recurrence of symptoms may represent recrudescence of resistant bacteria, reinfection with similar or different pathogens, or a post-infectious process. Reviewing the patient's vaccination history is also important; those

vaccinated against rotavirus and hepatitis A would be protected against infections caused by these agents. However, given the relatively low efficacy of typhoid fever vaccine (50-80% for both the oral and injectable forms), this diagnosis should remain in the differential diagnosis for returned travelers with signs and symptoms consistent with this.

Host factors can also play a role in the risk of TD, including age, reduced gastric acidity, blood type group O, and other genetic factors. Infants and toddlers can acquire infection though oral contact with nonfood items. Recent studies have suggested that individuals who produce higher amounts of the inflammatory mediators interleukin 8 AA, lactoferrin, and interleukin 10 may be more susceptible to TD. Polymorphisms in the CD14 receptor and osteoprotegerin have also been associated with increased susceptibility to TD. Immunocompromised individuals are at risk for prolonged illness with typical agents and infection with atypical organisms such as microsporidia.

Finally, the likelihood of the patient transmitting infectious diarrhea to other contacts must be explored. Food handlers and institutional caregivers with any type of diarrhea, including TD, should be evaluated regardless of the length of symptoms and their employment deferred until symptoms resolve to avoid a potential outbreak.

CLINICAL PRESENTATION AND EVALUATION

The severity of the illness dictates the level of evaluation required of an ill patient with diarrhea. The first consideration to be made is the need for hospitalization. The combination of orthostatic hemodynamic changes and an inability to maintain oral rehydration necessitates intravenous rehydration and possible hospitalization. If significant systemic toxicity is present, stool and blood cultures should be obtained along with initiation of empiric parenteral antibiotics.

That being said, the average case of TD is not severe (averaging 4.6 stools/day), and symptoms are often those of nausea and cramping abdominal pain. The presence of high fever and/or blood or pus in stool suggest an invasive pathogen such as *Salmonella*, *Shigella*, *Campylobacter*, STEC, or *Entamoeba histolytica* and decrease the likelihood of non-invasive pathogens such as ETEC, norovirus, *Giardia*, and *Vibrio* spp., which tend to cause profuse watery diarrhea and abdominal cramps, without fever or bloody stools. Blood may not be present in all patients infected with invasive organisms, and concurrent infection with more than one enteric pathogen may occur. Persistent watery diarrhea for longer than 14 days, without fever, suggests giardiasis as well as other parasitic infections such as *Cryptosporidium*, microsporidia, and *Cyclospora*.

The physical examination does not usually aid in determining the etiology of travel-acquired diarrhea, although it is useful to assess the general condition of the patient and to exclude other conditions that may present with diarrhea. In TD, the abdomen is typically not tender to palpation, and bowel tones are hyperactive. Focal abdominal tenderness dictates expanding the differential diagnosis to include appendicitis, biliary disease, peptic ulcer disease, pancreatitis, diverticulitis, small bowel perforation and inflammatory bowel disease (IBD). Hepatic tenderness in a traveler with diarrhea could be suggestive of acute viral hepatitis or amebic liver abscess. Unrecognized chronic human immunodeficiency virus infection may present with gastrointestinal complaints. Occult or gross blood in the stool indicates the presence of an invasive organism. If only non-invasive pathogens are identified in stool samples, a non-infectious cause for gastrointestinal bleeding should be considered, including IBD and malignancy.

Physicians seeing travelers with chronic gastrointestinal complaints need to maintain an index of suspicion for previously unrecognized gastrointestinal disease, particularly IBD and irritable bowel syndrome (IBS). Infections with *Salmonella*, *Shigella*, or *Campylobacter* may trigger or exacerbate IBD. Ulcerative colitis or Crohn disease should be suspected in patients with bloody diarrhea, accompanied by systemic signs such as weight loss, oral or perianal lesions, and extraintestinal manifestations including arthropathies or ophthalmologic symptoms. IBD becomes more likely when symptoms have persisted more than 2 months without a microbiologic diagnosis. IBS is one of the most common post-infectious diarrhea processes

TABLE 31.1 Potential Etiologies of Diarrhea in Returned Travelers Suggested by Clinical Presentations

Clinical Presentation	Microorganisms to Consider as Etiologies
Non-inflammatory diarrhea	Enterotoxigenic *Escherichia coli*
Acute watery diarrhea	Enteroaggregative *E. coli*
Absence of fever	*Vibrio cholera*
No blood or fecal WBCs	*Aeromonas*
	Noroviruses and other enteric viruses
	Giardia
	Cryptosporidium
	Cyclospora
Inflammatory diarrhea	*Salmonella*
Grossly bloody stool	*Campylobacter*
Fever present and other systemic symptoms	*Shigella*
Fecal WBCs present	Shiga-toxin producing *E. coli* (*E. coli* O157:H7)
	Vibrio parahaemolyticus
	Yersinia enterocolitica
	Plesiomonas shigelloides
	Entamoeba histolytica
Persistent diarrhea (lasting ≥14 days)	*Cyclospora*
	Cryptosporidium
	Entamoeba histolytica
	Giardia
	Microsporidia (immunocompromised)

WBCs, White blood cells.

in returning travelers and may be associated with intermittent cramping pain, bloating, and gas. If IBD or IBS is a consideration, further evaluation should be performed in consultation with a gastroenterologist.

