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### <u>Test report</u>

Date: Dec. 16th 2020.

#### Assignment:

Assessment of the effect of UV BENCH against SARS-CoV-2 viability

#### Attendees:

Assoc. Prof. PhD Thomas Emil Andersen, Research Assistant BSc Ditte Sandfelt Tornby, Biomedical Scientist MSc Line Bang.

#### Time period for test work:

Dec.10th-15th 2020.

#### Task description:

Single measurements of the viability of SARS-CoV-2 after 5 and 10 seconds of exposure to UVC in the center of the UV BENCH.

**Day one:** Culturing of host cells (VERO C1008 [Vero 76, clone E6, Vero E6] (ATCC® CRL-1586<sup>™</sup>).

**Day two:** Preparation of experiment, irradiation of test specimens, infection of VERO E6 cells and establishment of plaque assay.

Day five: Termination of plaque assay, collection of data.

#### Protocol:

SARS-CoV-2 viral freezer stock is diluted x33 i prewarmed (37°C) DMEM + 2% FBS (+Amp. B og PenStrep) and 120µL transferred to sterile quartz cuvette (Figure 1-3).

The quartz cuvette is closed with lid and sealed with para film, followed by exposure to UV light in the UV BENCH for 10 and 20 seconds.  $100\mu$ L treated viral suspension is transferred to an Eppendorf tube and mixed with  $200\mu$ L DMEM +2% FBS to achieve a total of  $10^2$  dilution relative to the stock. The  $300\mu$ L is transferred to the cell culture and infection allowed for a one-hour time period while placed on a tipping table at  $37^{\circ}$ C/5% CO<sub>2</sub>.

#### Results, UV BENCH: surviving virions relative to total treated virions:

Time period of exposure	10 seconds	20 seconds
Surviving virions	0/6 x 10 <sup>3</sup>	0/6 x 10 <sup>3</sup>

Conclusion: both exposure time periods inactivate virions to below the detection level, i.e. a >log3 or >99,9% reduction in active virions.

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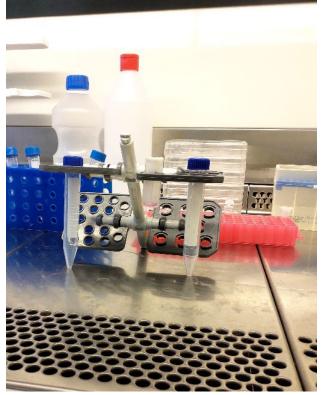


Figure 1. Experimental setup.



Figure 2. Experimental setup.

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Figure 3. Experimental setup.

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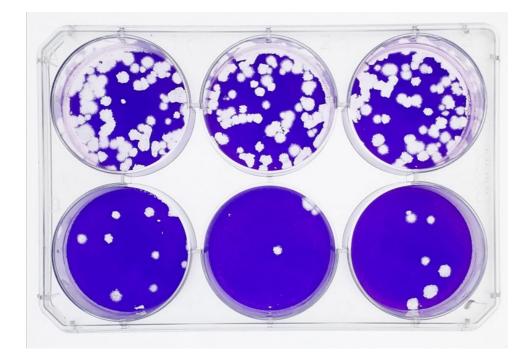
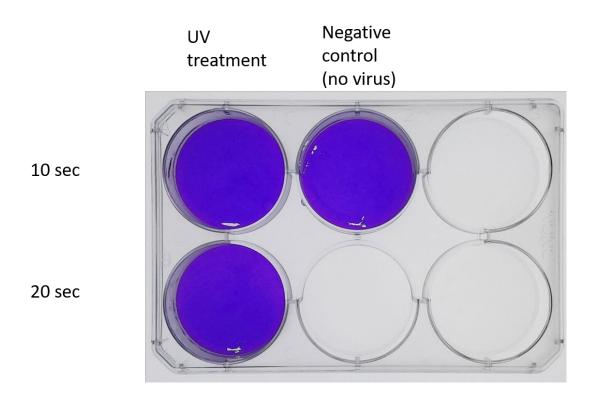


Figure 4. Plaque assay of untreated viral stock cultured in triplicates at  $10^4$  dilution (top row) and  $10^5$  dilution (bottom row). 300ul stock solution is added per well.

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Figur 5. Plaque assay conducted with viral stock suspension exposed to UV treatment (UV BENCH). The suspension was diluted  $10^2$  corresponding to 6 x  $10^3$  pfu/300 µL/well. No plaques could be detected.

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