Shiga Toxin

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Objectives

- Discuss the clinical significance of Shiga-toxin producing *E. coli* (STEC) and its effect on public health
- Demonstrate how STEC can be transmitted and what foods are at highest risk
- Review the ways in which STEC causes significant illness, especially in younger children
- Analyze the need for and how to test and treat for STEC
Top 7 Threats to the Human Race

Global Drivers
- Changed C/N cycles & rising atmospheric GHG concentration
- Increasing antibiotic resistance
- Increasing connectivity (economic, social, ecological)
- Rising human numbers & urbanization
- Increasing per capita resource use
- Nuclear proliferation
- International terrorism

Unwanted Outcomes
- CLIMATE
- ECOSYSTEM
- HUMAN HEALTH
- ECONOMIC

Source adapted from Science, Vol 325, September 2009
Available at http://www.sciencemag.org/content/325/5948.cover-expansion
1970: Surgeon General William Stewart said the US was “ready to close the book on infectious disease as a major health threat”

- Modern antibiotics, vaccination, and sanitation methods had done the job

1995: Infectious disease is the 3rd leading cause of death behind heart disease & cancer

2015: Infectious disease remains a critical concern as antimicrobial resistance increases
Antibiotic Overuse

One in every three inpatients will receive 2 or more antibiotics in the course of their hospital stay

Of the inpatients receiving antibiotics, 3/4 will receive unnecessary or redundant therapy

Each year, tens of millions of antibiotics are prescribed unnecessarily for viral upper respiratory infections

CDC – Get Smart Campaign
Costs of Antibiotic Resistance

Antibiotic resistance increases the economic burden on the entire US healthcare system

- Resistant infections cost more to treat and can prolong healthcare use

More than **$1.1 billion** is spent annually on *unnecessary* antibiotic prescriptions for respiratory infections in adults

In total, antibiotic resistance is responsible for:

- **$20 billion** in excess healthcare costs
- **$35 billion** in societal costs
- **8 million** additional hospital days

CDC – Get Smart Campaign
“A post-antibiotic era means, in effect, an end to modern medicine as we know it. Things as common as strep throat or a child’s scratched knee could once again kill.”

Margaret Chan, WHO Director General
Why Antimicrobial Stewardship?

- A balance of infection prevention and antibiotic management
  - Achieve optimal clinical outcomes
  - Decrease adverse drug events
    - C. difficile
  - Minimize development of antimicrobial resistance
  - Preserve antimicrobial resources
  - Reduce costs
Antimicrobial Stewardship Programs

- Guidelines for Developing an Institutional Program to Enhance Antimicrobial Stewardship – 2006
  http://www.idsociety.org

- Core members include:
  - Infectious Disease Physician
  - Clinical Pharmacist
  - Clinical Microbiologist
  - Infection Control Professional
  - Information System Specialist
Rapid Diagnostics

Test, Target, Treat

• Know the organism, know the appropriate treatment

Reduce antibiotic overuse & unwanted side effects

Shorten time to appropriate therapy

Provide targeted treatment with narrow-spectrum agents when possible

Reduced infection transmission increases infection prevention savings
Gastrointestinal Disease: Impossible but True

- Impossible to diagnose on clinical symptoms alone, but frequently done
- What’s the primary symptom of any GI disease?

DIARRHEA

- 100s of causes, often treated empirically with antibiotics
Diarrhea

Worldwide, it kills 1.5 to 2.5 million children per year

• 5 to 18 cases/year

US statistics

• Adults average 1 acute case/year
• Young children 2 acute cases/year
Diarrhea in the US

- 211-375 million diarrheal illnesses/year
  - 21-37 million episodes in children under five
- 73 million physician consultations
- 1.8 million hospitalizations
- 3100 deaths
- $25 billion in healthcare costs
Foodborne Illness in the US

- 76 million illnesses
- 325,000 hospitalizations
- 5000 deaths
Evaluate severity and duration
Obtain history and physical examination
Treat dehydration
Report suspected outbreaks
Check all that apply:

