SARS-CoV-2-Screen
SARS-CoV-2 real-time PCR Detection Kit

User manual

REF DP05.E

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IVD

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Abbreviations

RT-PCR	Reverse transcription polymerase chain reaction.
Real-time PCR-RT	Real-time reverse transcription polymerase chain reaction with fluorescence detection.
PC SARS-CoV-2	Positive control sample for steps of nucleic acid isolation and real-time PCR-RT.
NC	Negative control sample for steps of nucleic acid isolation and real-time PCR-RT.
QC	Quality control of taking biomaterial – endogenous internal control RT-PCR. The target RNA is human ribonuclease P.
cDNA	Complementary DNA – DNA synthesized on an RNA template in a reverse transcription reaction.

1. INTENDED USE

SARS-CoV-2 real-time PCR Detection Kit SARS-CoV-2-Screen is intended for the qualitative detection of SARS-CoV-2 coronavirus RNA by reverse transcription polymerase chain reaction (RT-PCR) with real-time fluorescence detection in clinical samples (nasopharyngeal swab, oropharyngeal swab, sputum).

For professional use in clinical laboratory diagnostics; for examination of male and female patients without age limitation with clinical symptoms of respiratory disease with suspected COVID-19 infection.

Demographic and population aspects of the application. SARS-CoV-2-Screen is recommended for examination of male and female patients without age limitation.

Target analyte. SARS-CoV-2 coronavirus: ORF1ab gene fragment of SARS-CoV-2 coronavirus, S gene fragment of SARS-CoV-2 coronavirus.

The intended purpose of the test performed using SARS-CoV-2-Screen is to support respiratory viral infection diagnostics, screening and epidemiological monitoring to identify the source of infection and assess the effectiveness of treatment.

The indications for the use of SARS-CoV-2-Screen are regulated by Surveillance case definitions for human infection with novel coronavirus (nCoV) WHO/2019-nCoV/Surveillance/2020.2 approved by World Health Organization and territorial legislative recommendations.

Application. Professional use in clinical laboratory diagnostics and research practice. For *in vitro* diagnostic only.

Users' qualification requirements. A specialist with a higher or secondary specialized medical education, trained in licensed laboratory diagnostics courses.

2. PRINCIPLES OF THE PROCEDURE

Working principle of SARS-CoV-2-Screen is based on the real-time PCR-RT method with hybridization-fluorescence detection. The detection of the target RNA, test validation through the real-time PCR and internal control samples, is carried out due to the release of FAM, ROX and HEX fluorophore molecules. They are released as a result of cleavaging the specific probes by polymerase during the amplification of cDNA strands synthesized from the RNA template of the SARS-CoV-2 coronavirus. Registration of the increase or absence of a specific fluorescent signal in the reaction mixture is carried out using three fluorescence detection channels simultaneously: FAM, ROX and HEX.

A necessary condition for a qualitative analysis is the fulfillment of all the requirements of the preanalytical stage of the study. An indicator of the correct sampling of biomaterial is sufficient amount of human genomic RNA in the sample that is detectable by the QC indicator (quality control of taking biomaterial). The source of this RNA is human cells that enter the sample with the correct technique for taking biomaterial. The composition of the DNA probe used for the detection of the QC amplification product includes a HEX/Yellow fluorescent dye (**Table 1**).

Table 1. Channels for detection of amplification products

FAM/Green	ROX/Orange	HEX/Yellow
Fragment of the SARS-CoV-2 coronavirus ORF1ab gene	Fragment of the coronavirus SARS-CoV-2 S gene	QC (Human ribonuclease P)

Detection of SARS-CoV-2 coronavirus RNA in test samples consists of three stages:

1) Extraction of RNA;

2) Synthesis of cDNA from the RNA template of the SARS-CoV-2 coronavirus;

3) Amplification of specific regions of cDNA by PCR with simultaneous hybridization-fluorescence detection in real-time mode.

The reagent kit detects simultaneously SARS-CoV-2 coronavirus RNA and human ribonuclease P (presence or absence of a signal through the corresponding fluorescence channel) and allows the user to interpret the result.

