

Cup O' Clostridium: A Laboratory Investigation of a Ramen Night *C. perfringens* Outbreak

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Background: On February 19, 2016, the Division of Consolidated Laboratory Services (DCLS) received patient specimens and leftover food from the Thomas Jefferson Health District for a suspected toxin-mediated outbreak at a restaurant. A collaborative laboratory investigation was initiated that involved clinical and food microbiology disciplines.

Methods: Stool samples were initially screened for norovirus, followed by culture for the toxin-producing bacteria *C. perfringens* and *B. cereus* based on epidemiologic data. To stabilize potential bacterial contaminants in the leftover food while awaiting the norovirus results, all samples were frozen in a 1:1 dilution of buffered glycerin-salt solution. After only 1 of 4 stools tested positive for Norovirus GI RNA, culture for the enumeration and isolation of both *B. cereus* and *C. perfringens* was initiated.

Results: Among 30 restaurant patrons with diarrheal illness, 24 (80%) reported consuming tonkotsu ramen at a special "Ramen Night" at Restaurant X. Clinical testing of the stools revealed elevated levels of *Clostridium spp.*, but only low levels of *Bacillus spp.* were observed. *C. perfringens* isolated from four stool samples and the leftover tonkotsu ramen composite were compared by pulsed-field gel electrophoresis (PFGE) and found to have indistinguishable DNA fingerprint patterns. Environmental Health Specialists learned that the ramen had not been properly cooled after preparation and observed limited accessibility to a hand sink and improper hand washing technique.

Conclusion: Collaborative efforts within DCLS and between DCLS and the health department were essential for stopping this outbreak. The enumeration and isolation of *C. perfringens* isolates and the PFGE evidence from the food and stool samples supported the hypothesis that consumption of a common source food from the restaurant caused the outbreak. The quick response by epidemiologists and environmental investigators resulted in a voluntary cancellation of a second "Ramen Night" by the restaurant owner, potentially preventing additional illnesses.