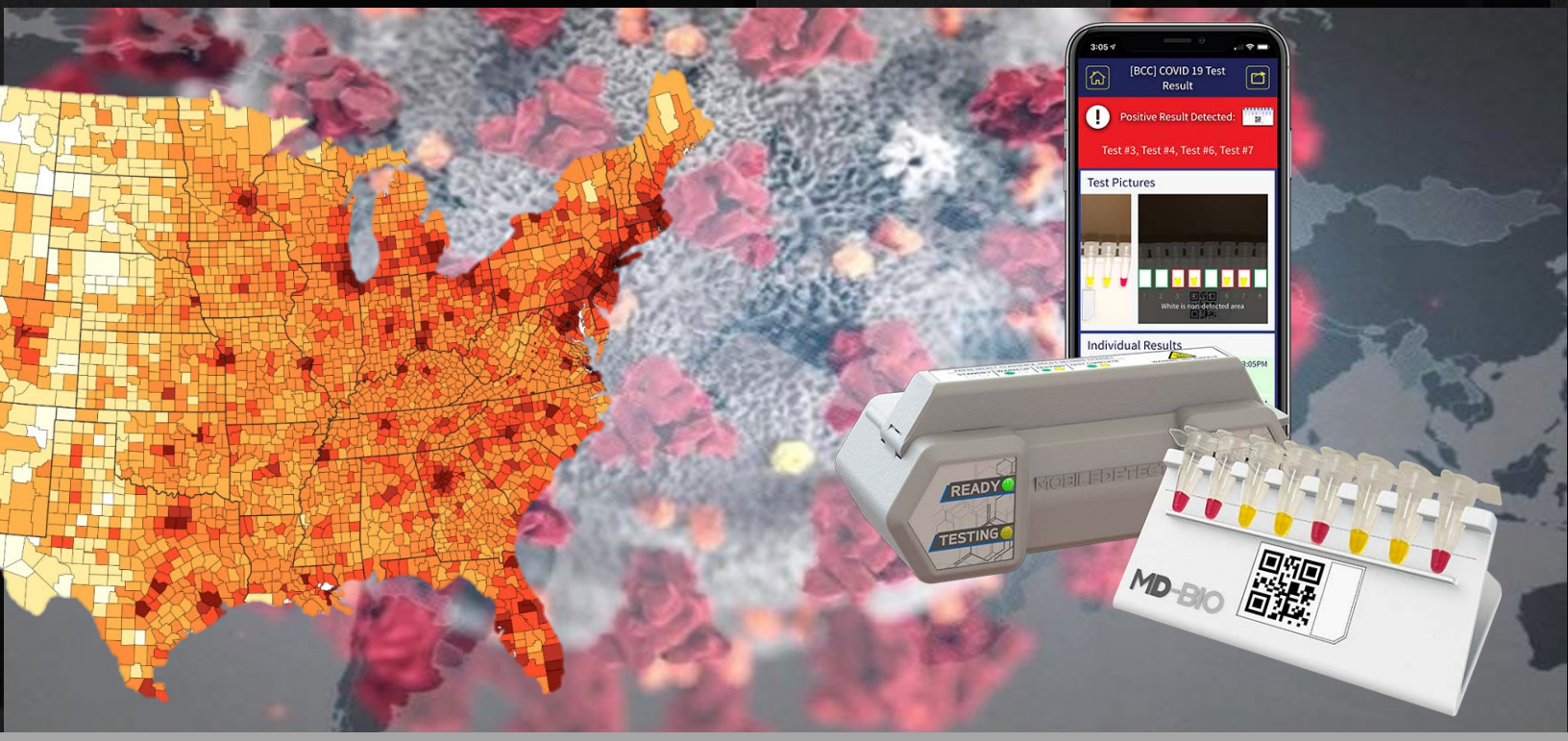
DETECTACHEM

MOBILEDETECT BIO

FDA EUA AUTHORIZED

BCC19 Test



COVID-19 SMARTPHONE TESTING KIT

The ground-breaking MobileDetect Bio BCC19 test kit combines laboratory-grade diagnostics with smartphone automated detection technology and rapid result reporting capabilities. This innovative solution enables high-capacity testing and actionable infection result data reporting anytime, anywhere.

Lab-Approved.

Accurate.

Fast.

Download on the App Store

ANDROID APP ON Google play

COVID19@DetectaChem.com | 1.844.433.8328 | www.DetectaChem.com/COVID19

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BCC19 Test

App Reporting

- -Instant Result Reports
- -Time, Date, Patient Info, Images, GPS and more
- -Send Reports via Email and Text

Mobile Field Labs

- -Complete Portable Lab Solution
- -96 or 960 Test Configurations
- -Run Up to 96 Tests at Once in 30 Minutes with with 96 Well Heater

MD-Bio Automated Analysis Station

MD-Bio Thermal Heater

Lab-Validated COVID-19 Detection Results in 8 Easy Steps

1. Use swab to sample interior of nasal cavity.
2. Apply swabbed nasal samples into buffer solution.
3. Apply buffered sample into MD-Bio cuvette.
4. Apply MD-Bio COVID-19 test assay to samples for reaction.
5. Heat reaction for 30 minutes with MD-Bio Thermal Heater.
6. Insert heated sample cuvette into Automated Analysis Station.
7. Scan QR code on Automated Analysis Station for infection results.
8. Instantly email or text results, images, notes, GPS information.

Technology	Chemical assay with color reaction
Sampling	Nasopharyngeal or Oralpharyngeal Swabbing
Detection Type	Active COVID-19 RNA or DNA strains
Operation	Bio sampling, buffer solution, assay exposure, heating, automated detection
Limit of Detection	<75 copies viral load
Reporting	Automated Detection and reporting with MobileDetect App on iOS and Android

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BCC19 TEST KITS

FDA EUA AUTHORIZED

Portable Lab-Quality COVID-19 Testing

DETECTACHEM

Background and Customers

- Founded in 2005
- Located in Houston, TX
- Supplies Explosive, Drug and Bio detection products to EVERY branch of DoD, DHS, DoJ and state and local agencies globally.
- All Products 100% Made in the USA.
- Massive production capabilities with fast delivery, 1M+ kits per week.

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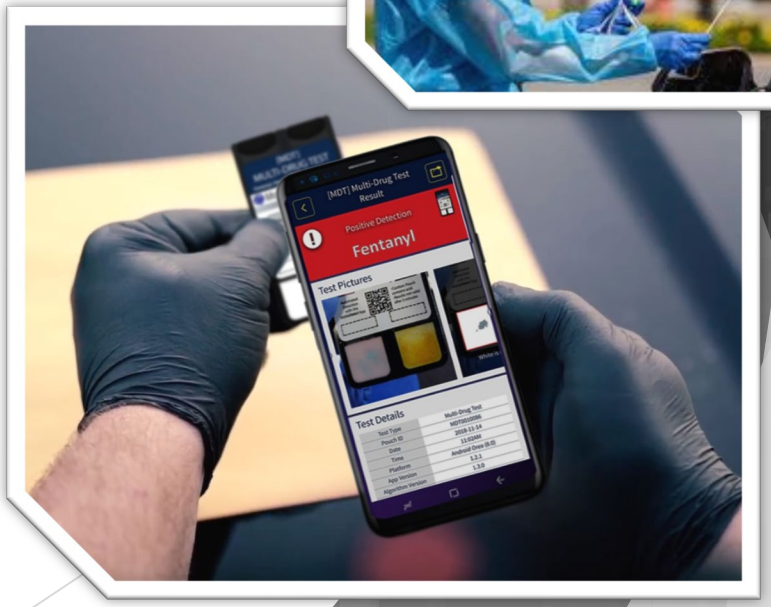
Just some of our customers…

- Army, Navy, Air Force, Marines, Coast Guard, SOCOM
- Customs & Border Protection, TSA, ICE, DEA, ATF, HSI
- NYPD, Port Authority, NJ, MA, LA, more
- NFL, NBA, Times Square, World Trade Center, Oculus, Boston Marathon, Ground Zero ceremonies, concerts, political venues, many more



COVID-19 DETECTION **SOLUTION**

MD-BIO BCC19

- **RT-LAMP technology with comparable results to laboratory standard RT-PCR.**
- **Only detects ACTIVE COVID-19 infections even if still asymptomatic.**
- **Only test kit to achieve 100% accuracy on FDA EUA submission testing. Report available.**
- **Uses commonly available nasal or oral swab.**
- **Run 8 to 96 tests at once in single 30 min cycle.**
- **Simple, easy-to-determine Yellow Positive or Red Negative results.**
- **FREE MobileDetect app for results with time, date, location, patient info, pics and more. Send PDF reports via email or text. (app use not required)**

Swab, Reagents, Heater, Analysis Stand

MobileDetect App

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HOW DOES THE BCC19 TEST WORK?

