*recom*Line SARS-CoV-2 IgG [Avidität]

MIKROGEN DIAGNOSTIK

IVD

Instructions for use (English)

1 Intended purpose

The *recom*Line SARS-CoV-2 IgG [Avidität] is a qualitative test to detect and determine the avidity of IgG antibodies against SARS-CoV-2 (**s**evere **a**cute **r**espiratory **s**yndrome **co**rona**v**irus-2) in human serum or plasma. The test is used as an aid to evaluate the adaptive humoral immune response that is induced by infection and/or vaccination.
To determine the avidity of the IgG antibodies, the avidity reagent can be requested separately.
The *recom*Line SARS-CoV-2 IgG [Avidität] can be performed manually or as an automated test by trained personnel in a suitable laboratory.

2 Field of application

SARS-CoV-2 belongs to the *Coronaviridae* family and is the causative pathogen for the COVID-19 pandemic. SARS coronaviruses spread primarily via droplets and aerosols in exhaled air to transmit from person to person. Symptoms range from fever, cough and dyspnoea to pneumonia and acute respiratory distress syndrome and ultimately death in persons with comorbidities. There are currently several approved vaccines against SARS-CoV-2.
The *recom*Line SARS-CoV-2 IgG [Avidität] is a line immunoassay. The separate line-up of individual antigens means that, unlike ELISA, specific antibodies against the individual antigens of the various coronaviruses can be identified in the same test. In the test, the following recombinant antigens are used for SARS-CoV-2: nucleocapsid (NP), RBD (receptor-binding domain of the spike protein) and S1 (S1 subunit of the spike protein). Antibodies against seasonal human coronaviruses (HCoV: 229E, NL63, OC43, HKU1) are also detected by the corresponding nucleocapsid antigens (NP).
IgG antibodies against SARS-CoV-2 can usually be detected at the latest 3 weeks after infection. The corresponding immune response is highly individual and decreasing titers can be observed after just a few months. Antigen-specific IgG antibodies can also be detected after vaccination whereby the duration of the detection period is still the subject of ongoing studies.
The avidity maturation of IgG after infection with SARS-CoV-2 is generally incomplete and very clearly differs in this regard from avidity maturation after infection with other viruses. Repeated vaccination can, however, achieve high avidity of the IgG against the spike protein and its receptor binding domain (RBD).
The test is used as an aid to evaluate the adaptive humoral immune response. It can contribute to our understanding of viral epidemiology and immunity after infection and/or vaccination and support the diagnosis of COVID-19. For acute infections, direct detection, e.g. using RT-PCR, is the standard, however.
The *recom*Line SARS-CoV-2 IgG [Avidität] can be used as a confirmatory test to clarify unclear screening results and as a screening test.

3 Test principle

Highly purified recombinant antigens (NP, RBD and S1 from SARS-CoV-2 as well as NP from 229E, NL63, OC43, HKU1) are fixed to nitrocellulose membrane test strips.

1. The test strips are incubated with the diluted serum or plasma sample with specific antibodies binding to the pathogen antigens on the test strips.
2. Unbound antibodies are then washed away.
3. The strips are incubated in a second step with anti-human immunoglobulin antibodies (IgG) that are coupled to horseradish peroxidase.
4. Unbound conjugate antibodies are then washed away.
5. Specific bound antibodies are detected in a colour reaction catalysed by the peroxidase. If an antigen-antibody reaction has taken place, a dark band appears on the strip at the corresponding location.

Control bands are located at the top end of the test strip:

a) The reaction control below the strip number for which every serum/plasma sample must show a reaction.
b) The conjugate control (IgG) is used to check the conjugate and strip type used (Ig class specific).
c) 'Cut-off control': The intensity of this band enables the reactivity of the individual antigen bands to be evaluated (see section 9.2 Analysis).

