EMERGENCY USE AUTHORIZATION (EUA) SUMMARY
P23 LABS TAQPATH SARS-COV-2 ASSAY
(P23 Labs)

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

(The P23 Labs TaqPath SARS-CoV-2 Assay will be performed in the P23 Labs, a Clinical Laboratory Improvement Amendments of 1988 (CLIA), 42 U.S.C. §263a certified high-complexity laboratory, per the Standard Operating Procedures that were reviewed by the FDA under this EUA).

INTENDED USE

The P23 Labs TaqPath SARS-CoV-2 Assay is a real-time reverse transcription polymerase chain reaction (rRT-PCR) test intended for the qualitative detection of nucleic acid from SARS-CoV-2 in oropharyngeal (throat) swab, nasopharyngeal swab, anterior nasal and mid-turbinate nasal swabs, nasopharyngeal wash/aspirate or nasal aspirate specimens as well as bronchoalveolar lavage (BAL) specimens from individuals suspected of COVID-19 by their healthcare provider.

This test is also for use with saliva specimens that are either self-collected at home or in a healthcare setting under the supervision of a healthcare provider (HCP), or collected by a HCP, using the OMNIgene·ORAL OM-505 Collection Device, when determined to be appropriate by a HCP.

Testing is limited to P23 Labs, LLC (P23), Little Rock, AR, that is certified under Clinical Laboratory Improvement Amendments of 1988 (CLIA), 42 U.S.C. §263a, to perform high complexity tests.

Results are for the detection and identification of SARS-CoV-2 RNA. The SARS-CoV-2 RNA is generally detectable in 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. Negative results for SARS-CoV-2 RNA from saliva should be confirmed by testing of an alternative specimen type if clinically indicated.

Testing with the P23 Labs TaqPath SARS-CoV-2 assay is intended for use by qualified laboratory personnel specifically instructed and trained in the techniques of real-time PCR assays. The assay is intended for use under the Food and Drug Administration's Emergency Use Authorization.

Please refer to FDA's FAQs on Diagnostic Testing for SARS-CoV-2 for additional information regarding the collection of appropriate specimen types for the detection of SARS-CoV-2.

DEVICE DESCRIPTION AND TEST PRINCIPLE

The P23 Labs TaqPath SARS-CoV-2 Assay is a real-time reverse transcription polymerase chain reaction (rRT-PCR) test. The assay uses primers and probes that were developed and validated under the Emergency Use Authorization (EUA) for the Thermo Fisher TaqPath COVID-19 Combo Kit and are designed to detect RNA from SARS-CoV-2 in respiratory specimens from patients as recommended for testing by public health authority guidelines. The purpose of this EUA request is to enable testing of additional specimen types, including saliva collected by or under the supervision of a healthcare provider in a healthcare setting (e.g., assisted living setting, doctor's office, drive through collection) or home environment setting. Patients are evaluated by their healthcare provider and determined to be appropriate for saliva collection using the CDC screening guidelines (https://www.cdc.gov/coronavirus/2019-nCoV/hcp/clinical-criteria.html).

Anterior nasal swabs, mid-turbinate nasal swabs, oropharyngeal (throat) swabs, nasopharyngeal swabs, nasopharyngeal wash/aspirate or nasal aspirate specimens, and BALs should be collected, transported and stored according to standard procedures. All swab and wash/aspirate/BAL collections (Copan ESwab™ 480C flocked swab in Amies Media [Cat # 480C.US], Sterile Container-Corning TP52C002, ThermoFisher Cat # 07-202-025) will be performed directly by a trained healthcare provider in a healthcare setting. The acceptable transport media for these collected swab types are UTM and liquid Amies (MTM). Once swab/wash/aspirate/BAL samples are received in the laboratory, specimens may be refrigerated (2-8°C) for an additional 3 days before they are extracted and processed if specimens cannot be processed immediately upon receipt. Saliva specimens must be collected, transported, and stored using the OMNIgene®·ORAL OM-505 collection device. Collection of saliva will occur under the supervision of a healthcare professional or by a trained HCP. Saliva specimens must be transported and stored at ambient temperature and tested within 120 hours of collection.

RNA extraction for all specimen types is performed using the MagMAX™ Viral/Pathogen Nucleic Acid Isolation Kit on the automated KingFisher™ Duo Primer Purification System (software v4.0). The input sample volume is 400µL, the elution volume is 50µL using RNase-free water.

Reverse transcriptase-PCR (RT-PCR) is performed using the Applied Biosystems TaqPath COVID-19 Combo Kit with 5µL of the extracted sample.

REAL-TIME PCR INSTRUMENT USED WITH THE TEST

The P23 Labs TaqPath SARS-CoV-2 Assay is for use with the ThermoFisher Applied Biosystems QuantStudio 5 Real-Time PCR System equipped with software v1.3 (Expression Suite).

REAGENTS AND MATERIALS

Table 1. Reagents and materials required for use of the P23 Labs TaqPath SARS-CoV-2 Assay

Reagent	Manufacturer	Catalogue #
MagMAX™ Viral/Pathogen Nucleic Acid Isolation Kit	ThermoFisher Scientific	A43252 or A48310
KingFisher™ Duo Prime Purification System	ThermoFisher Scientific	5400110
TaqPath COVID-19 Combo Kit	ThermoFisher Scientific	A47814
TaqPath COVID-19 Control Kit	ThermoFisher Scientific	A47816
TaqPath 1-Step Multiplex Master Mix	ThermoFisher Scientific	A28535
96 well Deep Well Plates	PerkinElmer	43001-0120
384 well PCR plate	ThermoFisher Scientific	4483273
Optical adhesive PCR plate cover	ThermoFisher Scientific	4311971
Nuclease-free water	--	--
Ethanol (96-100%)	--	--

CONTROLS TO BE USED WITH THE COVID-19 RT-PCR

The controls supplied with the ThermoFisher - Applied Biosystems TaqPath COVID-19 Combo Kit are described in Table 2.

