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RESEARCH ARTICLE

Screening for SARS-CoV-2 by RT-PCR: Saliva or nasopharyngeal swab? Rapid review and meta-analysis

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Abstract

Background

Diagnosis of COVID-19 in symptomatic patients and screening of populations for SARS-CoV-2 infection require access to straightforward, low-cost and high-throughput testing. The recommended nasopharyngeal swab tests are limited by the need of trained professionals and specific consumables and this procedure is poorly accepted as a screening method In contrast, saliva sampling can be self-administered.

Methods

In order to compare saliva and nasopharyngeal/oropharyngeal samples for the detection of SARS-CoV-2, we designed a meta-analysis searching in PubMed up to December 29th, 2020 with the key words "(SARS-CoV-2 OR COVID-19 OR COVID19) AND (salivary OR saliva OR oral fluid)) NOT (review[Publication Type]) NOT (PrePrint[Publication Type])" applying the following criteria: records published in peer reviewed scientific journals, in English, with at least 15 nasopharyngeal/orapharyngeal swabs and saliva paired samples tested by RT-PCR, studies with available raw data including numbers of positive and negative tests with the two sampling methods. For all studies, concordance and sensitivity were calculated and then pooled in a random-effects model.

Findings

A total of 377 studies were retrieved, of which 50 were eligible, reporting on 16,473 pairs of nasopharyngeal/oropharyngeal and saliva samples. Meta-analysis showed high concordance, 92.5% (95%CI: 89.5–94.7), across studies and pooled sensitivities of 86.5% (95%CI: 83.4–89.1) and 92.0% (95%CI: 89.1–94.2) from saliva and nasopharyngeal/oropharyngeal swabs respectively. Heterogeneity across studies was 72.0% for saliva and 85.0% for nasopharyngeal/oropharyngeal swabs.

Interpretation

Our meta-analysis strongly suggests that saliva could be used for frequent testing of COVID-19 patients and "en masse" screening of populations.

Introduction

Propagation of infections by SARS-CoV-2, the coronavirus causing the COVID-19 pandemic, occurs from asymptomatic as well as symptomatic carriers [1]. To reduce the circulation of the virus in the population, SARS-CoV-2 carriers need to be identified rapidly and isolated as soon as possible, ideally before the onset of symptoms. When the virus has disseminated throughout a whole country, massive testing becomes of utmost urgent importance to combat the pandemic [2–4].

The recommended diagnosis of SARS-CoV-2 infection from the World Health Organisation is based on real time RT-qPCR detection of viral RNA in respiratory specimen such as nasopharyngeal swabs (NP), bronchial aspiration (BA), throat swab and sputum [5]. The American Centres For Disease Control and Prevention and the European Centre for Disease Prevention and Control now recommend viral testing from the respiratory system such as nasal or oral swabs or saliva [6, 7]. The French health regulatory authority (*Haute Autorité de Santé*) has recently included in its recommendations the use of saliva samples for the detection of SARS-CoV-2 in symptomatic individuals for whom nasopharyngeal sampling is difficult, and for mass testing within schools, universities and among health workers [8].

To make the diagnostic acceptable to the largest number of people, especially asymptomatic individuals, massive testing should be based on a sampling procedure that is inexpensive, easy to set up and well accepted by the population [9]. In contrast to nasopharyngeal swabbing, saliva sampling meets these criteria. Saliva sampling is fast, non-invasive, inexpensive and painless. It does not require trained professional with personal protective equipment nor other material than a simple plastic tube, and can be self-administered.

To evaluate saliva sampling for the detection of SARS-CoV-2, we conducted a meta-analysis on studies published in peer-reviewed journals until the 29th of December 2020 comparing the detection of SARS-CoV-2 using RT-PCR on paired nasopharyngeal/oropharyngeal and saliva samples in the same individuals sampled at the same time.

Methods

Search strategy and selection criteria

Literature search in PubMed (https://pubmed-ncbi-nlm-nih-gov) run the 29th of December 2020 with search word (SARS-CoV-2 OR COVID-19 OR COVID19) AND (salivary OR saliva OR oral fluid)) NOT (review[Publication Type]) NOT (PrePrint[Publication Type]) identified 377 articles. We included publications if they met the following eligibility criteria:

- 1. Records published in peer reviewed scientific journals;
- 2. Records published in English;
- 3. Data curation based on examination of the title and abstract, searching for research articles assessing viral RNA presence in saliva vs nasopharyngeal and/or oropharyngeal swabs;

- 4. Availability of nasopharyngeal (and/or oropharyngeal) swabs and saliva data on specimens sampled on the same individuals at the same time;
- 5. Detection of SARS-CoV-2 using the same RT-PCR method on both samples;
- 6. More than 15 individuals included in the study.

Data analysis

From each eligible article, we extracted: the number of individuals positive for SARS-CoV-2 in both nasopharyngeal/oropharyngeal swabs and saliva (a), those positive only in nasopharyngeal/oropharyngeal swab (b), those positive only in saliva (c) and (d) those negative in both nasopharyngeal swab and saliva (Table 1). From these data, we calculated the concordance of the same test (RTqPCR for 49 studies and RTdPCR for 1 study) on the two types of sample (a +d)/(a+b+c+d). We also computed the sensitivity of the test on each type of sample. The estimation of the sensitivity of a test requires a reference diagnosis. Since nasopharyngeal swab sampling has been shown to produce false negatives by RTqPCR [10], sensitivities for the saliva and the nasopharyngeal swab are defined here respectively as (a+c)/(a+b+c) and (a+b)/(a+b+c), considering as true positive any individual with a positive result on one or the other sample. This definition of a positive individual is also in agreement with the US-CDC and the ECDC directives on SARS-Cov-2 testing.

The overall concordance and sensitivities have been estimated in a meta-analysis via Generalized Linear Mixed Models (GLMM), using a fixed-effect model, and also a random-effect model in case of over dispersion of the observations [11, 12]. Dispersion of effect sizes was evaluated using the Higgins I^2 estimate of heterogeneity along with the Cochran's Q in fixedeffect models [13] and using the tau² estimate (between-study variance which is the variance of the distribution of true effect size) in random-effects models. The tau² was calculated using the maximum likelihood estimator. As some studies have a sample size too small for the normality hypothesis, confidence intervals for each study were computed using the Clopper-Pearson method [14] also called "exact" binomial interval. Those for the overall estimates are based on normal approximation. Results are presented as forest plots.

All analyses were done with R version 4.0.3, using the package "meta" version 4.15–1 (2020-09-30) [15, 16].

Results

Forty-eight studies comparing SARS-CoV-2 loads in NP swabs and saliva samples collected concurrently in the same individuals using the same technique and providing positivity and negativity in both samples have been identified in PubMed [17–64]. We also included 2 articles not identified in the original Pubmed keywords search while fulfilling eligibility criteria (Table 1 and Fig 1) [65, 66]. In total 16,473 paired samples were analysed. The number of paired samples per study varied between 16 and 3834. Meta-analysis showed an overall concordance of 92.5% (95%CI: 89.5–94.7) across studies (Fig 2).