4 Reagents

4.1 Package contents

The reagents in one pack are sufficient for 20 assays.
Each set of reagents contains:

WASHBUF A 10 X	**100 ml** wash buffer A (**10× concentrate**) Contains phosphate buffer, NaCl, KCl, detergent, preservative: MIT (0.1%) and Oxypyrion (0.2%)
SUBS TMB	**40 ml** chromogenic substrate tetramethylbenzidine (**TMB, ready-to-use**)
MILKPOW	**5 g** skimmed milk powder
INSTRU	**1** instructions for use
EVALFORM	**1** evaluation sheet
TESTSTR	**2** tubes each with 10 consecutively numbered test strips
CONJ IgG	**500 µl** anti-human IgG conjugate **(100× concentrate, green cap)** From rabbit, contains NaN_3 (<0.1%), MIT (<0.1%) and chloroacetamide (<0.1%)

Avidity determination
To determine the avidity of SARS-CoV-2 IgG antibodies, the avidity reagent with the corresponding instructions for use can be supplied upon request as a supplement.

AVIDI Article no. 11010	**1** avidity reagent (solid 25 g) for 60 ml ready-to-use solution

4.2 Additionally required reagents, materials and equipment

- Incubation trays (can be purchased from MIKROGEN as required)
- Deionised water (high quality)
- Plastic forceps
- Horizontal shaker
- Vortex mixer or other rotatory device
- Vacuum pump or similar device
- Measuring cylinders, 50 ml and 1000 ml
- Micropipettes with single-use tips, 20 µl and 1000 µl
- 10 ml pipette or dispenser
- Timer
- Absorbent paper towels
- Single-use protective gloves
- Waste container for biohazardous substances

5 Shelf life and handling

- Store reagents between +2°C and +8°C before and after use; **do not freeze**.
- Before starting the test, allow all reagents to sit at room temperature (+18°C to +25°C) for at least 30 minutes. The test is performed at room temperature.
- Wash buffer, milk powder, dilution buffer, conjugates and TMB can be exchanged between the different *recom*Line and/or *recom*Blot test systems if these components carry the same symbol. Note the shelf life of these components when doing so.
- Before use, mix the concentrated reagents and patient sera thoroughly. Avoid foam formation.
- Only open the tubes with the test strips just before use to prevent water condensation. Strips that are not needed must be left in the tube and are continued to be stored at +2°C to +8°C (reseal tube firmly, test strips must not be moist before the start of the test!).
- The strips are identified with a consecutive number and a test abbreviation.
- The packages have an expiry date; after this date has passed no further guarantee of quality can be given.
- Keep the kit components away from direct sunlight throughout the test procedure. The substrate solution (TMB) is light sensitive.
- The test may only be performed by trained, authorised and qualified personnel.
- Substantial changes made by the user to the product or the directions for use may compromise the intended purpose of the test specified by MIKROGEN.
- Cross-contamination of the patient samples or conjugates in the kit can lead to false test results. Add patient samples, test strips and conjugate solution carefully. Make sure that any incubated liquids are not carried over to other wells. Carefully remove liquids.

- The strips must be completely wetted and submersed throughout the entire procedure.
- Automation is possible. Further details are available from MIKROGEN.

6 Warnings and safety precautions

- Only use for *in-vitro* diagnostics.
- All blood products must be treated as potentially infectious.
- The test strips were manufactured with inactivated whole-cell lysates and/or recombinant bacterial, viral or parasitic antigens.
- After adding patient or control material, the strips must be considered to be potentially infectious and handled appropriately as such.
- Suitable single-use gloves must be worn throughout the entire test procedure.
- The reagents contain the antimicrobial agents and preservatives sodium azide (NaN_3), MIT (methylisothiazolinone), Oxypyrion and chloroacetamide. Avoid contact with the skin or mucous membranes. Sodium azide (NaN_3) can form explosive azides if it comes into contact with heavy metals such as copper and lead.
- All aspirated liquids must be collected. All collection reservoirs must contain suitable disinfectants for inactivation of human pathogens or be autoclaved. All reagents and materials that come into contact with potentially infectious samples must be treated with suitable disinfectants or be disposed of according to laboratory guidelines. The concentrations and incubation times specified by the manufacturer must be followed.
- Only use incubation trays once.
- Handle strips carefully with a pair of plastic forceps.
- Do not replace or mix the reagents with reagents of other manufacturers.
- Read through and carefully follow all instructions before performing the test. Deviations from the test protocol described in the Instructions for Use can lead to false results.

