

Challenge Virus: Middle East Respiratory Syndrome Coronavirus (MERS-CoV)

Experimental Summary:

Veridical Efficacy Test for Device with AHPCO Technology against the challenge virus Middle East Respiratory Syndrome Coronavirus (MERS-CoV), BEI Resources, with host Vero E5 cells, ATCC CRL-1586. Contact times 1 hour, 2 hours and 4 hours.

Study Dates & Procedure:

A glass carrier was inoculated with 0.2 mL of virus in a 4 in2 area and dried for 18 minutes at 20°C. Exposure distance approximately 5 cm. The test device was assembled and operated safely according to the manufacturer or Sponsor's instructions. The laboratory phase of this test was performed at MicroBioTest, 105 Carpenter Drive, Sterling, VA 20164, from 05/26/16 to 06/03/16. Titer Results:

Sample	Replicate	Contact time	Titer (Log ₁₀ TCID ₅₀ /mL)	Volume (mL)	Viral Load (Log ₁₀ TCID ₅₀)
Cell viability/media sterility control	NA		no virus detected, cells viable; media sterile		
Virus Stock Titer Control			7.75	-	-
Theoretical load ^a					7.05
STR-Solution	1	1 hour	5.25	0.2	4.55
	2		5.00	0.2	4.30
	3		4.50	0.2	3.80
	1	2 hours	4.00	0.2	3.30
	2		4.00	0.2	3.30
	3		3.75	0.2	3.05
	1	4 hours	3.00	0.2	2.30
	2		2.30	0.2	1.60
	3		2.75	0.2	2.05
Initial Plate Recovery Control (T = 0 hours)	1	0 hours	7.00	0.2	6.30
	2		7.25	0.2	6.55
	3		7.25	0.2	6.55
	Average			6.48	
Final Plate Recovery Control (T = 4 hours)	1	4 hours	5.75	0.2	5.05
	2		6.00	0.2	5.30
	3		6.50	0.2	5.80
	Average			5.50	

^a The theoretical load is determined based on the Virus Stock Titer control and the volume of virus challenged per carrier.

NA = Not applicable

RESULTS (continued):

**Table 2
Cytotoxicity Controls**

Dilution of the Neutralized Sample	Cytotoxicity Control
10 ⁻²	no cytotoxicity observed in 4 out of 4 wells
10 ⁻³	no cytotoxicity observed in 4 out of 4 wells
10 ⁻⁴	no cytotoxicity observed in 4 out of 4 wells

**Table 3
Viral Reduction**

Test Agent	Contact Time	Replicate Number	Initial Viral Load* (Log ₁₀ TCID ₅₀)	Output Viral Load (Log ₁₀ TCID ₅₀)	Log ₁₀ Reduction	Percent Reduction
STR-Solution	1 hour	1	6.48	4.55	1.93	98.829%
		2		4.30	2.18	99.342%
		3		3.80	2.68	99.792%
		Mean Reduction				2.38
	2 hours	1	6.48	3.30	3.18	99.934%
		2		3.30	3.18	99.934%
		3		3.05	3.43	99.963%
		Mean Reduction				3.28
	4 hours	1	6.48	2.30	4.18	99.993%
		2		1.60	4.88	99.999%
		3		2.05	4.43	99.996%
		Mean Reduction				4.59

* Results represent the average of three replicates.

Conclusions/Observations:

When tested as described, the AHPCO device exhibited a 2.38, 3.28 and 4.59 Log₁₀ (99.582%, 99.948%, 99.997%) reduction when Middle East Respiratory Syndrome Coronavirus (MERS-CoV), containing 5% serum, was exposed to the test device for 1 hour, 2 hours and 4 hours respectively at 20°C. All of the controls met the criteria for a valid test. These conclusions are based on observed data.