Universidad de Oviedo
Universidá d'Uviéu
University of Oviedo

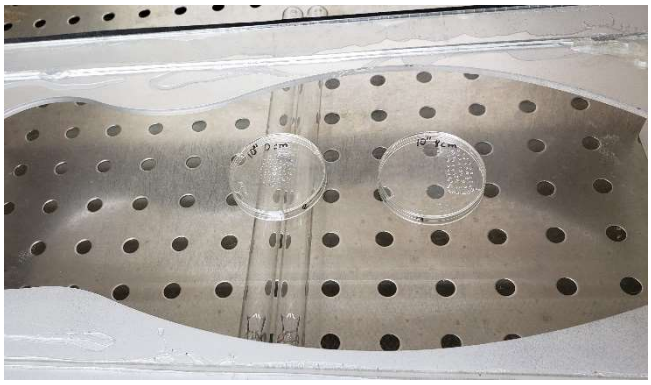
RESULTS REPORT (FUO-328-20)

STUDY OF THE VIRUCIDAL ACTIVITY AGAINST A HUMAN CORONAVIRUS OF A COMMERCIAL PROTOTYPE OF SHOES DECONTAMINATING APPARATUS BASED ON ULTRAVIOLET TYPE C LIGHT

Device: Commercial prototype of decontamination with type C UV- lamp.

Virus and cells: A recombinant 229E human coronavirus expressing fluorescent green protein (HCoV-229E/GFP) propagated in the Huh-7 human hepatoma cell line was used.

Test procedure: For each experimental condition, 10 µL of a coronavirus preparation with an approximate titre of 10^6 $TCID_{50}$/mL were used and seeded in 50-60 small droplets distributed similarly over the inside of a Petri dish 4 cm in diameter. The dishes were then reversed and placed side down, with the drops containing the virus facing the lamp in the corresponding test positions, as shown in the image below:



3 exposure times to ultraviolet light were tested: 10, 20 and 30 seconds. **Two different samples** located 0 and 8 cm away from the lamp were used for each exposure time.

In parallel, 10 µL of the viral preparation was placed in a similar way on a Petri dish that was not exposed to ultraviolet light, to be used as virus control.

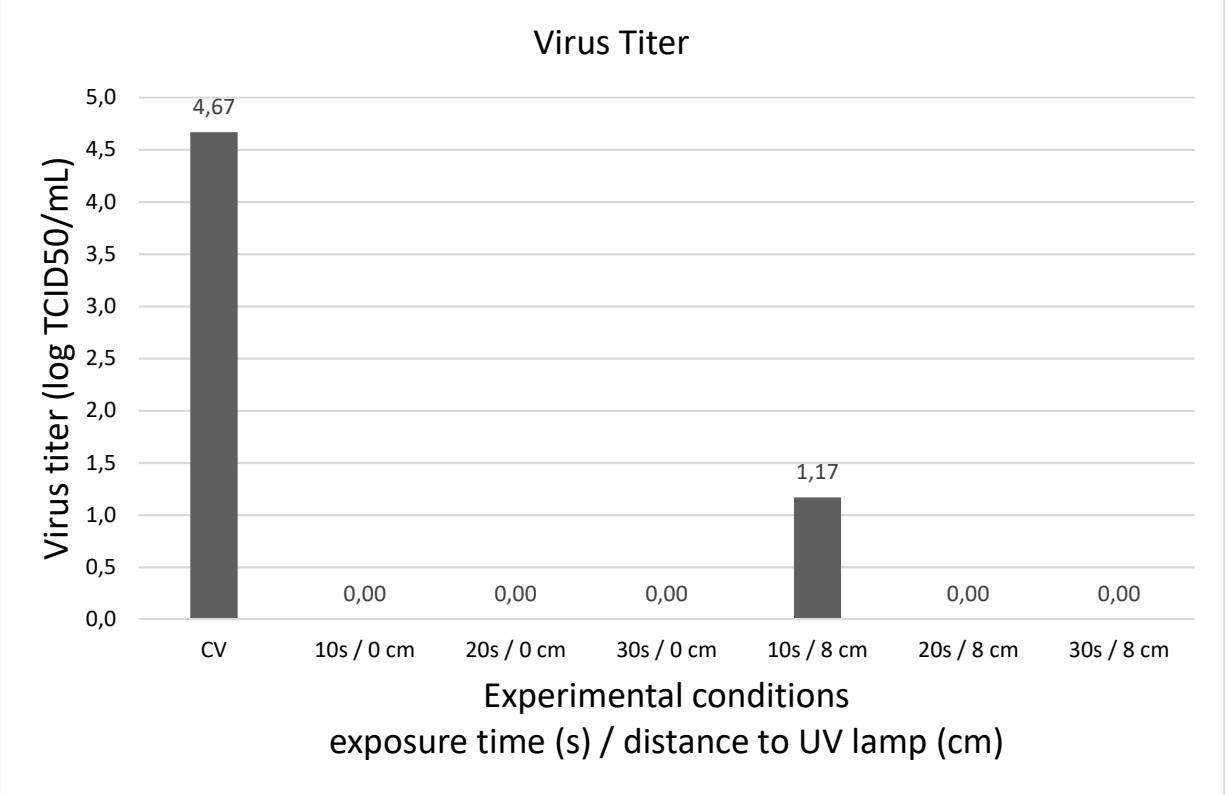
After exposure of the virus under different conditions, the plates were flipped again and 500 µL of DMEM culture medium supplemented with 2% fetal bovine serum were added over the virus droplets, mixing by pipetting to collect the virus. A similar procedure was used for the virus control.

