Food and Drug Administration
10903 New Hampshire Avenue
Silver Spring, MD 20993-0002
www.fda.gov

510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
INSTRUMENT ONLY

I Background Information:

A 510(k) Number

K191964

B Applicant

Becton, Dickinson and Company

C Proprietary and Established Names

BD Kiestra IdentifA

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
QQV	Class II	21 CFR 866.3378 - Clinical Mass Spectrometry Microorganism Identification And Differentiation System	MI - Microbiology
QBN	Class II	21 CFR 866.3378 - Clinical mass spectrometry microorganism identification and differentiation system	MI - Microbiology

II Submission/Device Overview:

A Purpose for Submission:

To obtain a substantial equivalence determination for the BD Kiestra IdentifA instrument.

B Type of Test:

Qualitative *in vitro* diagnostic device for identification and differentiation of microorganisms cultured from human specimens by automation of target preparation for mass spectrometry analysis.

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

The BD Kiestra IdentifA module is an automated in vitro diagnostic specimen preparation system for use with the BD Kiestra Laboratory Automation Solution to prepare MALDI targets for the Bruker MALDI Biotyper CA System for the qualitative identification and differentiation of microorganisms using matrix-assisted laser desorption/ionization-time of flight mass spectrometry (MALDI-TOF MS) analysis of colonies grown on plated culture media from human specimens.

The BD Kiestra IdentifA is indicated for use in the clinical laboratory with the ReadA Compact and Bruker MALDI Biotyper CA System to aid in the diagnosis of bacterial and fungal infections.

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only.

IVD - For In Vitro Diagnostic Use Only.

Special Instruments for Use:

1. BD Kiestra ReadA Compact, v 1.1
   The BD Kiestra ReadA Compact module is required for use in conjunction with the BD Kiestra IdentifA to obtain the digital image the IdentifA uses for colony selection and target preparation. The ReadA Compact is an essential component to the performance of the IdentifA and, together with the IdentifA and the Bruker MALDI Biotyper CA, comprises a test system for the qualitative identification and differentiation of microorganisms. Any change to the ReadA Compact will be assessed and appropriate verification and validation will be performed as applicable to assure proper function of the BD Kiestra IdentifA.

2. Bruker MALDI Biotyper for Clinical Applications (MBT-CA)
   When using the BD Kiestra IdentifA, refer to the most recent version of the Bruker MALDI Biotyper CA system labeling.

Performance of the BD Kiestra IdentifA was evaluated with the following culture media that are validated as compatible with the Bruker MALDI Biotyper CA:

Trypticase Soy Agar with 5% Sheep Blood
MacConkey Agar

Columbia CNA Agar with 5% Sheep Blood
Chocolate Agar
Sabouraud-Dextrose Agar
CDC Anaerobe Agar 5% Sheep Blood
Campylobacter Agar with 5 Antimicrobics and 10% Sheep Blood

Performance of the BD Kiestra IdentifA has not been established with the following culture media that are validated as compatible on the Bruker MALDI Biotyper CA:

Bacteroides Bile Esculin Agar with Amikacin
Bordet Gengou Agar with 15% Sheep Blood
Clostridium Difficile Agar with 7% Sheep Blood
Buffered Charcoal Yeast Extract Agar
Buffered Charcoal Yeast Extract Agar with Polymixin, Anisomycin and Vancomycin
Brucella Agar with 5% Horse Blood
CDC Anaerobe 5% Sheep Blood Agar with Phenylethyl Alcohol
CDC Anaerobe Laked Sheep Blood Agar with Kanamycin and Vancomycin
Modified Thayer-Martin Agar

IV Device/System Characteristics:

A Device Description:

The BD Kiestra IdentifA automates preparation of MALDI targets for the Bruker MALDI Biotyper CA System that is used in clinical laboratories for identification and differentiation of organisms grown on plated media by Matrix-Assisted Laser Desorption/Ionization Time-of-Flight Mass Spectrometry (MALDI-TOF MS). The system comprises the BD Kiestra IdentifA module (including the associated software and onboard nephelometers and pipetting system), formic acid and automation-compatible transfer vials (for HCCA matrix and Bacterial Test Standard (BTS), which are obtained directly from Bruker and manually transferred to the vials for use on the instrument), consumables (pipette tips and cuvette arrays for preparation of organism suspensions and fluid movement), and nephelometer calibration standards (McFarland standard vials for measuring turbidity of microbial suspensions).

When identification of an organism growing on a culture medium plate is required, a technologist designates specific colonies for picking by the BD Kiestra IdentifA module using a digital image of the plate obtained using the BD Kiestra ReadA Compact module. The BD Kiestra IdentifA automatically suspends the designated colonies in deionized water and uses an onboard nephelometer to determine the resulting turbidity. The organism concentration is adjusted automatically by picking additional designated colonies or by appropriate dilution of the suspension to achieve a turbidity within a targeted range of McFarland values. Based on the final organism concentration, the BD Kiestra IdentifA pipets one or more aliquots of the microbial suspension onto a MALDI target (either reusable 48-spot or disposable 96-spot targets) and dries the spots at elevated temperature.

The BD Kiestra IdentifA performs the extended Direct Transfer (eDT) Sample Preparation Procedure from Bruker whereby the instrument overlays the dried sample spot on the MALDI target with formic acid and matrix. The BD Kiestra IdentifA also spots the BTS used for quality control of MALDI-TOF MS organism identification. Once spots are dry, the technologist manually removes the target from the BD Kiestra IdentifA and loads it into the Bruker MALDI Biotyper CA System for analysis. Information regarding the location of each sample and BTS on the targets and the associated MALDI-TOF MS results are transmitted between the BD Kiestra IdentifA and Bruker MALDI Biotyper CA via the Synapsys Informatics, the main software interface, and the BD Kiestra BeA, the data interface hub module that communicates with all the other modules including the BD Kiestra IdentifA. In addition to preparing the MALDI target, if requested, the BD Kiestra IdentifA will also dilute the organism suspension to a standardized turbidity of 0.5 McFarland.

Modules of the BD Kiestra System each have their own operating software that communicates via the central BeA data interface hub module with the Synapsys user interface which in turn sends and receives information to/from the Laboratory Information System (LIS) (**Figure 1**).

