510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
INSTRUMENT ONLY

I Background Information:

A 510(k) Number

K223497

B Applicant

Spectrum Solutions, LLC

C Proprietary and Established Names

Spectrum Saliva Collection Device

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
QBD	Class II	21 CFR 866.2950 - Microbial Nucleic Acid Storage And Stabilization Device	MI - Microbiology

II Submission/Device Overview:

A Purpose for Submission:

To make a substantial equivalence determination for the Spectrum Solutions Saliva Collection Device for the collection, transport, and storage of viral specimens in saliva to the laboratory for downstream testing.

B Measurand:

Storage and stability of nucleic acids from SARS-CoV-2

C Type of Test:

Microbial nucleic acid storage and stabilization device

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

The Spectrum Solutions Saliva Collection Device is designed for use in the non-invasive collection, inactivation, and stabilization of viral nucleic acids for in vitro diagnostic testing of saliva samples. The device is intended to inactivate and stabilize human clinical saliva samples from the collection site to the laboratory at room temperature. The saliva sample is stabilized and suitable for use with legally marketed molecular diagnostic devices. The device is intended to be used by a health care provider for samples suspected of containing SARS-CoV-2.

C Special Conditions for Use Statement(s):

For Prescription Use Only

D Special Instrument Requirements:

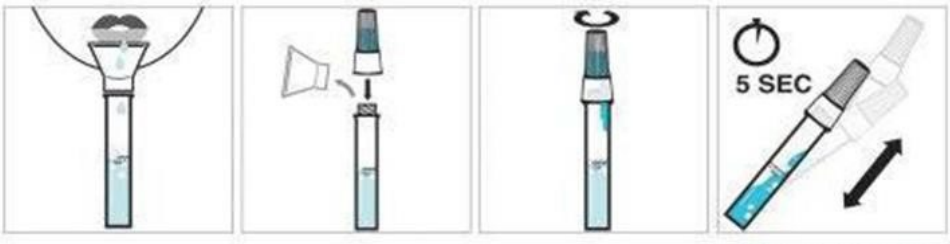
None

IV Device/System Characteristics:

A Device Description:

The Saliva Collection Device consists of a plastic tube designed for the collection of human saliva samples, a funnel, a cap with a stem flare, and a fluid chamber containing Spectrum's inactivating media. Sample collection is conducted under the supervision of a Health care provider. The user deposits their saliva into the collection tube with the aid of the attached funnel, the user removes the funnel and replaces it with the cap. Upon twisting and closing the cap, the inactivating media is released into the tube and mixes with the saliva.

Figure 1. Saliva Collection Device Sample Collection Steps



B Principle of Operation:

The components of the inactivating media for the Spectrum device are intended to inactivate SARS CoV-2 capsids, disrupt/lyse membranes, denature proteins, inactivate enzymes, and stabilize SARS CoV-2 RNA. The inactivating solution includes buffering salts, purified water, and surfactants as well as the following:

- Guanidine Thiocyanate
- Ethanol
- EDTA
- Blue dye

C Instrument Description Information:

1. Instrument Name:

   N/A

2. Specimen Identification:

   N/A

3. Specimen Sampling and Handling:

   N/A

4. Calibration:

   N/A

5. Quality Control:

   N/A

V Substantial Equivalence Information:

A Predicate Device Name(s):

Zymo Research DNA/RNA Shield Collection Tube

B Predicate 510(k) Number(s):

K202641

C Comparison with Predicate(s):

Feature:	Device: K223497	Predicate: K202641
Device Trade Name	Spectrum Solutions Saliva Collection Device	DNA/RNA Shield Collection Tube
General Device Characteristic Similarities		
Intended Use/Indications For Use	The Spectrum Solutions Saliva Collection Device is designed for use in the non-invasive collection,	The DNA/RNA Shield collection tube is intended for the stabilization and inactivation of upper and

