CLIA Waiver by Application Approval Determination

Decision Summary

A. Document Number

CW240016

B. Parent Document Number

K232377

C. CLIA Waiver Type:

CLIA Waiver by Application

D. Applicant

Healgen Scientific, LLC

E. Proprietary and Established Names

Healgen Rapid COVID-19 Antigen Test

F. Measurand (analyte)

Nucleocapsid protein antigen from SARS-Coronavirus 2 (SARS-CoV-2)

G. Sample Type(s)

Direct anterior nasal swab specimens

H. Type of Test

Qualitative lateral flow immunoassay

I. Test System Description

1. Overview

The Healgen Rapid COVID-19 Antigen Test is a lateral flow immunochromatographic membrane assay that uses monoclonal antibodies to detect nucleocapsid protein from SARS-CoV-2 virus in anterior nasal swab (ANS) specimens. The test strip is composed of a sample pad, reagent pad, reaction membrane, and absorbing pad housed within a test cassette. The reagent pad contains all necessary reagents (antibodies and dyes) to capture and detect the target analyte present in samples and to ensure reactivity of the internal control. The test strip has two distinct areas: the test line and the procedural control line.

An ANS specimen is collected by a healthcare professional (HCP). Following sample collection, the swab is transferred to the prefilled extraction tube to lyse the sample and solubilize the viral nucleoproteins. Four (4) drops of the lysed sample is loaded onto the test cassette sample well, and the test result is read by the HCP after 15 minutes. If antigen is present, a colloidal gold-conjugated monoclonal antibody (conjugate) recognizes and binds to the virus' nucleocapsid proteins in the reagent pad, and these antigen-antibody complexes then migrate up along the reaction membrane. In the test line (T), the reaction membrane is coated with a second antibody that recognizes another epitope of the nucleocapsid protein to capture the antigen-antibody immune complexes. If SARS-CoV-2 viral antigens are present, they will be captured by antibodies bound to the T line of the strip, and a colored line will be visible in the test cassette window. The sample then continues to migrate along the test strip up to the internal procedural control line (C). A red line will always appear at the C line, which is coated with an antibody recognizing unbound conjugate (the primary antibody) in the migrating sample.

External quality controls are required to be run with the test but are sold separately. The control swabs are processed according to the Instructions for Use (IFU) and are intended to demonstrate that the test is conducted correctly. The Negative Control Swab has a blue shaft and is composed of negative control buffer dried onto a swab. The Positive Control Swab has a red shaft and contains SARS-CoV-2 recombinant antigen extract dried onto a swab at a concentration of 0.1 ng/mL in addition to the negative buffer.

2. Test System Components

The following materials are provided with the test kits:

Component
20 test cassettes
20 sterile, single use swabs (polypropylene with flocked nylon tip)
20 prefilled extraction tubes
2 tube holders
Instructions for Use
Quick Reference Guide

The following components are provided in the separately sold *COVID-19 Antigen Control Kit,* that is available in two configurations:

Configuration	**Material**
1	5 each of Positive Control Swabs, Negative Control Swabs, and QC IFU
2	10 each of Positive Control Swabs, Negative Control Swabs, and QC IFU

J. Demonstrating "Simple"

- *Is a fully automated instrument or a unitized or self-contained test.*

The device is a unitized, self-contained test and only requires sample incubation in the extraction reagent prior to sample application.

- *Uses direct unprocessed specimens, such as capillary blood (fingerstick), venous whole blood, nasal swabs, throat swabs, or urine.*

  The test uses direct unprocessed anterior nasal swab specimens.

- *Needs only basic, non-technique-dependent specimen manipulation, including any for decontamination.*

  An untrained operator can conduct the test by performing seven simple steps without sample manipulation: (1) remove the swab from packaging and collect the anterior nasal sample, (2) remove seal and add the swab to the reagent tube with extraction buffer, (3) swirl the sample swab in the extraction buffer 6 times and snap the swab tip off at the breakpoint into the tube, (4) attach the dropper cap to the tube and allow to sit for one minute in the tube holder, (5) remove the cassette from pouch and use the tube to apply 4 drops of the sample to the sample well of the cassette, (6) wait 15 minutes, and (7) read the test results. No specialized equipment is needed for sample processing.