Table 2. Controls supplied with the Applied Biosystems TaqPath COVID-19 Combo Kit and used with the P23 Labs TaqPath SARS-CoV-2 Assay

Control Type	Purpose	Frequency of Testing
Positive Control	To monitor the integrity of the RT-PCR reagents and process	Once per run of RT-PCR
Internal (MS2 Phage)	To monitor the integrity of nucleic acid extraction and RT-PCR for each specimen	Added to each specimen and the Negative Control prior to extraction
No Template Control	To monitor for contamination of extraction and assay reagents	Once per run of RT-PCR

In addition to the controls in Table 2, a negative extraction control is also run per batch of specimens to monitor for cross-contamination during RNA extraction and RT-PCR. This control is no longer supplier with the TaqPath COVID-19 Combo Kit; however, P23 Labs uses a characterized negative human specimen as their negative extraction control.

The results from the controls are interpreted according to the criteria shown in Table 3. If the results obtained with the Positive, Negative, and No Template Controls do not meet the criteria shown, the results from the entire batch of samples are considered invalid and repeat testing must be performed using residual extracted nucleic acid. If any of the above controls do not exhibit the expected performance as described, the assay may have been improperly set up and/or executed improperly, or reagent or equipment malfunction could have occurred. Invalidate the run and re-test. Based on the scientific review of the failure, samples could be repeated from extraction or samples could be repeated from previously extracted material.

INTERPRETATION OF RESULTS

All test controls should be examined prior to interpretation of patient results. If the controls are not valid, the patient results cannot be interpreted (Refer to Table 3 for a summary of control results).

1) ***COVID-19 RT-PCR test Controls – Positive, Negative, and Internal:***

- **MS2 (Internal Positive Control);** MS2 in a sample indicates that PCR amplification occurred in the well. The presence of MS2 and no detectable SARS-CoV-2 during the analysis indicates that proper RNA extraction and amplification occurred, however, no SARS-CoV-2 is present. If SARS-CoV-2 is present in the specimen, amplification of the target RNA may reduce or abrogate MS2 amplification. In this case, the amplified SARS-CoV-2 indicates proper RNA extraction and amplification. Therefore, MS2 may or may not be detectable in a valid test on patient specimens.

- **External Positive Control;** The positive control must be positive for all three SARS-CoV-2 targets, i.e., the ORF1ab, the N Protein, and the S Protein genes and amplification must have a Ct <37 in order for the test result to be valid. The positive control does not contain MS2.

- **Nuclease-Free Water (Negative Control; NTC);** The negative control must be negative (undetermined) in order for the test result to be valid.

- **Negative Extraction Control (NEC);** Although not supplied with the Thermo TaqPath Combo Kit, P23 Labs runs a NEC with each batch of samples. The NEC should only show an amplification curve for MS2 with a Ct of less than or equal to 37 but must be negative for all SARS-CoV-2 targets (Ct undetermined).

Table 3. Ct values for controls that must be observed to obtain valid results

Control	Ct Value (Optical Channel)			
	N Gene (VIC)	S Gene (ABY)	ORF1ab (FAM)	MS2 Phage (JUN)
Negative Extraction Control	Undetermined	Undetermined	Undetermined	≤37
Positive Control	<37	<37	<37	Undetermined [1]
No Template Control	Undetermined	Undetermined	Undetermined	Undetermined [1]
MS2 Internal Control	Any	Any	Any	<37

[1] The MS2 Phage Internal Control is not added to the Positive Control or No Template Control and no signal should be obtained.

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. Please see the table below (Table 4) for guidance on interpretation and reporting of results.

Table 4. Result interpretation for patient samples

Ct Value (Optical Channel)				Result Interpretation
N Gene (VIC)	S Gene (ABY)	ORF1ab (FAM)	MS2 Phage (JUN)	
Undetermined	Undetermined	Undetermined	≤37	Negative
Two of three <37			Any value	Positive
One of three <37			Any value	Positive
Undetermined	Undetermined	Undetermined	Undetermined	Re-test [1]

[1] Re-test required from the residual extracted sample and by processing a new aliquot of the original sample if volume permits; if the re-test result is the same as the original then report result as "inconclusive," and consider collecting a new specimen.

PERFORMANCE EVALUATION

1) Analytical Sensitivity

The LoD was determined using SARS-CoV-2 genomic RNA fragments purchased from SeraCare (AccuPlex™ SARS-CoV-2, cat # 0505-0126) that was diluted in SARS-CoV-2 negative pooled nasopharyngeal swab matrix (eSwab; NP swab suspended in liquid Amies). An initial estimate of the LoD with the Applied Biosystems QuantStudio 5 Real-Time PCR System was obtained by testing ten replicates at each of four different target levels: 50, 20, 10, and 2.5 copies/µL. The lowest level at which all ten replicates were positive for all three SARS-CoV-2 targets was 10 copies/µL (Table 5). The estimated LoD was confirmed by testing an additional 20 extraction replicates at the same target level. All 20 replicates produced the expected results for each SARS-CoV-2 target, and the LoD was therefore confirmed to be 10 copies/µL (Table 6).