	inactivation, and stabilization of viral nucleic acids for in vitro diagnostic testing of saliva samples. The device is intended to inactivate and stabilize human clinical saliva samples from the collection site to the laboratory at room temperature. The saliva sample is stabilized and suitable for use with legally marketed molecular diagnostic devices. The device is intended to be used by a health care provider for samples suspected of containing SARS-CoV-2.	lower respiratory human specimens suspected of containing SARS-CoV-2. These devices can be used for collection transport and storage of specimens at ambient temperatures (20-25°C). Specimens collected and stored in a DNA/RNA Shield™ collection tube are suitable for use with legally marketed molecular diagnostic devices.
Additive/Reagent	Nucleic Acid Stabilization solution	Same
Analyte	RNA from SARS-CoV-2	Same
Limit of Detection	250 GEC/ml (20 GEC/reaction)	Same
Specimen Stability	SARS-CoV-2 for 28 days at 20-25 °C	Same
Transport Media	Disrupt/lyses lipid membranes, inactivates enzymes, and stabilizes nucleic acids	Same
Material	Medical-grade polypropylene	Same
Sterility	Non-Sterile	Same
General Device Characteristic Differences		
Specimen Type	Saliva only	Upper, lower respiratory, and saliva
Device Material	Tube - Polypropylene Tube and Funnel containing Spectrum's Nucleic Acid Stabilization Solution and a Polypropylene Cap.	Plastic Collection Tube pre-filled with DNA/RNA Shield transport media.
Inactivation test	Greater than 4 Log viral reduction in 10 seconds	Greater than 2 log viral reduction in 30 minutes

VI Standards/Guidance Documents Referenced:

Special controls that are applicable to regulation 21 CFR 866.2950

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

   N/A

2. Linearity:

   N/A

3. Analytical Specificity/Interference:

   N/A

4. Assay Reportable Range:

   N/A

5. Traceability, Stability, Expected Values (Controls, Calibrators, or Methods):

   Shelf life: The shelf life for the Spectrum Saliva Collection Device is 24 months at room temperature (15-30ºC) after the date of manufacture when stored at 5-30ºC. The stability of the Spectrum Saliva Collection Device was performed using real-time analysis on three (3) lots of device caps (tube tops) containing inactivating media. All of the 24-month-aged caps tested with inactivating media met the acceptance criteria for stability with regards to color appearance of dye (blue), pH, assays for guanidine thiocyanate, denatured alcohol, and EDTA, specific gravity, bacterial and fungal contamination, and weight. Analytical testing was run on finished commercial products after two years.

   Sterilization: The Spectrum Solutions Saliva Collection Devices are not sold as sterile nor are they intended to be sterilized by the user. These vials are single use devices that do not require cleaning by the operator.

   Sample Stability: Studies were conducted to evaluate the stability of SARS-CoV-2 in the Spectrum inactivating media for up to 28 days. Contrived samples containing SARS-CoV-2 at a concentration of 3x LoD (62.5 GEC/reaction; 1562.5 GEC/ml) were prepared in saliva matrix collected from a single negative donor. Sample stability was evaluated in a study that utilized two different device lots to collect two samples each (for a total of 4 test samples) that were then stored at ambient temperature (20-25ºC) for increasing lengths of time. Aliquots of each contrived sample was tested for SARS-CoV-2 using the TaqPath COVID-19 assay at T=0 and on Days 1, 2, 3, 4, 5, 6, 7, 14, 21, and 28. Aliquots of samples were removed and stored at -20ºC at various times and then all aliquots (total of 44 samples) were

tested as a single testing batch. The acceptance criteria for the stability study included a deviation of no more than +/- 3 Ct for each target at given time point from T=0

Results of stability testing are shown below in Tables 1 and 2.

Table 1. Specimen Stability Study Results

Time	Orf1ab Ct average	Orf1ab Ct SD	Orf1ab Positive Count	N gene Ct average	N gene Ct SD	N gene Positive Count	S gene Ct average	S gene Ct SD	S gene Positive Count
T0	33.1	0.98	4/4	31.7	0.32	4/4	32.8	0.34	4/4
D1	31.9	0.85	4/4	31.3	0.45	4/4	33.0	0.49	4/4
D2	32.4	0.33	4/4	31.4	0.33	4/4	33.8	0.61	4/4
D3	32.4	0.40	4/4	31.4	0.35	4/4	33.6	0.26	4/4
D4	32.7	1.23	4/4	31.6	0.46	4/4	34.0	1.74	4/4
D5	32.7	0.47	4/4	31.5	0.28	4/4	34.2	0.86	4/4
D6	32.7	0.43	4/4	31.6	0.29	4/4	33.4	0.47	3/4
D7	33.2	0.87	4/4	31.8	0.32	4/4	34.5	1.15	4/4
D14	33.7	0.84	4/4	32.1	0.19	4/4	33.8	NA	1/4
D21	34.4	0.79	4/4	33.1	0.68	4/4	NA	NA	0/4
D28	33.4	3.25	4/4	34.4	0.53	4/4	NA	NA	0/4