4 Simple Steps for Detection Results

1. Sample Collection

Nasal or oral swab and insert into UTM tube.

2. Add Reagents and Sample

Add reagents and sample into cuvette.

3. Thermal Heater

Heat up to 96 tests at once in 30 mins with portable thermal heater.

4. Analysis Stand

Insert tests into stand for automated analysis.

Result

YELLOW = POSITIVE

RED = NEGATIVE

Use App for Reporting (Optional)

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MD-BIO BCC19 **COMPARISON**

Closest Accuracy to Lab RT-PCR Testing

System	Sample Acquisition	Components	FDA Accuracy	Limit of Detection (viral copies)	Test Time	Specificity	Validated App and Reporting	Cost/Test
RT-PCR (Lab)	Nasal, Oral	12+	~99%	<5	1 per 24+ hrs	Molecular	No	$120
Serologic (Antibody)	Blood	6+	~60%	N/A	1 per 24+ hrs	Serological	No	$120
Lateral Flow Assay	Urine, Blood, Sweat, Other	6+	~70%	500	1 per 24+ hrs	Molecular	No	$100
Rapid Antigen	Bio material	8+	~70%	100	~15 mins each	Molecular	No	$90
BCC19 (RT-LAMP)	Nasal, Oral	3	97.6%	5	Up to 96 tests in 30 mins	Molecular	Yes	<$50

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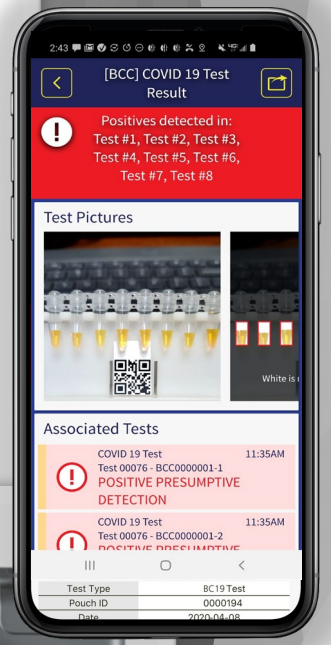
CONFIGURATIONS

MOBILE FIELD LABS

- **96** or **960** test capacity in a single portable case with everything needed for testing any time, any where.

REFILLS AND COMPONENTS

- **96** or **960** refill tests.
- **100** test positive control.
- Additional portable heaters: **8, 32, 96** test.
- Additional swabs, etc.



Large Volume Quantities in Stock and Ready for Immediate Shipping

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AVAILABLE:

- FDA Healthcare Provider Fact Sheet
- FDA Patient Fact Sheet
- FDA Instructions for Use
- FDA Letter of Authorization
- FDA Submission Testing Data, Cross Sensitivity, Etc.
- Patient Release Form
- Data Sheets, One-Pagers Promos

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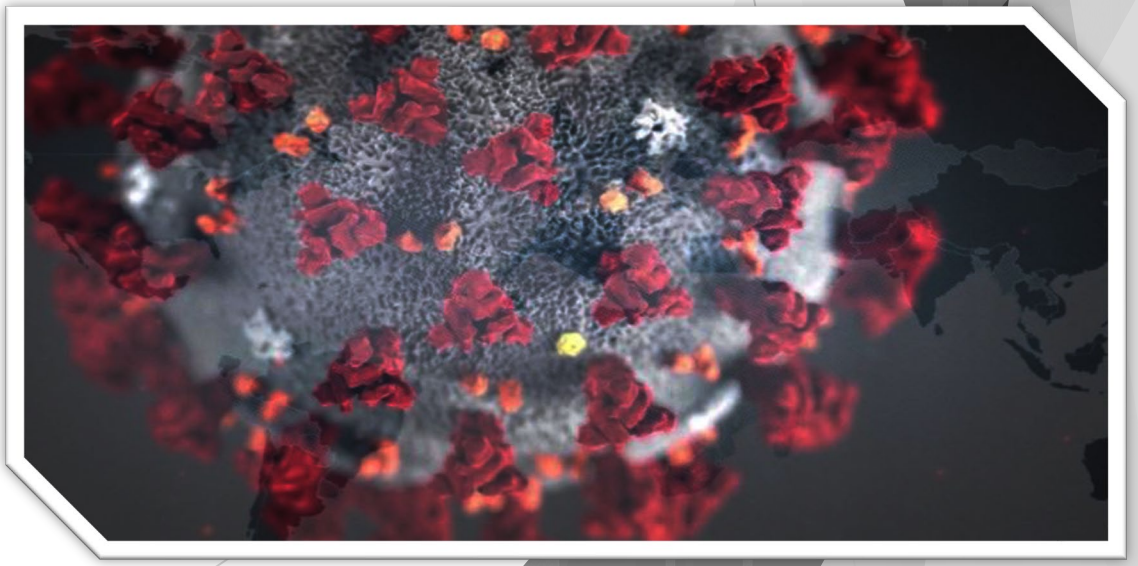

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U.S. FOOD & DRUG ADMINISTRATION

EMERGENCY USE AUTHORIZATION (EUA) SUMMARY FOR THE DETECTACHEM MOBILEDETECT BIO (MD-BIO) BCC19 TEST KIT

For *In vitro* Diagnostic Use
For use under Emergency Use Authorization (EUA) only

A. PURPOSE FOR SUBMISSION

Emergency Use Authorization (EUA) request for distribution and/or use of the Detectachem MobileDetect Bio (MD-Bio) BCC19 Test Kit for in vitro qualitative detection of RNA from the SARS-CoV-2 in nasopharyngeal, nasal, and oropharyngeal swabs from patients suspected of COVID-19. Additional testing and confirmation procedures should be performed in consultation with public health and/or other authorities to whom reporting is required. Positive results should also be reported in accordance with local, state, and federal regulations.

The MD-Bio BCC19 Test Kit is a simpler test than RT-PCR needing only a static-timed incubation cycle to detect the SARS-CoV-2 virus. This submission is for a kit that utilizes an isothermal incubation heat cycle. The test is designed to use the MD-Bio Heater along with various other Thermal Cyclers that are capable of performing this assay. The MD-Bio Heater is an easy-to-use and low-cost incubation solution that DetectaChem can manufacture in mass quantities to address the current pandemic diagnostic testing need.

