MDx-Chex® Controls Ensure Accuracy and Reliability for Molecular-Based Testing

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Introduction

In recent years, the use of automated DNA-based sepsis tests to complement gold standard culture-based methods have increased in frequency. While automation simplifies molecular diagnostic workflows, the integration of steps into a proverbial "black box" creates the potential for variability in instrument and test performance. To ensure expected performance, effective quality control measures are paramount. However, many commercially available controls are either solely comprised of synthetic materials that have been diluted in buffers or may have intact organisms that are not included in a patient-like matrix. Thus, these formulations fail to comprehensively measure the assay, as they are unable to detect all panel targets or cannot verify the system's ability to process and remove sample matrix effects from inhibitors in patient blood and culture media. Furthermore, internal controls do not effectively evaluate matrix inhibition when the quality control matrix doesn't reflect a patient sample.

The consequences of these gaps in coverage were highlighted in a recent medical device correction for the BIOFIRE® Blood Culture Identification 2 (BCID2) Panel that identified misdetection of *C. tropicalis* by a commercially available quality control kit that employs synthetic DNA targets. This example underscores the need to shift from "traditional" quality control designs to use of controls mirroring patient samples in composition and test experience. Patient-like, full process controls are essential to adequately evaluate4 system and test performance, including manufacturer changes to the assay.

Here, we present MDx-Chex® for BCID2, MDx-Chex for BC-GP and MDx-Chex for BC-GN, quality control solutions for routine molecular diagnostic sepsis testing. These FDA-cleared, class II assayed, patient-like full process controls are intended for use with the BIOFIRE BCID2 and Luminex VERIGENE® BC-GP and BC-GN sepsis panels, respectively. MDx-Chex controls contain intact, inactivated microorganisms constituted in a matrix representative of a blood culture sample.

Methods and Analysis

Sample Testing

Aliquots of each control sample were tested on the BIOFIRE FilmArray® Torch and 2.0 Systems using the BIOFIRE BCID2 Panel (#RFIT-ASY-0147), 200 µL, or Luminex VERIGENE System using the BC-GP (#20-005-018), 350 µL, or BC-GN (#20-005-021), 700 µL, Panels per the Instructions For Use.

Performance Evaluation

Precision of the controls was evaluated by testing three lots of MDx-Chex for BCID2, MDx-Chex for BC-GP or MDx-Chex for BC-GN using the respective panels mentioned above. Reproducibility (multi-site precision) of the controls was assessed internally and externally at three different clinical sites.

Results

While integration of rapid molecular sepsis tests into conventional testing workflows allows for faster turnaround times, the compilation of several complex steps into one process renders many current quality control measures insufficient. To address the gap in coverage left by "traditional" quality controls (i.e., those that contain synthetic gene fragments or off-target microorganisms), we developed MDx-Chex controls. MDx-Chex for BCID2, MDx-Chex for BC-GP and MDx-Chex for BC-GN are full-process, patient-like controls that contain intact, inactivated and test-specific microorganisms suspended in a matrix of blood culture media components (Figure 1 and Table 1).

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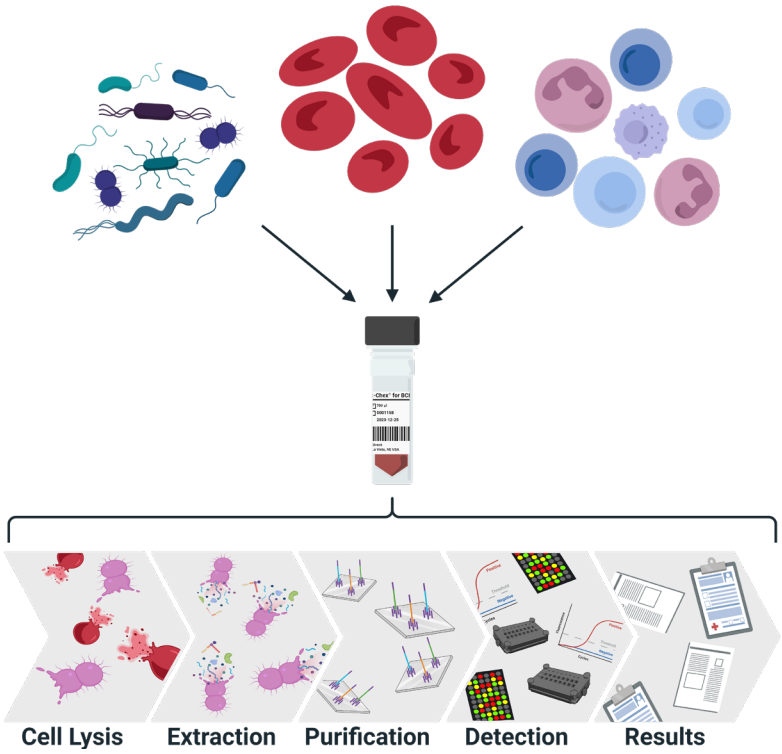

Figure 1: Composition of MDx-Chex® controls. Created with BioRender.com.