The overall sensitivity of the RT-PCR test from saliva samples was 86.5% (95%CI: 83.4– 89.1) (Fig 3) versus 92.0% from nasopharyngeal swabs (95%CI: 89.1–94.2) (Fig 4). There was no association between the sensitivities of the saliva (S1 Fig) or of nasopharyngeal swab (S2 Fig) estimated in each study and the prevalence of the virus in the same study. If the sensitivity of the saliva was lower in populations of asymptomatic individuals than in population of individuals with symptoms, one would expect to observe a lower sensibility of the saliva in the studies with a low prevalence of infection.

Reference	Number of tested individuals						Reference: S+ ou N+	
	Saliva + Nasoph. +	Saliva—Nasoph. +	Saliva + Nasoph	Saliva—Nasoph	Total		Sensitivity of S	Sensitivity of N
	a	b	с	d	$\mathbf{n} = \mathbf{a} + \mathbf{b} + \mathbf{c} + \mathbf{d}$	(a+d)/n	(a+c)/p	(a+b)/p
Aita	7	1	0	35	43	97.7%	87.5%	100.0%
Altawalah	287	57	18	529	891	91.6%	84.3%	95.0%
Azzi	22	4	33	54	113	67.3%	93.2%	44.1%
Babady	16	1	1	69	87	97.7%	94.4%	94.4%
Barat	30	7	1	421	459	98.3%	81.6%	97.4%
Berenger	52	11	6	6	75	77.3%	84.1%	91.3%
Bhattacharya	53	5	0	16	74	93.2%	91.4%	100.0%
Binder	10	1	1	7	19	89.5%	91.7%	91.7%
Borghi	79	28	7	187	301	88.4%	75.4%	93.9%
Braz-Silva	37	15	18	131	201	83,6%	78,6%	74,3%
Byrne	12	2	0	96	110	98.2%	85.7%	100.0%
Cassinarri	8	5	1	17	31	80.6%	64.3%	92.9%
Caulley	34	22	14	1869	1939	98.1%	68.6%	80.0%
Chau	19	0	1	7	27	96.3%	100.0%	95.0%
Chen	49	6	3	0	58	84.5%	89.7%	94.8%
Güçlü	23	4	4	33	64	87.5%	87.1%	87.1%
Hanege	29	9	0	0	38	76.3%	76.3%	100.0%
Hanson	75	5	6	268	354	96.9%	94.2%	93.0%
Hasanoglu	27	21	3	9	60	60.0%	58.8%	94.1%
Iwasaki	8	1	1	66	76	97.4%	90.0%	90.0%
Jamal 1	44	20	8	19	91	69.2%	72.2%	88.9%
Jamal 2	42	14	9	10	75	69.3%	78.5%	86.2%
Kandel	39	4	3	383	429	98.4%	91.3%	93.5%
Kojima	20	3	6	16	45	80.0%	89.7%	79.3%
Landry	28	5	2	89	124	94.4%	85.7%	94.3%
Leung	38	7	13	37	95	78.9%	87.9%	77.6%
Matic	15	6	1	52	74	90.5%	72.7%	95.5%
McCormick-Baw	47	2	1	106	156	98.1%	96.0%	98.0%
Migueres	34	7	3	79	123	91.9%	84.1%	93.2%
Moreno- Contreras	19	9	6	37	71	78.9%	73.5%	82.4%
Nagura-Ikeda	84	19	0	0	103	81.6%	81.6%	100.0%
Otto	45	0	4	43	92	95.7%	100.0%	91.8%
Pasomsub	16	3	2	179	200	97.5%	85.7%	90.5%
Procop	38	0	1	177	216	99.5%	100.0%	97.4%
Rao	73	11	76	57	217	59.9%	93.1%	52.5%
Sakanashi	15	0	4	9	28	85.7%	100.0%	78.9%
Senok	19	7	9	366	401	96.0%	80.0%	74.3%
Skolimowska	15	3	1	112	131	96.9%	84.2%	94.7%
Sorelle	32	7	0	44	83	91.6%	82.1%	100.0%
Sui	14	0	2	0	16	87.5%	100.0%	87.5%
Torres	46	54	8	835	943	93.4%	50.0%	92.6%
Uwamino	32	15	11	138	196	86.7%	74.1%	81.0%
Vaz	67	4	2	82	155	96.1%	94.5%	97.3%
Vogels	49	5	4	3776	3834	99.8%	91.4%	93.1%

Table 1. Studies comparing SARS-CoV-2 detection in paired saliva and nasopharyngeal samples meeting inclusion criteria.

(Continued)

Reference	Number of tested individuals C						Reference: S+ ou N+	
	Saliva + Nasoph. +	Saliva—Nasoph. +	Saliva + Nasoph	Saliva—Nasoph	Total		Sensitivity of S	Sensitivity of N
	a	b	с	d	n = a+b+c+d	(a+d)/n	(a+c)/p	(a+b)/p
Williams	33	6	1	49	89	92.1%	85.0%	97.5%
Wong	104	18	37	70	229	76.0%	88.7%	76.7%
Wyllie	34	9	13	13	69	68.1%	83.9%	76.8%
Yee	69	18	10	203	300	90.7%	81.4%	89.7%
Yokota	42	4	6	1872	1924	99.5%	92.3%	88.5%
Zhu	382	60	15	487	944	92.1%	86.9%	96.7%
Total	2412	525	376	13160	16473	94,5%	84,2%	88,7%

Table 1. (Continued)

https://doi.org/10.1371/journal.pone.0253007.t001

In our fixed-effect model meta-analysis, saliva gave I^2 of 72% and nasopharyngeal gave I^2 of 85%, both corresponding to high heterogeneity. Therefore, a random-effect model was performed to assess the overall sensitivities of the RTqPCR tests, taking into account the fact that the studies did not originate from one single population. The variance of saliva was 0.49 and that of nasopharyngeal was 1.03. As a sensitivity analysis, we also used various other methods to estimate the sensitivity, the confidence interval and the tau² and all yielded very similar results (Figs 2–4).

Discussion

This meta-analysis reviewed 50 studies and concluded to a high concordance between nasopharyngeal and saliva samples for the detection of SARS-CoV-2 by RT-PCR. Although sensitivity was slightly lower on saliva samples than on nasopharyngeal samples, both values are above the 80% sensitivity cut-off recommended by health regulatory authorities such as the French Haute Autorité de Santé [67].

For computing concordance and sensitivities, we considered here the reference as SARS--CoV-2 positivity by RT-PCR either in saliva and/or nasopharyngeal samples since the presence of the virus in any sample is indicative of virus carriage.

