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Evaluation of virucide activity against HCoV-OC43 of ESI system on a plastic surface

FINAL REPORT

INTRODUCTION

The virus used in this study is the Human Coronavirus HCov-OC43 which has an extremely high homology of structure with the virus responsible for CoViD-19, HCoV-SARS-2, both from a phylogenetic and a molecular point of view. In fact, they both belong to the β-Coronavirus group in an extremely close position of the phylogenetic tree. The homology is such that some highly specific antibodies against HCoV-OC43 also recognize SARS-2. This indicates that proteins, which are the main constituent of the viral particle and determine its resistance, are extremely similar between the two viruses. Since germicidal treatments act with non-specific mechanisms, morphologically similar viruses respond to inactivation in a similar way. Therefore, HCoV-OC43 has been used in several studies focused on viral persistence/inactivation as a substitute for the highly pathogenic Coronaviruses SARS-1, SARS-2 and MERS. In fact, HCoV-OC43 can be more easily manipulated, not requiring a laboratory with a biosafety level of 3 but 2, as the UNIMORE laboratory is.

EXPERIMENTAL PROTOCOL

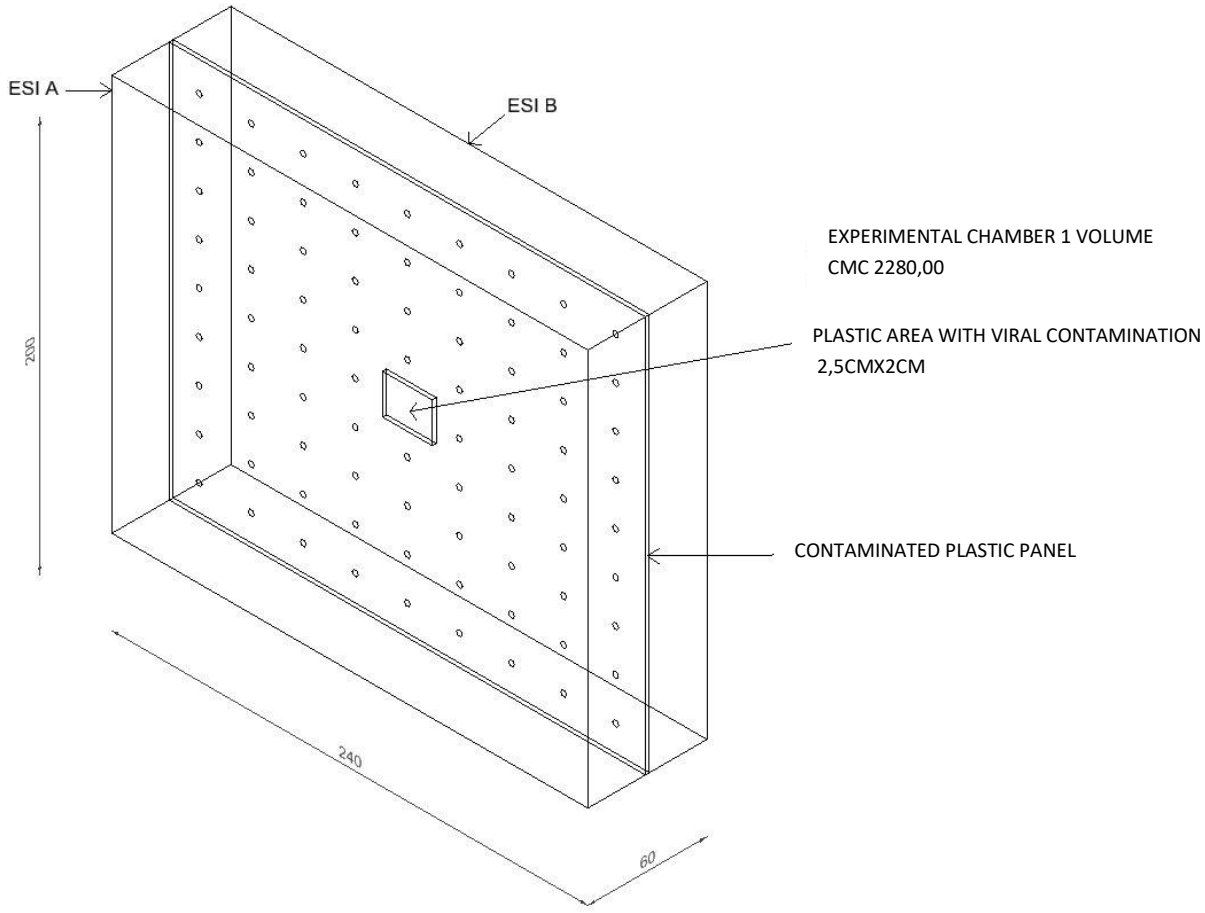
Before each experiment, the plastic panels were sterilized in order to eliminate any microbial contamination that would have interfered with the results by preventing subsequent analyses.

