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Short communication

Clinical performance of SARS-CoV-2 detection on the cobas Liat using water gargle samples<sup>☆</sup>Maxime Veillette<sup>a</sup>, Francine Tourangeau<sup>b</sup>, Judith Fafard<sup>c,d</sup>, Jeannot Dumaresq<sup>e,f</sup>, Annie-Claude Labbé<sup>d,g,h,\*</sup>, on behalf of the G-SPIT study group<sup>a</sup> Faculté de médecine, Université de Montréal, Québec, Canada<sup>b</sup> Service de microbiologie et d'infectiologie, CISSS Bas St-Laurent, Québec, Canada<sup>c</sup> Laboratoire de santé publique du Québec, Institut national de santé publique du Québec, Sainte-Anne-de-Bellevue, Québec, Canada<sup>d</sup> Département de microbiologie, infectiologie et Immunologie, Université de Montréal, Montréal, Québec, Canada<sup>e</sup> Département de microbiologie-infectiologie et d'immunologie, Faculté de Médecine, Université Laval, Québec, Québec, Canada<sup>f</sup> Service de microbiologie et d'infectiologie, CISSS de Chaudière-Appalaches, Lévis, Québec, Canada<sup>g</sup> Service de diagnostic moléculaire, département des laboratoires de biologie médicale et Service d'infectiologie, Grappe Optilab-CHUM, Centre hospitalier de l'Université de Montréal, Montréal, Québec, Canada<sup>h</sup> Service de maladies infectieuses, Département de Médecine, CIUSSS de l'Est-de-l'Île-de-Montréal, Montréal, Canada

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## ABSTRACT

Spring water gargle (SWG) is a suitable, non-invasive, alternative specimen for SARS-CoV-2 detection by RT-PCR. This study sought to evaluate the performance of the cobas Liat point-of-care system for the detection of SARS-CoV-2 in SWG samples. SWG samples and standard oral and nasopharyngeal swab (ONPS) were collected simultaneously from participants in a COVID-19 screening clinic, in November and December 2020. Both sample types were analyzed in parallel on the cobas Liat platform and with the Seegene Allplex 2019-nCoV assay. Among the 110 participants, 53% had compatible symptoms and 71% had a contact with a confirmed COVID-19 case. Only two (1.8%) individuals had neither symptoms nor contact. Amongst 110 paired samples, 25 (23%) were positive for SARS-CoV-2 on the cobas Liat for a least one sample type, with a kappa coefficient of 0.92. Agreement between the cobas Liat platform and the Seegene assay was also excellent (kappa coefficient values of 0.94 and 0.95). Two SWG samples failed to provide a positive result when their ONPS pair was positive, but their cycle threshold (Ct) values were >35 on the Seegene assay, reflecting a low viral load. Overall, the performance of the cobas Liat platform is excellent for the detection of SARS-CoV-2 in SWG samples in a high pre-test probability population.

## Introduction

Access to diagnostic methods is one of the most important measures to control the COVID-19 pandemic [1]. Although oral and nasopharyngeal swab (ONPS) remains one of the most recommended sample collection method for the molecular diagnosis of respiratory viral infections including SARS-CoV-2, it is susceptible to certain limitations including patient acceptability and availability of materials and trained professional healthcare workers to collect the specimen. Moreover, traditional platforms for nucleic acid amplification tests (NAAT) require dedicated personnel to perform them due to their inherent complexity [2]. The logistics and human resources required to perform those tests in suburban and remote communities can be limited. The automation of NAAT plat-

forms allowed the development of benchtop, point-of-care (POC) tests with some systems offering fast and easy platforms useable by unspecialized workers. Roche's cobas Liat platform uses a 'lab-in-a-tube' process to automatically perform all steps from reagents and sample preparation to nucleic acid extraction, amplification and readout [3]. It provides results in less than 20 min and was successfully used and validated to detect SARS-CoV-2 infection [4–6].