The interpretation is carried out in two stages:

1) Assessment of the quality of taking biomaterial (QC, HEX channel) – valid / invalid;

2) Registration of the result (SARS-CoV-2, FAM and ROX channels) – detected / not detected.

3. KIT DESCRIPTION

3.1. SARS-CoV-2-Screen description

SARS-CoV-2-Screen is available in one complete set (**Table 2**), designed for 96 tests, including control samples. SARS-CoV-2-Screen is not sterile.

Table 2. SARS-CoV-2-Screen description

Kit Component	Description	Amount
SARS-CoV-2 Buffer	Transparent liquid from colorless to pink	1.5 ml x 1 bottle
RT-polymerase	Colorless clear liquid	0.1 ml x 1 bottle
PC SARS-CoV-2	Colorless clear liquid	1.0 ml x 1 bottle
NC	Colorless clear liquid	1.0 ml x 1 bottle
User manual	-	1 piece
Certificate of quality	-	1 piece

3.2. Control materials included in SARS-CoV-2-Screen

PC SARS-CoV-2 – Positive control sample for the steps of nucleic acid isolation and real-time PCR-RT of SARS-CoV-2-Screen. It is applied to control the progress of the reaction. It is to be used in every study. The concentration of the recombinant phage is 10^5 copies/ml.

NC – Negative control sample for the steps of nucleic acid isolation and real-time PCR-RT of SARS-CoV-2-Screen. It is applied to control the progress of the reaction. It is to be used in every study.

QC – Quality control of sampling biomaterial – endogenous internal control for real-time PCR-RT. The target RNA is human ribonuclease P. It is necessary for quality control of biomaterial sampling and RNA extraction. Used to monitor the progress of the reaction in each study.

4. PERFORMANCE CHARACTERISTICS

4.1. Analytical specificity

The analytical specificity of SARS-CoV-2-Screen was checked *in silico* with the following infectious agents: Human coronavirus 229E, Human coronavirus OC43, Human coronavirus HKU1, Human coronavirus NL63, SARS-coronavirus, MERS-coronavirus, Adenovirus (e.g. C1 Ad. 71), Human Metapneumovirus (hMPV) , Parainfluenza virus 1-4, Influenza A & B, Enterovirus (e.g. EV 68), Respiratory syncytial virus, Rhinovirus, Chlamydia pneumoniae, Haemophilus influenzae, Legionella pneumophila, Mycobacterium tuberculosis, Streptococcus pneumoniae, Streptococcus pneumoniae, Streptococcus pyogenes, Pneumocystis jiroveci pneumonia (PJP), Candida albicans, Pseudomonas aeruginosa, Staphylococcus epidermis, Staphylococcus salivarius. The performed *in silico* analysis indicates that nonspecific amplification that could lead to a cross-reaction or affect the amplification of the SARS-CoV-2 target region is extremely unlikely.

The influence of potentially **interfering substances** on the performance of the reagent kit was tested in relation to potentially interfering substances that might occur during the procedure for

collecting biological material:

1. Hemoglobin 10% v/v
2. Mucin 5% v/v
3. "Ibuprofen" 0.04 mg/ml
4. "Ambrobene" 0.003 mg/ml
5. "Bromhexine" 0.016 mg/ml
6. "Kaletra" 0.02 mg/ml
7. "Interferon" 0.2 U/ml
8. "Teraflu" 0.071 mg/ml

These potentially interfering substances encountered in the procedure for isolating RNA from clinical material, evaluated at concentrations that would be expected to occur during normal use of the reagent kit, do not interfere with the assay result.

4.2. Analytical sensitivity

To determine analytical sensitivity, the EDX SARS-CoV-2 standard (BIO-RAD, USA) was used.

The sensitivity of the kit was measured to be $5*10^2$ copies of virus RNA (viral particles) per ml.

4.3. Diagnostic specificity

The diagnostic specificity of SARS-CoV-2-Screen (with a confidence level of 95%) based on the results of examining 25 samples of nasopharyngeal swabs characterized as not containing SARS-CoV-2 RNA was 100% (95% CI: 86.3 ~ 100.0).