An aliquot of 100µl of HCoV-OC43 viral suspension was deposited on 2,5x2 cm portion of the panels which were left to dry for about 30 minutes

The contaminated panels were placed in an experimental chamber expressly set up in order to keep the panels and the ESI plates in a steady vertical position. Once assembled, the chamber was put in a sterile laminar flow cabinet allowing to work in aseptic conditions. In parallel, similar plastic panels were contaminated in the same way but not exposed to ESI (controls) but left in the sterile cabinet as long as the experimental samples.

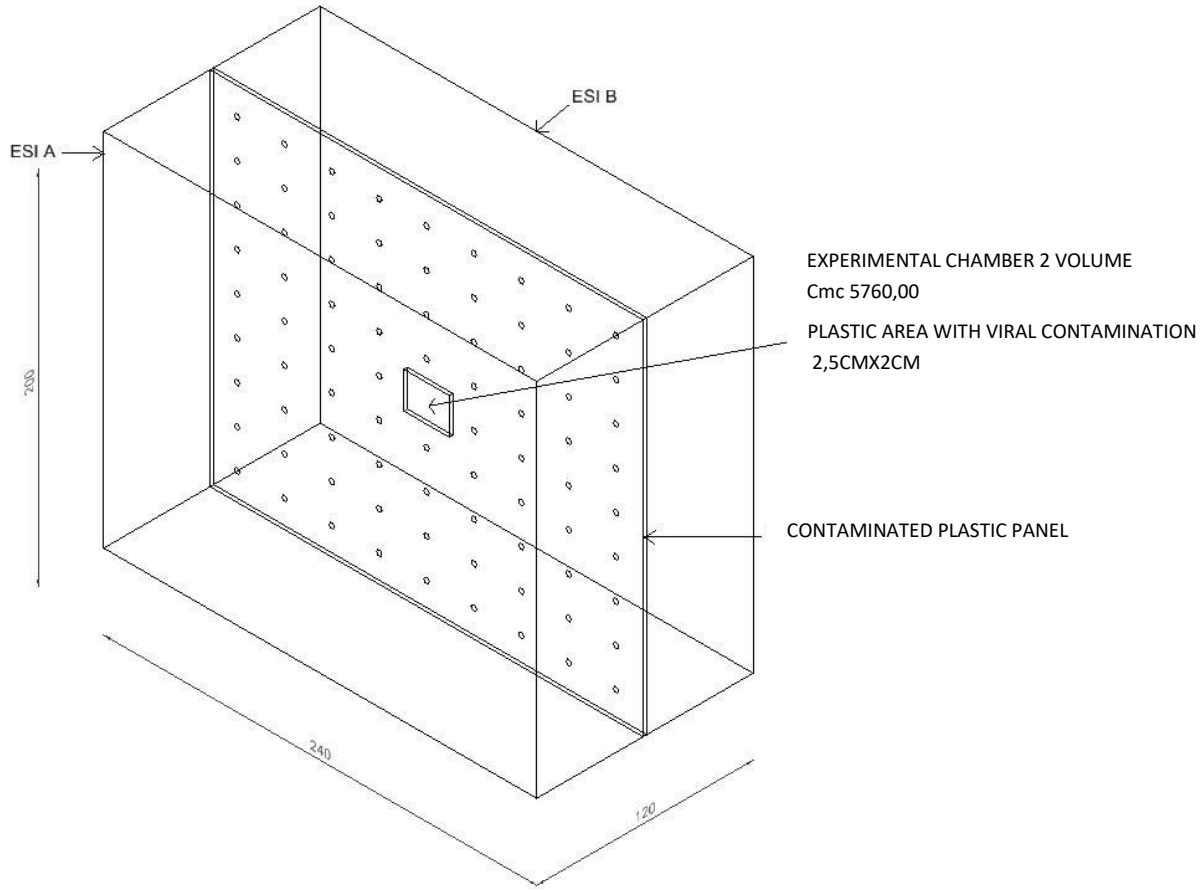
Experiment 1:

In a first set of experiments, the two ESI emitters (Esi "A" + Esi "B") were placed at the two sides of the chamber which had dimension of 240x200x60 mm, determining an air total volume of 2.280 cm^3. The contaminated plastic panel was set in the middle of the chamber with the contaminated side facing ESI "A".



Experiment 2:

In a second set of experiments the two ESI emitters were placed at the two sides of the chamber which had dimension of 240x200x120 mm, determining an air total volume of 5.760 cm^3. The contaminated plastic panel was set in the middle of the chamber with the contaminated side facing ESI "A".



In both cases, 3 time points were tested: 15", 30', 2h. At the end of the treatment, residual virus on the panel was rescued and titrated by the end point titration assay.

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RESULTS

The results obtained are shown in the table which reports the titre of the virus rescued from the treated and untreated (Ctrl) samples. Titres are expressed as TCID50. The reduction, calculated with respect to the untreated control, is expressed both as Log and as percentage. The results are the mean of 2 experiments in which each sample was tested in duplicate.

Table

Plastic		Ctrl	Treated		
			15''	30'	2h
2.280 cm3	Mean of the viral load values ($TCID_{50}$)	10^3	$10^{2,25}$	$10^{2,25}$	$10^{0,75}$
	Reduction expressed as Log		0,75	0,75	2,25
	Recudction expressed as %		82,2	82,2	99,4
5.760 cm3	Mean of the viral load values ($TCID_{50}$)	$10^{2,5}$	$10^{2,25}$	$10^{1,75}$	$10^{0,75}$
	Reduction expressed as Log		0,25	0,75	1,25
	Recudction expressed as %		43,8	82,2	94,4

COMMENTS

The ESI system assayed for virucidal activity against the Coronavirus HCoV-OC43 on a plastic surface proved effective on a contaminated plastic panel set between the 2 ESI

emitters in 2 different experimental chambers, the one with a volume of 2.280 cm^3 and the other with a volume 5.760 cm^3, for 3 time points (15", 30', 2h). The highest activity was observed when the emitters were placed in the experimental chamber 1 with a volume of 2.280 cm3 for a 2h treatment: in this case the viral load reduction was 2,25 Log (99.4%). With the experimental chamber 2 (volume 5.760 cm^3) the virus load reduction was 1,25 Log, 82,2%. At the other conditions, the virucide activity was modest (< 1 Log), but always higher in chamber 1 than in chamber 2.

Therefore, we can assume that a robust reduction can be achieved only with a long exposition (12h-24h). Duration rather than distance seems to be the most important factor influencing the virucide activity. Moreover, also the geometry of the chamber allocating the plastic panel and the emitters may influence our results creating alterations in the ionic emission. Further investigations will be carried out with longer treatments and using a different chamber.

The Research Supervisor

Prof. Claudio Cermelli

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