GI Pathogen Testing, New Guidelines

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Director, Microbiology & Molecular
Sutter Health Shared Laboratory
Microbe 2018
Objectives

• IDSA guidelines for diarrheal illnesses
  – Who should be tested
  – What pathogens should be targeted by the tests
  – When we should we test for parasites
  – Treatment

• IDSA guidelines – testing for *C. difficile*

• CIDT’s for stool pathogens
Sutter Health System (SHS)

2nd largest Northern-California health system
- Not-for-profit
- Serving >100 communities
- 24 acute care hospital systems
- 26 clinics, medical foundations
- 40,000 physicians
- 50,000 employees
Sutter Health Shared Laboratory (SHSL)

• Limited Chemistry, Hematology, Blood Bank, and Serology  
  – 3.9 million tests/year
• Major provider of Microbiology tests for the system  
  – 1 million tests/year
• Major provider of Molecular Dx (ID, Genetic)  
  – 367,000 tests/year
Gastroenteritis & Diarrheal Illnesses

- 2nd leading cause of infectious diseases morbidity
- 3rd leading cause of mortality
  - 1.4 million deaths in 2010
- 1.7 billion cases/year of childhood diarrhea worldwide
- 179 million cases/year in the US

1. Mandell's Principals of Infectious Diseases, 2015
2. WHO
3. DuPont, in NEJM 2014
Infectious Diarrhea

• **Definition:**
  - ≥3 unformed stools within 24 hours AND
  - Enteric symptoms (nausea, vomiting, pain, cramps,...)

• **Severity:**
  - Mild – active
  - Moderate – limited activity
  - Severe – serious disability

• **Duration:**
  - Acute <14 days
  - Persistent 14-30 days
  - Chronic >30 days – parasitic?
### Top 5 Most Frequent Pathogens

Numbers of acute gastroenteritis outbreaks and outbreak-associated outcomes caused by various etiologic agents reported in the National Outbreak Reporting System, United States, 2009–2010*

<table>
<thead>
<tr>
<th>Outbreak etiology</th>
<th>Confirmed</th>
<th>Suspected</th>
<th>Total</th>
<th>No. (%) outbreaks</th>
<th>No. (%) outbreak-associated outcomes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Single agent†</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Norovirus‡</td>
<td>1,355 (64.2)</td>
<td>553 (78.1)</td>
<td>1,908 (67.7)</td>
<td>69,145 (77.7)</td>
<td>1,093 (45.9)</td>
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<tr>
<td><em>Salmonella</em> spp.</td>
<td>344 (16.3)</td>
<td>11 (1.6)</td>
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<tr>
<td><em>Shigella</em> spp.§</td>
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<td>STEC</td>
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<td>52 (2.2)</td>
</tr>
</tbody>
</table>

Most Frequent Pathogens - caused **92.7%** of the illnesses

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<thead>
<tr>
<th>Outbreak etiology</th>
<th>Confirmed</th>
<th>Suspected</th>
<th>Total</th>
<th>Ilnesses</th>
<th>Hospitalizations</th>
<th>Deaths</th>
</tr>
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*Numbers of acute gastroenteritis outbreaks and outbreak-associated outcomes caused by various etiologic agents reported in the National Outbreak Reporting System, United States, 2009–2010*

Most Frequent Pathogens
- caused 96.6% of deaths

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Most Frequent Pathogens

Percentage of outbreaks of acute gastroenteritis transmitted by person-to-person contact, environmental contamination, and unknown mode of transmission by confirmed etiology — National Outbreak Reporting System, United States, 2009–2013

[MMWR, Dec. 11 2015 / 64]

https://www.cdc.gov/mmwr/preview/mmwrhtml/ss6412a1.htm?s_cid=ss6412a1_w#Tab1
2017 Infectious Diseases Society of America Clinical Practice Guidelines for the Diagnosis and Management of Infectious Diarrhea

Andi L. Shane, MD¹ Rajal K. Mody, MD² John A. Crump, MD³ Phillip I. Tarr,⁴ Theodore S. Steiner, MD⁵ Karen Kotloff, MD⁶ Joanne M. Langley, MD⁷ Christine Wanke, MD⁸ Cirle Alcantara Warren, MD⁹ Allen C. Cheng, PhD¹⁰ Joseph Cantey, MD¹¹ and Larry K. Pickering, MD¹²

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Clin Infect Dis. 2017 Nov 29;65(12):e45-e80
Who Should be Tested?