A. Community acquired or traveler's diarrhea
   (esp. if accompanied by significant fever or blood in stool)
   Culture or test for:
   Salmonella
   Shigella
   Campylobacter
   E. coli O157:H7 (if blood in stool also test for Shiga toxin and refer isolates if toxin pos.)
   C. difficile toxins A ± B (if antibiotics or chemotherapy taken in recent weeks)
   Consider quinolone for suspected shigellosis in adults (fever, inflammation); macrolide for suspected resistant Campylobacter; avoid antimotility or certain antimicrobial drugs if suspected STEC (afebrile, bloody diarrhea)

B. Nosocomial diarrhea
   (onset after >3 d in hospital)
   Test for
   C. difficile toxins A ± B
   (In suspect nosocomial outbreaks, in patients with bloody stools, and in infants, also add tests in panel A)
   Discontinue antimicrobials if possible; consider metronidazole if illness worsens or persists

C. Persistent diarrhea >7d
   (esp. if immunocompromised)
   Consider parasites
   Giardia
   Cryptosporidium
   Cyclospora
   Isospora belli
   + Inflammatory screen
   If HIV pos., add:
   Microsporidia
   (Gram-chromotrope)
   M. avium complex
   + panel A
   Treat per results of tests
Etiological Agents of Diarrhea

**Viral infections**
- Rotavirus, Norovirus, Cytomegalovirus (in immune compromised), Astrovirus, Adenovirus, etc.

**Bacterial infections**
- Campylobacter, Salmonella, Shigella, and *E. coli*

**Parasites**
- *Giardia lamblia*, *Entamoeba histolytica*, and *Cryptosporidium*
Escherichia coli (E. coli) Basic Information

Ubiquitous in the gut of healthy individuals & many animals

• Most strains are not only harmless, but beneficial

May cause illness

• Diarrhea
• Urinary tract infections
• Respiratory illness & pneumonia

Can be used to monitor water quality

• Fecal coliforms are bacteria that live in stool of warm-blooded animals
• Can have direct contamination of water with stool
• Can have run-off from farm irrigation
• Can have breaks in septic systems
What are Shiga-Toxin Producing *E. coli*?

**E. coli** that can produce Shiga toxin (STEC)

- Also called Entererohermorrhagic *E. coli* (EHEC) and verocytotoxic *E. coli* (VTEC)
- Identical toxin to what is produced in *Shigella*

Most famous is *E. coli* O157:H7

- Other non-O157 STEC serogroups that often cause illness in people in the United States include O26, O111, and O103.

**Shiga toxin**

- Comes in two main groups; Stx1 and Stx2
- Toxin that inhibits protein synthesis in target cells
- Stx2 more associated with complications like HUS
Additional Issues with STEC

- Bacteria are often acid tolerant
- Low infectious dose (<100 organisms)
- More common during the summer months
What are the Symptoms of STEC?

<table>
<thead>
<tr>
<th>Varies per person</th>
<th>Main symptoms</th>
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<tbody>
<tr>
<td></td>
<td>• Many can be subclinical</td>
</tr>
<tr>
<td></td>
<td>• Diarrhea (potentially bloody)</td>
</tr>
<tr>
<td></td>
<td>• Severe stomach cramps</td>
</tr>
<tr>
<td></td>
<td>• Vomiting</td>
</tr>
<tr>
<td></td>
<td>• May have fever</td>
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<table>
<thead>
<tr>
<th>Disease Resolution</th>
<th></th>
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<tr>
<td></td>
<td>• Usually 5 to 7 days</td>
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</table>
Complications of STEC

Of those diagnosed with STEC, around 5-10% can get hemolytic uremic syndrome (HUS)

Symptoms of HUS
- Bloody diarrhea
- Decreased urination
- Tired
- Loss of pink color in cheeks and inside of lower eyelid

Major Issue of HUS
- Kidneys may stop working among other complications
- People with HUS may recover in weeks, suffer permanent damage, or potentially death
How Do Patients Develop HUS?

Mechanism

- The toxin causes the blood cells to become misshapen
- Blood cells can clog the tiny vesicles in the kidneys known as glomeruli
- Filtration by the kidneys becomes blocked
- Kidneys may shut down

Usually children
Thrombotic Thrombocytopenic Purpura (TTP)

**Mechanism**

- Toxin changes shape of the blood cells
- Clots form in blood vessels
- Oxygen does not get to organs like brain, heart, and kidneys
- Platelets get used up
- Bleeding problems arise whether internally, under skin, or cut

More likely in adults
Incubation Period

Average time between consumption of STEC to disease is 3-4 days

• Time can vary from 1 to 10 days

Symptoms increase over time

• Stomach ache to mild diarrhea
• Increases to more significant diarrhea to bloody diarrhea

HUS, if it happens, usually is around a week after initial symptoms
STEC Stats

CDC estimates 265,000 cases per year in the US

- *E. coli* O157:H7 accounts for roughly 36% of these infections

Why aren’t they seen in hospitals?