Table 5. Preliminary LoD determination results

Concentration (copies/uL)	ORF1ab		N Gene		S Gene		MS2 (IC)	
	Detection Rate (%)	Mean Ct	Detection Rate (%)	Mean Ct	Detection Rate (%)	Mean Ct	Detection Rate (%)	Mean Ct
50	10/10 (100)	29	10/10 (100)	26	10/10 (100)	27	10/10 (100)	24
20	10/10 (100)	30	10/10 (100)	29	10/10 (100)	27	10/10 (100)	24
10	10/10 (100)	32	10/10 (100)	32	10/10 (100)	26	10/10 (100)	24
5	7/10 (70)	33	9/10 (90)	28	10/10 (100)	24	10/10 (100)	24
2.5	8/10 (80)	31	9/10 (90)	30	10/10 (100)	24	10/10 (100)	24
Negative	0/0 (0)	NA	0(0)	NA	0(0)	NA	10/10 (100)	24

Table 6. Confirmatory LoD study for nasopharyngeal swab specimen

Concentration (copies/uL)	ORF1ab		N Gene		S Gene	
	Detection Rate (%)	Mean Ct	Detection Rate (%)	Mean Ct	Detection Rate (%)	Mean Ct
10	19/20 (95)	32	20/20 (100)	31	20/20 (100)	29

To validate the use of saliva as an acceptable specimen type, a bridging study (confirmatory LoD study) was completed between nasopharyngeal swabs and healthcare provider supervised self-collection of saliva into the OM-505 device. This study used pooled, negative saliva

collected in OM-505 and pooled, negative NP swab matrix spiked with SeraCare material at 2X and 5X LoD (20 copies/µL and 50 copies/µL). Twenty individual extraction replicates of both specimen types at both 2X and 5X LoD were run on P23 Labs TaqPath COVID-19 Assay. The data demonstrated that the LoDs for NP swabs and saliva were equivalent.

Table 7. Bridging study between NP swab and saliva collected in OM-505 device (2X LoD)

Concentration (copies/uL)	ORF1ab		N Gene		S Gene		MS2 (IC)	
	Detection Rate (%)	Mean Ct	Detection Rate (%)	Mean Ct	Detection Rate (%)	Mean Ct	Detection Rate (%)	Mean Ct
NP Swab-eSwab (Liquid Amies)								
20	20/20 (100)	32	19/20 (95)	32	20/20 (100)	32	20/20 (100)	24
Saliva in OM-505								
20	19/20 (95)	32	20/20 (100)	32	20/20 (100)	32	20/20 (100)	24

Table 8. Bridging study between NP swab and saliva collected in OM-505 device (5X LoD)

Concentration (copies/uL)	ORF1ab		N Gene		S Gene		MS2 (IC)	
	Detection Rate (%)	Mean Ct	Detection Rate (%)	Mean Ct	Detection Rate (%)	Mean Ct	Detection Rate (%)	Mean Ct
NP Swab-eSwab (Liquid Amies)								
50	20/20 (100)	32	20/20 (100)	32	20/20 (100)	32	20/20 (100)	24
Saliva in OM-505								
50	20/20 (100)	32	20/20 (100)	32	20/20 (100)	32	20/20 (100)	24

2) Analytical Specificity

Inclusivity

The P23 Labs TaqPath SARS-CoV-2 Assay is a modification of the previously authorized ThermoFisher Applied Biosystems TaqPath COVID-19 Combo Kit. The assay targets specific genomic regions of the SARS-CoV-2 nucleocapsid (N) gene, spike (S) gene, and ORF1ab region. Inclusivity was demonstrated under the original ThermoFisher EUA and a right of reference to use their inclusivity data was provided to P23 Labs on May 12, 2020. Briefly, the primers and probes were mapped to 185 complete SARS-CoV-2 genomes that were available in the GenBank and GISAID (Global Initiative on Sharing All Influenza Data) databases as of March 5, 2020. For all primers and probes, there was 100% homology to each of the SARS-CoV-2 sequences analyzed, with one exception; a single base mismatch (95.6% homology) with the reverse primer for ORF1ab in sequence EPI_ISL_407084 (BetaCoronavirus/Japan/AI/I-004/2020). The mismatch is located at the 5' end of the primer and is not expected to affect test performance

Cross-reactivity

The analytical specificity of the P23 Labs TaqPath SARS-CoV-2 Assay was demonstrated *in silico* under the original EUA for the ThermoFisher Applied Biosystems TaqPath COVID-19 Combo Kit. As stated previously, a right of reference to use ThermoFisher's exclusivity data was given to P23 Labs. The analysis included evaluation of the primer and probe homology with the 43 organisms and viruses listed in Table 9. Based on this analysis, significant

amplification of non-target sequences that could result in cross-reaction (false-positive results) or interference (false-negative results were considered unlikely to occur.

