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A cluster of Clostridium difficile infections caused by a binary toxin-producing new PCR ribotype 826

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Background: Through *Clostridium difficile* infection (CDI) surveillance and outbreak management we identified a cluster of eight CDI episodes in five patients within a 4-month period (1 December 2015 until 31 March 2016). The cluster occurred at a gastro-intestinal surgical ward of a tertiary care hospital in the Netherlands with a low endemic CDI incidence rate and was associated with a high rate of recurrent and severe disease. Therefore, this prompted additional investigations.

Material/methods: We collected additional patient data supplementing surveillance data, and performed PCR ribotyping, MLVA typing, toxinotyping and antimicrobial susceptibility testing on cluster isolates.

Results: The CDI incidence rate at the gastro-intestinal surgical ward was 3.3 per 10,000 patient days until December 2015 and increased up to 19.8 per 10,000 patient days in the period between December 2015 and March 2016. The index case was an 83-year old male patient who had recently undergone a pancreaticoduodenectomy and developed hospital-acquired CDI during a readmission. An additional four cases with hospital-acquired CDI were detected on the ward within the following 4 months (figure). Different infection control measures were implemented to stop transmission (figure). Recurrent and severe disease was noted in 2 of 5 patients. Faecal samples from all five patients tested positive for toxin A, toxin B and binary toxin genes. The Tcd*C*Δ*117* deletion specific for ribotype

027 was not identified, but a 39-bp deletion in *TcdC* was detected. Isolates from all five patients displayed the same ribotyping profile that was not recognized in the Dutch Reference Library or in any of the databases of consulted international reference laboratories. The new strain was assigned as ribotype 826 by the UK Ribotyping Reference laboratory (Leeds). PCR analysis of a clade 5 specific DNA marker showed that ribotype 826 is part of the phylogenetic clade 5, and therefore closely related to the "hypervirulent" ribotype 078. Isolates were demonstrated to be 100% identical by MLVA and were resistant to ciprofloxacin and moxifloxacin (both MICs>32mg/L).

Conclusions: This cluster of CDI cases was remarkable as it occurred in a setting with only rare *C. difficile* transmission, was associated with a high rate of recurrent and/or severe disease and was due to a newly identified ribotype 826. This ribotype resembles the "hypervirulent" ribtoype 078 and we therefore assume that it has increased virulence, which might explain the cluster. The recognition of this cluster indicates that new *C. difficile* ribotypes with increased virulence still emerge, at unexpected locations and without a clear source. Therefore, ongoing CDI surveillance remains essential, and other institutions should now be aware of ribotype 826.

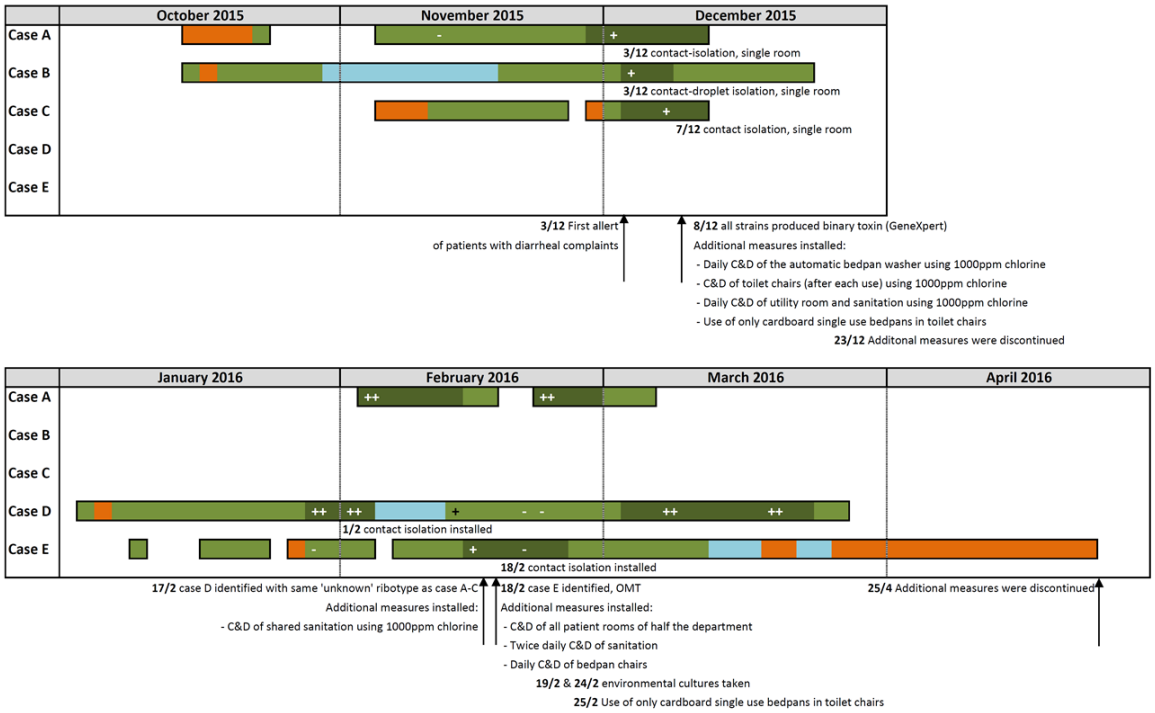

Figure. Epidemic curve of the 5 case patients infected with *C. difficile* caused by PCR ribotype 826. Green = outbreak non-ICU ward, Orange= other non-ICU ward, Blue= ICU, Dark green = diarrheal episode, White += positive culture for *C. difficile* and mild *C. difficile* infection, White ++= positive culture for *C. difficile* and severe *C. difficile* infection, Black + = Positive *C. difficile* culture without diarrhea, White – = Negative culture for *C. difficile* . Abbreviations: C&D= cleaning and disinfection, ICU= intensive care unit, OMT= outbreak management team.