

## REPORT BY THE DANISH TECHNOLOGICAL INSTITUTE

The following report is written by Stig Koust Hansen, Ph.D, Consultant at the Danish Technological Institute (DTI) about Rensair air cleaning technology. The DTI is a leading research and technology institute with 70 laboratories and 1,000 specialists. It works in close consultation with 800 research and development partners.

The objective of the test was to determine the Rensair unit's efficacy in reducing the concentration of active aerosolized MS2 bacteriophages (used as a proxy for SARS-CoV-2), deploying a modified ISO 16000-36:2018 method. The result was a particle reduction rate of 99.98% in 15 minutes and above 99.99 % in 30 minutes.

Kind regards



Christian Hendriksen  
Co-Founder and CEO  
Rensair





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Inactivation of aerosolized viruses: MS2 bacteriophages

**Rensair**



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# Inactivation of aerosolized viruses: MS2 bacteriophages

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## Assignment Description

The purpose of the test is to determine the efficacy of the air purifier to reduce the concentration of active aerosolized MS2 bacteriophages using a modified ISO 16000-36:2018 method. The tested air purifier is the filtration unit Rensair (HEPA-filter and UV-C photolysis device).

MS2 bacteriophages is chosen as virus surrogate as this is a recognized RNA and non-enveloped model virus, that furthermore is robust enough to survive aerosolization and air sampling.

## Conclusion

The reduction rate at 15 minutes is 99.98 % and above 99.99 % at 30 minutes. The reduction rates are calculated as described in ISO 16000-36:2018 section 8.3.

## Method and Materials

The purpose of this test is to determine the air purifier's ability to remove MS2 bacteriophages aerosolized in a test chamber. The natural decay rate of the concentration of active aerosolized MS2 is determined by sampling the air in the chamber over a 30-minute period and the enhanced decay rate due to the air purifier is determined in a similar manner.

The volume of the used chamber is 20 m<sup>3</sup> and it has an inert FEP lining for chemical resistance and easy cleaning. The room is airtight, and a fan is in the room to mix the air and secure a homogenous concentration of aerosols. The aerosol is generated within the test chamber using a nebulizer (Palas AGK 2000). The air purifier is placed on the floor in the center of the room. See the setup in Figure 1.

The room is cleaned thoroughly and heavily ventilated using clean air prior to the test.

The relative humidity in the test chamber during testing was  $50 \pm 10$  %RH and the temperature was  $21 \pm 0.5$  °C.