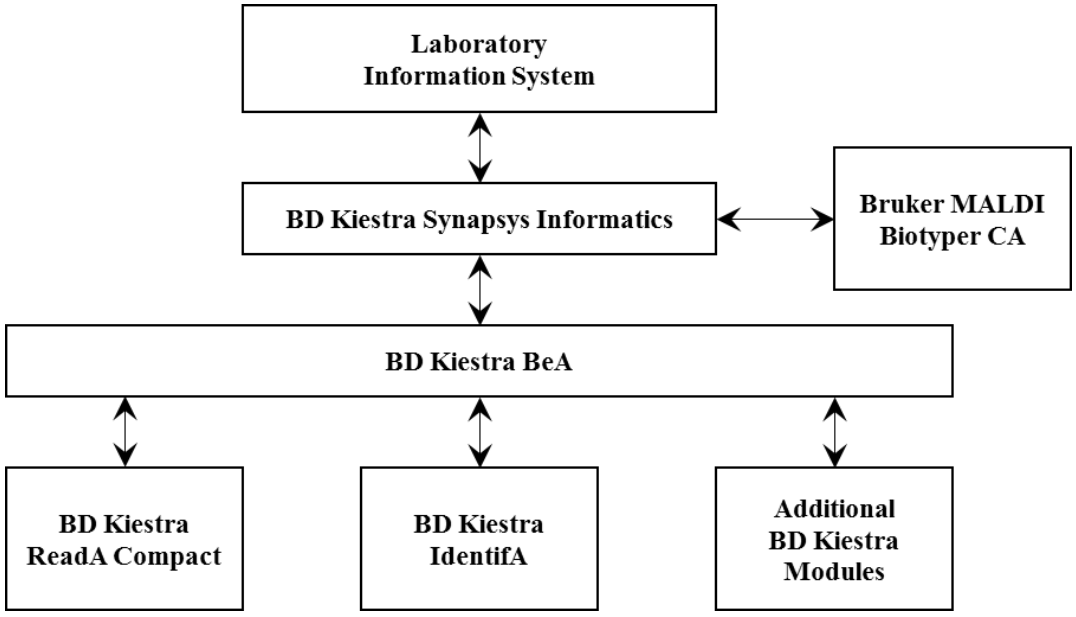

Figure 1. Overview of software interfaces for the BD Kiestra Laboratory Automation System.

The BD Kiestra ReadA Compact module is required for use in conjunction with the BD Kiestra IdentifA module for plate incubation and image capture. Additional software modules (the BD Synapsys Informatics and BD Kiestra BeA) are also required for the function of the BD Kiestra IdentifA, and these modules reside on the BD Kiestra Laboratory Automation Solution. The digital image is used by the BD Kiestra IdentifA for image analysis and colony designation by the operator.

B Instrument Description Information:

1. Instrument Name:

   BD Kiestra IdentifA

2. Specimen Sampling and Handling:

The BD Kiestra ReadA Compact has default parameters for image capture as well as user-defined settings. Images taken under the default parameters are always captured in addition to any additional images taken under conditions of illumination and background that are specified by the user. Each of the images is presented to the user via the Synapsis interface.

Culture plates for processing by the BD Kiestra IdentifA are loaded manually or via an automated conveyor, depending on the instrument configuration. Plates are identified by the BD Kiestra IdentifA by scanning a linear barcode on the side of the bottom half of each plate. The BD Kiestra IdentifA then queries the Synapsys software via the BeA data interface to obtain patient information and details of the testing to be performed. Colonies are identified by a technologist from a digital image of each culture plate obtained from the ReadA Compact and designated for picking via the BD Kiestra IdentifA. Specific colonies' locations are identified by polar coordinates that are calculated relative to the position of the barcode label. The BD Kiestra IdentifA automatically orientates each plate based on the location of the barcode, picks the designated colonies and prepares a homogenous suspension in deionized water. Mucoid colonies are detected automatically and processed using a modified procedure to reduce the potential for contamination and ensure homogeneity of the suspension. The turbidity of the final suspension is measured and adjusted as necessary to within a target range of McFarland values. One or more aliquots of the suspension is used to prepare a MALDI target, which is dried at elevated temperature and manually transferred to the Bruker MALDI Biotyper CA for analysis. The location into which each microbial suspension is spotted on the MALDI target together with other relevant tracking parameters for the microbial suspension prepared by the BD Kiestra IdentifA are posted to the Synapsis interface via the BeA module.

3. Specimen Identification:

MALDI targets are manually transferred to the Bruker MALDI Biotyper CA, which performs microbial differentiation and species identification of microorganisms via matrix-assisted laser desorption/ionization-time of flight mass spectrometry (MALDI-TOF MS) analysis. Two-way communication between the BD Kiestra IdentifA and Bruker MALDI Biotyper CA systems is mediated by the Synapsys interface to enable transfer of the MALDI target map to the MALDI Biotyper CA and subsequent receipt of microbial identification results that are displayed to the user.

4. Calibration:

Camera Calibration
Calibration of the BD Kiestra ReadA Compact camera must be performed each time the camera is cleaned or when the position of the camera is changed. Calibration and reference plates containing a plastic disk with red/green/blue print and a barcode label are provided with the system for use with the built-in software calibration wizard, OPTIS, to verify the accuracy of barcode reading, image dimensions and quality. Imaging area detection first

selects the appropriate area of the image that contains the plate. Black calibration determines the average grey value and signal-to-noise ratio (SNR) in black images (all lights off). Plate holder detection uses various inputs to determine the position of the plate holder. Lights calibration adjusts lighting power to ensure homogenous illumination. Linearity calibration ensures that increased exposure of an object results increased signal intensity in each channel of the Bayer filter. SNR calibration determines the SNR as a function of input grey value. White balance and light references calibration normalize RGB channel intensities so that a white object appears white when imaged by the camera system. Pixel calibration accounts for chromatic aberrations and geometrical distortions. When acquiring an image, multiple images of the same scene are evaluated to reduce potential noise and runtime checks are performed to ensure consistency with calibration. Additionally, Petri dishes are imaged with top, side, and bottom illumination utilizing black or white contrasting backgrounds to maximize information and allow reading of plates by microbiologists. After successful image capture, images are normalized and adjusted according to calibrated metrics to improve image quality and reduce variability across each instrument.

Nephelometer Calibration
Calibration of the BD Kiestra IdentifA nephelometers must be performed once a month using the provided calibration cuvette array standards (0.2, 0.5, 1.0 and 3.0 McFarland). Additional verification of the nephelometers must be performed daily using the 0.5 McFarland cuvette array. MALDI target preparation will not resume until daily verification passes.

5. Quality Control:

Use of the Bruker US IVD Bacterial Test Standard (BTS) is required to obtain valid identification results with the Bruker MALDI Biotyper CA System. The BD Kiestra IdentifA uses a modified workflow for preparation and spotting of the BTS solution whereby the volume of solvent is increased to improve the precision of automated pipetting. The modified BTS workflow was verified by conducting a study to validate equivalency of using a double volume of BTS solution (2 µL) compared to the standard volume (1 µL). BTS was prepared per the manufacturer's instructions for use (diluted in 50 µL solvent), and 1 µL was dispensed onto 48- and 96-spot MALDI targets for 102 spots. BTS was then prepared using twice the amount (100 µL) of solvent, and 2 µL was dispensed onto MALDI targets for a total of 430 spots. The modified BTS workflow for BTS spots prepared using the BD Kiestra IdentifA performs equivalently to the standard workflow with 423 (98.4%) spots yielding Log(score) values ≥ 2.00 (high-confidence identification) and no statistical difference in performance (paired chi square P value = 0.368) compared to the standard workflow. The results are acceptable.

