INTRODUCTION/BACKGROUND

Rapid and reliable detection of respiratory pathogens is a critical step in the diagnosis and treatment of respiratory infections. The FilmArray Respiratory Panel (RP) is a user-friendly, highly multiplexed in-vitro diagnostic test for the detection of dozens of agents in a single reaction, from nasopharyngeal aspirate (NPA) specimens in as few as four hours. One of the 17 total targets of the FilmArray RP test is Adenovirus. Adenoviruses are a genetically diverse family of viruses. Human adenoviruses have been classified into 46 species (A–46) and further distinguished into over 350 serotypes. Adenoviruses B, C, and D are associated with respiratory disease, while infection with other species (A, F, G, and Q) typically leads to conjunctivitis or gastroenteritis. The Adenovirus assay in the FilmArray RP will detect Adenovirus in respiratory samples, but not Adenovirus species and serotypes. The original FilmArray RP assay was labeled with a limitation on the detection of AdV2 (C2) and AdV3 (C3) serotypes and the sensitivity for detection of other respiratory adenoviruses was reported to be lower than some published or commercial molecular adenovirus assays. The FilmArray RP has recently been modified to improve detection of all respiratory adenoviruses from NPA specimens. The performance of this modified panel was validated with analytical and clinical studies. Analytical studies were performed to characterize the sensitivity (limit of detection), selectivity and specificity (cross-reactivity and performance in the presence of co-inhibiting organisms) for adenovirus in the modified panel. In addition, a clinical method comparison study was performed in which 222 archived clinical NPS specimens and 44 contrived specimens were tested with both the modified and unmodified FilmArray RP Performance was compared for detection of AdV and all analytes on the panel. These studies validate that detection of all respiratory adenoviruses, including C2 and C3, is improved with the modified FilmArray RP, while the performance of all other analytes was unchanged compared to the original version of the test.

METHODS

Analytical studies were performed by spiking quantified viruses and/or bacteria into a simulated NPS sample matrix (human nasal cells in Remel™ Nasal Transport medium) followed by testing with both the original and/or modified FilmArray RP test. The clinical comparison study was performed using 222 archived NPS specimens and 44 contrived specimens with both unmodified modified FilmArray RP versions. Adenovirus specimens were acquired from multiple source laboratories across the U.S. and Scotland and chosen based upon previous positive test results from various tests including culture, DFA, lab-developed tests, or original FilmArray RP to cover all analytes in the panel. Any specimen positive for Adenovirus by original or modified modified FilmArray RP was subject to co-directional sequencing (known gene target) to determine serotype. To supplement the archived specimens, control standards were prepared by spiking Chloroplastic DNA virus, Mumps virus, or Adenovirus serotypes C2 and C3 into archived NPS specimen. Operating parameters for the FilmArray RP testing were based on previous test results and continued spiking schemes.

RESULTS

Analytical Sensitivity (Limit of Detection): Adenovirus Detection Is Improved in Modified FilmArray RP As Compared to Original RP

Analytical Reactivity (Inclusivity): Modified FilmArray RP Detects More Adenovirus Serotypes at Lower Concentrations As Compared to Original FilmArray RP

Adenovirus & Other Analytes in the Presence of Potentially Competing FilmArray RP Viruses (Co-Infection)

Detection of Non-Adenovirus Analytes is Equivalent between the Original and Modified FilmArray RP at LoD Levels

Detection of Non-Adenovirus Analytes in Clinical NPS Samples is Equivalent in Original and Modified FilmArray RP

Detection of Non-Adenovirus Analytes is Equivalent between the Original and Modified FilmArray RP at LoD Levels

Detection of Non-Adenovirus Analytes in Clinical NPS Samples is Equivalent in Original and Modified FilmArray RP

SUMMARY

The modified FilmArray RP exhibited a 3-fold increase in analytical sensitivity (LoD) compared to the original panel and was shown to detect 17 different respiratory adenovirus serotypes within species B, C and D (including AdV2 and AdV3) at a concentration of 100 TCID₅₀/mL. No cross-reactivity with other respiratory pathogens was observed (data not shown) and the panel is able to accurately identify multiple pathogens from a single sample. In the clinical study, the modified FilmArray RP detected Adenovirus in 73% more archived specimens than the unmodified test (28/265 = 106%; 28/265 ± 12% relative detection in this specimen set). Directional sequencing was able to determine the serotype of Adenovirus in 24 specimens. Modified FilmArray RP demonstrated improved detection of C2, C5, C6, and C4 serotypes in archival NPS specimens.

ACKNOWLEDGEMENTS

BioFire Diagnostics, Inc., Salt Lake City, UT

Presented at the AMP 2013 Annual Meeting, November 2013

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