





## Vaccinia virus (VR-1549) Elstree strain Test Results

EN14476:2013 + A2:2019 Suspension test for the efficacy of Xtra Protect, BT-AQA-02FT(2) from Aqua Air against Vaccinia virus VR-1549 under Dirty conditions						
Test Results						
Concentration	1.0% (v/v)		4.0% (v/v)		8.0% (v/v)	
Exposure Time	data	TCID <sub>50</sub> /ml	data	TCID <sub>50</sub> /ml	data	TCID <sub>50</sub> /ml
t = 5 mins	2.67	1.48E+04	0.00	3.16E+01	2.00	3.16E+03
Raw Data	664000	1.48E+04	000000	3.16E+01	660000	3.16E+03
log		4.17		1.50		3.50
log difference		1.50		4.17		2.17

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Summary Table									
Product:	Interfering substance	Concentration	Level of cytotoxicity	lg TCID <sub>50</sub>					>4 lg reduction after 'X' Min
				0 min	5 min	15 min	30 min	60 min	
Xtra Protect	3.0g/l BSA + 3.0ml/l erythrocytes	8.0% (v/v)	3.50	3.50	3.50	n.a.	n.a.	n.a.	>5 mins
		4.0% (v/v)	3.50	n.a.	1.50	n.a.	n.a.	n.a.	<5 mins
		1.0% (v/v)	3.50	n.a.	4.17	n.a.	n.a.	n.a.	>5 mins
	3.0g/l BSA	8.0% (v/v)	3.50	n.a.	3.50	n.a.	n.a.	n.a.	>5 mins
		4.0% (v/v)	3.50	n.a.	1.50	n.a.	n.a.	n.a.	<5 mins
		1.0% (v/v)	3.50	n.a.	2.50	n.a.	n.a.	n.a.	>5 mins
Virus Control	DIRTY			5.67	5.67	5.50	n.a.	n.a.	n.a.
Virus Control	CLEAN			5.83	5.83	5.67	n.a.	n.a.	n.a.
							5 min	15 min	
Formaldehyde	PBS	0.7% (w/v)	3.50				3.67	3.50	>60 mins

***Vaccinia virus (VR-1549) Elstree strain Control Data***

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Controls												
Virus Recovery 0 min		Virus Recovery 5 mins		Virus Recovery 15 mins		Cytotoxicity		Disinfectant Suppression VS		Disinfectant Suppression VS2		
raw data	TCID <sub>50</sub> /ml	raw data	TCID <sub>50</sub> /ml	raw data	TCID <sub>50</sub> /ml	raw data	TCID <sub>50</sub> /ml	raw data	TCID <sub>50</sub> /ml	raw data	TCID <sub>50</sub> /ml	
4.17	4.68E+05	4.17	4.68E+05	4.00	3.16E+05	2.00	3.16E+03	2.00	3.16E+03	4.67	1.48E+06	
666610	4.68E+05	666610	4.68E+05	666600	3.16E+05	660000	3.16E+03	660000	3.16E+03	666640	1.48E+06	
	5.67		5.67		5.50		3.50		3.50		6.17	
									2.17		-0.50	
Formaldehyde reference inactivation controls												
Cytotoxicity		Exposure time	0.7% Formaldehyde				No column Control					
			5 mins		15 mins		5 mins					
raw data	TCID <sub>50</sub> /ml		raw data	TCID <sub>50</sub> /ml	raw data	TCID <sub>50</sub> /ml	raw data	TCID <sub>50</sub> /ml				
2.00	3.16E+03		2.17	4.68E+03	2.00	3.16E+03			4.67	1.48E+06		
660000	3.16E+03		661000	4.68E+03	660000	3.16E+03			666640	1.48E+06		
	3.50	log		3.67		3.50				6.17		
		log difference		1.83		2.00						
Interference control		Virus dilution						Stock Virus (TCID <sub>50</sub> )				
		-3	-4	-5	-6	-7	-8	5.67				
PBS Control		1	1	1	0.67	0	0	6.67E+09				
		3.16E+02	3.16E+02	3.16E+02	1.48E+02	3.16E+01	3.16E+01	6666631000				
Raw Data		2.50	2.50	2.50	2.17	1.50	1.50					
		6	6	6	4	0	0					
Product		1	1	1	0.67	0	0					
		3.16E+02	3.16E+02	3.16E+02	1.48E+02	3.16E+01	3.16E+01					
Raw Data		2.50	2.50	2.50	2.17	1.50	1.50					
		6	6	6	4	0	0					
Log Difference		0.00	0.00	0.00	0.00	0.00	0.00					
Product Cyt Dilution		-3	-3	-3	-3	-3	-3					
PBS Dilution		Neat	Neat	Neat	Neat	Neat	Neat					