V Substantial Equivalence Information:

A Predicate Device Name(s):
MALDI Biotyper CA System

B Predicate 510(k) Number(s):
DEN170081

C Comparison with Predicate(s):

Device & Predicate Device(s):	Device: K191964	Predicate: DEN170081
Device Trade Name	**BD Kiestra IdentifA**	**Bruker MALDI Biotyper CA System**
Intended Use/Indications For Use	The BD Kiestra IdentifA module is an automated *in vitro* diagnostic specimen preparation system for use with the BD Kiestra Laboratory Automation Solution to prepare MALDI targets for the Bruker MALDI Biotyper CA System for the qualitative identification and differentiation of microorganisms using matrix-assisted laser desorption/ionization - time of flight mass spectrometry (MALDI-TOF MS) analysis of colonies grown on plated culture media from human specimens. The BD Kiestra IdentifA is indicated for use in the clinical laboratory with the BD Kiestra ReadA Compact and Bruker MALDI Biotyper CA System to aid in the diagnosis of bacterial and fungal infections.	The MALDI Biotyper CA System is a mass spectrometer system using matrix-assisted laser desorption/ ionization - time of flight (MALDI-TOF) for the identification and differentiation of microorganisms cultured from human specimens. The MALDI Biotyper CA System is a qualitative *in vitro* diagnostic device indicated for use in conjunction with other clinical and laboratory findings to aid in the diagnosis of bacterial and fungal infections. (list of validated organisms omitted for brevity; refer to DEN170081)
General Device Characteristic Similarities		
Sample Type	Isolated colonies on plated culture media	Same
MALDI Target Preparation	Extended Direct Transfer (eDT) Sample Preparation Procedure	Same

Device & Predicate Device(s):	Device: K191964	Predicate: DEN170081
Amount of Organism on Target	Meets Bruker's Limit of Detection	Same
Quality Controls	US IVD Bacterial Test Standard (BTS)	Same
Matrix	US IVD HCCA Matrix	Same
Targets	MBT Biotarget 96 US IVD (96-spot disposable) target US IVD 48 Spot (48-spot reusable) target	Same
Target Loading on MALDI-TOF MS	Manual	Same
General Device Characteristic Differences		
Colony/Plate Visualization	Digital image from the BD Read A Compact module	Direct
Organism Preparation	Suspension of colonies prepared in deionized water by pipettor	Direct smear of colony onto target by stick
Number of Colonies Sampled	Up to 9 per microbial suspension or target spot	One per target per spot
Alternative Methods of MALDI Target Preparation	None	Direct Transfer (DT) and Extraction (Ext) Procedure
Sample and Reagent Application	Automated	Manual
Drying of Target Plate	35 ± 2°C	Ambient temperature
Results Achieved	Prepared MALDI target	Organism identification
Results Reported	None	Organism identification
Technology	Robotic x-y-z platform using pipetting system and onboard nephelometry	Mass spectrometer using matrix-assisted laser desorption/ionization-time of flight (MALDI-TOF)

VI Standards/Guidance Documents Referenced:

IEC 61010-1. Safety requirements for electrical equipment for measurement, control and laboratory use - Part 1: General Requirements.

IEC 61010-2-010. Safety requirements for electrical equipment for measurement, control and laboratory use - Part 2-010: Particular requirements for laboratory equipment for the heating of materials.

IEC 61010-2-081. Safety requirements for electrical equipment for measurement, control and laboratory use - Part 2-081: Particular requirements for automatic and semi-automatic laboratory equipment for analysis and other purposes.

IEC 61010-2-101. Safety requirements for electrical equipment for measurement, control and laboratory use - Part 2-101: Particular requirements for *in vitro* diagnostic (IVD) medical equipment.

IEC 61326-1. Electrical equipment for measurement, control and laboratory use - EMC requirements - Part 1: General requirements.

IEC 61326-2-6: Electrical equipment for measurement, control and laboratory use - EMC requirements - Part 2-6: Particular requirements - *In vitro* diagnostic (IVD) medical equipment.

VII Performance Characteristics (if/when applicable):

The isolates used in the analytical studies were obtained from the American Type Culture Collection (ATCC) as well as non-ATCC sources, i.e. clinical isolates from multiple sources, which were archived and identified via MALDI-TOF MS identification. These were designated as the expected identities of the isolates used to compare performance of the BD Kiestra IdentifA using the interpretive criteria for species identification described in the labeling for the Bruker MALDI Biotyper CA summarized in **Table 1**.

Table 1. Interpretive criteria for species identification using the Bruker MALDI Biotyper CA

Confidence Level	Log(Score) Value
High	≥ 2.00
Low	1.70 - 1.99 [1]
No identification	< 1.70 [1]

[1] Additional testing required to determine or confirm organism identity

A Analytical Performance:

1. Precision/Reproducibility:

Nephelometer Accuracy & Reproducibility

Studies were conducted to assess the accuracy and reproducibility of the BD Kiestra IdentifA on-board nephelometers. *Escherichia coli* was plated and incubated for 18 to 24 hours, and suspensions of the growing colonies were prepared manually to ~0.2, 0.5, 1 and 2 McFarland. BD Kiestra IdentifA nephelometer readings were compared to viable colony counts to evaluate accuracy and reproducibility. Testing was performed on three BD Kiestra IdentifA instruments using both the "pick" and "dilution" nephelometers (n = 8 per nephelometer). The Pick Neph nephelometer is utilized for determining the number of layers to spot on the MALDI target; the Dilution Neph nephelometer is utilized when a 0.5 McFarland is requested. On each instrument, the nephelometers were calibrated with a

different lot of turbidity standards. The results of the study are summarized in **Table 2** and show acceptable accuracy and reproducibility within and between nephelometers and BD Kiestra IdentifA instruments based on comparisons between target and measured McFarland values and the measured colony counts. The results of the study demonstrated a linear correlation between the mean measured McFarland value and the mean number of bacterial colony-forming units in suspension (mean CFU/mL) ($R^2 = 0.994$) considering that a 0.5 McFarland suspension of *E. coli* has a nominal value of 1-2 × 10^8 CFU/mL. [1]

Table 2. Reproducibility of nephelometer readings within and between BD Kiestra IdentifA instruments.

Target McFarland	BD Kiestra IdentifA	Nephelometer	Measured McFarland Value					
			Mean	% CV	Mean	% CV	Mean	% CV
0.2	**1**	**A**	0.19	17.3	0.20	13.0	0.21	10.4
		B	0.22	4.2				
	2	**A**	0.20	14.6	0.20	11.0		
		B	0.20	6.9				
	3	**A**	0.22	7.0	0.22	5.6		
		B	0.22	4.2				
0.5	**1**	**A**	0.50	7.0	0.51	6.4	0.51	7.4
		B	0.52	5.6				
	2	**A**	0.50	13.3	0.51	9.4		
		B	0.51	3.6				
	3	**A**	0.52	7.2	0.52	6.2		
		B	0.52	5.5				
1.0	**1**	**A**	1.02	4.6	1.00	4.4	1.00	4.7
		B	0.98	3.0				
	2	**A**	1.00	3.1	0.98	3.6		
		B	0.96	3.0				
	3	**A**	1.03	4.1	1.02	5.5		
		B	1.00	6.6				
2.0	**1**	**A**	2.08	5.2	2.07	4.9	2.07	4.4
		B	2.06	4.9				
	2	**A**	2.00	4.8	2.03	4.0		
		B	2.06	2.6				
	3	**A**	2.13	3.7	2.11	3.6		
		B	2.09	3.5				

% CV: Percent coefficient of variation

Reproducibility of McFarland values obtained with the BD Kiestra IdentifA was also evaluated by reading a stable calibration standard (40 Nephelometric Turbidity Units [NTU]) repeatedly with the "pick" and "dilution" nephelometers on each of three instruments over a period of three days (8 reads × 2 nephelometers × 3 instruments × 3 days = 144 reads). On

[1] CLSI. *Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically.* 11th ed. CLSI standard M07. Wayne, PA. Clinical and Laboratory Standards Institute; 2018.

each day, the nephelometers were calibrated with a different lot of turbidity standards. The percent coefficient of variation across all reads and instruments was 2.6%. These results are acceptable.