In the context of mass screening, most participants are asymptomatic. Among the 50 studies analysed, only one included exclusively asymptomatic participants [63] and 8 studies included both symptomatic and asymptomatic participants but it was impossible to separate the data between the two populations [17, 20, 25, 29, 51, 56, 58, 65]. One study of contact cases included a larger number of asymptomatic subjects as compared to study of symptomatic subjects [49]. We did not observe any difference in concordance of the tests in these particular studies involving asymptomatic participants (Table 1). Formal comparison of nasopharyngeal and saliva samples from asymptomatic individuals is challenging: it would require to screen a large population for a small number of positive cases detected since the prevalence is usually low in this population. On the contrary, the symptomatic population expectedly contains higher percentage of positive subjects, as symptoms usually timely correlates with the highest viral load, which allow an easier comparison of both sampling procedures. Anyhow, viral load in saliva of presymptomatic subjects remains in the range of detection of the RT-PCR test for several days both in saliva [68] and nasopharyngeal samples [69]. In addition, both asymptomatic and symptomatic subjects appear to be contagious [1] with similarities in their viral load evolution [70–72]. Moreover, S3 Fig shows that in France the proportion of positive cases in the symptomatic tested population is consistently about 5 times larger than in the asymptomatic tested population, independently of viral prevalence over time. This and the fact that



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neither saliva nor nasopharyngeal sensitivities are affected in a screening-like context (the low prevalence being taken as a proxy, <u>S1</u> and <u>S2</u> Figs) strongly predict that viral detection is expected to exhibit similar performance in both populations.

Our meta-analysis showed large heterogeneity between studies. Sources of heterogeneity are both biological and technical. Biological heterogeneity may come from the fact that a given individual may carry the virus in only one of the saliva or nasopharyngeal specimens, or from the timing of sampling during the course of contamination. Technical heterogeneity comes from differences in the sampling and in RT-PCR methods. Among the 50 studies meeting the

Study	Sample size	Concordance (%)	95% CI (%)	
Aita	43	97.7	[87.7, 99.9]	
Altawalah	891	91.6	89.6, 93.3	
Azzi	113	67.3	57.8, 75.8	
Babady	87	97.7	91.9. 99.71	i i i i i i i i i i i i
Barat	459	98.3	96.6. 99.2	1
Berenger	75	77.3	66.2 86.2	· · · · · · · · · · · · · · · · · · ·
Bhattacharva	74	93.2	84 9 97 81	i i i i i i i i .
Binder	19	89.5	66 9 98 7	
Borghi	301	88.4	84 2 91 8	i i i i i i i i i i <u>a</u> li
Braz Silva	201	83.6	77 7 88 41	
Burno	110	00.0	02 6 00 01	
Ogeneinerri	24	50.2	100.0, 00.01	
Cassinan	1 0 2 0	00.0	02.5, 92.5	
Caulley	1 9 3 9	90.1	91.4, 90.7	
Chau	21	96.3	[81.0, 99.9]	
Chen	58	84.5	72.6, 92.7	
Güçlü	64	87.5	76.8, 94.4	
Hanege	38	76.3	[59.8, 88.6]	
Hanson	354	96.9	[94.5, 98.4]	
Hasanoglu	60	60.0	46.5, 72.4	
Iwasaki	76	97.4	90.8, 99.7	i i i i i i i la la la la la <mark>la</mark> na
Jamal 1	91	69.2	58.7. 78.51	
Jamal 2	75	69.3	57 6 79 5	i i i i i i i i i i
Kandel	429	98.4	96 7 99 3	
Kojima	45	80.0	165 4 90 41	· · · · · · · · · · · · · · · · · · ·
Landny	124	94 4	88 7 97 7	
Loung	05	78.0	19 38 1 99	1 <u> </u>
Matia	74	10.5	101.4, 00.0]	
MaCarraiale Bass	14	90.5	01.5, 90.1	
Misuana A	100	90.1	94.5, 99.0	i i i i i i i i i i j
Migueres	123	91.9	[05.0, 90.0]	
Moreno - Contreras		78.9	67.6, 87.7	
Nagura-Ikeda	103	81.6	12.1, 88.5	
Otto	92	95.7	[89.2, 98.8]	· · · · · · · · · · · · · · · · · · ·
Pasomsub	200	97.5	[94.3, 99.2]	i i i i i i i i i i i i i i
Procop	216	99.5	[97.4, 100.0]	
Rao	217	59.9	[53.1, 66.5]	i i i i i i 🖛 i i i i
Sakanashi	28	85.7	[67.3, 96.0]	
Senok	401	96.0	93.6, 97.7	1 - 1 - 1 - 1 - 1 - 1 - 1 - 1 <mark>-</mark> 1 - 1
Skolimowska	131	96.9	92.4. 99.2	_ _ _ _ _ _ _ _ _ _
Sorelle	83	91.6	83 4 96 51	
Sui	16	87.5	61 7 98 4	
Torres	943	93.4	917 94 9	
Liwamino	196	86.7	181 2 01 11	i i i i i i i i <u></u>
Vaz	155	96.1	91 8 98 6	
Vaz	2 024	30.1	100 6 00 01	
Vogers	3 034	33.0	33.0, 33.3	
Wang	89	92.1	04.5, 90.8	
Wong	229	76.0	[09.9, 01.4]	
vvyille	69	68.1	55.8, 78.8	
Yee	300	90.7	86.8, 93.7	
Yokota	1 924	99.5	[99.0, 99.8]	
Zhu	944	92.1	[90.1, 93.7]	
Total fixed effect	16 473	94 5	[94.2. 94.9]	
Heterogeneity: 12 = 0.5%	10 4/0		[94.2, 94.0]	
Cochran $\Omega = 983$ (p<0.01)				
000111ali (2 - 000 (p<0.01)				
Total random effect		92.5	[89.5, 94.7]	
Heterogeneity: tau ² = 1.6				
				0 10 20 30 40 50 60 70 80 90 100
				Concordance (%)

Fig 2. Forest plot of the concordance between the results of RTqPCR tests on nasopharyngeal and saliva samples. The confidence intervals for each study are computed using the Clopper-Pearson method. Those for the overall estimates (fixed-effect or random-effect) are based on normal approximation. The blue box size is proportional to the number of positive tests. The red line corresponds to the value of the overall concordance of the random-effect model. This vertical line enables to locate the studies having an estimate concordance higher than 92.5%.

https://doi.org/10.1371/journal.pone.0253007.g002

inclusion criteria for the meta-analysis, 29 studies report saliva collection in sterile containers (urine tubes or vials) without any additional solution. The other studies diluted the saliva in various viral transport media or phosphate buffer saline with or without bovine serum albumin. Other sources of variability come from differences in the amplified region of SARS-Cov-2 or to different positivity threshold between studies; most studies do not present viral load data but only cycle thresholds (Ct) for specific amplification of SARS-Cov-2 sequences and not even Ct differences (Δ Ct) with a human reference gene.