7 Sampling and preparation

7.1 Sample material

The sample material can be either serum or plasma (EDTA, citrate, heparin, CPD) and must be separated as soon as possible after collection to avoid haemolysis. Microbial contamination of the sample must absolutely be avoided. Non-soluble substances must be removed from the sample prior to incubation. Use of icteric, haemolytic, lipaemic or cloudy samples is not recommended.

Caution!

If the tests are not carried out immediately, the sample material can be stored for up to 2 weeks between +2°C and +8°C. It is possible to store the samples for longer periods at −20°C or lower. Repeated freezing and thawing of the samples is not recommended due to the risk of false results. More than 3 freezing and thawing cycles should be avoided.

7.2 Preparation of solutions

7.2.1 *Preparation of the ready-to-use wash buffer A*

This buffer is required for the serum and conjugate dilution and the wash steps.

Before the dilution, the volume of wash buffer A must be determined for the corresponding number of tests to be carried out.

The skimmed milk powder is first pre-dissolved in wash buffer A concentrate and then this mixture is made up to the final volume with deionised water (dilution 1 + 9). The required quantity for a defined number of test strips is calculated using the following formulae (device-specific dead volumes are not taken into account):

Reagent	Formula	Example: 5 strips
Skimmed milk powder [g]	= strip number × 0.1	0.5 g
Wash buffer A concentrate [ml]	= strip number × 2	10 ml
Deionised water [ml]	= strip number × 18	90 ml
Ready-to-use wash buffer A [ml]	= strip number × 20	100 ml

Ready-to-use wash buffer A can be stored at +2°C to +8°C for four weeks. The ready-to-use wash buffer A has no odour and is slightly turbid.

7.2.2 *Preparation of conjugate solutions*

The conjugate solution must be prepared shortly before use; the ready-to-use conjugate solution must not be stored.

One part of the conjugate concentrate is diluted with 100 parts of the ready-to-use wash buffer A (1 + 100).

The required quantity for a defined number of test strips is calculated using the following formulae:

Reagent	Formula	Example: 5 strips
Conjugate concentration [µl]	= strip number × 20	100 µl
Ready-to-use wash buffer A [ml]	= strip number × 2	10 ml

The conjugate quantities are calculated without dead volumes. Depending on the processing (manually or with a device), please prepare additional conjugate solution for 1 to 3 strips.

8 Test procedure

No.	Implementation	Note
1	Before starting the test, allow all reagents to sit between 18°C and 25°C (room temperature) for at least 30 minutes.	The test is performed at room temperature.
2	Prepare the test strips Place strips in **2 ml** ready-to-use **wash buffer A** (see 7.2.1).	Do not handle the strips with bare hands – use forceps. The strip number faces upwards. For each strip one well in an incubation tray (see 4.2) is required. The strips must be completed submersed.
3 a) b)	Sample incubation **20 µl** of an undiluted sample (human serum or plasma) are added by pipette to the test strip for each incubation mixture (dilution 1 + 100). Incubate for **1 hour** with gentle shaking.	Pipette the sample onto one end of the submersed strip in wash buffer A and mix as quickly as possible by gently shaking the incubation tray. Cover the incubation tray with the plastic lid and place on the shaker.
4 a) b) c)	Wash Carefully remove the plastic lid from the incubation tray. Carefully aspirate the serum dilution from the individual wells. Pipette **2 ml** ready-to-use **wash buffer A** (see 7.2.1) into each well, wash for 5 minutes with gentle shaking and then aspirate off the wash buffer A.	Carry out wash steps 8.4a–8.4c a total of three times. Avoid cross-contamination. With automated processing, follow the directions of the device manufacturer for this step.
5	Incubation with conjugate Add **2 ml** ready-to-use **conjugate solution** (see 7.2.2) and incubate for **45 minutes** with gentle shaking.	Cover the incubation tray with the plastic lid and place on the shaker.
6	Wash See section 8.4	Carry out wash steps a total of three times (see 8.4a–8.4c).
7	Substrate reaction Add **1.5 ml substrate solution** and incubate for **8 minutes** with gentle shaking.	
8	Stopping the reaction Remove the substrate solution. Wash at least three times **briefly** with **deionised water**.	
9	Drying the strips Dry the strips before the evaluation for **2 hours** between 2 layers of absorbent paper.	Carefully remove the strips from the water with a pair of plastic forceps. Store the strips protected from light.
Caution! **Incubation solutions must not be carried over to other wells. Avoid splashing, particularly when opening and closing the cover.**		

9 Results

Caution:

Do not use the automated interpretation without taking note of the advice on the interpretation described below.

