NIRMIDAS BIOTECH

MidaSpot™ COVID-19 Antibody Combo Detection Kit (NBPC-0007)

Instructions for Use

25 Tests/Kit

For prescription use only

For in vitro diagnostic use only

For Emergency Use Authorization only

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1. PRODUCT NAME

MidaSpot™ COVID-19 Antibody Combo Detection Kit

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2. INTENDED USE

The MidaSpot™ COVID-19 Antibody Combo Detection Kit is a lateral flow immunoassay intended for the qualitative detection and differentiation of IgM and IgG antibodies to SARS-CoV-2 in human serum, plasma (dipotassium EDTA and lithium heparin), and fingerstick whole blood. The MidaSpot™ COVID-19 Antibody Combo Detection Kit is intended for use as an aid in identifying individuals with an adaptive immune response to SARS-CoV-2, indicating recent or prior infection. At this time, it is unknown for how long antibodies persist following infection and if the presence of antibodies confers protective immunity. The MidaSpot™ COVID-19 Antibody Combo Detection Kit should not be used to diagnose or exclude acute SARS-CoV-2 infection

Testing of serum and plasma specimens is limited to laboratories certified under the Clinical Laboratory Improvement Amendments of 1988 (CLIA), 42 U.S.C. 263a, that meet requirements to perform moderate or high complexity tests.

Testing of fingerstick whole blood specimens is limited to laboratories certified under the Clinical Laboratory Improvement Amendments of 1988 (CLIA), 42 U.S.C. §263a, that meet the requirements to perform high, moderate or waived complexity tests. Testing of fingerstick whole blood specimens is authorized for use at the Point of Care (POC), i.e., in patient care settings operating under a CLIA Certificate of Waiver, Certificate of Compliance, or Certificate of Accreditation.

Results are for the detection of SARS CoV-2 antibodies. IgM and IgG antibodies to SARS-CoV-2 are generally detectable in blood several days after initial infection, although the duration of time antibodies are present post-infection is not well characterized. Individuals may have detectable virus present for several weeks following seroconversion.

Laboratories within the United States and its territories are required to report all results to the appropriate public health authorities.

The sensitivity of the MidaSpot™ COVID-19 Antibody Combo Detection Kit early after infection is unknown. Negative results do not preclude acute SARS-CoV-2 infection. If acute infection is suspected, direct testing for SARS-CoV-2 is necessary.

False positive results for the MidaSpot™ COVID-19 Antibody Combo Detection Kit may occur due to cross-reactivity from pre-existing antibodies or other possible causes. Due to the risk of false positive results, confirmation of positive results should be considered using a second, different IgG or IgM assay.

The MidaSpot™ COVID-19 Antibody Combo Detection Kit is only for use under the Food and Drug Administration's Emergency Use Authorization.

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3. PRINCIPLE OF THE TEST

This test uses the principle of colloidal gold immunochromatography, and capture method to detect COVID-19 (SARS-CoV-2) IgM and IgG antibodies in human serum, plasma, and fingerstick whole blood. When the sample contains COVID-19 (SARS-CoV-2) IgM / IgG antibody and the concentration is greater than or equal to the minimum detection limit, the antibodies bind to antigen coated gold colloidal particles and migrate to the test line M / test line G and are captured by the secondary antibodies printed on the line to produce a colored (faint pink to red) reaction line. The result is considered positive when a colored reaction line appears in either test region. The result is considered negative when no colored reaction line is in either test region. The test is valid when the control line (C) produces a red reaction line.

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4. REAGENTS AND MATERIALS PROVIDED

The kit is comprised of 25 tests with 25 Individually foil-pouched MidaSpot™ COVID-19 Antibody Combo Detection Test Cards, with desiccant, 1 container tube of 25 non-sterile capillary tubes,1 bottle of Sample Diluent, and 1 bag containing 25 lancets (for use in fingerstick whole blood testing). The specifics for each item are listed below:

- The Test Card consists of the following:
    - A colloidal gold pad

- o Colloidal gold-labeled 2019-nCoV RBD Recombinant Protein and colloidal gold-labeled BSA-biotin
- A sample pad
- An absorbent paper
- A PVC board
- A Nitrocellulose membrane
  - o Streptavidin (C line),
  - o Anti-human IgM monoclonal antibody (region M),
  - o Anti-human IgG monoclonal antibody (region G)
- 1 container tube per 25 tests, each tube containing 25 non-sterile capillary tubes, with exact volume hydrophilic region; use 1 non-sterile capillary tube per Test Card.
- 1 dropper bottle of Sample Diluent per 25 tests. The Sample Diluent contains surfactant, bovine sourced serum albumin, blocking agent, macro-molecules, PBS and preservative.
- 1 bag containing 25 lancets (only needed for fingerstick whole blood testing)

5. MATERIALS REQUIRED BUT NOT PROVIDED

A timer and a permanent marker are needed to process Test Cards.
The following are needed for each fingerstick (capillary) test:

Materials for capillary draw	Qty / test
Alcohol pad	1
Gauze pad	1
Bandage	1

6. STORAGE AND EXPIRATION

The test kit should be stored at 4-25℃ in original packaging condition. Do not freeze. Properly stored kits are usable until the expiration date indicated on the kit labels. The Test Card should be used (apply sample and Sample Diluent) within 15 minutes after removing from the foil pouch and exposure to air.

7. SAMPLE REQUIREMENTS

- Serum, plasma (dipotassium EDTA or lithium heparin), and fingerstick whole blood can be used for testing.
- Serum and plasma samples should be tested within 4 hours of collection or stored at 2-8°C and tested within 3 days.
- Serum and plasma specimens which cannot be tested within 3 days after collection should be stored frozen at –20°C or lower for one month. Samples should be frozen and thawed only once. Avoid repeated freezing and thawing.

- Sample should be warmed to room temperature before testing.
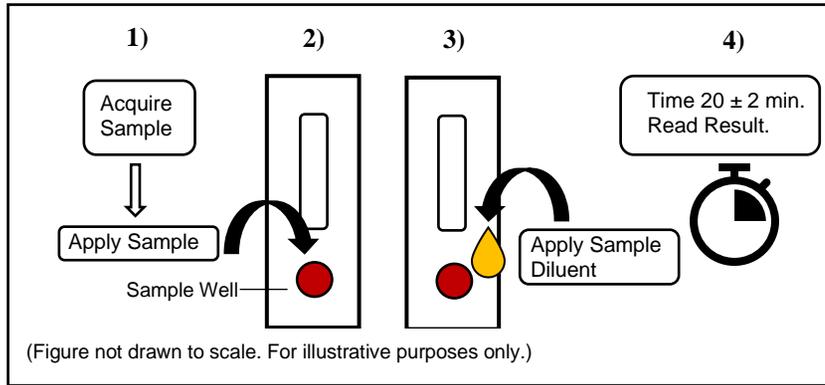- Avoid grossly hemolytic (bright red), lipemic (milky), or turbid samples (after centrifugation).
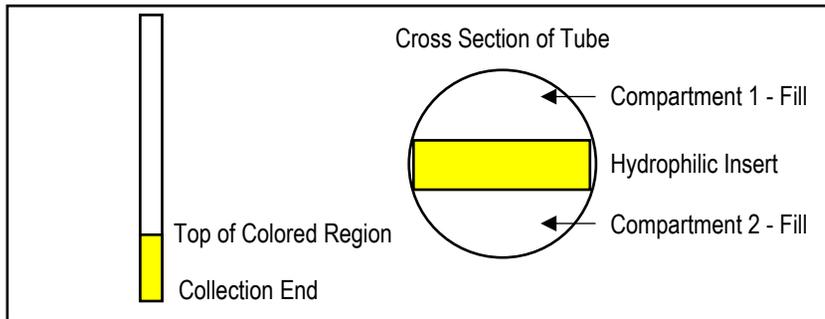
8. ASSAY PROCEDURE

- Bring specimen, Test Card, and Sample Diluent to room temperature (15-30°C) before testing.
- Remove the Test Card from the foil pouch and place on a flat surface. Do not use if foil pouch is not intact. Use a permanent marker to mark the Test Card with appropriate details.