Confirmation of the accuracy of BD Kiestra IdentifA nephelometer readings was performed with seven additional species of bacteria and yeast. Organisms were grown on plated media for 18 to 24 hours, and suspensions were prepared to ~0.5 McFarland. The goal was to verify that an ~0.5 McFarland suspension produced the expected colony count of approximately 1-2 × 10^8 CFU/mL for bacteria and 1-6 × 10^6 CFU/mL for yeast (**Table 3**). [1, 2] The results of the study are acceptable.

Table 3. Correlation of McFarland values determined by BD Kiestra IdentifA nephelometers and viable colony counts

Species	Measured McFarland Value		Mean CFU/mL
	Mean	% CV	
Acinetobacter baumannii	0.56	4.48	1.90×10^8
Enterococcus faecalis	0.60	4.17	1.41×10^8
Pseudomonas aeruginosa	0.70	4.45	1.20×10^8
Staphylococcus epidermidis	0.53	11.5	7.40×10^7
Streptococcus pneumoniae	0.61	4.19	2.31×10^7
Streptococcus pyogenes	0.56	5.68	1.63×10^8
Candida albicans	0.49	5.01	6.66×10^6

Reproducibility of Identification

Reproducibility of MALDI-TOF MS-based microbial identification when performed using the BD Kiestra IdentifA for sample preparation was evaluated in two separate studies described below.

Study 1:

In the first study, a panel of 10 strains of bacteria was tested in triplicate using each of three BD Kiestra IdentifA instruments over a period of three days and a single lot of Bruker MALDI Biotyper CA reagents (3 replicates × 3 instruments × 3 days = 27 data points per strain). The study was performed with the reusable 48-spot polished steel MALDI target. A list of the species and strains used in the study is provided in **Table 4**. MALDI identifications were compared to the expected identity for each isolate, i.e. the known species identifications for the reference isolates. For nine of the strains, all 27 replicates (100%) gave the expected identification results with Log (Scores) ≥ 2.00. For one strain of *E. coli*, 1/27 replicates gave an incorrect identification that was due to a user error in setting up the test, i.e. the technologist used the wrong culture plate to set up the sample. These results are acceptable.

Table 4. Strains of bacteria used to evaluate the reproducibility of MALDI identification results using the BD Kiestra IdentifA for sample preparation.

Species	Strain
Enterococcus faecalis	ATCC 29212
Escherichia coli	ATCC 25922
	ATCC 35218
Klebsiella aerogenes	ATCC 13048

[2] CLSI. *Reference Method for Broth Dilution Antifungal Susceptibility Testing of Yeasts; Approved Standard.* 4th ed. CLSI standard M27. Wayne, PA. Clinical and Laboratory Standards Institute; 2017.

Pseudomonas aeruginosa	ATCC 27853
Staphylococcus aureus	ATCC 29213
	ATCC 43300
Streptococcus agalactiae	POS 9812
Streptococcus pneumoniae	ATCC 49619
Streptococcus pyogenes	POS 537

Study 2:

To characterize the BD Kiestra IdentifA further, an additional Reproducibility Study was performed using the panel of species and strains listed in **Table 5**. The purpose of this study was to include organisms with a broader range of colony characteristics than the initial study.

Table 5. Strains of bacteria and yeast used to evaluate the reproducibility of MALDI identification results using the BD Kiestra IdentifA for sample preparation.

Species	Strain	Colony Characteristics
Alcaligenes faecalis	ENF 9139	Non-pigmented/grey, flat, irregular
Bacillus cereus	POS 1059	Dull, opaque, irregular
Enterococcus faecalis	ATCC 29212	Grey, smooth, entire margin, non-hemolytic
Candida albicans	ATCC 18804	Opaque, lobate
Corynebacterium jeikium	POS 11247	Grey/white, small, low, convex
Escherichia coli	ATCC 25922	Off-white/beige, shiny, slightly raised
	ATCC 35218	
Klebsiella aerogenes	ATCC 13048	Cream, shiny, smooth, convex
Proteus mirabilis	SCENF 10070	Grey, large, spreading
Pseudomonas aeruginosa	ATCC 27853	Green, large, slightly elevated, undulate
Staphylococcus aureus	ATCC 29213	Yellow, smooth, circular, β-hemolytic
	ATCC 43300	
Staphylococcus epidermidis	POS 274	Smooth, circular, no hemolysis
Streptococcus agalactiae	POS 9812	Non-pigmented, smooth, circular, β-hemolytic
Streptococcus pyogenes	POS 2979	Smooth, moist, convex, β-hemolytic

Each strain was tested in triplicate using three BD Kiestra IdentifA instruments over a period of three days, using three lots of Bruker MALDI reagents (3 replicates × 3 instruments × 3 days = 27 data points per strain). The study was performed with the reusable 48-spot polished steel MALDI target. The results are summarized in **Table 6**.

Table 6. Summary of results from reproducibility testing with the BD Kiestra IdentifA

Species	Bruker MALDI Log(Score) (*N* = 27 each)			
	Concordant, N (%)		No Identification, N (%)	
	≥2.00	1.70-1.99	<1.70	No Peaks
Alcaligenes faecalis	26 (96.3)	1 (3.7)	0	0
Bacillus cereus [1]	0	0	27 (100)	0
Candida albicans	20 (74.1)	7 (25.9)	0	0
Corynebacterium jeikeium [2]	10 (37.0)	11 (40.7)	0	6 (22.2)
Enterobacter aerogenes	27 (100)	0	0	0
Enterococcus faecalis	28 (100)	0	0	0
Escherichia coli	27 (100)	0	0	0
Escherichia coli	27 (100)	0	0	0
Proteus mirabilis	27 (100)	0	0	0
Pseudomonas aeruginosa	27 (100)	0	0	0
Staphylococcus aureus	27 (100)	0	0	0
Staphylococcus aureus	27 (100)	0	0	0
Staphylococcus epidermidis	27 (100)	0	0	0
Streptococcus agalactiae	27 (100)	0	0	0
Streptococcus pyogenes	26 (96.3)	1 (3.7)	0	0

