May 10, 2019

GenePOC Inc.
Dany Leblanc
VP QA/RA
360 rue Franquet
Quebec, G1P 4N3 Ca

Re: K190275
Trade/Device Name: GenePOC Carba
Regulation Number: 21 CFR 866.1640
Regulation Name: Antimicrobial Susceptibility Test Powder
Regulatory Class: Class II
Product Code: PMY, OOI
Dated: February 6, 2019
Received: February 8, 2019

Dear Dany Leblanc:

We have reviewed your Section 510(k) premarket notification of intent to market the device referenced above and have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (Act) that do not require approval of a premarket approval application (PMA). You may, therefore, market the device, subject to the general controls provisions of the Act. Although this letter refers to your product as a device, please be aware that some cleared products may instead be combination products. The 510(k) Premarket Notification Database located at https://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfpmn/pmn.cfm identifies combination product submissions. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration. Please note: CDRH does not evaluate information related to contract liability warranties. We remind you, however, that device labeling must be truthful and not misleading.

If your device is classified (see above) into either class II (Special Controls) or class III (PMA), it may be subject to additional controls. Existing major regulations affecting your device can be found in the Code of Federal Regulations, Title 21, Parts 800 to 898. In addition, FDA may publish further announcements concerning your device in the Federal Register.

Please be advised that FDA's issuance of a substantial equivalence determination does not mean that FDA has made a determination that your device complies with other requirements of the Act or any Federal statutes and regulations administered by other Federal agencies. You must comply with all the Act's requirements, including, but not limited to: registration and listing (21 CFR Part 807); labeling (21 CFR Part 801 and Part 809); medical device reporting (reporting of medical device-related adverse events) (21 CFR

803) for devices or postmarketing safety reporting (21 CFR 4, Subpart B) for combination products (see https://www.fda.gov/CombinationProducts/GuidanceRegulatoryInformation/ucm597488.htm); good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820) for devices or current good manufacturing practices (21 CFR 4, Subpart A) for combination products; and, if applicable, the electronic product radiation control provisions (Sections 531-542 of the Act); 21 CFR 1000-1050.

Also, please note the regulation entitled, "Misbranding by reference to premarket notification" (21 CFR Part 807.97). For questions regarding the reporting of adverse events under the MDR regulation (21 CFR Part 803), please go to http://www.fda.gov/MedicalDevices/Safety/ReportaProblem/default.htm.

For comprehensive regulatory information about medical devices and radiation-emitting products, including information about labeling regulations, please see Device Advice (https://www.fda.gov/MedicalDevices/DeviceRegulationandGuidance/) and CDRH Learn (http://www.fda.gov/Training/CDRHLearn). Additionally, you may contact the Division of Industry and Consumer Education (DICE) to ask a question about a specific regulatory topic. See the DICE website (http://www.fda.gov/DICE) for more information or contact DICE by email (DICE@fda.hhs.gov) or phone (1-800-638-2041 or 301-796-7100).

Sincerely,

for

Uwe Scherf, M.Sc., Ph.D.
Director
Division of Microbiology Devices
OHT7: Office of In Vitro Diagnostics
and Radiological Health
Office of Product Evaluation and Quality
Center for Devices and Radiological Health

Enclosure

5.0 510(K) SUMMARY

510K SUMMARY

510(k) Number: K190275

A. GENERAL INFORMATION

Submission Date: April 29, 2019

Submitter Information:

Submitted By: GenePOC Inc.
360 rue Franquet
Québec (Québec) G1P 4N3

Contact Person: Dany Leblanc
VP Quality Assurance and Regulatory Affairs
GenePOC Inc.
Telephone: +1 418 650-3535
Email: dany.leblanc@genepoc.ca

B. PURPOSE FOR SUBMISSION

To obtain a substantial equivalence determination for the GenePOC™ Carba assay on the revogene™ instrument for the qualitative detection and differentiation of the *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$ gene sequences from carbapenem-non-susceptible pure colonies of *Enterobacteriaceae*, *Acinetobacter baumannii*, or *Pseudomonas aeruginosa.*

C. MEASURAND

Target DNA sequence of the following genes: *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$

D. TYPE OF TEST

Qualitative real-time Polymerase Chain Reaction (PCR) assay

E. APPLICANT

GenePOC Inc.

F. PROPRIETARY AND ESTABLISHED NAMES

Proprietary Name: GenePOC™ Carba
Common Name : GenePOC™ Carba assay

G. REGULATORY INFORMATION

Trade Name:	GenePOC™ Carba
Classification:	Class II
Regulation:	21 CFR 866.1640
Regulation Name:	Antimicrobial susceptibility test powder
Product Code:	PMY, OOI
Panel:	83 - Microbiology

H. INTENDED USE

1. Intended Use and Indications for Use:

   The GenePOC™ Carba assay, performed on the revogene™ instrument, is a qualitative *in vitro* diagnostic test designed for the detection and differentiation of the *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$ gene sequences associated with carbapenem-non-susceptible pure colonies of *Enterobacteriaceae, Acinetobacter baumannii*, or *Pseudomonas aeruginosa*, when grown on blood agar or MacConkey agar. The test utilizes automated real-time Polymerase Chain Reaction (PCR).

   The GenePOC™ Carba assay should be used in conjunction with other laboratory tests including phenotypic antimicrobial susceptibility testing. A negative GenePOC™ Carba assay result does not preclude the presence of other resistance mechanisms.

   The GenePOC™ Carba assay is intended as an aid for infection control in the detection of carbapenem-non-susceptible bacteria that colonize patients in healthcare settings. The identification of a *bla*$_{IMP}$, *bla*$_{NDM}$ or *bla*$_{VIM}$ metallo-β-lactamase gene (i.e., the genes that encode the IMP, NDM and VIM metallo-β-lactamases, respectively) may be used as an aid to clinicians in determining appropriate therapeutic strategies for patients with known or suspected carbapenem non-susceptible infections.

2. Special conditions for use statement(s):

   Prescription Use Only

   Organisms should be identified and carbapenem non-susceptibility status should be determined prior to testing with the GenePOC™ Carba assay.

   The GenePOC™ Carba assay detects *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$ resistance genes from pure colonies and is not for bacterial identification.

   The performance of the GenePOC™ Carba assay with bacteria other than *Enterobacteriaceae, Acinetobacter baumannii* or *Pseudomonas aeruginosa*, has not been evaluated.

The GenePOC™ Carba assay is not a sub-typing tool and does not report variants of the *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$.

3. Special instrument requirements:

   The GenePOC™ Carba assay uses PCR technology on the revogene™ instrument platform, which extracts, amplifies, and detects the target DNA.

I. INDICATIONS FOR USE

Same as Intended Use.

J. DEVICE DESCRIPTION

The GenePOC™ Carba assay is an automated real-time polymerase chain reaction (PCR) *in vitro* diagnostic test for the qualitative detection and differentiation of the *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$ gene sequences in carbapenem-non-susceptible pure colonies of *Enterobacteriaceae, Acinetobacter baumannii*, or *Pseudomonas aeruginosa* grown on blood agar or MacConkey agar. The GenePOC™ Carba assay is designed to be performed on the revogene™ instrument. The revogene™ is an instrument that automates sample homogenization, sample dilution, cells lysis, DNA amplification, and detection of the amplified PCR products.

The GenePOC™ Carba assay kit is comprised of single-use, disposable microfluidic cartridges (PIEs), Sample Buffer Tubes (SBT), and Disposable Transfer Tools (DTT). These components are used to dilute the sample, extract, amplify, and simultaneously detect *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$ DNA. A Process Control (PrC) is also incorporated into each PIE to verify sample processing and amplification steps. The PrC allows for the verification of potential inhibitor substances as well as microfluidic, instrument or reagent failure.

Each GenePOC™ Carba assay kit contains 24 individual pouches. Each pouch has components for one (1) test including one (1) Carba PIE, one (1) SBT, and one (1) DTT. The single-use, multi-chambered fluidic cartridges are designed to complete sample preparation and real-time PCR for the detection of the *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$ gene sequences from isolates of pure cultures of carbapenem-non-susceptible gram-negative bacteria in approximately 70 minutes. User intervention is only required for preparing the standardized 0.5 McFarland bacterial suspension from characterized carbapenem-non-susceptible isolated colonies, inoculating the bacterial suspension into the Sample Buffer Tube (SBT), transferring the sample into the single-use disposable PIE, and loading/unloading the PIEs into the revogene™ carousel. Each PIE is a completely integrated closed device in which a sample is dispensed and processed through different microfluidic chambers and channels that allow for the sample processing and subsequent real-time PCR steps.

Upon completion of a run, the results are interpolated by the revogene™ from measured fluorescent signals and embedded calculation algorithms. The output results include positive, negative, indeterminate, and unresolved. Upon completion of a run, the user removes the used

cartridges and disposes of them in normal biological waste. The results are automatically generated at the end of the process in a report that can be viewed and printed.