Altogether, our meta-analysis of 50 studies including 16,473 paired samples shows high concordance (92.5%) between nasopharyngeal/oropharyngeal swabs and saliva, with a 5%

Study	Sample size	Sensitivity (%)	95% CI (%))
Aita	43	87.5	[47.3, 99.7]	
Altawalah	891	84.3	80.1. 87.91	
Azzi	113	93.2	83.5, 98.11	1 1 1 1 1 1 1 1 1 1 1
Babady	87	94.4	72 7 99 91	
Barat	459	81.6	65 7 92 3	
Berender	75	84 1	73 3 91 8	
Bhattachanya	74	01 /	181 0 07 11	
Bindor	10	01.4	61 5 00 9	
Dirider	201	31.1 75 A	GG E 92 01	
Dorgin Braz Ciluz	201	70.4	100.5, 05.0]	
Braz-Sliva	201	/0.6	67.1, 87.5	
Byrne	110	05.7	[57.2, 98.2]	
Cassinarri	31	64.3	35.1, 87.2	li i i i i i i i i i i i i
Caulley	1 939	68.6	[56.4, 79.1]	
Chau	27	100.0	[83.2, 100.0]	
Chen	58	89.7	[78.8, 96.1]	
Güçlü	64	87.1	70.2, 96.4	
Hanege	38	76.3	59.8, 88.6	
Hanson	354	94.2	187.0 98.11	· · · · · · · · · · · · · · · · · · ·
Hasanoglu	60	58.8	44 2 72 4	i i i i i i i i i i i i
wasaki	76	90.0	55 5 99 7	
Jamal 1	91	72.2	160 4 82 11	
Jamal 2	75	78.5	66 5 87 7	
Kandal	120	013	70 2 07 6	
Kaiima	425	90.7	72 6 07 01	
Kojima	40	09.7	12.0, 91.0	
Landry	124	00.7	109.7, 95.2	li i i i i i i i <mark>i</mark> i i
Leung	95	87.9	[/6./, 95.0]	
Matic	/4	12.1	49.8, 89.3	
McCormick-Baw	156	96.0	86.3, 99.5	
Migueres	123	84.1	[69.9, 93.4]	
Moreno - Contreras	71	73.5	[55.6, 87.1]	
Nagura-Ikeda	103	81.6	[72.7, 88.5]	· · · · · · · · · · · · · · ∎ + · · · ·
Otto	92	100.0	[92.7, 100.0]	
Pasomsub	200	85.7	[63.7, 97.0]	
Procop	216	100.0	[91.0, 100.0]	
Rao	217	93.1	188.0. 96.5	
Sakanashi	28	100.0	182 4 100 01	11 1 1 1 1 1 1 1 1 1 1 1 [1
Senok	401	80.0	163 1 91 6	
Skolimowska	131	84.2	60 4 96 61	
Sorelle	83	82.1	166 5 92 51	
Sui	16	100.0	79 4 100 01	
Torres	9/3	50.0	140 2 59 81	
lucraine	100	74.1	61 0 94 71	
Vez	190	14.1 04 E	01.0, 04.7	
Vaz	100	94.9	[00.0, 30.3]	
Vogels	3 634	91.4	01.0, 97.1	
Williams	09	05.0	10.2, 94.3	
vvong	229	88.7	[82.7, 93.2]	
VVyllie	69	83.9	/1.7, 92.4	
Yee	300	81.4	12.3, 88.6	li i i i i i i - <mark>-</mark> i i
Yokota	1 924	92.3	[81.5, 97.9]	
Zhu	944	86.9	[83.4, 89.8]	
Tadal Garad affa at	40.470			
lotal fixed effect	16 4/3	84.2	[82.9, 85.4]	
Heterogeneity: I ² = 72%				
Cochran Q = 176 (p<0.01)				
				
lotal random effect		86.5	[83.4, 89.1]	
Heterogeneity: tau ² = 0.49				
				0 10 20 30 40 50 60 70 80 90 100
				Sensitivity of saliva (%)

Fig 3. Forest plot of the sensitivity of RTqPCR test on saliva. The confidence intervals for each study are computed using the Clopper-Pearson method. Those for the overall estimates (fixed-effect or random-effect) are based on normal approximation. The blue box size is proportional to the number of positive tests. The difference between fixed-effect and random-effect overall sensitivity (respectively 84.2%, 86.5%) is low. The red line corresponds to the value of the overall sensitivity of the random-effect model. This vertical line enables to locate the studies having an estimate sensitivity higher than 86.5%. The heterogeneity estimator I² is equal to 72%, which means a higher level of heterogeneity.

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higher sensitivity for the nasopharyngeal/oropharyngeal (92.0%) as compared to saliva (86.5%). While that might have been a liability in the context of individual diagnosis, it is not such a concern for mass screening, especially given the major advantage of the saliva sampling in terms of logistics. Previous meta-analyses of paired nasopharyngeal and saliva samples included less than 15 peer-reviewed studies or preprints and reported average sensitivity of

Study	Sample size	Sensitivity (%)	95% CI (%)
Aita	43	100.0	[63.1, 100.0]	
Altawalah	891	95.0	[92.3, 97.0]	
Azzi	113	44.1	31.2, 57.6	
Babady	87	94.4	72.7, 99.9	
Barat	459	97.4	86.2, 99.9	
Berenger	75	91.3	82.0, 96.7	 -
Bhattacharva	74	100.0	[93.8, 100.0]	
Binder	19	91.7	[61.5, 99.8]	
Borahi	301	93.9	87.8 97.5	
Braz-Silva	201	74.3	62 4 84 01	
Byrne	110	100.0	76 8 100 01	
Cassinarri	31	92.9	166 1 99 8	
Caulley	1 939	80.0	68 7 88 6	
Chau	27	95.0	75 1 99 9	
Chen	58	94.8	185 6 98 91	
Güclü	64	87.1	70 2 96 4	
Hanege	38	100.0	100 7 100 01	
Hancon	254	02.0	105 / 07 /1	
Hanson	354	93.0	03.4, 37.4	
hasahogiu	76	94.1	65.6, 90.0	
lemal 1	10	50.0	170 2 05 11	
Jamal 2	31	00.9	75.3, 93.1	
	10	00.2	10.3, 33.5	
Kallesa	429	93.5	[02.1, 90.0]	
Kojima	45	19.3	100.3, 92.0	
Landry	124	94.3	00.0, 99.3	
Leung	95	11.6	[64.7, 87.5]	
Matic	14	95.5	11.2, 99.9	
McCormick-Baw	156	98.0	89.4, 99.9	
Migueres	123	93.2	[81.3, 98.6]	
Moreno - Contreras		82.4	[65.5, 93.2]	
Nagura-Ikeda	103	100.0	[96.5, 100.0]	
Otto	92	91.8	[80.4, 97.7]	
Pasomsub	200	90.5	69.6, 98.8	
Procop	216	97.4	86.5, 99.9	
Rao	217	52.5	[44.5, 60.4]	
Sakanashi	28	78.9	54.4, 93.9	
Senok	401	74.3	56.7, 87.5	
Skolimowska	131	94.7	[74.0, 99.9]	
Sorelle	83	100.0	[91.0, 100.0]	
Sui	16	87.5	61.7, 98.4	
lorres	943	92.6	[85.9, 96.7]	
Uwamino	196	81.0	68.6, 90.1	
Vaz	155	97.3	[90.5, 99.7]	
Vogels	3 834	93.1	[83.3, 98.1]	
Williams	89	97.5	[86.8, 99.9]	
Wong	229	76.7	[69.4, 83.1]	
Wyllie	69	76.8	[63.6, 87.0]	
Yee	300	89.7	[81.9, 94.9]	
Yokota	1 924	88.5	[76.6, 95.6]	
Zhu	944	96.7	[94.6, 98.2]	
Total fixed effect	16 473	89 7	187 5 89 71	
Hotorogonoity: 12 = 95%	10 4/0	00.7	[01.0, 00.1]	
Cochron $\Omega = 176 (p < 0.01)$				
Cocilian Q - 110 (p<0.01)				
Total random effect		92.0	[89.1. 94.2]	(1 1 1 1 1 1 1 1 1 1 1 1 6 1
Heterogeneity: tau ² = 1.03				
,				
				0 10 20 30 40 50 60 70 80 90 100
				Sensitivity of nasopharyngeal swab (%)