DIAGNOSTIC STUDIES

The decision to perform laboratory studies in the traveler with diarrhea should be based on the duration of symptoms, severity of the illness, and type of diarrhea present: inflammatory (blood or white blood cells present) versus non-inflammatory (primarily watery), as outlined in **Table 31.1**. The traveler seeking medical attention who has had fewer than 5 days of diarrhea does not require investigations unless there is significant fever, abdominal pain, dehydration, blood or mucus is present in the stool, or if the patient is immunocompromised. Before ordering stool studies, it is important to be aware of the capabilities and protocols of the clinical microbiology laboratory at your institution, including testing options, recommended stool sample volume, and preferred stool specimen transport method. **Table 31.2** lists the common causes of TD, associated symptoms, and diagnostic testing.

It is important to educate patients on proper collection of stool samples for laboratory testing, as submission of improperly collected specimens will compromise testing results. The stool specimen should be caught directly in a standard pint-sized specimen container or in a larger clean, dry container. Fecal specimens contaminated with urine or toilet paper or retrieved from the toilet bowl are not satisfactory. It is ideal for stool specimens to arrive in the laboratory within 1-2 hours post collection to ensure reliability of microscopy and culture results. If this is not feasible, specimens for bacterial culture should be placed in an

TABLE 31.2 Etiologic Agents of Diarrhea in Returned Travelers

Etiology	Incubation Period	Signs and Symptoms	Duration of Illnesses	Associated Exposure	Laboratory Testing
Aeromonas	Unknown	Abdominal cramps, watery diarrhea	2-10 days	Contaminated meats and water	Routine stool culture
Campylobacter jejuni	2-5 days	Fever, cramps, vomiting, diarrhea (possibly bloody)	2-10 days	Undercooked poultry, unpasteurized milk, contaminated water	Routine stool culture; requires special media and incubation conditions
Clostridium difficile	5 days to 5 weeks	Fever, abdominal cramps, diarrhea	4 days to weeks	Colonization and prior antimicrobials	Immunoassays or NAT for toxins A and B
Shiga-toxin producing *E. coli*	1-8 days	Abdominal pain, vomiting, severe, often bloody, diarrhea	5-10 days	Undercooked beef, unpasteurized milk, contaminated water and produce	Stool culture Immunoassays or NAT for Shiga toxin
Enterotoxigenic *E. coli*	1-3 days	Abdominal cramps, watery diarrhea	3-7 days	Contaminated water or food	Routine stool culture not useful. Multiplex NAT Specific testing by state or public health laboratories
Plesiomonas shigelloides	24-28 h	Abdominal pain and cramping, watery diarrhea	5-14 days	Contaminated seafood and water	Routine stool culture
Salmonella enteritidis	1-3 days	Fever, abdominal cramps, vomiting, diarrhea	4-7 days	Eggs, poultry, milk, raw fruits and vegetables	Routine stool culture
Shigella spp.	24-48 h	Fever, abdominal cramps, and diarrhea	4-7 days	Contaminated food or water. Also spread person to person	Routine stool cultures

Vibrio cholerae	24-72 h	Severe dehydration due to profuse watery diarrhea and vomiting	3-7 days	Contaminated water, fish, shellfish, street-vended food	Stool culture with specific request for isolation Antigen immunoassay
Vibrio parahaemolyticus	2-48 h	Watery diarrhea, abdominal cramps, nausea, vomiting	2-5 days	Undercooked or raw seafood	Stool culture with specific request for isolation
Vibrio vulnificus	1-7 days	Vomiting, diarrhea, abdominal pain	2-8 days	Undercooked or raw seafood	Stool cultures; request specific testing
Yersinia enterocolitica	24-48 h	Fever, abdominal pain, vomiting, bloody diarrhea (mimics appendicitis)	1-3 weeks	Undercooked pork, milk, water	Stool culture with specific request for isolation
Norovirus	12-48 h	Fever, myalgia, abdominal cramping, nausea, vomiting, diarrhea	12-60 h	Shellfish, contaminated water, contact with infected individuals	Not routinely available. Immunoassay and NAT by public health labs
Rotavirus	24-72 h	Fever, nausea, vomiting, watery diarrhea	4-10 days	Contact with infected individuals	Immunoassay

Adapted from Center for Disease Control. Diagnosis and management of foodborne illnesses: a primer for physicians and other healthcare providers. *MMWR.* 2004;53(RR-4):1–33.
NAT, Nucleic acid testing.

appropriate transport medium, such as Cary-Blair. When collecting stools to be examined for parasites, patients should be instructed to collect only one stool sample per day and at least three samples within a 10-day period if the first specimen is negative. It is recommended that stool specimens for ova and parasite examination be sent to the laboratory in a fixative. Preservative kits usually contain two different fixatives: formalin to preserve helminths and coccidian parasites and polyvinyl alcohol for staining to visualize protozoa. Single vial preservatives are now available, such as the alcohol-based Ecofix™ (Meridian Bioscience, Cincinnati, OH), which can be utilized for both the examination of helminths and staining for intestinal protozoa. It is important to note that the transport media used for bacterial cultures are not suitable for parasite examination and that fixed specimens cannot be used for bacterial culture. Thus, if bacterial culture and parasite examination are desired, separate fecal specimens are required, each in the appropriate transport container.