K. SUBSTANTIAL EQUIVALENCE INFORMATION

1. Predicate Device Name:
   Xpert® Carba-R Assay

2. Predicate 510(k) Number:
   K152614

3. Comparison with Predicate:
   The GenePOC™ Carba assay is substantially equivalent to the Xpert® Carba-R Assay (K152614). The GenePOC™ Carba and the Xpert Carba-R assays both detect target gene sequences from antibiotic-non-susceptible bacteria and use real-time PCR amplification and fluorogenic target-specific hybridization detection. The performance of the GenePOC™ Carba assay was determined in a multi-site clinical study. The table below shows the similarities and differences between the GenePOC™ Carba assay and the predicate device (Xpert® Carba-R Assay).

Item	GenePOC™ Carba assay	Xpert® Carba-R Assay (Predicate Device)
K Number	K190275	K152614
SIMILARITIES		
Classification	Class II	Class II
Intended Use	The GenePOC™ Carba assay, performed on the revogene™ instrument, is a qualitative *in vitro* diagnostic test designed for the detection and differentiation of the *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$ gene sequences associated with carbapenem-non-susceptible pure colonies of *Enterobacteriaceae*, *Acinetobacter baumannii*, or *Pseudomonas aeruginosa*, when grown on blood agar or MacConkey agar. The test utilizes automated real-time Polymerase Chain Reaction (PCR). The GenePOC™ Carba assay should be used in conjunction with other laboratory tests including phenotypic antimicrobial susceptibility testing. A negative GenePOC™ Carba assay result does not preclude the presence of other resistance mechanisms. The GenePOC™ Carba assay is intended as an aid for infection control in the detection of carbapenem-non-susceptible bacteria that colonize patients in healthcare settings. The identification of a *bla*$_{IMP}$, *bla*$_{NDM}$ or *bla*$_{VIM}$	The Xpert® Carba-R Assay, performed on the GeneXpert® Instrument Systems, is a qualitative *in vitro* diagnostic test for the detection and differentiation of the *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48}$, and *bla*$_{IMP}$ gene sequences associated with carbapenem-non-susceptible pure colonies of *Enterobacteriaceae*, *Acinetobacter baumannii*, or *Pseudomonas aeruginosa* grown on blood agar or MacConkey agar. The test utilizes automated real-time polymerase chain reaction (PCR). A negative Xpert Carba-R Assay result does not preclude the presence of other resistance mechanisms. The Xpert Carba-R Assay should be used in conjunction with other laboratory tests including phenotypic antimicrobial susceptibility testing. The Xpert Carba-R Assay is intended as an aid for infection control in detecting

Item	GenePOC™ Carba assay	Xpert® Carba-R Assay (Predicate Device)
	metallo-β-lactamase gene (i.e., the genes that encode the IMP, NDM and VIM metallo-β-lactamases, respectively) may be used as an aid to clinicians in determining appropriate therapeutic strategies for patients with known or suspected carbapenem non-susceptible infections.	and differentiating genetic markers of resistance to monitor the spread of carbapenem-non-susceptible organisms in healthcare settings. The Xpert Carba-R Assay is not intended to guide or monitor treatment for carbapenem-non-susceptible bacterial infections.
Technological Principles	Automated nucleic acid amplification; real-time PCR	Same
Test Cartridge	Disposable single-use, multi-chambered fluidic cartridge	Same
Detection Probes	TaqMan® Probes	Same
Assay Targets	Detects *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$ genes.	Same
Sample Preparation	Automated	Same
Interpretation of Test Results	Automated	Same
Sample Type	Bacterial isolates from culture	Same
Organisms	*Enterobacteriaceae*, *Pseudomonas aeruginosa*, *Acinetobacter baumannii*	Same
DIFFERENCES		
Instrument System	revogene™	GeneXpert Instrument System (includes GeneXpert Dx, Infinity-48, Infinity-48s, and Infinity-80)
Time to Obtain Test Results	Approximately 70 minutes	Approximately 50 minutes
Variant Type(s) Detected (based on analytical studies) [1]	KPC-2, 3, 4	KPC-2, 3, 4
	NDM-1, 4 to 7	NDM-1, 2, 4, 5
	IMP-1, 4, 8, 9, 11	IMP-1, 2, 4, 6, 10, 11
	OXA-48, 181, 204, 232	OXA-48, 181
	VIM-1, 2, 10, 19	VIM-1, 2, 4, 10, 19
Variant Types(s) Not Detected (based on analytical studies) [1]	None	IMP-7, 13, 14
Additional Detectable Variant Types(s) Predicted from *in silico* analysis[1]	KPC-5 to 38	KPC-5 to 16
	NDM-2, 3, 8 to 24	NDM-3, 6 to 9
	IMP-2, 5, 6, 10, 13 to 20, 23 to 26, 28 to 30, 32, 33, 37, 38, 40, 42, 45, 47 to 49, 53 to 56, 59, 60, 62, 66, 69 to 72, 74 to 79	IMP-3, 8, 9, 13, 19 to 22, 24, 25, 27, 28, 30, 31, 33, 37, 40, 42
	OXA-162, 199, 244, 245, 252, 370, 484, 505, 514, 515, 519, 546, 547, 566	OXA-162, 163, 204, 232, 244, 245, 247
	VIM-3 to 6, 8, 9, 11, 12, 14 to 18, 20, 23 to 46, 48 to 50, 52 to 55, 57, 59, 60	VIM 5 to 9, 11 to 18, 20, 23 to 38

[1] **Note:** The variants listed reflect the respective labeling of each device and the analytical studies and *in silico* analyses conducted at the time of 510(k) clearance.

The GenePOC™ Carba assay has the same general intended use as the predicate device and has the same technological characteristics as the predicate device. The differences between the GenePOC™ Carba assay and the predicate device do not raise new questions of safety and effectiveness.

L. STANDARDS/GUIDANCE DOCUMENTS REFERENCED

- CLSI Guideline EP25-A, Evaluation of Stability of In Vitro Diagnostic Reagents; Approved Guideline, 2009.
- CLSI EP28-A3c, Guideline Defining, Establishing, and Verifying Reference Intervals in the Clinical Laboratory; Approved Guideline, 2008.
- CLSI M02, Performance Standards for Antimicrobial Disk Susceptibility Tests; Approved Guideline, 13th edition, 2018.
- CLSI M100, Performance Standards for Antimicrobial Susceptibility Testing; Approved Guideline, 28th edition, 2018.

M. TEST PRINICIPLE

The GenePOC™ Carba assay is a single-use test for the qualitative detection and differentiation of the *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$ gene sequences in carbapenem-non-susceptible pure colonies of *Enterobacteriaceae, Acinetobacter baumannii,* or *Pseudomonas aeruginosa* grown on blood agar or MacConkey agar that utilizes an automated sample preparation and real-time polymerase chain reaction (PCR) technology with fluorogenic detection of the amplified DNA. The GenePOC™ Carba assay utilizes the GenePOC™ Carba microfluidic cartridge (PIE) for the simultaneous detection of the *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$ DNA and the internal process control (PrC) DNA (to verify sample processing, amplification, and the absence of reaction inhibitors).

Well-characterized *Enterobacteriaceae*, *Acinetobacter baumannii*, or *Pseudomonas aeruginosa,* carbapenem-non-susceptible strain is streaked for isolation on a blood agar plate (BAP) or a MacConkey agar plate with a 10-µg meropenem disc on the first quadrant. The plate is then incubated for 18-24 hours at 35°C ± 2°C under aerobic conditions. A standardized 0.5 McFarland (McF) suspension is prepared in saline solution by collecting isolated colonies selected from the incubated agar plate with a sterile swab. Fifteen (15) µL of the 0.5 McF suspension are transferred into the Sample Buffer Tube (SBT) containing 1 mL of Sample Buffer (SB). After a 15-second vortex step at maximal speed, a volume of SB lying between the two marks of the DTT is sampled from the SBT and transferred into a GenePOC™ Carba PIE to perform the assay. The loaded GenePOC™ Carba PIE is placed into the revogene™ for further sample processing. No operator intervention is necessary once the sample is loaded onto the revogene™.

Each Carba PIE is a completely integrated and self-contained device. Each sample is sequentially transferred by centrifugation from one microfluidic chamber to the next and all reagents specific for the PCR reaction are incorporated and dried within the PCR wells. The stepwise process includes sample homogenization, lysis of cells, and sample dilution followed by the subsequent real-time PCR steps. An internal PrC is contained in the homogenization chamber and is therefore present in every test to verify critical steps of the analytical process

(including sample lysis, dilution and nucleic acid amplification and detection) for the presence of potential inhibitory substances as well as system or reagent failures. The amplified products are detected in real time using target-specific TaqMan® chemistry-based probes. The results are interpolated by the system from measured fluorescent signals and embedded calculation algorithms. Results may be viewed, printed, transferred, and/or stored by the user.

N. PERFORMANCE CHARACTERISTICS

1. Analytical Performance

a. Reproducibility/Precision

The Reproducibility and Precision study was performed by testing a total of ten (10) positive bacterial carbapenem-non-susceptible strains, each one harboring resistance gene targeted by the GenePOC™ Carba assay (n=2 strains per resistance gene, i.e. *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$) and one (1) negative bacterial carbapenem-non-susceptible strain that does not carry any of the resistance genes targeted by the assay from standardized 0.5 McF suspensions.

