



Anti-SARS-CoV-2 ELISA (IgG)



- Specific detection of IgG against SARS-CoV-2 using the S1 domain of the spike protein including the immunologically relevant receptor binding domain (RBD)
- Excellent performance and good correlation with neutralisation tests confirmed in external studies
- Programmes for vaccine development worldwide are based on the spike protein as the antigen
- Validated for dried capillary blood as alternative sample material; fully automatable

Technical data

Antigen	Domain S1 of the SARS-CoV-2 spike protein expressed recombinantly in human cells
Calibration	Semiquantitative; calculation of a ratio from the extinction of the sample and that of the calibrator
Result interpretation	EUROIMMUN recommends interpreting results as follows: Ratio < 0.8: negative Ratio ≥ 0.8 to < 1.1: borderline Ratio ≥ 1.1: positive
Sample dilution	Serum or plasma, 1 : 101 in sample buffer, or 4.76 mm punched out membrane piece with dried capillary blood in 250 µl sample buffer
Reagents	Ready for use, with the exception of the wash buffer (10x); colour-coded solutions, in most cases exchangeable with those in other EUROIMMUN ELISA kits
Test procedure	60 min (37°C) / 30 min (37°C) / 30 min (RT) (sample/conjugate/substrate incubations), fully automatable
Measurement	450 nm, reference wavelength between 620 nm and 650 nm
Test kit format	96 break-off wells; kit includes all necessary reagents
Stability	6 months
Order number	EI 2606-9601 G; EI 2606-9620 G (designed especially for processing on the EUROLabWorkstation ELISA)

Clinical significance

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2, previously called 2019-nCoV) belongs to the family of coronaviruses and, like SARS-CoV, is classified in the genus *Betacoronavirus*. The new coronavirus originated in China, in the city of Wuhan, Hubei province. At the end of 2019 it caused an infection wave that has spread rapidly over the country and worldwide. Just a few days after the first report about patients with pneumonia of unclear origin, SARS-CoV-2 was identified as the causative pathogen.

SARS-CoV-2 is mainly transmitted via aerosols during coughing or sneezing or at close contact with an infected person. Health care personnel and family members are especially at risk. The zoonotic reservoir of the virus appears to be bats. The incubation time of SARS-CoV-2 is three to seven, maximally 14 days. The symptoms of SARS-CoV-2 infection are fever, coughing, breathing difficulties and fatigue. In most patients the infection manifests with symptoms of a mild febrile illness with irregular lung infiltrates. Some patients, especially elderly or chronically ill patients, develop acute respiratory distress syndrome (ARDS). The fatality rate is between 0.6 and 7.2%, depending on the country. In February 2020, the disease caused by SARS-CoV-2 was named COVID-19 by the WHO.

Suitable methods for the diagnosis of SARS-CoV-2 infections are the detection of viral RNA by reverse transcriptase polymerase chain reaction (RT-PCR) or of virus protein by means of ELISA primarily in sample material from the upper (nasopharyngeal or oropharyngeal swab) or lower respiratory tract (bronchoalveolar lavage fluid, tracheal secretion, sputum, etc.). The determination of antibodies enables confirmation of SARS-CoV-2 infection in patients with typical symptoms and in suspected cases. It also contributes to monitoring and outbreak control. For significant serological results, two patient samples should be investigated, one from the acute phase (week 1 of the illness) and one from the convalescent phase (3 to 4 weeks later).

Cross reactions with antibodies within the genus *Betacoronavirus* have been described. Currently, there is no medication or vaccine available against infection with this new virus.



Diagnostic sensitivity (prevalence)

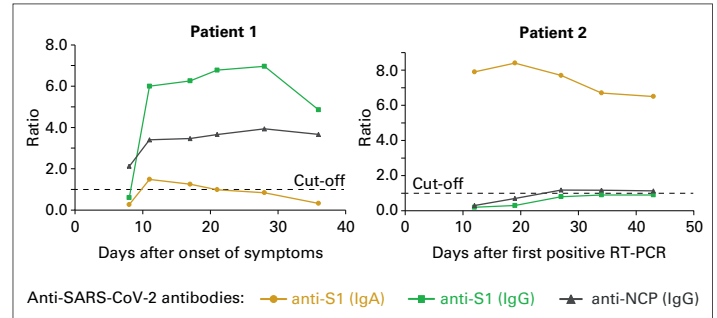
The sensitivity was determined by investigating 166 samples from 152 European patients using the Anti-SARS-CoV-2 ELISA (IgG). In these patients, infections with SARS-CoV-2 had been confirmed by RT-PCR¹ based on a sample taken at the early phase of infection. The serological examination was based on samples collected during the further course of the infection. In samples taken until day 10 (time point after symptom onset or positive direct detection), the Anti-SARS-CoV-2 ELISA (IgG) showed a sensitivity of 43.7%. The sensitivity of the Anti-SARS-CoV-2 ELISA (IgG) in samples collected after day 10 was 94.4%.