- Many patients don’t seek medical attention
- Those patients that do may not give a stool sample
- Laboratories may not be able to test stool samples
Infecting Others

Typically, toxin-producing bacteria go away with symptoms.

For some individuals, symptoms may resolve, but can still have STEC for months.

Therefore, important to keep up appropriate handwashing!
Transmission of STEC

**Fecal oral route**
- Directly from people not washing hands and touching others
- Indirectly from people not washing hands and handling food
- Daycares with diapers

**Food exposure**
- Meat can be directly contaminated during processing
- Other foods like water runoff from cow pasture to farm

**Environmental Exposure**
- Lake/pool/water park
- Petting zoos & other animal exhibits
STEC Transmission

- Community acquired
- Food & water borne
- Outbreaks
E. coli O157:H7 Outbreaks
Worldwide, 1982–present

Printed with permission from Ellin Doyle. From: Human Illness Caused by E. coli O157:H7 from Food and Non-food Sources. http://fri.wisc.edu/docs/pdf/FRIBrief_EcoliO157H7humanillness.pdf
Multistate *E. coli* O157:H7 in Ground Beef – June 2014

**Major statistics**
- 12 cases with 7 being hospitalized
- Spanning 4 states
- Zero deaths

**Why**
- Contaminated ground beef at Wolverine Packing company
- 1.8 million pounds distributed to restaurants and retail throughout the nation

**Action**
- Recall happened AFTER use-by date
- Consumers should check their frozen meat
Multistate *E. coli* O121 in Raw Clover Sprouts – August 2014

**Major Statistics**
- 19 cases with 44% hospitalization
- 6 states
- Zero deaths

**Why?**
- Evergreen Fresh Sprouts, LLC of Idaho had contaminated raw clover sprouts
- In interviews, 81% of people ate raw clover sprouts the week before, often from local restaurants

**Action**
- FDA inspection saw multiple unsanitary conditions often involving corroded metal in contact with food
- The suspect seed lot was discontinued
Multistate *E. coli* O121 in Farm Rich Brand Frozen Food – May 2013

Major Statistics
- 35 cases with 9 hospitalizations
- 19 states
- Zero deaths, but 82% were under 21 and 2 developed HUS

Why?
- O121 found in frozen mini pizza slices

Action
- Recall expanded to include all Farm Rich, Market Day, and Schwan’s brand frozen food products
Diagnostic Recommendations
Why Should People Test?

Hydration

• May want to treat with parenteral volume expansion if it is early in the infection

Do not take antibiotics

• Increases risk of HUS
• Antidiarrheal agents like Imodium may also increase risk of HUS
Antibiotics and STEC

- Potentially fatal renal failure
- More prevalent in pediatrics
- Antibiotics may increase risk of HUS due to toxin release from dying STEC cells
Who should be tested?

- CDC & Joint Commission: All acute community acquired diarrheas

### Screening Criteria

<table>
<thead>
<tr>
<th>Screening Criteria</th>
<th>Test if</th>
<th>Risk</th>
</tr>
</thead>
<tbody>
<tr>
<td>Visible blood</td>
<td>Present</td>
<td>Not all STEC positives have visible blood</td>
</tr>
<tr>
<td>Season</td>
<td>Summer-Fall</td>
<td>Can occur year-round, esp. with imported produce</td>
</tr>
<tr>
<td>Patient Age</td>
<td>Pediatrics, Elderly</td>
<td>Not all cases are peds &amp; elderly</td>
</tr>
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</table>

- Antibiotic use is common for GI disease, may be deadly in a STEC patient
- Rapid identification of potential outbreaks
O157 and non-O157 strains

O157:H7 is the most common US strain

- Traditionally labs only tested for O157

Over 100 non-O157 strains cause disease

- April 2013 outbreak of O121
- 2011 German outbreak of O104
  - Over 3,500 cases, 23% developed HUS
Detecting O157 vs. non-O157