9.1 Validation – Quality control

The test can only be analysed when the following criteria are satisfied:

1. Reaction control band (upper line) is clearly stained, dark band detectable.
2. Antibody class (second band): the IgG conjugate control band must be clearly stained.
3. Cut-off control (third band): weak but visible staining.

9.2 Analysis

The test strips can be analysed visually or with a computer using the *recom*Scan test strip analysis software. The *recom*Scan software is intended to help with test strip interpretation. Additional information and corresponding instructions for computer-aided analysis are available from MIKROGEN upon request. The following instruction refers to the visual analysis.

9.2.1 *Evaluation of the band intensity*

1. Note the date and batch and tube numbers along with the antibody class that was detected in the attached evaluation sheet.
2. Enter the sample identification numbers in the evaluation sheet.
3. Now adhere the associated test strip with a glue stick into the corresponding field in the evaluation sheet. Align the test strip with the reaction control band at the indicated marker line. Then adhere the test strip to the left of the marking line using transparent adhesive tape (do not stick over the reaction control band!). Adhering the entire test strip with glue stick or adhesive tape can lead to changes in the staining.
4. Now identify the bands for the developed test strips using the printed control strip from the evaluation sheet and enter these into the evaluation sheet. Carry out the evaluation of the intensity of the emergent bands separately for the relevant immunoglobulin classes using Table 1.

Table 1: Evaluation of the band intensity relative to the cut-off band

Colour intensity of the bands	Evaluation
No reaction	-
Very low intensity (lower than the cut-off band)	+/-
Low intensity (equivalent to the cut-off band)	+
Strong intensity (stronger than the cut-off band)	++
Very strong intensity	+++

Evaluation of the avidity: see section 9.4.

9.3 Interpretation schematic

Table 2: Test interpretation of *recom*Line SARS-CoV-2 IgG [Avidität]

Test interpretation	Antigen reactivity
SARS-CoV-2 IgG positive	One or more SARS-CoV-2-specific antigen bands (NP, RBD and/or S1) are positive, that is, they react with the same (+) or a stronger intensity than the cutoff band (regardless of the reactivity of the HCoV antigens).
SARS-CoV-2 IgG negative	All SARS-CoV-2-specific antigen bands (NP, RBD and S1) are negative, that is, they do not show any bands (−) or bands with a lower (+/−) intensity than the cutoff band (regardless of the reactivity of the HCoV antigens).

Detecting antibodies can indicate a recent or past SARS-CoV-2 infection. To detect an acute infection, direct detection of the pathogen using, e.g., RT-PCR, is considered the gold standard. Usually, after an infection IgG antibodies against all three antigens used (NP, RBD and S1) can be detected.
After vaccination, IgG antibodies can also be detected. Depending on the vaccine used, the immune response can in some cases be differentiated from an infection. Because most of the approved vaccines (e.g. Comirnaty from BioNTech/Pfizer) are directed against the spike protein, only antibodies against RBD and S1 are induced in naive persons as a result.

Along with SARS-CoV-2, reactivities against the seasonal human coronaviruses (HCoV: 229E, NL63, OC43, HKU1) are also determined. A total seroprevalence of 70%–90% can be expected (see also Table 6). The reactivities of the HCoVs determined do not allow differentiation or any conclusions to be drawn about the immune status regarding the particular coronaviruses or about cross-reactivity with SARS-CoV-2.

9.4 Avidity determination

By processing two IgG solutions in parallel, one of which is treated with the avidity reagent, it can be determined whether the IgG antibodies have low, intermediate or high avidity. Low-avidity antibodies can be washed off the binding site on the strip using an avidity reagent while high-avidity antibodies cannot be detached by the reagent.