- *Needs only basic, non-technique-dependent reagent manipulation, such as "mix reagent A and reagent B."*

  The test requires only basic reagent handling to obtain accurate results. The reagent is provided pre-measured in sealed, single-use vials. No measuring or processing of reagents is needed prior to combining the sample and the reagent. The test kits are unitized and contain all the reagents required for analysis.

- *Needs no operator intervention during the analysis steps.*

  The test does not require any operator intervention during the analysis step. After application of 4 drops of the extracted sample to the test cassette, the test develops without user intervention for 15 minutes. Upon completion of the run time, the user interprets the test results by visual interpretation using the guidance provided in the IFU/Quick Reference Guide (QRG).

- *Needs no technical or specialized training with respect to troubleshooting or interpretation of multiple or complex error codes.*

  The test does not require any instrumentation or machinery, and therefore does not need any technical or specialized training with respect to troubleshooting or interpretation of multiple or complex error codes.

- *Needs no electronic or mechanical maintenance beyond simple tasks, e.g., changing a battery or power cord.*

  The test does not require any instrumentation or machinery, and therefore does not need any electronic or mechanical maintenance.

- *Produces results that require no operator calibration, interpretation, or calculation.*

The test does not require any operator calibration or calculation to interpret results. Operator visual interpretation of test results is required.

- *Produces results that are easy to determine, such as 'positive' or 'negative', a direct readout of numerical values, the clear presence or absence of a line, or obvious color gradations.*

  The test gives three simple, easy to determine results: positive, negative or invalid based on the presence or absence of colored test and control lines. A positive result will present two colored lines beside the "C" and "T" regions of the test window. A negative result will present only one colored line next to the "C" region of the test window. If no line is seen in the "C" region after 20 minutes, then the result is invalid. The IFU/QRG provided with the test kit include descriptions of how to interpret test results, including visual examples and next steps to take based on the results.

- *Contains a quick reference instruction sheet that is written at no higher than a 7th grade reading level.*

  The test contains a QRG written at a 7th grade comprehension level.

K. Demonstrating "Insignificant Risk of an Erroneous Result" – Failure Alerts and Fail-safe Mechanisms

1. Risk Analysis

   Risk management of the Healgen Rapid COVID-19 Antigen Test has been conducted in accordance with *ISO 14971 Medical devices - Application of risk management to medical devices* and the FDA guidance, "*Recommendations for Clinical Laboratory Improvement Amendments of 1988 (CLIA) Waiver Applications for Manufacturers of In Vitro Diagnostic Devices*".

   Risk analysis was performed by the sponsor using Device Hazard Analysis and the Failure Mode Effects Analysis (FMEA) methods to assess the risks of failure that may occur during use or misuse of the device; the detailed analysis was included in the submission. The FMEA includes use-related (i.e., influenced by the user) failure modes associated with incorrect diagnosis, invalid results, unable to run test, and no result and evaluates potential failures that may be caused by the user during use or misuse of the product. Elements considered include the intended user, intended use environment (e.g., physician's offices, laboratories, hospitals), human factors/potential human errors, and historical field data from similar devices.

   Potential sources of errors that could adversely affect system performance were identified and mitigated first through system design and then through additional cautions in the labeling. The following safety risk control methods were used to eliminate or reduce the probability of occurrence of identified safety hazards:

   - *Design* – Incorporate design decisions to ensure a fail-safe architecture, including

providing redundant hardware or selection of high reliability components to minimize potential safety risks.

- *Manufacturing Procedures* – Procedures that are used in the manufacturing process are compliant with applicable quality system regulations to ensure product safety and efficacy.
- *Testing and Verifications* – Specific device test or verification activities to verify product performance of in-process and final released product consistent with the cleared performance described in the labeling.
- *Labeling / Instructions for Use* – Develop external labels for the device and instructions for use that define intended use and required operational use procedures and contra-indicate improper device use.

The identified risks which could result in erroneous test results were evaluated in flex studies that stressed the functional limits of the test system (Section K.3).

2. Fail-Safe and Failure Alert Mechanisms

The Healgen Rapid COVID-19 Antigen Test was designed to include numerous features and fail-safe mechanisms built into the system to prevent erroneous results.

