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SARS-CoV-2 inactivation testing: interim report

Report identifier	HCM/CoV2/005/v3
Report date	15 June 2020
Undertaken by High Containment Microbiology, NIS Laboratories, National Infection Service, Public Health England N.B. This is an interim report and may be updated as further results are obtained	

Product/treatment details	
Product/treatment	Tween® 20 (synonym: polysorbate 20)
Concentration	0.1% (v/v) 0.5% (v/v)

Sample details	
Sample type tested	Tissue culture fluid containing 5% (v/v) foetal calf serum
Virus strain tested	SARS-CoV-2 England 2
Ratio of spiked virus stock to sample matrix	Not applicable; tissue culture fluid used undiluted

Experimental conditions	
Contact time	30 minutes
Temperature of incubation	Room temperature
Brief description of tests performed	Triplicate samples were treated with test buffer for indicated contact time/s or mock-treated in triplicate with an equivalent volume of PBS. All samples were then subjected to a purification step to remove cytotoxic buffer components. PBS-treated samples were subjected to the same purification procedure in parallel. **Test 1:** Purified samples were immediately titrated on Vero E6 cells to establish virus titre. This test is quantitative and reports the titre of virus in each treatment condition in TCID50 per ml. Reduction in virus titre following treatment is given as the difference between the mean log_{10} TCID50/ml for treated conditions and the PBS control. **Test 2:** In parallel, purified samples were seeded onto Vero E6 monolayers to amplify any remaining virus over the course of up to four serial passages. Virus amplification over each passage was detected by visual (microscopic) examination of monolayers for cytopathic effect, and confirmed by SARS-CoV-2-specific real-time PCR. This test is qualitative and reports either the presence or absence of virus amplification. This test may detect levels of virus that are below the detection limit of the titration assay (Test 1) due to a greater sample plating volume and the opportunity for any virus present to amplify over serial passages.

Table of results			
Maximum detectable virus reduction in test (log_{10} TCID50/ml)			5.2
	Test 1: Virus titration post-treatment		**Test 2: Passage of samples in cell culture**
	Mean virus titre (log_{10} TCID50/ml)	Titre reduction (log_{10} TCID50/ml)	Virus detected/ Virus not detected
PBS-treated	5.9	-	Virus detected (all replicates)
0.1% Tween® 20-treated	6.0	None detected	Virus detected (all replicates)
0.5% Tween® 20-treated	5.9	None detected	Virus detected (all replicates)

Interpretation

Test 1: No reduction in virus titre was detected with either treatment condition.

Test 2: Infectious virus was recoverable from all sample treatment conditions.

These data do not support use of 0.1% or 0.5% Tween® 20 for SARS-CoV-2 inactivation for treatment times of 30 minutes or less.

These tests have been performed on tissue culture fluid containing 5% (v/v) foetal calf serum. The effectiveness of this treatment against SARS-CoV-2 may vary when used to inactivate clinical samples or other types of sample matrix. Any results of inactivation testing using other sample matrices will be released as they become available.

Inactivation reagents should not be assumed to be 100% effective against SARS-CoV-2.

Suitability of products and treatments for inactivation of other pathogens has not been evaluated in this study.

All COVID-19 laboratory testing workflows must be subjected to suitable and sufficient risk assessment, with consideration given to any inactivation step. Risk assessments should be reviewed regularly as new information on the inactivation of SARS-CoV-2 becomes available.

The impact of chosen inactivation method on the sensitivity of subsequent SARS-CoV-2 detection should also be assessed locally.

Disclaimer

PHE's evaluations of commercial products and treatments for inactivating SARS-CoV-2 have been carried out primarily for PHE's own internal use and the reports of such evaluations are shared solely for readers information; PHE does not in any way recommend any particular product for virus inactivation; and PHE shall not be responsible for the choice of product or treatment for virus inactivation, and it is the responsibility of the testing laboratory to ensure that any such product or treatment implemented has undergone the necessary verification and validation; and PHE shall not be liable, to the greatest extent possible under any applicable law, for any claim, loss or damage arising out of or connected with use of this and related reports and choice of virus inactivation products or treatments.

Summary of revisions

Version 1:	New document
Version 2:	Header and disclaimer edited; date issued to PHE's COVID Incident Virology Cell added; key guidance points added to interpretation
Version 3:	Reformatted for publication

Queries regarding this report or HCM inactivation testing should be directed to HCMgroup@phe.gov.uk

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