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New pathogen discovery and diagnostic evaluation

Clostridium difficile infection point-prevalence study (Switzerland): a two-step protocol with GDH/PCR is equally sensitive as PCR alone for detection of toxigenic C. difficile

Sarah Tschudin-Sutter*[1], Adrian Egli[2], Ruth Schindler[3], Violeta Spaniol[4], Daniel Goldenberger[5], Reno Frei[6], Andreas F. Widmer[7]

[1]*., Basel, Switzerland*

[2]*University Hospital Basel, Clinical Microbiology, Laboratory Medicine, Basel, Switzerland*

[3]*University Hospital Basel, Abtlg. Infektiologie und Spitalhygiene, Base, Switzerland*

[4]*University Hospital Basel, Basel, Switzerland*

[5]*Department of Clinical Microbiology, University Hospital Basel, Basel, Switzerland*

[6]*University Hospital Basel, Department of Laboratory Medicine, Division of Clinical Microbiology, Basel, Switzerland*

[7]*University Hospital Basel, DIV. of Infectious Diseases & Hospital Epidemiology, Basel, Switzerland*

Background: *Clostridium difficile* has emerged as one of the most important healthcare-associated pathogens worldwide. Recent studies regarding *C. difficile* infection (CDI) in Europe report a mean of 7 cases of CDI per 10 000 patient-days and a high proportion of undiagnosed cases. The ideal diagnostic algorithm for rapid and cost-effective detection of toxigenic *C. difficile* still remains to be determined. As data regarding frequency of CDI and distribution of different strain types of *C. difficile* in Switzerland are lacking, we conducted a point-prevalence study in a variety of different Swiss hospitals and herein report on the performance of different diagnostic algorithms in place for detection of toxigenic *C. difficile*.

Material/methods: On two different days (one in winter and one in summer) in 2015, all unformed stool specimens submitted to the microbiology laboratory as standard of care from 84 Swiss hospitals were collected. In addition, stool samples tested positive for toxigenic *C. difficile* within the prior week were also collected from all participating institutions. The following diagnostic algorithms for detection of toxigenic *C. difficile* were compared: a two-stage algorithm consisting in an enzyme immunoassay (EIA) for detection of glutamate dehydrogenase (GDH) and toxins A and B (C.DIFF QUIK CHEK COMPLETE™, Techlab/ALERE, USA), and a two-stage algorithm consisting in EIA for detection of GDH followed by PCR for detection of toxins A and B (RealStar® *Clostridium difficile*, altona Diagnostics, Germany) in GDH positive samples. PCR for detection of toxin B as stand-alone test was considered as reference method.

Results: Overall, 354 stool samples were collected (252 from both point prevalence days and 102 from the two prior weeks). Toxigenic *C. difficile* was detected by PCR in 116 stools (32.8%). The gene encoding for toxin A was diagnosed in 95/116 samples (81.9%).

As compared to PCR, the two-stage algorithm consisting in EIA for detection of GDH followed by PCR for detection of toxin B had a sensitivity, specificity, positive and negative predictive value of 99.1%, 100.0%, 100.0%, and 99.6% respectively, while the two-stage algorithm consisting in EIA for detection of GDH and toxins A and B had a sensitivity, specificity, positive and negative predictive value of 78.4%, 99.2%, 97.8%, and 90.4%.

Conclusions: The two-stage diagnostic algorithm consisting in first performance of EIA for detection of GDH and subsequent PCR for detection of the toxin B gene in GHD-positive samples, showed very high performance characteristics. This diagnostic approach combines the advantages of the high sensitivity of PCR while reducing associated costs by first screening for GDH. In contrast, the two-stage algorithm using EIA for GDH and toxins A and B showed low sensitivity.