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Date
24 March 2021

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Subject
evaluation of the reported viricidal performance tests of the Shycocan system

Dear [redacted],

Please find attached the review document concerning the evaluation of the reported viricidal performance tests of the Shycocan system.

Yours faithfully,

[redacted]
Senior scientist
Department of Microbiology and Systems Biology

Intermediate Report

Disclaimer

According to the TNO's General Terms and Conditions, article 10 applies to the use of the final report: "Final Report, as well as a (possible) preliminary or an intermediate report or any subsequent explanatory notes to the Final Report, is solely for the Client's own use. Any other use requires TNO's prior written consent".

Assignment

Scalene EU Ltd assigned TNO to evaluate tests that were done with the Shycocan system in various laboratories considering the current pandemic Corona crisis and to advise on further tests if necessary.

Objective

The objective is to establish whether the Shycocan could be a valuable tool in combatting Corona viruses thereby providing an option for opening social economic activities, and to define what further tests would be useful to support the manufacturers claim the device reduces the viral load with more as 99%.

Preliminary Conclusion

Based on the tests done so far. it can be concluded that the Shycocan system is antiviral against several viruses including corona viruses that are considered as surrogates for SARS COV-2.

The proof of functionality in relation to the current pandemic lies in a test with the SARS COV2 virus. This test will be done by TNO shortly.
A surrogate virus tested is Equine Arteritis Virus The virucidal effect on Equine arteritis virus was demonstrated to be 3.1 log units for the 15 minutes exposure at 50 cm and 3.4 log units for the 120 minutes exposure at 500 cm. Therefore, a more than 99.9% reduction in infectious Equine arteritis virus particles was concluded.

Another more likely surrogate is the Avian corona virus. The efficacy of the Shycocan system was tested with the airborne virus (Aerosol) in a closed chamber of 6,3 m3. Viruses collected in this chamber were inoculated in embryonated chicken eggs. The eggs that were inoculated with viruses that were not exposed to the Shycocan system showed impaired embryo viability while eggs inoculated with viruses exposed to the active Shycocan system during 15, 60 and 120 minutes, respectively, all showed viable embryos without defects. Thus the Shycocan system realized a strong virus reduction to a non-lethal level.
The conclusion of these 2 tests is that the Shycocan reduces surrogate SARS-COV2 viruses in spaces with more than 99,7% within 5 to 15 minutes.

Recommendation

It is recommended to repeat this kind of tests with the SARS-COV2 virus itself to support the manufacturers claim that the Shycocan system deactivates more as 99,7% of the SARS-COV2 virus instantaneously.

TNO assignment

Scalene EU Ltd assigned TNO to carry out these tests. TNO expects to complete this test and the report before 1 May 2021.

Management Summary

TNO was asked by the European importer of the Shycocan to validate the manufacturers claim that the device neutralizes more as 99% of Corona viruses instantaneous. As a first step the laboratory tests done so far were reviewed. TNO's preliminary conclusion is that the manufacturers claim that the device reduces the viral load caused by Corona viruses instantaneously with more as 99% is probably correct. However not all tests were done with the same high scientifically standards as TNO itself applies. Therefore, to be completely sure TNO will do additional tests in its own laboratories.

Working of the device

The manufacturer explained they have a hypothesis about the working of the device but were not yet able to proof this hypothesis. The hypothesis is that the working of the device is based on the way Corona viruses cause infections. Corona viruses are characterized by spikes on the surface of the virus. These spikes have a positive charge and in this way the virus is attracted to and penetrates mammal cells. The device causes clouds of negatively charged electrons that neutralizes the positive charged spikes, in this way preventing infections.
Normally electrons cannot travel far in air, but photons being weightless particles of light can. The manufacturer claims the device creates photons that converts into electrons upon hitting mass.
The tests being done so far don't contradict this hypothesis.

Tests reviewed.

AQUADIAGNOSTICS, IAMPO group, Bangalore, India evaluated the SHYCOCAN system with respect to the decontamination of the bacterium *Escherichia coli* smeared on acrylic sheets and *E. coli* bacteriophage MS2 (ATCC15597B1) on the same type of surface and on planks (see document 1). Regarding the Shycocan system, exposure of *E. coli* to the system during 1 and 2 hours at short distance and longer distance (=12 feet) did not yield any reduction in viable counts of *E. coli*. The Shycocan system at 12 feet from MS2 *E. coli* bacteriophage on acrylic sheets, did reduce the infectious capacity of the MS2 *E. coli* bacteriophage by a percentage of about 99.9% irrespective distance and the exposure time up to 2 hours. Unfortunately, the description of the experimental setup was brief. Appropriate description of non- exposed controls was absent. Therefore, it is highly probable but not absolute sure whether bacteriophage inactivation was caused by the Shycocan system.