Table 9. Organisms and viruses evaluated for potential cross-reaction and/or interference with the Applied Biosystems TaqPath COVID-19 Combo Kit

Viruses	Bacteria
Adenovirus	*Bacillus anthracis*
Enterovirus	*Bordetella pertussis*
Human coronavirus 229E	*Chlamydophila pneumoniae*
Human coronavirus HKU1	*Chlamydophila psittaci*
Human coronavirus NL63	*Corynebacterium diphtheriae*
Human coronavirus OC43	*Coxiella burnetii*
Human Metapneumovirus (hMPV)	*Haemophilus influenzae*
Influenza A, B and C	*Legionella* (non-pneumophila)
MERS-coronavirus	*Legionella pneumophila*
Parainfluenza 1-4	*Leptospira* sp.
Parechovirus	*Moraxella catarrhalis*
Respiratory Syncytial Virus A and B	*Mycobacterium tuberculosis*
Rhinovirus/Enterovirus	*Mycoplasma pneumoniae*
SARS-coronavirus	*Neisseria elongata* and *Neisseria meningitidis*
Yeast/Fungus	*Pseudomonas aeruginosa*
Candida albicans	*Staphylococcus aureus*
Pneumocystis jirovecii	*Staphylococcus epidermidis*
	Streptococcus pneumoniae
	Streptococcus pyogenes
	Streptococcus salivarius

3) Clinical Evaluation

Saliva (Paired NP Swab and Saliva Clinical Study)

A study was performed to evaluate the use of saliva as a specimen type for detection of SARS-CoV-2 in patients who were suspected of COVID-19. The study was conducted with symptomatic patients from one ambulatory care center who were each provided with instructions for self-collection of saliva using the OMNIgene®·ORAL OM-505 collection device. Self-collection of saliva samples was performed under the observation of a healthcare provider, without intervention, who subsequently (within 10 minutes) also collected a nasopharyngeal swab from each patient for parallel testing for SARS-CoV-2. Patients were given the option to ask the HCP for assistance or to complete the collection, if they had remaining questions not covered by the instructions. For this study, patients with previously detected SARS-CoV-2 positive results were re-tested between 7-21 days of an initial positive test using paired samples collected in the clinic as previously described within 10 minutes of each other. The swabs were placed in viral transport medium for shipment to the testing laboratory. Both the saliva and swabs were transported at ambient temperature and tested using the P23 Labs TaqPath SARS-CoV-2 Assay within 48 hours of collection. A summary of the results of the study is presented in Tables 10 and 11 below.

There was 100% positive and negative agreement between the results obtained from testing saliva and those obtained from nasopharyngeal swab. Of the 31 previously reported positive NP swab samples, all 31 paired NP and saliva specimens produced positive results for the N gene (31/31; 100%), whereas the S target was positive for 30/31 samples (96.8%) for both NP and saliva samples. For the ORF1ab target, 30/31 (96.8%) NP samples were positive and 29/31 tested saliva samples were positive. According to the result algorithm described in Table 4 above, a sample is considered positive for SARS-CoV-2 RNA if amplification is detected with at least one of the three SARS-CoV-2-specific target sequences. The results of the clinical evaluation with paired nasopharyngeal swabs and saliva were therefore considered acceptable.

Table 10. Summary of results obtained from parallel testing of nasopharyngeal swab samples and saliva from patients suspected of COVID-19, stratified by measurand

Number of Patients	Sample Type	Analysis	Target (Optical Channel)			
			N Gene (VIC)	S Gene (ABY)	ORF1ab (FAM)	MS2 (JUN)
31 NP positive	NP swab	Positive (%)	31/31 (100)	30/31 (96.8)	30/31 (96.8)	31/31 (100)
		Median Ct	28.3	28.6	29.0	24.8
	Saliva	Positive (%)	31/31 (100)	30/31 (96.8)	29/31 (93.5)	31/31 (100)
		Median Ct	28.1	28.4	29.9	24.6
11 NP negative	NP swab	Positive (%)	0 (0)	0 (0)	0 (0)	11/11 (100)
		Median Ct	N/A	N/A	N/A	24.9
	Saliva	Positive (%)	0 (0)	0 (0)	0 (0)	11/11 (100)
		Median Ct	N/A	N/A	N/A	24.8

NP: Nasopharyngeal; N/A: Not applicable

Table 11. Summary of qualitative results obtained from parallel testing of nasopharyngeal swab samples and saliva from patients suspected of COVID-19

		Nasopharyngeal Swab		
		Positive	Negative	Total
Saliva	Positive	31	0	31
	Negative	0	11	11
	Total	31	11	42
Positive Agreement		100% (31/31); 88.98-100.00% [1]		
Negative Agreement		100% (11/11); 74.12-100.00%		

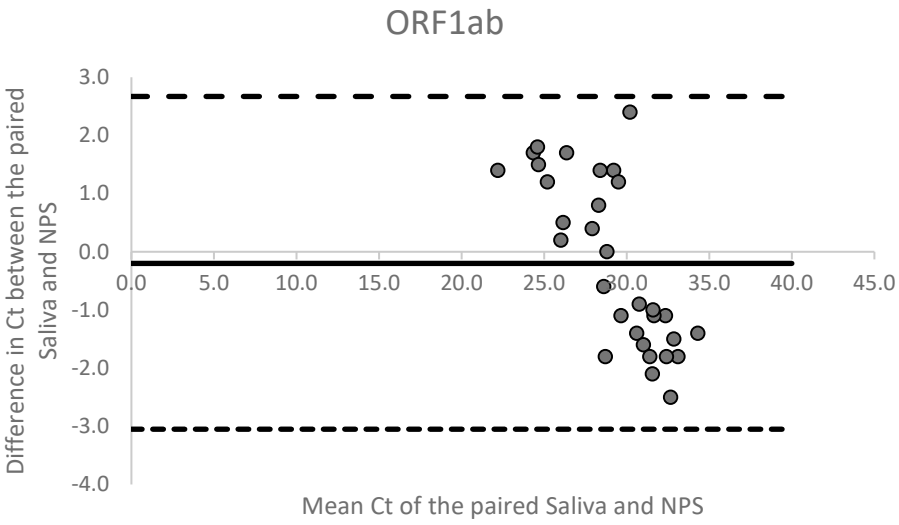
[1]Two-sided 95% score confidence intervals

