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Clinitest rapid COVID-19 antigen test for the diagnosis of SARS-CoV-2 infection: A multicenter evaluation study

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ABSTRACT

Objectives: RT-PCR assay is the reference method for diagnosis of COVID-19, but it is also a laborious and time-consuming technic, limiting the availability of testing. Rapid antigen-detection tests are faster and less expensive; however, the reliability of these tests must be validated before they can be used widely. The objective of this study was to determine the performance of the Clinitest Rapid COVID-19 Antigen Test (ClinitestRT) (SIEMENS) for SARS-CoV-2 in nasopharyngeal swab specimens.

Methods: This prospective multicenter study was carried out in three Spanish university hospitals including individuals with clinical symptoms or epidemiological criteria for COVID-19. Only individuals with ≤7 days from the onset of symptoms or from exposure to a confirmed case of COVID-19 were included. Two nasopharyngeal samples were taken to perform the ClinitestRT, as a point-of-care test, and a diagnostic RT-PCR test.

Results: Overall sensitivity and specificity for the ClinitestRT among the 450 patients studied were 93.3% (CI 95%: 89.7–96.8) and 99.2% (CI 95%: 97.2–99.8), respectively. Sensitivity in participants with ≤5 days of the clinical course was 93.6% (CI 95%: 89.2–96.3), and in participants who had a $C_T < 25$ for the RT-PCR test was 98.4% (CI 95%: 94.5–99.6). Agreement between techniques was 96.7% (kappa score: 0.93; CI 95%: 0.90–0.97).

Conclusions: The ClinitestRT provides good clinical performance, with more reliable results for patients with a higher viral load. The results must be interpreted based on the local epidemiological context.

1. Introduction

There are already multiple CE-marked commercial rapid antigen-detection tests (RADT) for the diagnosis of SARS-CoV-2 infection [1]. RADT detect SARS-CoV-2 proteins produced by replicating viruses in respiratory secretions [2], and they are compatible with both laboratory and near-patient use. RADT allow the generalization of decentralized diagnostic tests for SARS-CoV-2 infection, which are one of the key points for the control of the COVID-19 pandemic. However, independent evaluation studies comparing RADT to the RT-PCR, using different swabs and strictly following manufacturers' instructions, are frequently lacking [1].

Clinitest Rapid COVID-19 Antigen Test (ClinitestRT) (Siemens, Healthineers, Erlangen, Germany) is a rapid in vitro test for the qualitative detection of the SARS-CoV-2 nucleocapsid protein in nasopharyngeal or nasal swabs to diagnose COVID-19 within the first week after symptoms onset.

ClinitestRT is a lateral-flow-format test that uses

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immunochromatography with colloidal gold, and it is designed to be performed at the patient care site by trained healthcare personnel, with results in 15 min.

The objective of this study is to determine the performance of this test, with CE marking, in nasopharyngeal swabs.

2. Materials and methods

2.1. Hospitals and individuals participating

Between November 2020 and January 2021, we carried out an independent, prospective multicenter diagnostic evaluation study across three independent university hospitals in two Spanish autonomous communities (Madrid and Galicia). The hospitals participating in the study were Hospital Clínico Universitario San Carlos and Hospital Universitario La Paz from Madrid, and Complejo Hospitalario Universitario de A Coruña from Galicia.

Individuals with clinical symptoms or epidemiological criteria (asymptomatic close contacts) for COVID-19 in whom a diagnostic RT-PCR test was indicated were offered to participate in this study. All participants were reported as part of the study and verbal informed consent was obtained prior to their inclusion. The ClinitestRT result did not influence the clinical management of the patients, who were informed and treated only based on the RT-PCR result. The symptoms, number of days since the onset of symptoms or exposure, threshold cycle (C_T) values for PCR, and demographic data were collected for all participants. Participant's data were coded and no samples were stored after the ClinitestRT was performed. Only individuals with ≤7 days from the onset of symptoms or from exposure to a confirmed case of COVID-19 were included, according to recommendations of the WHO [2]. The study was presented to the Research Ethics Committee, which responded favorably.