Design Features

- Each test cassette is packaged in a foil pouch with desiccant to maintain the integrity of the test device and reagents.
- The foil pouch of the cassette is printed with the assay name, test analyte, lot number, and expiration date to ensure clarity and appropriate use.
- Each test cassette features distinct position marks to facilitate clear and accurate test performance and results interpretation. The cassette displays a clear letter "S" next to the sample well that visually guides the user where to add the sample. The cassette displays the control line denoted as "C" and the test line denoted as "T" within the results window to guide the operator in differentiating the test result correctly.
- The extraction buffer is sealed in a separate tube with foil to maintain the integrity of the reagent.

Fail-safe Features

- *Internal Quality Control* – The test contains a built-in procedural control. The internal procedural control "C" line is designed to control for the adequate sample volume, flow of reagents, adequate sample migration, and integrity of the assay. A visible band must be present in the control "C" region of the results window. If the control "C" line does not develop within 30 minutes, the test result is considered invalid, and retesting with a new sample and new device is recommended.
- *External Quality Control* – Testing of one Positive Control Swab (coated with SARS-CoV-2 recombinant antigen) and one Negative Control Swab (coated with

control buffer dried onto a swab) is required to ensure that the reagents and cassette are functioning properly and to demonstrate proper use and performance by the operator.

External control swabs are provided ready to use in a separate kit and are extracted and processed using the same procedure as for patient samples. Each control swab should produce the expected positive or negative results to validate test performance. These controls monitor the entire assay and serve to detect product defects or deterioration between the manufacturer's lot release date and the date of use as well as operator use to detect any errors in the performance of the test. Users are instructed to run the controls for each new lot, shipment, or operator and as required to follow local, state, and federation regulations regarding quality control procedures. If the controls do not perform as expected, users are instructed to repeat testing and not to report patient results after repeated invalid results. Users are instructed not to use expired external controls.

3. Flex Studies

The operational limits of the Healgen Rapid COVID-19 Antigen Test were evaluated in a series of "stress" experiments, including variations in workflow and operating environment outside of those recommended in the IFU that may reasonably be expected to occur with untrained operators in a CLIA-waived setting, to support the CLIA Waiver Application. Flex studies were conducted in 3 replicates each of low positive (inactivated virus prepared at 2x LoD) and negative (negative clinical matrix (NCM) alone) samples prepared and tested under various conditions to evaluate test robustness on one test lot. Fifty (50) µL of the prepared sample was applied to kit swab and eluted into the extraction buffer tube, and tests were subjected to the following flex studies.

a. Environmental factors

Extreme temperature and humidity/Opened kit stability – The test was stored under the following controlled conditions under closed pouch storage for 5 days and a further 2 hours after test pouches and buffer vials were opened: low temperature and humidity (5°C, 5% RH), low temperature/high humidity (5°C, between 85 to 95% RH), high temperature/low humidity (45°C, 5% RH), high temperature and high humidity (45°C, between 85 to 95% RH). For these studies, negative samples were composed of 0.1x LoD virus in negative clinical matrix. All samples gave the expected result.

Light source – This study evaluated the impact of lighting source on the user's ability to perform the test and interpret the results. The test was run in fluorescent, incandescent, and natural lighting. All samples gave the expected result.

b. Human factors / Operator error

Incorrect handling of specimen or reagents

Incorrect application of the specimen to the device (Swab Transfer Method Study) – Swabs were swirled 0, 2, 4, 6, 8 or 10 times in the buffer extraction tube; the test

recommends 6 swirls. All samples gave the expected result.

Incorrect application of the specimen to the device (Sample Volume Study) – Volumes of 1, 2, 3, 4, 5 or 6 drops of sample were added to the well of the test cassette; four drops are recommended. The one drop volume gave an invalid result; all other samples gave the expected result.

Incorrect handling of reagents (Bubbles in Sample Study) – The extraction tube containing samples in extraction buffer was shaken thoroughly for about 20 seconds until bubbles were observed and then added to the sample well of the test cassette. For this study, negative samples were composed of nasal cavity wash, and each sample was tested in 5 replicates using three kit lots. All samples gave the expected result.