The diagnostic specificity of SARS-CoV-2-Screen (with a confidence level of 95%) based on the results of examining 25 samples of oropharyngeal swabs characterized as not containing SARS-CoV-2 RNA was 100% (95% CI: 86.3 ~ 100.0).

The diagnostic specificity of SARS-CoV-2-Screen (with a 95% confidence level) based on the results of examining 25 sputum samples characterized as not containing SARS-CoV-2 RNA was 100% (95% CI: 86.3 ~ 100.0).

4.4. Diagnostic sensitivity

The diagnostic sensitivity of SARS-CoV-2-Screen (with a confidence level of 95%) based on the results of examining 25 samples of nasopharyngeal swabs characterized as containing SARS-CoV-2 RNA was 100% (95% CI: 86.3 ~ 100.0).

The diagnostic sensitivity of SARS-CoV-2-Screen (with a confidence level of 95%) based on the results of examining 25 samples of oropharyngeal swabs characterized as containing SARS-CoV-2 RNA was 100% (95% CI: 86.3 ~ 100.0).

The diagnostic sensitivity of SARS-CoV-2-Screen (with a confidence level of 95%) based on the results of examining 25 sputum samples characterized as containing SARS-CoV-2 RNA was 100% (95% CI: 86.3 ~ 100.0).

4.5. Reproducibility

The study of precision under reproducibility conditions was carried out on three instruments with three different operators with each configuration.

According to the test results, complete intra-stage, inter-stage and inter-series reproducibility was observed, the coefficient of variation does not exceed 2%.

5. CLINICAL MATERIAL RECOMENDATION

Clinical samples, including oropharyngeal swab, nasopharyngeal swab or sputum are to be used as clinical material for the extraction of nucleic acids.

According to WHO guidelines «Diagnostic testing for SARS-CoV-2» the optimal specimen depends on clinical presentation and time since symptom onset. Respiratory specimens:

• Upper respiratory specimens are adequate for testing early-stage infections, especially in asymptomatic or mild cases. Testing combined nasopharyngeal and oropharyngeal swabs from one individual has been shown to increase sensitivity for detection of respiratory viruses and improve the reliability of the result. Two individual swabs can be combined in one collection tube or a combined nasopharyngeal and oropharyngeal swab can be taken. A few studies have found that individual

nasopharyngeal swabs yield a more reliable result than oropharyngeal swabs.

• Lower respiratory specimens are advised if collected later in the course of the COVID-19 disease or in patients with a negative URT sampling and there is a strong clinical suspicion of COVID-19. LRT specimens can consist of sputum, if spontaneously produced (induced sputum is not recommended as this poses an increased risk of aerosol transmission). Before implementing other respiratory or oral fluid sampling methods, the sampling method should first pass validation in the laboratory for the intended patient groups.

Specimens for virus detection should reach the laboratory as soon as possible after collection. Correct handling of specimens during transportation and in the laboratory is essential.

Specimen type	Collection materials	Recommended temperature for storage and/or shipment to the laboratory and until testing (from date of specimen collection) #
Nasopharyngeal and oropharyngeal swab	Dacron or polyester flocked swabs with VTM *	2–8 °C if ≤12 days* minus 70 °C (dry ice) if > 12 days
Sputum	Sterile container	2–8 °C if ≤ 2 days minus 70 °C (dry ice) if > 2 days

 # Avoid repeated freezing and thawing of specimens. If no access to minus 70 °C, consider storing at minus 20 °C.

** For transport of specimens for viral detection, use preferentially viral transport medium (VTM) containing antifungal and antibiotic supplements. If VTM is not available, other solutions may be used after validation. Such solution may include phosphate buffered saline (PBS), 0.9% sterile saline, minimum essential medium (with storage at +4 °C up to 7 to 14 days). In case other viruses such as influenza should also be tested, do not store samples for more than 5 days at 4–8 degrees but minus 70 °C or dry ice.*