2.2. SARS-CoV-2 testing

Two nasopharyngeal samples were taken per patient, one in each choana. One of them was taken with the swab provided by ClinitestRT, and the other one with a suitable swab for taking a virus sample including a universal transport medium for RT-PCR (Copan flocked swabs with UTM™, Universal Transport Medium). ClinitestRT was performed immediately, under point-of-care conditions (regulations of quality systems ISO 15,189) [3,4], and according to the manufacturer's instructions (lot number 2011072) by physicians and nurses from emergency and primary care services trained by microbiology specialists. The second swab was used for a molecular diagnostic (RT-PCR) by each hospital according to its standard procedures for COVID-19 diagnosis. The commercial RT-PCR methods used for the participants hospitals in this study were Allplex™ 2019-nCoV Assay (Seegene, Seoul, South Korea), GENOMICA S.A.U. (Madrid, Spain), TaqPath COVID-19 Combo Kit (Thermo Fisher, Waltham, MA, USA), GeneXpert (Cepheid, Sunnyvale, CA, USA), and Cobas 6800 (Roche, Indianapolis, USA) Due to the high diagnostic demand, in some hospitals different PCR techniques were used.

2.3. Statistical analyses

Specificity and sensitivity, with 95% confidence intervals (CI), of ClinitestRT were calculated using the RT-PCR results as the standard; or which is the same as proportion of negative and positive agreement, respectively. Sensitivity was calculated for all patients and for specific groups of patients according to the time of onset of symptoms or exposure, RT-PCR C_T values and symptoms. The level of agreement between the tests was evaluated using Cohen's kappa score [5]. Statistical analyses were performed using GraphPad Prism software v.7.02 (GraphPad Software Inc., San Diego, CA, USA).

3. Results

A total of 450 individuals who had at least one symptom compatible with COVID-19 ($n = 410$) or had been in close contact with a diagnosed COVID-19 patient ($n = 40$) were included in this study (Table 1). There were between 124 and 200 individuals from each participating hospital, with a median age of 65 years (interquartile range: 44); 58.4% were women (Table 1). There were not cases in pediatric patients (≤14 years old).

RT-PCR was positive in 192 (42.7%) and negative in 258 (57.3%); while ClinitestRT was positive in 181 (40.2%) and negative in 269 (59.8%) (Table 2) The agreement between both methods was 96.7% (kappa score: 0.93; CI 95%: 0.90–0.97). In 15 patients the results differed between the two tests, 13 of them were false negatives results with ClinitestRT (2.9% of total cases) (Table 2). Twelve (92.3%) false negatives had $C_T \geq 25$ values for RT-PCR.

Based on these data, the overall sensitivity and specificity of the ClinitestRT were 93.3% (CI 95%: 89.7–96.8) and 99.2% (CI 95%: 97.2–99.8), respectively (Table 3). Sensitivity was higher in patients who had a $C_T < 25$ for the RT-PCR (98.4%; CI 95%: 94.5–99.6) (Table 3), than in those with $C_T \geq 25$ (80%; CI 95%: 68.9.−91.1). Sensitivity was also higher in patients with ≤5 days of the clinical course of the disease (93.6%; CI 95%: 89.2–96.3) (Table 3) than in those with 6–7 days of clinical evolution (66.7%; CI 95%: 13.3–120); although only three positive cases were included in this last group.

Among the 40 asymptomatic participants, 23 (92%) of the 25 positive by RT-PCR were also positive by ClinitestRT, and there was full agreement in the 15 that were negative.

The negative predictive value (NPV) and positive predictive value (PPV) in the study cohort, were 95.2% and 98.9%, respectively (Table 3).

4. Discussion

This study showed very good clinical performance values of the ClinitestRT, with 93.3% sensitivity and 99.2% specificity; moreover, sensitivity was even improved in samples with $C_T < 25$ (98.4%) which, is probably closer to the limit of infectivity, as previously reported [6].

The strengths of this study include the large study size and the prospective nature of the study, the inclusion of multiple centers, and its use according to the manufacturer's instructions under point-of-care conditions. A limitation of the study may be the use of different RT-PCR protocols because the C_T values can vary slightly between techniques; however, all of them are used for routine diagnosis in participating hospitals, and all of them are validated and widely used worldwide.

The gap between the number of diagnostic needs and the testing capacity are still evident in epidemic waves [7]. RADT are simple to perform and interpret by minimally trained health workers at the point-of-care, they do not require specific equipment, they are less expensive than RT-PCR, and they improve the turnaround time for results [8,9]. Although during the first wave of the pandemic, RADT were

Table 1
Study cohort included in the validation study of the Clinitest Rapid COVID-19 Antigen Test of SIEMENS.

Total N (valid PCR results)	450
Positive PCR [% (n)]	**42.7% (192)**
Age [median (interquartile range)]	**65 (44)**
Gender [% F, (n/N)]	**58.4% (263/450)**
Symptoms present [% Yes, (n/N)]	**91.1% (410/450)**
Days from symptom onset or from exposure [mean (N)]	**2.9 (450)**
Days ≤ 5 [n/N (%)]	**431/450 (95.8%)**
Days 6–7 [(n/N (%)]	**19/450 (4.2%)**
PCR C_T (n)	**(178)**
$C_T \geq 25$ [n, (%)]	**[50, (28.1%)]**
$C_T < 25$ [n, (%)]	**[128, (71.9%)]**

Table 2
Summary of the results of the Clinitest Rapid COVID-19 Antigen Test compared to RT-PCR.

