Point of Care (POC) Workflow of The BioeXsen SARS-CoV-2 Triple Gene RT-qPCR Kit

Sampling

1- Take nasopharyngeal swab, oropharyngeal swab, oral/saliva swab, and nasal swab samples. *Use only the swab provided with the vNAT Transfer Tube*.

2- Place the sample into the vNAT Transfer Tube, close the tube cap and incubate for 5 minutes to *inactivate the pathogens*. Afterwards, the sample can *directly be used in RT-qPCR*.

Before RT-qPCR

3- Wear personal protective equipment (e.g., Mask, Gloves, Lab. Coat…).

4- Clean the surface with 0.5-1% (w/v) sodium hypochlorite (10-20% v/v bleach) or use Whatman Filter Paper to provide *clean work bench*.

Reaction Set-up

5- Combine the "2x Prime Script" and the "Oligo Mix" kit components in a microcentrifuge tube to obtain the "Master Mix".

Number of Samples	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
2x Prime Script (µL)	15	20	25	30	35	40	45	50	60	65	70	75	80	85	90	95
Oligo Mix (µL)	7.5	10	12.5	15	17.5	20	22.5	25	30	32.5	35	37.5	40	42.5	45	47.5
Number of Samples	**17**	**18**	**19**	**20**	**21**	**22**	**23**	**24**	**25**	**26**	**27**	**28**	**29**	**30**	**31**	**32**
2x Prime Script (µL)	100	115	120	125	130	135	140	145	150	155	160	165	170	175	180	185
Oligo Mix (µL)	50	57.5	60	62.5	65	67.5	70	72.5	75	77.5	80	82.5	85	87.5	90	92.5
Number of Samples	**33**	**34**	**35**	**36**	**37**	**38**	**39**	**40**	**41**	**42**	**43**	**44**	**45**	**46**		
2x Prime Script (µL)	190	195	200	205	210	215	220	225	230	235	240	245	250	255		
Oligo Mix (µL)	95	97.5	100	102.5	105	107.5	110	112.5	115	117.5	120	122.5	125	127.5		

6- *Vortex and spin* the "Master Mix" quickly.

7- Calculate the number of necessary micPCR tube as follows: *Number of Samples + 2 Controls*.

CAUTION: When transferring any liquid into the micPCR tube, always *use only the first stop* of the pipette, don't use the second stop (purge) *not to cause bubbles* at the bottom of the tube.

8- Dispense 7.5 µL of the "Master Mix" *into the bottom* of the each micPCR tube.

9- Add 2.5 µL of NTC into the bottom of the micPCR tube as a negative control reaction and *close it quickly*.

10- Vortex and transfer 2.5 µL of each sample into the bottom of the respective micPCR tube. After dispensing all the samples, close the tubes quickly.

11- Add 2.5 µL of PC into the bottom of the micPCR tube as a positive control reaction and *close it quickly*.

RT-qPCR

12- Place the tubes into the micPCR. There are 48 positions in the micPCR. If there are less than 48 tubes to be placed into the instrument, *place the balance tubes into the empty positions*.

13- Start RUN.

Reporting

14- After the run is completed, online *FastFinder software automatically evaluates and reports* the results.

After RT-qPCR

15- *Immediately after the run, put the reaction tubes into a plastic bag, close it tightly. Place the plastic bag into another plastic bag and close it tightly and discard it.*