- **For Processing Fingerstick Samples:**
  (1) Using an alcohol pad, clean the fingertip and let dry.
  (2) Remove cap on lancet.
  (3) Squeeze the finger and push the lancet firmly into the side of the finger pad.
  (4) Follow Steps 1-4 of the procedure *For Processing All Sample Types*, below.

- **For Processing All Sample Types:**
  (1) Using the provided non-sterile capillary tube and holding by the clear end, place the colored (yellow-green) collection end in contact with the sample to allow the sample to fill the tube by capillary action up to **the top of the colored region** on the capillary. Capillary tubes are exact volume* and will stop filling when the desired volume is reached. The colored region is made of a hydrophilic insert. Ensure both compartments created by this insert are filled with sample.
  (2) Deliver the sample directly on the sample well of the Test Card by **gently** touching the capillary tip to the membrane in the Sample Well until sample is fully released. Repeat touching if necessary, for full release.
  (3) Add 4 drops (total 100μL) of Sample Diluent to the sample well. Do not add more than 4 drops, or less than 4 drops. Dispense drops at a distance with enough space above the Test Card so that the liquid will form complete, free falling drops. Hold the bottle vertically when dispensing and ensure there are no bubbles in the tip. Wipe away the first drop off the bottle if bubbles are forming. Squeeze continuously, but slowly until indicated number of drops is reached.
  (4) Start a timer. Read the test result **20 ± 2 minutes** after Sample Diluent is added. Do not read the result before 18 minutes. Do not read the result after 25 minutes. Discard the Test Card after result is read to avoid confusion.

- **For Processing External Positive and Negative Controls** – Treat Positive and Negative Controls in the same manner as a serum or plasma sample, using the processing steps above.

*The sample volume collected by the non-sterile capillary tube is 10μL. Micropipettes can be used instead of the non-sterile capillary tube for serum and plasma sample application. Please contact Nirmidas for more information.

Assay Protocol Summary

1) Acquire Sample → Apply Sample — Sample Well
2)
3) Apply Sample Diluent
4) Time 20 ± 2 min. Read Result.

(Figure not drawn to scale. For illustrative purposes only.)

Capillary tube diagram (left is side view, right is cross section view from bottom). Sample collection is from the yellow end of the tube.

Cross Section of Tube

Compartment 1 - Fill
Hydrophilic Insert
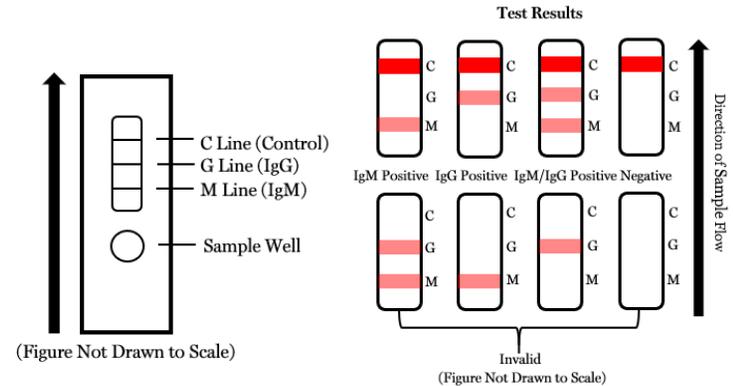Compartment 2 - Fill

Top of Colored Region
Collection End

9. INTERPRETATION OF RESULTS

- **Positive for COVID-19 (SARS-CoV-2) IgM:**
  A red line appears in test region M as well as at control line (C).
- **Positive for COVID-19 (SARS-CoV-2) IgG:**
  A red line appears in test region G as well as at control line (C).
- **Positive for Both COVID-19 (SARS-CoV-2) IgM and IgG:**
  A red line appears in both test region M and G, as well as at control line (C).
- **Negative for COVID-19 (SARS-CoV-2) IgM/IgG:**
  A red line appears at control line (C) but no line appears in test region M or test region G.
- **Faint lines in test regions M or G are considered positive.**

- **Invalid Result:**
  If no red line appears at control line (C), the test is invalid. It is recommended that the sample be retested with a new test device. Retesting of a fingerstick sample should be performed using a freshly collected fingerstick capillary blood sample.