[1] Species not within the validated Bruker MALDI Biotyper CA Reference Library; no identification result is expected
[2] Plates incubated for 48 hours; plates for all other species were incubated for 24 hours prior to testing

Although no incorrect identifications were observed, neither *Corynebacterium jeikium* nor *Candida albicans* met the acceptance criterion of ≥ 95% concordant results with the expected identity and Log(score) ≥ 2.00. An investigation was conducted to assess further. The same two strains were tested by manual preparation using extended Direct Transfer (eDT) Sample Preparation Procedure for the Bruker MALDI Biotyper CA System. For *C. jeikeium*, 23/27 replicates (85.1%) prepared manually gave the expected identification with a Log(score) ≥ 2.00, while for *C. albicans*, 0/27 replicates (0%) after 24 hours of incubation and 4/27 (14.8%) after 48 hours produced the expected identification with a Log(score) ≥ 2.00. It was therefore determined that the failure to meet the acceptance criteria with *C. jeikeium* and *C. albicans* was not due to the performance of the BD Kiestra IdentifA but to the specific characteristics of the isolates used in the study. If a low-confidence identification or no identification result is obtained using eDT method, the operator is instructed to perform additional testing by an alternative method to determine the identity of the organism (e.g., using Bruker's Extraction (Ext) Sample Preparation Procedure). These instructions were also included in the BD Kiestra IdentifA User's Manual. Because this low confidence identification is a property of the original Bruker's system and not the BD Kiestra IdentifA, the results of the Reproducibility Study were determined to be acceptable.

2. Linearity:

   Not applicable.

3. Analytical Specificity/Interference:

   Not applicable.

4. Accuracy (Instrument):

Colony Picking Accuracy

The ability of the BD Kiestra IdentifA to pick the colonies designated by the operator from the digital image obtained by the BD Kiestra ReadA Compact was evaluated in two separate studies described below.

Study 1:

In the first study, three BD Kiestra IdentifA instruments were used to pick designated colonies from 200 mixed cultures of *Escherichia coli* (ATCC 25922; as representative of large bacterial colonies) and *Streptococcus pyogenes* (ATCC 19615; as representative of small bacterial colonies) plated on TSA with 5% sheep blood agar and incubated aerobically for 24 and 48 hours at 35°C. Colonies were picked and used to prepare targets for analysis using a Bruker MALDI Biotyper Compass Research Use Only (RUO) version of the MALDI reference database. Picking of the correct colonies was confirmed by visual inspection of the plates and comparison to the original digital images. All (100%) of 1200 colonies were picked successfully and all (100%) of 400 MALDI target spots provided the expected identification, with Log(score) values ≥ 2.00.

Because an RUO version of the MALDI reference database was inadvertently used for the first study, the accuracy of organism identification was verified by manually preparing a total of 88 target spots from the original culture plates and analyzing them using both the Bruker MALDI Biotyper CA System (IVD) and Bruker MALDI Biotyper Compass (RUO). Two microbial suspensions were prepared, at < 0.8 and > 1.6 McFarland for a total of 44 target spots per bacterial strain per McFarland preparation, and applied to the target mimicking the workflow of the BD Kiestra IdentifA. All results were as expected, with Log(score) ≥ 2.00.

Study 2:

To supplement the data from the first study, a second Colony Picking Accuracy Study was performed in which the initial study protocol was repeated using the same bacterial strains, culture medium, incubation conditions and sample preparation but instead using the IVD Bruker MALDI Biotyper CA System for analysis. As before, the accuracy of colony picking was verified by visual inspection of the culture plates and comparison of the observed and expected MALDI-TOF MS results. All (100%) of 1200 colonies were picked successfully and all (100%) of 400 MALDI target spots provided the expected identification, with Log(score) values ≥ 2.00. These results demonstrate the accuracy of colony picking and are acceptable.

Accuracy of Identification

Accuracy of MALDI-TOF MS organism identification with samples prepared by the BD Kiestra IdentifA was evaluated in the Identification Equivalency Study. A total of 464 isolates of Gram-positive bacteria, Gram negative bacteria and yeasts were tested using 3 BD Kiestra IdentifA instruments and compared to results by manual sample preparation, i.e. performing the extended Direct Transfer (eDT) Procedure and spotting on a MALDI target according to the previously FDA-cleared Bruker MALDI Biotyper CA user manual. Results

from both methods of preparation were compared to the expected result for each isolate to determine agreement, and performance was compared between both methods. As before, expected identity is the reference method used to determine agreement, which is based on the known species identifications of the isolates used in this study.

Tables 7, **8** and **9** show percent agreement for the BD Kiestra IdentifA compared to the expected identity for Gram-negative bacteria, Gram-positive bacteria and yeasts, respectively. The selected isolates were chosen to represent a diverse range of species and colony morphologies that are encountered in clinical microbiology. A substantially lower proportion of concordant results for yeast species than for either Gram-positive or Gram-negative bacteria was noted; however, the original Bruker's system shows that yeasts cannot be reliably identified by manual preparation using the extended direct transfer (eDT) method. Consistent with the original Bruker's system, the BD Kiestra IdentifA User's Manual will recommend that yeast species or any samples that produce a Low Confidence Identification or No Identification Result should be manually prepared using the Bruker's Extraction (Ext) Test Procedure and/or an alternative method of organism identification.

Table 7. Agreement for Gram-negative bacteria using the BD Kiestra IdentifA

Expected Identity	Number	Log(score)					
		Concordant		Discordant		No Identification	
		≥ 2.00	1.70-1.99	≥ 2.00	1.70-1.99	< 1.70	No Peaks
Acinetobacter baumannii/nosocomialis group	10	10	0	0	0	0	0
Bacteroides fragilis	2	2	0	0	0	0	0
Campylobacter coli	3	3	0	0	0	0	0
Campylobacter jejuni	3	3	0	0	0	0	0
Campylobacter lari	1	0	0	0	0	1	0
Citrobacter amalonaticus complex	1	1	0	0	0	0	0
Citrobacter freundii complex	4	4	0	0	0	0	0
Citrobacter koseri	9	9	0	0	0	0	0
Eikenella corrodens	2	2	0	0	0	0	0
Enterobacter aerogenes	9	9	0	0	0	0	0
Enterobacter cloacae complex	7	7	0	0	0	0	0
Escherichia coli	37	37	0	0	0	0	0
Gardnerella vaginalis	2	0	1	1 [2]	0	0	0
Haemophilus influenzae	4	4	0	0	0	0	0
Haemophilus parainfluenzae	2	2	0	0	0	0	0
Hafnia alvei	1	1	0	0	0	0	0
Klebsiella oxytoca/Raoultella ornithinolytica	13	12	0	0	0	0	1
Klebsiella pneumoniae	20	19	0	0	0	0	1
Moraxella sg *Branhamella catarrhalis*	2	2	0	0	0	0	0
Morganella morganii	12	12	0	0	0	0	0
Neisseria gonorrhoeae	1	1	0	0	0	0	0
Pantoea agglomerans	1	0	0	0	1 [6]	0	0
Porphyromonas gingivalis	1	0	0	1 [3]	0	0	0
Prevotella oralis	1	0	0	0	1 [7]	0	0
Proteus mirabilis	9	9	0	0	0	0	0
Proteus vulgaris group	14	11	0	0	0	0	3
Providencia stuartii	10	8	0	1 [4]	0	0	1
Pseudomonas aeruginosa	15	15	0	0	0	0	0
Salmonella sp.	4	4	0	0	0	0	0
Serratia liquefaciens	2	2	0	0	0	0	0
Serratia marcescens	14	13	0	1 [5]	0	0	0
Shigella sonnei	3	3 [1]	0	0	0	0	0
Stenotrophomonas maltophilia	5	5	0	0	0	0	0
Total (%)	**224**	**210 (93.8)**	**1 (0.4)**	**4 (1.8)**	**2 (0.9)**	**1 (0.4)**	**6 (2.7)**
		211 (94.2)		**6 (2.7)**		**7 (3.1)**	