• Test based on:
  – History of illness
    • Travel
    • Long-term care
    • Childcare
    • Healthcare Associated (HCA)
  – Immunocompromised
  – Zoonoses, Outbreak-associated, or Public Health risk
  – Severe Diarrhea
    • Fever, dehydration, dysentery
    • > 7 Days$^{1,2}$ or persistent

1. Riddle MS, in AJG April 2016
• “The optimal specimen for laboratory diagnosis of infectious diarrhea is a diarrheal stool sample (i.e., a sample that takes the shape of the container). For detection of bacterial infections, if a timely diarrheal stool sample cannot be collected, a rectal swab may be used (weak, low).”
“The optimal specimen for laboratory diagnosis of infectious diarrhea is a diarrheal stool sample (i.e., a sample that takes the shape of the container). For detection of bacterial infections, if a timely diarrheal stool sample cannot be collected, a rectal swab may be used (weak, low).”

- Diarrheal Stool

- Rectal Swab
IDSA – Target Oriented Testing

- Organism detection depends on what pathogens are tested and which tests are used
- Factors that determine tests and targets are:
  - History (exposure)
  - Clinical presentation (signs and symptoms)
  - Immune status

Clin Infect Dis. 2017 Nov 29;65(12):e45-e80
## IDSA – Test For Which Pathogens?

<table>
<thead>
<tr>
<th>Condition</th>
<th>Target</th>
</tr>
</thead>
<tbody>
<tr>
<td>Severe Cramping/Tenderness, Fever, Bloody or Mucoid stool, Sepsis?</td>
<td><em>Salmonella, Shigella, Campylobacter, Yersinia, STEC, C. difficile</em></td>
</tr>
<tr>
<td>• Children w/ persistent abdominal pain, exposure to pork products</td>
<td>• <em>Yersinia enterocolitica</em></td>
</tr>
<tr>
<td>• Travel to endemic region or Shellfish consumption</td>
<td>• <em>Vibrio</em></td>
</tr>
<tr>
<td>Possibility of Outbreaks</td>
<td>Broad Panel (bacterial, viral, parasitic)</td>
</tr>
<tr>
<td>Immunocompromised</td>
<td>Broad Panel (bacterial, viral, parasitic)</td>
</tr>
<tr>
<td>Uncomplicated Traveler’s Diarrhea, which treatment is not indicated</td>
<td>Testing not recommended</td>
</tr>
<tr>
<td>Traveler’s Diarrhea &gt;14 days</td>
<td>Parasites</td>
</tr>
</tbody>
</table>

* usually not bloody.

Clin Infect Dis. 2017 Nov 29;65(12):e45-e80
• “In immunocompetent children and adults, empiric antimicrobial therapy for bloody diarrhea while waiting for results of investigations is **not** recommended (strong, low).”

IDSA – Who Should be Empirically Treated?

• “In immunocompetent children and adults, empiric antimicrobial therapy for bloody diarrhea while waiting for results of investigations is not recommended (strong, low).”

• Exceptions:
  o <3 months old
  o High severity
    ▪ Fever
    ▪ Abdominal pain
    ▪ Dysentery (bloody stool, cramps, tenesmus, fever ...)
  o Recent travel history
    ▪ Fever
    ▪ Signs of sepsis
  o Immunocompromised

Clin Infect Dis. 2017 Nov 29;65(12):e45-e80
IDSA – Who Should be Treated?

- Usually has fever and bloody diarrhea
- Any signs or symptoms of sepsis
- Epidemiological or travel link
- Immunocompromised with severe illness
- *Shigella*
- Some *Campylobacter* and *Salmonella*
# IDSA – Testing for Parasites

[Exposure or Condition Associated with Pathogens]