The graph shows two individual immune responses in COVID-19 patients measured with the EUROIMMUN Anti-SARS-CoV-2 ELISAs (IgA, IgG) based on the recombinantly expressed spike protein domain S1 and the EUROIMMUN Anti-SARS-CoV-2 NCP ELISA (IgG) using the modified nucleocapsid protein (NCP) as the antigen. **Patient 1 (47 years old):** The anti-SARS-CoV-2 NCP IgG antibody level was elevated as early as day 8 after the onset of symptoms. Anti-SARS-CoV-2 S1 antibodies (IgA and IgG) were not yet detectable. The follow-up sample taken on day 11 after symptom onset showed an increase in the antibody levels for both Ig classes. **Patient 2 (58 years old):** The Anti-SARS-CoV-2 S1 IgA antibody level was already highly elevated 12 days after the first positive RT-PCR. In contrast, the levels of anti-SARS-CoV-2 S1 and anti-SARS-CoV-2 NCP IgG antibodies increased only slowly until day 43 after positive RT-PCR.

The time course of antibody secretion and the antibody activity at specific time points can vary significantly. In most patients, antibodies are detectable after day 10 after symptom onset or positive direct detection. In individual cases, however, a significantly delayed synthesis of IgG (> 4 weeks after symptom onset or positive detection) has been reported.

Days after symptom onset or positive direct detection	EUROIMMUN Anti-SARS-CoV-2 ELISA (IgG)		
	Positive	Negative	Sensitivity (prevalence)*
≤ 10	38	49	43.7 %
> 10	68	4	94.4 %

* Borderline results were not included (n=7). The sensitivity depends on the prevalence of specific IgG antibodies in persons with COVID-19 infection.



Specificity

The specificity of the Anti-SARS-CoV-2 ELISA (IgG) was determined by investigating 222 patient sera that were positive, for instance, for antibodies against other human pathogenic coronaviruses, other pathogens or for rheumatoid factors. Additionally, 1122 samples from blood donors, children and pregnant women obtained before the occurrence of SARS-CoV-2 (before January 2020) were analysed. Cross reactions with other human pathogenic coronaviruses were not observed. The specificity of the Anti-SARS-CoV-2 ELISA (IgG) thus amounted to 99,6%.

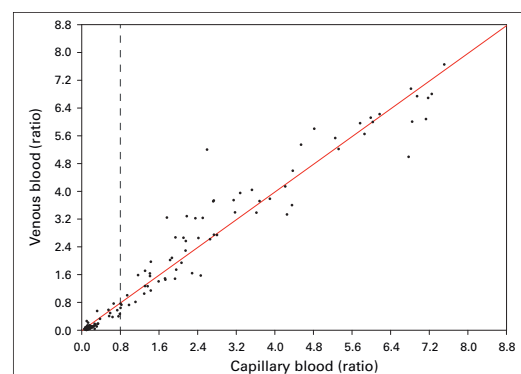
Panel	EUROIMMUN Anti-SARS-CoV-2 ELISA (IgG)	
	n	Specificity
Blood donors	849	99.5 %
Pregnant women	199	99.5 %
Children	74	100 %
Elderly patients	97	100 %
Infections with other human pathogenic coronaviruses	23	100 %
Influenza (freshly vaccinated, incl. courses)	40	100 %
Acute EBV infection & heterophilic antibodies	22	100 %
Rheumatoid factors	40	100 %
Total	1344	99.6 %

* Borderline results (n=7) were not included.

Method comparison

The correlation between extracts of dried blood spots (DBS) from capillary blood and serum from venous blood was determined by analysing 215 patient samples collected in Europe, using the Anti-SARS-CoV-2 ELISA (IgG). For each patient, one capillary blood sample and one venous blood sample were available.

The agreement between the results of the dried capillary blood spots and the venous blood samples was 100%. Borderline samples were excluded from the calculation.



Literature

1. Corman VM, et al. **Detection of 2019 novel coronavirus (2019-nCoV) by real-time RT-PCR.** Euro Surveill 25(3): pii=2000045 (2020-01-23).
2. WHO: **Laboratory testing for 2019 novel coronavirus (2019-nCoV) in suspected human cases.** Interim guidance (2020-03-19).
3. Matushek S, et al. **Evaluation of the EUROIMMUN Anti-SARS-CoV-2 ELISA Assay for detection of IgA and IgG antibodies.** bioRxiv preprint (2020-05-13).