The determination of white blood cells (WBC) in the stool can be helpful in the evaluation of patients with diarrhea. The presence of WBC suggests an inflammatory process in the gastrointestinal tract, possibly due to an invasive infection. The detection of fecal leukocytes in stool samples can be achieved by microscopic examination of a stool smear stained with methylene blue or by using a commercially available enzyme immunoassay (EIA) for lactoferrin. The sensitivity/specificity of fecal leukocytes by microscopy or lactoferrin for predicting inflammatory diarrhea are 73%/84%, and 92%/79%, respectively.

TD studies have shown that the most commonly identified bacterial pathogens are ETEC, *Salmonella*, *Shigella*, and *Campylobacter*, which account for 45-50% of cases. Because bacteria are the most commonly reported cause of TD, stool culture has traditionally served as the backbone for a TD diagnostic work-up. While stool cultures are a commonly ordered test, they are *not* performed the same way at every hospital laboratory. Stool cultures are designed to optimize the recovery of *Salmonella*, *Shigella*, *Campylobacter*, and STEC, including *E. coli* O157:H7. Some laboratories will also isolate *Aeromonas*, *Plesiomonas*, *Yersinia*, and/or *Vibrio* from stool, whereas others will do so only on special request. In view of this, it is crucial to know how your laboratory performs stool cultures to ensure that the enteric pathogens being considered will be recovered. It is important to note that the most common cause of TD, ETEC, is difficult to isolate in stool culture, as there is no reliable way to discriminate between ETEC and endogenous *E. coli* by culture alone. Nucleic acid testing (NAT) platforms are available that will detect ETEC in stool samples as a single target or as part of a multiplex panel along with *Salmonella*, *Shigella*, and *Campylobacter*. Many laboratories will include an antigen-based assay or NAT to detect Shiga toxin in all stool cultures to aid in detecting STEC. However, STEC is an uncommon cause of TD, although it has been seen in outbreaks such as the one in Germany and France in 2011. Another illness to consider in patients with TD is *Clostridium difficile* colitis, based on history of antibiotics use or previous *C. difficile* infections. Detection of *C. difficile* from stool specimens is achieved by detecting toxins A and B using commercially available EIA or NAT methodologies. In addition to feces, cultures of blood, bone marrow, and/or urine samples should be performed in patients with symptoms consistent with disseminated *Salmonella*. Two negative stool cultures from specimens collected on separate days, rules out the majority of bacterial pathogens in patients with diarrhea.

Identification of bacteria and yeast using matrix-assisted laser desorption ionization time of flight mass spectroscopy (MALDI-TOF MS) has become increasingly available in clinical laboratories throughout the world including the United States. Currently, there are two commercially available systems: VITEK® MS (BioMérieux Inc.) and the MALDI Biotyper CA System (Bruker Daltonics Inc.). Each system includes a mass spectrometer, software, and database including a list of microorganisms that are cleared for identification. MALDI-TOF MS based bacterial identification takes minutes to perform and is significantly cheaper to operate than nucleic acid based identification methods such as 16S rRNA sequencing and multiplex panels, leading to a de-emphasis of biochemical-based bacterial identification. However, despite the robust nature of this technology, it cannot reliable differentiate E.

coli from Shigella spp as these are highly related organisms at the molecular level. Thus, there may always be a need for biochemical identification in certain situations.

One major advantage of isolating stool bacterial pathogens is the ability to perform antimicrobial susceptibility testing. This is particularly important in travelers returning from parts of the world known to harbor multidrug-resistant (MDR) bacterial pathogens. MDR *Salmonella* have become more common in Asia and sub-Saharan Africa. *Campylobacter* resistance to fluoroquinolones has become a concern in Southeast Asia, with resistance rates up to 80%. Additionally, Enterobacteriaceae producing extended spectrum beta-lactamases and, more recently, carbapenem-resistant Enterobacteriaceae are becoming more prevalent in many parts of the world.

Parasitic infections are estimated to be responsible for 5-10% of TD cases. The index of suspicion for an enteric parasite should increase in travelers with diarrhea lasting longer than 1 week. Microscopic examination of stool for ova and parasites indicating infection is performed by examination of a stool specimen on two glass slides prepared using two different methods. One slide is prepared using concentrated, preserved stool and examined with or without the addition of iodine, looking for helminths and their eggs, or ova. The second slide is of preserved stool stained with trichrome, which facilitates identification of intestinal flagellates and amebic parasites. Microsporidia, *Cryptosporidium*, *Cystoisospora*, and *Cyclospora* are best visualized using special stains other than trichrome. If these organisms are suspected, the laboratory must be notified so the appropriate stains are included. Reliable detection of parasites by microscopy requires examination of multiple (at least three) fecal specimens, since parasites may be excreted intermittently and infections with multiple parasites may not be detected with one or two specimens. *Strongyloides* may require up to seven stool examinations and the use of special methods such as the Baermann technique or agar plate culture.