We and others [4,7–9] previously described how natural spring water gargle (SWG) is a suitable specimen for RT-PCR detection of SARS-CoV-2 infection while being well tolerated, readily accessible and allowing patient self-collection of sample. In this study, we sought to validate SWG samples on the POC cobas Liat system for the detection of SARS-CoV-2.

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**Table 1**  
Detection of SARS-CoV-2 RNA from oropharyngeal swab and gargle specimens on the cobas® Liat® system (n = 110).

Results	Agreement (95% CI)					
	ONPS		PPA	NPA	OPA	Kappa
SWG	+	-				
+	22	1	91.7 (74.2 – 97.7)	98.8 (93.7 – 99.8)	97.3 (92.3 – 99.1)	0.92 (0.83 – 1.00)
-	2	85				

SWG: spring water gargle; ONPS: oronasopharyngeal swab; CI: confidence interval; PPA: positive percentage agreement; NPA: negative percentage agreement; OPA: overall percentage agreement; Kappa: Cohen's Kappa

## Materials and methods

### Study population, ethics, and questionnaire

This study is part of the G-SPIT multicentre project comparing the performance of SWG to ONPS for SARS-CoV-2 detection on various NAAT platforms. In this arm of the project, participants were recruited prospectively in designated COVID-19 screening clinics in Rimouski, Canada. Individuals older than 5 years old who had a recent confirmed contact or symptoms compatible with COVID-19 were recruited. After obtaining verbal informed consent, participants were asked to complete a short questionnaire: age, contact with COVID-19 cases, presence and duration of symptoms.

This study obtained ethics board approval from appropriate regional health boards (CIUSSS de l'Est-de-l'Île-de-Montréal and CISSS du Bas-Saint-Laurent).

### Sample collection

Participants were asked not to eat, drink or smoke for 15 min prior to sample collection. For each participant, ONPS and SWG specimens were collected as previously described [8,9]. Briefly, a trained healthcare professional collected the ONPS by first swabbing the posterior oropharynx before inserting the same flexible swab through one nostril and rotating for five seconds before removing. The ONPS was transported in 3 ml of molecular grade water. SWG were collected by having participants swish 5 ml of natural spring water (ESKA, St-Mathieu-d'Harricana, Canada) in their mouth for 5 s and their throat for 5 s and repeating once for a total of 20 s before spitting as much as possible in a cup. The content of the cup was transferred into a 15 ml conical tube. Both samples were sent to the laboratory of the Hôpital régional de Rimouski (Rimouski, Canada).

### SARS-CoV-2 detection by NAAT

Both samples were stored at 4 °C and first tested within 24 to 48 h for SARS-CoV-2 RNA on the cobas Liat system using the cobas SARS-CoV-2 & Influenza A/B kit (Roche) at the Hôpital régional de Rimouski, according to manufacturer's protocol. Samples were then frozen (-70 °C) and transported to the laboratory of the Hôpital Hôtel-Dieu de Lévis (Lévis, Canada). They were analyzed within two months with the Seegene Allplex 2019-nCoV Assay kit (Seegene, Seoul, Republic of Korea) ran on CFX-96 thermocyclers (Bio-Rad, CA, USA), as previously described.

### Statistical analysis

Statistical analyses were performed using R [10] and figures were produced using the ggplot2 package [11]. A confusion matrix was created with the results and positive percent agreement (PPA), negative percent agreement (NPA), positive predictive value (PPV) and negative predictive value (NPV) were calculated with the epiR package using the Wilson-Brown method [12].