B. MEASURAND

Specific nucleic acid sequences from the genome of the SARS-CoV-2 N-Gene and E-Gene sequence as shown below:

N-Gene
AAGGAAATTTTGGGGAGGAACTAATCAGACAAGGAACTGATTACAAGGCCGCAAAT
TGCACAATGCCCCCAGCGCTTCAGCGTTCTAATGTCGCGCATTGGCATGGAAGTCAC
ACCTTCGGGTGGTTGACCTACATGCCATCAAATTGGATGACAAAGATCCAAATTTCA
AAGATCAAGTCATTTTGCTGAATAAGCATATTGACGCAT

E-Gene
ATGTACTCATTCGTTTCGGAAGAGACAGGTACGTTAATAGTTAATAGCGTACTTCTT
TTTCTTGCGTGGTACTTGCTAGTTACACTAGCCATCACTGCGCTTCGATTGTGTGCGT
ACTGCTGCATGTTAACGTGAGTCTTGTAAAACCTTCTTTTTACGTTTACTCGTGTTAA
AAATCTGAATTCTTCTAGAGTTCCTGATCTTCTGGTCTAA

C. APPLICANT

Detectachem Inc
4100 Greenbriar Dr., Suite 180
Stafford TX, 77477
DUNS #: 785979662, CAGE code: 4KC19
Phone Number: 7132987400
Email: tkisner@detectachem.com

D. PROPRIETARY AND ESTABLISHED NAMES

Proprietary Name - MobileDetect Bio BCC19 Test Kit
Proprietary Name – MD-Bio BCC19
Established Name - MobileDetect Bio BCC19 Test Kit
Established Name – MD-Bio BCC19

E. PROPOSED INTENDED USE

1) Intended Use:

The MobileDetect Bio BCC19 Test Kit is an RT-LAMP (reverse transcriptase loop-mediated isothermal amplification) test intended for the qualitative detection of RNA from the SARS-CoV-2 in nasopharyngeal, nasal, and oropharyngeal swab specimens from individuals suspected of COVID-19 by their healthcare provider. Testing is limited to laboratories - certified under the Clinical Laboratory Improvement Amendments of 1988 (CLIA), 42 U.S.C. §263a, to perform high complexity tests, or by similarly qualified non-U.S. laboratories.

Results are for the identification of SARS-CoV-2 RNA. The SARS-CoV-2 RNA is generally detectable in upper respiratory specimens during the acute phase of infection. Positive results are indicative of the presence of SARS-CoV-2 RNA; clinical correlation with patient history and other diagnostic information is necessary to determine patient infection status. Positive results do not rule out bacterial infection or co-infection with other viruses. The agent detected may not be the definite cause of disease. Laboratories within the United States and its territories are required to report all positive results to the appropriate public health authorities.

Negative results do not preclude SARS-CoV-2 infection and should not be used as the sole basis for patient management decisions. Negative results must be combined with clinical observations, patient history, and epidemiological information.

The MobileDetect Bio BCC19 Test Kit is intended for use by trained clinical laboratory personnel specifically instructed and trained in the techniques of in vitro diagnostic procedures. The

MobileDetect Bio BCC19 Test Kit is only for use under the Food and Drug Administration's Emergency Use Authorization.

2) Special Conditions for Use Statements:

For Emergency Use Authorization (EUA) only
For in vitro diagnostic use only

3) Special Instrument Requirements:

The Detectachem MD-Bio BCC19 Test Kit test is to be used with a MD-Bio Heater or Thermal Cycler (from the approved list), MD-Bio Analysis Station, and (optional) MobileDetect Smartphone App as displayed in Figure 1.

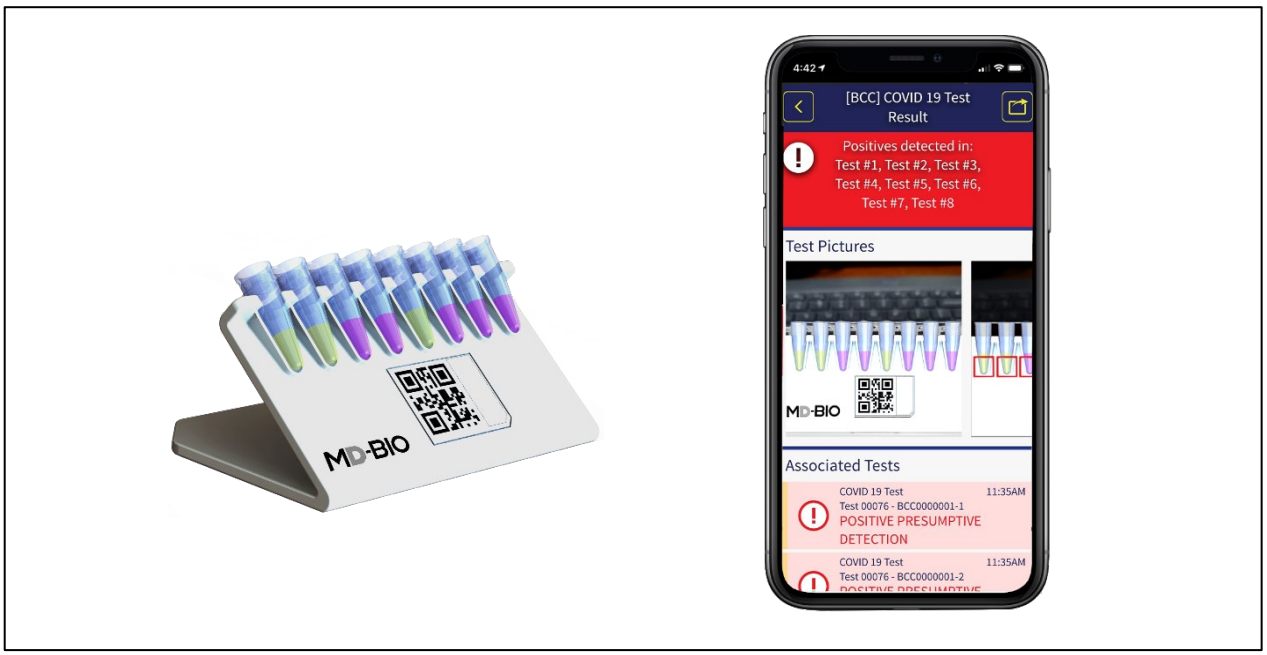

Figure 1: MD-Bio Analysis Station and Smart Phone Detection

G. DEVICE DESCRIPTION AND TEST PRINCIPLE

1) Product Overview/Test Principle:

The MD-Bio BCC19 Test utilizes a loop mediated isothermal amplification polymerase reaction (LAMP) test intended for the detection of SAR-CoV-2 viral RNA. The test uses a set of specific primers designed to uniquely detect SARS-CoV-2 RNA.

The MD-Bio BCC19 Test utilizes an isothermal nucleic acid amplification technique wherein DNA amplification is carried out at a constant temperature of 65°C. The assay/test uses a set of primers, and a DNA polymerase with high strand

displacement and replication activity. The assay primers are targeting the SARS-CoV-2 N and E gene, as seen in measurand. Also incorporated is a reverse transcriptase (RT) polymerase, which creates complimentary cDNA from RNA, and is in turn amplified by the DNA polymerase. These two polymerases work in tandem enabling the detection of viral DNA and RNA in the same reaction. The amplified DNA byproducts drive the pH of the reaction down to the point where it pushes the pH sensitive dyes to turn yellow, from their starting red color. Reactions displaying a visual color shift indicate that the target sequence is present.

The primer sequences used in the test are included in Table 1 and Table 2 below.

Table 1: N-Gene Primer Names and Their Respective Sequence

Primer Name	Sequence
N-F3	[redacted]
N-B3	[redacted]
N-FIP	[redacted]
N-BIP	[redacted]
N-Loop-F	[redacted]
N-Loop-B	[redacted]

Table 2: E-Gene Primer Names and Their Respective Sequence

Primer Name	Sequence
E-F3	[redacted]
E-B3	[redacted]
E-FIP	[redacted]
E-BIP	[redacted]
E-Loop-F	[redacted]
E-Loop-B	[redacted]

2) ***Description of Test Steps:***

NOTE: Every test set should include a positive and a negative control. All tests from the same set of Reagents can run a single positive and negative control per heat test cycle.

