Molecular epidemiology of *Clostridium difficile* infection in British Columbia, Canada

Agatha Jassem, PhD
Senior Scientist, BCCDC Public Health Laboratory
Objectives

• Molecular typing methods for *C. difficile*

• Prevalence & incidence of *C. difficile* infection (CDI)

• Risk factors & sources associated with CDI

• Molecular epidemiology of CDI in British Columbia from recent studies
Clostridium difficile infection (CDI)

• *C. difficile* is becoming the most common pathogen of healthcare–associated (HA) infections

• The epidemiology of CDI is changing

An understanding of *C. difficile* epidemiology is necessary to understand CDI diagnosis and guide infection prevention and control practices.
THE PAST
Historical Burden of HA-CDI

1970’s: report of severe diarrhea and pseudomembranous colitis associated with clindamycin use in a hospital setting, followed by association with toxin-producing *Clostridium*


Historical Burden of HA-CDI

• The hospital environment is contaminated with *C. difficile* spores that can persist for weeks and are resistant to detergent-based cleaners

• *C. difficile* can be transmitted from patients with CDI and from asymptptomatically colonized patients through the hands of healthcare workers or contaminated surfaces and objects

Molecular Epidemiology

Evaluation of strain diversity & transmission dynamics

Limitation: no universal, sensitive, reproducible method

• Toxinotyping
  – DNA patterns based on toxin genes

• Pulsed field gel electrophoresis (PFGE)
  – DNA pattern based on genome fragmentation by an enzyme

• Restriction endonuclease analysis (REA)
  – DNA pattern based on genome fragmentation by an enzyme, more frequent cutting than in PFGE

• PCR Ribotyping
  – DNA patterns based on 16S and 23S ribosomal RNA

• Multilocus variable number tandem repeat analysis (MLVA)
  – DNA pattern based on the copy number of parts of a gene
Historical Burden of HA-CDI

• *C. difficile*: diverse group of organisms

• New strains are frequently introduced to hospitals, some of which result in hospital transmission to multiple patients, whereas most are sporadic and result in few or no transmissions
  – Variations in types geographically and among institutions
  – Variations in types between years and seasons

Historical Burden of HA-CDI

BI/NAP1/027 emergence in early 2000’s

• Increases in CDI incidence, particularly in the Northeast within North America, largely attributed to the emergence of this strain

• North American pulsotype 1 (NAP1) strain is fluoroquinolone-resistant and associated with severe disease and death

Gould and McDonald (2012) Clin Infect Dis
Global Spread of NAP1 *C. difficile*

By whole-genome sequencing & phylogenetic analysis: two lineages emerged in North America

He et al. (2013) Nature Genetics
Characteristics of NAP1 C. difficile

Produces toxin A, toxin B, binary toxin, and contains deletions in the repressor of toxins A/B (tcdC):
“hypervirulent” strain?

Murray (2009) BMC Infect Dis
NAP1 *C. difficile* in British Columbia

At BCCDC Public Health Laboratory: DNA fingerprinting by pulsed field gel electrophoresis (PFGE)
NAP1 in BC: 2008 Molecular Epidemiology Study

From 13 hospital labs & 1 province-wide community lab

• Collected toxin A/B positive stool or isolates in March
• N=341 isolates: 271 (79.5%) and 70 (20.5%), respectively
• **NAP1 designation accounted for 42.7% of isolates**
  – NAP2 for 10.2%, NAP4 for 13.5%, non-NAP for 21.3%


In collaboration with BC Provincial Infection Control Network & BC Association of Medical Microbiologists
2008 BC-wide Molecular Epidemiology

From 13 hospital labs & 1 province-wide community lab

• Mean patient age was 69.3 years (median 77 years)
• Patients at community sites were 15.4 years younger (mean 57.1 years) than those at hospital sites
• NAP1 isolates were identified proportionally more at hospital sites (49.6% vs 15.7%), while community sites had more non-NAP isolates (37.1% vs 17.3%)


In collaboration with BC Provincial Infection Control Network & BC Association of Medical Microbiologists
Established Model for CDI Acquisition