Table 2. Specimen Stability Study – Ct Changes Observed

Time	Orf1ab Ct change from T0	N gene Ct change from T0	S gene Ct change from T0
T0	NA	NA	NA
D1	-1.25	-0.38	0.29
D2	-0.73	-0.34	1.04
D3	-0.72	-0.29	0.85
D4	-0.47	-0.15	1.19
D5	-0.43	-0.26	1.45
D6	-0.47	-0.10	0.69
D7	0.05	0.05	1.75
D14	0.52	0.38	1.04
D21	1.28	1.40	ND
D28	0.24	2.71	ND

43/44 of the samples tested positive for SARS-CoV-2 at all time points during initial testing. One Day 21 sampled failed to produce a conclusive result when first tested, but upon retesting the sample (per the TaqPath COVID-19 protocol) the sample gave a positive result. All samples gave expected internal control results (amplification of MS2 phage nucleic acid) to monitor the integrity of nucleic acid extractions and RT-PCR for each sample. There were minimal Ct changes over the 28 days of the experiment with the highest deviation shown for N gene amplification on day 28, however the observed difference was still within the study's acceptance criteria. The S gene target did show a decreased overall reactivity, however this is consistent and expected due to SARS-CoV-2 viral mutation as discussed above. Since the

TaqPath COVID-19 assay requires 2/3 targets to amplify for positive results, this decreased reactivity did not impact assay performance.

These test results fall within the established acceptance criteria and indicate samples are stable in the Spectrum inactivating media for up to 28 days.

6. Detection Limit:

Limit of Detection (LoD) testing was conducted to determine the lowest concentration of whole genome (Genome Equivalent Copies; GEC) that contains measurable nucleic acids that can be repeatedly recovered from the transport media with greater than 95% accuracy. LoD was obtained by spiking the Spectrum SDNA-1000 collecting device containing negative matrix (saliva + preservative) with various concentrations of SARS-CoV-2 before performing RNA extractions using the MagMAX Viral/Pathogen II (MVP II) Nucleic Acid Isolation Kit on the KingFisher Flex Purification System. Viral RNA was subsequently amplified using the TaqPath COVID-19 Combo Kit using the Applied Biosystems QuantStudio 5 (QS5) Fast Real-Time PCR System. The RNA extraction and RT-PCR kits contained an internal MS2 Phage control to monitor the integrity of nucleic acid extraction and RT-PCR for each specimen. Ct values and detection of virus were determined using the EUA IFU guidelines and the Applied Biosystems COVID-19 Interpretative Software v2.5.

For the COVID-19 TaqPath assay, Ct values <37 are considered positive for SARS-CoV-2. Ct values of 37 to <40 were considered inconclusive and those samples retested to determine SARS-CoV-2 status, as per the protocol Qualitative Detection of Nucleic Acid from SARS-CoV-2 using the COVID-19 TaqPath Combo Kit (JB-PRO-000105, v.2.1). Three gene targets in SARS-CoV-2 were subjected to amplification (ORF1ab, N gene, and S gene), with a sample considered positive if 2/3 of these gene targets were amplified.

Preliminary LoD testing was initially performed by spiking multiple concentrations of SARS-CoV-2 into clinically negative matrix (saliva) to achieve final concentrations of 0, 10, 20, 50, 100, 250, and 500 GEC/reaction. Five samples of each of these contrived concentrations were tested with results shown below in Table 3.

Table 3. Preliminary LoD Estimation Results

GEC/reaction	Orf1ab detection	ORF1a Mean Ct	N gene detection	N gene Mean Ct	S gene detection	S gene Mean Ct	SARS-CoV-2 positive result
0	0/5	ND	0/5	ND	0/5	ND	0/5
10	5/5	34.45	0/5	ND	0/5	ND	5/5
20	5/5	32.89	5/5	33.97	0/5	ND	5/5
50	5/5	31.81	5/5	31.90	5/5	33.45	5/5
100	5/5	30.43	5/5	30.30	5/5	31.66	5/5
250	5/5	29.44	5/5	29.06	5/5	30.13	5/5
500	5/5	29.05	5/5	28.84	5/5	29.67	5/5

The estimated LoD based on the preliminary results was identified as 20 GEC/reaction, but one of the three gene targets was successfully amplified (ORF1ab) results from the 10

GEC/reaction dilution. To further confirm this preliminary LoD assessment, both 15 and 20 GEC/reactions were evaluated in a set of 20 replicates (see Table 4). A positive result for a sample was defined as amplification of at least 2 of the 3 gene targets from SARS-CoV-2 (ORF1ab, N gene, S gene) per the IFU from the authorized device.