9.4.1 *Analysis of the avidity in the recomLine SARS-CoV-2 IgG [Avidität]*

- Only carry out the avidity determination with an overall positive IgG result.
- Bands on the IgG strip that have a lower reactivity than the cut-off are not considered in the avidity determination.
- Compare the intensities of the corresponding bands on the two test strips (IgG strip and avidity strip) that were incubated with the same patient sample. Note whether the intensities have changed.
- A reduction in the intensity of the SARS-CoV-2 bands (NP, RBD and S1) of more than 60% (avidity index ≤0.4) is defined as low avidity while a reduction of between 40% and 60% is defined as intermediate (avidity index >0.4 to <0.6).
- With high avidity, the band intensity of the avidity strip reduces by less than 40% (avidity index ≥0.6) and the IgG antibodies are defined as high avidity.
- In general, the avidity maturation after SARS-CoV-2 infections is highly variable and is frequently incomplete. This contrasts with the possibility of complete avidity maturation after optimal vaccination.

9.4.2 *Significance of the avidity determination*

The following information about the avidity of SARS-CoV-2 IgG antibodies was obtained and can contribute to better evaluation of the immune response:

Avidity after infection (see also Tables 7 and 8)

- In patients with symptomatic SARS-CoV-2 infection who do not need to be hospitalised, usually IgG (NP, RBD, S1) of only low or intermediate avidity develops in the subsequent 2 months. In hospitalised patients, high-avidity IgG develops in about 12% of patients in this period.
- Unlike many other viral infections, a reliable indication of the likely time of the infection cannot be determined from a single avidity determination. Determining the avidity again at a later time can identify whether there has been a further increase in the avidity. Therefore, avidity measurement is not suitable for determining the time of infection.

Avidity after vaccination (see also Table 9)

- Usually, only low-avidity IgG against RBD and S1 is induced after the first vaccination (vector vaccines or mRNA vaccines). In 55% to 75% of those vaccinated, IgG with higher avidity is achieved after the second vaccination.
- There is generally a further increase in the avidity following the third vaccination. This is particularly important for persons who did not achieve high avidity after the second vaccination.
- To achieve high avidity, current information suggests that not only can homologous vaccination with vector vaccines or mRNA vaccines be used but heterologous vaccination is also possible, that is, vaccines can be combined.
- A prior SARS-CoV-2 infection initially leads only to low or intermediate avidity. A single subsequent vaccination can be highly effective, however, and rapidly achieve high avidity of the IgG against RBD and S1 with no change in the avidity of the IgG against NP.

10 Limits of the method, restrictions

- By determining and measuring the avidity of SARS-CoV-2-specific IgG antibodies, *recom*Line SARS-CoV-2 IgG [Avidität] enables a better evaluation of the humoral immune response after infection and/or vaccination. However, there are currently no officially recognised guidelines that permit a definition of the presence or duration of immune protection.
- Serological test results for *recom*Line SARS-CoV-2 IgG [Avidität] do not permit a direct statement abpit or prediction of the severity of a current infection. A persistent lack of avidity maturation after infection or vaccination may be an indication that protective immunity is lacking.
- Serological test results must always be viewed within the context of the clinical presentation. The therapeutic consequences of the serological finding must be related to the clinical data.
- In case of unclear serological results, repeat testing over the course of the infection is recommended.
- To detect an acute infection, direct detection of the pathogen using RT-PCR is considered the gold standard. Detection of antibodies can confirm the result of the RT-PCR test but a negative RT-PCR test result may also indicate a recent or past infection.
- A negative result does not rule out the possibility of a SARS-CoV-2 infection because, particularly in the early stage of infection, antibodies may not be present or they may not be present in detectable quantities. If a SARS-CoV-2 infection is clinically suspected and the serological results are negative, RT-PCR (e.g. with tests from the *ampli*Cube portfolio from MIKROGEN) should be carried out and/or another sample should be collected and tested after 2 weeks.