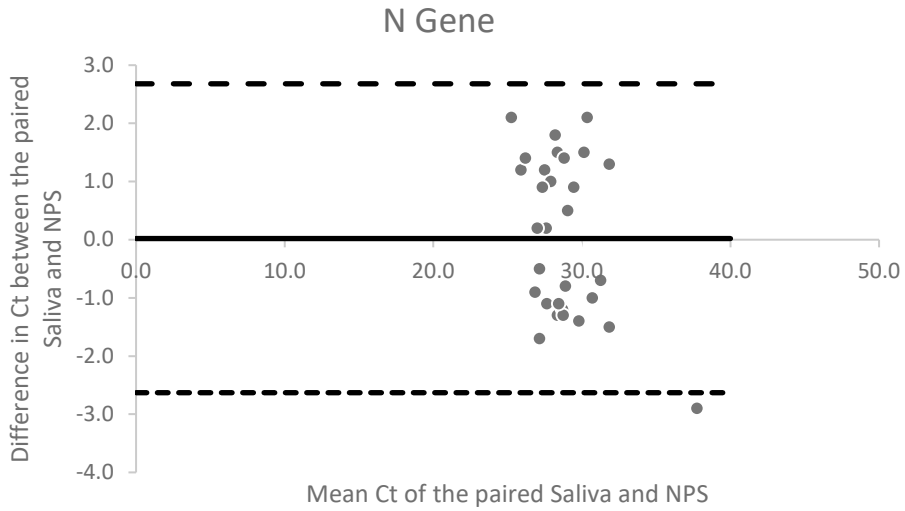
Evaluation of Ct Cycle Differences Among Paired Samples

The difference between Ct values from the paired samples are shown in Figure 1. Overall median Ct values were similar for saliva and nasopharyngeal swab and demonstrated that there was no systematic difference between testing saliva and NP swab specimens collected from the same patient within 10 minutes of one another. Figure 2 shows a

correlation analysis among the individual Ct values for the 31 paired positive NP and saliva specimens. According to the regression analysis in Figure 2, there appears to be a correlation between the Ct values obtained with the two sample types. The slope of each regression line is close to 1, indicating that the Ct values between the paired NP swabs and saliva samples trend in the same direction as supported by the Bland Altman plots in Figure 1. Based on the R squared values for the 3 assay targets (ORF1ab, N, and S genes), there is evidence of a correlation between Ct values from the different sample types collected from the same patient. The results support the use of saliva as a specimen type for use with the P23 Labs TaqPath SARS-CoV-2 Assay.