Incorrect placement of device

Disturbance during operation – Test cassettes were dropped from the lab bench from a height of 35" after sample loading. For this study, negative samples were composed of 0.1x LoD virus in negative clinical matrix. All cassettes were intact following dropping except for one, which had its membrane slightly dislocated, but it did not lead to incorrect interpretation of the test result. All samples gave the expected result.

Non-level surface – Four cassette positions were tested as follows and read after an incubation of 15 minutes: sample loading well on top, sample loading well at bottom, sample loading well on the left, and sample loading well on the right. For this study, negative samples were composed of 0.1x LoD virus in negative clinical matrix. All samples gave the expected result.

Incorrect timing of test procedures

Incorrect reading of test results – Samples were added to the well of the test cassette and read at 5, 10, 15, 20, 25, 30 or 60 minutes; 15 to 20 minutes is recommended. All samples gave the expected result.

Incorrect extraction time – Samples were incubated at 0, 1, 5 and 10 minutes after swab was transferred to the buffer extraction tube; one minute is recommended. All samples gave the expected result.

4. Specimen and Reagent Integrity

a. Specimen stability

A specimen stability study in which 3 replicates each of low positive (prepared at 2x LoD) and negative samples (NCM) were prepared and tested on 3 test lots. Fifty (50) µL of the prepared sample were applied to kit swab and incubated at room temperature (RT). Swabs were tested at 0, 2, 4 and 6 hours. All samples gave the expected result, demonstrating that specimens are stable up to 4 hours after collection when stored at RT.

b. Reagent stability

A real-time reagent stability study was conducted on the Healgen Rapid COVID-19 Antigen Test to support a shelf-life of 18 months. Three lots were stored at 2 to 8°C and 30 ± 3°C. At each timepoint test devices were visually inspected and tested with 5 replicates of a low positive sample (prepared at 2x LoD) and one replicate each of a high negative (prepared at 0.1x LoD) and a negative sample. Testing was performed at 0, 3, 6, 9, 12, 15, 18 and 20 months. All samples at all timepoints gave the expected results, supporting a shelf-life of the test device for 18 months under the intended storage condition.

Three lots of the COVID-19 Antigen Control Kit were stored at 2 ± 3°C and 30 ± 3°C and tested in duplicate on 2 test kit lots at Time 0 and every 3 months to 27 months. All samples at all timepoints and storage conditions gave the expected results, supporting a shelf-life of the control kit for 24 months under the intended storage condition.

L. Demonstrating "Insignificant Risk of an Erroneous Result" – Accuracy

1. Comparison Study

a. Study Design

i. Study Sites and Duration

The clinical performance of the Healgen Rapid COVID-19 Antigen Test was evaluated in a multi-center, prospective study conducted from May 2022 to July 2022 using 32 untrained operators at six different CLIA-waived sites; comparator samples were tested at one reference laboratory. Testing was performed in a simulated home environment. The table below describes the clinical study sites and number of operators per site.

Table 1. Overview of Study Sites

Site	Site Type	# Operators
1	CLIA-waived clinical research center (Miami, FL)	3
2	CLIA-waived emergency department (St. Louis, MO)	7
3	CLIA-waived urgent care or medical clinic / Clinical research center (Sarasota, FL)	4
4	CLIA-waived emergency department (Ann Arbor, MI)	8
5	CLIA-waived urgent care or medical clinic / Clinical research center (Ft. Lauderdale, FL)	7
6	CLIA-waived urgent care or medical clinic / Clinical research site (Morgantown, WV)	3

ii. Operators

There were a total of 32 operators representative of users in a CLIA-waived setting across the six clinical sites. The table below describes the study operators that were responsible for enrollment, collection and testing of samples.