The Between-Laboratory Reproducibility study was performed by two (2) operators at three (3) sites with one (1) GenePOC™ Carba assay kit lot over a total of five (5) days (consecutive or not) per panel member tested. Two (2) runs were performed by each operator on each day. On each day of testing, four (4) panel members of each positive strain were tested on each site and by each operator for a total of 120 replicates per strain (*5 days of testing x 4 panel members per strain x 3 sites x 2 operators = 120 replicates per strain*). Eight (8) negative panel members were also tested on each site and by each operator, on each respective day of testing for a total of 240 replicates (*5 days of testing x 8 panel members x 3 sites x 2 operators = 240 replicates*).

To assess the Between-Lot Reproducibility, testing was pursued at one (1) site with two (2) additional kit lots for a total of 15 days of testing (five (5) days per kit lot, consecutive or not) per panel member tested for each kit lot. Two (2) runs per kit lot were performed by each operator on each day. On each day of testing, four (4) panel members of each positive strain were tested by each operator for a total of 120 replicates per strain (*5 days of testing x 4 panel members per strain x 3 kit lots x 2 operators = 120 replicates per strain*). Eight (8) negative panel members were also tested with each kit lot and by each operator, on each respective day of testing for a total of 240 replicates (*5 days of testing x 8 panel members x 3 kit lots x 2 operators = 240 replicates*).

The Within-Laboratory Precision for each panel member was calculated based on one (1) GenePOC™ Carba assay kit lot (kit lot 1) that was tested by two (2) operators at site 1 over a total of five (5) days (consecutive or not). A total of 40 replicates for each of the ten (10) positive bacterial carbapenem-non-susceptible strains, each one harboring one (1) resistance gene targeted by the GenePOC Carba assay (n=2 strains per resistance gene), were considered for the analysis (*5 days of testing x 4 panel members per strain x 2 operators = 40 replicates*). A total of 80 replicates of the

negative bacterial carbapenem-non-susceptible strain that does not carry any of the resistance genes targeted by the assay were also considered (*5 days of testing x 8 panel members x 2 operators = 80 replicates*).

For the negative samples tested, there was agreement of 98.8% (237/240; 95% CI: [96.4-99.6%], 99.6% (239/240; 95% CI: [97.7-99.9%]) and 100.0% (80/80; 95% CI: [95.4-100.0%], respectively for the Between-Laboratory Reproducibility study, the Between-Lot Reproducibility study and for the Within-Laboratory Precision study. For the positive samples, agreement of 100.0% (120/120; 95% CI: [96.9-100.0%]), 100.0% (120/120; 95% CI: [96.9-100.0%]) and 100.0% (40/40; 95% CI: [91.2-100.0%]), was achieved for each panel member in the Between-Laboratory Reproducibility study, the Between-Lot Reproducibility study and for the Within-Laboratory Precision study, respectively.

Tested Target Results and Overall Agreement for the Between-Laboratory Reproducibility Qualitative Analysis

Resistance Gene and Variant	Results/Total									Overall % Agreement [95% CI]
	Site 1			Site 2			Site 3			
	Operator A	Operator B	Site	Operator A	Operator B	Site	Operator A	Operator B	Site	
IMP-1	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
IMP-4	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
KPC-4	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
KPC-2	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
NDM-1	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
NDM-5	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
OXA-181	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
OXA-48	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
VIM-1	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
VIM-1	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
Negative	40/40	40/40	80/80	39/40	40/40	79/80	39/40	39/40	78/80	237/240 (98.8%) [96.4-99.6]

Tested Target Results and Overall Agreement for the Between-Lot Reproducibility Qualitative Analysis

Resistance Gene and Variant	Results/Total									Overall % Agreement [95% CI]
	Kit Lot 1			Kit Lot 2			Kit Lot 3			
	Operator A	Operator B	Site	Operator A	Operator B	Site	Operator A	Operator B	Site	
IMP-1	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
IMP-4	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
KPC-4	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
KPC-2	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
NDM-1	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
NDM-5	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
OXA-181	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
OXA-48	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
VIM-1	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
VIM-1	20/20	20/20	40/40	20/20	20/20	40/40	20/20	20/20	40/40	120/120 (100.0%) [96.9-100.0]
Negative	40/40	40/40	80/80	40/40	39/40	79/80	40/40	40/40	80/80	239/240 (99.6%) [97.7-99.9]

Tested Target Results and Overall Agreement for the Within-Laboratory Precision Qualitative Analysis

Resistance Gene and Variant	Results/Total		Overall % Agreement [95% CI]
	Site 1 and Kit Lot 1		
	Operator A	Operator B	
IMP-1	20/20	20/20	40/40 (100.0%) [91.2-100.0]
IMP-4	20/20	20/20	40/40 (100.0%) [91.2-100.0]
KPC-4	20/20	20/20	40/40 (100.0%) [91.2-100.0]
KPC-2	20/20	20/20	40/40 (100.0%) [91.2-100.0]
NDM-1	20/20	20/20	40/40 (100.0%) [91.2-100.0]
NDM-5	20/20	20/20	40/40 (100.0%) [91.2-100.0]
OXA-181	20/20	20/20	40/40 (100.0%) [91.2-100.0]
OXA-48	20/20	20/20	40/40 (100.0%) [91.2-100.0]
VIM-1	20/20	20/20	40/40 (100.0%) [91.2-100.0]
VIM-1	20/20	20/20	40/40 (100.0%) [91.2-100.0]
Negative	40/40	40/40	80/80 (100.0%) [95.4-100.0]

Ct Values for the Between-Laboratory Reproducibility Quantitative Analysis

Resistance Gene and Variant	Mean Ct[1]	N	Between-Laboratory		Between-Operator		Between-Day		Repeatability		Overall	
			SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%
IMP-1	29.7	120	1.2	4.0	0.6	2.2	0.0	0.0	0.9	3.1	1.6	5.5
IMP-4	29.6	120	0.7	2.2	0.4	1.4	0.2	0.7	1.0	3.3	1.3	4.3
KPC-4	30.5	120	1.3	4.1	0.7	2.3	0.2	0.8	1.6	5.1	2.1	7.0
KPC-2	30.4	120	1.6	5.2	0.4	1.4	0.1	0.2	1.5	5.0	2.2	7.3
NDM-1	30.3	120	0.3	0.9	0.1	0.2	0.0	0.0	0.9	3.0	1.0	3.1
NDM-5	30.2	120	0.6	2.1	0.5	1.6	0.2	0.5	1.2	4.0	1.5	4.8
OXA-181	28.5	120	0.9	3.2	0.3	0.9	0.2	0.8	1.0	3.4	1.4	4.8
OXA-48	30.5	120	1.4	4.5	0.5	1.5	0.3	0.9	1.1	3.7	1.9	6.1
VIM-1	27.6	120	0.3	1.1	0.4	1.3	0.0	0.0	0.9	3.1	1.0	3.5
VIM-1	28.5	120	0.3	1.0	0.5	1.7	0.0	0.0	1.1	3.8	1.2	4.3
Negative	34.9	237	1.1	3.1	0.7	2.0	0.4	1.2	1.4	3.9	1.9	5.5

N: Number; SD: Standard Deviation; CV: Coefficient of Variation
[1] For resistance genes and variants, Ct values reported are for the specified gene. For negative samples, Ct values reported are for the PrC.

Ct Values for the Between-Lot Reproducibility Quantitative Analysis

Resistance Gene and Variant	Mean Ct[1]	N	Between-Lot		Between-Operator		Between-Day		Repeatability		Overall	
			SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%
IMP-1	29.0	120	0.8	2.6	0.3	1.2	0.1	0.4	0.7	2.5	1.1	3.8
IMP-4	29.2	120	0.8	2.8	0.3	0.9	0.1	0.4	1.1	3.6	1.4	4.7
KPC-4	29.6	120	1.6	5.4	0.3	1.1	0.0	0.0	1.5	5.0	2.2	7.5
KPC-2	30.8	120	1.2	3.9	0.3	1.1	0.3	1.0	1.9	6.1	2.3	7.4
NDM-1	29.5	120	1.0	3.2	0.1	0.3	0.1	0.4	1.1	3.6	1.4	4.8
NDM-5	28.9	120	1.1	3.7	0.3	1.2	0.0	0.0	1.0	3.4	1.5	5.1
OXA-181	28.2	120	1.2	4.1	0.3	0.9	0.2	0.8	0.9	3.0	1.5	5.2
OXA-48	28.6	120	1.3	4.6	0.2	0.7	0.0	0.0	1.0	3.3	1.6	5.7
VIM-1	27.5	120	0.4	1.5	0.4	1.4	0.1	0.5	0.8	2.9	1.0	3.6
VIM-1	28.1	120	0.9	3.0	0.3	1.0	0.0	0.0	1.0	3.6	1.4	4.8
Negative	33.0	239	2.5	7.5	0.7	2.1	0.4	1.3	1.3	3.9	2.9	8.8

N: Number; SD: Standard Deviation; CV: Coefficient of Variation

[1] For resistance genes and variants, Ct values reported are for the specified gene. For negative samples, Ct values reported are for the PrC.