<table>
<thead>
<tr>
<th>Exposure or Condition</th>
<th>Pathogens</th>
</tr>
</thead>
<tbody>
<tr>
<td>Swimming in recreational water facility with treated water</td>
<td><em>Cryptosporidium</em> and other potentially waterborne pathogens when disinfectant concentrations are inadequately maintained</td>
</tr>
<tr>
<td>Healthcare, long-term care, prison exposure, or employment</td>
<td><em>Norovirus</em>, <em>Clostridium difficile</em>, <em>Shigella</em>, <em>Cryptosporidium</em>, <em>Giardia</em>, <em>STEC</em>, rotavirus</td>
</tr>
<tr>
<td>Child care center attendance or employment</td>
<td>Rotavirus, <em>Cryptosporidium</em>, <em>Giardia</em>, <em>Shigella</em>, <em>STEC</em></td>
</tr>
<tr>
<td>Recent antimicrobial therapy</td>
<td><em>C. difficile</em>, multidrug-resistant <em>Salmonella</em></td>
</tr>
<tr>
<td><strong>Travel to resource-challenged countries</strong></td>
<td><em>Escherichia coli</em> (enteroaggregative, enterotoxigenic, enteroinvasive), <em>Shigella</em>, <em>Typhi</em> and nontyphoidal <em>Salmonella</em>, <em>Campylobacter</em>, <em>Vibrio cholerae</em>, <em>Entamoeba histolytica</em>, <em>Giardia</em>, <em>Blastocystis</em>, <em>Cyclospora</em>, <em>Cystoisospora</em>, <em>Cryptosporidium</em></td>
</tr>
<tr>
<td>Exposure to house pets with diarrhea</td>
<td><em>Campylobacter</em>, <em>Yersinia</em></td>
</tr>
<tr>
<td>Exposure to pig feces in certain parts of the world</td>
<td><em>Balantidium coli</em></td>
</tr>
<tr>
<td>Contact with young poultry or reptiles</td>
<td>Nontyphoidal <em>Salmonella</em></td>
</tr>
<tr>
<td>Visiting a farm or petting zoo</td>
<td><em>STEC</em>, <em>Cryptosporidium</em>, <em>Campylobacter</em></td>
</tr>
<tr>
<td><strong>Exposure or condition</strong></td>
<td><strong>Pathogens</strong></td>
</tr>
<tr>
<td>Age group</td>
<td>Rotavirus (6–18 months of age), nontyphoidal <em>Salmonella</em> (infants from birth to 3 months of age and adults &gt;50 years with a history of atherosclerosis), <em>Shigella</em> (1–7 years of age), <em>Campylobacter</em> (young adults)</td>
</tr>
<tr>
<td>Underlying immunocompromising condition</td>
<td>Nontyphoidal <em>Salmonella</em>, <em>Cryptosporidium</em>, <em>Campylobacter</em>, <em>Shigella</em>, <em>Yersinia</em></td>
</tr>
<tr>
<td>Hemochromatosis or hemoglobinopathy</td>
<td><em>Y. enterocolitica</em>, <em>Salmonella</em></td>
</tr>
</tbody>
</table>
| **AIDS, immunosuppressive therapies** | *Cryptosporidium*, *Cyclospora*, *Cystoisospora*, microsporidia, *Mycobacterium avium–interven-

IDSA – Testing for Parasites

- Travel to resource-challenged area
- AIDS and immunosuppressive therapy
  - Swimming in treated water
    - Cryptosporidium
  - Childcare related
    - Cryptosporidium, Giardia
  - Underlying immunocompromised condition
    - Cryptosporidium
  - Anal-genital, oral-anal, digital-anal contact
    - E. histolytica, Giardia, Cryptosporidium

• **Travel to resource challenged area**
  – “... *Entamoeba histolytica*, *Giardia*, *Blastocystis*, *Cyclospora*, *Cystoisospora*, *Cryptosporidium* ...”

“Travelers with diarrhea lasting 14 days or longer should be evaluated for intestinal parasitic infections (strong, moderate).”

IDSA – Testing for Parasites

- Travel to resource challenged area

- AIDS and Immunosuppressive Therapy
  - “Cryptosporidium, Cyclospora, Cystoisospora, microsporidia...”
Clinical Practice Guidelines for *Clostridium difficile* Infection in Adults and Children: 2017 Update by the Infectious Diseases Society of America (IDSA) and Society for Healthcare Epidemiology of America (SHEA)

L. Clifford McDonald,1 Dale N. Gerdin,2 Stuart Johnson,23 Johan S. Bakken,4 Karen C. Carroll,5 Susan E. Coffin,6 Erik R. Dubberke,7 Kevin W. Garey,8 Carolyn V. Gould,1 Ciaran Kelly,9 Vivian Loo,10 Julia Shaklee Sammons,6 Thomas J. Sandora,11 and Mark H. Wilcox12