[1] Identified as *Escherichia coli* in accordance with a known Limitation of the Bruker MALDI Biotyper CA
[2] Identified as *Lactobacillus rhamnosus*; concordant with the result obtained with manual sample preparation
[3] Identified as *Veillonella parvula*; concordant with the result obtained with manual sample preparation
[4] Identified as *Citrobacter freundii* complex; concordant with the result obtained with manual sample preparation
[5] Identified as *Enterobacter cloacae* complex; concordant with the result obtained with manual sample preparation
[6] Identified as *Escherichia* species; manual result provided *Escherichia vulneris* (log score of 2.03)
[7] Identified as *Bacteroides caccae* (Low Confidence); manual result provided unspecified (log score of 1.6)

Table 8. Agreement for Gram-positive bacteria using the BD Kiestra IdentifA

Expected Identity	Number	Log(score)					
		Concordant		Discordant		No Identification	
		≥ 2.00	1.70-1.99	≥ 2.00	1.70-1.99	< 1.70	No Peaks
Corynebacterium jeikeium	3	3	0	0	0	0	0
Corynebacterium urealyticum	1	0	0	0	0	1	0
Enterococcus avium	2	2	0	0	0	0	0
Enterococcus casseliflavus	3	3	0	0	0	0	0
Enterococcus faecalis	12	11	0	0	0	0	1
Enterococcus faecium	19	17	1	1 [2]	0	0	0
Enterococcus gallinarum	5	5	0	0	0	0	0
Propionibacterium acnes	2	0	1	0	0	1	0
Rothia dentocariosa	3	1	0	0	0	0	2
Staphylococcus aureus	21	20	0	0	0	0	1
Staphylococcus cohnii	1	0	1	0	0	0	0
Staphylococcus epidermidis	16	15	0	0	0	1	0
Staphylococcus haemolyticus	6	1	4	0	0	1	0
Staphylococcus hominis	3	3	0	0	0	0	0
Staphylococcus lugdunensis	1	1	0	0	0	0	0
Staphylococcus saprophyticus	3	3	0	0	0	0	0
Staphylococcus sciuri	1	0	0	0	0	1	0
Staphylococcus simulans	2	2	0	0	0	0	0
Staphylococcus warneri	1	1	0	0	0	0	0
Staphylococcus xylosus	1	1	0	0	0	0	0
Streptococcus agalactiae	25	24	0	1 [3]	0	0	0
Streptococcus anginosus	1	1	0	0	0	0	0
Streptococcus dysgalactiae	8	8	0	0	0	0	0
Streptococcus gordonii	1	0	1	0	0	0	0
Streptococcus infantarius	1	1 [1]	0	0	0	0	0
Streptococcus mitis/oralis group	4	2	1	0	0	1	0
Streptococcus parasanguinis	1	1	0	0	0	0	0
Streptococcus pneumoniae	32	19	6	1 [4]	0	3	3
Streptococcus pyogenes	10	9	0	1 [5]	0	0	0
Total (%)	**189**	**154 (81.5)**	**15 (7.9)**	**4 (2.1)**	**0 (0.0)**	**9 (4.8)**	**7 (3.7)**
		169 (89.4)		**4 (2.1)**		**16 (8.5)**	

[1] Identified as *Streptococcus lutetiensis* in accordance with a known Limitation of the Bruker MALDI Biotyper CA
[2] Identified as *Enterococcus faecalis*; concordant with the result obtained with manual sample preparation
[3] Identified as *Streptococcus pyogenes*; concordant with the result obtained with manual sample preparation
[4] Identified as *Streptococcus mitis/oralis* group; concordant with the result obtained with manual sample preparation
[5] Identified as *Streptococcus agalactiae*; concordant with the result obtained with manual sample preparation

Table 9. Agreement for yeasts using the BD Kiestra IdentifA

Expected Identity	Number	Log(score)					
		Concordant		Discordant		No Identification	
		≥ 2.00	1.70-1.99	≥ 2.00	1.70-1.99	< 1.70	No Peaks
Candida albicans	8	6	2	0	0	0	0
Candida dubliniensis	2	2	0	0	0	0	0
Candida glabrata	5	3	0	0	0	2	0
Candida guilliermondii	1	1	0	0	0	0	0
Candida kefyr	2	0	2	0	0	0	0
Candida parapsilosis	3	1	2	0	0	0	0
Candida pelliculosa	1	0	1	0	0	0	0
Candida sphaerica	1	0	0	0	0	1	0
Cryptococcus gattii	3	3	0	0	0	0	0
Cryptococcus neoformans var. *grubii*	2	1	0	0	0	0	1
Cryptococcus neoformans var. *neoformans*	3	0	0	1 [2]	0	1	1
Cryptococcus neoformans var. Not Known	1	1 [1]	0	0	0	0	0
Geotrichum candidum	3	0	1	0	0	2	0
Geotrichum capitatum	1	0	0	0	0	1	0
Pichia angusta	2	0	0	0	0	1	1
Saccharomyces cerevisiae	2	1	1	0	0	0	0
Trichosporon aquatile	1	0	0	0	0	1	0
Trichosporon asahii	4	3	0	0	0	1	0
Trichosporon inkin	2	2	0	0	0	0	0
Trichosporon mucoides group	2	0	0	0	0	1	1
Trichosporon ovoides	1	0	0	0	0	0	1
Trichosporon pullulans	1	0	0	0	0	1	0
Total (%)	**51**	**24 (47.1)**	**9 (17.6)**	**1 (2.0)**	**0 (0)**	**12 (23.5)**	**5 (9.8)**
		33 (64.7)		**1 (2.0)**		**17 (33.2)**	

[1] Identified as *Cryptococcus neoformans* var. *grubii*
[2] Identified as *Candida lusitaniae*; with the result obtained with manual sample preparation

Of the 489 isolates included in the study, 25 isolates represented species that were not listed in the Bruker MALDI Biotyper CA Reference Library. A total of 464 isolates represented species that are included in the Bruker MALDI Biotyper CA Reference Library, and therefore, the primary analysis included only those isolates. Of these, 388 (83.6%) were correctly identified using the BD Kiestra IdentifA to prepare targets, with Log(score) Values ≥ 2.00 (high-confidence identification) and 25 (5.4%) gave the expected identification result but with a low-confidence identification (Log(score) 1.70 to 1.99). Results from both methods of preparation were compared to the expected result for each isolate to determine % agreement, and agreement for both methods was compared to demonstrate equivalent performance of the BD Kiestra IdentifA vs. manual preparation of targets (**Table 10**).