The viral suspensions obtained from both virus control and the various treatments were titred by the Reed and Muench method of endpoint dilution, to determine residual infectivity (TCID50/mL) after treatments with respect to untreated virus control. The assessments were performed on 96 well culture plates seeded with confluent Huh-7 cells. For the assessment of the outcome of infections, cultures that had at least one green fluorescent cell, indicative of recombinant coronavirus infection, were considered positive.

RESULTS

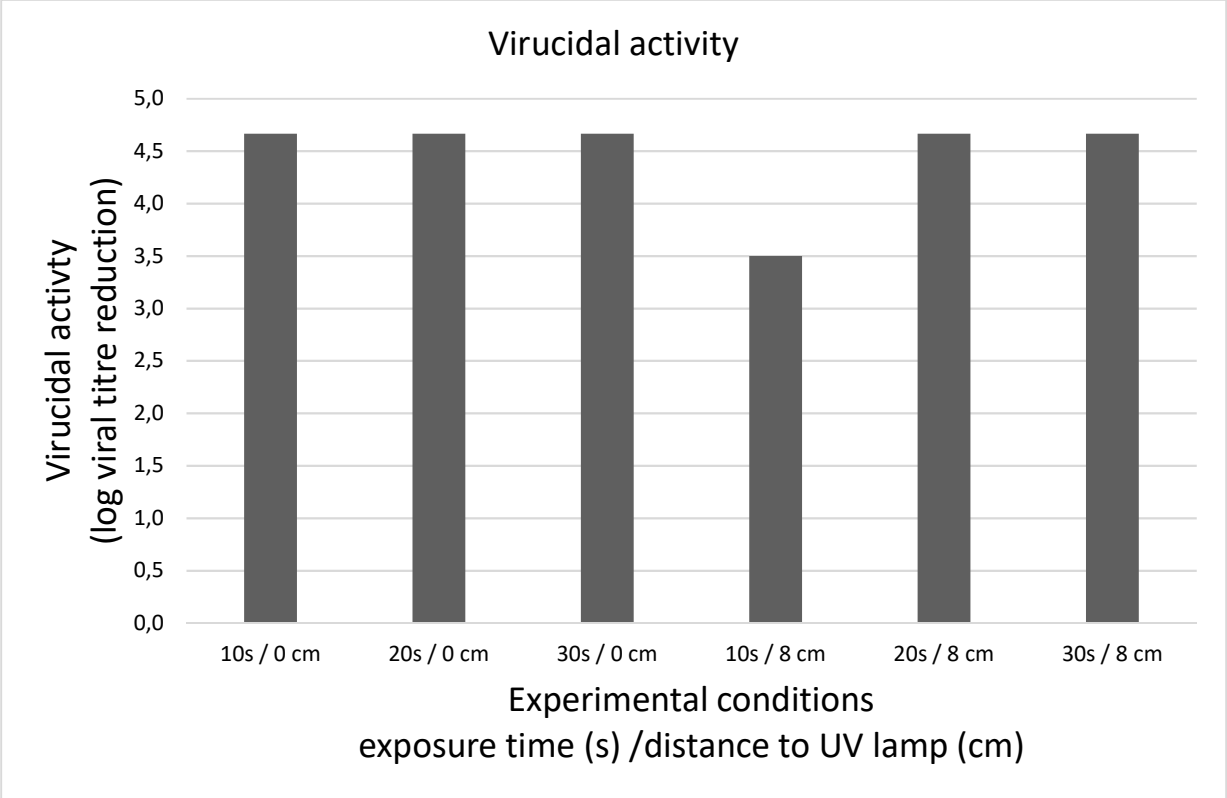
Green infected cells were observed in virus-control wells up to dilution 10^{-4}, while in most UV treatments no green fluorescence, indicative of viral replication, was observed even in the lowest dilution (10^{-1}). Only positive wells were observed in 4 of the 6 experimental replicas of dilution 10^{-1} for the mildest treatment condition (shorter exposure time and longer lamp distance, 10s / 8 cm). However, as can be seen in the graphs of viral titre and virucidal activity, even in this treatment the reduction of viral titre was more than 3 logarithm units, indicative of a virucidal activity of 99.9%.

The graphs with the values of infective titres obtained from untreated virus controls (CV) and the different time and distance (t/d) treatments are shown below. The infective titre turned out to be null for most exposure conditions, except the one mentioned above of 10s / 8cm.



The virucidal activity results from subtracting the logarithm from the infective titre of virus control (CV) and the logarithm of viral titres after the various exposures. In all cases this reduction was always greater than 3, a value considered to be 99.9% inactivation of viral particles.

Universidad de Oviedo
Universidá d'Uviéu
University of Oviedo



CONCLUSION

The tested shoe-decontaminating device had a **virucidal activity of more than 99.9%** under all tested conditions, from a minimum exposure time of 10 seconds to a distance of 8 cm between the virus sample and the UV light emitting source.

Oviedo a 25 November 2020

PARRA FERNANDEZ JOSE FRANCISCO - 71612214N

Firmado digitalmente por PARRA FERNANDEZ JOSE FRANCISCO - 71612214N
Nombre de reconocimiento (DN): c=ES, serialNumber=IDCES-71612214N, givenName=JOSE FRANCISCO, sn=PARRA FERNANDEZ, cn=PARRA FERNANDEZ JOSE FRANCISCO - 71612214N
Fecha: 2020.11.25 17:04:49 +01'00'

Signed:. Dr. Francisco Parra Fernández

ÁLVAREZ RODRÍGUEZ, ÁNGEL LUIS (FIRMA)
2020.11.25 15:40:49 +01'00'

Dr. Ángel Luís Álvarez Rodríguez