1Centers for Disease Control and Prevention, Atlanta, Georgia; 2Edward Hines Jr Veterans Administration Hospital, Hines, and 3Loyola University Medical Center, Maywood, Illinois; 4St Luke's Hospital, Duluth, Minnesota; 5Johns Hopkins University School of Medicine, Baltimore, Maryland; 6Children's Hospital of Philadelphia, Pennsylvania; 7Washington University School of Medicine, St Louis, Missouri; 8University of Houston College of Pharmacy, Texas; 9Beth Israel Deaconess Medical Center, Harvard Medical School, Boston, Massachusetts; 10McGill University Health Centre, McGill University, Montréal, Québec, Canada; 11Boston Children's Hospital, Massachusetts; and 12Leeds Teaching Hospitals NHS Trust, United Kingdom
IDSA - *C. difficile* Infection (CDI) Definition

**Symptoms:**
- Diarrhea (usually)
  - Unexplained (e.g., exclude laxative)
  - New-onset
  - ≥3 within 24 hours
  - Unformed stool

**Tests:**
- Detection of *C. difficile* toxins
- Detection of Toxigenic *C. difficile*
- Colonoscopy
- Histopathology

1. “VI. When should testing be performed for Clostridium difficile? Recommendation.
   18. Testing may be considered for *C. difficile* in people >2 years of age who have a history of diarrhea following antimicrobial use and in people with healthcare-associated diarrhea.” \(^1\)

2. “Patients with unexplained and new-onset ≥3 unformed stools in 24 hours are the preferred target population for testing for CDI.” \(^2\)

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IDSA – When Should We Test for *C. difficile*?

- >2 years old
- History of antimicrobial use
- Healthcare-associated diarrhea
- Persistent with no apparent etiology or risk factor
- Test only once
- Testing either toxin (EIA?) or toxigenic strain (NAAT) are acceptable

# SHSL Gastroenteritis Testing

<table>
<thead>
<tr>
<th>Bacterial cult. (19K 4.4% POS)</th>
<th>Viral (2,500)</th>
<th>Parasites (EIA 5,300)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Campylobacter</em> (407)</td>
<td>Rotavirus-EIA (6.5%)</td>
<td><em>Cryptosporidium</em> (1.2%)</td>
</tr>
<tr>
<td><em>Salmonella</em> (259)</td>
<td>Norovirus-PCR (10.7%)</td>
<td><em>Giardia</em> (0.9%)</td>
</tr>
<tr>
<td><em>Shigella</em> (30)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Aeromonas</em> (78)</td>
<td></td>
<td>Other parasites (50K)</td>
</tr>
<tr>
<td><em>Plesiomonas</em> (13)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>E. coli</em> 0157, STEC (36)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Vibrio</em> (5)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Yersinia</em> (4)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>C. difficile</em> (12,000)</td>
<td></td>
<td></td>
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<tr>
<td>EIA (5.9%) – PCR (25.2%)</td>
<td></td>
<td></td>
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</tbody>
</table>

* Needs special request & media
Stool Bacterial Culture

- GN-Broth (only if Shiga-toxin ordered)
- BAP
- MAC
- MAC/Sorb
- *Salmonella, Shigella (SS)*
- Hektoen Enteric (HE)
- Campy-CVA agar

https://www.tuyenlab.net/2018/02/microbiology-hektoen-enteric-agar.html
Stool Bacterial Culture (Confirmation)

- Shiga-toxin – EIA
- *E. coli* O157, H7 – EIA
- Latex agglutination: Wellcolex® Color
  - *Shigella*
  - *Salmonella*

Links:
- [https://www.google.com/search?q=wellcolex+shigella&safe=active&rlz=1C1GGRV](https://www.google.com/search?q=wellcolex+shigella&safe=active&rlz=1C1GGRV)
- [https://www.thermofisher.com/order/catalog/product/R24250](https://www.thermofisher.com/order/catalog/product/R24250)
Culture Independent Diagnostic Tests (CIDT)
[Marder EP, et al. MMWR 2017]

https://www.cdc.gov/mmwr/volumes/66/wr/mm6615a1.htm
“V. Which diagnostic tests should be performed when enteric fever or bacteremia is suspected? Recommendation.