Ct Values for the Within-Laboratory Precision Quantitative Analysis

Resistance Gene and Variant	Mean Ct[1]	N	Between-Day		Between-Run		Repeatability		Overall	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV
IMP-1	30.0	40	0.0	0.0	0.0	0.0	0.8	2.5	0.8	2.5
IMP-4	30.2	40	0.5	1.5	0.0	0.0	1.0	3.4	1.1	3.7
KPC-4	31.5	40	0.0	0.0	0.0	0.0	1.6	5.0	1.6	5.0
KPC-2	32.2	40	0.1	0.4	0.0	0.0	1.7	5.1	1.7	5.1
NDM-1	30.6	40	0.3	0.9	0.1	0.3	0.9	3.1	1.0	3.2
NDM-5	30.2	40	0.4	1.4	0.0	0.0	0.9	2.8	1.0	3.2
OXA-181	29.5	40	0.4	1.2	0.0	0.0	0.9	3.2	1.0	3.4
OXA-48	30.1	40	0.0	0.0	0.5	1.6	1.0	3.5	1.1	3.8
VIM-1	28.1	40	0.1	0.3	0.0	0.0	0.9	3.3	0.9	3.3
VIM-1	29.1	40	0.0	0.0	0.2	0.7	1.0	3.4	1.0	3.5
Negative	35.9	80	0.6	1.7	0.4	1.1	1.8	5.1	2.0	5.5

N: Number; SD: Standard Deviation; CV: Coefficient of Variation
[1] For resistance genes and variants, Ct values reported are for the specified gene. For negative samples, Ct values reported are for the PrC.

b. Linearity/Assay Reportable Range

Not Applicable.

c. Traceability, Stability, Expected Values (controls, calibrators, or methods)

Internal Process Control (PrC)

Each PIE contains a PrC that controls for amplification inhibition, assay reagents, and sample processing effectiveness.

External Controls

External Controls should be prepared by the end user. These are recommended but not required to perform the GenePOC™ Carba assay. The user should select the most appropriate controls for their laboratory quality control program to comply with applicable regulations and the requirements of accrediting agencies.

As a Positive External Control, GenePOC recommends using a standardized 0.5 McF suspension of *Klebsiella pneumoniae* or *Escherichia coli* prepared from a commercially available strain that harbors one of the targeted carbapenemase genes (NCTC 13476 for *bla*$_{\text{IMP-1}}$, CCUG 59348 for *bla*$_{\text{KPC-2}}$, ATCC® BAA-2146™ for *bla*$_{\text{NDM-1}}$, ATCC® BAA-2523™ for *bla*$_{\text{OXA-48}}$ and NCTC 13440 for *bla*$_{\text{VIM-1}}$).

As a Negative External Control, GenePOC recommends using a standardized 0.5 McF suspension of isolated colonies from a representative carbapenem-non-susceptible bacterial strain of *Enterobacteriaceae, A. baumannii* or *P. aeruginosa* that does not carry any of the resistance genes targeted by the GenePOC™ Carba assay.

Sample Stability in the PIE

The GenePOC™ Carba assay PIE (microfluidic cartridge) stability study was verified by testing five (5) positive carbapenem-non-susceptible bacterial strains, each carrying a different carbapenemase gene (*bla*$_{\text{KPC}}$, *bla*$_{\text{NDM}}$, *bla*$_{\text{VIM}}$, *bla*$_{\text{OXA-48-like}}$, and *bla*$_{\text{IMP}}$) targeted by the GenePOC™ Carba assay. In addition, a carbapenem-non-susceptible *Enterobacter cloacae* strain that does not carry any of the carbapenemase resistance genes targeted by the assay was also tested.

Both positive and negative samples were tested with the GenePOC™ Carba assay from standardized 0.5 and 4 McF suspensions, respectively. The results of the study support the recommended maximum interval of one (1) hour at 25 ± 2°C from opening of the PIE pouch and sample addition into the GenePOC™ Carba assay PIE to processing on the revogene™ instrument.

Sample Stability

Sample stability was evaluated with Sample Buffer Tube (SBT) inoculated with a standardized 0.5 McF suspension of a bacterial strain carrying one of the resistance genes targeted by the GenePOC™ Carba assay (*bla*$_{\text{KPC}}$, *bla*$_{\text{NDM}}$, *bla*$_{\text{VIM}}$, *bla*$_{\text{OXA-48-like}}$, and *bla*$_{\text{IMP}}$) and with a standardized 0.5 McF suspension of a carbapenem-non-susceptible bacterial strain that does not carry any of the resistance genes targeted by the GenePOC™ Carba assay. The product labeling indicates that SBT inoculated with a standardized 0.5 McF suspension prepared from carbapenem non-susceptible bacterial isolates can be stored at 2-8°C for up to seven (7) days or at 25 ± 2°C for up to four (4) days prior to testing with the GenePOC™ Carba assay.

d. Detection Limit

Not applicable.

e. Analytical Reactivity

The analytical reactivity (inclusivity) of the GenePOC™ Carba assay was evaluated with 58 carbapenem-non-susceptible isolates of *Enterobacteriaceae, Pseudomonas aeruginosa* and *Acinetobacter baumannii* from various geographic origins representing

temporal diversity and harboring variants of the *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$ carbapenemase genes:

- Two (2) strains with two (2) resistance genes;
- 11 strains (including five [5] variants) with the *bla*$_{IMP}$ resistance gene;
- 11 strains (including at least three [3] variants) with the *bla*$_{KPC}$ resistance gene;
- 14 strains (including five [5] variants) with the *bla*$_{NDM}$ resistance gene;
- 11 strains (including three [3] variants) with the *bla*$_{OXA-48-like}$ resistance gene;
- 9 strains (including four [4] variants) with the *bla*$_{VIM}$ resistance gene.

Each strain was tested from a standardized 0.5 McF bacterial suspension. Three (3) replicates per strain were tested using three (3) different GenePOC™ Carba kit lots (one (1) replicate per kit lot). The ability of the GenePOC™ Carba assay to detect multiple variants of the *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$ carbapenemase genes in the 58 *Enterobacteriaceae*, *Pseudomonas aeruginosa* and *Acinetobacter baumannii* strains carrying these genes was demonstrated.

Carbapenem-Non-susceptible Bacterial Isolates Tested for Analytical Reactivity (Inclusivity) with the GenePOC™ Carba Assay

Species	Collection Number	Geographical and Temporal Origin		Reported Resistance Gene and Variant (When Available)
Isolates Harboring Multiple Genes				
Pseudomonas aeruginosa	ATCC® BAA-2793™	Chile	2014	KPC-2, VIM-2
Klebsiella pneumoniae	CCRI-23061	Switzerland[2]	2015	OXA-232, NDM-1
KPC Isolates				
Klebsiella pneumoniae	NCTC 13438	N/A[1]	N/A[1]	KPC-3
Klebsiella pneumoniae	ATCC® BAA-1705™	USA	2007	KPC-2
Pseudomonas aeruginosa	CCRI-21587	Canada[2]	2011	KPC-2
Enterobacter cloacae	CCRI-21578	Canada	2011	KPC-4
Klebsiella oxytoca	CCRI-21581	Canada	2011	KPC-3
Klebsiella pneumoniae	CCRI-19587	Canada[2]	2009	KPC-3
Klebsiella pneumoniae	CCRI-19570	USA	2003	KPC-2
Klebsiella pneumoniae	CCUG 59413	Sweden	2010	KPC-3
Klebsiella pneumoniae	CCUG 59348	Norway	2010	KPC-2
Klebsiella pneumoniae	CCUG 56233	Sweden	2088	KPC-2
Escherichia coli	ATCC® BAA-2340™	USA (State Public Health laboratories)	N/A[1]	KPC
NDM Isolates				
Klebsiella pneumoniae	NCTC 13443	N/A[1]	N/A[1]	NDM-1
Klebsiella pneumoniae	ATCC® BAA-2146™	USA (State Public Health laboratories)	N/A[1]	NDM-1
Escherichia coli	CCRI-22255	France[2]	2013	NDM-1
Klebsiella pneumoniae	CCRI-21711	Canada	2011	NDM-1
Klebsiella pneumoniae	CCRI-22199	Canada	2012	NDM-1
Providencia rettgeri	CCRI-22257	France[2]	2013	NDM-1
Providencia stuartii	CCRI-22256	France[2]	2013	NDM-1
Klebsiella pneumoniae	CCRI-22254	France[2]	2013	NDM-4
Escherichia coli	CCRI-23064	Switzerland[2]	2015	NDM-5