6. MATERIALS REQUIRED BUT NOT SUPPLIED*

- Reagent kit for the isolation of DNA/RNA from clinical material (for example, ExtraMAG 1, ExtraMAG 2 or ExtraMAG 3 reagent kit, LabPack LLC, Russia);
- Detecting amplifier for polymerase chain reaction in real time (DTlight 4S1, DNA-Technology, Russia; DT96, DNA-Technology, Russia; DTprime, DNA-Technology, Russia; Rotor – Gene Q, QIgene, USA; CFX96 Optics Module, Bio-Rad, USA; QuantStudio 5, Thermo Fisher Scientific);
- Laminar box II or III class of biological safety (for example, "BAVp-01- "Laminar-S"-1,2", "Laminar systems", Russia);
- Thin-walled tubes for PCR with a volume of 0.2 ml with a convex or flat optically transparent cap or test tubes with a volume of 0.2 ml in strips of 8 pcs with transparent lids (for example, Axygen, USA) – when using a tablet-type device;
- Thin-walled tubes for PCR with a volume of 0.2 ml with a flat cap (for example, Axygen, USA) or tubes for PCR to Rotor-Gene with a volume of 0.1 ml in strips of 4 pcs with covers (for example, QIAGEN GmbH, Germany) – when using a rotary device;
- Racks for test tubes with a volume of 0.2 ml or 0.1 ml (in accordance with the used reagent kits) (for example, Axygen, USA);
- Tight-fitting 1.5 ml tubes (for example, Axygen, USA) – for preparation of the reaction mixture;
- Packs for test tubes with a volume of 1.5 ml (for example, Axygen, USA);
- Mini-Centrifuge/Vortex (for example, FV-2400 Micro-Spin, SIA Biosan, Latvia);
- Refrigerator with chambers that maintain a temperature from +2 to +8 °C and with a freezer that maintain a temperature not higher than minus 16 °C;
- 1-channel mechanical pipettors with a variable dosing volume, certified by the average dose value and the repeatability of pipetting results (error no more than 3%) (for example, Sartorius Biohit,

Finland);

- Disposable tips with a filter for semi-automatic pipettors marked "RNAase-free, DNAase-free" in volumes of 1 – 20 µl, 5 – 50 µl, 20 – 200 µl, 100 – 1000 µl (for example, Axygen, USA);
- Personal Protective Equipment (PPE): disposable dressing gown, hat, mask, gloves;
- Container with a lid for a disinfectant solution.

** In the absence of the above equipment and materials, it is allowed to use other means with characteristics no worse than those given.*

7. INSTRUCTIONS FOR USE

To achieve the declared sensitivity, it is recommended to use ExtraMAG 1, ExtraMAG 2 or ExtraMAG 3 DNA/RNA Isolation Kits, LabPack LLC, Russia.

7.1. Preparation of the components of SARS-CoV-2-Screen for analysis

Table 3. Preparing components for analysis

Kit Component	Preparation of the components
SARS-CoV-2 Buffer	Ready to use
RT-polymerase	Ready to use
PC SARS-CoV-2	Ready to use
NC	Ready to use

7.2. Real-time PCR-RT stage

ATTENTION! The choice of tubes for amplification depends on the used thermocycler with a real-time detection system. Disposable filter tips have to be used to add reagents, RNA and control samples to tubes.

7.2.1. SARS-CoV-2 buffer should be brought to room temperature (+18...+25 °C) before analysis, stirred thoroughly and the drops precipitated on a vortex. The RT-polymerase is to be transferred from the freezer just before use, stirred thoroughly, and the droplets precipitated on a vortex.

7.2.2. Prepare the reaction mixture: mix the required amount of SARS-CoV-2 Buffer with the specified amount of RT-polymerase (**Table 4**). Prepare the mixture on the basis of $n + 3$, where n is the number of samples to be analyzed, *3* is the number of additional tubes. Additional tubes are necessary for the positive (PC SARS-CoV-2), negative (NC) control sample and the minimum reserve for dosing error (1 sample). An example of the calculation of the required amount of reagents is given in **Table 4**. After preparing the reaction mixture, stir it thoroughly on a vortex and precipitate the drops from the cap of the test tube.