Fig 4. Forest plot of the sensitivity of RTqPCR test on nasopharyngeal sample. The confidence intervals for each study are computed using the Clopper-Pearson method. Those for the overall estimates (fixed-effect or random-effect) are based on the normal approximation. The blue box size is proportional to the number of positive tests. The red line corresponds to the value of the overall sensitivity of the random-effect model. This vertical line enables to locate the studies having an estimate sensitivity higher than 92.0%. The heterogeneity estimator 1² is equal to 85%, which means a higher level of heterogeneity.

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91%, 85% and 83.4% in 4, 16 and 5 studies respectively [73–75]. However, these studies used nasopharyngeal positivity as the reference, which did not seem relevant in our context.

To prevent a shortage of analytic reagents and to cut the costs necessarily associated to mass screening strategies, several recent publications have proposed mass testing methods based on saliva sampling either through extraction-free protocols [76–78] or through pre-extraction sample pooling [79–81]. In addition, sample pooling has gained a recognized interest for recurrent screening programs from the Centres of Disease Control recommendations

[82], and surveillance protocols implemented in higher education institutions across the world, e.g. the State University of New York (United States) [83], Liège University (Belgium)
[84], Heidelberg University (Germany) [85] as well as at Nottingham University (United Kingdom) [86].

In conclusion, this meta-analysis conclusively demonstrates that saliva is as valid as nasopharyngeal sampling for the detection of SARS-CoV-2 infections in symptomatic as well as asymptomatic carriers. In contrast to nasopharyngeal swabs, saliva sampling is simple, fast, non-invasive, inexpensive, painless and it thus uniquely applicable for surveillance, screening and diagnosis.

Supporting information

S1 Fig. Saliva sensitivity in each of the 50 studies as a function of SARS-CoV2 prevalence. (TIF)

S2 Fig. Nasopharyngeal sample sensitivity in each of the 50 studies as a function of SARS--CoV2 prevalence.

(TIF)

S3 Fig. Prevalence in the population tested in France by symptomatic status. Week 28 corresponds to the results published the 13th of July 2020. Sources: Points épidémiologiques hebdomadaires.

(TIF)

S1 Checklist. PRISMA 2009 checklist. (DOC)

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References

- Ferretti L, Wymant C, Kendall M, Zhao L, Nurtay A, Abeler-Dorner L, et al. Quantifying SARS-CoV-2 transmission suggests epidemic control with digital contact tracing. Science. 2020; 368(6491). <u>https:// doi.org/10.1126/science.abb6936</u> PMID: 32234805
- 2. Larremore DB, Wilder B, Lester E, Shehata S, Burke JM, Hay JA, et al. Test sensitivity is secondary to frequency and turnaround time for COVID-19 screening. Sci Adv. 2020.
- Lavezzo E, Franchin E, Ciavarella C, Cuomo-Dannenburg G, Barzon L, Del Vecchio C, et al. Suppression of a SARS-CoV-2 outbreak in the Italian municipality of Vo'. Nature. 2020; 584(7821):425–9. https://doi.org/10.1038/s41586-020-2488-1 PMID: 32604404
- Xing Y, Wong GWK, Ni W, Hu X, Xing Q. Rapid Response to an Outbreak in Qingdao, China. N Engl J Med. 2020; 383(23):e129. https://doi.org/10.1056/NEJMc2032361 PMID: 33207089
- Laboratory testing for 2019 novel coronavirus (2019-nCoV) in suspected human cases. https://www. who.int/publications-detail/laboratory-testing-for-2019-novel-coronavirus-in-suspected-human-cases-20200117 (accessed Jan. 3, 2021).
- Overview of Testing for SARS-CoV-2 (COVID-19). https://www.cdc.gov/coronavirus/2019-ncov/hcp/ testing-overview.html?CDC_AA_refVal=https%3A%2F%2Fwww.cdc.gov%2Fcoronavirus%2F2019ncov%2Fhcp%2Fclinical-criteria.html (accessed Jan. 3, 2021).
- Diagnostic testing and screening for SARS-CoV-2. https://www.ecdc.europa.eu/en/covid-19/latest-evidence/diagnostic-testing (accessed Jan. 4, 2021).
- Detection of SASR-CoV-2 in saliva. https://www.has-sante.fr/upload/docs/application/pdf/2021-02/ac_ 2021_0007_rt-pcr_salivaire_covid-19.pdf accessed Apr. 21, 2021).
- Siegler AJ, Hall E, Luisi N, Zlotorzynska M, Wilde G, Sanchez T, et al. Willingness to Seek Diagnostic Testing for SARS-CoV-2 With Home, Drive-through, and Clinic-Based Specimen Collection Locations. Open Forum Infect Dis. 2020; 7(7):ofaa269.
- Woloshin S, Patel N, Kesselheim AS. False Negative Tests for SARS-CoV-2 Infection—Challenges and Implications. N Engl J Med. 2020; 383(6):e38. https://doi.org/10.1056/NEJMp2015897 PMID: 32502334
- Schwarzer G, Chemaitelly H, Abu-Raddad LJ, Rucker G. Seriously misleading results using inverse of Freeman-Tukey double arcsine transformation in meta-analysis of single proportions. Res Synth Methods. 2019; 10(3):476–83. https://doi.org/10.1002/jrsm.1348 PMID: 30945438
- 12. Stijnen T, Hamza TH, Ozdemir P. Random effects meta-analysis of event outcome in the framework of the generalized linear mixed model with applications in sparse data. Stat Med. 2010; 29(29):3046–67. https://doi.org/10.1002/sim.4040 PMID: 20827667
- Higgins JP, Thompson SG, Deeks JJ, Altman DG. Measuring inconsistency in meta-analyses. BMJ. 2003; 327(7414):557–60. https://doi.org/10.1136/bmj.327.7414.557 PMID: 12958120
- 14. Agresti A, Coull BA. Approximate Is Better than "Exact" for Interval Estimation of Binomial Proportions. The American Statistician. 1998; 52(2):119–26.
- **15.** Team RC. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. 2020.
- Balduzzi S, Rucker G, Schwarzer G. How to perform a meta-analysis with R: a practical tutorial. Evid Based Ment Health. 2019; 22(4):153–60. https://doi.org/10.1136/ebmental-2019-300117 PMID: 31563865
- Aita A, Basso D, Cattelan AM, Fioretto P, Navaglia F, Barbaro F, et al. SARS-CoV-2 identification and IgA antibodies in saliva: One sample two tests approach for diagnosis. Clin Chim Acta. 2020; 510:717– 22. https://doi.org/10.1016/j.cca.2020.09.018 PMID: 32946791
- Altawalah H, AlHuraish F, Alkandari WA, Ezzikouri S. Saliva specimens for detection of severe acute respiratory syndrome coronavirus 2 in Kuwait: A cross-sectional study. J Clin Virol. 2020; 132:104652. https://doi.org/10.1016/j.jcv.2020.104652 PMID: 33053493