Species	Collection Number	Geographical and Temporal Origin		Reported Resistance Gene and Variant (When Available)
Escherichia coli	CCRI-23464	Canada	2016	NDM-5
Escherichia coli	CCRI-23065	Switzerland[2]	2015	NDM-6
Escherichia coli	CCRI-23066	Switzerland[2]	2015	NDM-7
Enterobacter cloacae	ATCC® BAA-2468™	USA (State Public Health laboratories)	N/A[1]	NDM-1
Klebsiella pneumoniae	CCUG 60138	Sweden	2010	NDM-1
VIM Isolates				
Pseudomonas aeruginosa	NCTC 13437	N/A[1]	N/A[1]	VIM-10
Klebsiella pneumoniae	NCTC 13439	N/A[1]	N/A[1]	VIM-1
Klebsiella pneumoniae	NCTC 13440	N/A[1]	N/A[1]	VIM-1
Klebsiella pneumoniae	CCRI-19585	France	2009	VIM-1
Klebsiella pneumoniae	CCRI-22258	France[2]	2013	VIM-1
Pseudomonas aeruginosa	CCRI-21588	Canada[2]	2011	VIM-2
Serratia marcescens	CCRI-22261	France[2]	2013	VIM-2
Pseudomonas aeruginosa	CCRI-22720	Argentina	2014	VIM-2
Klebsiella pneumoniae	CCRI-22259	France[2]	2013	VIM-19
OXA-48-like Isolates				
Klebsiella pneumoniae	NCTC 13442	N/A[1]	N/A[1]	OXA-48
Klebsiella pneumoniae	CCRI-22263	France[2]	2013	OXA-48
Escherichia coli	CCRI-22265	France[2]	2013	OXA-48
Enterobacter cloacae	CCRI-22266	France[2]	2013	OXA-48
Klebsiella pneumoniae	CCRI-22264	France[2]	2013	OXA-181
Providencia rettgeri	CCRI-22267	France[2]	2013	OXA-181
Klebsiella pneumoniae	CCRI-23060	Switzerland[2]	2015	OXA-204
Citrobacter freundii	CCRI-23374	Canada	2016	OXA-204
Escherichia coli	ATCC® BAA-2523™	human clinical isolates	N/A[1]	OXA-48
Klebsiella pneumoniae	ATCC® BAA-2524™	human clinical isolates	N/A[1]	OXA-48
Klebsiella pneumoniae	CCUG 64452	Sweden	2013	OXA-48
IMP Isolates				
Escherichia coli	NCTC 13476	N/A[1]	N/A[1]	IMP-1
Acinetobacter baumannii	CCRI-19488	Canada[2]	2003	IMP-1
Klebsiella pneumoniae	CCRI-19569	Japan	2003	IMP-1
Klebsiella pneumoniae	CCRI-19582	Turkey	2009	IMP-1
Pseudomonas aeruginosa	CCRI-21589	Canada[2]	2011	IMP-1
Klebsiella pneumoniae	CCRI-19583	Taiwan	2009	IMP-4
Klebsiella pneumoniae	CCRI-19588	Taiwan	2009	IMP-4
Citrobacter youngae	CCRI-21591	Canada[2]	2011	IMP-4
Klebsiella pneumoniae	CCRI-19584	Taiwan	2009	IMP-8
Pseudomonas aeruginosa	CCRI-21590	China	2000	IMP-9
Serratia marcescens	CCRI-22262	France[2]	2013	IMP-11

[1] N/A = Not Available
[2] Isolation country

An *in silico* analysis was performed to assess inclusivity of the primers and probes of the GenePOC™ Carba assay. For each target, all variants documented in the National Center for Biotechnology Information (NCBI) nucleotide database were analyzed on November 7th, 2018. One representative sequence of each known variant was aligned. Variants for each target, predicted to be detected, are summarized in the table below.

GenePOC™ Carba Assay Target Variants Coverage

Targeted Resistance Genes	Tested with GenePOC™ Carba			*in silico* Prediction		
	# of Samples	Variants Detected	Variants Not Detected	Detectable[1]	Potentially Detectable[2]	Not Detectable
bla$_{KPC}$	12	2, 3, 4	None	2 to 38	None	N/A
bla$_{NDM}$	15	1, 4, 5, 6, 7	None	1 to 24	None	N/A
bla$_{IMP}$	11	1, 4, 8, 9, 11	None	1, 2, 4 to 6, 8 to 10, 13 to 20, 23 to 26, 28 to 30, 32, 33, 37, 38, 40, 42, 45, 47 to 49, 53 to 56, 59, 60, 62, 66, 69 to 72, 74 to 79	3, 7[3], 11[4], 21, 22, 27, 34, 41, 43, 44, 51, 52, 58, 61, 64, 67, 68, 73	12, 31, 35, 63
bla$_{OXA-48-like}$	12	48, 181, 204, 232	None	48, 162, 181, 199, 204, 232, 244, 245, 252[5], 370, 484, 505, 514[5], 515[5], 519, 546[5], 547[5], 566	None	54[5], 163[6], 247[6], 405[6], 416[5], 436, 438[6], 439[6], 517, 535[5], 538[5], 567
bla$_{VIM}$	10	1, 2, 10, 19	None	1 to 6, 8 to 12, 14 to 20, 23 to 46, 48 to 50, 52 to 55, 57, 59, 60	51, 56, 58	7, 13, 47

[1] Based on alignments with identity and query cover ≥ 95% and E-values < 0.01.

[2] Based on alignments presenting not more than two (2) nucleotides mismatches.

[3] A recombinant strain carrying a *bla*$_{IMP-7}$ gene was tested with GenePOC™ Carba, in complement to the *in silico* study, and its detection was confirmed.

[4] A clinical strain carrying a *bla*$_{IMP-11}$ gene was tested with GenePOC™ Carba in the analytical inclusivity study and its detection was confirmed.

[5] Variants only identified in the rare opportunistic human pathogens of the *Shewanella* genus.

[6] Variants presenting a deletion in their sequence resulting in no carbapenamase activity.

f. Analytical Specificity

The analytical specificity study was evaluated by testing 50 bacterial strains of *Enterobacteriaceae*, *Acinetobacter baumannii*, or *Pseudomonas aeruginosa* exhibiting non-susceptibility or susceptibility to the four (4) carbapenems (ertapenem, meropenem, imipenem, doripenem), but not carrying any of the resistance genes targeted by the GenePOC™ Carba assay (*bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$). Among the 50 strains which carried *bla*$_{CTX-M}$, *bla*$_{AmpC}$, *bla*$_{SHV}$, *bla*$_{SME}$, *bla*$_{TEM}$ and/or *bla*$_{SPM}$ β-lactamase resistance genes that are different from those targeted by the GenePOC™ Carba assay, only one (1) was observed to cross-react with the GenePOC™ Carba assay. The *Klebsiella pneumoniae* CCUG 59359 *bla*$_{TEM-52}$ strain yielded one positive result when tested at $1.13x10^7$ CFU/mL of SB. However, no cross-reactivity was observed for a 10-fold dilution of the same sample ($1.13x10^6$ CFU/mL of SB). All other strains analyzed yielded negative results.

Bacterial strains were characterized for their β-lactamase gene content by whole genome sequencing, except for the *Pseudomonas aeruginosa* C72 strain which was characterized using an SPM gene-specific PCR method.

Bacterial Strains Tested for Analytical Specificity with the GenePOC™ Carba Assay

Species (Species identified following sequencing, if different)	**Collection Number**	**β-Lactamase Gene(s) Identified (100% Coverage)**	**Resistance Phenotype (S/I/R)[1]**			
			ETP[1]	**MEM[1]**	**IMP[1]**	**DOR[1]**
Enterobacter cloacae	CCRI-3852	ACT-7	I	S	S	S
Enterobacter cloacae	CCRI-3854	ACT-4[2]	I	S	I	S
Proteus mirabilis	CCRI-21789	No β-Lactamase Gene Identified	S	S	I	S
Serratia marcescens	CCRI-21537	SRT-1	S	S	I	S
Acinetobacter baumannii	CCRI-1016	TEM-90, Mbl, BlaA2, OXA-65[3], Zn-dependent hydrolase	N/A[4]	S	S	S
Acinetobacter baumannii	CCRI-1017	TEM-206, SCO-1, Mbl, BlaA2, OXA-67[3], Zn-dependent hydrolase	N/A[4]	S	S	S
Enterobacter aerogenes (*Klebsiella aerogenes*)	CCRI-19495	SHV-5, AmpC	R	R	R	R
Enterobacter amnigenus (*Enterobacter cloacae*)	CCRI-22353	ACT-15	R	R	R	R
Enterobacter cloacae	CCRI-23473	No β-Lactamase Gene Identified	R	R	R	R
Enterobacter cloacae	CCRI-21536	ACT-5	R	R	R	R
Enterobacter cloacae	CCRI-21540	ACT-7	R	S	I	S
Enterobacter cloacae	CCRI-21603	ACT-7	R	R	R	R
Enterobacter cloacae	CCRI-21692	ACT-14	R	S	R	I

