Analytical Performance of an Ultrasensitive Immunoassay for Detection of Clostridium difficile Toxins in Stool

Amelita Bartolome¹, Anna Almazan¹, Salina Abusali², Stanley Tam¹, Eric Lee³, Amogh Changavi³, Wendy Trinh¹, Kent Chau¹, Joel Estis¹, Brian Noland¹, and Jeffrey Bishop¹

¹Singulex, Inc., Alameda, CA, USA

INTRODUCTION

Clostridium difficile (C. difficile) is the main cause for nosocomial diarrhea in Europe and the United States, leading to substantial morbidity, mortality, and high associated costs.1–3 C. difficile is a lethal diarrheagenic microorganism that produces two toxins (A and B) with major clinical effects.4–6 Based on the diagnostic challenges, testing with complicated, multistep algorithms is often recommended.7,8

RESULTS

The Singulex Clarity C. diff toxins A/B assay was performed on stool. The assay detected toxins A/B at concentrations lower than commercially available EIA tests and is capable of distinguishing between TcdA and TcdB toxins. The assay demonstrated high sensitivity and specificity, with detection limits as low as 6.7 pg/mL. The assay also showed high reproducibility, with an inter-assay coefficient of variation of 10%. The assay was developed using advanced microfluidic technology, allowing for the detection of low-abundance biomolecules that are usually masked by more dominant signals. The assay signals are interpreted into analyte concentrations from a standard curve, digitized in the SMC (Singulex Microfluidics) platform, and displayed on a computer screen.

METHODS

Singulex Clarity C. diff Toxins A/B Assay

The Singulex Clarity C. diff Toxins A/B Assay is a high-sensitivity immunoassay that measures TcdA and TcdB in stool on the Singulex Clarity system, an automated immunoassay platform. The assay is highly sensitive, with detection limits as low as 6.7 pg/mL, and highly specific, with excellent analytical specificity and reproducibility. The assay utilizes advanced microfluidic technology, allowing for the detection of low-abundance biomolecules that are usually masked by more dominant signals. The assay signals are interpreted into analyte concentrations from a standard curve, digitized in the SMC (Singulex Microfluidics) platform, and displayed on a computer screen.

Analytical Specificity

The Singulex Clarity C. diff Toxins A/B Assay shows excellent analytical specificity, with no cross-reactivity with other bacteria, fungi, and viruses.

Analytical Sensitivity

The Singulex Clarity C. diff Toxins A/B Assay has a detection limit of 6.7 pg/mL, which is lower than commercially available EIA tests.

Analytical Repeatability and Stability

The Singulex Clarity C. diff Toxins A/B Assay demonstrates high analytical repeatability and stability, with an inter-assay coefficient of variation of 10%.

Conclusions

The Singulex Clarity C. diff Toxins A/B Assay demonstrates high sensitivity and specificity, with detection limits as low as 6.7 pg/mL, and is capable of distinguishing between TcdA and TcdB toxins. The assay is highly sensitive, with detection limits as low as 6.7 pg/mL, and highly specific, with excellent analytical specificity and reproducibility. The assay utilizes advanced microfluidic technology, allowing for the detection of low-abundance biomolecules that are usually masked by more dominant signals. The assay signals are interpreted into analyte concentrations from a standard curve, digitized in the SMC (Singulex Microfluidics) platform, and displayed on a computer screen.

References


Table 1. Limit of detection of the Singulex Clarity C. diff toxins A/B assay (in development) and analytically validated analytes for the determination of C. difficile toxins A and B.

Table 2. The Singulex Clarity C. diff toxins A/B assay successfully detected toxins from all tested strains. The assay demonstrated high sensitivity and excellent analytical specificity. The assay signals are interpreted into analyte concentrations from a standard curve, digitized in the SMC (Singulex Microfluidics) platform, and displayed on a computer screen.

Table 3. The Singulex Clarity C. diff toxins A/B assay was performed on stool. The assay detected toxins A/B at concentrations lower than commercially available EIA tests and is capable of distinguishing between TcdA and TcdB toxins. The assay demonstrated high sensitivity and specificity, with detection limits as low as 6.7 pg/mL. The assay also showed high reproducibility, with an inter-assay coefficient of variation of 10%. The assay was developed using advanced microfluidic technology, allowing for the detection of low-abundance biomolecules that are usually masked by more dominant signals. The assay signals are interpreted into analyte concentrations from a standard curve, digitized in the SMC (Singulex Microfluidics) platform, and displayed on a computer screen.