Test Results

C Line (Control)
G Line (IgG)
M Line (IgM)
Sample Well

IgM Positive   IgG Positive   IgM/IgG Positive   Negative

Direction of Sample Flow

Invalid
(Figure Not Drawn to Scale)

(Figure Not Drawn to Scale)

10. External Positive and Negative Controls

External Positive Control / Negative Control kits are sold separately. They are manufactured by Nirmidas, Inc. and should be purchased using Catalog #NBPC-0010. It is recommended that testing of external positive and negative controls be conducted on the MidaSpot™ COVID-19 Antibody Combo Detection Kit under the following circumstances:

- A new operator uses the test kits for the first time.
- A new shipment of test kits is received.
- Device storage falls out of the 4-25°C range.
- To verify a higher or lower than expected frequency of positive or negative results.
- To investigate the cause of repeated invalid results
- A new test environment is used.
- If the expected control results are not achieved, repeat the control solution with a new test. If still not correct, contact Nirmidas at support@nirmidas.com

11. WARNING AND PRECAUTIONS

- This test has not been FDA cleared or approved but has been authorized for emergency use by FDA under an EUA. Testing of serum, plasma and venous whole blood specimens is limited to laboratories certified under CLIA that meet requirements to perform moderate or high complexity tests. Testing of fingerstick whole blood specimens is limited to laboratories certified under CLIA that meet the requirements to perform high, moderate or waived complexity tests. Testing of fingerstick whole blood specimens is authorized for use at the Point of Care (POC), i.e., in patient care settings

operating under a CLIA Certificate of Waiver, Certificate of Compliance, or Certificate of Accreditation.
- This test has been authorized only for detecting the presence of IgM and IgG antibodies against SARS-CoV-2, not for any other viruses or pathogens.
- The emergency use of this test is only authorized for the duration of the declaration that circumstances exist justifying the authorization of emergency use of in vitro diagnostic tests for detection and/or diagnosis of COVID-19 under Section 564(b)(1) of the Federal Food, Drug and Cosmetic Act, 21 U.S.C. § 360bbb3(b)(1), unless the declaration is terminated or authorization is revoked sooner.
- It is recommended that all specimens be handled in accordance with Biosafety Level 2 practices as described in the CDC/NIH Publication, Biosafety in Microbiological and Biomedical Laboratories or other equivalent guidelines.
- Always wear gloves when performing this procedure and treat all specimens and used devices as potentially infectious.
- The foil pouch containing the Test Card must be completely sealed. Do not use if foil pouch seal is not intact.
- Do not use the test device beyond the expiration date.
- Do not reuse the test device. It is for single-use.
- Contact the manufacturer if you have any questions during the use of this product.

12. LIMITATIONS

1. Use of the MidaSpot™ COVID-19 Antibody Combo Detection Kit is limited to personnel who have been trained. Not for home use.
2. Performance has only been established with the specimen types listed in the Intended Use. Other specimen types have not been evaluated and should not be used with this assay.
3. It is unknown at this time if the presence of antibodies to SARS-CoV-2 confers immunity to re-infection.
4. Results from antibody testing should not be used to diagnose or exclude acute COVID-19 infection or to inform infection status.
5. False positive results may occur due to cross-reactivity from pre-existing antibodies or other possible causes.
6. A positive result may not indicate previous SARS-CoV-2 infection. Consider other information including clinical history and local disease prevalence, in assessing the need for a second but different serology test to confirm an immune response.
7. Pedigreed specimens with direct evidence of antibodies to non-SARS-CoV-2 coronavirus (common cold) strains such as HKU1, NL63, OC43, or 229E, have not been evaluated with this assay.
8. A negative result for an individual subject indicates absence of detectable anti-SARS-CoV-2 antibodies. Negative results do not preclude SARS-CoV-2 infection and should not be used as the sole basis for patient management decisions. The sensitivity of this assay early after infection is unknown.
9. A negative result can occur if the quantity of the anti-SARS-CoV-2 antibodies present in the specimen is below the detection limits of the assay, or the antibodies that are detected are not present during the stage of disease in which a sample is collected.
10. Not for screening of donated blood.
11. SARS-CoV-2 IgG antibodies may be below detectable levels in patients who have been exhibiting symptoms for less than 15 days.
12. The performance of this device has not been established using serum, plasma, and fingerstick samples collected from individuals less than 8 days following the onset of symptoms. Samples should be collected from individuals greater than 7 days following the onset of symptoms. Samples should not be tested if collected from individuals less than 8 days post symptom onset.
13. Elevated environmental temperature combined with high humidity (40°C + 95% Relative Humidity) may produce false negative results.
14. The performance of this device has not been established in individuals that have received a COVID-19 vaccine. The clinical significance of a positive or negative antibody result following COVID-19 vaccination has not been established, and the results from this assay should not be interpreted as an indication or degree of protection from infection after vaccination.
15. The performance of this test was established based on the evaluation of a limited number of clinical specimens. The samples for the negative agreement study were all collected prior to December 2019. The samples from the positive percent agreement were collected from the United States from March 2020 to December 2020. The clinical performance has not been established in all circulating variants but is anticipated to be reflective of the prevalent variants in circulation at the time and location of the clinical evaluation. Performance at the time of testing may vary depending on the variants circulating, including newly emerging strains of SARS-CoV-2 and their prevalence, which change over time.