Species (Species identified following sequencing, if different)	**Collection Number**	**β-Lactamase Gene(s) Identified (100% Coverage)**	**Resistance Phenotype (S/I/R)[1]**			
			ETP[1]	**MEM[1]**	**IMP[1]**	**DOR[1]**
Enterobacter cloacae	CCRI-22075	ACT-7	R	R	R	R
Enterobacter cloacae	CCRI-22097	ACT-16	R	R	R	R
Enterobacter cloacae	CCRI-23318	CTX-M-15, TEM-206, CMH-1	R	R	R	R
Escherichia coli	CCRI-21970	AmpC1, AmpC2, MrdA, AmpH, CMY-44	R	I	R	I
Escherichia coli	CCUG 58541	CTX-M-14, TEM-104, MrdA, AmpC2, AmpH	R	S	S	S
Klebsiella pneumoniae	CCUG 58546	SHV-44, AmpH	R	S	S	S
Pseudomonas aeruginosa	CCRI-873	OXA-50[3]	N/A[4]	S	S	S
Pseudomonas aeruginosa	CCRI-1228	OXA-50[3]	N/A[4]	S	S	S
Pseudomonas aeruginosa	C72	SPM	N/A[4]	R	R	R
Serratia marcescens	CCRI-23334	SME-4, SRT-1	R	R	R	R
Enterobacter aerogenes	CCRI-3853	AmpC	S	S	S	S
Enterobacter aerogenes	CCRI-3879	AmpC	S	S	S	S
Escherichia coli	NCTC 13441	CTX-M-15, TEM-198, MrdA, OXA-1[3], AmpC2	S	S	S	S
Escherichia coli	CCRI-21710	AmpC2, MrdA, CTX-M-15, AmpH, OXA-1[3]	S	S	S	S
Escherichia coli	CCRI-778	AmpC2, MrdA	S	S	S	S
Escherichia coli	CCRI-779	TEM-206, AmpC1, MrdA, AmpC2, AmpH	S	S	S	S
Escherichia coli	CCRI-785	TEM-206, AmpC1, MrdA, AmpC2, AmpH	S	S	S	S
Escherichia coli	CCRI-878	AmpC2, MrdA, AmpH	S	S	S	S
Escherichia coli	CCUG 55970	CTX-M-9, AmpC2, TEM-206, MrdA, AmpC1, AmpH	S	S	S	S
Escherichia coli	CCUG 55971	CTX-M-15, TEM-143, AmpC2	S	S	S	S
Escherichia coli	CCUG 55972	CTX-M-2, AmpC1, AmpC2, AmpH	S	S	S	S
Escherichia coli	CCUG 58540	CTX-M-15, AmpC2, TEM-206, MrdA, AMPH, OXA-1[3]	S	S	S	S
Escherichia coli	CCUG 58542	CTX-M-15, MrdA, OXA-1[3], AmpC2	S	S	S	S
Klebsiella pneumoniae	NCTC 13465	SHV-85, TEM-206, AmpH	S	S	S	S
Klebsiella pneumoniae	CCUG 54718	CTX-M-15, TEM-33, OXA-1[3], AmpH	S	S	S	S
Klebsiella pneumoniae	CCUG 59358	SHV-14, OXA-1[3], LAP-2	S	S	S	S

Species (Species identified following sequencing, if different)	**Collection Number**	**β-Lactamase Gene(s) Identified (100% Coverage)**	**Resistance Phenotype (S/I/R)[1]**			
			ETP[1]	**MEM[1]**	**IMP[1]**	**DOR[1]**
Klebsiella pneumoniae	CCUG 59349	CTX-M-15, AmpH, OXA-1[3], TEM-105, SHV-11	S	S	S	S
Klebsiella pneumoniae	CCUG 59359	TEM-15, SHV-70, AmpH	S	S	S	S
Klebsiella pneumoniae	CCUG 59360	SHV-12, TEM-168, AmpH, OXA-9[3]	S	S	S	S
Klebsiella pneumoniae	CCRI-784	SHV-27, AmpH	S	S	S	S
Klebsiella pneumoniae (*Klebsiella quasipneumoniae*)	CCRI-806	OKP-B-11	S	S	S	S
Klebsiella pneumoniae	CCRI-1015	TEM-171, SCO-1, PER-2, OXA-9[3], SHV-39, AmpH	S	S	S	S
Proteus mirabilis	CCRI-825	TEM-33, CTX-M-2, OXA-2[3]	S	S	S	S
Proteus mirabilis	CCRI-826	TEM-215	S	S	S	S
Proteus mirabilis	CCRI-831	TEM-206, CTX-M-2, OXA-2[3]	S	S	S	S
Salmonella sp.	CCRI-8892	CTX-M-5, TEM-166, OXA-1[3]	S	S	S	S
Salmonella sp.	CCRI-8893	CTX-M-5, TEM-95, OXA-1[3]	S	S	S	S

[1] S/I/R: Sensitive/Intermediate/Resistant, ETP: Ertapenem, MEM: Meropenem, IMP: Imipenem, DOR: Doripenem

[2] Resistance gene identified with 99.9% homology.

[3] These *bla*$_{OXA}$ variants are not part of the *bla*$_{OXA\text{-}48\text{-}like}$ family, but are part of the Ambler class D.

[4] N/A: Not Applicable, *A. baumannii* and *P. aeruginosa* possess intrinsic resistance to ertapenem.

The cross-reactivity with primers and probes of the GenePOC™ Carba assay was evaluated by an *in silico* analysis performed on sequences contained in the National Center for Biotechnology Information (NCBI) database on September 6th, 2018. No other resistance gene than the assay target genes (i.e. *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA\text{-}48\text{-}like}$, and *bla*$_{IMP}$) was found to have significant level of homology with the primers and probes of the GenePOC™ Carba assay.

g. Assay Cut-off

Thresholds and cut-offs for the GenePOC™ Carba assay are embedded within the Assay Definition File that also encodes the instrument settings required to perform the test. The assay cut-offs were determined by testing a total of 599 samples.

h. Interfering Substances

The interfering substances study for the GenePOC™ Carba assay evaluated the potential interference effects of substances from culture media and/or saline (colony diluent) on GenePOC™ Carba assay performance. A total of 12 potentially interfering

combinations of solid media plates and sterile saline solutions including two (2) types of agar plates (i.e. Blood Agar Plates [BAP] and MacConkey Agar Plates [MAP]) each from three (3) manufacturers and sterile saline solutions from two (2) manufacturers were used during this study.

Five (5) positive bacterial strains harboring each of the resistance genes detected by the GenePOC™ Carba assay and one (1) negative bacterial carbapenem-non-susceptible strain that does not carry any of the resistance genes targeted by the assay were tested from standardized 0.5 McF suspensions.

List of Combinations of Agar Plates and Saline Solutions Tested with the GenePOC™ Carba

Combinations of Agar Plates and Saline Solutions (Manufacturer)
Colombia Blood Agar 5% (Hardy Diagnostics) / BBL™ Prepared Saline Solution (BD)
Colombia Blood Agar 5% (Hardy Diagnostics) / Saline 0.85% (Thermo Scientific™ Oxoid™)
Prepared Media BD BBL™ Columbia Agar with 5% Sheep Blood (BD) / BBL™ Prepared Saline Solution (BD)
Prepared Media BD BBL™ Columbia Agar with 5% Sheep Blood (BD) / Saline 0.85% (Thermo Scientific™ Oxoid™)
Columbia Agar with 5% Sheep Blood (bioMérieux) / BBL™ Prepared Saline Solution (BD)
Columbia Agar with 5% Sheep Blood (bioMérieux) / Saline 0.85% (Thermo Scientific™ Oxoid™)
MacConkey Agar (Hardy Diagnostics) / BBL™ Prepared Saline Solution (BD)
MacConkey Agar (Hardy Diagnostics) / Saline 0.85% (Thermo Scientific™ Oxoid™)
MacConkey Agar Medium (Thermo Scientific™ Remel™) / BBL™ Prepared Saline Solution (BD)
MacConkey Agar Medium (Thermo Scientific™ Remel™) / Saline 0.85% (Thermo Scientific™ Oxoid™)
MacConkey Agar (Thermo Scientific™ Oxoid™) / BBL™ Prepared Saline Solution (BD)
MacConkey Agar (Thermo Scientific™ Oxoid™) / Saline 0.85% (Thermo Scientific™ Oxoid™)

The interfering substances study demonstrated the compatibility of the GenePOC™ Carba assay with culture media and colony diluent from different manufacturers.

i. Carry-over and Cross-Contamination Studies

The purpose of the carry-over and cross-contamination study was to determine the carry-over rate of contamination in negative samples due to the nucleic acid amplification of high positive samples. *Klebsiella pneumoniae* CCUG 59348, a carbapenem-non-susceptible strain, which harbors the bla_{KPC} gene, was used to prepare positive samples. *Enterobacter cloacae* CCRI-22760, a representative carbapenem-non-susceptible strain that does not carry any of the carbapenemase resistance genes targeted by the assay, was used to prepare negative samples. The concentration of bacterial suspensions was standardized to 4 McF ($\geq 1.14 \times 10^7$ CFU/mL of SB), higher than the nominal concentration of 0.5 McFarland that is specified for use in the assay.

For the carry-over study, a run of eight (8) replicates of high positive samples followed by a run of eight (8) replicates of negative samples was performed by two (2) operators with the GenePOC™ Carba assay, for a total of ten (10) runs on one (1) revogene.

For the cross-contamination study, a total of ten (10) runs were performed by two (2) operators on one (1) revogene. Testing was performed with alternating high positive and negative samples.

No unexpected positive results for the *bla*$_{KPC}$ target were detected in negative samples tested in the carry-over study (n=40) nor in the cross-contamination study (n=40).

2. Comparison Studies

a. Method Comparison with predicate device

Refer to Section 3 Clinical Studies

b. Matrix Comparison

Not Applicable.

3. Clinical Studies

The clinical study was designed to assess the clinical performance of the GenePOC™ Carba assay for its use in the detection and differentiation of the *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$ gene sequences associated with carbapenem-non-susceptibility on isolated colonies. Three (3) clinical centers (one (1) in Canada and two (2) in the United States of America) participated in this clinical study as testing sites. The Reference Method for the study consisted of a high-performing FDA-cleared Nucleic Acid Amplification Test (NAAT) for the detection of the targeted carbapenemase genes from isolated colonies of known carbapenem non-susceptible organisms, the performance of which was established in comparison to PCR/bidirectional sequencing and which was used according to the manufacturer's instructions. All isolates in the study were tested with the Reference Method and with the GenePOC™ Carba assay. Organism susceptibility status (susceptible, intermediate, or resistant) to meropenem, ertapenem, doripenem, and/or imipenem was determined using CLSI standard test methods (M02, 13th Edition) and the interpretive criteria found in the FDA drug labels and CLSI M100 28th Edition.