17. Culture-independent, including panel-based multiplex molecular diagnostics from stool and blood specimens, and, when indicated, culture-dependent diagnostic testing should be performed when there is a clinical suspicion of enteric fever (diarrhea uncommon) or diarrhea with bacteremia (strong, moderate).”
IDSA – Culture-Independent Diagnostic Tests

• Now CIDT’s are recommended when:
  – Suspected sepsis
  – Immunocompromised patients

• Results should be interpreted with cautions and/or be confirmed by culture if:
  – non-viable organisms are suspected
  – susceptibility testing is indicated
  – outbreak investigation at Public Health level required

• CIDT’s should not be used for patient management or test-of-cure

Clin Infect Dis. 2017 Nov 29;65(12):e45-e80
CIDT (Continued)

- Advantages:
  - Turnaround time (Days vs. Hours)
  - Easy to perform (Moderate complexity)
  - Reliability (higher sensitivity) \(^1,^3\)
  - Reduces ER admission\(^2\)
  - Reduces length of stay\(^2\)
  - Less dependent on the specimen quality, viability\(^3\)
  - Recommended by IDSA latest guidelines\(^4\)

2. McNabb, K. 2017
3. Alexander J, 2018
CIDT (Continued)

• Disadvantages:
  – $ Cost ??
  – May detect nonviable organisms; not the best fit for follow-up or treatment monitoring¹
    • Public Health studies
    • Susceptibility
      – Rarely indicated
      – Ciprofloxacin resistance in 2015²
        » *Salmonella* = 4%
        » *Shigella* = 2.5%

## FDA-cleared GI Panels

<table>
<thead>
<tr>
<th>Test</th>
<th>Instrument</th>
<th>Manufacturer</th>
<th>Targets</th>
<th>Time (h)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Enteric Panel (EP)</td>
<td>VERIGENE®</td>
<td>Luminex</td>
<td>9</td>
<td>2</td>
</tr>
<tr>
<td>GI Pathogen</td>
<td>xTAG®</td>
<td>Luminex</td>
<td>14</td>
<td>~5</td>
</tr>
<tr>
<td>FilmArray® GI Panel</td>
<td>Torch®</td>
<td>BioFire Dx</td>
<td>22</td>
<td>1</td>
</tr>
<tr>
<td>EBP</td>
<td>BD Max®</td>
<td>BD</td>
<td>4</td>
<td>~3</td>
</tr>
<tr>
<td>ProGastro</td>
<td>GenProbe®</td>
<td>Hologic</td>
<td>4</td>
<td>4</td>
</tr>
</tbody>
</table>
## BioFire FilmArray® - GI Panel

<table>
<thead>
<tr>
<th>Bacteria</th>
<th>Parasites</th>
<th>Virus</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Campylobacter</em></td>
<td><em>Cryptosporidium</em></td>
<td>Adenovirus</td>
</tr>
<tr>
<td><em>C. difficile</em></td>
<td><em>Cyclospora</em></td>
<td><em>Astrovirus</em></td>
</tr>
<tr>
<td><em>Plesiomonas</em></td>
<td><em>E. histolytica</em></td>
<td><em>Norovirus</em></td>
</tr>
<tr>
<td><em>Salmonella</em></td>
<td><em>Giardia</em></td>
<td><em>Rotavirus</em></td>
</tr>
<tr>
<td><em>Y. enterocolitica</em></td>
<td></td>
<td><em>Sapovirus</em></td>
</tr>
<tr>
<td><em>Vibrio</em></td>
<td></td>
<td></td>
</tr>
<tr>
<td>STEC, ETEC, EPEC, EAEC, Shigella/EIEC</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

# VERIGENE® - Enteric Panel

**Bacteria**
- Campylobacter Group
- Salmonella spp.
- Shigella spp.
- Vibrio Group
- Y. enterocolitica

**Toxins**
- Shiga Toxin 1
- Shiga Toxin 2

**Viruses**
- Norovirus
- Rotavirus

Factors to Consider CIDT’s

• Reliability
  – Results accuracy, analytical specificity, and sensitivity
  – Vulnerability to contamination
• Ease-of-use
• Ability to detect major pathogens
• Flexibility to match the right-patient/right-test concept
• Avoid over-diagnosis, not matching the clinical picture such as C. difficile
• Cost:
  – Capital
  – Per test
Summary

• Testing should be based on history, severity, duration, epidemiological links of the disease, and immune status of the patient
• Utilization and value of CIDT’s for diarrheal illnesses
• Over-diagnosis has to be dealt with by choosing the right patient, the right test, and the right specimen
• Consider testing for parasites when the clinical features (lack of fever, chronicity…) and exposure links (travel, daycare ...) are established or patient is immunocompromised
• Consider testing for C. difficile when there is a history of antimicrobial use or healthcare association of diarrhea
• There are indications for broad syndromic panels, but they’re rare