Figure 1. Bland Altman Plot for each SARS-CoV-2 target gene





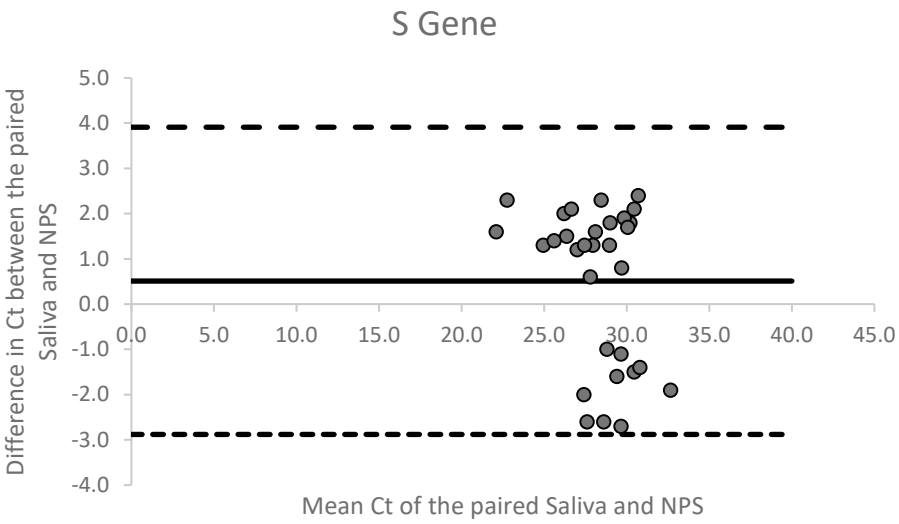
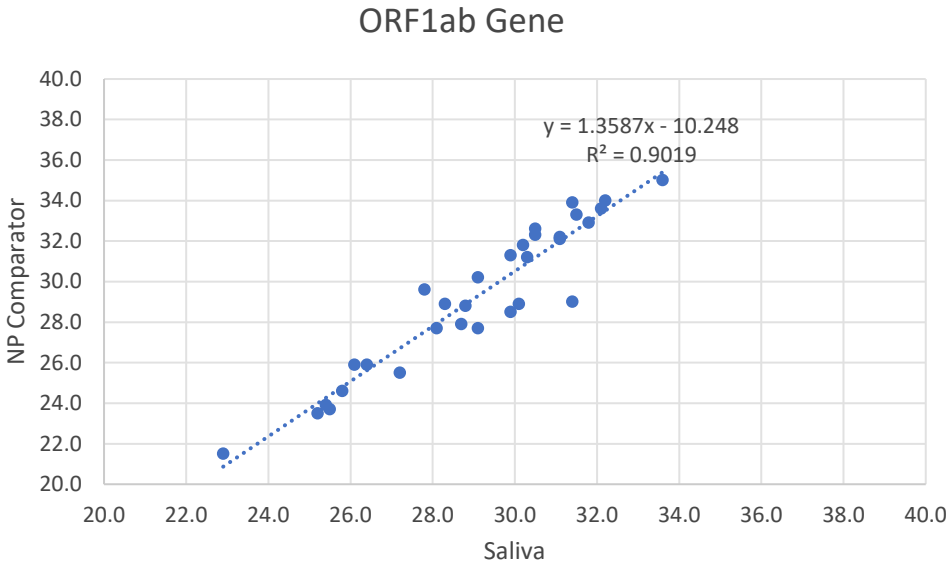
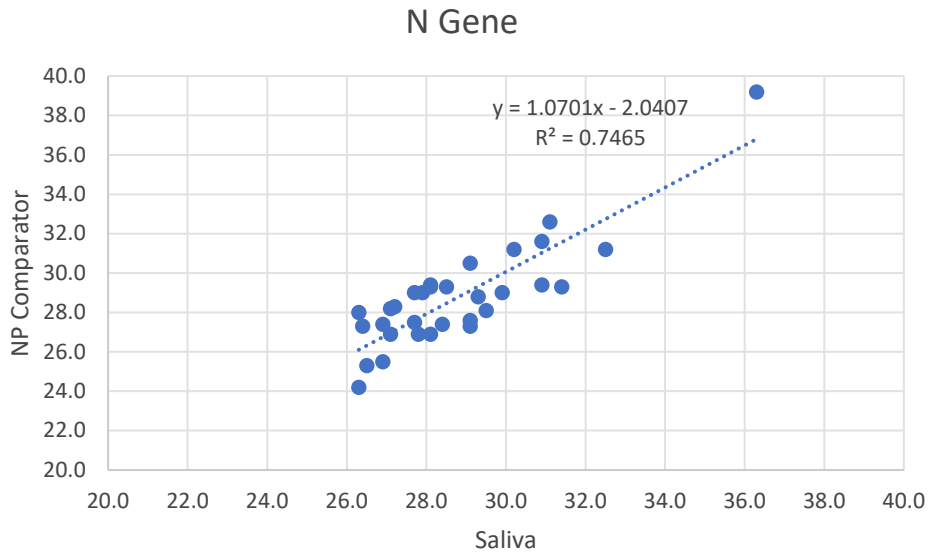
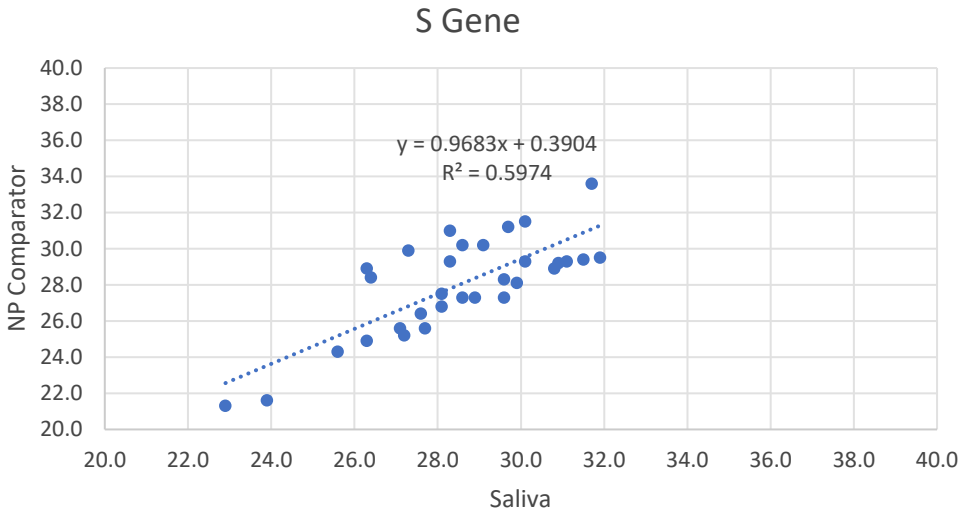


Figure 2. Regression analysis for individual Ct values for the paired NP and saliva specimens







Contrived Clinical Study

The performance of the P23 Labs SARS-CoV-2 Assay was also evaluated in a contrived clinical study using leftover, negative clinical nasopharyngeal swab matrix and saliva (collected in the OM-505 collection device) spiked with the SeraCare material. Individual, unique NP swab specimens and saliva specimens were used for spiking. A total of 30 contrived positives prepared at 2X and 5X LoD and 40 contrived negative

samples for both NP swabs and saliva were tested with the P23 Labs assay. A summary of the results of the study is provided in Table 12. All 40 (100%) contrived negative NP and saliva results were non-reactive and produced the expected results. Of the 30 contrived positive NP and saliva samples, all 30 produced positive results for all three assay targets (N, S, and ORF1ab). The results of the contrived clinical evaluation with nasopharyngeal swabs and saliva were considered acceptable.