Table 2. Overview of Study Operators

Site	#	Operator	Education	Occupation	Comments
1	3	1	High School	Medical Assistant/Phlebotomist	Provide clinical and administrative support; interview patients, measure and record vital signs, performing phlebotomy and other routine tests (e.g. ECG), document patient plan of care, tests, and examination results in patient's chart.
		2	High School	Medical Assistant/Phlebotomist	
		3	High School	Medical Assistant/Phlebotomist	
2	7	1	Bachelor's Degree	Medical Assistant	Provide clinical and administrative support; interview patients to obtain medical histories, measure and record vital signs, performing phlebotomy and other routine tests (e.g. ECG), obtain urine samples, obtain nasal and NP samples, obtain ECGs, room patients and place IVs; documentation of symptoms, medical history, tests and examination results in patient chart.
		2	Master's Degree	Medical Assistant/Paramedic	
		3	High School	Medical Assistant/EMT	
		4	Medical Degree	Medical Assistant	
		5	Bachelor's Degree	Medical Assistant/ Registered Nurse	
		6	Associate Degree	Medical Assistant/ Registered Nurse	
		7	Bachelor's Degree	Medical Assistant	
3	4	1	Doctor in Surgery	Medical Assistant	Interviewing patients, performing other routine tests such as ECGs, documenting patient plan care, tests, and examination of results.
		2	Bachelor's Degree	Clinical Assistant	Interviewing patients, documenting patient plan care, tests, and examination of results.
		3	Bachelor's Degree	Data Entry Specialist	Provides administrative and data entry support to the study.
		4	Associate Degree	Clinical Assistant	Interviewing patients, documenting patient plan care, tests, and examination of results.
4	8	1	Master's Degree	Clinical Research Coordinator	Oversee training, create workflow, and direct instruction of research staff, interview patients/participants, measure and record vital signs.
		2	High School	Research Assistant	Interview patients/participants, and measure and record vital signs.
		3	High School	Research Assistant	Interview patients/participants, and measure and record vital signs.
		4	Bachelor's Degree	Project Manager	Interview patients/participants, and measure and record vital signs.
		5	High School	Research Assistant	Interview patients/participants, and measure and record vital signs.
		6	Bachelor's Degree	Research Specialist	Interview patients/participants, and measure and record vital signs.
		7	Bachelor's Degree	Research Assistant	Interview patients/participants, and measure and record vital signs.
		8	PhD	Post-doctoral Fellow	Interview patients/participants, measure and record vital signs, and perform phlebotomy.

Site	#	Operator	Education	Occupation	Comments
5	7	1	Medical Degree	Clinical Research Coordinator/Medical Assistant	Assists with interviewing patients, measuring and recording vital signs, performing phlebotomy and other routine tests such as ECGs, document patient plan of care, tests, and examinations.
		2	Bachelor's Degree	Clinical Research Coordinator/Clinical Assistant	Provide clinical and administrative support.
		3	Doctor in Surgery	Clinical Research Coordinator/Medical Assistant	Assists with interviewing patients, measuring and recording vital signs, performing phlebotomy and other routine tests such as ECGs, document patient plan of care, tests, and examinations.
		4	Bachelor's Degree	Clinical Research Coordinator/Medical Scribe	Provides clinical and administrative support, assists with interviewing patients, measuring and recording vital signs, document patient plan of care, tests, and examinations.
		5	Bachelor's Degree	Clinical Research Coordinator/Clinical Assistant	Provide clinical and administrative support.
		6	Bachelor's Degree	Clinical Research Coordinator/Data Entry Specialist	Provide clinical and administrative support.
		7	Bachelor's Degree	Clinical Research Coordinator/Clinical Assistant	Assists with interviewing patients, measuring and recording vital signs, document patient plan of care, tests, and examinations.
6	3	1	Medical Assistant	Clinical Research Coordinator/Medical Assistant	Provides clinical and administrative support in the clinic, interview patients, measuring and recording vital signs and other routine testing.
		2	Medical Assistant	Clinical Research Coordinator/Medical Assistant	
		3	Bachelor's Degree	Clinical Research Coordinator/Medical Assistant	

iii. Instructions for Use

The operators selected for the study were untrained in the use of the Healgen Rapid COVID-19 Antigen Test and provided only with the Quick Reference Guide (QRG).

iv. Subjects (Patients)

A total of 867 subjects were enrolled in the clinical study of which 843 were evaluable. Twenty-four (24) subject samples were removed due to failure to meet inclusion criteria or unplanned protocol deviations at the study sites. Enrolled subjects were aged 2 years or older symptomatic individuals within six (6) days post symptom onset (DPSO) who still exhibited symptoms at the time of collection. Symptomatic individuals were defined

as exhibiting symptoms of fever or 2 or more of the following: chills, cough, shortness of breath/difficulty breathing, fatigue, muscle/body aches, headache, new loss of taste or smell, sore throat, congestion or runny nose, nausea or vomiting, and diarrhea. Patient demographics and specimen positivity are summarized in the following tables.

