P0788

Paper Poster Session IV

Hot topics in Clostridium difficile diagnostics

Application of a novel nuclear magnetic resonance platform to detect *C. difficile*

K. Park[1], J. Pettersson[1], S. Hash[2], P. Yang[2], C. Petti[3], P. Ward[1], R. She[1]

[1]*University of Southern California, Los Angeles, USA*

[2]*Menon Biosensors- Inc., San Diego, USA*

[3]*HealthSpring Global- Inc., Bradenton, USA*

Objective: Accurate and prompt identification of pathogens from complex biological matrices at points of need is important for both clinical medicine and biosurveillance. Nuclear magnetic resonance (NMR) nanotechnology is a next generation diagnostic method that can detect nucleic acids or antigens of microorganisms with high sensitivity and specificity. We evaluated an early prototype of a patented NMR detection platform using molecular mirroring (M^2) technology, [Menon Biosensors, San Diego, CA] to detect *C. difficile* toxin from stool. In the absence of a nucleic acid gold standard test, our objective was to assess relative diagnostic concordance between the M^2 platform and an FDA cleared PCR based test for *C. difficile* toxin from stool.

Methods: Patient stool specimens submitted to the laboratory for *C. difficile* toxin testing were flagged by laboratory staff. After the clinical specimen was tested by a standard FDA cleared PCR assay and results reported, remaining specimen was either tested fresh if refrigerated < 5 days, or frozen at minus 70°C and then thawed immediately prior to testing. Keck Medical Center's Ethics Board approved the study. DNA was isolated from stool, and amplified by a combination of DNA and signal amplification methods using proprietary protocols. The M^2 assay utilized customized oligonucleotide probes conjugated with magnetic nanoparticles. Presence of *C. difficile* DNA was detected by quantitatively measuring the NMR spin-spin relaxation time T2, reflecting the distribution of nanoparticles. When no target is present, uniform distribution of nanoparticles occurs resulting in lower T2. When target DNA is present, structures of nanoparticles are formed resulting in higher T2. The cut-off value for positive results was a T2 difference of 150 milliseconds (ms). An internal control was included for each specimen. Positive and negative controls were included for each batch of specimens.

Results: Of 124 patient specimens tested, 116 were evaluated with 8 specimens being excluded for failed internal controls. For PCR-positive specimens, T2 signal measurements ranged from 4 to 1293 ms. For PCR-negative specimens, T2 signal measurements ranged from -6.5 to 586 ms. Concordance was observed for 79% of specimens (Table). After DNA extraction, approximate time for amplification and T2 signal detection was approximately 60-90 minutes for batch of 8 specimens.

FDA Cleared Test	Molecular Mirrroring Technology		
	Positive	Negative	Total
Positive	56	11	67
Negative	13	36	49
Total	69	47	116

Conclusions: The prototype M^2 platform demonstrates promising performance to detect *C. difficile* toxin from clinical stool specimens. Further testing of specimens of discordant results and for analytical performance is ongoing. The platform design offers a novel, low cost alternative to approved nucleic acid based tests, and when the automated, portable instrument is completed with additional assay optimization, the M^2 platform has potential for a wide variety of applications at point of need.