Table 12. Contrived Clinical Study Summary Results for NP and Saliva Samples

Type of Sample	Concentration (copies/µL)	Number of samples	Detection Rate (%)		
			N Gene	ORF1ab	S Gene
Nasopharyngeal Swab	2X LoD (20 copies/µL)	25	25/25 (100%)	25/25 (100%)	25/25 (100%)
	5X LoD (50 copies/µL)	5	5/5 (100%)	5/5 (100%)	5/5 (100%)
	Negative Clinical Samples	40	0/40; UND	0/40; UND	0/40; UND
Saliva collected in OM-505	2X LoD (20 copies/µL)	25	25/25 (100%)	25/25 (100%)	25/25 (100%)
	5X LoD (50 copies/µL)	5	5/5 (100%)	5/5 (100%)	5/5 (100%)
	Negative Clinical Samples	40	0/40; UND	0/40; UND	0/40; UND

Clinical Confirmation

The first 5 positive and first 5 negative nasopharyngeal specimens as determined by P23 Labs using the P23 Labs TaqPath SARS-CoV-2 Assay were also tested by the Arkansas State Health Department using the previously authorized CDC 2019-Novel Coronavirus (2019-nCoV) Real-Time RT-PCR Diagnostic Panel. There was 100% (5/5) positive and negative agreement for the specimens tested. These results are acceptable and support use of the by P23 Labs TaqPath SARS-CoV-2 Assay for testing clinical specimens.

4) Simulated Shipping Study with the OMNIgene®·ORAL OM-505 Saliva Collection Device

To support home use of the OMNIgene®·ORAL OM-505 collection device, a simulated shipping study was performed that was designed to evaluate the effect of temperature variation on the stability of SARS-CoV-2 RNA during transport of saliva specimens. The shipping study was designed to simulate shipping at room temperature as well as the extreme temperature conditions that could be experienced during the summer and winter months. See Tables 13 and 14 for summer and winter thermal profiles evaluated in this study. Room temperature stability was evaluated by physically shipping samples via UPS from Little Rock, AR to Memphis, TN over 3 days (72 hours) and 5 days (120 hours) at ambient (room temperature) conditions.

Simulated sample stability and shipping studies were performed using both contrived positive saliva specimens at 2X and 5X LoD concentrations as well as previously reported clinical positive and negative patient specimens. After the samples underwent

the thermal excursions, they were equilibrated to room temperature, extracted, and tested with the P23 Labs TaqPath assay.

Table 13. Summer temperature excursion

Temperature	Cycle Period	Cycle Period Hours	Total Time Hours[1]
40°C	1	8	8
22°C	2	4	12
40°C	3	2	14
30°C	4	36	50
40°C	5	6	56

[1] Sum of cycle periods

Table 14. Winter temperature excursion

Temperature	Cycle Period	Cycle Period Hours	Total Time Hours[1]
-10°C	1	8	8
18°C	2	4	12
-10°C	3	2	14
10°C	4	36	50
-10°C	5	6	56

[1] Sum of cycle periods

Shipping Study Using Contrived Saliva Samples

Contrived samples were prepared using pooled known negative patient sample matrix and spiking with quantified SeraCare material to establish low positives of 2x LoD (LoD previously established as 10 copies/µL) and high positive saliva samples of 5x LoD. For the spiked saliva, donated pathogen free saliva was pooled. Saliva was shown to be negative as each donor was asymptomatic and a prior NP swab was tested and shown to be negative. The donor saliva was screened by an outside laboratory using the TaqPath Assay 48 hours before use in the contrived study.

Testing included 25 contrived spiked saliva samples; 20 low positive and 5 high positive saliva samples collected in the OM-505 collection device. The contrived positive and negative saliva samples were either physically shipped at room temperature conditions or stored for the duration of each simulated shipping study as shown in Table 12 and 13. At the conclusion of each thermal profile, the samples were equilibrated to room temperature, extracted using the MagMAX™ kit, and retested with the P23 Labs TaqPath SARS-CoV-2 Assay. Results were compared to those reported upon initial testing when specimens were received at time 0 (day 0, room temperature).

Twenty out of 20 low positive samples (100%) and 5/5 high positive contrived samples (100%) were reported as positive after exposure to room temperature as well as the summer and winter temperature cycles. The mean and standard deviation of the Ct values for each gene target were similar before and after each simulated shipping scenario (within ~3 Cts), with no evidence of significant degradation of the SARS-CoV-2 RNA. All SARS-CoV-2 negative specimens were reported as negative after enduring ambient

temperature and extreme temperature conditions ((no amplification of N, ORF1ab, or S genes).

A summary of the mean Ct values observed for each SARS-CoV-2 specific target gene is provided in Table 15.