Confirmatory testing supported an LoD of 20 GEC/reaction with 20/20 replicates for amplification of ORF1ab and 20/20 replicates for N gene, which met the pre-defined criteria for RT-PCR reactions (greater than 95% accuracy for detecting 2/3 targets in samples). However, for the 15 GEC/reaction dilution only 16/20 samples met the requirement of 2/3 targets amplified (80% accuracy), which fell below the pre-defined criteria. Therefore, 20 GEC/reaction is the established LoD.

Table 4. Confirmatory LoD Study Results

GEC/reaction	Orf1ab detection	ORF1a Mean Ct	N gene detection	N gene Mean Ct	S gene detection	S gene Mean Ct	SARS-CoV-2 positive result
15	20/20	33.92	16/20	35.29	0/20	ND	0/5
20	20/20	33.81	20/20	35.23	2/20	36.77	5/5

Ct values for each of the three amplified genes of the 20 replicates of the confirmatory tests for the 20 GEC/reaction concentration are shown in Table 5.

Table 5. LoD Confirmation Study - Individual Replicate Ct Values

	C_t Value		
Replicate	**ORF1ab**	**N gene**	**S gene**
1	36.14	34.83	
2	33.09	35.84	
3	33.35	36.09	
4	30.03	36.79	
5	34.50	34.48	
6	32.18	36.48	
7	34.60	34.79	36.66
8	33.55	34.42	
9	33.86	35.72	
10	34.71	36.63	
11	34.06	33.37	
12	34.24	36.01	36.87
13	33.85	34.47	37.61
14	32.80	35.45	37.83
15	34.17	34.02	
16	33.73	34.94	37.64
17	33.93	36.54	39.65
18	35.94	34.37	
19	33.15	35.12	
20	34.30	34.28	
AVG:	33.81	35.23	37.71
SD:	1.29	0.99	1.06

Note: The 69-70del mutation interferes with the detection of the S gene target by the TaqPath COVID-19 assay, which may have rendered the S gene target as not detected ("S gene target failure"). But due to the multi-target test design, the overall test performance of the TaqPath assay is not impacted.

In summary, the testing performed established 250 GEC/ml (20 GEC/reaction) as the LoD or the lowest concentration tested that resulted in > 95% accuracy of detection of contrived samples.

7. Inactivation

An inactivation study was conducted to verify that the Spectrum SDNA-1000 collecting device inactivates SARS-CoV-2 as efficiently as the predicate device. Specifically, viral inactivation testing of SARS-CoV-2 was conducted using a Virucidal Suspension Test. Viral preparations containing SARS-CoV-2 isolate (B.1.1.529 *Omicron* variant) of 3.2 X 10^6 $TCID_{50}$ (approximately 2.24 X 10^6 PFU) or lower were combined with various combinations of Spectrum inactivating media and/or neutralization fluid (for the inactivating media).

Following treatment of virus, 10-fold dilutions were made of each preparation and incubated with Vero E6 kidney cells grown to 80-90% confluency. After a one hour incubation with the treatments listed above, cells were returned to culture conditions and incubated before cytopathic effects were determined visually with an inverted compound microscope.

The acceptance criteria for the inactivation study included at least a 4 $\log_{10}$ of $TCID_{50}$ would be recovered from the virus control (no inactivation) andat least a 3 $\log_{10}$ reduction in titer could be demonstrated in Spectrum media

Three different lots of devices were tested for viricidal activity, with each test performed in two replicates per device. Plating of each replicate was performed four times.

Under the experimental conditions described above the Spectrum inactivating media inactivates SARS-CoV-2 by an average of $\geq$4.00 $\log_{10}$ (>99.99%) following a 10-second exposure and by an average of $\geq$4.00 $\log_{10}$ (>99.99%) following a 60-minute exposure.

8. Assay Cut-Off:

   N/A

9. Accuracy (Instrument):

   N/A

10. Carry-Over:

    N/A

B Comparison Studies:
N/A

C Clinical Studies:
N/A

D Clinical Cut-Off:
N/A

E Expected Values/Reference Range:
N/A

F Other Supportive Instrument Performance Characteristics Data:
N/A

VIII Proposed Labeling:

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

IX Conclusion:

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