Patients exposed to *C. difficile* spores through contact with hospital environment or healthcare workers

Risk factors for developing HA-CDI

- Antimicrobial exposure
- Prior hospitalization
- Long duration of hospitalization
- Increased patient age
- Comorbidity
- PPIs and H2 antagonists
- Abdominal surgery

http://www.illorem.com
THE PRESENT
Current Burden of HA-CDI in US

- *C. difficile* is the most commonly reported pathogen (12.1% of HAIs in 10 states in one study), surpassing *Staphylococcus aureus*
  - Rates of CDI in US hospitals have increased steadily since 1993 to more than 336,000 in 2009

Lucado, Gould, Elixhauser (2011) US Dpnt Health Human Services
Current Burden of HA-CDI in Canada

*Clostridium difficile* Associated Disease (CDAD) Surveillance

Number of Healthcare-Associated-*Clostridium difficile* infection cases and incidence rate per 10,000 patient days by region

<table>
<thead>
<tr>
<th>Year</th>
<th>Western No. cases</th>
<th>Western Rate</th>
<th>Central No. cases</th>
<th>Central Rate</th>
<th>Eastern No. cases</th>
<th>Eastern Rate</th>
<th>Overall No. cases</th>
<th>Overall Rate</th>
</tr>
</thead>
<tbody>
<tr>
<td>2007</td>
<td>1,180</td>
<td>6.06</td>
<td>1,831</td>
<td>8.22</td>
<td>260</td>
<td>3.95</td>
<td>3,271</td>
<td>6.77</td>
</tr>
<tr>
<td>2008</td>
<td>1,060</td>
<td>8.06</td>
<td>1,597</td>
<td>7.36</td>
<td>256</td>
<td>3.86</td>
<td>2,913</td>
<td>7.02</td>
</tr>
<tr>
<td>2009</td>
<td>683</td>
<td>6.87</td>
<td>1,401</td>
<td>5.91</td>
<td>161</td>
<td>3.19</td>
<td>2,245</td>
<td>5.80</td>
</tr>
<tr>
<td>2010</td>
<td>1,251</td>
<td>6.84</td>
<td>1,266</td>
<td>6.76</td>
<td>155</td>
<td>2.27</td>
<td>2,672</td>
<td>6.09</td>
</tr>
<tr>
<td>2011</td>
<td>1,170</td>
<td>6.10</td>
<td>1,910</td>
<td>8.37</td>
<td>101</td>
<td>2.88</td>
<td>3,181</td>
<td>6.99</td>
</tr>
</tbody>
</table>
Current Burden of HA-CDI in Canada

From 2007 to 2012:

- Overall rates peaked in 2008, then remained stable
- NAP1 strain type were the most dominant type followed by NAP4

2012: estimated 37,900 CDI episodes (27% recurrence)

Canadian Nosocomial Infection Surveillance Program (CNISP)
Current Burden of HA-CDI in BC

• 2,260 CDI cases reported in 2014/15
  – A 4.9% decrease from 2013/14 and the lowest annual number since 2009/10

• Rate of new CDI in 2014/15 was 4.2 per 10,000 inpatient days
  – A 51.2% decrease from 2009/10

• No diagnostic changes in past two years

• Decreases attributed to intervention measures?
Current Burden of HA-CDI in BC

But...
Changing epidemiology of HA-CDI

• ≥30% of diarrheal deaths in the elderly occur in nonhospital care settings, like **long-term care facilities** (LTCFs)

• Incidence and recurrence rates of CDI in LTCFs are comparable to those of acute-care hospitals

• There have been several outbreaks of CDI in LTCFs

• CDI risk factors in LTCFs are previous antibiotic use and frequent transitions between hospitals and LTCFs

Community-associated (CA) CDI

- In the 2000’s: first reports of CDI in the community

- Incidence of CA-CDI suggested to be increasing across Europe & North America and occurring in younger (median 53 vs 78 years), healthier persons, with fewer risk factors associated with HA-CDI