Table 4. Example of calculating the amount of reagents for preparing the reaction mixture

Number of tubes	96	90	80	70	60	50	40	30	20	10
SARS-CoV-2 Buffer, µl	1500	1350	1200	1050	900	750	600	450	300	150
RT-polymerase, µl	100	90	80	70	60	50	40	30	20	10

ATTENTION! It is recommended to prepare reaction mixtures just before analysis. ***The prepared reaction mixture cannot be stored!***

7.2.3. Place the required number of tubes or strips in the rack for amplification of the test and control samples.

7.2.4. Pipette 15 µl of the reaction mixture into the appropriate tubes.

7.2.5. Add 10 µl of RNA solution of the analyzed sample and the negative control sample (NC) that has passed the stage of nucleic acid extraction to the prepared tubes with the activated buffer. Close all lids except for positive control tubes.

7.2.6. In a separate PCR box for adding positive controls, add 10 µl of PC SARS-CoV-2 sample that has passed the nucleic acid isolation step to the corresponding tube and close the lid.

7.2.7. Install all tubes (strips) into the detecting thermocycler unit.

7.2.8. Program the amplifier according to the manufacturer's instructions and run the appropriate

amplification program (**Tables 5** and **6**).

Table 5. SARS-CoV-2-Screen program for DTlight 4S1, DTprime, DT-96 (DNA-Technology), CFX-96 (Bio-Rad), QuantStudio 5 (Thermo Fisher Scientific) devices

Block no.	Temperature, ° C	Min	Sec	Cycles	Detection	Block type
1	48	30	00	1		Cycling
2	95	5	00	1		Cycling
3	95	0	10	5		Cycling
	60	0	20			
4	95	0	10	40		Cycling
	60	0	20		V	

For DTlight, DTprime, DT-96 (DNA-Technology) devices, set the following exposure values in the "Measurements exposure" tab: on the FAM channel – 2500; on the HEX channel – 3000; on the ROX channel - 2000. Set the "Method" – "Threshold Ct" and the threshold line value of 10%, obtained for the PC SARS-CoV-2 sample in the last amplification cycle. If necessary, the threshold can be adjusted in the range of 0 – 20%.

For CFX96 (Bio-Rad) devices, set the following Baseline Threslhold values in the Settings → Baseline threshold → User defined tab for FAM channel – 50, for ROX channel – 50, for channel HEX – 50.

Table 6. SARS-CoV-2-Screen program for Rotor Gene 3000, 6000 (Corbett Research) and Rotor-Gene Q (QIAGEN) devices

Block no.	Temperature, °C	Min	Sec	Cycles	Detection	Block type
1	48	30	00	1		Hold
2	95	5	00	1		Hold
3	95	0	10	5		Cycling
	60	0	20			
4	95	0	10	40		Cycling 2
	60	0	30		V	

For rotary instruments Rotor Gene 3000, 6000 (Corbett Research) and Rotor-Gene Q (QIAGEN), it is necessary to calibrate before the first measurement (activate Perform Calibration Before 1st Acquisition/Perform Optimization Before 1st Acquisition/Perform optimization at 1st step of detection).In the Auto gain calibration channel settings section, set the calibrations for each channel from 5Fl to 10Fl. Select linear scale type (Linear scale). In the menu of each main window (Quantitation analysis), the Dynamic tube and Slope Correct buttons must be activated. In the CT calculation menu (in the right part of the window), for each of the main windows set the threshold line level Threshold = 0.05. Select the More settings / Outlier Removal parameter and set the value of the negative samples threshold (NTC Threshold) equal to 10%.

Fluorescence detection (Acquiring to Cycling A/Detection on Cycling A) via the FAM/Green, ROX/Orange and HEX/Yellow channels is switched on at the second step (+60 °C) of the second cycling block.

7.2.9. When the program is ended, start analyzing and interpreting the results.

7.3. Amplification results accounting

Fluorescence detection of the amplification products is carried out in real time using a detecting amplifier. To interpret the test results, software is used that is not included in the kit and is provided by the manufacturer of the kit additionally. After the end of amplification, a study report will be automatically generated, including data on the magnitude of the indicator cycle for each analyzed type of microorganism.

8. CONTROL OF REACTION PROGRESS

8.1. In the Certificate of quality attached to each lot of SARS-CoV-2-Screen, the cut-off values of the threshold (indicator) Ct cycles for positive (PC SARS-CoV-2), negative control samples (NC) and biomaterial control for the test samples (QC) are indicated through the corresponding fluorescence channels.

