

## CoV Screen

### Detection system for SARS-CoV viral RNA by RT-qPCR

#### For Research Use Only!

#### Indication

**CoV Screen** is a broad RT-qPCR detection system for the RNA of the Severe Acute Respiratory Syndrome Coronaviruses (SARS-CoV). Combined with our universal one-step RT-PCR enzyme mix ConviFlex™ RT-Taq Mix (Cat. No. 192-0025/192-0100/192-0250), this primers/probes system allows qualitative detection of coronaviruses in extracted RNA samples.

This product is designed on the basis of the broad-spectrum SARS-CoV detection assays and sequences (both primers and probes) indicated by the WHO reference diagnostic laboratory in Germany (Ref. 1; <https://www.who.int/emergencies/diseases/novel-coronavirus-2019/technical-guidance/laboratory-guidance>). SARS-CoV-viruses are the causative agents of various infectious respiratory diseases.

#### Principle of the Method

**CoV Screen** contains lyophilized primers/probes, which amplify the RNA-dependent RNA polymerase (RdRp) gene of SARS-CoV, bat-SARS-related CoVs, and SARS-CoV-2 (also named 2019-nCoV). For the specific virus-targeting sequences of the primers and probe, please see Refs. 1, 2. A broad range of SARS-CoV viruses are detected by these primers/probes (Refs. 1, 2). Please note that this kit is therefore not discriminating between a general SARS-CoV virus and SARS-CoV-2. Specific detection of SARS-CoV-2 can be qualitatively achieved by RT-qPCR with our **SARS-CoV-2 Confirm** detection system (Cat. No. 171-02-0100).

This product is used in combination with the universal one-step RT-PCR enzyme mix ConviFlex™ RT-Taq Mix (Cat. No. 192-0025/192-0100/192-0250) for qualitative detection of SARS-CoV in RNA samples. RNA may be extracted from diverse starting materials like tissue, biopsies and swabs (including respiratory specimens), mammalian cells and bacteria with a commercially-available RNA extraction kit (see ExtractNow™ Virus RNA Kit Cat. No. 611-1010/-1050) or a well-established extraction method.

The amplification of the SARS-CoV target (RdRp) is detected in the FAM™ channel. The mix includes also an amplification system for the human RNase P gene (process control), detected in the ROX™ channel, to assess the performance of the sample preparation procedure. Included is also a lyophilized Positive Control RNA containing a synthetic RNA sequence for the Wuhan-specific SARS-CoV-2 virus strain.

**Reagents**

Each kit contains reagents and components for 100 reactions (100 reactions / vial). The expiry date of the unopened package is marked on the package label. The kit components should be stored at +2 - +8 °C, and after rehydration at ≤ -18 °C. Protect the CoV Screen Mix from light. We recommend storing the rehydrated components in aliquots to avoid multiple freeze-thaw cycles.

Component	Quantity	
	100 Reactions	Cap Color
<b>Cat. No. 171-01-0100</b>		
CoV Screen Mix	1 vial, lyophilized	red
Positive Control RNA	1 vial, lyophilized	green
PCR Grade Water	1 vial	white

**User-supplied consumables and equipment**

The kit contains some of the components required for RT-PCR amplification. Additional consumables and equipment are supplied by the user:

- ConviFlex™ RT-Taq Mix (Cat. No. 192-0025/192-0100/192-0250). Please note that ConviFlex™ RT-Taq Mix is a lyophilized enzyme mix containing a reverse transcriptase and a Taq Polymerase. Resuspend as indicated in the Instructions for Use of ConviFlex™ RT Taq Mix.
- qPCR thermocycler
- Suitable qPCR reaction tubes (DNase- and RNase-free)
- Pipettes with corresponding filter tips (DNase- and RNase-free)
- Microcentrifuge
- User-specific RNA extracts or samples
- Optional for sample preparation: ExtractNow™ Virus RNA Kit Cat. No. 611-1010/-1050

**Precautions**

CoV Screen is intended for research use only. CoV Screen should be used by trained laboratory staff only.

All samples should be handled with all due care and attention, according to the specimen type. Always wear appropriate protective clothing and disposable gloves.

This kit does not contain hazardous substances. Remnants can be discarded according to local regulations.

**Additional Notes**

- ⇒ These instructions must be understood to successfully use the CoV Screen. The reagents supplied should not be mixed with reagents from different lots but used as an integral unit. The reagents of the kit must not be used beyond shelf life.
- ⇒ Follow the exact protocol when preparing CoV Screen RT-qPCR. Deviations may affect the test method and results.
- ⇒ We recommend including controls on a regular basis to monitor the reliability of your results. Set up at least one positive, one negative extraction control sample, and one non-template control (NTC) in each PCR, in duplicates. The controls must be processed in the same manner as the test samples. You may want to include other lab-specific control samples such as high, median, and low RNA levels.
- ⇒ To avoid DNA and unspecific RNA cross-contaminations during the procedure, we recommend performing the RT-PCR under RNA- and DNA-free conditions, and according to the specimen type. The degradation of the extracted RNA greatly limits the performance of RT-PCR reactions. Minimizing the number of freeze-thaw cycles of RNA samples or RNase contaminations helps preventing RNA degradation. DNA contamination can also greatly reduce the RT-PCR yield. Please ensure high-quality intact RNA is used for the test.

