FINAL REPORT

Evaluation of the virucidal activity of the Urban Air Purifier (UAP400) system against SARS-CoV-2

Test Facility

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IRTA-CReSA study number: SA-11060-20

Version: Final
Date: 18th January 2021

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1. STUDY DATES

EXPERIMENTAL PHASE: December 2020

2. OBJECTIVE

To test the virucidal efficacy of the Urban Air Purifier system against SARS-CoV-2. On the one hand, UV-C light system was evaluated and on the other hand the air flow capabilities regarding inactivation was also tested.

3. MATERIALS AND METHODS

3.1 Test Item

3.1.1 Urban Air Purifier (UAP 400)

An UAP400 was received at IRTA-CReSA facilities before the experiment. The system consisted of an air purifier with a combination of filters (PPI, M6, and HEPA 14) and two UV-C lights. The air flow was set up at 50% during the whole experiment.

3.2 Test System

3.2.1 Virus

Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) with ID EPI_ISL_510689 (isolated from a nasopharyngeal swab collected from an 89-year-old male patient) was used. The virus was propagated, and a virus stock was prepared collecting the supernatant culture fluid from Vero E6 cells. On the day of use the appropriate number of aliquots were removed, thawed, and maintained at a refrigerated temperature until used in the assay.

3.2.2 Cell Culture and Test Culture Media

Vero E6 cells (ATCC CRL-1586) were cultured in Dulbecco's modified Eagle medium, supplemented with 5% fetal calf serum (FCS), 100 U/mL penicillin, 100 µg/mL streptomycin, and 2 mM glutamine.

3.3 Test Method

3.3.1 Testing Schedule

1) SARS-CoV-2 on cellulose paper + UV-C (before HEPA filter): replicate 1; 2 hours
2) SARS-CoV-2 on cellulose paper + UV-C (before HEPA filter): replicate 2; 2 hours
3) SARS-CoV-2 on cellulose paper + UV-C (before HEPA filter): replicate 3; 2 hours
4) SARS-CoV-2 on cellulose paper (no UV-C, after HEPA filter): replicate 1; 2 hours
5) SARS-CoV-2 on cellulose paper (no UV-C, after HEPA filter): replicate 2; 2 hours
6) SARS-CoV-2 on cellulose paper (no UV-C, after HEPA filter): replicate 3; 2 hours
7) SARS-CoV-2 on cellulose paper (no UAP treatment): replicate 1; 2 hours
8) SARS-CoV-2 on cellulose paper (no UAP treatment): replicate 1; 2 hours
9) SARS-CoV-2 on cellulose paper (no UAP treatment): replicate 1; 2 hours
10) SARS-CoV-2 inoculum (no UAP treatment): 1 replicate; 2 hours

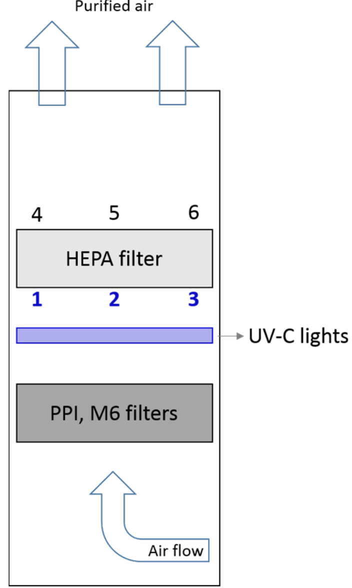

Figure 1. Scheme of the sample locations inside the UAP400 system. Samples 1, 2 and 3 before HEPA filter, under UV-C exposition. Samples 4, 5 and 6 above the HEPA filter in order to measure the effect of the air flow (no UV-C exposition).

3.3.2 Preparation of experimental items

A total of 100 µL of SARS-CoV-2 at 10^6 $TCID_{50}$/mL was spiked onto sterilized cellulose paper pieces per each replicate. A total of 10 paper pieces spiked with virus were prepared under a Biosafety Cabinet. This procedure was performed in the CR/1035 Laboratory at CReSA High Biocontainment Unit using the appropriate PPE (double glove, double gown and PAPR Sundström equipment).

3.3.3 Virucidal assay

After the virus was absorbed by the cellulose paper pieces, 3 samples (1,2,3) were located immediately after the UV-C lamps, just before the HEPA filter, one on the left side, one on the middle and the other on the right side, respectively. The same distribution but upstream the HEPA filter was used for 3 other samples (4,5,6) to evaluate the air flow inactivation capabilities (with no concurrent UV-C light action). Non-exposed samples (7,8,9) were left under the flow of the Biosafety Cabinet and were used as controls.

Once the samples were placed inside the system, the UAP-400 was turned on at 50% of its air flow capcity.

After exposure time (2 hours), the cellulose papers pieces were recovered and desiccated viruses were gently eluted with 1 mL of media (DMEM, 1% FBS). All samples were individually stored at -75°C until virus titration assay.

3.3.4 Sample titration

The day before titration, 96-well plates were prepared with Vero E6 cells. The day of titration, serial ten-fold dilutions of the samples were performed and added to Vero E6 cells. Additionally, a bulk of approximately 50% of the recovered eluate was also titrated. Microplates were incubated at 37°C and 5% CO_2 for 6-7 days and afterwards, cytopathic effect was determined under light inverted microscope.

4. RESULTS

Mean of the assayed triplicates was calculated per each sample and is shown in **Table 1**. No virus was detected in any of the samples (1,2,3) exposed to UV-C light, at least at the detection limit of the technique (0.45 $TCID_{50}$/mL). Taking into account the bulk titration (50% of the final eluate), inactivation reached ≥ 98.55% for all replicates at 2 hours of UV-C exposition.

Air flux samples located after the HEPA filter (4,5,6) showed partial inactivation. Samples on both sides (4 left and 6 right) showed less inactivation than sample number 5 (at the center), 11% vs 77%, respectively.

Table 1. SARS-CoV-2 titration on VERO E6 cells expressed in mean (±standard deviation) of $TCID_{50}$/mL and percentage of inactivation (%) according to the mean of control virus (non-UV exposed SARS-CoV-2 on cellulose paper) after 2 hours.

Samples	$TCID_{50}$/mL	logR	% Inactivation
UV-C exposed samples (1,2,3)	**≤0.45 ± 0.00**	**≥1.84 ± 0.44**	**≥ 98.55**
Air flow samples (4,5,6)	2.06 ± 0.36	0.23 ± 0.40	29.5 ± 41.7
Desiccated Control Virus (7,8,9)	2.29 ± 0.44	-	-
Inoculum (10)	5.24 ± 0.26	-	-

5. CONCLUSIONS

Regarding the evaluation of the UV-C light system (samples 1, 2 and 3), this assay allowed to show an inactivation of SARS-CoV-2 at least until the detection limit of the technique with a logR of ≥ 1.84, which means a inactivation rate of ≥ 98.55%. This indicates that potential virus reaching the HEPA filter just upstream UV-C lamps will be totally inactivated by UV-C exposition, in our experimental design.

Regarding the evaluation of the air flow (with no consideration of the UV-C inactivation system, samples 4, 5 and 6), the results suggest that the air flow might be not sufficient to inactivate potential virus with no UV-C light treatment. However, in a realistic situation, potential virus would be retained by the HEPA filter, desiccated. Thus, the obtained results also suggest that air flow inside the UAP400 is not lineal and inactivation might be more effective at the center of the air flow, although no statistical significant differences were found.