8.2. For each run, a conclusion about the reliability of the test results is made on the basis of comparison of the indicators obtained for the PC SARS-CoV-2 and NC with the Certificate of quality indicators.

8.3. If the threshold cycle values exceeding the PC SARS-CoV-2 threshold are obtained, the results of the entire series are considered unreliable. All samples should be reanalyzed starting from the amplification step.

8.4. If the values of the threshold cycle are lower than the maximum permissible for NC, the results of the whole staging series are considered unreliable. It is necessary to repeat the study starting from the RNA extraction step.

9. INTERPRETATION OF RESULTS

9.1. To interpret the results, it is necessary to analyze the obtained data – fluorescent signal accumulation curves through FAM/Green, ROX/Orange and HEX/Yellow channels.

Table 7. Channels of fluorescence detection

FAM/Green	For SARS-CoV-2 coronavirus ORF1ab gene fragment amplification product in the tested samples, PC SARS-CoV-2 (positive result – Ct is defined and does not exceed the passport value) and NC (negative result – Ct is not defined or exceeds the passport value)
ROX/Orange	For SARS-CoV-2 coronavirus S gene fragment amplification product in the tested samples, PC SARS-CoV-2 (positive result – Ct is defined and does not exceed the passport value) and NC (negative result – Ct is not defined or exceeds the passport value)
HEX/Yellow	For the endogenous internal control amplification product – human ribonuclease P RNA fragment (quality control of biomaterial collection) in the tested samples, PC SARS-CoV-2 (positive result – Ct is defined and does not exceed the passport value) and NC (negative result – Ct is not defined or exceeds the passport value)

9.2. The results are to be interpreted in the following order:

9.2.1. Analysis of Ct threshold cycle values for PC SARS-CoV-2 and NC:

The results are interpreted only if the Ct threshold cycles of the FAM, ROX and HEX channels for the positive (PC SARS-CoV-2) and negative (NC) control samples correspond to the passport values (see the Certificate of quality of the kit lot).

9.2.2. Analysis of the results of human ribonuclease P detection (quality control of biomaterial sampling) by HEX/Yellow channel:

If the HEX/Yellow channel Ct QC threshold cycle value in the results table is defined and less than or equal to 33, the result is considered valid and can be proceeded to the next stage of results analysis (see 9.2.3.).

If no HEX/Yellow Ct value is detected for a given sample or the HEX/Yellow Ct QC threshold cycle value is greater than 33, the result is considered invalid. In this case, it is necessary to repeat the PCR study of the respective sample. An exception is the samples for which the Ct values of the FAM/Green and ROX/Orange channels are defined and do not exceed the threshold values (see **Table 9**).

Table 8. Accounting for QC results by HEX/Yellow channel

Biomaterial quality assessment result (QC)	Ct (QC) for nasopharyngeal swab, oropharyngeal swab, sputum
The result is **valid** (no inhibition, enough	The value is defined and does not exceed the threshold.

of biomaterial)	The exceptions are samples for which Ct values for the FAM / Green and ROX / Orange channels are defined and do not exceed the threshold
Invalid result (inhibition or insufficient amount of biomaterial)	The value is undefined or exceeds the threshold

9.2.3. Analysis of the results of detecting RNA fragments of the SARS-CoV-2 coronavirus by the FAM/Green and ROX/Orange channel:

• SARS-CoV-2 coronavirus RNA is **detected** if Ct values for the FAM/Green and ROX/Orange channels are defined for a given sample in the table of results. In this case, the fluorescence curve of this sample should cross the threshold line in the area of the characteristic exponential rise in fluorescence.

• RNA of the SARS-CoV-2 coronavirus was **not detected** if the Ct value for a given sample in the table of results was not defined for at least one of the FAM/Green or ROX/Orange channels.

Table 9. Calculation of results for FAM / Green and ROX / Orange channels.

