

Challenge M171-2

May 2017

Stool: *Campylobacter coli*

HISTORY

A simulated stool collected from a 25 year old traveler returning from Finland was sent to category A and B laboratories.

Participants were expected to identify *Campylobacter coli*.

CMPT QA/QC/STATISTICS

All simulated stool samples are produced at CMPT according to CMPT internal protocols. The sample contained a mixed culture of *Campylobacter coli* and *Escherichia coli* as a background organism.

The samples are assessed for homogeneity and stability using in-house quality control methods and random selection of samples before and during production, and post sample delivery. The number of random samples selected is 15% of the total production batch.

The challenge sample lot was confirmed to be homogeneous and stable for at least 18 days.

Organism identification was confirmed by a reference laboratory.

All challenge components have in-house assigned values based on the most clinically appropriate result; the most clinically appropriate result is determined by expert committee evaluation. No further statistical analysis is performed on the results beyond that described under "Suitability for grading."

SURVEY RESULTS

Reference laboratories: 12/13 (92%) laboratories reported *Campylobacter coli*, 1 laboratory reported *Campylobacter* species, refer. 12/13 (92%) laboratories indicated they would report to Public Health; 1 laboratory is a PH laboratory.

Participants (Tables 1 and 2): All participants processing the sample identified the organism at least to the genus level. 27/57 laboratories reported *Campylobacter coli*; 1 laboratory reported *C. jejuni/coli*; 18 participants reported species, refer. See table 1 for reports and grades.

MAIN EDUCATIONAL POINTS from M171-2

1. Hydrolysis of sodium hippurate is the major test for distinguishing *C. jejuni* from other *Campylobacter* species, especially *C. coli*.
2. MALDI-TOF has been shown to be a reliable means of identification for *Campylobacter* species.
3. *Campylobacter* infections are among the most common causes of bacterial diarrhea both in the developed and developing worlds.
4. Reporting to Public Health agencies is required in every province in Canada.

54/57 (95%) participants indicated they would report the isolate to Public Health and thus, received a grade of 4 (table 2).

Suitability for Grading

A challenge is considered suitable for grading if agreement is reached by 80 percent of selected reference group and at least 50 percent of the participants.

Organism identification was correctly performed by at least 80 percent of reference laboratories and greater than 50 percent of all laboratories and was thus, determined to be suitable for grading.

Grading

Maximum grade: 8

Reporting *C. coli*, *C. jejuni/coli* was graded 4

Reporting *Campylobacter* species with referral was graded 4 and downgraded to 3 if not referred.

Reporting *Campylobacter lari* was graded 3.

Reporting NSSEYVC was graded 0.

Reporting the isolate to PH was graded 4.

Not reporting a *Campylobacter* to PH was graded 0.

Table 1. Identification results

Reported	A labs	B labs	Total	Grade
<i>Campylobacter coli</i> , ± refer	27		27	4
<i>Campylobacter jejuni/coli</i>	1		1	4
<i>Campylobacter</i> spp, refer ± presumptive	17	1	18	4
<i>Campylobacter</i> species, not <i>C. jejuni</i>	2		2	3
<i>Campylobacter</i> species	8		8	3
<i>Campylobacter lari</i>	1		1	3
NSSEYVC	1			0
Unable to ID due to equipment malfunction, refer	1			ungraded
snp ± refer	6		8	ungraded
Total	64	1	65	

NSSEYVC: no *Salmonella*, *Shigella*, *Edwardsiella tarda*, *Yersinia*, *Vibrio*, or *Campylobacter*

Table 2. Public Health notification

Reported	Total	Grade
yes	52	4
Infection control department notified	2	4
no report	1	0
refer (lab is a Public Health lab)	1	ungraded
refer	1	ungraded
snp	8	ungraded
Total	65	

COMMENTS ON RESULTS

A grade of 4 was given for the correct identification of the organism, and a grade of 4 was also given for having reported to public health. Reporting *Campylobacter* species without referral or incorrect species name without referral were downgraded to 3. Laboratories that do not normally process this sample type were not graded. Not reporting to public health was graded 0.

ISOLATION and IDENTIFICATION

Campylobacter species are curved, S-shaped or spiral, non-spore forming, gram negative rods.

Organisms are usually motile and are generally microaerobic; some strains grow aerobically or anaerobically. An atmosphere containing increased hydrogen is required by some species for microaerobic growth.¹

Although *Campylobacter* species can grow quite slowly, 72 to 96 hrs may be required for primary isolation from stool samples. With the use of selective media and incubation of selective plates at 42°C, isolates are commonly detected at 24 hours. Isolation from blood may take longer² but growth of enteric isolates is supported by commercial blood culture systems.

Isolation from stool samples is usually accomplished by the use of media containing a cephalosporin (e.g. cephalothin or cefoperazone) as most *Campylobacter* species are resistant to these agents while most other stool flora are susceptible.^{1,2} Likewise, the use of incubation temperatures of 42°C to exploit the thermotolerant nature of *C. jejuni* and *C. coli* both promotes their growth and represses other bacterial species.³

Because some non-*jejuni* *Campylobacter* species do not grow on selective media, the filter method and antibiotic-free media can be used if initial results of cultures are negative and the suspicion of *Campylobacter* infection remains high.²

For stool isolates, oxidase-positive colonies exhibiting a characteristic Gram stain appearance isolated from selective media incubated at 42°C under microaerobic conditions can be reliably reported as *Campylobacter* species; hydrolysis of sodium hippurate is the major test for distinguishing *C. jejuni* from other *Campylobacter* species (especially *C. coli*). Occasional strains of *C. jejuni* are hippurate negative, making them more difficult to identify.¹ MALDI – TOF has been shown to be a reliable means of identification for *Campylobacter* species.⁴

If identification of other species is required, isolates should be examined by reference laboratories. The former practice of using resistance to nalidixic acid and cephalothin in the identification of *Campylobacter* species is not reliable because of the prevalence of resistance to antimicrobial agents.