Table 3. Overview of Patient Demographics

Demographic Distribution (N = 806, %)		
Gender	Female	475 (57.7)
	Male	348 (42.3)
Age Group (Years)	2-13	80 (9.7)
	14-24	119 (14.4)
	25-64	525 (63.8)
	≥ 65	99 (12.0)
Race	American Indian or Alaskan Native	1 (0.1)
	Asian	13 (1.6)
	Black/African American	156 (19.0)
	Native Hawaiian or Pacific Islander	1 (0.1)
	White/Caucasian	651 (79.1)
	Unknown/Prefer not to answer	0 (0.0)
	Other	1 (0.1)
Ethnicity	Hispanic/Latino	486 (59.1)
	Not Hispanic/Latino	336 (40.8)
	Unknown/Prefer not to answer	1 (0.1)

v. Samples

A total of 806 samples were tested and evaluated for clinical performance; of the 843 evaluable subjects, 20 asymptomatic subjects and 17 subjects at 7 DPSO were further excluded.

Table 4. Overview of Samples Tested and Specimen Positivity by DPSO

DPSO	# Samples	# Positive Samples	% Positive
0	26	3	11.5
1	92	16	17.4
2	213	46	21.6
3	219	39	17.8
4	150	29	19.3
5	75	19	25.3
6	31	12	38.7
TOTAL	**806**	**164**	**20.3**

Two swabs were collected from each subject, one ANS was immediately tested on the candidate device, and one nasopharyngeal swab (NPS) was collected into 3 mL of viral transport media (VTM), stored at 2-8°C and tested within 120 hours using a composite comparator method (or frozen at -70°C for long-term storage). Samples were randomly selected for either HCP collection or self collection under the supervision of an HCP. Some samples, mostly from pediatric subjects, were also lay-collected. Sample collection

order was not randomized since candidate and comparator samples were collected from different anatomical body sites. The table below provides additional information about sample collection and sample types at the different study sites.

Table 5. Overview of Samples Tested and Collection Method

Site	Samples Tested	HCP-Collected	Self-Collected/ Lay-Collected	# Positives	% Positivity
1	399	199	200	76	19.0
2	95	48	47	23	24.2
3	72	39	33	9	12.5
4	46	24	22	15	32.6
5	28	14	14	10	35.7
6	166	81	85	31	18.7
TOTAL	**806**	**405**	**401**	**164**	**23.8**

vi. Comparative Method (CM)

For comparator testing, samples were tested using a composite reference method. First, testing was performed on two highly sensitive RT-PCR assays; testing on a third highly sensitive molecular test would then be performed if discordant results were observed between the first two tests. The final comparator result for those samples was determined based on a 2 out of 3 rule.

b. Results and Analysis

i. Statistical Analysis of Comparison Study Results

Results obtained on the candidate device were compared to the composite comparator method and performance was determine as a clinical sensitivity/positive percent agreement (PPA) of 85.4% with a 95% Confidence Interval (CI) of 79.1% and a specificity/negative percent agreement (NPA) of 99.7% with a 95% CI of 98.9%.

Table 6. Overview of Clinical Performance

Candidate device	Composite Comparator Method		
	Detected	**Not Detected**	**Total**
Reactive	140	2	142
Non-reactive	24	640	664
Total	164	642	806
Positive Percent Agreement: 85.4% (95% CI: 79.1 to 90.0%)			
Negative Percent Agreement: 99.7% (95% CI: 98.9 to 99.9%)			

Table 7. Summary of Clinical Performance by DPSO

DPSO	Number of Samples	Candidate Positive	Comparator Positive	PPA (%)
Day 0*	26	3	3	100.0
Day 1	92	14	16	87.5
Day 2	213	36	46	78.3

DPSO	Number of Samples	Candidate Positive	Comparator Positive	PPA (%)
Day 3	219	31	39	79.5
Day 4	150	27	29	93.1
Day 5	75	17	19	89.5
Day 6	31	12	12	100

*Day 0: Day symptoms start, that is symptoms exist <24 hours.