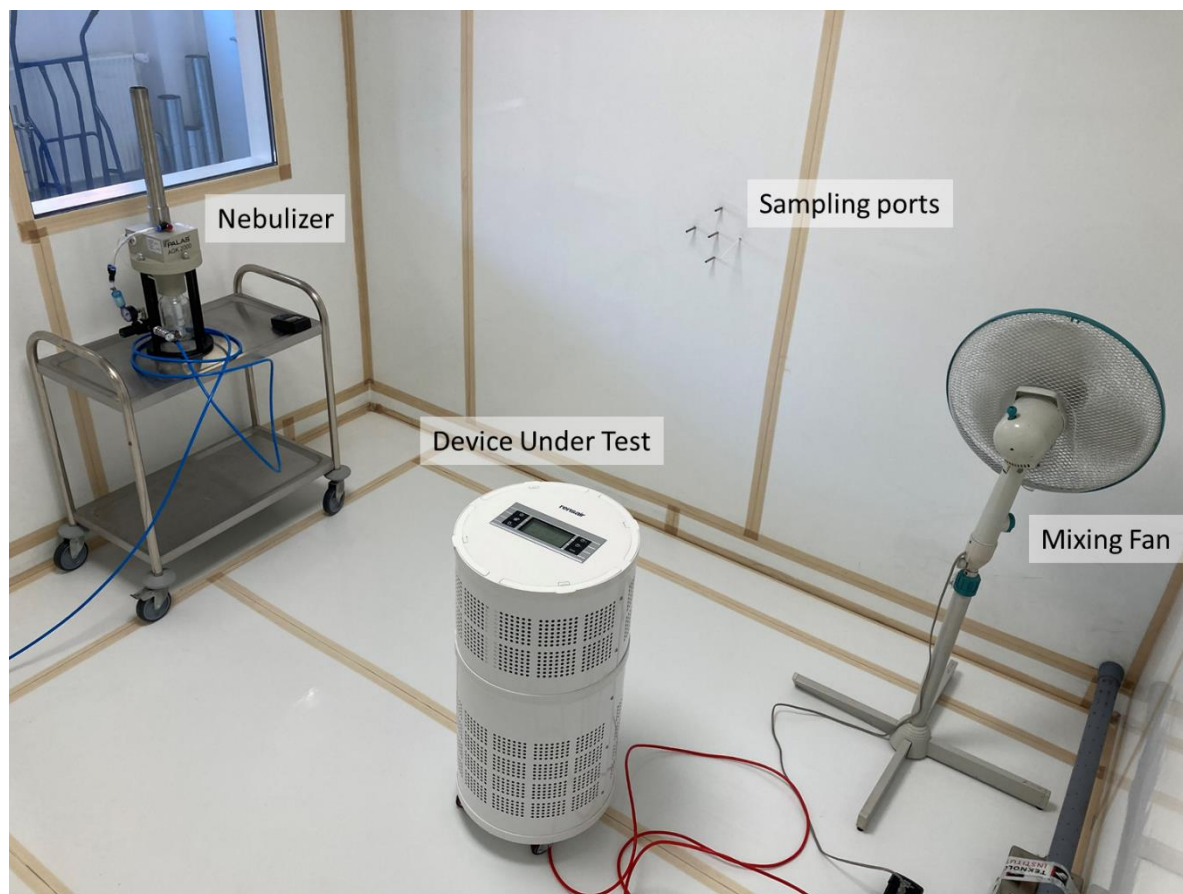


Figure 1: Image of setup in the test chamber.





Figure 2: Device under test - Rensair.

The sampling from the air is done through 6mm stainless steel tubes in the sidewall of the room using GilAir plus pump at 4.0 L/min. For each time stamp three air samples are extracted simultaneously at different locations in the test chamber.

A total of 20 L is extracted per sample into an impinger with 60mL SM-buffer. The timing of sampling is: 0, 15 and 30 minutes after finishing the aerosolization. The exact time for the sampling is defined by the beginning of the sampling time of about 5 minutes. The start of the first sample ( $t = 0$  minutes) is less than a minute after the nebulizer is stopped.



The procedure is the following:

1. A suspension of MS2 in SM-buffer is prepared and the concentration is determined.
2. A background sample is taken before the test and injection of aerosol.
3. The reference test of the natural decay is carried out *without* the air purifier turned on. The Palas nebulizer is working at 2 bar pressure for a total time of 15 minutes before the reference test is started.
4. The sampling is carried out according to the timing plan.
5. After the 30-minute test with the air purifier *off*, the room is flushed with clean air for 45 minutes.
6. The same procedure is followed for the air purifier test. After injection of the MS2 containing aerosol and sampling of the t=0 minutes sample the air purifier is remotely turned on ("Max" fan speed).
7. The sampling is carried out according to the timing plan.
8. The concentration of active MS2 is evaluated for each sample by mixing dilutions series with a fresh culture of the host bacteria, cultivation, and enumeration of plaque-forming unit (PFU) following incubation.

The test is performed 18<sup>th</sup> – 24<sup>th</sup> March 2021.

### Experimental conditions for air cleaning

Test organism:	MS-2 bacteriophage, ATCC 15597-B1
Host organism for MS2:	<i>Escherichia coli</i> , ATCC 15597
Growth conditions for enumeration of pfu:	Coliform agar at 37±2°C for 18-24 h
Growth conditions for host organism:	First on TSA plates and then in TSB at 250 rpm. at 37±2°C for 20-24 h.
Sampling and dilution solution:	SM-buffer
Sample volume (SM-buffer):	60 mL per bottle
Test suspension for aerosolization:	SM buffer with $3.3 \cdot 10^{10}$ pfu/mL



