



Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Elsevier hereby grants permission to make all its COVID-19-related research that is available on the COVID-19 resource centre - including this research content - immediately available in PubMed Central and other publicly funded repositories, such as the WHO COVID database with rights for unrestricted research re-use and analyses in any form or by any means with acknowledgement of the original source. These permissions are granted for free by Elsevier for as long as the COVID-19 resource centre remains active.



Short communication

Comparison of the SARS-CoV-2 Rapid antigen test to the real star Sars-CoV-2 RT PCR kit

Alexander Krüttgen^a, Christian G. Cornelissen^b, Michael Dreher^b, Mathias W. Horne^c, Matthias Imöhl^a, Michael Kleines^{a,*}^a Laboratory Diagnostic Center, University Hospital RWTH Aachen, Germany^b Department of Pneumology and Intensive Care Medicine, University Hospital RWTH Aachen, Germany^c Institute of Medical Microbiology, University Hospital RWTH Aachen, Germany

ARTICLE INFO

Keywords:

COVID-19

Lateral flow assay

Antigen

SARS-CoV-2

ABSTRACT

There is an ongoing need for reliable antigen assays for timely and easy detection of individuals with acute SARS-CoV-2 infection. Using 75 swabs from patients previously tested positive by SARS-CoV-2 PCR and 75 swabs from patients previously tested negative by SARS-CoV-2 PCR, we investigated the sensitivity and specificity of the SARS-CoV-2 Rapid Antigen Test (Roche). We determined a specificity of 96 %. The assay's sensitivity with samples with a cycle threshold of < 25, 25 - <30, 30 - <35, and > = 35 was 100 %, 95 %, 44.8 % and 22.2 %, respectively. We conclude that sensitivity and specificity of the antigen assay is inferior to the PCR assay. However, the antigen assay may be a quick and easy to perform alternative for differentiation of individuals contagious for SARS-CoV-2 from non-contagious individuals.

SARS-CoV-2, a novel coronavirus, emerged end of 2019 (Zhou et al., 2020; Zhu et al., 2020) causing an ongoing pandemic. Correct and timely identification of acute and past infections with SARS-CoV-2 are an urgent need. Nucleic acid amplification assays are the gold standard for detection of acute infections (Eis-Hübing et al., 2020). Sensitive and specific serologic assays are available for detection of past infections (Krüttgen et al., 2020a, Krüttgen et al. 2020b). Standard PCR protocols are sensitive and specific but they are time consuming. Nucleic acid amplification protocols with time to result of less than 1 h have become available, however they are expensive and the production capacity for these assays is far too low.

SARS-CoV-2 specific antigen assays, a simple and fast alternative for nucleic acid amplification assays, have become available recently. Here we describe the comparison of the SARS-CoV-2 Rapid Antigen Test (Roche, Switzerland) and the Real Star SARS-CoV-2 RT PCR Kit (Altona, Germany) (van Kasteren et al., 2020).

75 nasopharyngeal swabs from patients admitted to the RWTH Aachen University hospital (approval of Medical Ethics Committee EK093/2020) tested SARS-CoV-2 RNA-negative with the Real Star SARS-CoV-2 RT-PCR Kit as recommended by the manufacturer were collected and investigated using the SARS-CoV-2 Rapid Antigen Test, a lateral flow assay, without intermitting freeze-thaw cycle. Briefly,

350 µL of swab transport medium were mixed with extraction buffer provided by the manufacturer. Three drops of the mixture were applied to the lateral flow device. After an incubation at room temperature for 15–30 min the test result was read out. Samples showing both, control line and test line, were regarded SARS-CoV-2 antigen-positive, samples showing only the control line were regarded SARS-CoV-2 antigen-negative. 75 further nasopharyngeal swabs from patients tested SARS-CoV-2 RNA-positive with the Real Star SARS-CoV-2 RT-PCR Kit and representing a wide range of cycle thresholds (Ct) were investigated using the SARS-CoV-2 Rapid Antigen Test without intermitting freeze-thaw cycle as described above (Ct < 20: n = 5, Ct 20–<25: n = 12, Ct 25–<30: n = 20, Ct 30–<35: n = 29, Ct > = 35: n = 9).

