EURx® MOLECULAR BIOLOGY PRODUCTS

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Product intended for in vitro diagnostic use only.

CE IVD

SARS-CoV-2 qRT-PCR Detection KIT

SARS-CoV-2 qRT-PCR Detection KIT is designed to detect SARS-CoV-2 coronavirus in samples from patients with symptoms of COVID-19 infection. The purified genetic material of the virus from throat, nasopharyngeal, saliva, etc. is amplified by real-time RT-PCR and detected using SARS-CoV-2-specific probes, labeled with fluorescent dyes. Virus identification is based on highly conserved regions in the ORF1ab and NP genes characteristic for SARS-CoV-2. In addition, the kit includes primers specific for the human ACTB gene (gene encoding beta-actin), which makes it possible to verify the correctness of the collected swab and the quality of RNA purification. The test meets the requirements of the WHO recommendation "Laboratory testing for coronavirus disease 2019 (COVID-19) in suspected human cases" dated March 2, 2020. The kit shows no cross-reactions against other microorganisms causing respiratory diseases.

The kit includes:

Component	**Cat. no. E0430—01** **100 reactions, 25 µl each**	**Cat. no. E0430—02** **500 reactions, 25 µl each**
Cov Buffer Mix (2x) * brown tube	2 x 625 µl	10 x 625 µl
Cov Enzyme Mix orange cap	100 µl	5 x 100 µl
Positive Control ** black cap	50 µl	2 x 50 µl
RNase free water colourless cap	1200 µl	5 x 1200 µl

All kit components should be stored at a temperature of -20°C

*Avoid repeated thawing and freezing (>2), as this may reduce assay sensitivity. Freeze the component in aliquots if they are to be used intermittently.

**Positive Control should be stored separately, away from other kit components.

Preparation of the sample taken from the patient:

RNA from a swab, saliva or other secretion fluids should be isolated using kits dedicated to viral RNA purification. Follow the instructions recommended by the kit manufacturer.

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Protocol

Reaction components:
Preparation of PCR reaction:

Reaction component	Patient's RNA sample	Negative control	Positive control
Cov Buffer Mix (2x)	12.5 µl	12.5 µl	12.5 µl
Cov Enzyme Mix	1 µl	1 µl	1 µl
Positive Control	-	-	5 µl
Sample of purified RNA	5 µl	-	-
RNase free water	6.5 µl	11.5 µl	6.5 µl
Reaction volume	25 µl	25 µl	25 µl

1. Determine the number of reactions. Reactions can be prepared at room temperature. Cov Enzyme Mix and Cov Buffer Mix (2x) should be kept "on ice".
2. Prepare one reaction Master Mix, i.e. mix together the appropriate number of reaction ingredients according to above table except for RNA from the patient: mix appropriate amounts of Cov Buffer Mix (2x), Cov Enzyme Mix and water in the tube. Before taking Cov Buffer Mix (2x), mix the buffer by pipetting it several times.
3. The components of the Master Mix should be mixed by pipetting several times. Do not vortex the mixture.
4. Distribute the Master Mix into strip tubes or onto a plate by transferring 20 µl of the solution.
5. Prepare the reactions in the following sequence:

    5a. Negative control: add 5 µl of water and close the tube to avoid contamination by the patient's RNA.

    5b. Patient RNA samples: add 5 µl of purified RNA and close the tubes.

    5c. Positive control: add 5 µl of Positive Control.

6. One positive and one negative control should be set in addition to the patient samples to be tested for each test cycle.
7. Put the prepared reactions into the thermal cycler and program it as follows:

Fluorescence is measured at the annealing/extension stage in three channels:

Step	Temperature	Time	Number of cycles
Reverse transcription	50°C	15 min	1
Initial denaturation	95°C	2 min	1
Denaturation	95°C	10 s	40-45
Annealing / extension	60°C	40 s	

FAM for ORF1ab gene; HEX for ACTB gene and **ROX** for NP gene.

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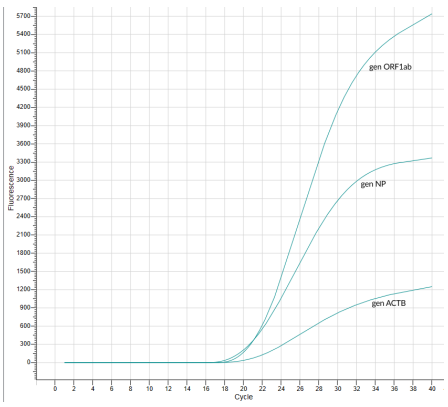
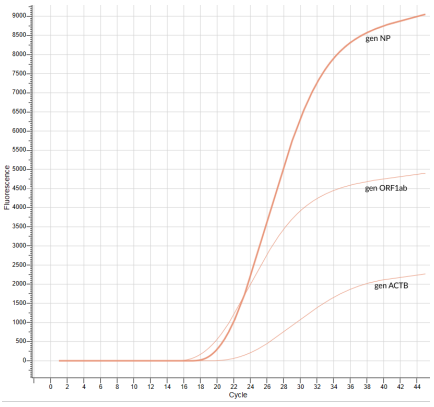
Interpretation of results:

Sample type	FAM channel	HEX channel	ROX channel	Result
Negative control	-	-	-	correct
Positive control	+	+	+	correct
1	-	+	-	negative for SARS-CoV-2
2	+	+	+	positive for SARS-CoV-2
3	+	+	-	positive for SARS-CoV-2
4	-	+	+	positive for SARS-CoV-2
5	-	-	-	incorrect, repeat the test
6	+	-	+	incorrect, repeat the test

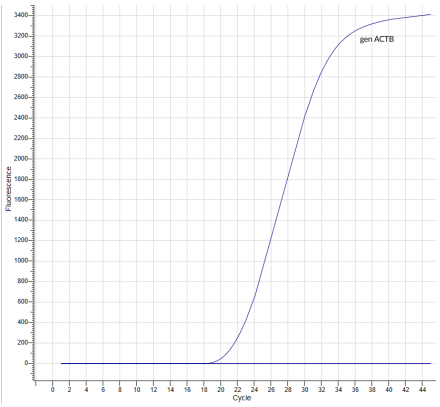
The test sample is considered positive if it shows a fluorescence signal in the HEX channel and at least in one of the other two channels, where Ct should be less than or equal to 38.



Positive control



Positive for SARS-CoV-2



Negative for SARS-CoV-2

Note:

If you receive an incorrect or a doubtful result, it is recommended to repeat the test.

All tests should be performed by qualified personnel and assessed in the context of clinical symptoms and medical history.

During the test, correct collection of the swab and the quality of purified RNA is very important.

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