As an adjunct to microscopic stool examination, many laboratories utilize EIAs or fluorescent antibody tests for the detection of *Giardia* and *Cryptosporidium* antigens in stool because they are more sensitive than microscopic examination and are easier to perform. Stool antigen testing typically requires only one specimen to be diagnostic and effectively rules out infection with two negative results from separate specimens collected on different days. The sensitivity and specificity of *Giardia* EIAs are greater than 95%, while the performance of the *Cryptosporidium* EIAs are more variable, with sensitivity and specificity ranging from 80 to 99%.

It is estimated that 5-15% of cases of TD are due to viral infections, the majority of which are norovirus and rotavirus. Diagnostic work-up in these patients is unnecessary, since viral infections are self-limited and there are no directed therapies that will shorten the duration of illness or decrease viral shedding. Immunocompromised individuals are a patient group in whom diagnostic work-up may be beneficial, because determining the etiology of the diarrhea would limit further diagnostic work-up. In travelers in whom viral diagnostic work-up is warranted, stool can be assessed for rotavirus using commercially available EIAs; some laboratories offer NAT for norovirus and rotavirus. Hepatitis A is another viral cause of diarrhea that would be important to consider in unvaccinated individuals and is diagnosed using serology.

The causes of protracted diarrhea and ongoing gastrointestinal complaints will usually reveal themselves after clinical examination and laboratory studies outlined in this section. However, in approximately 30% of cases of traveler's diarrhea, no pathogen can be identified despite multiple, thorough laboratory evaluations. In these instances, consultation with a gastroenterologist is recommended to assist with further diagnostic evaluation. A complete blood count should be obtained to assess for signs of systemic inflammation, suggested by a high WBC and/or eosinophilia, and to assess for anemia from ongoing gastrointestinal blood loss or malabsorption. If tropical sprue or other malabsorption syndromes are being considered, testing for lactose tolerance and D-xylose absorption may be considered. If deemed appropriate, a gastroenterologist can perform a colonoscopic examination to inspect the mucosa and obtain biopsies to assess for Crohn disease, ulcerative colitis, schistosomiasis, and amebiasis.

Currently, there are three FDA-cleared, multiplex NAT panels available for the detection of enteric pathogens from stool specimens: xTag® GI Pathogen Panel (Luminex Corporation, Toronto, Canada), the FilmArray™ GI panel (BioFire, Inc., Salt Lake City, UT), and Verigene® Enteric Pathogens (EP) Test (Nanosphere, Inc., Northbrook, IL). All three panels detect bacterial pathogens commonly isolated in stool culture (*Campylobacter* spp., *Salmonella* spp., *Shigella* spp., *Vibrio* spp., and *Yersinia enterocolitica*) and the enteric viral pathogens Norovirus and Rotavirus. The xTag® and FilmArray™ GI panels can also detect the protozoan parasites *Cryptosporidium* spp., *Entamoeba histolytica*, and *Giardia lamblia*. These assays yield results within hours and require less hands-on time than the laborious, conventional methods for identification of bacteria and parasites. However, these assays may be cost prohibitive for some laboratories and patient populations and none of the currently available multiplex NAT assays can detect helminths. While molecular assays offer improved sensitivity over culture, use of these assays remains controversial due to concerns regarding specificity, clinical correlation and lack of an isolate for antimicrobial susceptibility testing and subtyping analysis to support identification during outbreak investigations. It is important to maintain an open dialog with the clinical microbiology laboratory to ensure that the tests being ordered are the appropriate tests for the patient.

INFECTIOUS DIARRHEA SYNDROMES

The goal of this section is to highlight the important features of bacterial diarrhea syndromes caused by pathogens that may cause protracted symptoms and are more likely to be encountered by a healthcare professional evaluating a patient with TD. The invasive bacterial pathogens *Salmonella*, *Shigella*, and *Campylobacter* are more common in patients with prolonged diarrhea and will be the focus of this section. In addition, several bacterial pathogens that are rare but important causes of TD and those that are emerging organisms of interest, including viral agents, will be discussed. Parasitic infections are discussed in Chapter 32 and will not be addressed in this section. Antibiotic resistance trends will be mentioned, but specific antibiotic treatment options are discussed in Chapter 8.

SALMONELLA

Salmonelle are ubiquitous Gram-negative bacteria that cause a range of disease in humans. The nomenclature of *Salmonella* is quite confusing, as there are several subspecies and serotypes (also referred to as serovars). Of the 2500 known serotypes, only about 100 are known to cause human disease and are in the genus *Salmonella*, species *enterica*, subspecies *enterica* (or subspecies I). The other species of *Salmonella* and other subspecies within *S. enterica* rarely infect humans. Included in the *S. enterica* subspecies *enterica* are the serotypes Typhi and Paratyphi, which cause typhoid and paratyphoid fever, respectively. The remaining serotypes are referred to as nontyphoidal and primarily cause intestinal infections. The most common serotypes in the United States are Enteritidis, Typhimurium, Newport, and Javiana, but other serotypes may present in travelers returning from other countries. It is important to note that nontyphoidal *Salmonella* strains can cause extraintestinal infections (e.g., bacteremia, meningitis, and osteomyelitis), particularly in children under 2 years of age, the elderly, and immunocompromised individuals including patients with sickle cell disease.