Steps to run the MD-Bio BCC19 Test:

1. **Sample Preparation:** A nasopharyngeal swab is used to collect the upper respiratory specimen from the patient. This swab is then placed in Viral Transport Media (VTM). 1µL of this solution is used as the analyte in the reaction.

2. Reaction Preparation: [redacted] of Reagent 1, and [redacted] of Reagent 2 is pipetted into each of the 8 wells of the provided MD-Bio PCR Strip. Analyte sample will come from viral transport media containing one of the clinical matrices with patient samples mentioned above. Each well of the PCR strip will receive [redacted] of its analyte sample for investigation.
3. Heat Cycle: Place the prepared PCR Strip in the MD-Bio heater (or approved Thermal Cycler). For the MD-Bio heater, momentarily push the select button for the incubation heat cycle to begin. For an approved Thermal Cycler, set the well plate temperature to [redacted] and the lid temperature to [redacted]. Set the incubation time to 30 minutes, set solution volume to [redacted], and incubate the prepared MD-Bio PCR Strip.
4. Cool Down: Place the PCR Strip in the MD-Bio Analysis Station and allow it to cool for approximately 1 minute before color interpretation. This will allow the chemistry to come to thermal equilibrium and more clearly show the yellow positive and red negative results.
5. Results Interpretation: If user is analyzing manually, use eyesight color recognition per the user manual to identify whether the result is positive or negative based on color change. Optionally, the MobileDetect Smartphone App can interpret the color change of each of the 8 wells in the PCR Strip. After Cool Down, open the MobileDetect Smartphone App and scan the QR code on the Analysis Station. The MobileDetect App will automatically analyze each of the PCR Strip wells and provide positive or presumptive negative results. After the analysis, a report is generated that can be exported as desired.

Control Material(s) to be Used with MobileDetect Bio BCC19 Test Kit:

Controls that will be provided with the test kit include:

a) A negative control is needed to validate that the chemistry has not been contaminated. Frequency of suggested use is to run a negative control with every 8 strip PCR tube or cluster of PCR tubes run in the same heat cycle used.
   i) A negative control is [redacted] of virgin VTM or [redacted] of nuclease free water.

b) A positive template control is needed to validate that the chemistry is active. Frequency of suggested use is to run a positive control with every 8 tube PCR strip or cluster of PCR tubes run in the same heat cycle used.
   i) A positive control is [redacted] copies of SARS-CoV-2 RNA using [redacted] of [redacted] copies/µL, which is 5x the LoD of the assay.
   ii) Positive Controls that are required but not provided with the test kit include:
      1. [redacted] Synthetic SARS-CoV-2 RNA Control 1 [redacted]

H. INTERPRETATION OF RESULTS

All test controls should be examined prior to interpretation of patient results. If the controls are not valid, test controls should be remedied, per procedures, before proceeding with patient testing.

Results can be interpreted by the operator by using the guide below.

1) ***MobileDetect Bio BCC19 Test Kit Controls – Positive, Negative and Internal***

   The MD-Bio BCC19 Test Kit should include a single positive and negative control on every PCR strip to determine if the test is working appropriately. Positive controls should turn yellow and alarm as positive, negative controls should remain red and present the result as negative. An example of what these controls look like are displayed in Figure 2.

Figure 2: Negative Control (Left) and Positive Control (Right)

   In the event of an invalid negative control, the lab should redouble its sterilization procedures, use virgin pipettes and filtered tips, and use new chemistry. In the event of an invalid positive control, the lab should verify use of stable positive control material, as well as verify the expiration date of the reagents.

2) ***Examination and Interpretation of Patient Specimen Results:***

   Assessment of clinical specimen test results should be performed after the positive and negative controls have been examined and determined to be valid and acceptable. If the controls are not valid, the patient results cannot be interpreted.

   Manual – Use eyesight color recognition, per the user manual, to identify whether the result is positive or negative based on color change. See Figure 3 below.

Figure 3: Example of Test Results (4 Left Negative, 4 Right Positive)

Automated – Optional

The MobileDetect App is an optional capability of the MD-Bio product, which can be used in addition to manual color determination of the result. The App uses the Apple or Android device's optics (camera) to read the color in a calibrated relationship. The MD-Bio BCC19 Test Kit detection algorithm on the MobileDetect App works by first registering the location of the QR code on the MD-Bio Analysis Station. The algorithm uses the QR code location as a reference point to draw a region around each of the 8 wells of the PCR Strip. A white balance calibration measurement is taken from a white box directly to the side of the QR code on the MD-Bio Analysis Station. This calibration area serves to baseline the color spectrum that may differ due to variation in lighting and smartphones. The HSV (Hue, Saturation, Value) color value for each pixel in the identified boxes is then analyzed. If the reaction creates a positive color indication for a "Positive" sample, the algorithm will identify that well/box as positive. Detectachem is the industry leader in Automated Colorimetric Recognition on various platforms and has been deploying Automated Colorimetric Detection equipment for the past 15 years.

After performing the result interpretation, the app generates an individual result for each well in the PCR Strip. These results can be saved as a document and printed or exported, along with their image data, as desired by the user. Figure 4 below shows examples of positive and presumptive negative individual tests. Negative results should not be used as the sole basis for patient management decisions. Negative results must be combined with clinical observations, patient history, and epidemiological information.

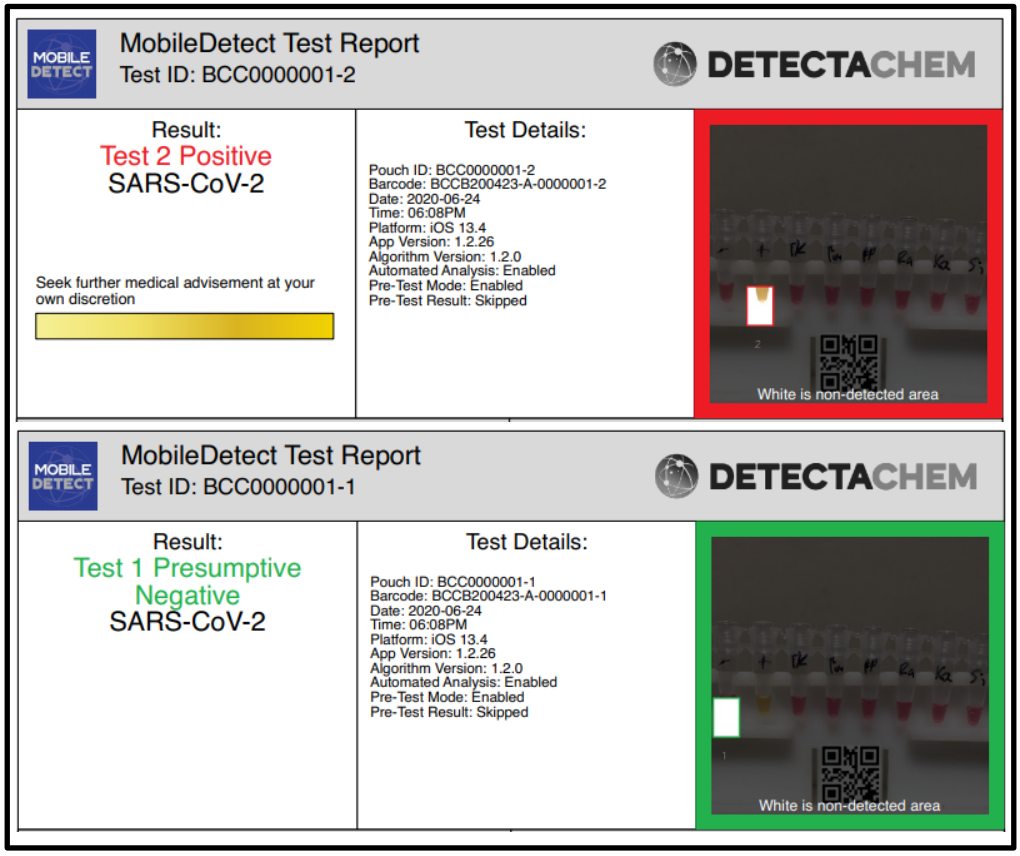

Figure 2: MobileDetect Detection Screen for Positive (Top) and Negative (Bottom) Test

I. PRODUCT MANUFACTURING

The Detectachem MD-Bio BCC19 Test Kit has been validated using only the components referenced in this submission and shall not be changed without prior concurrence from the FDA.