Overall, similar agreement with the expected results was observed with the manual method of sample preparation, although the Log(score) Values observed with samples prepared using the BD Kiestra IdentifA tended to be lower for yeast species. These results were determined

to be acceptable because, according to the Instructions for Use of the Bruker MALDI Biotyper CA System, if a low-confidence identification or no identification result is obtained using the eDT method of sample preparation, the operator is instructed to repeat testing of the isolate manually using the Extraction (Ext) Sample Preparation Procedure. Similar instructions are included in the BD Kiestra IdentifA User's Manual.

Table 10. Summary of Log(Scores) observed with samples prepared manually and with the BD Kiestra IdentifA

Organism Group	Disposition vs. Expected	MALDI Log(score)	Number of Isolates (%)	
			BD Kiestra IdentifA	Manual
Gram Negative Bacteria (n = 224)	Concordant	≥ 2.00	210 (93.8)	201 (89.7)
		1.70-1.99	1 (0.4)	7 (3.1)
	Discordant	≥ 2.00	4 (1.8) [1]	5 (2.2)
		1.70-1.99	2 (0.9) [2]	0 (0.0)
	No Identification	< 1.70	1 (0.4)	6 (2.7)
		No Peaks	6 (2.7)	5 (2.2)
Gram Positive Bacteria (n = 189)	Concordant	≥ 2.00	154 (81.5)	155 (82.0)
		1.70-1.99	15 (7.9)	10 (5.3)
	Discordant	≥ 2.00	4 (2.1) [1]	4 (2.1)
		1.70-1.99	0 (0.0) [2]	0 (0.0)
	No Identification	< 1.70	9 (4.8)	8 (4.2)
		No Peaks	7 (3.7)	12 (6.3)
Yeast (n = 51)	Concordant	≥ 2.00	24 (47.1)	31 (60.8)
		1.70-1.99	9 (17.6)	11 (21.6)
	Discordant	≥ 2.00	1 (2.0) [3]	1 (2.0)
		1.70-1.99	0 (0.0)	0 (0.0)
	No Identification	< 1.70	12 (23.5)	7 (13.7)
		No Peaks	5 (9.8)	1 (2.0)
All Isolates Combined (n = 464)	Concordant	≥ 2.00	388 (83.6)	387 (83.4)
		1.70-1.99	25 (5.4)	28 (6.0)
	Discordant	≥ 2.00	9 (1.9)	10 (2.2)
		1.70-1.99	2 (0.4)	0 (0.0)
	No Identification	< 1.70	22 (4.7)	21 (4.5)
		No Peaks	18 (3.9)	18 (3.9)

[1] 7/8 discordant results reported using the BD Kiestra IdentifA agreed with the species identity reported using the manual method of sample preparation

[2] 2/2 Low Confidence identification results obtained with the BD Kiestra IdentifA agreed with High Confidence identifications using the manual method of sample preparation

[3] The result reported using the BD Kiestra IdentifA agreed with that reported using the manual method of sample preparation

A summary of results for isolates included in the study that belonged to species that were not listed in the Bruker MALDI Biotyper CA Reference Library using the BD Kiestra IdentifA method of sample preparation is provided in **Table 11**. These results were determined to be acceptable.

Table 11. Results obtained using the BD Kiestra IdentifA with isolates not included in the Bruker MALDI Biotyper CA Reference Library

Reference Identity	Number	Log(score)			
		≥ 2.00	1.70-1.99	< 1.70 [2]	No Peaks
Bacillus cereus	6	0	0	6	0
Providencia alcalifaciens	6	0	5 [1]	1 [3]	0
Staphylococcus gallinarum	4	0	0	4	0
Staphylococcus hyicus	6	0	0	6	0
Candida sphaerica	1	0	0	1	0
Pichia angusta	2	0	0	1	1
Total	**25**	**0**	**5**	**19**	**1**

[1] Reported as *Providencia rettgeri* (Low Confidence)
[2] No identification reported
[3] Reported as *Providencia alcalifaciens* (Low Confidence) using the manual method of target preparation

Per the recommendation that approximately the same number of samples are processed on each type of MALDI target, updated line listing data were provided to include the target type and showing 285 samples/spots were performed on the 48-spot MALDI target and 179 samples/spots on the 96-spot target.

5. Carry-Over:

Cross-Contamination

Studies were conducted to evaluate the potential for cross-contamination using the BD Kiestra IdentifA within and between culture plates and between spots on the MALDI target.

Study 1

Two hundred nine (209) culture plates were inoculated with two bacterial species (*S. aureus* and a mucoid strain of *K. pneumoniae*) and incubated for 18 to 24 hours to obtain isolated colonies, after which they were used with the BD Kiestra IdentifA to prepare MALDI targets. For processing on the BD Kiestra IdentifA, plates with colonies were alternated on the instrument with uninoculated culture plates. Both 48-spot and 96-spot targets were used for a total of 360 samples/spots performed on the 48-spot MALDI target and 62 samples/spots on the 96-spot target. In total, 213 uninoculated culture plates were processed and all (100%) produced results of either "No Identification" or "No Peaks." From the inoculated cultures, 102 samples of *S. aureus* and 107 samples of *K. pneumoniae* produced the expected results (high confidence identifications with Log(scores) ≥ 2.00 100% agreement to the expected identity in each case). These results demonstrate that the potential for cross-contamination with the BD Kiestra IdentifA is acceptably low.

Handling of Mucoid Colonies

A study was performed to verify the ability of the BD Kiestra IdentifA to prepare suspensions from mucoid colonies using a single, mucous-producing strain of *K. pneumoniae*. One hundred (100) replicate culture plates were inoculated and incubated for 18 to 48 hours, after which 2 BD Kiestra IdentifA instruments were used to prepare both 48- and 96-spot MALDI targets using up to 9 colonies from each culture plate. The presence of mucoid strings, their detection by the instrument and automatic

implementation of a modified mixing protocol to homogenize the bacterial suspension were documented by observation of the colony picking process.

All (100%) of 100 mucoid samples were successfully detected and processed by the instrument and all identification results were as expected with Log(scores) ≥ 2.00. These results are acceptable.