**1. Reagent preparation**

1.1.	CoV Screen Mix	red cap	Spin down for 5 sec at maximum speed
	Positive Control RNA	green cap	
1.2.	CoV Screen Mix	red cap	Add 500 $\mu$ l PCR Grade Water (white cap).
1.3.	Positive Control RNA	green cap	Add 100 $\mu$ l PCR Grade Water (white cap).
1.4.	CoV Screen Mix	red cap	Incubate at room temperature for 5 min, vortex briefly and spin down for 5 sec
	Positive Control RNA	green cap	

**2. RT-PCR reaction mix preparation (20  $\mu$ l reaction volume)**

Pipetting scheme:			
		For 1 reaction	For 100 reactions
2.1.	ConviFlex™ RT-Taq Mix (Cat. No. 192-OXXX)	10 $\mu$ l	1000 $\mu$ l
	CoV Screen Mix	5 $\mu$ l	500 $\mu$ l

Important: ConviFlex™ RT-Taq Mix has to be purchased separately. Resuspend the ConviFlex™ RT-Taq Mix as indicated in the corresponding “Instructions for Use”.

- 2.2. Mix by tapping carefully against the tube or pipetting up and down 4 - 5 times.
- 2.3. Aliquot 15  $\mu$ l PCR master mix to each PCR reaction tube.
- 2.4. **Samples:** Add 5  $\mu$ l of RNA template.
- 2.5. **Negative Controls:** Add 5  $\mu$ l elution buffer from RNA extraction kit as negative control for extraction (NEC).  
Add 5  $\mu$ l PCR grade water as No-Template Control (NTC).
- 2.6. **Positive Control:** Add 5  $\mu$ l Positive Control RNA.
- 2.7. Close PCR tubes tightly and spin down briefly.
- 2.8. Place PCR tubes in the cycler and close the lid tightly.
- Program the cycler or load a stored cycler program, as in (1):
- 2.9.
- |           |                               |
|-----------|-------------------------------|
| 1 cycle   | 55 °C for 10 min              |
| 1 cycle   | 94 °C for 3 min               |
| 45 cycles | 94 °C for 15 sec              |
|           | 58 °C for 30 sec              |
|           | Hold between +4 °C and +10 °C |
- 2.10. Start the program.

## Data Interpretation

FAM™ channel	ROX™ channel	Interpretation
Positive	Positive	Detection of SARS-CoV RNA
Positive	Negative	Detection of SARS-CoV RNA
Negative	Positive	No detection of SARS-CoV RNA
Negative	Negative	Invalid sample preparation procedure

**Important:** In case of negative results in the FAM™ or ROX™ channel for the Positive Control, repeat PCR. In case of positive results in the FAM™ or ROX™ channel for the Negative Controls, consider checking for contaminations.

## References

1. Corman VM, Bleicker T, Brünink S, Schneider J, Drosten C. Diagnostic detection of 2019-nCoV by real-time RT-PCR. [https://www.who.int/docs/default-source/coronaviruse/protocol-v2-1.pdf?sfvrsn=a9ef618c\\_2](https://www.who.int/docs/default-source/coronaviruse/protocol-v2-1.pdf?sfvrsn=a9ef618c_2)
2. Corman VM, Landt O, Kaiser M, Molenkamp R, Meijer A, Chu DKW, Bleicker T, Brünink S, Schneider J, Schmidt ML, Mulders DGJC, Haagmans BL, van der Veer B, van den Brink S, Wijsman L, Goderski G, Romette JL, Ellis J, Zambon M, Peiris M, Goossens H, Reusken C, Koopmans MPG, Drosten C. Detection of 2019 novel coronavirus (2019-nCoV) by real-time RT-PCR. Euro Surveill. 2020 25(3). doi: 10.2807/1560-7917.ES.2020.25.3.2000045.

## Appendix

### Limited Product Warranty

This warranty limits our liability for replacement of this product. No warranties of any kind, express or implied, including, without limitation, implied warranties of merchantability or fitness for a particular purpose, are provided. Minerva Biolabs shall have no liability for any direct, indirect, consequential, or incidental damages arising from the use, the results of use, or the inability to use this product.

### Trademarks

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## Related Products

192-0025/-0100/-0250	ConviFlex™ RT-Taq Mix (25 reactions / vial)	25/100/250 Reactions
191-0025/-0100/-0250	ConviFlex™ DNAmix (25 reactions / vial)	25/100/250 Reactions
611-1010/-0050	ExtractNow™ Virus RNA Kit	10/50 Extractions
171-02-1100	SARS-CoV-2 Confirm Kit	100 Reactions
171-02-0100	SARS-CoV-2 Confirm	100 Reactions
171-01-1100	CoV Screen Kit	100 Reactions



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