- Azzi L, Carcano G, Gianfagna F, Grossi P, Gasperina DD, Genoni A, et al. Saliva is a reliable tool to detect SARS-CoV-2. J Infect. 2020; 81(1):e45–e50. https://doi.org/10.1016/j.jinf.2020.04.005 PMID: 32298676
- Babady NE, McMillen T, Jani K, Viale A, Robilotti EV, Aslam A, et al. Performance of Severe Acute Respiratory Syndrome Coronavirus 2 Real-Time RT-PCR Tests on Oral Rinses and Saliva Samples. J Mol Diagn. 2020. https://doi.org/10.1016/j.jmoldx.2020.10.018 PMID: 33217552
- Barat B, Das S, Giorgi V, Henderson DK, Kopka S, Lau AF, et al. Pooled Saliva Specimens for SARS-CoV-2 Testing. medRxiv. 2020. https://doi.org/10.1101/2020.10.02.20204859 PMID: 33052363
- Berenger BM, Conly JM, Fonseca K, Hu J, Louie T, Schneider AR, et al. Saliva collected in universal transport media is an effective, simple and high-volume amenable method to detect SARS-CoV-2. Clin Microbiol Infect. 2020. https://doi.org/10.1016/j.cmi.2020.10.035 PMID: 33160035
- Bhattacharya D, Parai D, Rout UK, Dash P, Nanda RR, Dash GC, et al. Saliva for diagnosis of SARS-CoV-2: first report from India. J Med Virol. 2020. https://doi.org/10.1002/jmv.26719 PMID: 33295640
- Binder RA, Alarja NA, Robie ER, Kochek KE, Xiu L, Rocha-Melogno L, et al. Environmental and Aerosolized Severe Acute Respiratory Syndrome Coronavirus 2 Among Hospitalized Coronavirus Disease 2019 Patients. J Infect Dis. 2020; 222(11):1798–806. https://doi.org/10.1093/infdis/jiaa575 PMID: 32905595
- Borghi E, Massa V, Carmagnola D, Dellavia C, Parodi C, Ottaviano E, et al. Saliva sampling for chasing SARS-CoV-2: A Game-changing strategy. Pharmacol Res. 2020:105380. <u>https://doi.org/10.1016/j.</u> phrs.2020.105380 PMID: 33338623
- Braz-Silva PH, Mamana AC, Romano CM, Felix AC, de Paula AV, Fereira NE, et al. Performance of athome self-collected saliva and nasal-oropharyngeal swabs in the surveillance of COVID-19. J Oral Microbiol. 2020; 13(1):1858002. https://doi.org/10.1080/20002297.2020.1858002 PMID: 33391631
- Byrne RL, Kay GA, Kontogianni K, Aljayyoussi G, Brown L, Collins AM, et al. Saliva Alternative to Upper Respiratory Swabs for SARS-CoV-2 Diagnosis. Emerg Infect Dis. 2020; 26(11):2770–1. <u>https://doi.org/ 10.3201/eid2611.203283</u> PMID: 32917294
- Cassinari K, Alessandri-Gradt E, Chambon P, Charbonnier F, Gracias S, Beaussire L, et al. Assessment of multiplex digital droplet RT-PCR as a diagnostic tool for SARS-CoV-2 detection in nasopharyngeal swabs and saliva samples. Clin Chem. 2020.
- Chau NVV, Thanh Lam V, Thanh Dung N, Yen LM, Minh NNQ, Hung LM, et al. The natural history and transmission potential of asymptomatic SARS-CoV-2 infection. Clin Infect Dis. 2020.
- Chen L, Zhao J, Peng J, Li X, Deng X, Geng Z, et al. Detection of SARS-CoV-2 in saliva and characterization of oral symptoms in COVID-19 patients. Cell Prolif. 2020; 53(12):e12923. <u>https://doi.org/10.1111/cpr.12923</u> PMID: 33073910
- Guclu E, Koroglu M, Yurumez Y, Toptan H, Kose E, Guneysu F, et al. Comparison of saliva and oronasopharyngeal swab sample in the molecular diagnosis of COVID-19. Rev Assoc Med Bras (1992). 2020; 66(8):1116–21. https://doi.org/10.1590/1806-9282.66.8.1116 PMID: 32935807
- Hanege FM, Kocoglu E, Kalcioglu MT, Celik S, Cag Y, Esen F, et al. SARS-CoV-2 Presence in the Saliva, Tears, and Cerumen of COVID-19 Patients. Laryngoscope. 2020.
- Hanson KE, Barker AP, Hillyard DR, Gilmore N, Barrett JW, Orlandi RR, et al. Self-Collected Anterior Nasal and Saliva Specimens versus Health Care Worker-Collected Nasopharyngeal Swabs for the Molecular Detection of SARS-CoV-2. J Clin Microbiol. 2020; 58(11). <u>https://doi.org/10.1128/JCM.</u> 01824-20 PMID: 32817233
- Hasanoglu I, Korukluoglu G, Asilturk D, Cosgun Y, Kalem AK, Altas AB, et al. Higher viral loads in asymptomatic COVID-19 patients might be the invisible part of the iceberg. Infection. 2020.
- Iwasaki S, Fujisawa S, Nakakubo S, Kamada K, Yamashita Y, Fukumoto T, et al. Comparison of SARS-CoV-2 detection in nasopharyngeal swab and saliva. J Infect. 2020; 81(2):e145–e7. https://doi.org/10.1016/j.jinf.2020.05.071 PMID: 32504740
- Jamal AJ, Mozafarihashjin M, Coomes E, Anceva-Sami S, Barati S, Crowl G, et al. Sensitivity of midturbinate versus nasopharyngeal swabs for the detection of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). Infect Control Hosp Epidemiol. 2020:1–3.
- Jamal AJ, Mozafarihashjin M, Coomes E, Powis J, Li AX, Paterson A, et al. Sensitivity of nasopharyngeal swabs and saliva for the detection of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). Clin Infect Dis. 2020.
- Kandel C, Zheng J, McCready J, Serbanescu MA, Racher H, Desaulnier M, et al. Detection of SARS-CoV-2 from Saliva as Compared to Nasopharyngeal Swabs in Outpatients. Viruses. 2020; 12(11). https://doi.org/10.3390/v12111314 PMID: 33212817
- Kojima N, Turner F, Slepnev V, Bacelar A, Deming L, Kodeboyina S, et al. Self-Collected Oral Fluid and Nasal Swab Specimens Demonstrate Comparable Sensitivity to Clinician-Collected Nasopharyngeal