Table 15. Summary of results from the simulated shipping study using contrived samples

Sample Group	Test Point	N	Mean Ct (Standard Deviation)			Positive (%)
			N Gene	ORF1ab	S Gene	
Negative	Day 0 (RT)[1]	20	Und[6]	Und[6]	Und[6]	0 (0)
	Day 3 (RT)[2]	20	Und[6]	Und[6]	Und[6]	0 (0)
	Day 5 (RT)[3]	20	Und[6]	Und[6]	Und[6]	0 (0)
	Summer[4]	20	N/A	N/A	N/A	0 (0)
	Winter[5]	20	N/A	N?A	N/A	0 (0)
Low Positive 2X LoD 20 copies/µL	Day 0 (RT)	20	22.9 (1.9)	24.3 (2.8)	26.1 (1.6)	20/20 (100)
	Day 3 (RT)	20	23.3 (1.6)	25.5 (1.0)	26.9 (2.1)	20/20 (100)
	Day 5 (RT)	20	26.9 (0.9)	27.4 (1.8)	28.1 (1.3)	20/20 (100)
	Summer	20	26.8 (1.8)	27.8 (1.3)	27.1 (1.9)	20/20 (100)
	Winter	20	26.3 (2.1)	26.9 (0.6)	26.9 (1.7)	20/20 (100)
High Positive 5X LoD 50 copies/µL	Day 0 (RT)	5	22.3 (1.1)	23.9 (1.6)	24.7 (2.0)	5/5 (100)
	Day 3 (RT)	5	24.1 (1.6)	24.8 (1.2)	25.3 (1.9)	5/5 (100)
	Day 5 (RT)	5	25.1 (0.9)	25.9 (13)	27.3 (1.3)	5/5 (100)
	Summer	5	25.9 (1.1)	25.5 (1.8)	24.3 (2.3)	5/5 (100)
	Winter	5	25.9 (1.8)	25.4 (1.1)	24.8 (1.4)	5/5 (100)

[1] Day 0 (RT) = within 2 hours of collection at room temperature shipping conditions
[2] Day 3 (RT) = 72 hours at room temperature shipping conditions
[3] Day 5 (RT) = 120 hours at room temperature shipping conditions
[4] Testing performed at the conclusion of the thermal excursions described in Table 13
[5] Testing performed at the conclusion of the thermal excursions described in Table 14
[6] Und = Undetermined value (no detectable Ct value)

Shipping Study Using Confirmed Positive Clinical Saliva Samples

A total of 31 previously confirmed clinical SARS-CoV-2 positive saliva specimens were evaluated in the simulated shipping study. The clinical samples were either physically shipped at room temperature conditions or stored for the duration of each simulated temperature excursion, followed by equilibration to room temperature, extracted using the MagMAX™ kit, and tested with the P23 Labs Assay.

Results of the simulated shipping studies when using previously confirmed clinical positive and negative saliva specimens showed that 31/31 positive samples were positive by the P23 Labs SARS-CoV-2 Assay (Table 16). The mean and standard deviation of the Ct values for each gene target were similar before and after simulated shipping scenario (within ~3 Cts), with no evidence of significant degradation of the SARS-CoV-2 RNA. Eleven out of 11 negative clinical saliva specimens were negative by the P23 Labs Assay (no amplification of N, ORF1ab, or S genes).

Table 16. Summary of results from the simulated shipping study using clinical saliva samples

Saliva Sample Group	Test Point	N	Mean Ct (Standard Deviation)			Positive (%)
			N Gene	ORF1ab	S Gene	
Negative	Day 0 (RT)[1]	11	Und[6]	Und[6]	Und[6]	0 (0)
	Day 3 (RT)[2]	11	Und[6]	Und[6]	Und[6]	0 (0)
	Day 5 (RT)[3]	11	Und[6]	Und[6]	Und[6]	0 (0)
	Summer[4]	11	N/A	N/A	N/A	0 (0)
	Winter[5]	11	N/A	N/A	N/A	0 (0)
Positive	Day 0 (RT)[1]	31	25.3 (0.8)	23.6 (1.5)	26.1 (1.2)	31/31 (100)
	Day 3 (RT)[2]	31	26.9 (1.1)	24.4 (0.9)	28.1 (1.3)	31/31 (100)
	Day 5 (RT)[3]	31	27.3 (1.9)	24.9 (1.8)	27.7 (1.6)	31/31 (100)
	Summer[4]	31	28.9 (1.8)	25.9 (1.1)	27.1 (2.3)	31/31 (100)
	Winter[5]	31	25.9 (1.3)	27.3 (1.5)	27.1 (0.9)	31/31 (100)

[1] Day 0 (RT) = within 2 hours of collection at room temperature shipping conditions
[2] Day 3 (RT) = 72 hours at room temperature shipping conditions
[3] Day 5 (RT) = 120 hours at room temperature shipping conditions
[4] Testing performed at the conclusion of the thermal excursions described in Table 13
[5] Testing performed at the conclusion of the thermal excursions described in Table 14
[6] Und = Undetermined value (no detectable Ct value)

These results demonstrate that SARS-CoV-2 RNA positive saliva specimens are stable in the OMNIgene®·ORAL OM-505 collection device when exposed to a broad range of temperature conditions. P23 Labs TaqPath Assay performance was equivalent when using saliva collected in OM-505 and spiked with SARS-CoV-2 RNA versus patient self-collected saliva in OM-505 under HCP supervision (i.e., contrived specimens versus clinical samples). These data support the use of the OMNIgene®·ORAL OM-505 for transport and storage of specimens following collection of saliva under HCP supervision in the home or healthcare setting.

LIMITATIONS

- Testing of saliva specimens is limited to patients with symptoms of COVID-19.

Note: This test is only authorized for the duration of the declaration that circumstances exist justifying the authorization of emergency use of in vitro diagnostics for detection and/or diagnosis of COVID-19 under Section 564(b)(1) of the Act, 21 U.S.C. § 360bbb-3(b)(1), unless the authorization is terminated or revoked sooner.