Table 8. Summary of Clinical Performance by Site

Site	Number of Samples	Candidate Positive	Comparator Positive	PPA (%)
1	399	64	76	84.2
2	95	20	23	86.9
3	72	7	9	77.8
4	46	13	15	86.7
5	28	8	10	80.0
6	166	28	31	90.3

2. Device Performance with Analyte Concentrations Near the Cutoff (Multi-Site Precision (Reproducibility) and Near Cutoff Evalution)

a. Multi-Site Precision

Precision/reproducibility of the Healgen Rapid COVID-19 Antigen Test was evaluated at 3 external, CLIA-waived testing sites with intended users representative of those typically found at CLIA-waived sites that provide patient care. The tables below describe the test sites and operators.

Table 9. Overview of Study Sites

Site	Site Type	# Operators
1	CLIA-waived family practice clinic/Clinical research center	3
2	CLIA-waived urgent care clinic/Clinical research site	3
3	CLIA-waived internal medicine clinic/Clinical research site	3

Testing was performed by 3 untrained operators per site over 5 days. Each operator tested 3 replicates of each sample concentration each day, i.e., 3 replicates × 3 operators × 3 sites × 5 days = 135 replicates per concentration. The following test sample concentrations were prepared: positive (prepared at 4.9x LoD), low positive (prepared at 1x LoD), and negative. Three test lots were used in this study; lots were distributed to all sites and each of 3 operators per site used a different lot per day, alternating different lots each day. Fifty (50) µL of the prepared sample were applied to kit swabs, shipped and stored frozen at -20°C until testing.

The results were ≥ 90% agreement between expected and read result within run, by lot, by operator, by day, between sites and overall.

Table 10. Summary Results of Multisite Precision Study (Reproducibility)

Site	Negative		Weak Positive		Positive	
	Correct Reads / Total	NPA	Correct Reads / Total	PPA	Correct Reads / Total	PPA
1	44 / 45	97.8%	45 / 45	100.0%	45 / 45	100.0%
2	45 / 45	100.0%	45 / 45	100.0%	45 / 45	100.0%
3	45 / 45	100.0%	45 / 45	100.0%	45 / 45	100.0%
Total	**134 / 135**	**99.2%**	**135 / 135**	**100.0%**	**135 / 135**	**100.0%**

b. Near Cutoff Evaluation

An additional study was performed to evaluate device performance with weakly reactive samples near the assay cutoff and to assess lot-to-lot variability. For this, low positive (1x LoD), high negative (0.1x LoD), and negative (NCM alone) samples were tested and compared between trained and untrained operators, i.e., 3 replicates × 3 sites × 4 operators (3 untrained + 1 trained) × 5 days = 180 data points for each analyte level. Samples were randomized into 180 panels each containing 1 weak positive, 1 high negative/true negative, and 1 high negative sample, with 90 panels each split between the untrained and the trained operators. That is, each of 12 untrained operators at 3 sites tested 10 panels each (10 weak positive, 20 high negative/true negative replicates), and each of 3 trained operators at one site tested 30 panels (30 weak positive, 60 high negative/true negative replicates).

Table 11. Summary Results of Near Cutoff Study by Lot

Lot	Negative		High Negative		Low Positive	
	Correct Reads / Total	NPA	Correct Reads / Total	PPA	Correct Reads / Total	PPA
1	25 / 25	100.0%	32 / 32	100.0%	26 / 28	92.9%
2	36 / 36	100.0%	20 / 22	90.9%	28 / 28	100.0%
3	36 / 36	100.0%	26 / 26	100.0%	31 / 31	100.0%

Table 12. Summary Results of Near Cutoff Study by Operator

Operator	Negative		High Negative		Low Positive	
	Correct Reads / Total	NPA	Correct Reads / Total	PPA	Correct Reads / Total	PPA
Untrained	97 / 97	100.0%	78 / 80	97.5%	85 / 87	97.7%
Trained	99 / 99	100.0%	81 / 81	100.0%	90 / 90	100.0%