Swab Specimens for the Detection of SARS-CoV-2. Clin Infect Dis. 2020. https://doi.org/10.1093/cid/ ciaa1589 PMID: 33075138

- Landry ML, Criscuolo J, Peaper DR. Challenges in use of saliva for detection of SARS CoV-2 RNA in symptomatic outpatients. J Clin Virol. 2020; 130:104567. <u>https://doi.org/10.1016/j.jcv.2020.104567</u> PMID: 32750665
- Leung EC, Chow VC, Lee MK, Lai RW. Deep throat saliva as an alternative diagnostic specimen type for the detection of SARS-CoV-2. J Med Virol. 2020. https://doi.org/10.1002/jmv.26258 PMID: 32621616
- Matic N, Stefanovic A, Leung V, Lawson T, Ritchie G, Li L, et al. Practical challenges to the clinical implementation of saliva for SARS-CoV-2 detection. Eur J Clin Microbiol Infect Dis. 2020. <u>https://doi.org/10.1007/s10096-020-04090-5</u> PMID: 33236269
- 43. McCormick-Baw C, Morgan K, Gaffney D, Cazares Y, Jaworski K, Byrd A, et al. Saliva as an Alternate Specimen Source for Detection of SARS-CoV-2 in Symptomatic Patients Using Cepheid Xpert Xpress SARS-CoV-2. J Clin Microbiol. 2020; 58(8). https://doi.org/10.1128/JCM.01109-20 PMID: 32414838
- Migueres M, Mengelle C, Dimeglio C, Didier A, Alvarez M, Delobel P, et al. Saliva sampling for diagnosing SARS-CoV-2 infections in symptomatic patients and asymptomatic carriers. J Clin Virol. 2020; 130:104580. https://doi.org/10.1016/j.jcv.2020.104580 PMID: 32781366
- 45. Moreno-Contreras J, Espinoza MA, Sandoval-Jaime C, Cantu-Cuevas MA, Baron-Olivares H, Ortiz-Orozco OD, et al. Saliva Sampling and Its Direct Lysis, an Excellent Option To Increase the Number of SARS-CoV-2 Diagnostic Tests in Settings with Supply Shortages. J Clin Microbiol. 2020; 58(10). https://doi.org/10.1128/JCM.01659-20 PMID: 32703816
- 46. Nagura-Ikeda M, Imai K, Tabata S, Miyoshi K, Murahara N, Mizuno T, et al. Clinical Evaluation of Self-Collected Saliva by Quantitative Reverse Transcription-PCR (RT-qPCR), Direct RT-qPCR, Reverse Transcription-Loop-Mediated Isothermal Amplification, and a Rapid Antigen Test To Diagnose COVID-19. J Clin Microbiol. 2020; 58(9).
- 47. Pasomsub E, Watcharananan SP, Boonyawat K, Janchompoo P, Wongtabtim G, Suksuwan W, et al. Saliva sample as a non-invasive specimen for the diagnosis of coronavirus disease 2019: a cross-sectional study. Clin Microbiol Infect. 2020.
- Procop GW, Shrestha NK, Vogel S, Van Sickle K, Harrington S, Rhoads DD, et al. A Direct Comparison of Enhanced Saliva to Nasopharyngeal Swab for the Detection of SARS-CoV-2 in Symptomatic Patients. Journal of Clinical Microbiology. 2020; 58(11):e01946–20. <u>https://doi.org/10.1128/JCM.</u> 01946-20 PMID: 32883744
- **49.** Rao M, Rashid FA, Sabri F, Jamil NN, Zain R, Hashim R, et al. Comparing nasopharyngeal swab and early morning saliva for the identification of SARS-CoV-2. Clin Infect Dis. 2020.
- Sakanashi D, Asai N, Nakamura A, Miyazaki N, Kawamoto Y, Ohno T, et al. Comparative evaluation of nasopharyngeal swab and saliva specimens for the molecular detection of SARS-CoV-2 RNA in Japanese patients with COVID-19. J Infect Chemother. 2021; 27(1):126–9. https://doi.org/10.1016/j.jiac. 2020.09.027 PMID: 33060046
- Senok A, Alsuwaidi H, Atrah Y, Al Ayedi O, Al Zahid J, Han A, et al. Saliva as an Alternative Specimen for Molecular COVID-19 Testing in Community Settings and Population-Based Screening. Infect Drug Resist. 2020; 13:3393–9. https://doi.org/10.2147/IDR.S275152 PMID: 33061486
- Skolimowska K, Rayment M, Jones R, Madona P, Moore LSP, Randell P. Non-invasive saliva specimens for the diagnosis of COVID-19: caution in mild outpatient cohorts with low prevalence. Clin Microbiol Infect. 2020; 26(12):1711–3. https://doi.org/10.1016/j.cmi.2020.07.015 PMID: 32688069
- SoRelle JA, Mahimainathan L, McCormick-Baw C, Cavuoti D, Lee F, Thomas A, et al. Saliva for use with a point of care assay for the rapid diagnosis of COVID-19. Clin Chim Acta. 2020; 510:685–6. https://doi.org/10.1016/j.cca.2020.09.001 PMID: 32910978
- Sui Z, Zhang Y, Tu J, Xie J, Huang W, Peng T, et al. Evaluation of saliva as an alternative diagnostic specimen source for SARS-CoV-2 detection by RT-dPCR. J Infect. 2020. https://doi.org/10.1016/j.jinf. 2020.11.023 PMID: 33248219
- 55. Torres M, Collins K, Corbit M, Ramirez M, Winters CR, Katz L, et al. Comparison of saliva and nasopharyngeal swab SARS-CoV-2 RT-qPCR testing in a community setting. J Infect. 2020.
- 56. Uwamino Y, Nagata M, Aoki W, Fujimori Y, Nakagawa T, Yokota H, et al. Accuracy and stability of saliva as a sample for reverse transcription PCR detection of SARS-CoV-2. J Clin Pathol. 2021; 74 (1):67–8. https://doi.org/10.1136/jclinpath-2020-206972 PMID: 32928941
- Vaz SN, Santana DS, Netto EM, Pedroso C, Wang WK, Santos FDA, et al. Saliva is a reliable, non-invasive specimen for SARS-CoV-2 detection. Braz J Infect Dis. 2020; 24(5):422–7. https://doi.org/10.1016/ j.bjid.2020.08.001 PMID: 32888905

