Assay Validation

Product name	**SARS-CoV-2 Antibody Test (Lateral Flow Method)**
Manufacturer	SUSTech Technology Shenzhen, China)
Distributor	TAmiRNA GmbH
LOT Nr.	20200428

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Summary

Sensitivity 98.4%
Specificity 100%
Detection limit approx. 10 times higher than ELISA

Methode

The SARS-CoV-2 Antibody Test from TAmiRNA is an immunochromatographic assay for rapid, qualitative detection of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) IgM & IgG antibody in human whole blood, serum or plasma sample.

Samples

312	Plasma samples of blood donors from the previous year (2019)
141	Plasma samples from SARS-CoV-2 PCR-positive patients
48	24 blood samples and 24 plasma samples obtained therefrom

Sensitivity

For the validation of the antibody sensitivity, samples from 141 persons were used, which had previously been tested positive for SARS-CoV-2 using PCR.

In order to better characterize the sensitivity of the TAmiRNA Antibody Test, one of these samples was also tested in five 10-fold dilution levels.

Specificity

For the specificity calculation, 312 plasma samples from blood donors from 2019 – assuming that they have not yet been able to generate antibodies (IgM, IgA or IgG) against the novel coronavirus SARS-CoV-2 – were defined as negative standards and used for the calculation.

Comparison whole blood v. plasma

A parallel test with whole blood and plasma was also carried out on 24 people. The test was carried out with whole blood immediately after taking the blood and then with plasma obtained by centrifugation.

Results

Sensitivity

141 samples from persons with PCR confirmed SARS-CoV-2 infections were used to determine the diagnostic sensitivity. The period between PCR testing and blood sampling was also taken into account.

Of the 141 PCR confirmed blood samples, 132 were recognized as positive with the TAmiRNA Antibody Test. The sensitivity for the detection of an existing or previous SARS-CoV-2 infection is therefore 93,6%. After the exclusion of 19 blood samples in which the blood sample were taken 7 days or less after positive PCR, the sensitivity increases to 95%. The sensitivity is highest from the 22nd day after the diagnostic PCR (Table 1)

Table 1. Sensitivity of TAmiRNA SARS-CoV-2 Antibody Tests (n=141)

days post-PCR (n)	positive	negative	total	Sensitivity
0-7	15	4	19	78.9%
8-14	22	3	25	88%
15-21	43	2	45	95.6%
22-28	10	0	10	100%
>28	42	0	42	100%
	132	9	141	93.6%

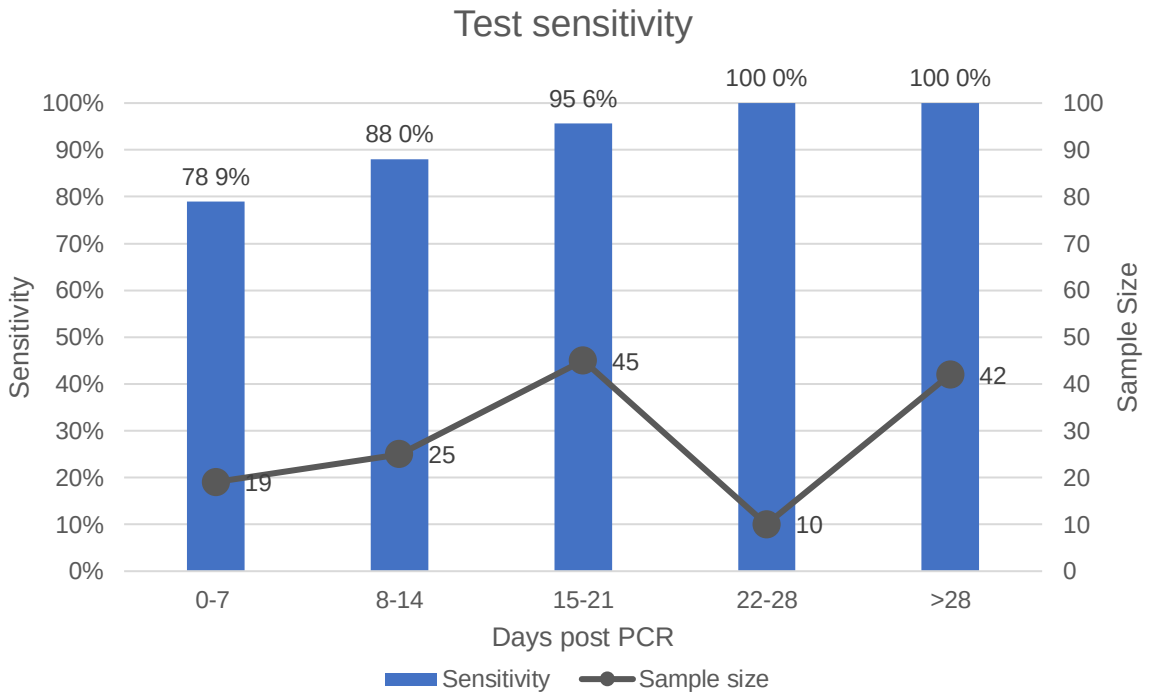

Fig 1.: Sensitivity of the TAmiRNA Antibody Test depending on the stage of disease.

The performance of the SARS-CoV-2 Antibody Test was also examined using the results of other serological procedures. Samples were evaluated as positive if they were tested positive in addition to an already existing positive PCR result in at least one serological procedure.