Typing of *Campylobacter* stool isolates is not routinely performed, although typing methods have been used to identify outbreaks.

Alternative methods of detection, including EIA and PCR assays, have been described for detection of *Campylobacter* species in stool specimens. A comparison of commercial kits and culture, which used a PCR method as the reference method found their sensitivities compared favorably to culture, with high specificity.⁵

ANTIMICROBIAL SUSCEPTIBILITY

Symptomatic *C. jejuni* infections are most commonly self-limited and do not require antibiotic therapy. Therapy is restricted to severe infections (fever, bloody stool, severe cramping) or in patients with comorbidities.⁶

When antimicrobial therapy is indicated, macrolides remain the frontline agents for treating culture-confirmed *Campylobacter* cases. Fluoroquinolones are also commonly used.⁷ The increase in fluoroquinolone resistance prompted studies to evaluate the efficacy of single-dose macrolide antibiotic, azithromycin, which has superior efficacy to fluoroquinolone antibiotics.⁶

A Canadian study published in 2015⁸ reported the prevalence of antibiotic resistance among clinical isolates of *C. jejuni* and *C. coli* recovered in Ontario during 2011–2013. The authors found 30% of *C. jejuni* and 41% of *C. coli* resistant to ciprofloxacin; 4% and 13% resistant to erythromycin and 64% and 54% resistant to tetracycline respectively.

CLINICAL RELEVANCE

Campylobacter infections are among the most common causes of bacterial diarrhea both in the developed and developing worlds. Human infections are usually transmitted by the ingestion of undercooked poultry or contact with farm animals.⁶

A variety of animals (chicken, cattle, sheep, and pigs) have been implicated as reservoirs; *Campylobacter* species may also be present in domestic pets.¹ In 2010, Canadian Integrated Program for Antimicrobial Resistance Surveillance (CIPARS) reported that 42.4% of retail chicken in British Columbia, 27.3% in Saskatchewan, 27.6% in Ontario, 21.3% in Quebec, and 35.8% in Maritimes were infected with *Campylobacter* spp.⁹

Campylobacter infects the small intestine and colon, causing diarrhea (occasionally bloody), abdominal cramps, nausea and fever; onset is usually within 1-7 days and symptoms can last several days to more than 1 week and occasionally is prolonged.⁶ *Campylobacter* infections may mimic acute appendicitis and result in unnecessary surgery.

Infection with *Campylobacter* has been associated with the development of Guillain-Barre syndrome (GBS), an immune-mediated demye-

linating polyneuropathy of peripheral nervous system (PNS). Onset occurs at a mean of 9 days following infection and is not caused by direct infection, but by the immune response.¹⁰ This is a result of a cross-reactive immune response between epitopes present in the *Campylobacter*'s lipopolysaccharide (LPS) and the ganglioside targets on peripheral nerves.¹¹

REFERENCES

1. Fitzgerald C., Nachamkin I. *Campylobacter* and *Arcobacter*. In: Jorgensen ea, ed. *Manual of Clinical Microbiology*. Vol 1. 10th ed. ed. Washington, DC.: ASM; 2015:998.
2. Acheson D, Allos BM. *Campylobacter jejuni* Infections: Update on Emerging Issues and Trends. *Clinical Infectious Diseases*. 2001;32:1201-1206.
3. On SL. Isolation, identification and subtyping of *Campylobacter*: where to from here? *J Microbiol Methods*. 2013;95:3-7.
4. Bessede E, Delcamp A, Sifre E, Buissonniere A, Megraud F. New methods for detection of campylobacters in stool samples in comparison to culture. *J Clin Microbiol*. 2011;49:941-944.
5. Granato PA, Chen L, Holiday I, et al. Comparison of Premier CAMPY Enzyme Immunoassay (EIA), ProSpecT *Campylobacter* EIA, and ImmunoCard STAT! CAMPY Tests with Culture for Laboratory Diagnosis of *Campylobacter* Enteric Infections. *J Clin Microbiol*. 2010;48:4022-4027.
6. Kirkpatrick BD, Tribble DR. Update on human *Campylobacter jejuni* infections. *Curr Opin Gastroenterol*. 2011;27:1-7.
7. Ge B, Wang F, Sjolund-Karlsson M, McDermott PF. Antimicrobial resistance in *Campylobacter*: susceptibility testing methods and resistance trends. *J Microbiol Methods*. 2013;95:57-67.
8. Riley A, Eshaghi A, Olsha R, Allen VG, Patel SN. Antibiotic susceptibility of clinical isolates of *Campylobacter jejuni* and *Campylobacter coli* in Ontario, Canada during 2011–2013. *Diagnostic Microbiology and Infectious Disease*. 2015;83:292-294.
9. Agunos A, Leger D, Avery BP, et al. Ciprofloxacin-resistant *Campylobacter* spp. in retail chicken, western Canada. *Emerg Infect Dis*. 2013;19:1121-1124.
10. Rees J, S Soudain, N Gregson, R Hughes. *Campylobacter* infection and Guillain Barre syndrome. *NEJM* Nov 23, 1995. 1374-1379.
11. Nyati KK, Nyati R. Role of *Campylobacter jejuni* infection in the pathogenesis of Guillain-Barre syndrome: an update. *Biomed Res Int*. 2013;2013:852195.