- Patients with CA-CDI are less likely to recur than HA-CDI (10% vs 20%)

CDC (2005) MMWR Morb Mortal Wkly Rep
Dumyati et al. (2012) Emerg Infect Dis
Lessa et al. (2012) Clin Infect Dis
Prevalence of CA-CDI in BC

<table>
<thead>
<tr>
<th>Year</th>
<th>Unknown</th>
<th>CA</th>
<th>HCA with another facility, relapse</th>
<th>HCA with reporting facility, relapse</th>
<th>HCA with another facility, new cases</th>
<th>HCA with reporting facility, new cases</th>
<th>Total number of CDI cases</th>
</tr>
</thead>
<tbody>
<tr>
<td>2009/10</td>
<td>63</td>
<td>516</td>
<td>62</td>
<td>390</td>
<td>171</td>
<td>2,210</td>
<td>3,422</td>
</tr>
<tr>
<td>2010/11</td>
<td>72</td>
<td>812</td>
<td>63</td>
<td>362</td>
<td>143</td>
<td>2,152</td>
<td>3,604</td>
</tr>
<tr>
<td>2011/12</td>
<td>104</td>
<td>753</td>
<td>54</td>
<td>302</td>
<td>188</td>
<td>2,212</td>
<td>3,613</td>
</tr>
<tr>
<td>2012/13</td>
<td>96</td>
<td>794</td>
<td>71</td>
<td>297</td>
<td>153</td>
<td>1,835</td>
<td>3,246</td>
</tr>
<tr>
<td>2013/14</td>
<td>63</td>
<td>636</td>
<td>46</td>
<td>201</td>
<td>121</td>
<td>1,309</td>
<td>2,376</td>
</tr>
<tr>
<td>2014/15</td>
<td>62</td>
<td>674</td>
<td>47</td>
<td>139</td>
<td>132</td>
<td>1,206</td>
<td>2,260</td>
</tr>
</tbody>
</table>
Changing epidemiology of CDI

CDI acquisition is occurring in the outpatient setting and in those with no healthcare exposure

% of CDI cases (n=10,342) by inpatient or outpatient status and type/location of exposure

CDC (2012) MMWR Morb Mortal Wkly Rep
Shifts in *C. difficile* strain types

- In some European hospitals the circulation of BI/NAP1/027 strains is decreasing
- **Several countries are reporting emergence of non-BI/NAP1/027 stains**, mostly in healthcare settings
  - Ribotypes 078 (NAP7), 244, 012, 017, 018, 014/20, 106, 015
  - Data on strain types in persons in the community are sparse
  - In North America, still NAP1 dominance?

---

Lim *et al.* (2014) Clin Infect Dis  
Bauer *et al.* (2011) Lancet  
Cairns *et al.* (2015) J Clin Microbiol  
De Almeida *et al.* (2013) NZ Med J  
Hensgens *et al.* (2009) Euro Surveill  
Diversity of *C. difficile*

- *C. difficile* strains are naturally diverse and can evolve rapidly into those with increased pathogenicity.

- Epidemic strains result in outbreaks, but then diminish in frequency and are replaced by new types.

- New *C. difficile* strains are frequently introduced to hospitals from a large pool of strains.

He *et al.* (2010) Proc Natl Acad Sci
2013 BC Follow-Up Study

Specimens from 1 hospital lab within Island Health
- Collected toxin B positive stools from March-July (n=68)
- Proportion (%) of community-associated inpatient cases:

<table>
<thead>
<tr>
<th>Year</th>
<th>2008</th>
<th>2009</th>
<th>2010</th>
<th>2011</th>
<th>2012</th>
<th>2013</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>11.7</td>
<td>17.0</td>
<td>22.0</td>
<td>28.3</td>
<td>34.5</td>
<td>35.6</td>
</tr>
</tbody>
</table>

2013 BC Follow-Up Study

Specimens from 1 hospital lab within Island Health

• N=68 isolates: 38 (55.9%) HA- and 30 (44.1%) CA-CDI

• Patients with CA-CDI were 16.2 years younger (mean 58.7 years) than those at hospital sites

• The proportion of non-NAP isolates was high in CA-CDI (50.0%) and HA-CDI (39.5%) cases

Case definitions: medical record review discriminated between inpatients with CA-CDI and HA-CDI. Outpatients and inpatients with CA-CDI were collectively referred to as cases.

