Comparison of the Biozek lateral-flow test against the Vircell IgG Elisa

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Introduction

The SARS-CoV-2 virus, emerged in China in the province of Wuhan, rolled over the world in just a few month enabling a pandemic. And it is, as it seems, a very unpredictable virus. The general method for detecting an active infection is the PCR. After the first onset of the illness, it is possible to detect antibodies to confirm an ongoing infection.

Also, a quick method for detecting antibodies to detect whether a patient was infected with the coronavirus in the past is useful. The Biozek lateral-flow test can detect IgM and IgG antibodies simultaneously within 15 minutes out of capillary blood, venous blood or serum. In practice the IgM and IgG develop in time after infection. At the onset of the disease, IgM antibodies develop as the first immune response. Recent studies showed IgG antibodies development also depends how severe the patients had the virus infection. In a recent study, patients with confirmed Covid-19 infection failed in the RT-PCR. These false negative results were possible because of inadequate sample collection or inappropriate timing of sample collection. The sensitivity of the nasal swab was 63% and from pharyngeal swab 32%. The specificity of the RT-PCR test is >99%.(9)

In this evaluation project, we use the Vircell IgG Elisa as the standard and not PCR. The Vircell ELISA was validated against the PCR. (Vircell **G1032:** Indirect immunoenzyme assay to test IgG antibodies against SARS-CoV-2 in human serum/plasma) The serum panel used is from patients having SARS-CoV-2 like complaints approximately 14-28 days prior to blood sampling. The decision was made to use known serum samples to avoid false negatives due to the fact not all patients develop antibodies in spite of a positive SARS-CoV-2 PCR (10,11) and there is no gold standard for SARS-CoV-2 antibody testing.

Due to the low amount of IgM positive samples, this antibody is not completely evaluated yet.

Principle of the Biozek lateral-flow test.

The 2019-nCoV IgG/IgM Rapid Test Cassette (Whole Blood/Serum/Plasma) is a qualitative membrane-based immunoassay for the detection of IgG and IgM antibodies to 2019-nCoV in whole blood, serum or plasma specimen. This test consists of two components, an IgG component and an IgM component. In the IgG component, anti-human IgG is coated in IgG test line region. During testing, the specimen reacts with 2019-nCoV antigen-coated particles in the test cassette. The mixture then migrates upward on the membrane chromatographically by capillary action and reacts with the anti-human IgG in IgG test line region, if the specimen contains IgG antibodies to 2019-nCoV. A coloured line will appear in IgG test line region as a result of this. Similarly, anti-human IgM is coated in IgM test line region and if specimen contains IgM antibodies to 2019-nCoV, the conjugate-specimen complex reacts with anti-human IgM. A coloured line appears in IgM test line region as a result.
Therefore, if the specimen contains 2019-nCoV IgG antibodies, a coloured line will appear in IgG test line region. If the specimen contains 2019-nCoV IgM antibodies, a coloured line will

appear in IgM test line region. If the specimen does not contain 2019-nCoV antibodies, no coloured line will appear in either of the test line regions, indicating a negative result. To serve as a procedural control, a coloured line will always appear in the control line region, indicating that the proper volume of specimen has been added and membrane wicking has occurred. The Biozek lateral flowtests are purchased from Clindia B.V. Netherlands.

Principle of the Vircell Elisa test.

The ELISA method is based upon the reaction of antibodies in the sample tested with the antigen adsorbed on the polystyrene surface. All unbound immunoglobulins are washed off. An enzyme-labelled anti-human globulin binds the antigen-antibody complex in a second step. After a new washing step, bound conjugate is developed with the aid of a substrate solution (TMB) to render a blue coloured soluble product which turns into yellow after adding the acid stopping solution. The Vircell Elisa test is purchased from Selinion B.V. Netherlands.

Vircell Elisa test: sensitivity / specificity against PCR. The sensitivity of the Vircell IgG ELISA, based on the data from the largest participating lab, is 98% from 14 days after complaints started (n = 53). This picture is also partly reflected in the RIVM report itself, at> 14 days (100%, n = 15). Within infection serology, 14 days for an IgG determination is a common period, and also more towards the 4 weeks that is increasingly apparent in the international consultations of the WHO working group.
The subgroup of participating peripheral hospitals, and who also subjected the samples to the prescribed heat inactivation, establish a specificity of 95 - 97% (n = 102). This specificity is therefore more representative and is generally regarded as an acceptable and accepted specificity within infection serology.

Introduction evaluation method:

Capillary blood samples are gathered from about 1000 patients. All samples are from patients which had a mild to severe illness, but were not admitted into a hospital.
212 samples could be used for performing the Vircell Elisa IgG/IgM and Biozek lateral IgG/IgM flowtest due to the fact that not all samples were tested for both methods.

Analytical results.

Total positive elisa tests	86
Total positive lat.flow tests	87
Total negative elisa tests	126
Total neg.lat flow tests	125

Evaluation of results:

From 212 samples we measured 126 negative Elisa tests and 125 negative flow tests.
According to these findings, the IgG specificity is calculated as 99,2%.
Furthermore, 86 samples tested positive for Elisa IgG and 87 lateral flow tests tested positive resulting in a sensitivity of 98,8%

Preliminary result: Due to the low amount of samples the IgM sensitivity and specificity could not be calculated. Figures of IgM elisa and IgM lateral-flowtest on 39 samples resulted in a IgM specificity of 100%.

Conclusion:

The antibody test by Biozek is comparable to the Vircell IgG Elisa and can be used to test for IgG Covid-19 antibodies.

Comparison with only SARS-CoVid-2 PCR positive patients is not applicable due to the fact not all patients develop antibodies. This would result in to many false negative antibody results. We think, from a scientific point of view, that using the antibody test for comparison is more useful.

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Literature:

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