**Vaccinia virus (VR-1549) Elstree strain Control Data**

Parallel Control Test												
Controls						Test Results						
Virus Recovery 0 min		Virus Recovery 5 mins		Virus Recovery 15 mins		Concentration	1.0% (v/v)		4.0% (v/v)		8.0% (v/v)	
raw data	TCID <sub>50</sub> /ml	raw data	TCID <sub>50</sub> /ml	raw data	TCID <sub>50</sub> /ml	Exposure Time	data	TCID <sub>50</sub> /ml	data	TCID <sub>50</sub> /ml	data	TCID <sub>50</sub> /ml
4.33	6.76E+05	4.33	6.76E+05	4.17	4.68E+05	t = 5 mins	1.00	3.16E+02	0.00	3.16E+01	2.00	3.16E+03
666620	6.76E+05	666620	6.76E+05	666610	4.68E+05	Raw data	600000	3.16E+02	000000	3.16E+01	660000	3.16E+03
	5.83		5.83		5.67	log		2.50		1.50		3.50
						log difference		3.33		4.33		2.33

## CONCLUSION

### Verification of the methodology

A test is only valid if the following criteria are fulfilled:

- a) The titre of the test suspension of at least  $10^8$  TCID<sub>50</sub> /ml is sufficiently high to at least enable a titre reduction of 4 lg to verify the method.
- b) Detectable titre reduction is at least 4 log<sub>10</sub>.
- c) Difference of the logarithmic titre of the virus control minus the logarithmic titre of the test virus in the reference inactivation test is between:
  - Between 0.5 and 2.5 after 30 min and between 2.0 and 4.5 after 60 min for poliovirus
  - Between 3.0 and 5.0 after 30 min and between 3.5 and 5.5 after 60 min for adenovirus
  - Between 1.0 and 3.0 after 30 min and between 2.0 and 4.0 after 60 min for murine norovirus
  - Between 0.0 and 2.0 after 30 min and between 0.5 and 2.5 after 60 min for parvovirus
  - Between 0.75 and 3.5 after 5 min and between 2.0 and 4.0 after 15 min for Vaccinia virus
- d) Cytotoxicity of the product solution does not affect cell morphology and growth or susceptibility for the test virus in the dilutions of the test mixtures which are necessary to demonstrate a 4 log<sub>10</sub> reduction of the virus.
- e) The interference control result does not show a difference of < 1.0 log<sub>10</sub> of virus titre for test product treated cells in comparison to the non-treated cells.
- e) Neutralisation validation. This is called the disinfectant suppression test in this protocol. The disinfectant was neutralised by column chromatography through an Illustra Microspin S-400 HR column to achieve the best possible neutralisation available for this test. The difference for virus is greater than 0.5 log<sub>10</sub> indicating rapid irreversible virucidal activity of the disinfectant by dilution at a concentration of 8.0% v/v for VS1. This neutralisation validation has been verified by VS2, which shows the product has been successfully neutralised.

According to EN 14476:2013 + A2:2019, **XtraProtect POSSESSES VIRUCIDAL** activity at a concentration of **4.0% v/v** of the working concentration as tested after **5 MINUTES** at **20°C** under **DIRTY** conditions (3.0 g/l bovine albumin + 3.0 ml/l erythrocytes) against *Vaccinia virus* VR-1549 Elstree strain / Vero cells.

The cytotoxicity of the product has prevented at 4.0 log reduction being observed at 8.0% v/v.

**This product therefore is effective against all enveloped viruses as defined in EN 14476:2013 + A2:2019. This therefore includes all coronaviruses and SARS-CoV-2.**

Signed



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### DISCLAIMER

The results in this test report only pertain to the sample supplied.

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