75 SARS-CoV-2 positive and 75 SARS-CoV-2 negative nasopharyngeal swabs were analyzed using the SARS-CoV-2 Rapid Antigen Test (Roche). Of 75 specimens tested SARS-CoV-2 RNA negative, three had a positive result in the SARS-CoV-2 Rapid Antigen Test (Table 1). This calculates to a specificity of 96 %. Of the 75 specimens tested SARS-CoV-2 RNA positive 53 had a positive result in the SARS-CoV-2 Rapid Antigen Test. This results in an overall sensitivity of 707%. However, the overall sensitivity is strictly dependent on the distribution of cycle thresholds (Ct) within the population of specimens and does not allow a realistic evaluation of the assay. Thus, the sensitivity was recalculated in a Ct-

* Corresponding author at: Laboratory Diagnostic Center, University Hospital RWTH Aachen, Pauwelsstr. 30, 52074 Aachen, Germany.

E-mail address: mkleines@ukaachen.de (M. Kleines).

<https://doi.org/10.1016/j.jviromet.2020.114024>

Received 9 October 2020; Received in revised form 17 November 2020; Accepted 18 November 2020

Available online 20 November 2020

0166-0934/© 2020 Elsevier B.V. All rights reserved.

Table 1

Number of SARS-CoV-2 antigen positive- and negative samples in a population of 75 SARS-CoV-2 positive and 75 SARS-CoV-2 negative specimens.

	SARS-CoV-2 RNA positive	SARS-CoV-2 RNA negative	
SARS-CoV-2 antigen positive	53	3	56
SARS-CoV-2 RNA negative	22	72	94

dependent manner (Fig. 1). For specimens with a high viral load (Ct < 25) the sensitivity of the SARS-CoV-2 Rapid Antigen Test was 100 %. The sensitivity for specimens with a medium (Ct 25–<30), low (Ct 30–<35) and very low (Ct >35) viral load was calculated to 95 %, 44.8 %, and 22.2 %, respectively.

Sensitivity and specificity of the SARS-CoV-2 Rapid Antigen Test were published in the manufacturer's recommendations to be 96.5 % and 99.7 %. These values differ markedly from the values determined by us. This difference may be due to the selection of the specimens. Our sample collection contained clinical specimens only and the SARS-CoV-2 RNA positive subpopulation was characterized by a wide range of Ct-values with medium and low Ct-values dominating. This allowed the calculation of sensitivity and specificity values with higher relevance for clinical practice. The Ct-dependent evaluation shows a very good sensitivity for highly and moderately SARS-CoV-2 positive samples (Ct < 30). In contrast, the sensitivity of the assay with specimens containing only a limited viral load is low. The overall sensitivity and the specificity of the SARS-CoV-2 Rapid Antigen Test are comparable to the recently published SARS-CoV-2 antigen assay of SD Biosensor (Cerutti et al., 2020). Thus, the SARS-CoV-2 Rapid Antigen Test may have a limited suitability for the determination of the SARS-CoV-2 infection status of patients. COVID-19 infection would not be detected in patients in the early or late phase of the infection typically associated with a low viral load. However, differentiation between contagious and non-contagious individuals may be possible with this assay. Samples with Ct-values >30 usually do not allow culturing of the virus indicating low infectivity. Such individuals may be regarded non-contagious despite carrying low virus loads. This differentiation of individuals may be of particular importance for the decision on access to susceptible individuals e.g. in nursing homes or at many other circumstances.

The significance of this assay's results is, as the significance of any other assay aiming at determination of the SARS-CoV-2 infection status, severely affected by the method of sample collection. The viral load of a specimen does not necessarily reflect the viral load in the patient's respiratory tract. Meaningful results can only be obtained with an efficient sample collection method. Otherwise the viral load in the patient may be underestimated.

Taken together, we conclude that sensitivity and specificity of the antigen assay is inferior to the PCR assay. However, the antigen assay may be a quick and easy-to-perform approach to allow differentiation of individuals contagious for SARS-CoV-2 from non- or less contagious individuals.