Conditions of Authorization for the Laboratory

The MidaSpot™ COVID-19 Antibody Combo Detection Kit Letter of Authorization, along with the authorized Fact Sheet for Healthcare Providers, the authorized Fact Sheet for Patients, and authorized labeling are available on the FDA website:https://www.fda.gov/medical-devices/coronavirus-disease-2019-covid-19-emergency-use-authorizations-medical-devices/vitro-diagnostics-euas

Authorized laboratories using the MidaSpot™ COVID-19 Antibody Combo Detection Kit ("your product" in the conditions below), must adhere to the Conditions of Authorization indicated in the Letter of Authorization as listed below:

1. Authorized laboratories* using your product must include with test result reports, all authorized Fact Sheets. Under exigent circumstances, other appropriate methods for disseminating these Fact Sheets may be used, which may include mass media.
2. Authorized laboratories using your product must use your product as outlined in the Instructions for Use. Deviations from the authorized procedures, including the

authorized clinical specimen types, authorized control materials, authorized other ancillary reagents and authorized materials required to use your product are not permitted.

3. Authorized laboratories that receive your product must notify the relevant public health authorities of their intent to run your product prior to initiating testing.

4. Authorized laboratories using your product must have a process in place for reporting test results to healthcare providers and relevant public health authorities, as appropriate.

5. Authorized laboratories must collect information on the performance of your product and report to DMD/OHT7-OIR/OPEQ/ CDRH (via email: CDRH-EUA-Reporting@fda.hhs.gov) and Nirmidas Biotech, Inc. (via email: COVID-Reporting@nirmidas.com) any suspected occurrence of false positive or false negative results and significant deviations from the established performance characteristics of your product of which they become aware.

6. All laboratory personnel using your product must be appropriately trained in immunoassay techniques and use appropriate laboratory and personal protective equipment when handling this kit and use your product in accordance with the authorized labeling. All laboratory personnel using the assay must also be trained in and be familiar with the interpretation of results of the product.

7. Nirmidas Biotech, Inc., authorized distributors, and authorized laboratories using your product must ensure that any records associated with this EUA are maintained until otherwise notified by FDA. Such records will be made available to FDA for inspection upon request.

*The letter of authorization refers to "authorized laboratories" as follows: Testing of serum and plasma (dipotassium EDTA and lithium heparin) is limited to laboratories certified under the Clinical Laboratory Improvement Amendments of 1988 (CLIA), 42 U.S.C. 263a, that meet requirements to perform moderate or high complexity tests. Testing of fingerstick whole blood specimens is limited to laboratories certified under the Clinical Laboratory Improvement Amendments of 1988 (CLIA), 42 U.S.C. §263a, that meet the requirements to perform high, moderate or waived complexity tests. Testing of fingerstick whole blood specimens is authorized for use at the Point of Care (POC), i.e., in patient care settings operating under a CLIA Certificate of Waiver, Certificate of Compliance, or Certificate of Accreditation.