For Reference Method testing, well-isolated colonies grown on blood agar plates were diluted to a standardized 0.5 McFarland (McF) suspension before testing. For GenePOC™ Carba assay testing, well-isolated colonies grown on each of the agar types were suspended to a standardized 0.5 McF suspension. If discordant results between the GenePOC™ Carba assay and Reference Method were observed, discrepant testing was performed using alternative PCR for each of the five (5) assay analytes followed by bi-directional sequencing on isolates from blood agar plates.

A total of 532 bacterial isolates (475 clinical stock isolates and 57 prospectively collected fresh isolates) were enrolled in the clinical study. Sixteen (16) isolates were excluded from the analysis of performance for the following reasons: eleven (11) did not meet the inclusion criteria for species identification (either they were not among the targeted organisms or organism identification was inconclusive), three (3) were unavailable for analysis due to laboratory error and two (2) were found to be susceptible to all four (4) carbapenems tested. Four (4) additional isolates were also excluded because they were associated with a Negative External Control failure on initial testing and produced Indeterminate results upon repeat. Thus, 512 compliant isolates remained in the final analysis.

A total of 512 fully compliant isolates were tested with both the Reference Method and the GenePOC™ Carba assay for the performance estimation study. The primary objective of the trial was to establish the performance characteristics of the GenePOC™ Carba assay for its use in determining the presence of *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$ gene DNA sequences in carbapenem-non-susceptible pure colonies of *Enterobacteriaceae*, *Acinetobacter baumannii*, or *Pseudomonas aeruginosa*, when grown on blood agar and MacConkey agar, and to establish the sensitivity and the specificity in comparison to the Reference Method.

External controls for the GenePOC™ Carba assay consisted of one (1) negative control and five (5) different positive controls each containing a single target analyte of the assay. The clinical sites ran a negative control and rotated testing one (1) of the positive controls on each day that study samples were tested (i.e. one specific positive control every five (5) days of testing). Study sample results were not valid until expected results were obtained for each control. External control data were compiled across all sites and overall QC results were acceptable.

Isolates grown on both blood agar and MacConkey agar types were evaluated with the GenePOC™ Carba assay for a total of 165 runs. Performance of the GenePOC™ Carba assay was assessed separately for each type of agar and resistance gene target relative to Reference Method. The study showed that 98.4% (1016/1032) of isolates yielded results on the first run. The initial non-reportable rates (combining Unresolved and Indeterminate rates), at the media level, were 1.7% (9/516) from blood agar, and 1.4% (7/516) from MacConkey agar. At the target level, from blood agar, the rates were 1.7% (9/516) for *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{IMP}$ and *bla*$_{OXA-48-like}$ gene targets and 1.6% (8/516) for *bla*$_{VIM}$ gene target, with an overall rate of 1.7% (9/516). From MacConkey agar, the non-reportable rates were 1.4% (7/516) across all targets. Out of the 16 isolates that initially yielded non-reportable results, eight (8) resulted in a non-reportable result upon repeat testing. The final non-reportable rates were 0.8% (4/516) across all media and gene targets.

Discrepant analysis was performed for every sample that produced discordant results between the GenePOC™ Carba assay and the Reference Method. The discrepant analysis comprised five (5) alternative PCR assays, one (1) for each resistance gene targeted by the GenePOC™ Carba assay, followed by bi-directional Sanger sequencing.

Of 31 isolates grown on blood agar with discrepant results, 21 were positive by the alternative PCR/bidirectional sequencing for the same target as the GenePOC Carba assay (see table footnotes below for details on all discrepancies). Seventeen (17) out of 19 samples with False Positive results for IMP target were found to be IMP positive by alternative PCR and bi-directional sequencing, including one (1) variant of IMP-4 (from Australia [2010]), 11 IMP-13/IMP-37 variants (one (1) from Argentina [2006], one (1) from North America [2014], and nine (9) from Europe [2005-2015]), one (1) variant of IMP-27/IMP-64 (from Canada [2017]), one (1) variant of IMP-15 (from Argentina [2004]), one (1) variant of IMP-16 (from Brazil [2004]), and two (2) variants of IMP-62 (from Argentina [2006]). The results of the discrepant analysis suggest that the GenePOC™ Carba assay and Reference Method may exhibit different coverage with regard to variants of IMP. Similar results were observed with colonies grown on MacConkey agar.

A total of 24 isolates contained more than one carbapenemase gene as determined by the GenePOC™ Carba assay. The reported carbapenemase gene content of 21 of these isolates was confirmed by the Reference Method. However, the Reference Method only detected one carbapenemase gene in the other three (3) isolates.

Performance by target and performance by organism group for the GenePOC™ Carba assay with isolates grown on blood agar is shown in the two (2) following tables.

Performance of GenePOC™ Carba Assay on Isolated Colonies Grown on Blood Agar Relative to Reference Method

Target	N	TP	FP	TN	FN	Sensitivity [95% CI]	Specificity [95% CI]
NDM	512	186	2[1]	322	2[6]	98.9% [96.2 - 99.7%]	99.4% [97.8 - 99.8%]
KPC	512	113	3[2]	395	1	99.1% [95.2 - 99.8%]	99.2% [97.8 - 99.7%]
OXA-48-like	512	65	3[3]	444	0	100.0% [94.4 - 100.0%]	99.3% [98.0 - 99.8%]
IMP	512	27	19[4]	466	0	100.0% [87.5 - 100.0%]	96.1% [94.0 - 97.5%]
VIM	512	52	1[5]	459	0	100.0% [93.1 - 100.0%]	99.8% [98.8 - 100.0%]

N: Number; TP: True Positive; FP: False Positive; TN: True Negative; FN: False Negative; CI: Confidence Interval

Note: Discrepant testing consisted of five (5) alternative PCR followed by bi-directional sequencing and was performed for every discrepant target result. The results from discrepant testing of 21/31 samples agreed with those of the GenePOC™ Carba assay. The notes below summarize the discrepant testing results for each target.

[1] One (1) out of two (2) was NDM-1 positive.

[2] Two (2) out of three (3) were KPC-3/KPC-38 positive.

[3] One (1) out of three (3) was OXA-48 positive. Investigation suggested an OXA cross-contamination at the step of sample preparation in one (1) out of three (3) isolates. Discrepant testing did not produce a sequence match with the OXA-48-like target.

[4] 17 out of 19 were found IMP positive including one (1) variant of IMP-4 (from Australia [2010]), 11 variants IMP-13/IMP-37 (one (1) from Argentina [2006], one (1) from North America [2014], and nine (9) from Europe [2005-2015]), one (1) variant of IMP-27/IMP-64 (from Canada [2017]), one (1) variant of IMP-15 (from Argentina [2004]), one (1) variant of IMP-16 (from Brazil [2004]), and two (2) variants of IMP-62 (from Argentina [2006]). The discrepant analysis pointed out potential differences in IMP variant coverage between the GenePOC™ Carba assay and the Reference Method. Investigation suggested an IMP cross-contamination at the step of sample preparation in two (2) out of 19 isolates for which discrepant testing did not produce a sequence match with the IMP target.

[5] Investigation suggested a VIM cross-contamination at the step of sample preparation. Discrepant testing did not produce a sequence match with the VIM target but produced a sequence match for the NDM target.

[6] Discrepant testing did produce a sequence match with the NDM-1 target in one (1) out of two (2) isolates and produced a sequence match for the OXA-48-like target in one (1) out of two (2) isolates. The OXA-48- positive isolate was classified as FP.

Performance of GenePOC™ Carba Assay by Organism Category and by Target Gene, for Isolated Colonies Grown on Blood Agar Relative to Reference Method

Organisms	Target	N	TP	FP	TN	FN	Sensitivity [95% CI]	Specificity [95% CI]
Entero-bacteriaceae	NDM	306	85	1	219	1	98.8% [93.7 - 99.8%]	99.5% [97.5 - 99.9%]
	KPC	306	112	3	190	1	99.1% [95.2 - 99.8%]	98.4% [95.5 - 99.5%]
	OXA-48-like	306	64	3	239	0	100.0% [94.3 - 100.0%]	98.8% [96.4 - 99.6%]
	IMP	306	14	5	287	0	100.0% [78.5 - 100.0%]	98.3% [96.1 - 99.3%]
	VIM	306	12	0	294	0	100.0% [75.8 - 100.0%]	100.0% [98.7 - 100.0%]
Pseudomonas aeruginosa	NDM	107	26	1	80	0	100.0% [87.1 - 100.0%]	98.8% [93.3 - 99.8%]
	KPC	107	0	0	107	0	-	100.0% [96.5 - 100.0%]
	OXA-48-like	107	1	0	106	0	100.0% [20.7 - 100.0%]	100.0% [96.5 - 100.0%]
	IMP	107	5	14	88	0	100.0% [56.6 - 100.0%]	86.3% [78.3 - 91.6%]
	VIM	107	39	0	68	0	100.0% [91.0 - 100.0%]	100.0% [94.7 - 100.0%]
Acinetobacter baumannii	NDM	99	75	0	23	1	98.7% [92.9 - 99.8%]	100.0% [85.7 - 100.0%]
	KPC	99	1	0	98	0	100.0% [20.7 - 100.0%]	100.0% [96.2 - 100.0%]
	OXA-48-like	99	0	0	99	0	-	100.0% [96.3 - 100.0%]
	IMP	99	8	0	91	0	100.0% [67.6 - 100.0%]	100.0% [96.0 - 100.0%]
	VIM	99	1	1	97	0	100.0% [20.7 - 100.0%]	99.0% [94.4 - 99.8%]

N: Number; TP: True Positive; FP: False Positive; TN: True Negative; FN: False Negative; CI: Confidence Interval
Multiple target results were observed for some isolates.