1) Overview of Manufacturing and Distribution:

The product will be manufactured at Detectachem Incorporated, by Detectachem Incorporated personnel consistent with practices for the production of medical diagnostic devices based on the Detectachem Medical Device Quality Management System. Sub components of reagents are provided and manufactured by [redacted]. Raw components come from accredited vendors such as [redacted]. No products involved in the manufacturing are procured from unknown manufacturers.

The current manufacturing capabilities include the ability to manufacture approximately 700,000 products per week, however in the event of a surge in demand this could be increased to 2,100,000 product per week within a month timeframe.

The product will be distributed by Detectachem Incorporated.

2) ***Components Included with the Test***

Components manufactured by Detectachem Incorporated and supplied with the test are displayed in Table 2. These items include:

- 12 MD-Bio PCR Strips
- 1 MD-Bio Heater
- 1 MD-Bio Preparation Station
- 1 1.2mL vial of BCC19 Reagent 1
- 1 1.2mL vial of BCC19 Reagent 2
- 1 MD-Bio Analysis Station
- Optional: 1 100uL vial of BCC19 Positive Control

BCC19 Reagent Composition:

BCC19 Reagent 1: Colorimetric LAMP Master Mix for DNA & RNA

- [redacted] Colorimetric LAMP 2X Master Mix (DNA&RNA) Product Number: [redacted]

BCC19 Reagent 2: Primer/Oligonucleotide Mix in Nuclease Free Water

- Oligonucleotides synthesized by [redacted].
  - N-FIP Primer, Final Concentration [redacted]
  - N-BIP Primer, Final Concentration [redacted]
  - N-F3 Primer, Final Concentration [redacted]
  - N-B3 Primer, Final Concentration [redacted]
  - N-LOOP F Primer, Final Concentration [redacted]
  - N-LOOP B Primer, Final Concentration [redacted]
  - E-FIP Primer, Final Concentration [redacted]
  - E-BIP Primer, Final Concentration [redacted]
  - E-F3 Primer, Final Concentration [redacted]
  - E-B3 Primer, Final Concentration [redacted]
  - E-LOOP F Primer, Final Concentration [redacted]
  - E-LOOP B Primer, Final Concentration [redacted]
- [redacted] Nuclease Free Water, Product Number: [redacted]

BCC19 Positive Control:

- 0.125µL [redacted], Synthetic SARS-CoV-2 RNA Control 1 [redacted]
- 99.875µL [redacted] Nuclease Free Water, Product Number: [redacted]

Table 2: Products Included with the MD-Bio Test Kit

Image	Quantity	Description
	12	**MD-Bio PCR Strip** 8 reactions each
	1	**MD-Bio Heater**
	1	**1.2 mL BCC19 Reagent 1** 96 Rxn Total 12.5 µL/Rxn
	1	**1.2 mL BCC19 Reagent 2** 96 Rxn Total 11.5 µL/Rxn
	1	**MD-Bio Analysis Station**
	1	**MD-Bio Preparation Station**
	1	**Optional: 100 µL BCC19 Positive Control** 100 Rxn Total 1 µL/Rxn

3) ***Optional Components Not Included with the Test***

Components that are optional but not included with the test:

1. Smartphone - (Android or Apple based Smart Devices displayed in Table 3)
2. MobileDetect App.
3. If not purchased as part of the MD-Bio BCC19 Testing Kit, a lab must maintain a Positive Control Sample. Examples of known positive samples are shown below. Positive Control samples should be diluted to [redacted].
    a. [redacted], Synthetic SARS-CoV-2 RNA Control 1 [redacted]

Table 3: Hardware and Software Requirements to Use the Mobile Detect App

Operating System	Architecture	Hardware Requirements	Phones Tested
iOS 12 or later	Arm64	Memory: 1GB+ Processor: 1.3GHz+ Storage 16GB+	iPhone 6s/6s+ 7/7+ 8/8+ 10/X/XR/XS/MAX 11/Pro/Pro MAX
Android 6.0 or later	Armeabi, Armeabi-7a, Arm64-v8	Memory: 4GB+ Processor: 1.6GHz+ Storage 32GB+	Samsung Galaxy S7/Edge/Active S8/Plus/Active S9/ Plus S10/Plus/5G/E/Lite S20/Plus/Ultra

4) ***Testing Capabilities***

Total time required to perform one 8 strip test: 41 minutes (6:50 minutes per patient sample). One MD-Bio Heater instrument can process 90+ samples in an 8-hour period.

1. Reagent preparation of one 8 strip: 3 minutes
2. Addition of patient sample and controls: 3 minutes
3. Heat cycle: 33 minutes
4. Cool down: 1 minute
5. Results interpretation: 1 minute

5) ***Reagent Stability:***

BCC19 Reagent 1: Stable for at least 2 years when stored at -20°C

BCC19 Reagent 2: Stable for at least 2 years when stored at -20°C

BCC19 Positive Control: Stable for up to 2 years when stored at -80°C

Future Stability Plan:

i) **Unopened kit - BCC19 Reagent 1 & 2 and Positive Control:**
The stability test will use reagent from 3 different lots. The reagent will be aliquoted into 50 tests per lot. The reagent will remain unopened and stored at -20°C. Every 3 months for the next 27 months, 5 tests per lot will be performed on the stored chemicals. One set of 5 tests from each of the lots will include 2 control negatives and 3 positive contrived samples at 5x LoD. The first test will take place within one week of prepping the aliquoted samples. The remaining tests will take place in 90-day increments from the initial run.

ii) **Shipping Stability:**
All shipments will be overnight shipments for every reagent. All reagents will be shipped with cold packs to keep the temperature of the product between 2-8°C. Testing will include shipping the reagents in cold packs to a designated location and return to testing facility. The received product will then be used in 20 tests at LoD.

iii) **BCC19 Positive Control:**
The stability test will use positive control from 3 different lots. The positive control will be aliquoted into 40 tests per lot. One set of 5 test from each of the lots will include 2 negative controls and 3 positive controls. The first test will take place at test initiation. The test will be repeated every hour for the next 9 hours. Every test will use BCC19 Reagents 1 & 2 from the same aliquot.

iv) **Freeze-Thaw Stability:**
The stability test will use BCC19 Reagent 1 & 2 and BCC19 Positive Control from 3 different lots. The reagent will be aliquoted into 64 test sets, and the positive control will be aliquoted into 32 test sets. All of the test sets will be frozen and thawed 16 times. A test will take place after each freeze-thaw cycle. One set of 4 tests will include two positive controls from the freeze-thaw stability aliquot, one positive control that has been thawed once, and one control negative test. All tests will occur within 3 days of the previous freeze-thaw cycle.

J. PERFORMANCE EVALUATION

1) ***Limit of Detection (LoD) - Analytical Sensitivity:***

A Limit of Detection Determination test series was conducted to determine the lower limit of detection using the MD-Bio Heater. The positive control used to determine LoD was [redacted], Synthetic SARS-CoV-2 RNA Control 1 [redacted] which contains the entire N-gene. This analyte was spiked into Viral Transport Media (VTM): 500 copies/µL, 375 copies/µL, 250 copies/µL, 125 copies/µL, 100 copies/µL, 75 copies/µL, 50 copies/µL, ands 25 copies/µL. Then 1µL of each of the contrived analytes was used to yield the total number of copies per reaction below. All measurements were performed using 3 individual samples. The conclusion of the LoD determination test series is that 75 copies/µL is the LoD for this assay. This value was the lowest analyte value attempted that correctly agreed with all 3 samples yielding positive results. For concentrations near the LoD, 20 additional samples were performed to determine the concentration at which at least 95% of the samples (19/20) were positive. All results from the LoD determination test series can be viewed in Table 4.