## Results

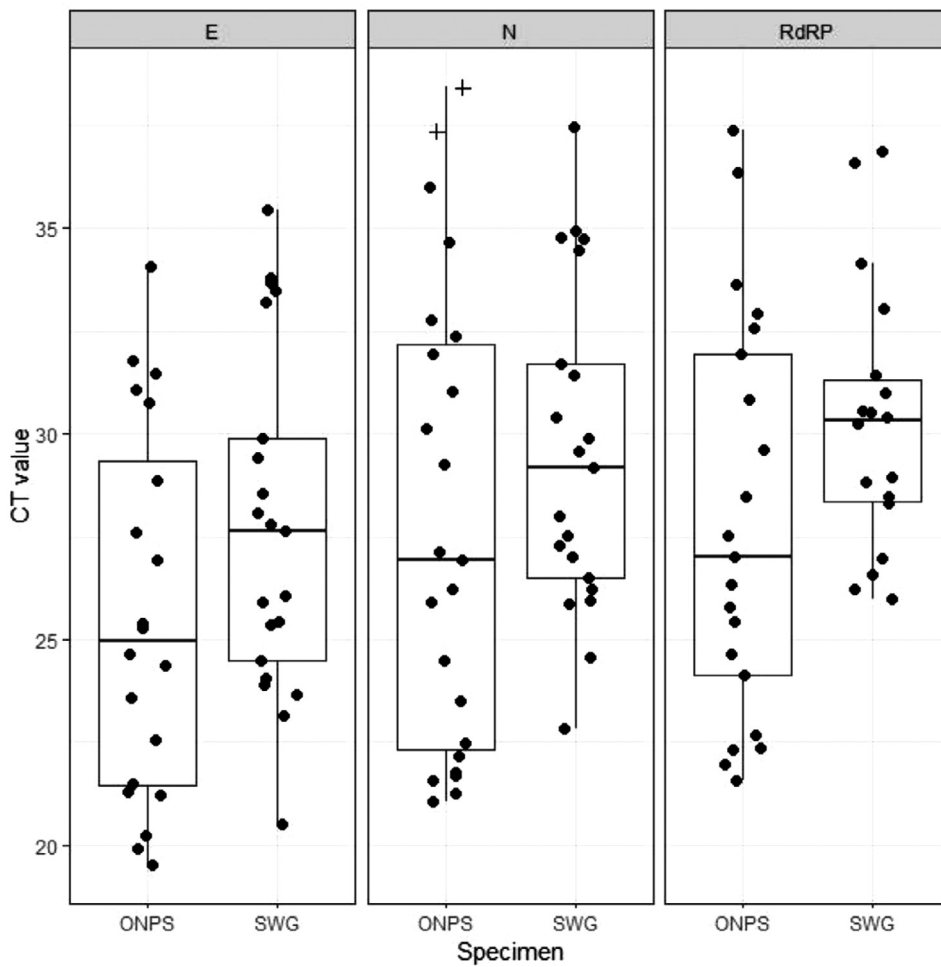
Between November 25 and December 3, 2020, 110 paired samples were obtained. Mean age of the participants was 37 years (range: 6–88), 53% had symptoms compatible with COVID-19 with a mean duration of two days, 71% reported a contact with a confirmed COVID-19 case and 26% had both compatible symptoms and a confirmed positive contact. Only two (1.8%) individuals had neither symptoms nor a confirmed contact and tested negative.

On the cobas Liat, all samples were negative for Influenza A and B; at least one of the two samples was positive for SARS-CoV-2 in 25/110 (22.7%) participants. Agreement between the two sample types was excellent (Table 1), as reflected by a kappa coefficient ( $\kappa$ ) of 0.92. Two individuals had negative results from the SWG sample, but positive results from the ONPS. These two SWG samples were also negative when tested with the Allplex assay and their paired ONPS sample resulted in a positive detection only in the N gene [Cycle threshold (Ct) values of 37 and 38; Fig. 1]. One of these two individuals had fever and malaise for two days and the other was asymptomatic. Conversely, on cobas Liat, one individual had a positive result from the SWG sample, but a negative result from the ONPS. Similar results were obtained using the Allplex assay, with only two of the three targets detected on the SWG (Ct value of 28 for the E gene and of 30 for the N gene) and a negative result from the ONPS. This individual had fever and malaise for four days.