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Two serological test kits were used as a reference:

1. Euroimmun, anti-SARS-CoV-2 -IgG ELISA (anti-S1 proof, Euroimmun, Lübeck, Germany)
2. Euroimmun, anti-SARS-CoV-2 -IgA ELISA (anti-S1 proof, Euroimmun, Lübeck, Germany)
3. Abbott, SARS-CoV-2 IgG immunoassay (anti-N proof, Abbott, Illinois, USA)

The TAmiRNA Antibody Test identified 126 of 128 samples that were positive in one of the two serological methods (Abbott, Euroimmun) (Table 2). Using this procedure, the test shows a **sensitivity of 98.4%** for the detection of antibodies against SARS-CoV-2 and a congruence of 94.3%.

Table 2. Comparison between TAmiRNA SARS-CoV-2 Antibody Test and ELISA in PCR positive samples

		Standard	
		Positive*	Negative**
TAmiRNA	Positive	126	6
	Negative	2	7

*Abbott and/or Euroimmun IgG positive (n=128)
**Abbott and Euroimmun IgG negative (n=13)

When testing with dilution series, the SARS-CoV-2 Antibody Test showed a detection limit of 1:100. The limit of the Euroimmun ELISA is 1:1000 (Fig.2)

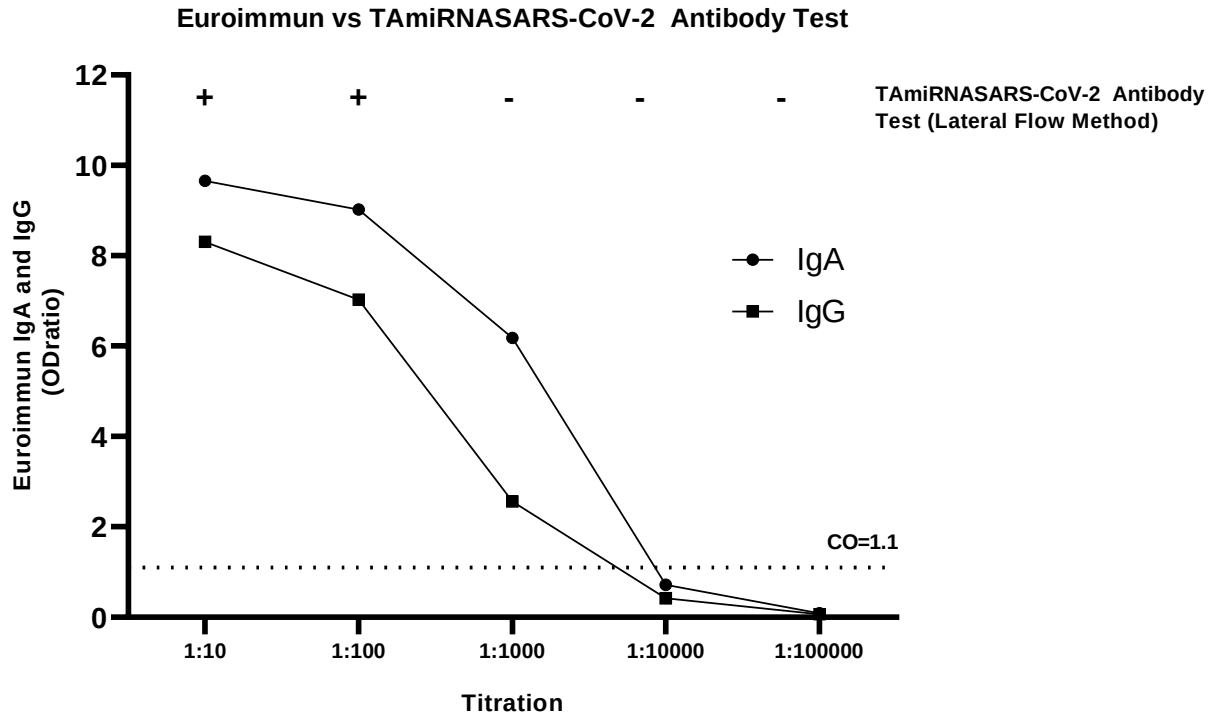

Fig 2.: Sensitivity analysis using dilution series

Specificity

To determine the specificity, 312 blood samples from blood donors from the previous year, which usually should be negative, were tested. All 312 blood donors were recognized negative with the SARS-CoV-2 Antibody Test, resulting in a specificity of 100%

Comparison whole blood vs. plasma

When comparing whole blood vs. plasma, the parallel tests gave congruent results in 23 out of 24 tests, the **agreement is therefore 95.8%.** One sample showed a negative result with whole blood a borderline result with plasma (Table 3).

Table 3. Übersicht whole blood vs. plasma

	whole blood +	whole blood -
plasma +	5	1
plasma -	0	18

Remarks

Borderline results were rated positive in this analysis.

Conclusion

A sensitivity of 98.4% was measured in the 128 PCR and antibody positive samples. The specificity in the 312 samples from the previous year was 100%. In a dilution series, the detection limit of the Euroimmun anti-S1 IgG/IgA ELISA was 10 times lower than that of the TAmiRNA Antibody Test (last detectable dilution step Euroimmun 1:1000 vs. TAmiRNA Antibody Test 1:100).

Due to the testing on more than 300 blood donors from the previous year and the high specificity determined from it, the TAmiRNA Antibody Test seems to be reliable for use on seroprevalence studies to analyze populations with low seroprevalence (<5%) and is suitable for individual immunity tests.