13. PERFORMANCE SUMMARY

The clinical validation evaluated 55 banked clinical specimens (47 serum, and 8 plasma samples) collected from 50 unique study subjects with respiratory samples confirmed to be positive for SARS-2-CoV (by PCR), and 100 clinical specimens (51 serum, and 49 plasma samples) collected prior to the pandemic and presumed to be negative for SARS-2-CoV. The results of a total of 5 of the 55 positive specimens were not included in the data analysis because they represented serial bleeds. All samples were randomized and all test operators were blinded to the status of each sample.

The clinical performance summary data are illustrated in Tables 1a – 2d below. The confidence intervals are calculated using the Clopper-Pearson interval method.

Table 1a. Combined IgM/IgG Negative Percent Agreement for 100 Pre-Pandemic Serum and Plasma Samples.

Number of Samples Tested	MidaSpot™ COVID-19 Antibody Combo Detection Kit Results	
	IgM/IgG Negative Results	NPA (95% CI)
100	100	100.0% (96.4% - 100.0%)

Table 2a. IgG Positive Percent Agreement for 48 Serum and Plasma Samples by 'Days from Symptom Onset'

Days from Symptom Onset	Number of Samples Tested	MidaSpot™ COVID-19 Antibody Combo Detection Kit Results		
		IgG Positive Results	IgG PPA	95% CI
0-7 days	0	0	N/A	N/A
8-14 days	13	10	76.9%	49.7 – 91.8%
≥15 days	35*	35	100%	90.1 – 100%

*Ten samples did not have an associated 'date from symptom onset', however these 10 samples were collected from patients with prior reported symptoms at least 16 days after the PCR positive results, and therefore are presumed to be ≥ 15 days post symptom onset. Two additional samples for which symptom onset was unknown and could not be presumed were not included in this analysis.

Table 2b. IgM Positive Percent Agreement for 48 Serum and Plasma Samples by 'Days from Symptom Onset'

Days from Symptom Onset	Number of Samples Tested	MidaSpot™ COVID-19 Antibody Combo Detection Kit Results		
		IgM Positive results	IgM PPA	95% CI
0-7 days	0	0	N/A	N/A
8-14 days	13	13	100%	77.2 – 100%
≥15 days	35*	35	100%	90.1 – 100%

* Ten samples did not have an associated 'date from symptom onset', however these 10 samples were collected from patients with prior reported symptoms at least 16 days after the PCR positive results, and therefore are presumed to be ≥ 15 days post symptom onset. Two additional samples for which symptom onset was unknown and could not be presumed were not included in this analysis.

Table 2c. IgG Positive Percent Agreement for 12 Serum and Plasma Samples by 'Days from PCR Result'
(Performance data excludes 38 samples without "date of PCR positive" information)

Days from PCR Positive Result	Number of Samples Tested	MidaSpot™ COVID-19 Antibody Combo Detection Kit Results		
		IgG Positive Results	IgG PPA	95% CI
0-7 days	0	0	N/A	N/A
8-14 days	2	2	100%	34.2 – 100%
≥15 days	10	10	100%	72.2 – 100%

Table 2d. IgM Positive Percent Agreement for 12 Serum and Plasma Samples by 'Days from PCR Result'
(Performance data excludes 38 samples without "date of PCR positive" information)

Days from PCR Positive Result	Number of Samples Tested	MidaSpot™ COVID-19 Antibody Combo Detection Kit Results		
		IgM Positive results	IgM PPA	95% CI
0-7 days	0	0	N/A	N/A
8-14 days	2	2	100%	34.2 – 100%
≥15 days	10	10	100%	72.2 – 100%

Independent Clinical Agreement Evaluation

The MidaSpot™ COVID-19 Antibody Combo Detection Kit was tested on December 14, 2020 at the Frederick National Laboratory for Cancer Research (FNLCR) sponsored by the National Cancer Institute (NCI). The test was validated against a panel of previously frozen samples consisting of 30 SARS-CoV-2 antibody-positive serum samples and 80 antibody-negative serum and plasma (ACD) samples. Each of the 30 antibody-positive samples were confirmed with a nucleic acid amplification test (NAAT) and both IgM and IgG antibodies were confirmed to be present in all 30 samples. The presence of antibodies in the samples was confirmed by several orthogonal methods prior to testing with the MidaSpot™ COVID-19 Antibody Combo Detection Kit. The presence of IgM and IgG antibodies specifically was confirmed by one or more comparator methods. Antibody-positive samples were selected at different antibody titers.