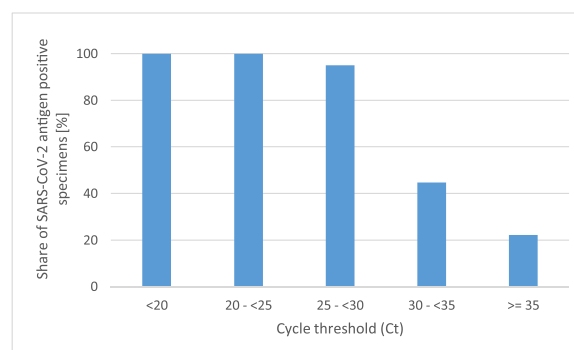


Fig. 1. Sensitivity of the SARS-CoV-2 Rapid Antigen Test in respect to the viral load of clinical specimens.

CRediT authorship contribution statement

Alexander Krüttgen: Investigation, Data curation, Writing. **Christian G. Cornelissen:** Resources, Writing. **Michael Dreher:** Resources, Writing. **Mathias W. Hornef:** Writing - review & editing. **Matthias Imöhl:** Resources, Writing - review & editing. **Michael Kleines:** Conceptualization, Data curation, Writing.

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.

References

- Cerutti, F., Burdino, E., Milia, M.G., Allice, T., Gregori, G., Bruzzzone, B., Ghisetti, V., 2020. Urgent need of rapid tests for SARS CoV-2 antigen detection: evaluation of the SD-Biosensor antigen test for SARS-CoV-2. *J. Clin. Virol.* <https://doi.org/10.1016/j.jcv.2020.104654>.
- Eis-Hübinger, A.M., Hönemann, M., Wenzel, J.J., Berger, A., Widera, M., Schmidt, B., Aldabbagh, S., Marx, B., Streeck, H., Ciesek, S., Liebert, U.G., Huzly, D., Hengel, H., Panning, M., 2020. Ad hoc laboratory-based surveillance of SARS-CoV-2 by real-time RT-PCR using minipools of RNA prepared from routine respiratory samples. *J. Clin. Virol.* 127, 104381.
- Krüttgen, A., Cornelissen, C.G., Dreher, M., Hornef, M., Imöhl, M., Kleines, M., 2020a. Comparison of four new commercial serologic assays for determination of SARS-CoV-2 IgG. *J. Clin. Virol.* 128, 104394.
- Krüttgen, A., Cornelissen, C.G., Dreher, M., Hornef, M., Imöhl, M., Kleines, M., 2020b. Determination of SARS-CoV-2 antibodies with assays from Diasorin, Roche and IDvet. *J. Virol. Methods.* <https://doi.org/10.1016/j.jviromet.2020.113978>.
- van Kasteren, P., van der Veer, B., van den Brink, S., Wijsman, L., de Jonge, J., van den Brandt, A., Molenkamp, R., Reusken, C., Meijera, A., 2020. Comparison of seven commercial RT-PCR diagnostic kits for COVID-19. *J. Virol. Methods.* <https://doi.org/10.1016/j.jcv.2020.104412>.
- Zhou, P., Yang, X.L., Wang, X.G., Hu, B., Zhang, L., Zhang, W., Si, H.R., Zhu, Y., Li, B., Huang, C.L., Chen, H.D., Chen, J., Luo, Y., Guo, H., Jiang, R.D., Liu, M.Q., Chen, Y., Shen, X.R., Wang, X., Zheng, X.S., Zhao, K., Chen, Q.J., Deng, F., Liu, L.L., Yan, B., Zhan, F.X., Wang, Y.Y., Xiao, G.F., Shi, Z.L., 2020. A pneumonia outbreak associated with a new coronavirus of probable bat origin. *Nature* 579, 270–273.
- Zhu, N., Zhang, D., Wang, W., Li, X., Yang, B., Song, J., Zhao, X., Huang, B., Shi, W., Lu, R., Niu, P., Zhan, F., Ma, X., Wang, D., Xu, W., Wu, G., Gao, G.F., Tan, W., 2020. A novel coronavirus from patients with pneumonia in China, 2019. *N. Engl. J. Med.* 382, 727–733.