Ct HEX/Yellow (QC)	Ct FAM/Green	Ct ROX/Orange	Result
Value is **defined / undefined**	Value is **defined**	Value is **defined**	SARS-CoV-2 coronavirus RNA **detected**
Value is **defined** and does not exceed the passport value	Value is **undefined**	Value is **defined**	SARS-CoV-2 coronavirus RNA **not detected**
	Value is **defined**	Value is **undefined**	SARS-CoV-2 coronavirus RNA **not detected**
	Value is **undefined**	Value is **undefined**	SARS-CoV-2 coronavirus RNA **not detected**

10. REAGENT HANDLING AND STORAGE

10.1. Storage conditions

Store SARS-CoV-2-Screen at temperatures ranging from minus 16 °C to minus 25 °C. Do not use the kits stored in violation of the regulated regime.

10.2. Transportation conditions

SARS-CoV-2-Screen can be transported by covered transport (road, rail or air) in thermal containers containing refrigeration elements at temperatures from +2 to +8 °C not more than 5 days.

After receiving store the kit in accordance with the storage temperatures indicated on the package. Do not use the kits transported in violation of the temperature regime.

10.3. Product shelf life*

SARS-CoV-2-Screen shelf life is 12 months.

10.4. Storage conditions

Table 10. Storage conditions and shelf life of opened product components

№	Kit Component	Storage conditions and shelf life
1	SARS-CoV-2 Buffer	1 month at temperatures from minus 16 °C to minus 25 °C
2	RT-polymerase	1 month at temperatures from minus 16 °C to minus 25 °C
3	PC SARS-CoV-2	1 month at temperatures from minus 16 °C to minus 25 °C
4	NC	1 month at temperatures from minus 16 °C to minus 25 °C

*- *Data from the accelerated assessment of medical device stability are not supported by real-time studies.*

11. WARNINGS AND SPECIAL PRECAUTIONS REGARDING THE SAFE DISPOSAL OF REAGENT KIT

11.1. Only personnel trained in the methods of molecular diagnostics and the rules of work in the

clinical diagnostic laboratory in the established manner are allowed to work with reagents kit.

11.2. To prevent contamination with amplification products (amplicons), nucleic acid samples or biomaterials of newly investigated samples, reagents and consumables, and, as a consequence, the appearance of false positive results, the laboratory process should be unidirectional. Separate rooms (zones) are used for different stages of the analysis. Work should start in the isolation zone, continue in the amplification and detection zone. Do not return samples, equipment and reagents to the area where the previous step of the process was carried out.

11.3. Always consider the samples under study to be infectious-hazardous, organize work and storage in accordance with the regulatory documents.

11.4. SARS-CoV-2-Screen is intended for single use in the study of the specified number of samples (see 3.1.)

11.5. It is allowed to use the kit for the intended purpose only, in accordance with these instructions and within the indicated expiration date.

11.6. Do not use the reagent kit if the inner packaging is broken or the appearance of the reagent does not correspond to the description.

11.7. Do not use the reagent kit if the conditions of transportation and storage have not been observed according to the instructions.

11.8. It is imperative to use personal protective equipment: disposable gloves, laboratory coats. Wash hands thoroughly after finishing work.

11.9. Avoid contact of the kit components with the skin, eyes and mucous membranes. In case of contact, rinse immediately the affected area with water and seek medical attention.

11.10. Disposal of the reagent kit. Unused reagents, expired reagents, as well as used reagents, packaging, biological material, including materials, tools and items contaminated with biological material, should be disposed in accordance with the requirements of Directive 2008/98/EC in EU or relevant regulations in the consumer's country.

Wastes containing pathogens (materials, tools and items contaminated with biological fluids) have to be decontaminated / neutralized.

The choice of the method of disinfection / neutralization is determined by the capabilities of the organization carrying out medical or biological activities. In the absence of a disinfection / neutralization site or a centralized medical waste disposal system adopted in the administrative territory, the kits are disinfected by the personnel on their workplaces by chemical / physical methods.

The manufacturer, suppliers, sellers, importers can dispose the kits that have lost their consumer properties or have expired, in accordance with Directive 2008/98/EC as class H9 medical waste using methods agreed with the territorial authorities responsible for sanitary and epidemiological well-being of the population. The personnel carrying out the disposal of the kits have to comply with the safety rules for carrying out one or another method of destruction. To obtain precise instructions, the relevant regulations in the consumer's country have to be used.