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<tr>
<th>O157-specific Assays</th>
<th>Assays for Toxins</th>
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<tbody>
<tr>
<td>Test for O157 strain only</td>
<td>Test for toxins Stx1 &amp; Stx2</td>
</tr>
<tr>
<td>Will miss all non-O157 STEC strains</td>
<td>Will detect all toxin-producing strains</td>
</tr>
<tr>
<td>Plate culture (SMAC, mac-sorb) most common</td>
<td>Rapid assay or ELISA most common (may require broth enrichment)</td>
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CDC Recommendations

Test all stools from community-acquired diarrhea for STEC

Concurrent testing

• Culture stool for O157
• Testing for Shiga toxin

Actions

• All O157 STEC isolates should be forwarded to appropriate public health lab for confirmatory testing & additional characterization
• Results should be reported promptly to physician
Rationale for CDC Recommendation

There are publications supporting that STEC may be as common as other bacterial pathogens

- STEC (0% - 4.1%)
- Salmonella (1.9% - 4.8%)
- Campylobacter (0.9% - 9.3%)

Early diagnosis is important for proper treatment

- Volume expansion for O157
- No antibiotics to lessen risk of HUS

Help detect outbreaks earlier
Why Every CA Stool?

Many infections can be missed when having selective strategies

• Just children
• Just summer

Some STEC don’t have visible blood and other pathogens may

Pathogens will be more difficult to recover from stools at a later date

Help prevent transmission to others

• Children may not be able to return to daycare facilities
• Food service workers
E. coli O157 Culture

Plate on sorbitol-MacConkey agar plate or chromogenic agar

- O157 does not ferment sorbitol within 24 hours

Wait 16-24 hours @ 37°C

Look for color

- SMAC plates have colorless colonies
- Chromogenic agar has specific colony colors

Test well-isolated colony on antiserum or latex

- Screen at least 3 colonies

Preliminary findings reported

Confirmatory testing
Shiga Toxin Assays

**Major benefit**
- Detects all Shiga-toxin producing *E. coli* rather than just O157

**What to test?**
- See manufacturer’s product inserts on what is approved

**Direct stool**
- Fresh
- Frozen

**Cultures**
- Broth culture
- Plate culture
Stool testing For Shiga Toxin

Major Benefit – Detects toxin producers beyond *E. coli* O157

Direct stool detection provides real-time actionable results

- Initiate proper treatment while patient is in the healthcare setting
- Avoid antibiotic exposure

Ideal for emergency situations, pediatrics with bloody diarrhea

Identify potential outbreaks sooner
Molecular Assays

No single Shiga-toxin assay available, but two multiplex assays

Public health laboratories may use home brew molecular

Can be done on isolated colonies or clinical samples
How Does Broth Culture Compare?

2 published studies from Alberta, Canada looked at performance of rapid assays vs. in-house PCR

- “EHEC Assay” study compared market leader in 2013 to PCR
- “STEC Assay” wasn’t available then, study done in 2014
- Similar regional demographics, methods, similar sample size, prevalence rate

Both assays used broth enrichment culture

STEC Assay also tested directly on stool specimens

Broth Culture Comparison

- EHEC Assay not recommended as a routine screening test for STEC from broth cultures due to low sensitivity and risk of false positives.

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<th>PPV</th>
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<td>EHEC Assay (broth culture)</td>
<td>35%</td>
<td>54.5%</td>
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- STEC Assay is a viable alternative to molecular testing for frontline labs as a primary screening method for STEC.

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<tr>
<td>STEC Assay (broth culture)</td>
<td>85%</td>
<td>100%</td>
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Broth Culture Comparison

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- Majority of true positives not detected
- Almost half of all positive results were false positive
- More positives detected
- All positive results were confirmed as true positive

PPV = Positive Predictive Value
Preventing STEC

Hand washing!
- After restroom
- Changing diapers
- Prior to preparing food
- Contact with animals

Prepare food appropriately
- Make sure meat – especially ground beef – is cooked thoroughly
- Don’t use cutting board for meat and then other food
- Clean cutting boards

Avoid drinks that are unpasturized that are supposed to be
- Raw milk
- Dairy products
- Apple cider
Discussion

Shiga toxin producing *E. coli* are a significant health risk

Diagnostic testing can help better direct therapy

Appropriate therapy can reduce complications, such as HUS and TTP