- It cannot currently be ruled out that an infection with immune escape variants of SARS-CoV-2 leads to a reduced sensitivity of the antibody detection.
- Due to the close relationship between the SARS coronaviruses (SARS-CoV and SARS-CoV-2), a cross-reaction with antibodies against SARS-CoV is possible. Cross-reactions with other human pathogenic coronaviruses (HCoV) cannot be ruled out completely but could not be determined in the evaluation of the *recom*Line SARS-CoV-2 IgG [Avidität].
- Individual cross-reactions with samples from pregnant women, patients with acute EBV infections and rheumatoid-factor-positive and lipaemic samples may occur in rare cases. See also Table 5.
- Dark test strips: Some patient samples can produce a dark generalised or patterned staining over the entire nitrocellulose strip (e.g. with sera from patients with milk protein allergies). There are a number of different factors associated with the patient serum that are responsible for this. Evaluating these strips is generally only possible with limitations. Such 'inverse' bands (white bands on a dark background) must be rated negatively. The corresponding serum should be checked using another serological method.

11 Performance characteristics

11.1 Diagnostic sensitivity

To determine the diagnostic sensitivity, 54 samples from people with RT-PCR confirmed SARS-CoV-2 infection were examined.

Table 3: Diagnostic sensitivity for *recom*Line SARS-CoV-2 IgG [Avidität]

***recom*Line SARS-CoV-2 IgG [Avidität]**	**Days after onset of symptoms**		
	Early <12 days	**Middle 12–23 days**	**Late >23 days**
Positive	6	20	26
Negative	1	1	0
Diagnostic sensitivity	**85.7%**	**95.2%**	**100%**
	96.3%		

11.2 Diagnostic specificity

To determine the diagnostic specificity, samples from German blood donors (n = 300) that were collected at different times before the start of the SARS-CoV-2 pandemic as well as potentially cross-reactive (n = 191) or interfering samples (n = 80) were examined.

Table 4: Diagnostic specificity for *recom*Line SARS-CoV-2 IgG [Avidität]

***recom*Line SARS-CoV-2 IgG [Avidität]**	**Blood donors (n = 300)**	**Potentially cross-reactive samples* (n = 191)**	**Potentially interfering samples** (n = 80)**
Positive	1	4	2
Negative	299	187	78
Diagnostic specificity	**99.7%**	**97.9%**	**97.5%**
	98.8%		

* Samples positive for seasonal coronaviruses, influenza A/B virus, RSV, adenoviruses, *Mycoplasma pn., Chlamydia pn.*, EBV, CMV, auto-antibodies as well as from pregnant women

** Lipaemic, haemolytic and icteric samples, RF-positive samples

11.3 Analytical specificity

The analytical specificity is defined as the suitability of the test to precisely determine the analytes in the presence of potential interference factors in the sample matrix or cross-reactions with potentially interfering antibodies.

a) Interference: Control studies on potentially interfering factors have shown that the test performance is not influenced by anticoagulants (sodium citrate, EDTA, heparin, CPD), haemolysis, lipaemia or bilirubinaemia of the sample.

b) Cross-reactions: Potential interference of antibodies against other organisms that can induce clinical symptoms similar to those of a SARS-CoV-2 infection (e.g. seasonal coronaviruses, influenza A/B virus, RSV, adenoviruses, *Mycoplasma pn.*, *Chlamydia pn.*) were investigated in control studies. In addition, conditions were tested that can be attributed to the development of atypical immune system activity (e.g. EBV, CMV, anti-nuclear autoantibodies, pregnancy, rheumatoid factor). The results of the tests can be seen in Table 5.

Table 5: Testing of cross-reactivities for *recom*Line SARS-CoV-2 IgG [Avidität]

Collective (n = 271)	***recom*Line SARS-CoV-2 IgG [Avidität]**
	Positive
Seasonal coronaviruses (HCoV) (n = 9)	0
Influenza A virus (n = 9)	0
Influenza B virus (n = 5)	0
Respiratory syncytial virus (RSV) (n = 10)	0
Adenoviruses (n = 6)	0
Mycoplasma pneumoniae (n = 10)	0
Chlamydia pneumoniae (n = 25)	0
Epstein-Barr virus (EBV) (n = 31)	2
Cytomegalovirus (CMV) (n = 11)	0
ANA-autoantibody-positive (n = 15)	0
Pregnant women (n = 60)	2
Rheumatoid-factor-positive (n = 50)	1
Haemolytic samples (n = 10)	0
Lipaemic samples (n = 10)	1
Icteric samples (n = 10)	0

11.4 Prevalence

To determine the prevalence of HCoV, 300 samples from German blood donors that were collected at different times before the start of the SARS-CoV-2 pandemic were examined.