Additional Cross-Contamination Studies

An additional cross-contamination study was performed prior to design verification and validation testing following various software updates, which did not meet the predetermined acceptance criteria, i.e. all (100%) of uninoculated spots should provide No Identification/No Peaks results and no more than 10% of inoculated spots should fail to give the expected identifications with Log(scores) ≥ 2.00. A root cause analysis attributed this to failure to remove the cap-sense logging tool following EMC testing. Following removal of the tool, a study was performed using 1,200 samples in a checkerboard pattern; 600 high McFarland and 600 low McFarland mixtures of *Lactobacillus*, *Streptococcus pneumoniae* and *Corynebacterium jeikeium* which showed improvement in the cross-contamination rate but was still above the acceptance criteria. An analysis determined that 28/29 of the cross-contamination failures occurred when a high sample preceded a low sample with a value < 0.1 McFarland. As a result, BD implemented a software change to prevent all samples below 0.1 McFarland from being spotted for analysis using the BD Kiestra IdentifA, so study results now fell within acceptance criteria. During the root cause investigation, a high-speed camera also determined that when the pipette tip dispensed onto the MALDI target, there was a delay prior to retracting up from the target that created a droplet bridge between the material and pipette tip, creating an opportunity for cross-contamination. Therefore, an additional change was made to the software control architecture that forced the tip to retract immediately upon droplet contact with the target. Upon subsequent testing, this change was shown to reduce the cross-contamination rate to an acceptable level and the software change implementing a 0.1 McFarland cutoff was therefore removed. Validation of the final software version was performed according to the original study protocol: preparing 100 *S. aureus* and 100 *K. pneumoniae* culture plates, spotting each plate onto targets and alternating with spots prepared using uninoculated media. Results were also consistent with those from the original protocol and showed 100% agreement with expected identity.

To address the question of why this issue of cross-contamination was not identified earlier, an examination was conducted of historical information from instruments in the field. Three (3) BD Kiestra IdentifA instruments have been actively running in Europe since 2020: one system has been running in the Netherlands for over 19 months, and the other 2 systems have been running in Switzerland for over 10 months. In that time, there have been no complaints or failure modes related to the cross-contamination event noted above. Over 58,000 samples have been processed across the 3 instruments since January 2020, without any cross-contamination events reported. The assessment is that incremental software changes, particularly the MAS (Synapsys) integration with the BD Kiestra IdentifA software, potentially had an additive effect that could have resulted in delays seen in BD's pipetting system.

The additional cross-contamination study results demonstrated that the additional change made to the software control architecture appears to have addressed the issue of cross-contamination on the BD Kiestra IdentifA.

B Other Supportive Instrument Performance Characteristics Data:

Limit of Detection

The target turbidity for microbial suspensions prepared by the BD Kiestra IdentifA is a McFarland value of 1.6. Technologist-designated colonies (up to a maximum of nine) may be picked and added to the suspension to achieve the targeted turbidity. If the final concentration is below a 0.2 McFarland after all the designated colonies are picked, the target is still spotted but a warning message is displayed that the turbidity was low.

The ability to obtain the expected organism identification with microbial suspensions at the lower extreme of the acceptable concentration for MALDI-TOF MS analysis as defined for the BD Kiestra IdentifA (i.e., 0.2 McFarland) was verified by preparing targets using 10 representative microbial species (3 Gram-positive bacteria, 6 Gram-negative bacteria and 1 yeast; **Table 12**). Strains were plated and grown for 18 to 24 hours, and microbial suspensions were prepared in deionized water of 0.2 to 0.3 McFarland by the BD Kiestra IdentifA. Eight target spots were manually prepared for each organism (4 layers of 3 µL each). Suspensions were plated, and colony counts were performed to estimate the CFU/spot. For each organism, 6/8 replicates should result in a correct identification. All of the organisms obtained 8/8 acceptable MALDI identifications except *Saccharomyces cerevisiae*. For *S. cerevisiae*, 3/8 replicates produced the expected result and 5/8 were reported as "No Peaks." There were no incorrect identifications for any organism.

Table 12. Summary of results from the Limit of Detection Study for the BD Kiestra IdentifA

Organism Group	Species	CFU/spot	Expected Identification Result
Gram-positive Bacteria	*Enterococcus faecalis* ATCC 29212	1.13×10^6	8/8 (100%)
	Enterococcus faecium ATCC 19434	7.89×10^5	8/8 (100%)
	Staphylococcus aureus ATCC 25923	1.30×10^6	8/8 (100%)
Gram-negative Bacteria	*Acinetobacter baumannii* ATCC 19606	1.35×10^6	8/8 (100%)
	Enterobacter cloacae ATCC 13047	1.41×10^6	8/8 (100%)
	Escherichia coli ATCC 25922	1.09×10^6	8/8 (100%)
	Klebsiella pneumoniae ATCC 13883	1.29×10^6	8/8 (100%)
	Proteus vulgaris ATCC 13315	1.65×10^6	8/8 (100%)
	Pseudomonas aeruginosa ATCC 27853	1.24×10^6	8/8 (100%)
Yeast	*Saccharomyces cerevisiae* YST 1125A	1.79×10^4	3/8 (37.5%) [1]

[1] 5/8 samples reported as "No Peaks" (No Organism Identification Possible)

The inability to obtain valid mass spectra when the concentration of yeast in a sample is at the low end of the specified range for turbidity is noted as a Limitation in the device labeling. This was determined to be acceptable because, as described in the Package Insert for the Bruker MALDI Biotyper CA System Reference Library, if an isolate produces a low confidence Log(score) [1.70-1.99], or a result of either "No Identification" [Log(score) 0-1.70] or "No Peaks" [Log(score) = 0] the operator is instructed to repeat the test manually using the Extraction (Ext) Sample Preparation Procedure and/or to perform organism identification by an alternative method.

Reagent Deck Stability

The stability of MALDI-TOF MS reagents (formic acid, US IVD BTS [Bacterial Test Standard] solution and matrix [US IVD HCCA portioned]) onboard the BD Kiestra IdentifA was evaluated under different conditions of temperature and humidity (18°C/20% relative humidity [RH]; 27°C/20% RH and 27°C/80% RH). Eight (8) representative species, 3 Gram-positive bacteria, 4 Gram-negative bacteria and 1 yeast, were grown for 18 to 24 hours. At intervals, colonies of representative species were designated for automated picking and preparation of MALDI targets in duplicate (16 samples/spots in total) by the BD Kiestra IdentifA using reagents stored on the deck of the instrument. All 16 samples were run on both 48- and 96-spot targets; a total of 7 batches were processed at time zero to 8 hours. In addition, targets were prepared using *E. coli* and 3 lots of matrix and formic acid held on deck up to 24 hours (at the three environmental conditions stated above). The concentration of formic acid was also verified by High Performance Liquid Chromatography (HPLC). The results of the study demonstrated that the BTS solution is stable up to 8 hours and the matrix and formic acid are stable up to 24 hours on the deck of the BD Kiestra IdentifA instrument.

All BTS spots (42 total) passed QC on the MALDI Biotyper CA instrument, and 308 of 310 sample spots prepared using the onboard reagents were correctly identified with Log(score) ≥ 2.00. These results are acceptable. To further mitigate the risks associated with potential use of an expired reagent, the time elapsed since placement of each reagent on the deck of the BD Kiestra IdentifA is tracked automatically by the system software and the user is alerted when a reagent is about to expire.

In a separate study, bottles of deionized water used for preparation of microbial suspensions were shown to be stable for 24 hours on the deck of the BD Kiestra IdentifA prior to use. These results are acceptable.

VIII Proposed Labeling:

The labeling **supports** the finding of substantial equivalence for this device.

IX Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.