Salmonella infections are usually acquired through ingestion of contaminated food or water. Eating raw, unpeeled fruits and vegetables, undercooked meat, and unpasteurized dairy products is particularly risky. Infections can also be transmitted by direct contact with infected animals or their excreta. Oysters and shellfish grown in contaminated waters can transmit *Salmonella*. Meals can be contaminated during preparation through unsanitary kitchen practices or infected food handlers. Infections are dependent on the size of the inoculum, the vehicle of transmission, and the susceptibility of the host. An oral inoculum of 10^6 bacteria or greater, ingestion of contaminated foods high in protein and fat, and impaired host status tend to promote infection.

The clinical presentation of *Salmonella* enterocolitis can range from a mild to severe diarrheal illness with cramps, nausea, vomiting, and fever. The incubation period is usually

1-3 days. The acute illness usually lasts for 1-2 weeks, although *Salmonella* are shed in the feces for 4-6 weeks in untreated persons and for up to several months in patients treated with antibiotics. *Salmonella* enterocolitis can result in a bacteremia that can lead to focal infections outside the gastrointestinal tract. Sepsis and even death can occur in patients with pre-existing comorbidities.

Stool specimens from patients suffering from *Salmonella* enterocolitis are usually positive for fecal leukocytes. Clinical microbiology laboratories can isolate *Salmonella* spp. from stool cultures and occasionally blood. If a biopsy or sterile fluid specimen (e.g., synovial or spinal fluid) is sent to the microbiology laboratory with concern for *Salmonella* infection, it is important to alert the laboratory to ensure appropriate cultures are set up. Once an isolate is identified as *Salmonella* by biochemical or molecular analysis, confirmation is performed using *Salmonella*-specific antisera directed at the O antigen. Full serotyping is beyond the scope of most clinical laboratories but can be performed by public health laboratories as part of epidemiologic surveillance.

In general, antibiotic treatment is not indicated for patients with uncomplicated *Salmonella* enterocolitis but may be considered for those who are severely ill or at risk for severe infections, such as neonates, the elderly, and immunocompromised individuals. However, antibiotic treatment may prolong infectious carriage of *Salmonella*. Treatment should be guided by antibiotic sensitivity testing, since drug resistance to multiple antibiotics is prevalent among some *Salmonella* strains.

SHIGELLA

Shigellosis is an acute gastrointestinal infection caused by one of four *Shigella* subgroups that have been historically treated as species: *S. dysenteriae* (subgroup A), *S. flexneri* (subgroup B), *S. boydii* (subgroup C), and *S. sonnei* (subgroup D). This subgroup classification is based on the type of O antigen present on the bacteria. Shigellosis in the United States and other developed countries is primarily caused by *S. sonnei* (subgroup D). In the developing world, the majority of endemic *Shigella* dysentery is due to *S. flexneri* (subgroup B); this is the species most commonly identified in travelers with shigellosis. *S. dysenteriae* serotype 1 is associated with severe disease and high rates of mortality.

Shigella is a strictly human pathogen, can remain viable on inanimate objects for weeks, and can cause infections with an inoculum of just 200 bacteria. Shigellosis outbreaks have been associated with conditions favoring human fecal-oral transmission, such as poor sanitation, inadequate water supplies, and crowded living conditions. Shigellosis is commonly seen among travelers returning from developing areas of the world and among people living or working in refugee camps and institutional settings, such as daycare centers, prisons, and facilities for the developmentally disabled.

Clinical illness ranges from severe inflammatory diarrhea with systemic toxicity, commonly called bacterial dysentery, to mild, nonspecific diarrhea. The incubation period of shigellosis is usually 24-48 hours, with a presentation characterized by fever, abdominal cramps, and watery diarrhea. The illness may progress to a more serious condition with the passage of blood, mucus, and pus, accompanied by left lower quadrant pain and tenesmus. *Shigella* invade the mucosa and submucosa of the colon during this stage of the disease. The duration of symptoms is usually 1 week or less, although *Shigella* may persist in the stools for 1-3 months after cessation of clinical symptoms.

The presence of fecal leukocytes supports the diagnosis of an inflammatory infection but is not specific for shigellosis. Isolation of *Shigella* spp. can be achieved through routine stool culture media. Specific antisera to the O antigen are available for subgroup classifications, and isolates may be sent to public health laboratories for further serotyping analysis. Most laboratories employ a nonculture assay for the detection of Shiga toxin 1 (Stx1) and 2 (Stx2) to enhance the detection of STEC. The toxin produced by *S. dysenteriae* serotype 1 is highly homologous to Stx1 and would be detected by these assays as well.

Treatment of shigellosis is warranted, as it shortens disease and terminates bacterial carriage, and should be based on antimicrobial susceptibilities. Rates of antibiotic resistance are

higher in isolates from developing countries. This is particularly true for *S. dysenteriae* 1 isolates acquired during international travel to parts of Africa and Asia where most strains are multidrug resistant. Post-infection complications are rare but can occur in patients with shigellosis. The most notorious complication is hemolytic-uremic syndrome (HUS). While this is associated with Shiga toxin exposure, it is more commonly due to STEC infection rather than shigellosis. Reactive arthritis is another complication that may follow *Shigella* infection, particularly with *S. flexneri*, and does not appear to be associated with antibiotic treatment.