Performances with isolates grown on MacConkey agar are shown in the following two (2) tables.

Performance of GenePOC™ Carba Assay on Isolated Colonies Grown on MacConkey Agar Relative to Reference Method

Target	N	TP	FP	TN	FN	Sensitivity [95% CI]	Specificity [95% CI]
NDM	512	186	2[1]	322	2[6]	98.9% [96.2 - 99.7%]	99.4% [97.8 - 99.8%]
KPC	512	114	3[2]	395	0	100.0% [96.7 - 100.0%]	99.2% [97.8 - 99.7%]
OXA-48-like	512	65	2[3]	445	0	100.0% [94.4 - 100.0%]	99.6% [98.4 - 99.9%]
IMP	512	27	21[4]	464	0	100.0% [87.5 - 100.0%]	95.7% [93.5 - 97.2%]
VIM	512	52	1[5]	459	0	100.0% [93.1 - 100.0%]	99.8% [98.8 - 100.0%]

N: Number; TP: True Positive; FP: False Positive; TN: True Negative; FN: False Negative; CI: Confidence Interval

Note: Discrepant testing consisted of five alternative PCR followed by bi-directional sequencing and was performed for every discrepant target result. The results from discrepant testing of 21/31 samples agreed with those of the GenePOC™ Carba assay. The notes below summarize the discrepant testing results for each target.

[1] One (1) out of two (2) was NDM-1 positive.

[2] Two (2) out of three (3) were KPC-3/KPC-38 positive.

[3] One (1) out of two (2) was OXA-48 positive. Investigation suggested an OXA cross-contamination at the step of isolate preparation in one (1) out of two (2) isolates. Discrepant testing did not produce a sequence match with the OXA-48-like target.

[4] 17 out of 21 were found IMP positive including one (1) variant of IMP-4 (from Australia [2010]), 11 variants IMP-13/IMP-37 (one (1) from Argentina [2006], one (1) from North America [2014], and nine (9) from Europe [2005-2015]), one (1) variant of IMP-27/IMP-64 (from Canada [2017]), one (1) variant of IMP-15 (from Argentina [2004]), one (1) variant of IMP-16 (from Brazil [2004]), and two (2) variants of IMP-62 (from Argentina [2006]). The discrepant analysis pointed out potential differences in IMP variant coverage between the GenePOC™ Carba assay and the Reference Method. Investigation suggested an IMP cross-contamination at the step of isolate preparation in four (4) out of 21 isolates for which discrepant testing did not produce a sequence match with the IMP target.

[5] Investigation suggested a VIM cross-contamination at the step of sample preparation. Discrepant testing did not produce a sequence match with the VIM target but produced a sequence match for the KPC target.

[6] Discrepant testing did produce a sequence match with the NDM-1 target in one (1) out of two (2) isolates and produced a sequence match for the OXA target in one (1) out of two (2) isolates. The OXA-48 positive isolate was classified as FP.

Performance of GenePOC™ Carba Assay by Organism Category and by Target Gene, on Isolated Colonies Grown on MacConkey Agar Relative to Reference Method

Organisms	Target	N	TP	FP	TN	FN	Sensitivity [95% CI]	Specificity [95% CI]
Entero-bacteriaceae	NDM	306	85	1	219	1	98.8% [93.7 - 99.8%]	99.5% [97.5 - 99.9%]
	KPC	306	113	3	190	0	100.0% [96.7 - 100.0%]	98.4% [95.5 - 99.5%]
	OXA-48-like	306	64	2	240	0	100.0% [94.3 - 100.0%]	99.2% [97.0 - 99.8%]
	IMP	306	14	5	287	0	100.0% [78.5 - 100.0%]	98.3% [96.1 - 99.3%]
	VIM	306	12	1	293	0	100.0% [75.8 - 100.0%]	99.7% [98.1 - 99.9%]
Pseudomonas aeruginosa	NDM	107	26	1	80	0	100.0% [87.1 - 100.0%]	98.8% [93.3 - 99.8%]
	KPC	107	0	0	107	0	-	100.0% [96.5 - 100.0%]
	OXA-48-like	107	1	0	106	0	100.0% [20.7 - 100.0%]	100.0% [96.5 - 100.0%]
	IMP	107	5	14	88	0	100.0% [56.6 - 100.0%]	86.3% [78.3 - 91.6%]
	VIM	107	39	0	68	0	100.0% [91.0 - 100.0%]	100.0% [94.7 - 100.0%]
Acinetobacter baumannii	NDM	99	75	0	23	1	98.7% [92.9 - 99.8%]	100.0% [85.7 - 100.0%]
	KPC	99	1	0	98	0	100.0% [20.7 - 100.0%]	100.0% [96.2 - 100.0%]
	OXA-48-like	99	0	0	99	0	-	100.0% [96.3 - 100.0%]
	IMP	99	8	2	89	0	100.0% [67.6 - 100.0%]	97.8% [92.3 - 99.4%]
	VIM	99	1	0	98	0	100.0% [20.7 - 100.0%]	100.0% [96.2 - 100.0%]

N: Number; TP: True Positive; FP: False Positive; TN: True Negative; FN: False Negative; CI: Confidence Interval
Multiple target results were observed for some isolates.

4. Clinical Cut-off

Not Applicable.

5. Expected Values/Reference Range

A total of 512 isolates of *Enterobacteriaceae, Acinetobacter baumannii*, or *Pseudomonas aeruginosa* covering a large geographical and temporal diversity and reported to be carbapenem-non-susceptible based on conventional phenotypic (AST) methods have been tested with GenePOC™ Carba assay. After their growth on blood agar, 449 isolates were determined to have one (1) or more of the *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, and *bla*$_{IMP}$ gene targets by the GenePOC™ Carba assay. Results obtained from isolates grown on MacConkey agar plates were similar, with 449/512 carbapenem non-susceptible isolates positive for *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{OXA-48-like}$, or *bla*$_{IMP}$ gene targets.

O. INSTRUMENT NAME

revogene™

P. SYSTEM DESCRIPTIONS

1. Modes of Operation:

Real-time PCR with fluorogenic detection of amplified DNA.

2. Software:

FDA has reviewed applicant's Hazard Analysis and software development processes for this line of product types:

Yes ___X____ or No _____

3. Specimen Identification:

Barcodes are used to identify patient specimens. The GenePOC™ Carba assay's Sample Buffer Tube (SBT) and microfluidic cartridge (PIE) are both pre-labeled with a unique barcode to identify both the specimen and assay. The instrument has two barcode readers to identify reagents and patient specimens. It provides traceability of the sample ID to the PIE ID, SBT ID, and assay ID.

4. Specimen Sampling and Handling:

User intervention is required for preparing the standardized 0.5 McFarland (McF) bacterial suspension from characterized carbapenem-non-susceptible isolated colonies, inoculating the bacterial suspension into the Sample Buffer Tube (SBT), transferring the sample into the microfluidic cartridge (PIE), and loading/unloading the microfluidic cartridge into the revogene. All further sample handling is automated.

5. Calibration:

   The system is factory calibrated by the manufacturer. The calibration is verified annually. Upon the verification, maintenance is performed if required.

6. Quality Control:

   An Internal Process Control (PrC) is provided in each microfluidic cartridge (PIE) of the GenePOC™ Carba assay. The PrC is lysed, amplified, and detected along with each sample tested and verifies the efficacy of the DNA extraction and PCR amplification processes.

   Commercially available strains (NCTC 13476 for *bla*$_{\text{IMP-1}}$, CCUG 59348 for *bla*$_{\text{KPC-2}}$, ATCC® BAA-2146™ for *bla*$_{\text{NDM-1}}$, ATCC® BAA-2523™ for *bla*$_{\text{OXA-48}}$ and NCTC 13440 for *bla*$_{\text{VIM-1}}$) can be used as a Positive External Control. A carbapenem non-susceptible strain of *Enterobacteriaceae*, *A. baumannii* or *P. aeruginosa* that does not carry any of the resistance genes targeted by the GenePOC™ Carba assay can be used as a Negative External Control.

Q. OTHER SUPPORTIVE INSTRUMENT PERFORMANCE CHARACTERISTICS DATA NOT COVERED IN THE "PERFORMANCE CHARACTERISTICS" SECTION

Not applicable.

R. PROPOSED LABELING

The labeling is sufficient and satisfies the requirements of 21 CFR Part 809.10.

S. CONCLUSION

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