P0825
Poster Session III
New developments in molecular diagnosis of C. difficile

RAPID RANDOM ACCESS DIAGNOSIS OF CLOSTRIDIUM DIFFICILE IN THE LABORATORY USING DIASORIN® LIAISON ANTIGEN AND TOXIN TESTING AND ISOTHERMAL AMPLIFICATION METHODS IN A TWO-STEP ALGORITHM.

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Objectives: Since antigen tests for *Clostridium difficile* toxin lack the required sensitivity to be used as a stand-alone test for *C. difficile* disease (CDAD) the suggested diagnostic algorithm is a screening test often *C. difficile*-specific GDH, followed by a highly specific toxin test. The LIAISON® *C. difficile* GDH and C. difficile Toxins A&B (DiaSorin) tests are automated random-access tests providing a result in 90 minutes. The specificity and resulting positive predictive value of this combination is approximately 85 %. We undertook to compare the use of the LIAISON® tests with isothermal amplification and a possible combination in a two- or three-step algorithm to provide a quick result in a random access manner.

Methods: Stool samples sent for *C. difficile* testing were subjected to LIAISON® *C. difficile* GDH and Toxin A&B testing and in addition to two isothermal amplification methods using the Illumigene® and AmpliVue®. Discrepant results were clarified using real-time PCR for *C. difficile*-specific gene (16s rDNA) and *tcdA*/*tcdB* genes and toxigenic culture. The hands-on time for the tests was analysed in order to provide information as to the practicality of a possible algorithm.

Results: 251 stool specimens were tested by all methods. The overall prevalence of positive stools was 19.5 %. The sensitivity and specificity of the LIAISON® GDH test was 100 % i.e. there was complete accordance in the results between GDH testing and isothermal amplification. The sensitivity and specificity of the LIAISON® Toxin A&B test was 65.3 % and 100 % respectively. A total of 32 specimens were positive in all tests. 31 specimens were GDH-positive and LIAISON® Toxin A&B-negative, of which 16 (53.3 %) were positive and 14 (46.7 %) were negative by isothermal amplification. The results were confirmed by real-time PCR. The cytotoxic neutralisation test was equivalent in sensitivity to the toxin antigen detection.

Conclusion: The LIAISON® *C. difficile* GDH test is a sensitive screening method to identify negative stool specimens requiring no further testing. The LIAISON® Toxin A&B is, however, no better than the cytotoxicity neutralisation test and as other ELISA not sensitive enough to used alone test for CDAD. In our study to date, the LIAISON® Toxin A&B missed 14 of 49 (28.6 %) positive specimens. Isothermal amplification is a suitable method for processing a reasonably small number of specimens – up to approximately 10 specimens per run is manageable. For many laboratories this is not feasible and thus a faster, more efficient screening is necessary. The LIAISON® GDH test is quick and requires little hands-on time. Confirmation of the positive results is necessary and this can easily be done using an isothermal amplification method. The total time to positive result is approximately 2 hours and less than 1 hour for a negative result.