2013 BC Follow-Up Study

Specimens from 1 hospital lab within Island Health

- **Isolates not matching a NAP type accounted for 44.1%**
  - NAP1 for 11.8%, NAP4 for 22.1%, NAP6 for 11.8%
  - Among no NAPs, no dominant pattern or ribotype

- **Between 2008-13, there was a 28.9% decrease in NAP1 and 20.0% increase in isolates without NAP designation**

2013 BC Follow-Up Study

• Taken together, the high proportion of isolates not matching described NAP types among the 2008 community site-collected isolates & the 2013 CA-CDI isolates could relate to a shift of strains from community sources to the healthcare setting.

• Therefore, the observed shift in NAP distribution between 2008 and 2013 in the Island Health Authority region could reflect the steady increase in the proportion of CA-CDI inpatient cases identified.

THE FUTURE
Role of Whole Genome Sequencing

- WGS: a new method of *C. difficile* molecular typing
  - Most sensitive and specific typing method
- Pairing patient interaction and WGS data, a hospital in the UK found that for non-outbreak cases distinct sources other than symptomatic CDI cases are involved in most (62%) cases of CDI. *C. difficile* transmission

Role of Whole Genome Sequencing

Study the dynamics of *C. difficile* transmission and CDI recurrence among symptomatic patients

Understanding Sources of CDI

• Hospital exposures, including asymptomatic carriers

• Outpatient health care exposures
  – Many CA-CDI cases have outpatient health care exposures (doctor/dentist offices, outpatient surgeries, etc) prior to CDI & *C. difficile* has been isolated from environmental surfaces in outpatient clinics

• Exposure to household members with CDI & infants
  – Association with increased risk of CA-CDI (infants have a high rate of *C. difficile* colonization)

*Chitins et al.* (2013) JAMA Intern Med  
Understanding Sources of CDI

- **C. difficile** has been isolated from food and animals
  - Prevalence in retail meats <7%, in vegetables 3-5%
  - Widespread in dogs, cats, pigs, calves, horses, sheep
  - Ribotype 078 (NAP 7) mostly among animal sources
  - WGS showed that farmers and pigs were colonised with identical & nearly identical *C. difficile* clones

- **C. difficile** has been isolated from soil, sewage, marine sediments, swimming pools, etc.

Knetsch *et al.* (2014) Eurosurveill  
Actual Model of CDI?

1. Infectious Agent

+ strain type

Ex. NAP7 in animals

Ex. Toxin A-negative ribotype 017

2. Reservoir

3. Exit Portal

4. Transmission

Contact

Droplets

Vector

Vehicle

Fomite

5. Entry Portals

Nasal / Oral

Blood

GI / Genital

Skin

6. Susceptible Host

The Chain of Infection

Cairns et al. (2015) J Clin Microbiol

www.atrainceu.com Toni Thompson
Molecular Epid for Prevention of CDI

- It is likely that humans ingest *C. difficile* frequently
- Comprehensive surveillance defining CA- and HA-CDI cases will elucidate sources of acquisition and transmission and identify emerging strains

- Strain description
- Sensitive techniques

www.vch.ca
Bomers *et al.* (2012) BMJ
Acknowledgements

- **BC Provincial Infection Control Network** – Bruce Gamage, BSN
- **BC Association of Medical Microbiologists** – Dr. Sylvie Champagne
- **Island Health Authority** – Dr. Pamela Kibsey, Dr. Kennard Tan
- **National Microbiology Lab** – Dr. George Golding, Dr. Michael Mulvey
- **BC Centre for Disease Control** - Dr. Bonnie Henry, Dr. Fawziah Marra
- **BCCDC Public Health Laboratory**
  – Patricia Umlandt, Dr. Natalie Prystajecky, Dr. Linda Hoang