Table 4: LoD Testing for MD-Bio Reagents

Concentration (copies/µL)	Positive Samples Detected
500	3/3
375	3/3
250	3/3
125	3/3
100	20/20
75	20/20
50	14/20
25	9/20

2) ***Inclusivity (analytical sensitivity):***

The assay targets specific genomic regions of the SARS-CoV-2 nucleocapsid (N) gene and small envelope (E) gene. Inclusivity was demonstrated under the original EUA by mapping the primers and probes to the complete SARS-CoV-2 genomes that were available in GISAID (Global Initiative on Sharing All Influenza Data) database as of June 26, 2020. For all primers, there was 100% with an overwhelming majority of COVID-19 strains, equating to a 97.22% match rate for the N-gene, and 98.65% match rate for the E-gene. The rate for just 1 mismatch was 2.6% for the N-gene, and 1.3% for the E-gene, and there were no mismatches of more than 3. Previous work on MERS-CoV has demonstrated that a single nucleotide mismatch in one primer set had a negligible impact on the detection using LAMP assays (PMID 25103205). This data is displayed in Table 5.

Table 5: In silico Inclusivity Analysis

	N-gene	E-gene
Total Primer Length (nt)	171	174
Total # of Strains Evaluated	5557	7454
100% Match	97.22%	98.65%
1 Mismatch	142	98
2 Mismatches	9	2
3 Mismatches	2	1
>3 Mismatches	0	0

3) ***Cross-reactivity (Analytical Specificity):***

In silico cross-reactivity analysis was performed by aligning the N-gene and E-gene LAMP primer sequences against sequences of coronaviruses related to SARS-Cov-2, as well as common viruses. See Table 6 and Table 7 below for the organisms assessed *in silico* for potential cross-reactivity to the MobileDetect Bio (MD-Bio) BCC19 Test Kit.

Table 6: N Primer Cross Reactivity

Organism Name	Organism ID	N_B3 25 bp	N_BIP 40 bp	N_F3 21 bp	N_FIP 42 bp	N_LB 20 bp	N_LF 21 bp
SARS coronavirus 2	NC_045512.2	**100 %**	53%	**100 %**	48%-55%	**100 %**	**100 %**
SARS coronavirus	NC_004718	64%	53%	**90%**	55%	**85%**	**90%**
Human coronavirus NL63	NC_005831.2	0%	0%	0%	0%	0%	0%
Human coronavirus HKU1	NC_006577.2	0%	0%	0%	0%	0%	0%
Human coronavirus OC43 strain ATCC VR-759	NC006213.1	0%	0%	0%	0%	0%	0%
Pneumocystis jirovecii RU7 chromosome Unknown supercont1.8	NW_017264782.1	64%	35%				
Streptococcus pneumoniae NCTC7465 chromosome: 1	NZ_LN831051.1	60%-80%		**95%**		75%	62%-71%
Rothia mucilaginosa DY-18 DNA	NC_013715.1				40%	**75%-90%**	
Chlamydia pneumonia TW-183	NC_005043.1	56%-76%		76%	36%	65%	
Staphylococcus epidermidis ATCC 12228	NC_004461.1	60%-68%		62%			62%
Candida albicans SC5314 chromosome 1 sequence	NC_032089.1	60%-72%	35%-45%	71%-76%			62%-76%
Legionella pneumophila subsp. pascullei strain NCTC12273, chromosome: 1	NZ_LR134380.1	60%-76%	43%		36%-55%		62%-76%
Streptococcus pyogenes NCTC8198, chromosome: 1	NZ_LN831034.1	60%-64%	43%-50%				
Bordetella pertussis 18323	NC_018518.1		35%			65%-80%	**81%**

Haemophilus influenzae NCTC8143, chromosome: 1	NZ_LN831035.1	60%-68%	50%		43%		62%-67%
Mycoplasma pneumoniae FH chromosome	NZ_CP010546.1	60%				65%-75%	76%
Mycobacterium tuberculosis H37Rv	NC_000962.3						67%
Pseudomonas aeruginosa PAO1	NC_002516.2		35%			75%	
Human parainfluenza virus 1	NC_003461.1			71%			

* All above blank cells are <50% and are considered negligible

Table 7: E Primer Cross Reactivity

Organism Name	Organism ID	E_B3 22 bp	E_BIP 44 bp	E_F3 23 bp	E_FIP 42 bp	E_LB 19 bp	E_LF 18 bp
SARS coronavirus 2	NC_045512.2	**100 %**	45%-57%	**100 %**	45%-62%	**100 %**	**100 %**
SARS coronavirus	NC_004718	**95%**	55%	**100%**	43%-62%	**100%**	**100%**
Human coronavirus NL63	NC_005831.2	0%	0%	0%	0%	0%	0%
Human coronavirus HKU1	NC_006577.2	0%	0%	0%	0%	0%	0%
Human coronavirus OC43 strain ATCC VR-759	NC006213.1	0%	0%	0%	0%	0%	0%
Pneumocystis jirovecii RU7 chromosome Unknown supercont1.8	NW_017264782.1				40%		
Streptococcus pneumoniae NCTC7465 chromosome: 1	NZ_LN831051.1	59%-68%			40%	68%	72%
Chlamydia pneumonia TW-183	NC_005043.1	0%-59%			0%-36%		
Staphylococcus epidermidis ATCC 12228	NC_004461.1		45%		43%-48%		72%-78%
Candida albicans SC5314 chromosome 1 sequence	NC_032089.1	59%-73%			38%-48%		72%
Legionella pneumophila subsp. pascullei strain NCTC12273, chromosome: 1	NZ_LR134380.1	59%-73%			36%-40%		**78%-83%**
Streptococcus pyogenes NCTC8198, chromosome: 1	NZ_LN831034.1	73%	41%		48%		72%
Haemophilus influenzae NCTC8143, chromosome: 1	NZ_LN831035.1	77%			36%-43%		
Mycoplasma pneumoniae FH chromosome	NZ_CP010546.1				43%		
Mycobacterium tuberculosis H37Rv	NC_000962.3		34%	57%			
Pseudomonas aeruginosa PAO1	NC_002516.2					84%	72%

* All above blank cells are <50% and are considered negligible

SARS-CoV was the only organism analyzed that showed a higher than 80% overall match to the N-gene primer set. Per FDA recommendation, wet testing was performed to test cross-reactivity in SARS-CoV.

Cross-Reactivity Wet Testing:

Wet testing was performed for SARS-CoV due to its higher than 80% match rate to the N-gene and E-gene primer sets. Samples were prepared by spiking SARS-CoV RNA (BEI Resources, Catalog: 011N-03) at 200,000 copies/µL. SARS-CoV synthetic RNA had no Cross-Reactivity with the assay, as seen in Table 8 below.

Table 8. Cross-Reactivity/Exclusivity Wet Testing Results

Organism	Strain	N-gene Detected Replicates	E-gene Detected Replicates
SARS-CoV	N/A	0/8	0/8

Interfering Substances Study:

Interfering substances that could be found endogenously or exogenously were tested to evaluate any potential effects on the assay. Nasopharyngeal swabs were collected in triplicate as negative control samples, to establish a baseline of expected negative results. Then, the interfering substances were added in quantities as recommended by the manufacturer (if applicable), imitating a relevant dose. In the case of whole blood and mucin, the potential interferents were added separately into a negative clinical matrix in triplicate. Once the negative interferent samples were used, nasopharyngeal swabs were collected immediately after in triplicate and spiked with synthetic SARS-CoV-2 RNA ([redacted] SARS-CoV-2 RNA Control) at 5X LoD with the potential interfering substances. Negative swabs not containing potential interferants were spiked with synthetic RNA at 5X LoD to establish a baseline of expected positive results. None of these potential interferents were found to inhibit the performance of the assay, as all swabs with and without the potential interferents were found to have expected results. These results can be seen in Table 9.