AQUADIAGNOSTICS also studied the efficacy of the SHYCOCAN system against MS2 bacteriophage in office room settings (see document 2). MS2 bacteriophage contaminated planks were in the corners and middle of the room and exposed to the system during 1, 5 and 15 minutes. Non-exposed controls were briefly mentioned. The results indicate a gradual increased percentage of reduction in plaque forming units (pfu) from about 99.6% to 99.7% upon 1 to 15 minutes exposure to the SHYCOCAN system.

Document 3 describes briefly the tests performed by AQUADIAGNOSTICS with the SHYCOCAN system 7 feet above floor level on MS2 bacteriophage present on planks at different distances from the SHYCOCAN system (ranging from 10 to 24 feet distance and locate 11 feet above the floor) in two different rooms: one with pillar in the room and one without. Exposure times were ranging from 5 to 60 minutes. The test results were compared and calculated as percentage reduction relative to the pfu counts before treatment. The results show a slightly increased reduction percentage in pfu with increased exposure time to the SHYCOCAN system. The presence of the pillar in the room hardly affected the reduction. Reductions observed ranged from 99.82% to about 99.96% and at the backside of the pillar about 99.75% on average. At about 10 feet, planks with MS2 placed in glass chambers also showed a reduction of pfu of about 97% relative to the "before treatment" MS2 pfu counts. In addition, the description of the test settings, the nature of the negative controls should be improved in future tests.

Tres Monos Lab de R.L. de C.V., Mexico, studied the SHYCOCAN system against two viruses, Equine arteritis virus ATCC VR-796 (enveloped RNA virus; surrogate for SARS-CoV-2 virus) and Influenza B virus ATCC VR1535 (enveloped RNA virus; surrogate for SARS-CoV-2 virus). These tests were reported in document 4 and 5, respectively.

According to their reports, the SHYCOCAN system is an electron emission device meant to be applied for environmental and surface sanitation against coronavirus and analogues. Both reported studies are well documented with adequate description of experimental conditions, analysis methodology, data processing and interpretation. The SHYCOCAN system was tested in a room of 6 x 3 x 2 meters that was treated for 120 minutes by the active SHYCOCAN system. Thereafter, three plates with Equine arteritis virus suspension for each distance were placed at 50 and 500 cm distance from the SHYCOCAN system and exposed. In addition, non-exposed plates were included. The virucidal effect on Equine arteritis virus was demonstrated to be 3.1 log units for the 15 minutes exposure at 50 cm and 3.4 log units for the 120 minutes exposure at 500 cm. Therefore, a more than 99.9% reduction in infectious Equine arteritis virus particles was concluded (see document 4).

In conclusion, the experiments described in document 4 are executed in a sound way and presented accordingly. The SHYCOCAN system showed viricidal effects on Equine arteritis virus present on surfaces at different locations in a room. Document 5 shows the report of Tres Monos Lab de R.L. de C.V., Mexico, on the viricidal effect the SHYCOCAN system against the Influenza B virus ATCC VR1535 (enveloped RNA virus; surrogate for SARS-CoV-2 virus). As for the Equine arteritis virus the reported methodological approach was solid.

In conclusion, the experiments described in document 5 are executed in a sound way and presented accordingly as far as the surrogate SARS-COV2 viruses are concerned. However, a claim that The SHYCOCAN system also deactivates the Influenza B virus is not sufficiently proven. The test showed marginal (50%) viricidal effects on Influenza B virus present on surfaces.

The Indian Institute of Technology Guwahati evaluated the SHYCOCAN system against airborne avian coronavirus (see document 6). For this purpose, a small chamber of about 6.3 cubic meter was used to test viricidal activity against airborne virus particles under contained conditions. The setup of testing and methodology was adequately described for generating airborne virus particles in aerosol, homogenization of aerosol in the chamber, virus loaded aerosol sampling

by using a 0.01 µm filter and bioassay on embryonated chicken egg. However, the procedure for real time PCR and use for obtaining virus titration data was unclear; this needs further explanation. The viricidal effect of the SHYCOCAN system against Avian coronavirus was demonstrated in a small room. The strength of the conclusion based on the bioassay is acceptable but can be made stronger with appropriate information on the qPCR data.
In conclusion, the experiments described in document 6 are executed in a sound way. Unfortunately, only the bioassay was well described, which did not apply to the description of the real time PCR. Nevertheless, the SHYCOCAN system showed viricidal active against airborne Avian coronavirus present in a small chamber of 6.3 cubic meter based on the bioassay. If the real time PCR was presented in a more concise way, provided data in the document with respect to the drop in virus presence as a result of the SHYCOCAN system could be accepted.

Overall, it can be concluded that the SHYCOCAN system is antiviral against Equine arteritis virus and Avian coronavirus while displaying limited antiviral activity against Influenza B virus.
These tests are sufficient basis for TNO to continue with the assignment. The next step will be tests with the “real’ SARS-COV 2 virus.