Campylobacter

Campylobacter jejuni is a short, comma-shaped, Gram-negative rod that is a common cause of diarrhea worldwide and may exceed *Salmonella* and *Shigella* as an etiologic agent of acute infectious diarrhea. Of the 22 species within the genus *Campylobacter*, the two responsible for the majority of gastrointestinal disease are *C. jejuni* and *Campylobacter coli*, with *C. jejuni* responsible for about 85% of infections. *Campylobacter fetus* subsp. *fetus* infection is much less common but may cause serious systemic infections in human hosts with impaired immunity. Extraintestinal *Campylobacter* infections can occur but are primarily reported in neonates, the elderly, and immune-compromised individuals.

Campylobacter species are found worldwide and are primarily zoonotic, inhabiting the intestinal tracts of livestock and domesticated pets but rarely causing disease in these animals. The organisms are shed in animal feces, and humans can become infected by close contact with animals. The consumption of poorly cooked meat or dairy products contaminated with animal feces is the most common source of campylobacteriosis.

After an incubation period of 2-4 days, patients develop cramping abdominal pain, fever, and watery diarrhea typically lasting less than 1 week, although recurrent attacks in untreated individuals have been noted. In some cases, colitis symptoms predominate with bloody stools and lower abdominal pain that can mimic appendicitis. Rarely, colonic ulcerations and systemic infections may occur. *C. fetus* subsp. *fetus* is more frequently associated with systemic infections, with diarrhea occurring in only one-third of patients. Infants and immunocompromised patients appear to be more susceptible to the complications of fulminant sepsis, endocarditis, and meningitis caused by this organism.

The diagnosis of campylobacteriosis is made by isolation of the organism from stool, blood, or other tissues. Isolation of *Campylobacter* from clinical specimens requires special growth conditions that are created using unique, commercially available media and incubation under microaerophilic conditions at 42° C. These growth conditions assist in selecting for the thermo-tolerant *C. jejuni* and *C. coli* from other stool flora. However, such conditions may be not optimal for the recovery of other *Campylobacter* species, such as *C. fetus*. Standard stool culture procedures incorporate the appropriate growth conditions for *C. jejuni* and *C. coli*. However, routine work-up for blood, urine, and other specimens do not. Therefore, if the clinician suspects extraintestinal manifestations of *Campylobacter*, the laboratory must be notified so that appropriate culturing conditions are included.

Severe or prolonged gastrointestinal *Campylobacter* infection should be treated with antibiotics, especially in pregnant women or immunosuppressed patients. Fluoroquinolone-resistant *Campylobacter* has become widespread in many parts of the world, particularly in Asia. It is important to be aware of the autoimmune complications such as Guillain-Barré syndrome (GBS) that may occur following *Campylobacter* infection. GBS is an acute, paralytic disease of the peripheral nervous system that is seen in approximately 0.1% of *Campylobacter* infections. GBS has been associated primarily with *C. jejuni*, which expresses lipooligosaccharides that mimic human gangliosides, resulting in autoantibodies that react with epitopes in the peripheral nerves. The onset of GBS usually occurs within 2-21 days of the diarrheal illness. Reactive arthritis affects 2-4% of patients post-infection and is characterized by joint pain and swelling, commonly in the knees, that last for several weeks to a year. Symptoms typically begin days to weeks following intestinal illness; in about 5% of cases the arthritis can be chronic or relapsing.

VIBRIO

The vibrios are Gram-negative bacilli that are widely distributed in marine and estuarine environments, with *Vibrio cholerae* also found in bodies of fresh water. Of the many *Vibrio* species, gastrointestinal infections primarily occur with *V. cholerae* and *Vibrio parahaemolyticus*. While *V. cholerae* is the only species that causes endemic, epidemic, and pandemic cholera, only a small subset of *V. cholerae* strains carry the requisite genes to cause cholera. Serotyping studies have identified more than 200 different O groups within the species *V. cholerae*, with almost all cholera-causing strains belonging to O group 1 or 139. The majority of non-O1/non-O139 *V. cholera* strains are nonpathogenic, or cause mild illness, although some of these strains have been implicated in outbreaks of cholera-like illness. *Vibrio parahaemolyticus* infections are most commonly transmitted by contaminated food. The pathogenicity of *V. parahaemolyticus* has been correlated with production of one of two thermostable direct hemolysins, Vp-TDH and Vp-TRH. Less commonly, gastroenteritis can also be caused by *Vibrio mimicus*, *Vibrio fluvialis*, *Vibrio vulnificus*, and the related organism, *Grimontia hollisae*.

Cholera is primarily seen in severely resource-limited settings where there is inadequate access to potable water. Tourists from developed countries rarely acquire cholera because they are not typically exposed to these environments and have access to potable water. In contrast, *V. parahaemolyticus* is a common cause of acute gastroenteritis in areas where raw, undercooked, or improperly stored and handled seafood is consumed. *V. parahaemolyticus* outbreaks have been reported in Japan and Asia and on cruise ships. Sushi, pre-cooked shellfish, and raw oysters have been implicated in outbreak reports. A relatively large inoculum (10^{10} organisms or more) is necessary to establish an infection in normal human hosts because *Vibrio* is exquisitely sensitive to gastric acid. In patients with decreased gastric acidity, the infectious inoculum is lower ($\leq 10^6$ organisms). *V. vulnificus* gastroenteritis risks include the ingestion of shellfish, especially raw oysters. In addition to vomiting and diarrhea, *V. vulnificus* can become an invasive pathogen, leading to septicemia and intractable septic shock. Pre-existing hepatic disease was present in over 75% of *V. vulnificus* septicemic patients in one report. Therefore, patients with hepatic disease should avoid eating raw oysters.