All antibody-negative samples were collected prior to 2020 and include: i) Seventy (70) samples selected without regard to clinical status, "Negatives" and ii) Ten (10) samples selected from banked serum from HIV+ patients, "HIV+". Testing was performed by one operator using one lot of the MidaSpot™ COVID-19 Antibody Combo Detection Kit. Confidence intervals for sensitivity and specificity were calculated per a score method described in CLSI EP12-A2 (2008).

For evaluation of cross-reactivity with HIV+, it was evaluated whether an increased false positive rate among antibody-negative samples with HIV was statistically higher than the false positive rate among antibody-negative samples without HIV (for this, a confidence interval for the difference in false positive rates was calculated per a score method described by Altman). The results and data analysis are shown in Table A below.

Table A. FNLCR/NCI Independent Evaluation – Summary Results

MidaSpot COVID-19 Antibody Combo Detection Kit (NBPC-0007)	Comparator Method			Collected pre-2020		Total
	Antibody Positive			Antibody Negative		
	IgM+, IgG+	IgM+, IgG-	IgM-, IgG+	Negative	HIV+	
IgM+, IgG+	29					29
IgM+, IgG-	1			1		2
IgM-, IgG+				2		2
IgM-, IgG-				67	10	77
Total	30			70	10	110

Table 2: Summary Statistics

Measure	Estimate	Confidence Interval
IgM Sensitivity	100% (30/30)	(88.7%; 100%)
IgM Specificity	98.8% (79/80)	(93.3%; 99.8%)
IgG Sensitivity	96.7% (29/30)	(83.3%; 99.4%)
IgG Specificity	97.5% (78/80)	(91.3%; 99.3%)
Combined Sensitivity	100% (30/30)	(88.7%; 100%)
Combined Specificity	96.2% (77/80)	(89.5%; 98.7%)
Combined PPV for prevalence = 5.0%	58.4%	(30.9%; 80.4%)
Combined NPV for prevalence = 5.0%	100%	(99.3%; 100%)
Cross-reactivity with HIV+	0.0% (0/10), not detected	

Limitations of the FNLCR/NCI Independent Evaluation:
- Samples were not randomly selected, and sensitivity and specificity estimates may not be indicative of the real-world performance of the device.
- These results are based on serum and plasma samples only and may not be indicative of performance with other sample types, such as whole blood, including finger stick blood.
- The number of samples in the panel is a minimally viable sample size that still provides reasonable estimates and confidence intervals for test performance, and the samples used may not be representative of the antibody profile observed in patient populations.

Class Specificity

Using 5 samples (testing with 2 replicates each), DTT removal experiments were conducted to assess IgM removal from samples positive for both IgG and IgM. After DTT treatment, the IgM results were negative, and IgG results remained positive. This was in 100% agreement with expected outcomes. Study summary data is illustrated in Table 3.

Table 3. Class Specificity Results.

Sample ID	Replicate	Result NO DTT Treatment (IgM/IgG)	Result DTT Treatment (IgM/IgG)	Expected Result with DTT Treatment (IgM/IgG)	Result Agreement
FL 205	1	+/+	-/+	-/+	yes
FL 205	2	+/+	-/+	-/+	yes
FL 217	1	+/+	-/+	-/+	yes
FL 217	2	+/+	-/+	-/+	yes
FL 221	1	+/+	-/+	-/+	yes
FL 221	2	+/+	-/+	-/+	yes
FL 448	1	+/+	-/+	-/+	yes
FL 448	2	+/+	-/+	-/+	yes
FL 452	1	+/+	-/+	-/+	yes
FL 452	2	+/+	-/+	-/+	yes

Matrix Equivalency

Studies were performed with venous serum, and venous plasma (lithium heparin and dipotassium EDTA) using contrived negative, low positive, and moderate/high positive samples. Five complete sets of matrices were used in the study, and each assay was performed twice. The agreement was 100% between both plasma matrices compared to serum. Study summary data are illustrated in Tables 4a. and 4b.