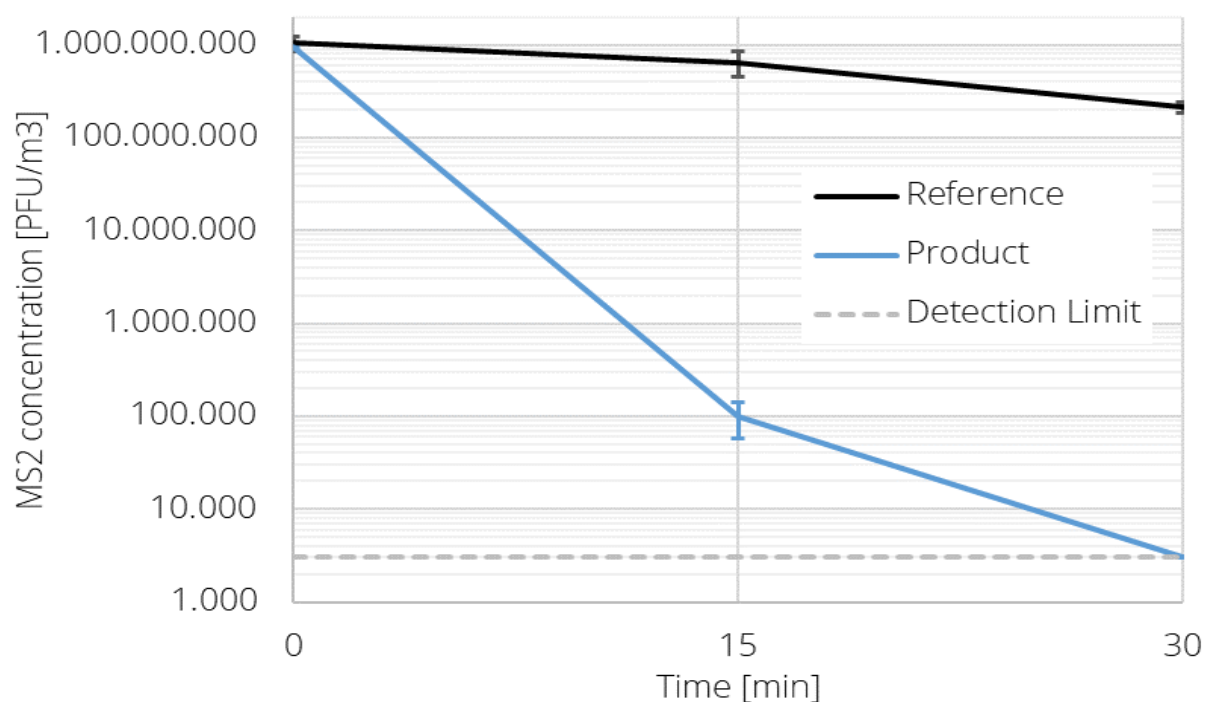
## Results

The concentration of active MS2 expressed as PFU/m<sup>3</sup> is shown in Table 1 and in graph in Figure 3. The room background is measured before the first injection of aerosols.

**Table 1: The concentration of active MS2 for the Natural decay and the Product test. The concentration is calculated as the average of triplicates. The standard deviation between the triplicates are presented in parentheses.**

Time (min)	Natural decay (PFU/m <sup>3</sup> )	Product test (PFU/m <sup>3</sup> )
Background	0	-
0	$1.04 \times 10^9$ ( $1.89 \times 10^8$ )	$9.49 \times 10^9$ ( $3.47 \times 10^7$ )
15	$6.44 \times 10^8$ ( $1.90 \times 10^8$ )	$9.99 \times 10^4$ ( $4.29 \times 10^4$ )
30	$2.14 \times 10^8$ ( $2.73 \times 10^7$ )	$< 3.00 \times 10^3$ (0.00)

\* Sample concentration below detection limit (defined as  $< 60$  PFU/sample).



**Figure 3: Concentration of active MS2 over time for the product test and the reference experiment. Note logarithmic y-axis.**





The air purifier's capacity to reduce active MS2 is calculated based on the difference in relative concentration at individual sampling points as specified in ISO 16000-36:2018.