The incubation period for *Vibrio*-associated diseases ranges from less than 1 day to several days. Patients with cholera present with severe, voluminous watery ("rice-water") diarrhea, abdominal cramps, nausea, and vomiting. *V. parahaemolyticus* usually causes a self-limited, 72-hour, cholera-like gastroenteritis but some strains can penetrate the lamina propria, resulting in dysentery that resembles shigellosis.

A definitive diagnosis of *Vibrio* infection is based on isolation of the organism from clinical samples, which also allows the determination of antibiotic susceptibility. The isolation of *Vibrio* from stool is accomplished using selective salt-containing media that is *not* part of the standard stool culture protocol in most microbiology laboratories. Therefore, if *Vibrio* infection is in the differential, this needs to be communicated to the laboratory. Once cultured, *Vibrio* species can be determined by biochemical tests; serogroup and serotyping can be performed with specific antibodies by public health laboratories. There is a rapid diagnostic test for *V. cholerae* that has been used as part of cholera outbreak investigations (Crystal VC™ Span Diagnostics, Surat, India).

The cornerstone of clinical management for severe gastroenteritis due to *Vibrio* infection is the aggressive replacement of fluid and electrolytes lost in the diarrheal stools. Antibiotic treatment may shorten the duration of *Vibrio* infections by eradicating organisms in the stool, although symptoms may persist because of toxins already bound to the mucosal surface.

OTHER NOTABLE BACTERIAL PATHOGENS

Yersinia enterocolitica is a zoonotic enteric pathogen that is increasingly recognized as a cause of human bacterial enteritis. This pathogen appears to be ubiquitous in nature, with acquisition by ingestion of contaminated food or water. These organisms are able to multiply at room temperature and survive at low temperatures (4° C) for many months under a variety of environmental conditions. Infection requires ingestion of a relatively large inoculum (10^9

bacteria) followed by invasion and involvement of the mesenteric lymph nodes. Patients can present with cramping abdominal pain, fever, and diarrhea, which may last from 1 to 3 weeks. Occasionally, pain localized in the right lower quadrant caused by ileitis and mesenteric lymphadenitis may be severe enough to mimic appendicitis. The laboratory should be notified of a possible *Yersinia* infection to ensure the use of selective media that may not be included in routine stool cultures. *Yersinia* may be present in the stool for weeks after symptoms resolve. Usefulness of antimicrobial therapy in enterocolitis and lymphadenitis caused by *Y. enterocolitica* is uncertain but may be warranted in severe cases and guided by antibiotic susceptibility testing. Post-infectious complications consist of autoimmune disorders, including arthritis, erythema nodosum, Reiter syndrome, and ankylosing spondylitis, which may be more likely to develop in patients with the HLA-B27 histocompatibility tissue type.

Plesiomonas shigelloides is a Gram-negative bacillus found in aquatic environments that has been primarily associated with sporadic cases and outbreaks of diarrheal disease. Risk factors for *P. shigelloides* gastroenteritis include travel to tropical regions, consumption of raw or undercooked shellfish or contaminated water, and exposure to reptiles and tropical fish. *Plesiomonas* enteritis typically presents as a watery diarrhea but may manifest as a dysenteric syndrome. The organism readily grows in the laboratory on standard media, and its presence is usually screened for as part of standard stool culturing. Most infections are characterized by self-limiting diarrhea with blood or mucus, abdominal cramps, vomiting, and fever and do not require antibiotic therapy. Antimicrobial therapy may be warranted with prolonged infections and infections associated with severe illness.

Aeromonas spp. are ubiquitous in freshwater aquatic habitats, and concentrations peak when water temperatures rise during the summer months. While *Aeromonas* spp. are not considered part of the normal gastrointestinal flora, their role as enteropathogens is somewhat contested. Consumption of contaminated meats or water is the most common source of infection. *Aeromonas*-associated diarrhea most commonly presents as an acute watery diarrhea but may be associated with a more invasive disease resembling dysentery or enterocolitis. *Aeromonas* spp. will grow in media used in routine stool culture, but its presence may be overlooked because its growth in culture is difficult to distinguish from endogenous flora. Therefore, it is important to alert the laboratory to screen for this agent in patients suffering from TD. Most infections with *Aeromonas* spp. are self-limiting and require only supportive care. Cases of HUS have been associated with *Aeromonas* diarrheal illness thought to be secondary to the organism expressing Shiga toxin.

Colitis caused by *C. difficile* is not expected to be a primary pathogenic process in travel-acquired infectious diarrhea. However, *C. difficile* diarrheal illness should be considered in cases of diarrhea persisting after travel in patients who recently received of antimicrobials. Clinical features of *C. difficile* colitis include persistent and profuse watery diarrhea, sometimes containing blood and mucus. Systemic toxicity with fever and malaise may be present in severe cases. Diagnosis is made by detecting the presence of *C. difficile* toxins A or B in stools by immunoassay or NAT. Isolation of the organism by culture is rarely performed. Treatment consists of discontinuing the causative antimicrobial and starting either oral vancomycin or metronidazole.

