AddMedi SARS-CoV-2 Sequencing Kit

Product Information

Product Code ADM-SCS100

Provided with

In Box-1

1. AddScript RT-PCR Master (2x conc.) 0.95 mL x 4 tubes
2. 10x Oligo Mixture-1 (10x OM-1) 0.25 mL
3. 10x Oligo Mixture-2 (10x OM-2) 0.25 mL
4. 10x Oligo Mixture-3 (10x OM-3) 0.25 mL

In Box-2

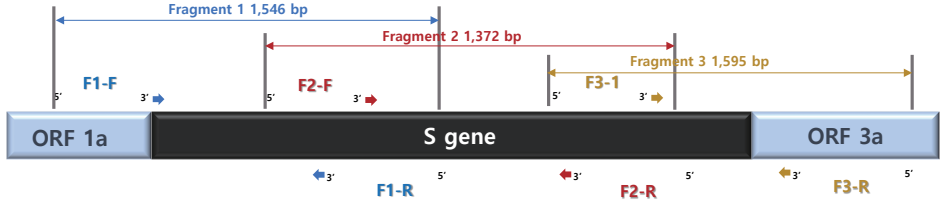
5. 5 μM Fragment-1 Forward Primer for Sequencing (F1-F) 0.1 mL
6. 5 μM Fragment-1 Reverse Primer for Sequencing (F1-R) 0.1 mL
7. 5 μM Fragment-2 Forward Primer for Sequencing (F2-F) 0.1 mL
8. 5 μM Fragment-2 Reverse Primer for Sequencing (F2-R) 0.1 mL
9. 5 μM Fragment-3 Forward Primer for Sequencing (F3-F) 0.1 mL
10. 5 μM Fragment-3 Reverse Primer for Sequencing (F3-R) 0.1 mL
11. Nuclease-free Water 0.1 mL

Storage Conditions -10°C ~ -30°C

Stability Stable for 1 year from manufacturing date.

Description

AddMedi SARS-CoV-2 Sequencing Kit can be used for Sanger sequencing the full spike gene (S gene) of SARS-CoV-2 RNA extracted from clinical positive samples. This kit is composed 3 primer sets for amplification of 3 fragments which 3 parts of S gene. The amplicon size of fragment 1 is 1,546 bp, fragment 2 is 1,372 bp and Fragment 3 is 1,595 bp. Especially the sequence of variable variants in S gene can be confirmed by sequencing analysis of fragment 1 and fragment 2.
The location of primers each is expressed in figure below.



Applications

- SARS-CoV-2 S gene Sequencing

Nucleic Acid Amplification and Sequencing Procedure

1. Add the following components to a thin-walled PCR tube:

AddScript RT-PCR Master (2x conc.)	12.5 μl
10x Oligo Mixture	2.5 μl
RNA template (or Positive control or Nuclease-free Water)	10.0 μl
Total reaction volume	25 μl

2. PCR cycling

cDNA synthesis	50°C, 20 min
Initial denaturation	95°C, 10 min
PCR cycling (40 cycles)	95°C, 30 sec
	60°C, 45 sec
	72°C, 60 sec
Final extension	72°C, 5 min
Hold	12°C, ∞

3. Electrophoresis

PCR product loading volume: 5 μl in 1.5% 0.5X TBE (or 1.0X TAE) agarose gel
* The loading dye for electrophoresis is not included in RT-PCR Master (2x conc.) for sequencing reaction and analysis. If the electrophoresis processing, any loading dye for electrophoresis can be used.

4. Sequencing

For sequencing, sequencing primer(5 μM) can be used 1 μl per 1 reaction with PCR product.

Manufacture

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