Table 6: Prevalence in Germany before the SARS-CoV-2 pandemic with *recom*Line SARS-CoV-2 IgG [Avidität]

Blood donors (n = 300)	***recom*Line SARS-CoV-2 IgG [Avidität]**
Prevalence	HCoV (229E, NL63, OC43, HKU1)
	81.9%

11.5 Avidity

To determine the avidity of SARS-CoV-2 IgG antibodies, samples from people with SARS-CoV-2 infection confirmed by RT-PCR and samples from vaccinated people were examined.

Table 7: Avidity in people infected with SARS-CoV-2 after onset of symptoms

SARS-CoV-2 infected*	**Days after onset of symptoms**					
	0–50 days (n = 52)			**51–100 days (n = 32)**		
Avidity**	l	i	h	l	i	h
NP [%]	98.1	-	1.9	87.4	6.3	6.3
RBD [%]	96.4	3.6	-	71.9	21.9	6.2
S1 [%]	96.4	3.6	-	78.1	12.5	9.4

* Sera from SARS-CoV-2 RT-PCR-positive people

** Avidity: l = low, i = intermediate, h = high

Table 8: Avidity in people infected with SARS-CoV-2 by severity of disease

SARS-CoV-2 infected*	**Severity of the disease**								
	Progression without hospitalisation (n = 14)			**Progression with hospitalisation on normal ward (n = 24)**			**Progression with hospitalisation in ICU (n = 11)**		
Avidity**	l	i	h	l	i	h	l	i	h
NP [%]	100	-	-	91.6	4.2	4.2	90.9	9.1	-
RBD [%]	92.9	7.1	-	70.8	16.7	12.5	81.8	-	18.2
S1 [%]	100	-	-	83.3	12.5	4.2	72.7	9.1	18.2

* Sera from SARS-CoV-2 RT-PCR-positive people

** Avidity: l = low, i = intermediate, h = high

Table 9: Avidity in people vaccinated against SARS-CoV-2

SARS-CoV-2 vaccinated*	**Time after vaccination**								
	1st dose (n = 28)			**2nd dose (n = 40)**			**3rd dose (n = 20)**		
Avidity**	l	i	h	l	i	h	l	i	h
NP [%]	-	-	-	-	-	-	-	-	-
RBD [%]	96.4	3.6	-	4.2	12.5	83.3	-	-	100
S1 [%]	96.4	3.6	-	4.2	12.5	83.3	-	-	100

* Sera from people after SARS-CoV-2 vaccination which were collected 10 to 50 days after vaccine dose 1, 2 or 3. The collective includes vaccinated people without known previous SARS-CoV-2 infection, and the following vaccine was used: BioNTech/Pfizer Comirnaty®

** Avidity: l = low, i = intermediate, h = high

12 Literature

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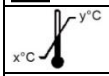
We will be pleased to send you additional literature on request.

13 Explanation of symbols

	Content is sufficient for <n> formulations Number of formulations
WASHBUF A 10 X	Wash buffer A (10× concentrate)
SUBS TMB	Chromogenic substrate tetramethylbenzidine
MILKPOW	Skimmed milk powder
TESTSTR	Test strips
CONJ IgG	Anti-human IgG conjugate
AVIDI	Avidity reagent
EVALFORM	Evaluation sheet
INSTRU	Instructions for use
	Follow the instructions for use
CONT	Content, contains
IVD	In-vitro diagnostic medical device
LOT	Batch/version number
	Do not freeze
REF	Order number
	Use by Expiry date
	Store between x°C and y°C
	Manufacturer

14 Manufacturer and version data

***recom*Line SARS-CoV-2 IgG [Avidität]**	Article no. **7374**
Instructions for use Valid from	GARLCS004EN 2023-03

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