EMERGING BACTERIAL PATHOGENS

Enteroaggregative *E. coli* (EAEC) is another pathotype of *E. coli* that appears to be playing an emerging role in TD. It possesses the ability to aggressively attach to intestinal mucosal epithelial cells and mediate inflammation through a variety of adhesins and toxins. As EAEC is similar to ETEC, accurate identification of EAEC in stool cultures is challenging. The introduction of NAT methods has the potential to improve the diagnosis of these pathogens.

Arcobacter spp. are *Campylobacter*-like organisms that have been isolated from the feces of animals with enteritis. *Arcobacter butzleri* has been reported as a cause of TD in travelers

to Mexico, Guatemala, and India and is likely transmitted through the ingestion of contaminated food or water. *Arcobacter* may be an underestimated etiologic agent of TD since its presence in stool cultures is rarely sought, but it can be recovered using the same media to isolate *Campylobacter* incubated under microaerophilic conditions at 37° C rather than 42° C.

Bacteroides spp. are a major component of the normal human fecal flora. Enterotoxigenic *Bacteroides fragilis* (ETBF) have been identified as a cause of acute watery diarrhea and colonic inflammation. In a recent study, ETBF was identified via NAT in the stool of 13% of patients with TD returning from India. The ability to discriminate ETBF from normal anaerobic intestinal flora requires NAT. More study is required to better understand the role of ETBF as a cause of diarrheal illness in those who travel abroad.

VIRAL AGENTS

Norovirus is the leading cause of food-borne infection and the cause of half of all gastroenteritis outbreaks worldwide. This virus may persist in the environment for prolonged periods and is highly contagious, with an infectious dose of only 20 viral particles. Its ability to cause explosive outbreaks in closed communities has been reported in all-inclusive resorts and on cruise ships. A single genotype of norovirus (genogroup II, genotype 4) has been the predominant norovirus strain that has been associated with gastroenteritis outbreaks in many countries. Norovirus may be transmitted by ingestion of contaminated water and foods such as salads, clams, and oysters and from contact with contaminated fomites. Gastroenteritis caused by norovirus is characterized by watery diarrhea, abdominal cramps, and vomiting; fever is rarely present. The disease is self-limited, and the gastrointestinal symptoms last from 1 to 4 days. Antigen detection immunoassays and nucleic acid tests can be used to confirm the clinical diagnosis. Treatment consists of replenishing fluids and electrolytes.

Rotavirus has been implicated as an important cause of TD among adults and children, especially among those visiting Central America and the Caribbean. Transmission is fecal-oral and is typically through direct contact with infected individuals such as caregivers caring for ill children. Symptoms begin with fever and vomiting for 2-3 days followed by profuse nonbloody, non-inflammatory diarrhea. The incidence of rotavirus has decreased secondary to the advent of effective vaccines combined with massive vaccination campaigns throughout the developing world, particularly in Latin America. Nevertheless, unvaccinated travelers to underdeveloped regions remain at risk. Rotaviral antigen can be detected in stool using commercially available immunoassays or by NAT; treatment is achieved through supportive care.

FURTHER READING

Anderson, E.J., Weber, S.G., 2004. Rotavirus infection in adults. Lancet Infect. Dis. 4 (2), 91–99.

Binnicker, M.J., 2015. Multiplex molecular panels for the diagnosis of gastrointestinal infection: performance, result interpretation and cost-effectiveness. J. Clin. Microbiol. pii: JCM.02103-15. [Epub ahead of print]; PMID: 26311866.

de la Cabada Bauche, J., Dupont, H.L., 2011. New developments in traveler's diarrhea. Gastroenterol. Hepatol. (NY) 7 (2), 88–95.

Estrada-Garcia, T., Navarro-Garcia, F., 2012. Enteroaggregative *Escherichia coli* pathotype: a genetically heterogeneous emerging foodborne enteropathogen. FEMS Immunol. Med. Microbiol. 66 (3), 281–298.

Gomi, H., Jiang, Z.D., Adachi, J.A., et al., 2001. In vitro antimicrobial susceptibility testing of bacterial enteropathogens causing traveler's diarrhea in four geographic regions. Antimicrob. Agents Chemother. 45 (1), 212–216.

Hill, D.R., Beeching, N.J., 2010. Travelers' diarrhea. Curr. Opin. Infect. Dis. 23 (5), 481–487.

Humphries, R.M., Linscott, A.J., 2015. Laboratory diagnosis of bacterial gastroenteritis. Clin. Microbiol. Rev. 28 (1), 3–31.

Jiang, Z.D., Dupont, H.L., Brown, E.L., et al., 2010. Microbial etiology of travelers' diarrhea in Mexico, Guatemala, and India: importance of enterotoxigenic *Bacteroides fragilis* and *Arcobacter* species. J. Clin. Microbiol. 48 (4), 1417–1419.

Patel, R., 2015. MALDI-TOF MS for the diagnosis of infectious diseases. Clin. Chem. 61 (1), 100–111.

Ross, A.G., Olds, G.R., Cripps, A.W., et al., 2013. Enteropathogens and chronic illness in returning travelers. N. Engl. J. Med. 368 (19), 1817–1825.

Steffen, R., Hill, D.R., DuPont, H.L., 2015. Traveler's diarrhea: a clinical review. JAMA 313 (1), 71–80.