Table 9. Endogenous and Exogenous Substances Evaluated for Potential Assay Interference

Substance	Active Ingredient	Concentration	% Agreement with Expected Results
Whole Blood	N/A	5X LoD	3/3
		Negative	3/3
Mucin	N/A	5X LoD	3/3
		Negative	3/3
Alcohol	Ethanol	5X LoD	3/3
		Negative	3/3
Chloraseptic spray	Phenol, Glycerin	5X LoD	3/3
		Negative	3/3
Mouthwash		5X LoD	3/3

	Eucalyptol, menthol, Methyl Salicylate, Thymol	Negative	3/3
Nasal allergy spray	Triamcinolone acetonide	5X LoD	3/3
		Negative	3/3
Nasal congestion spray	Oxymetazoline HCl	5X LoD	3/3
		Negative	3/3
Nyquil	Acetaminophen, Doxylamine succinate, Dextromethorphan HBr	5X LoD	3/3
		Negative	3/3
Saline nasal spray	NaCL, Phenylcarbinol, Nemalkonium Chloride	5X LoD	3/3
		Negative	3/3
Sore throat and cough lozenges	Benzocaine, Dextromethorphan HBr	5X LoD	3/3
		Negative	3/3
Tobacco	Nicotine, Tobacco	5X LoD	3/3
		Negative	3/3
THC	N/A	5X LoD	3/3
		Negative	3/3
Zinc	Zinc	5X LoD	3/3
		Negative	3/3

4) ***Clinical Evaluation:***

Performance of the DetectaChem MobileDetect Bio BCC19 Test Kit was evaluated using both contrived samples as well as confirmed clinical nasopharyngeal samples.

Contrived Testing

In the contrived clinical evaluation test series 30 contrived positive samples were tested in conjunction with 30 contrived negative samples to determine effective true positive and true negative rates. The positives and negatives were mixed randomly as shown below. The analyte used to spike the positive samples was [redacted], Synthetic SARS-CoV-2 RNA Control 1 [redacted] which contained the entire N genome. This analyte was spiked into nuclease-free water. 30 non-reactive specimens were prepared in conjunction with 30 contrived reactive specimens. Of the contrived reactive specimens 20 were spiked with RNA at 2x LoD (150 copies/µL), and 10 spiked at amounts spanning the range of the assay from 1x LoD (75 copies/µL) through 10x LoD (750 copies/µL). Of the 30 contrived positive or reactive specimens all 30 were

accurately diagnosed as positive. Of the 30 contrived negative or non-reactive specimens, all 30 were accurately diagnosed as negative. The results of this test are denoted below in Table 9. Detailed results can be found in Appendix A.

Table 10: Contrived RNA Sample Test

Detectachem Test	RNA Positives	Control Negatives	Total
Positives	30/30	0/30	30
Negatives	0/30	30/30	30
Total	30	30	60
Positive Agreement	100% (30/30)		
Negative Agreement	100% (30/30)		

Clinical Study with Confirmed Positive and Negative Samples

Live patient nasopharyngeal samples were tested simultaneously alongside EUA approved RT-PCR using Thermo Fisher Scientific's TaqPath COVID-19 Combo Kit at Oklahoma State University OADDL lab. The LoD Ct value from the data given to us was determined to be 35. At this LoD only 1 false negative was found. Table 11 below shows the summation of the OSU Patient Sample Test Report. For more detailed data on the OSU Patient Sample Test Report test view Appendix B.

Table 11: OSU Patient Sample Test Report (Ct <= 35)

TaqPath PCR Result	DetectaChem Positive	DetectaChem Negative	Total
Positive	29	1	30
Negative	0	30	30
Total	29	31	60
Positive Agreement	96.7% (29/30)		
Negative Agreement	100% (30/30)		

5) Additional Thermal Cycler Verification

In addition to testing this assay with the MD-Bio Heater, various additional thermal cyclers were verified at LoD to confirm use. These results can be seen in Table 12 below. For more detailed data on the LoD tests ran on the approved thermal cyclers view Appendix C.

Table 12: Thermal cycler LoD Verification

Thermal cycler	Concentration (copies/µL)	% Replicates Detected
Bio-Rad T100	75	100% (20/20)
Detectachem MD-Bio Heater	75	100% (20/20)
Benchmark Scientific T5005-3205	75	100% (20/20)
Techne 5Prime/02	75	100% (20/20)
EVDOtek EDVOcycler 2	75	100% (20/20)
EVDOtek EDVOcycler Jr.	75	100% (20/20)
Applied Biosystems Veriti	75	100% (20/20)
Applied Biosystems MiniAmp	75	100% (20/20)

P. RECORD KEEPING AND REPORTING INFORMATION TO FDA:

Detectachem Inc. will track adverse events and report to FDA under 21 CFR Part 803. A website is available to report on adverse events, and this website is referenced in the Fact Sheet for Health Care providers as well as through the Detectachem Inc. Product Support website: www.detectachem.comEach report of an adverse event will be processed according to Detectachem's Non-Conformance Reporting Requirements, and Medical Device Reports will be filed with the FDA as required. Through a process of inventory control, Detectachem Inc. will also maintain records of device usage/purchase. Detectachem Inc. will collect information on the performance of the test, and report to FDA any suspected occurrence of false positive or false negative results of which Detectachem Inc. becomes aware. Detectachem Inc. will maintain records associated with this EUA and ensure these records are maintained until notified by FDA. Such records will be made available to FDA for inspection upon request.

Q. APPENDIX

Appendix A: Contrived Test with Twist RNA

Test	Result	Specimen	Correct	Test	Result	Specimen	Correct	Test	Result	Specimen	Correct
1	Negative	1 µl VTM	Correct	21	Negative	1 µl VTM	Correct	41	Positive	525 copies	Correct
2	Negative	1 µl VTM	Correct	22	Positive	150 copies	Correct	42	Negative	1 µl VTM	Correct
3	Negative	1 µl VTM	Correct	23	Negative	1 µl VTM	Correct	43	Positive	150 copies	Correct
4	Negative	1 µl VTM	Correct	24	Positive	150 copies	Correct	44	Positive	375 copies	Correct
5	Positive	150 copies	Correct	25	Positive	150 copies	Correct	45	Negative	1 µl VTM	Correct
6	Positive	150 copies	Correct	26	Negative	1 µl VTM	Correct	46	Positive	75 copies	Correct
7	Negative	1 µl VTM	Correct	27	Negative	1 µl VTM	Correct	47	Negative	1 µl VTM	Correct
8	Positive	600 copies	Correct	28	Negative	1 µl VTM	Correct	48	Positive	150 copies	Correct
9	Positive	150 copies	Correct	29	Positive	150 copies	Correct	49	Negative	1 µl VTM	Correct
10	Negative	1 µl VTM	Correct	30	Positive	150 copies	Correct	50	Negative	1 µl VTM	Correct
11	Positive	150 copies	Correct	31	Positive	750 copies	Correct	51	Positive	150 copies	Correct
12	Positive	150 copies	Correct	32	Negative	1 µl VTM	Correct	52	Negative	1 µl VTM	Correct
13	Positive	450 copies	Correct	33	Negative	1 µl VTM	Correct	53	Positive	150 copies	Correct
14	Negative	1 µl VTM	Correct	34	Negative	1 µl VTM	Correct	54	Positive	225 copies	Correct
15	Positive	150 copies	Correct	35	Positive	300 copies	Correct	55	Negative	1 µl VTM	Correct
16	Negative	1 µl VTM	Correct	36	Positive	150 copies	Correct	56	Positive	500 copies	Correct
17	Negative	1 µl VTM	Correct	37	Negative	1 µl VTM	Correct	57	Positive	675 copies	Correct
18	Positive	150 copies	Correct	38	Positive	150 copies	Correct	58	Negative	1 µl VTM	Correct
19	Negative	1 µl VTM	Correct	39	Positive	150 copies	Correct	59	Negative	1 µl VTM	Correct
20	Negative	1 µl VTM	Correct	40	Negative	1 µl VTM	Correct	60	Positive	150 copies	Correct
Percentage True Positives			100%	Percentage True Negatives			100%				
Percentage False Positives			0%	Percentage False Negatives			0%				