	Clinitest Rapid COVID-19 antigen test of SIEMENS.		
RT-PCR	Positive	Negative	TOTAL
Positive	179	13	192
Negative	2	256	258
TOTAL	181	269	450

Table 3
Estimation of clinical performance of the Clinitest Rapid COVID-19 Antigen Test compared to RT-PCR.

Relative Sensitivity (CI 95%)	93.3% (89.7–96.8)
Sensitivity days ≤ 5 (CI 95%)	93.6% (89.2–96.3)
Sensitivity $C_T < 25$ (CI 95%)	98.4% (94.5–99.6)
Relative Specificity (CI 95%)	99.2% (97.2–99.8)
Agreement (kappa index; CI 95%)	96.7% (0.93; 0.90–0.97)
Positive predictive value (CI 95%)	98.9% (96.1–99.7)
Negative predictive value (CI 95%)	95.2% (91.9–97.2)

not recommended due to poor reliability [10,11], recent studies have shown a significant improvement in the performance of newly developed RADTs [6,12–15].

In a recent Spanish study with ClinitestRT, including 178 symptomatic patients with suspected COVID-19 and 92 asymptomatic closed contacts [15], the sensitivity observed in each group was 80.3% and 60%, respectively. In that study [15], the number of participants with C_T values > 30, although it was not detailed, probably was high, as can be deduced by the difference in the sensitivity between patients with C_T values $\leq$ 30 ($S = 94.6\%$) and patients with C_T values < 33 ($S = 75.9\%$); this fact may explain the different sensitivity obtained with respect to the study presented here.

WHO guidelines require that SARS-CoV-2 RADT demonstrate ≥80% sensitivity and ≥97% specificity compared to the RT-PCR reference assay [2], while ECDC suggests aiming to use tests with a performance closer to RT-PCR, i.e. ≥90% sensitivity and ≥97% specificity [16]. According to these criteria, our study support the clinical use of the ClinitestRT in patients with symptoms of COVID-19 with a short clinical course (≤5–7 days) of the disease. Although in this study susceptibility obtained in asymptomatic patients was 92%, general conclusions cannot be drawn from these results given the small number of cases included of this group. In general, previous studies have shown a lower sensitivity of RADTs in asymptomatic close contacts than in patients with symptoms [16,17]. This is consistent with the advice from the WHO against using RADT for screening asymptomatic individuals in populations with low COVID-19 prevalence [2] due to the potential increase of higher incidence of false positives.

The performance of an RADT may depend on the epidemiological situation of the population being tested; therefore, how the test is used and how the results are interpreted will depend on local epidemiological factors [2]. As with all diagnostic tests, to correctly interpret and act on the results of the RADT, the prevalence of disease must be previously estimated, since this determines the PPV and NPV values. In an epidemiological setting of high prevalence, the pre-test probability of COVID-19 disease is relatively high, and positive test results have a high predictive value. On the contrary, if the NPV is low, it should advise patients with respiratory symptoms to exercise infection control practices, and consider repeat RADT if symptoms persist or progress [2].

In conclusion, this study showed that the ClinitestRT provides very good clinical performance as a point-of-care test. The use of RADT can have an undoubted impact on diagnostic strategies for COVID-19, helping to prevent an overload on health care services and laboratories saturation at the peaks of epidemic waves. However, the results of RADT performance must be interpreted based on the local epidemiological context.

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CRediT authorship contribution statement

Paloma Merino-Amador: Data curation, Writing – original draft, Investigation, Writing – review & editing. **Patricia González-Donapetry:** Investigation, Data curation. **Mercedes Domínguez-Fernández:** Investigation, Data curation. **Fernando González-Romo:** Investigation, Data curation. **Miguel Ángel Sánchez-Castellano:** Investigation, Data curation. **Alejandro Seoane-Estevez:** Investigation, Data curation. **Alberto Delgado-Iribarren:** Investigation. **Julio García:** Investigation. **Germán Bou:** Investigation. **Manuel Cuenca-Estrella:** Conceptualization, Supervision, Investigation. **Jesús Oteo-Iglesias:** Conceptualization, Methodology, Writing – original draft.

Declaration of Competing Interest

Authors have none to declare.

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