ATTENTION! When disposing waste after amplification, it is unacceptable to open test tubes and spray the contents since this can lead to the contamination of laboratory area, equipment and reagents with PCR products.

12. LIMITATION

Contamination at the stage of RNA isolation or real-time PCR-RT reaction is a possible reason for obtaining a false positive result during the subsequent procedure for detecting and identifying RNA by reverse transcription polymerase chain reaction (RT-PCR).

Negative results do not exclude the possibility of infection with the SARS-CoV-2 coronavirus and should not be used as the sole basis for deciding on patient treatment. Negative results should be correlated with clinical observations, medical history and epidemiological information.

The test does not detect the presence of SARS-CoV-2 coronavirus if its concentration is lower

than the declared analytical sensitivity ($5*10^2$ viral particles per ml).

13. MANUFACTURER'S WARRANTIES

13.1. SARS-CoV-2-Screen is manufactured by LabPack LLC. Address: Karpovka River Embarkment, 5, letter I, room 3-N, 197376 Saint-Petersburg, Russia.
Phone: +7 (812) 490 75 93.

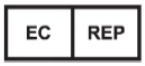
13.2. The manufacturer guarantees the compliance SARS-CoV-2-Screen with the requirements of the technical documentation if the conditions of transportation, storage and use established by this instruction for use are observed.

13.3. The user manual is provided by the manufacturer on paper and in the form of an electronic document via online publishing. The latest revision of the instruction manual: 31/08/2021.

13.4. Complaints about the quality of SARS-CoV-2-Screen should be sent to an authorized representative of the manufacturer in the European Community:

MagnaLab LLC, Address: Ludwika Rydygiera Str. 8/4, Warsaw 01-793, Poland.
Phone: +48 531 261 817, e-mail: info@magnalab.pl

Appendix A. Symbols to be used with medical device labels

Symbol	Title of symbol	Symbol	Title of symbol
EN ISO 15223-1:2016			
	Use-by date		Do not re-use
	Date of manufacture		Consult instructions for use
	Batch code		Caution
	Catalogue number		*In vitro* diagnostic medical device
	Keep away from sunlight		Do not use if package is damaged
	Keep dry		Contains sufficient for <n> tests
	Temperature limit		Manufacturer
	Authorized representative in the European Community		

Appendix B. Harmonised standarts

1	EN ISO 13485:2016	Medical devices – Quality management systems – Requirements for regulatory purposes
2	EN 13612:2002	Performance evaluation of in vitro diagnostic medical devices
3	EN 13641:2002	Elimination or reduction of risk of infection related to in vitro diagnostic reagents
4	EN 13975:2003	Sampling procedures used for acceptance testing of in vitro diagnostic medical devices – Statistical aspects
5	EN ISO 14971:2012	Medical devices – Application of risk management to medical devices
6	EN ISO 15223-1:2016	Medical devices – Symbols to be used with medical device labels, labelling and information to be supplied – Part 1: General requirements
7	EN ISO 18113-1:2011	In vitro diagnostic medical devices – Information supplied by the manufacturer (labelling) – Part 1: Terms, definitions and general requirements
8	EN ISO 18113-2:2011	In vitro diagnostic medical devices – Information supplied by the manufacturer (labelling) – Part 2: In vitro diagnostic reagents for professional use
9	EN ISO 23640:2015	In vitro diagnostic medical devices – Evaluation of stability of in vitro diagnostic reagents
10	EN 62366-1-2015	Medical devices – Part 1: Application of usability engineering to medical devices
11	Directive 98/79/EC	Directive 98/79/EC of the European Parliament and of the Council on in vitro diagnostic medical devices
12	Directive 2008/98/EC	Directive 2008/98/EC of the European Parliament and of the Council of 19 November 2008 on waste and repealing certain Directives

The above standards were valid at the time the instructions for use were approved. In the future, when using the document, it is advisable to check the validity of the reference normative documents at the current moment. If the referenced document is replaced or changed, then when applying this document, the replaced (modified) document should be used.