- Vogels CBF, Watkins AE, Harden CA, Brackney DE, Shafer J, Wang J, et al. SalivaDirect: A simplified and flexible platform to enhance SARS-CoV-2 testing capacity. Med. <u>https://doi.org/10.1016/j.medj.</u> 2020.12.010 PMID: 33521748
- Williams E, Bond K, Zhang B, Putland M, Williamson DA. Saliva as a Noninvasive Specimen for Detection of SARS-CoV-2. J Clin Microbiol. 2020; 58(8). <u>https://doi.org/10.1128/JCM.00776-20</u> PMID: 32317257
- Wong S, Tse H, Siu HK, Kwong TS, Chu MY, Yau FYS, et al. Posterior oropharyngeal saliva for the detection of SARS-CoV-2. Clin Infect Dis. 2020.
- 61. Wyllie AL, Fournier J, Casanovas-Massana A, Campbell M, Tokuyama M, Vijayakumar P, et al. Saliva or Nasopharyngeal Swab Specimens for Detection of SARS-CoV-2. N Engl J Med. 2020.
- 62. Yee R, Truong T, Pannaraj PS, Eubanks N, Gai E, Jumarang J, et al. Saliva is a Promising Alternative Specimen for the Detection of SARS-CoV-2 in Children and Adults. J Clin Microbiol. 2020. <u>https://doi.org/10.1101/2020.10.25.20219055</u> PMID: 33140064
- Yokota I, Shane PY, Okada K, Unoki Y, Yang Y, Inao T, et al. Mass screening of asymptomatic persons for SARS-CoV-2 using saliva. Clin Infect Dis. 2020. https://doi.org/10.1093/cid/ciaa1388 PMID: 32976596
- Zhu J, Guo J, Xu Y, Chen X. Viral dynamics of SARS-CoV-2 in saliva from infected patients. J Infect. 2020; 81(3):e48–e50. https://doi.org/10.1016/j.jinf.2020.06.059 PMID: 32593658
- Caulley L, Corsten M, Eapen L, Whelan J, Angel JB, Antonation K, et al. Salivary Detection of COVID-19. Ann Intern Med. 2020. https://doi.org/10.7326/M20-4738 PMID: 32857591
- Otto MP, Darles C, Valero E, Benner P, Dutasta F, Janvier F. Posterior oropharyngeal salivafor the detection of SARS-CoV-2. Clin Infect Dis. 2020. https://doi.org/10.1093/cid/ciaa1181 PMID: 32770241
- COVID-19: HAS positions antigenic tests in three situations Press release—Posted on 09 Oct 2020 https://www.has-sante.fr/jcms/p_3212125/fr/covid-19-la-has-positionne-les-tests-antigeniques-danstrois-situations.
- Winnett A, Cooper MM, Shelby N, Romano AE, Reyes JA, Ji J, et al. SARS-CoV-2 Viral Load in Saliva Rises Gradually and to Moderate Levels in Some Humans. medRxiv. 2020:2020.12.09.20239467. https://doi.org/10.1101/2020.12.09.20239467 PMID: 33330885
- Mina MJ, Parker R, Larremore DB. Rethinking Covid-19 Test Sensitivity—A Strategy for Containment. N Engl J Med. 2020; 383(22):e120. https://doi.org/10.1056/NEJMp2025631 PMID: 32997903
- 70. Arons MM, Hatfield KM, Reddy SC, Kimball A, James A, Jacobs JR, et al. Presymptomatic SARS-CoV-2 Infections and Transmission in a Skilled Nursing Facility. N Engl J Med. 2020; 382(22):2081–90. https://doi.org/10.1056/NEJMoa2008457 PMID: 32329971
- He X, Lau EHY, Wu P, Deng X, Wang J, Hao X, et al. Temporal dynamics in viral shedding and transmissibility of COVID-19. Nat Med. 2020; 26(5):672–5. <u>https://doi.org/10.1038/s41591-020-0869-5</u> PMID: 32296168
- 72. Widders A, Broom A, Broom J. SARS-CoV-2: The viral shedding vs infectivity dilemma. Infect Dis Health. 2020; 25(3):210–5. https://doi.org/10.1016/j.idh.2020.05.002 PMID: 32473952
- 73. Czumbel LM, Kiss S, Farkas N, Mandel I, Hegyi A, Nagy A, et al. Saliva as a Candidate for COVID-19 Diagnostic Testing: A Meta-Analysis. Front Med (Lausanne). 2020; 7:465. <u>https://doi.org/10.3389/</u> fmed.2020.00465 PMID: 32903849
- Kivela JM, Jarva H, Lappalainen M, Kurkela S. Saliva-based testing for diagnosis of SARS-CoV-2 infection: A meta-analysis. J Med Virol. 2020. https://doi.org/10.1002/jmv.26613 PMID: 33079392
- 75. Ricco M, Ranzieri S, Peruzzi S, Valente M, Marchesi F, Balzarini F, et al. RT-qPCR assays based on saliva rather than on nasopharyngeal swabs are possible but should be interpreted with caution: results from a systematic review and meta-analysis. Acta Biomed. 2020; 91(3):e2020025. https://doi.org/10. 23750/abm.v91i3.10020 PMID: 32921721
- 76. Ott IM, Strine MS, Watkins AE, Boot M, Kalinich CC, Harden CA, et al. Simply saliva: stability of SARS-CoV-2 detection negates the need for expensive collection devices. medRxiv. 2020. <u>https://doi.org/10.1101/2020.08.03.20165233</u> PMID: 32793924
- Ranoa DRE, Holland RL, Alnaji FG, Green KJ, Wang L, Brooke CB, et al. Saliva-Based Molecular Testing for SARS-CoV-2 that Bypasses RNA Extraction. bioRxiv. 2020:2020.06.18.159434.
- Vogels C.B.F. B DE, Wang J., Kalinich C.C., Ott I.M., Kudo E., Lu P., et al. SalivaDirect: simple and sensitive molecular diagnostic test for SARS-Cov-2 surveillance. medRxive. 2020.
- 79. Gavars D, Gavars M, Perminovs D, Stasulans J, Stana J, Metla Z, et al. Saliva as testing sample for SARS-CoV-2 detection by RT-PCR in low prevalence community setting. medRxiv. 2020:2020.10.20.20216127.

- Sahajpal NS, Mondal AK, Ananth S, Njau A, Ahluwalia P, Chaubey A, et al. SalivaAll: Clinical validation of a sensitive test for saliva collected in healthcare and community settings with pooling utility for SARS-CoV-2 mass surveillance. medRxiv. 2020:2020.08.26.20182816.
- Watkins A.E. F EP, Weinberger Daniel M., Vogels Chantal B.F., Brackney Doug E., Casanovas-Massana Arnau, Campbell Melissa, et al. Pooling saliva to increase SARS-CoV-2 testing capacity. MedRxiv. 2020. https://doi.org/10.1101/2020.09.02.20183830 PMID: 32909003
- Interim Guidance for Use of Pooling Procedures in SARS-CoV-2 Diagnostic, Screening, and Surveillance Testing. https://www.cdc.gov/coronavirus/2019-ncov/lab/pooling-procedures.html (accessed Jan. 4, 2021).
- 83. SUNY COVID-19 Case Tracker. https://www.suny.edu/COVID19-tracker/ (accessed Jan. 4, 2021).
- 84. Screening in practice. https://www.coronavirus.uliege.be/cms/c_12199182/en/screening-in-practice (accessed Jan. 4, 2021).
- **85.** New Corona testing strategies for the general public. <u>https://www.virusfinder.de/en/</u> (accessed Jan. 4, 2021).
- Fogarty A, Joseph A, Shaw D. Pooled saliva samples for COVID-19 surveillance programme. Lancet Respir Med. 2020; 8(11):1078–80. https://doi.org/10.1016/S2213-2600(20)30444-6 PMID: 32976755