Appendix B: OSU Patient Sample Test Report

Sample #	Detectachem – N gene		Thermo	Ct - N gene	Ct - ORF1ab	Ct - S gene
1	Negative		Negative			
2	Negative		Negative			
3	Negative		Negative			
4	Negative		Negative			
5	Positive		Positive	28.89	27.71	28.38
6	Negative		Negative			
7	Negative		Negative			
8	Negative		Negative			
9	Negative		Negative			
10	Negative		Positive	31.15	20.67	19.67
11	Negative		Negative			
12	Negative		Negative			
13	Positive		Positive	22.79	21.49	22.23
14	Positive		Positive	19.96	18.39	19.14
15	Positive		Positive	14.08	14.1	14.32
16	Positive		Positive	24.15	22.39	23.11
17	Positive		Positive	19.08	17.34	18.05
18	Positive		Positive	23.73	22.25	23.16
19	Positive		Positive	24.08	24.82	25.29
20	Positive		Positive	17.03	15.77	16.32
21	Positive		Positive	14.89	14.25	14.46
22	Positive		Positive	32.04	31.36	32.08
23	Positive		Positive	30.98	29.89	30.18
24	Positive		Positive	23.22	22.02	22.66
25	Positive		Positive	22.48	27.79	23.95
26	Negative		Negative			
27	Negative		Negative			
28	Negative		Negative			
29	Negative		Negative			
30	Negative		Negative			
31	Negative		Negative			
32	Positive		Positive	25.03	24.39	25.25
33	Positive		Positive	20.27	18.85	20.19
34	Positive		Positive	31.12	31.07	31.10
35	Positive		Positive	32.27	31.44	31.68
36	Positive		Positive	22.33	21.26	21.36
37	Positive		Positive	17.11	15.71	15.91
38	Positive		Positive	16.03	15.26	15.27
39	Positive		Positive	16.77	17.24	17.65
40	Positive		Positive	22.26	21.39	21.55
41	Positive		Positive	29.7	27.89	29.53
42	Positive		Positive	32.86	31.42	32.35
43	Positive		Positive	30.62	28.30	29.30
44	Positive		Positive	30.15	28.89	29.45
45	Negative		Negative			
46	Negative		Negative			
47	Negative		Negative			
48	Negative		Negative			
49	Negative		Negative			
50	Negative		Negative			

51	Negative		Negative			
52	Negative		Negative			
53	Negative		Negative			
54	Negative		Negative			
55	Negative		Negative			
56	Negative		Negative			
57	Negative		Negative			
58	Negative		Negative			
59	Positive		Positive	21.79	20.00	20.89
60	Positive		Positive	15.99	14.84	15.26

Appendix C: LoD Test for Recommended Thermocyclers

Bio-Rad T100			MD-Bio Heater			Benchmark Scientific T5005-3205		
	Concentration (copies/µL)	Result		Concentration (copies/µL)	Result		Concentration (copies/µL)	Result
1	75	Positive	1	75	Positive	1	75	Positive
2	75	Positive	2	75	Positive	2	75	Positive
3	75	Positive	3	75	Positive	3	75	Positive
4	75	Positive	4	75	Positive	4	75	Positive
5	75	Positive	5	75	Positive	5	75	Positive
6	75	Positive	6	75	Positive	6	75	Positive
7	75	Positive	7	75	Positive	7	75	Positive
8	75	Positive	8	75	Positive	8	75	Positive
9	75	Positive	9	75	Positive	9	75	Positive
10	75	Positive	10	75	Positive	10	75	Positive
11	75	Positive	11	75	Positive	11	75	Positive
12	75	Positive	12	75	Positive	12	75	Positive
13	75	Positive	13	75	Positive	13	75	Positive
14	75	Positive	14	75	Positive	14	75	Positive
15	75	Positive	15	75	Positive	15	75	Positive
16	75	Positive	16	75	Positive	16	75	Positive
17	75	Positive	17	75	Positive	17	75	Positive
18	75	Positive	18	75	Positive	18	75	Positive
19	75	Positive	19	75	Positive	19	75	Positive
20	75	Positive	20	75	Positive	20	75	Positive
Total Correct		20/20	Total Correct		20/20	Total Correct		20/20
Techne 5Prime/02			Edvotek EDVOcycler 2			Edvotek EDVOcycler Jr.		
	Concentration (copies/µL)	Result		Concentration (copies/µL)	Result		Concentration (copies/µL)	Result
1	75	Positive	1	75	Positive	1	75	Positive
2	75	Positive	2	75	Positive	2	75	Positive
3	75	Positive	3	75	Positive	3	75	Positive
4	75	Positive	4	75	Positive	4	75	Positive
5	75	Positive	5	75	Positive	5	75	Positive
6	75	Positive	6	75	Positive	6	75	Positive
7	75	Positive	7	75	Positive	7	75	Positive
8	75	Positive	8	75	Positive	8	75	Positive
9	75	Positive	9	75	Positive	9	75	Positive

10	75	Positive	10	75	Positive	10	75	Positive
11	75	Positive	11	75	Positive	11	75	Positive
12	75	Positive	12	75	Positive	12	75	Positive
13	75	Positive	13	75	Positive	13	75	Positive
14	75	Positive	14	75	Positive	14	75	Positive
15	75	Positive	15	75	Positive	15	75	Positive
16	75	Positive	16	75	Positive	16	75	Positive
17	75	Positive	17	75	Positive	17	75	Positive
18	75	Positive	18	75	Positive	18	75	Positive
19	75	Positive	19	75	Positive	19	75	Positive
20	75	Positive	20	75	Positive	20	75	Positive
Total Correct		20/20	Total Correct		20/20	Total Correct		20/20

Applied Biosystems Veriti			Applied Biosystems MiniAmp					
	Concentration (copies/µL)	Result		Concentration (copies/µL)	Result		Concentration (copies/µL)	Result
1	75	Positive	1	75	Positive	1		
2	75	Positive	2	75	Positive	2		
3	75	Positive	3	75	Positive	3		
4	75	Positive	4	75	Positive	4		
5	75	Positive	5	75	Positive	5		
6	75	Positive	6	75	Positive	6		
7	75	Positive	7	75	Positive	7		
8	75	Positive	8	75	Positive	8		
9	75	Positive	9	75	Positive	9		
10	75	Positive	10	75	Positive	10		
11	75	Positive	11	75	Positive	11		
12	75	Positive	12	75	Positive	12		
13	75	Positive	13	75	Positive	13		
14	75	Positive	14	75	Positive	14		
15	75	Positive	15	75	Positive	15		
16	75	Positive	16	75	Positive	16		
17	75	Positive	17	75	Positive	17		
18	75	Positive	18	75	Positive	18		
19	75	Positive	19	75	Positive	19		
20	75	Positive	20	75	Positive	20		
Total Correct		20/20	Total Correct		20/20	Total Correct		