

SARS-CoV-2 inactivation testing: interim report

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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	Sodium dodecyl sulphate (SDS)
Concentration	0.1% (w/v)
	0.5% (w/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 times	30 minutes			
Temperature of incubation	Room temperature			

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 log₁₀ TCID50/ml for treated conditions and the PBS control.

Brief description of tests performed

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.

Test 3: Treated, unpurified samples were extracted and subjected to SARS-CoV-2 specific real-time PCR, in triplicate for each treatment condition. This test reports Ct values for each treatment condition and indicates the effect on SARS-CoV-2 RNA stability. This test measures levels of viral RNA only and does not reflect virus infectivity.

Table of results								
Maximum detectable TCID50/ml)	6.5							
,		st 1: post-treatment	Test 2: Passage of samples in cell culture	Test 3: Effect on viral RNA				
	Mean virus titre (log ₁₀ TCID50/ml)	Titre reduction (log ₁₀ TCID50/ml)	Virus detected/Virus not detected	Mean Ct (standard error)				
PBS-treated	7.2	-	Virus detected (all replicates)	11.1 (0.2)				
0.1% SDS	1.4	5.7	Virus detected (all replicates)	12.4 (0.2)				
0.5% SDS	≤0.7	≥6.5	Virus detected (2 replicates)	10.4 (0.2)				

Interpretation

Test 1: 0.1% SDS reduced infectious virus titre by 5.7 \log_{10} but virus was still detected in all treated sample replicates. 0.5% SDS reduced virus titre by \geq 6.5 \log_{10} , with virus detected in one replicate. The maximum detectable titre reduction in this test was 6.5 \log_{10} .

Test 2: Infectious virus was recoverable from all samples treated with 0.1% SDS and from 2 sample replicates treated with 0.5% SDS, indicating that inactivation at these concentrations was incomplete.

Demonstrating complete inactivation is dependent on the starting titre of virus used for testing, and it is likely that complete inactivation could be achieved under the above conditions if samples contained lower levels of infectious virus than those tested here. Conversely, sample treatments that inactivate virus effectively in our testing may fail to inactivate samples containing higher levels of virus than those evaluated in this study.

Test 3: We observed minimal effects of SDS treatment on Ct value in SARS-CoV-2 specific real-time PCR (+1.3 Cts for 0.1% SDS and -0.7 Cts for 0.5% SDS for treated samples compared to untreated samples).

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

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Summary of revisions

Version 1: New document

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

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