Table 1. Comparison of MDx-Chex controls to commercially available kits.

	Barcode Tracking	Intact Organisms	Human Blood Cells	Removal of Inhibitors Hemoglobin Human gDNA Culture media	Amplification/ Detection
MDx-Chex Controls	✓	✓	✓	✓	✓
Competitor A	X	X	X	X	✓
Competitor B	X	✓	X	X	✓

To internally evaluate control precision, we tested three independent lots of MDx-Chex for BCID2, MDx-Chex for BC-GP or MDx-Chex for BC-GN on the Filmarray® Torch and 2.0 Systems using the BIOFIRE® BCID2 Panel (for MDx-Chex for BCID2) or the Luminex VERIGENE® System using the BC-GP or BC-GN Panels for sepsis (for MDx-Chex for BC-GP and MDx-Chex for BC-GN, respectively). Overall precision of MDx-Chex for BCID2 controls was ≥ 95% positive agreement and 100% negative agreement (Table 2). Similar trends were observed with MDx-Chex for BC-GP and MDx-Chex for BC-GN, where the positive and negative agreements were 100% for MDx-Chex for BC-GP and 98% and 100% for MDx-Chex for BC-GN (Tables 3 and 4). Taken together, these data support that MDx-Chex controls perform appropriately and similarly across separately manufactured lots.

Table 2. Lot-to-lot precision of MDx-Chex® for BCID2.

Category	#Observed/#Expected Results	Percent Agreement	95% Confidence Interval
Positive: Control 1-GN and Control 2-GPY, combined	114/120	95%	94 – 100%
Negative: Control 1-GN and Control 2-GPY, combined	120/120	100%	97 – 100%

Table 3. Lot-to-lot precision of MDx-Chex for BC-GP.

Category	#Observed/#Expected Results	Percent Agreement	95% Confidence Interval
Positive	60/60	100%	94 – 100%
Negative	60/60	100%	94 – 100%

Table 4. Lot-to-lot precision of MDx-Chex for BC-GN.

Category	#Observed/#Expected Results	Percent Agreement	95% Confidence Interval
Positive	59/60	98%	91 – 100%
Negative	60/60	100%	94 – 100%

To test reproducibility, MDx-Chex controls were sent to three external testing sites, where they were run on different days with different operators. Positive and negative agreement for MDx-Chex for BCID2 was ≥ 96% and ≥ 99%, respectively (Table 5). MDx-Chex for BC-GP and MDx-Chex for BC-GN data demonstrate 100% positive and negative agreement and 99% positive agreement and 100% negative agreement, respectively (Tables 6 and 7). Overall, these results support the reproducibility of MDx-Chex controls across sites, days, operators and systems.

Table 5. Reproducibility of MDx-Chex for BCID2.

Category	Site #1 #Observed/ #Expected Results	Site #2 #Observed/ #Expected Results	Site #3 #Observed/ #Expected Results	Site #4 #Observed/ #Expected Results	Percent Agreement (all sites)	95% Confidence Interval
Positive: Control 1-GN and Control 2-GPY, combined	59/60	57/60	58/60	56/60	95.8% (230/240)	92 – 99%
Negative: Control 1-GN and Control 2-GPY, combined	60/60	60/60	60/60	59/60	99.6% (239/240)	98 – 100%

Table 6. Reproducibility of MDx-Chex for BC-GP.

Category	Site #1 #Observed/ #Expected Results	Site #2 #Observed/ #Expected Results	Site #3 #Observed/ #Expected Results	Percent Agreement (all sites)	95% Confidence Interval
Positive	30/30	30/30	30/30	100% (90/90)	96 – 100%
Negative	30/30	30/30	30/30	100% (90/90)	96 – 100%

Table 7. Reproducibility of MDx-Chex® for BC-GN.

Category	Site #1 #Observed/ #Expected Results	Site #2 #Observed/ #Expected Results	Site #3 #Observed/ #Expected Results	Percent Agreement (all sites)	95% Confidence Interval
Positive	29/30	30/30	30/30	99% (89/90)	94 – 100%
Negative	30/30	30/30	30/30	100% (90/90)	96 – 100%

Conclusion

Collectively, MDx-Chex controls ensure reliable and accurate BIOFIRE BCID2 and VERIGENE BC-GP and BC-GN Panel-based sepsis testing. Use of these controls enables physicians and healthcare professionals to maintain assay quality while improving compliance with accrediting standards and guidelines.