As shown in Table 2 excellent agreement was also obtained when comparing paired samples between the cobas Liat and the Allplex assay for both SWG ( $\kappa=0.94$ ) and ONPS ( $\kappa=0.95$ ). All positive SWG samples on the Allplex assay were also positive on the cobas Liat. Two additional SWG samples were identified as positive on the cobas Liat, but negative on the Allplex assay. Their paired ONPS samples were also positive on the cobas Liat. Overall, when defining a true positive as at least one positive sample (SWG or ONPS) on the Allplex assay (gold standard), sensitivity and negative predictive value of the cobas Liat on SWG were respectively estimated at 91.7% (95% CI 74.2–97.7) and 97.7% (95% CI 92.0–99.4).

## Discussion

We evaluated the clinical performance of identifying SARS-CoV-2 infection from SWG samples on the POC cobas Liat compared to 1) ONPS samples on the cobas Liat and 2) SWG on another NAAT (Allplex 2019-nCoV assay). We demonstrated excellent overall agreement (97.3% and 98.2%, respectively) for both comparisons. The use of paired samples ran on both platforms allowed us to identify two individuals who had positive results on both assays using the ONPS sample, while their paired SWG sample resulted in a negative result. Interestingly, those individuals had low viral loads in their samples as reflected by high Ct values on the Allplex assay. This is consistent with ONPS samples being more sensitive in detecting low viral loads compared to SWG samples, at least for certain SARS-CoV-2 variants. Interestingly, some [13,14], but not all [15] studies report increased sensitivity for the detection of Omicron



**Fig. 1.** Seegene Allplex 2019-nCoV assay cycle threshold values distribution, by sample type. Ct: Cycle threshold; SWG: spring water gargle; ONPS: oronasopharyngeal swab. Note: The two cross symbols for N gene (Ct values of 37 and 38) indicate ONPS samples paired with negative SWG samples on both platforms. The other two targets were not detected from the two ONPS samples that were negative.

**Table 2**  
Detection of SARS-CoV-2 RNA with the cobas® Liat® system and the Seegene Allplex 2019-nCoV assay, by sample type.

Liat	Results		PPA	NPA	Agreement (95% CI)	
	Seegene				OPA	Kappa
	SWG +	SWG -				
SWG +	21	2	100	97.8	98.2	0.94
SWG -	0	87	(84.5–100)	(92.2–99.4)	(93.6–99.5)	(0.87–1.00)
ONPS +	23	1	95.8	98.8	98.2	0.95
ONPS -	1	85	(79.8–99.3)	(93.7–99.8)	(93.6–99.5)	(0.87–1.00)
SWG + or ONPS +	22	1	91.7	98.8	97.3	0.92
SWG - and ONPS -	2	85	(74.2–97.7)	(93.7–99.8)	(92.3–99.1)	(0.83–1.00)
ONPS +	23	1	95.8	98.8	98.2	0.95
ONPS -	1	85	(79.8–99.3)	(93.7–99.8)	(93.6–99.5)	(0.87–1.00)

SWG: spring water gargle; ONPS: oronasopharyngeal swab; CI: confidence interval; PPA: positive percentage agreement; NPA: negative percentage agreement; OPA: overall percentage agreement; Kappa: Cohen’s Kappa.

variant in saliva samples. While our study was performed when most circulating strains were of the Alpha lineage, we believe that limiting false negatives especially in a population with high pre-test probability is of utmost importance to help curtail the spread of SARS-CoV-2 infection. In this regard, the use of SWG samples on the cobas Liat platform showed an overall sensitivity consistent with other reported NAAT tests and an excellent NPV of 97.7%. Fast and accurate results coupled with the simplification of both sample collection and analytical steps makes

POC platforms such as the cobas Liat ideal for rapid testing of populations with high pre-test probability especially in remote communities.

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## Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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