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Application Note: Use of Z03730 with L00847-A SARS-CoV-2 Surrogate Virus Neutralization Test (sVNT) Kit

Description of SARS-CoV-2 Spike protein RBD-HRP, Omicron Variant, His Tag (Cat. No.: Z03730)

SARS-CoV-2 (Severe Acute Respiratory Syndrome Coronavirus 2) also known as 2019-nCoV (2019 Novel Coronavirus) is a virus that causes illnesses ranging from the common cold to severe diseases. Recently, the new B.1.1.529 variant was confirmed in South Africa and preliminary evidence suggests an increased risk of reinfection with this variant. The B.1.1.529 variant was first reported to WHO on 24 November 2021 and WHO has designated this variant as a VOC (Variant of Concern), named Omicron. There are more than 30 mutations in the spike protein.

Neutralizing antibodies against SARS-CoV-2 can block the interaction between SARS-CoV-2 RBD and ACE2. The blocking ability of neutralizing antibodies is affected by the virus strain. This product has been validated to bind with human ACE2 in functional ELISA assay. Users can design assays to detect SARS-CoV-2 neutralizing antibodies against Omicron RBD using this product with the SARS-CoV-2 Surrogate Virus Neutralization Test (sVNT) (GenScript, Cat.No L00847-A).

Due to the immune escape feature of the Omicron variant,¹⁻³ significant reduction in neutralization was observed when using the Omicron RBD-HRP with the positive control in sVNT.

Results from in-house testing

	OD450 (Z03730 Omicron RBD-HRP)
L00847-Positive Control (PC)	1.2249
L00847-Negative Control (NC)	2.3246

Recommended test procedure of Z03730 with SARS-CoV-2 Surrogate Virus Neutralization Test (sVNT) Kit (GenScript, Cat.No L00847-A)

 Use the same protocol for the Omicron RBD-HRP as per L00847-A manual. It is recommended to perform the test for Wildtype RBD-HRP (supplied in the sVNT kit) and Omicron RBD-HRP on the same plate as shown in the test configuration below.



Test Configuration

	Wildtype RBD-HRP						Omicron RBD-HRP					
	1	2	3	4	5	6	7	8	9	10	11	12
Α	NC	sample 5	sample 13				NC	sample 7	sample 15			
В	NC	sample 6	sample 14				NC	sample 8	sample 16			
С	PC	sample 7	sample 15				sample 1	sample 9	sample 17			
D	PC	sample 8	sample 16				sample 2	sample 10	sample 18			
Е	sample 1	sample 9	sample 17				sample 3	sample 11	sample 19			
F	sample 2	sample 10	sample 18				sample 4	sample 12	sample 20			
G	sample 3	sample 11	sample 19				sample 5	sample 13	sample 21			
Н	sample 4	sample 12	sample 20				sample 6	sample 14	sample 22			

NC: Negative control PC: Positive control

2. The NC and PC of wildtype (A1-D1) serve as the **quality control** to determine the validity of the assay as per L00847-A manual.

OD450 values for quality control

Items	OD450 value	Control Result for Valid Assay				
	> 1.0	Negative Control				
Quality Control	< 0.3	Positive Control				

Note: The controls in the table are only intended to evaluate the performance of the kit.

3. Use the averaged OD value of NC wildtype (A1, B1) and NC Omicron (A7, B7) for calculating % inhibition of samples tested for wildtype and omicron neutralization, respectively, as per L00847-A manual.

References

1 Wilhelm, A. et al. Reduced Neutralization of SARS-CoV-2 Omicron Variant by Vaccine Sera and Monoclonal Antibodies. medRxiv, 2021.2012.2007.21267432 (2021).

2 Cele, S. et al. SARS-CoV-2 Omicron has extensive but incomplete escape of Pfizer BNT162b2 elicited neutralization and requires ACE2 for infection. medRxiv, 2021.2012.2008.21267417 (2021).

3 Sokal, A. et al. Immune escape of SARS-CoV-2 Omicron variant from mRNA vaccination-elicited RBDspecific memory B cells. bioRxiv, 2021.2012.2021.473528 (2021).

Caution

For Research Use Only. Not for Use in Diagnostic Procedures.