Table 4a. Dipotassium EDTA plasma compared to serum.

Plasma (Dipotassium EDTA) compared to Serum			
IgG		IgM	
True Positives	False Positives	True Positives	False Positives
20	0	20	0
False Negatives	True Negatives	False Negatives	True Negatives
0	10	0	10
PPA	NPA	PPA	NPA
100%	100%	100%	100%

Table 4b. Lithium Heparin plasma compared to serum.

Plasma (Li Heparin) compared to Serum			
IgG		IgM	
True Positives	False Positives	True Positives	False Positives
20	0	20	0
False Negatives	True Negatives	False Negatives	True Negatives
0	10	0	10
PPA	NPA	PPA	NPA
100%	100%	100%	100%

Point of Care (POC) use

Clinical Performance Studies
Nirmidas conducted a prospective clinical study using natural fingerstick whole blood samples at two sites to assess the clinical agreement of the MidaSpot™ COVID-19 Antibody Combo Detection Kit results versus clinical status determined by PCR. To assess positive percent agreement, samples from PCR confirmed SARS-2-CoV positive participants were tested. To assess negative percent agreement, samples from both presumed healthy donors and PCR confirmed SARS-CoV-2 negative participants were tested. Study site 1 was a hospital in Maryland. Samples from a total of 38 PCR positive, and 11 PCR negative participants were tested. Five (5) operators performed the fingerstick testing. Study site 2 was in California. Samples from a total of 44 healthy participants were tested. Five (5) operators performed the fingerstick testing. The performance summary data are illustrated in Tables 5a – 5d.

Table 5a. POC Study – Site 1 – Negative Percent Agreement for 11 PCR Confirmed Negative Participants Using Fingerstick Whole Blood

Number of Samples Tested	MidaSpot™ COVID-19 Antibody Combo Detection Kit Results	
	IgG Negative Results	IgG NPA (95% CI)
11	11	100% (74.1 – 100%)
	IgM Negative Results	IgM NPA (95% CI)
11	11	100% (74.1 – 100%)

Table 5b. POC Study – Site 2 – Negative Percent Agreement for 44 Presumed Healthy Participants Using Fingerstick Whole Blood

Number of Samples Tested	MidaSpot™ COVID-19 Antibody Combo Detection Kit Results	
	IgG Negative Results	IgG NPA (95% CI)
44	44	100% (92.0 – 100%)
	IgM Negative Results	IgM NPA (95% CI)
44	43	97.7% (91.8 – 100%)

Table 5c. POC Study – IgG Positive Percent Agreement for 38 PCR Positive Participants Using Fingerstick Whole Blood by Days from Symptom Onset

Days from Symptom Onset	Number of Samples Tested	MidaSpot™ COVID-19 Antibody Combo Detection Kit Results		
		IgG Positive results	IgG PPA	95% CI
0-7 days	0	0	NA	NA
8-14 days	8	6	75%	40.9 – 92.7%
≥15 days	30	30	100%	88.6 – 100%

Table 5d. POC Study – IgM Positive Percent Agreement for 38 PCR positive participants Using Fingerstick Whole Blood.

Days from Symptom Onset	Number of Samples Tested	MidaSpot™ COVID-19 Antibody Combo Detection Kit Results		
		IgM Positive Results	IgM PPA	95% CI
0-7 days	0	0	N/A	N/A
8-14 days	8	8	100.0%	63.1% - 100.0%
≥15 days	30	30	100.0%	88.4% - 100.0%

Robustness

Studies were performed to test the robust use of this test in a point of care setting. Varying amounts of sample, varying amounts of Sample Diluent, temperature, humidity, and lighting conditions were assessed. The results from this testing indicate that the test will perform as expected across environmental and use variations that may occur in POC settings. Testing in elevated environmental temperature combined with high humidity (40°C + 95% Relative Humidity) resulted in 2 false negative IgG results out of 5 samples tested.

14. CONTACT INFORMATION

For technical and product related questions, please contact
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