

# **Product information**

**BIO BASIC** 

QF 24 V4 V26 2020

# EZ-10 Spin Column COVID-19 Viral RNA Extraction Kit

 Catalog #:
 V T 9200

 Size:
 50 preps, 250 preps, 500 preps

 Storage:
 4°C to 25°C\*

\*: Product will be shipped at ambient temperature. Once recons tuted, carrier RNA to be stored at -20°C, remaining kit components to be stored at 4°C to 25°C. Components have a one year expira on from me of purchase.

### **Product Description:**

The kit is **especially designed for COVID-19 (SARS-CoV-2)**. It simplifies isola on of COVID-19 viral RNA from cell-free body fluids with fast spin-column format. No phenol/chloroform extrac on is required. Viral RNA binds specifically to the silica membrane while contaminants are removed in the flow-through. PCR inhibitors such as divalent ca ons and proteins are completely removed in two efficient wash steps, leaving pure viral R NA to be eluted in RN ase-free Wa ter. Purified RNA is ready to use in RT-PCR, Nort hern blo ng or other downstream applica ons.

#### **Features:**

- Fast: Using a rapid spin column format, the en re procedure takes about 20 minutes.
- High Yield: The recovery yield of viral RNA is generally >85%.
- Versa le: Suitable for purifica on of viral RNA from Nasopharyngeal swabs (dry) or Transport medium/Viral transport medium (VTM) containing Nasopharyngeal swabs, Nasopharyngeal aspirates, and bronchoalveolar lavage (BAL) samples.
- **Non-toxic:** No phenol/chloroform are used.

#### **Materials Supplied by User:**

- Microcentrifuge capable of at least 12,000 × g
- RNase-Free pipe es and pipette ps
- Vortexer
- RNase-Free Ethanol (96-100%)
- RNase-Free Microcentrifuge tubes (1.5 ml or 2 ml)

#### **Before Starting:**

i Freshly Prepare the following *Lysis Mastermix*:

Compo nent	Volume/rxn	Volume/50 rxns	Volume/250 rxns	Volume/500 rxns
Carrier RNA	2 μl	100 μl	500 μl	500 ul <b>x2</b>
Buffer Rlysis-VG	600 μl	30 ml	150 ml	150 ml <b>x2</b>

ii. Mix thoroughly by vortexing.

#### **Composition:**

Components	50 Prep Kit	250 Prep Kit	500 Prep Kit
Buffer Rlysis-VG	30 ml	15 0 m l 1	150 m l <b>x2</b>
Universal RPE Solu on **	12 ml	30 ml <b>x2</b>	40 ml <b>x3</b>
Nuclease-Free Water***	5 ml	25 ml	50 ml
EZ-10 Spin Columns	50	250	500
2 ml Collec on Tubes	50	250	500
Carrier RNA***	1 mg	1 mg	1 mg
Protocol		1	

\*\*: Universal RPE Solu on is supplied in a concentrated form. Before use, add 48 ml 96-100% ethanol to 12ml concentrated universal RPE solu on and mix well.

\*\*\*: Add Nuclease-Free Water to the tube containing lyophilized

Gam RNA to obtain a solu on of 1  $\mu$ g/ $\mu$ L. Dissolve the carrier RNA thoroughly, divide it into conveniently sized aliquots, and store it at -20°C. Do not freeze-thaw the aliquots of Carrier RNA more than 3 mes.

#### Storage:

Upon recons tu on, Carrier RNA should be divided into conveniently sized aliquots and stored

at –20°C. Do not freeze–thaw the aliquots of Carrier RNA more than 3 mes. All remaining kit components to be stored at 4°C to 25°C. The kit is valid for 1 year.



Distribución Autorizada

Dibbiotek

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## **Procedure:**

#### 1. Sample prepara on:

A) Nasopharyngeal swabs (dry):

Select between method a.1 (without Proteinase K) OR a.2 (with Proteinase K - op onal):

- a.1. Place the swab into a clean 1.5 ml microtube, and snap off the handle. Add 0.2 ml physiological saline, vortex for 30 sec. Then transfer 0.2 ml solu on to a new 1.5 ml microtube and proceed to step 2.
- **a.2.** If using Proteinase K (recommended for be er yield):

i) Add 300 µl of PBS (user supplied) and 10ul of 20mg/ml Proteinase K solu on (user supplied, *Bio Basic cat. #401*) to each swab sample.
ii) Incubate at 56°C for 5 min. with occasional mixing.
iii) Centrifuge at 10,000 X g (or max. speed) for 30 sec.

- iv) Use 200  $\mu$ l from this step as a sample and proceed to step **2**.
- B) <u>Transport medium/Viral transport medium (VTM)</u> <u>containing Nasopharyngeal swabs, Nasopharyngeal</u> <u>aspirates and bronchoalveolar lavage (BAL) samples:</u>\*\*\*

i. Vortex the tubes containing the swab at max. speed for 1 min.

ii. Use 200  $\mu$ l from this step as a sample and proceed with step **2**.

\*\*\*<u>Note:</u> If transfer medium is very dilute i.e. if the volume is more than 400 μl, perform the below steps for virus enrichment instead of "i" and "ii": iii. Transfer appropriate liquid sample to a new 1.5 ml microtube,

iv. Centrifuge at 24,000 g for 60 min. at 4°C. Then keep approx. 0.2 ml solu on in the tube but discard the remaining and con nue to step 2.

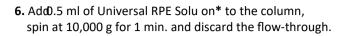
**2.** Add 0.6 ml of the recently prepared *Lysis Mastermix* into the tube from step **1**, vortex vigorously for 30 sec.; incubate at room temp. for 10 min.

NOTE: Lysis-Buffer-VG may form precipitate at 4°C, please dissolve it at 65°C and mix well before use.

**3.** Add equal volume of ethanol, mix by inver ng the tube.

**4.** Transfer the mixture into the spin column; keep at room temp. for 2 min.

5. Spin at 10,000 g for 1 min., discard the flow-through.

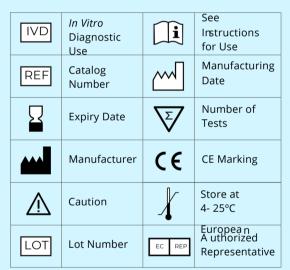


\*: Universal RPE Solu on is supplied in a concentrated form. Before use, add 48 ml 96-100% ethanol to 12ml concentrated universal RPE solu on and mix well.

7. Repeat the Step 6 once.

**8.** Centrifuge at 10,000 g for 1 min., discard the flow-through residue.

- **9.** Transfer the column to a new 1.5 ml RNase-free microtube. Add 35-60  $\mu$ l of Nuclease-Free Water onto the centre of the column; keep at room temp. for 2 min.
- **10.** Spin at 10,000 g for 1 minute. Purified viral RNA is ready for use or keep at -20°C for long term storage.



**AAA** 

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