The percentage-wise reduction is calculated as the relative difference in MS2 concentration over time for the product test and the reference experiment (natural decay). The MS2 concentration at t=0 for each run is defined as index 100% (see Table 2 and Figure 4).

The results show that after 15 minutes 0.011% of the initial airborne MS2 was left in the product test as compared to 61.9 % for the natural decay. Hence, the concentration is 99.98% lower after 15 minutes, when comparing the product test and the natural decay. After 30 minutes, the concentration of active MS2 in the air in the product test is below the detection limit. Thus, the product attribution is calculated from the concentration defined as the detection limit.

**Table 2: Calculated reduction in MS2-concentration**

Time, minutes	Relative Concentration (%)		Reduction in MS2-concentration (%)
	Natural Decay	Product Test	Product Attribution
0	100	100	-
15	61.9	0.011	99.98
30	20.5	< 0.001*	> 99.99*

\* Calculated from the concentration defined as the detection limit

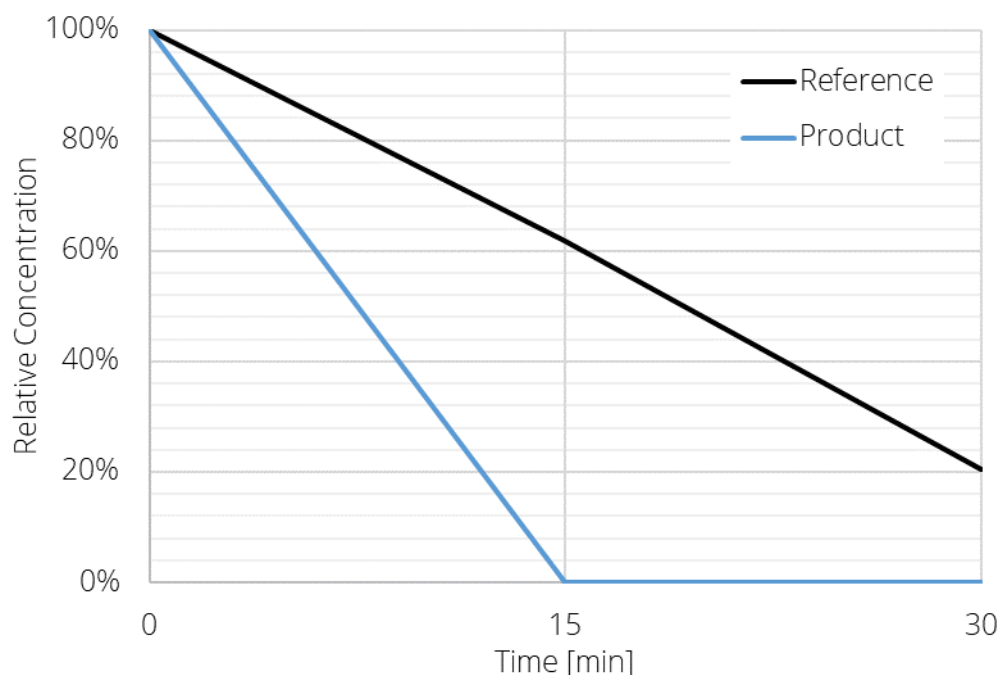


Figure 4: Relative concentration of active MS2 over time for the product test and the reference experiment. The concentration at  $t=0$  for each test run is defined to 100%.

#### Reduction of airborne Particles:

During this test of the Rensair air purifier (inactivation of active MS2 in the air) the size of aerosols produced by the nebulizer is continuously measured, as well as the concentration during the test of the product, using an Optical Particle Sizer (TSI OPS 3330). The efficiency to capture particles in the size range  $0.3 - 5 \mu\text{m}$  showed a  $>99.98\%$  reduction across the entire range after a 15 minute-period.

Note, that it is unclear which aerosol size that “carries” the active MS2 bacteriophage, hence the reduction is not directly comparable to the MS2-reduction.

Particle Size [ $\mu\text{m}$ ]	Reduction (20 m <sup>3</sup> test chamber in 15 minutes)
0.3	99.98%
0.579	99.99%
0.721	100.00%
1.117	100.00%
1.732	100.00%
2.685	100.00%
3.343	100.00%
4.162	100.00%

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