Table 13. Near Cutoff Study Results by Operator

Operator Type	Operator	Correct Reads / Total			Overall Agreement (%)
		Negative	High Negative	Weak Positive	
Trained	1	33 / 33	27 / 27	30 / 30	100.0
	2	33 / 33	27 / 27	30 / 30	100.0
	3	33 / 33	27 / 27	30 / 30	100.0

Untrained	4	11 / 11	9 / 9	10 / 10	100.0
	5	10 / 10	9 / 9	9 / 9	100.0
	6	11 / 11	9 / 9	10 / 10	100.0
	7	11 / 11	9 / 9	9 / 10	96.7
	8	11 / 11	8 / 9	10 / 10	96.7
	9	11 / 11	9 / 9	10 / 10	100.0
	10	10 / 10	8 / 8	7 / 8	96.2
	11	11 / 11	8 / 9	10 / 10	96.7
	12	11 / 11	9 / 9	10 / 10	100.0

3. Operator Questionnaire

Usability and User Comprehension (Sample Collection): At the end of the clinical evaluation, a subset of study participants completed a questionnaire. Of the 823 evaluable subjects, 435 subjects completed a questionnaire. The questions were to do with ease of sample collection. Of the subjects that participated in the assessment, 98.4% found the instructions clear and easy to follow, 98.6% found sample collection easy to perform, and 92.6% rated the swab collection from neutral to very comfortable. Overall, 96.5% of all subjects collected the samples correctly.

Usability and User Comprehension (Sample Processing): At the end of the Precision/Reproducibility studies, each of the nine untrained operators completed a questionnaire to provide feedback on the Healgen Rapid COVID-19 Antigen Test. The questionnaire contained 8 questions on the ease of use of the test and interpreting the results and whether the IFU and QRG were easy to read and understand. The individuals completing the questionnaire overwhelmingly chose "Strongly Agree" or "Agree" that the test was easy to perform and the written instructions were easy to understand. One operator chose "Neutral" that the sample was easy to prepare for testing.

M. Labeling for Waived Devices

The labeling for the Healgen Rapid COVID-19 Antigen Test consists of:

1. Instructions for Use
2. Quick Reference Guide
3. Quality Controls Instructions for Use

The following elements are appropriately present:

- The Quick Reference Guide and the Instructions for Use are written at no higher than a 7th grade reading level.
- The Instructions for Use and Quick Reference Guide identify the test as CLIA waived.

- The Instructions for Use and test cartridge package insert contain a statement that a Certificate of Waiver is required to perform the test in a waived setting.
- The Instructions for Use and Quick Reference Guide contain a statement that laboratories with a Certificate of Waiver must follow the manufacturer's instructions for performing the test per 42 CFR 493.15(e)(1).
- The Instructions for Use and Quick Reference Guide provide instructions for conducting quality control procedures.

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

N. Benefit-Risk Considerations

The evidence provided in this submission indicates that the Healgen Rapid COVID-19 Antigen Test will diagnose SARS-CoV-2 within the intended use population. This assay was validated to support a full clearance and classification as a moderate complexity device. The Clinical Evaluation demonstrated a clinical sensitivity/positive percent agreement (PPA) of 85.4% and a clinical specificity/negative percent agreement (NPA) of 99.7%. The study took place when Omicron was the predominant circulating variant. The performance estimates therefore reflect the expected device performance under current real-world use at the time of granting this CLIA Waiver by Application.

The risks associated with the device, when used as intended, are those related to the risk of false test results, failure to correctly interpret the test results, and failure to correctly operate the device. The clinical benefits outweigh the probable risk of erroneous results for the proposed assay, considering the product labeling, special controls, and general controls. The clinical benefits of the assay include ease of use for the healthcare provider. The results of the CLIA Waiver Clinical Evaluation, Reproducibility Study, and Operator Questionnaire suggest that errors will be uncommon and are mitigated by the device labeling, which will facilitate accurate assay implementation and interpretation of results.

Thus, granting of a CLIA Waiver for the Healgen Rapid COVID-19 Antigen Test will provide substantial benefits to patients and healthcare providers as an aid in the diagnosis of SARS-CoV-2 when used in conjunction with other laboratory results and clinical information, and will be a benefit to public health.

O. Conclusion:

The submitted information in this CLIA waiver application supports an approval decision.