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An Institution of National Importance
(Ministry of Home Affairs, Government of India)

COVID -19 Testing Facility (ICMR APPROVED)

ANALYSIS REPORT

EFFICACY TESTING OF STYLAM ANTIVIRAL LAMINATE FOR SARS COV-2 VIRUS AS PER ISO 21702:2019 (MODIFIED) TEST METHOD

Name of Company:

Stylam Industries Limited.

Report No.

NFSU/IFS/AntiViral Testing/12/2021

Date: 19-07-2021

Ref. Email **Date: 21-06-2021**

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19/7/2021

Objective

To evaluate the antiviral activity on the surface of one sample as demonstrated by the ISO 21702:2019 (Modified) test method.

Test Sample Details

Name of the Product:	Antiviral High Pressure Laminate
Type of the product:	High Pressure Laminate
State of Product:	High Pressure Laminate
Name of Company	Stylam Industries Limited
Address	SCO 14, Sector 7C, Madhya Marg, Chandigarh - 160019, India
Phone	+91 7508009901
Email	sachinbhatla@stylam.com

Brief Note on Test Organism and Target Genes

- SARS-CoV-2 is a positive-sense, single-stranded RNA (ssRNA), group IV virus. CoV genomes code for a ORF1a / ORF1ab polyprotein and four structural proteins (e.g. S gene and N gene) and the non-structural genes include the RNA dependent RNA polymerase (RdRP) which are widely studied as major drug and detection targets.

Test Procedure Summary

- The test organism was adjusted and diluted to obtain the starting inoculum concentration of C_T value of 27 for all the targets i.e. N, RNAseP and ORF genes.
- Each sample piece was placed in a sterile Petri dish, inoculated and then covered with the sterile plastic in order to spread the inoculum evenly over the sample surface and hold it in place and incubated at 35°C till the sample got dried.
- Then the samples were incubated at 35°C and a relative humidity of at least 90%. At the appropriate time the entire area of the sample piece was swabbed using the sterile wet cotton swab to collect the inoculum from the surface and mixed in 200 µL nuclease free water and treated with Ambion™ RNase A (As per the manufacturer protocol).
- Then the RNAse Treated sample was processed for RNA extraction using MagMAX™ RNA Extraction Kit (Thermo Fisher Scientific) method.
- After RNA extraction, the CoviPath (Thermo Fisher Scientific) kit was used to measure the C_T value of S, N and ORF genes for each sample.
- The C_T value of each sample was recorded. The results are found in the "Test Results" section below. These results pertain only to the samples tested. All the samples were run in triplicate.

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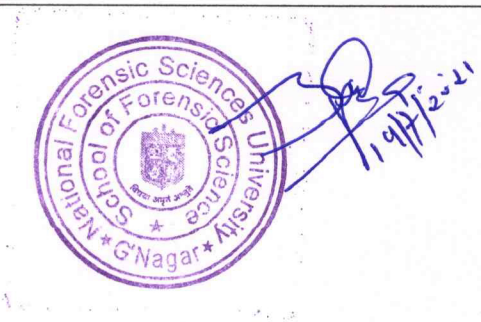
Test Variables

No.	Variable	Details
1	Sample Submission Date	21-06-2021
2	Sample Testing Date	16-07-2021
3	Report Date	19-07-2021
4	Sample to be tested	Antiviral Laminate (Batch No. 1020SD)
5	Test Organism	COVID-19 Virus
6	Sample Size	50 mm x 50 mm x 1 mm
7	Method of Sterilization / Pre-Cleaning	NA
8	Control Sample	Non-Antiviral Laminate
9	Dilution Medium Used	Viral Transport Medium
10	Starting Inoculum Concentration	C_T value 27 for N, S, and ORF gene
11	Amount of Inoculum	70 µL
12	Contact Time	5, 10, 20, 30, 40, 120 min
13	Deviations from Standard Test Method	Yes. The method (ISO 21702:2019) was modified for the test organism COVID-19 Virus.



Test Results (These results pertain only to the samples tested Batch No. 1020SD)

No.	Sample Code	Time Interval (min)	Antiviral High Pressure Laminate									High Pressure Laminate (Untreated)			
			**N Gene C_T (Average)	Delta C_T (13-3)	% Reduction in viral Load	**ORF Gene C_T (Mean of Triplicate)	Delta C_T (14-3)	% Reduction in viral Load	**RNAseP C_T (Mean of Triplicate)	Delta C_T (15-3)	% Reduction in viral Load	**N Gene C_T (Average)	**ORF Gene C_T (Average)	**RNAseP C_T (Average)	% Reduction in viral Load
1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
1	Stylam Antiviral Laminate	05	32	01	90.0	33	02	99.0	32	02	99.0	31	31	30	0.0
		10	33	02	99.0	34	03	99.9	33	03	99.9	31	31	30	0.0
		20	33	02	99.0	34	03	99.9	33	03	99.9	31	31	30	0.0
		30	34	03	99.9	34	03	99.9	34	04	99.99	31	31	30	0.0
		40	35	04	99.99	35	04	99.99	34	04	99.99	31	31	30	0.0
		120	35	04	99.99	35	04	99.99	34	04	99.99	31	31	30	0.0
2	Positive Control	-	27	-	-	27	-	-	27	-	-	-	-	-	-
3	Negative Control	-	ND	-	-	ND	-	-	-	-	-	ND	ND	ND	-

** = Mean of Triplicates, ND=Not Detected, NS=Not Significant

Conclusion (These results pertain only to the samples tested with Batch No. 1020SD)

Based on the test results, it is concluded that the tested products (tested samples) are effective to reduce viral load of SARS CoV-2 up to 99.9% at time interval of more than 10 min.

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Test Results Interpretation

The value of the antiviral activity was calculated according to the formula listed below and recorded as log reduction.

$$R = (U_t - U_0) - (A_t - U_0) = U_t - A_t$$

Where,

R : antiviral activity

U_0 : average of logarithm numbers of viable viral particles from untreated control at Time = 0 h

U_t : average of logarithm numbers of viable viral particles from untreated control at Time = t Min

A_t : average of logarithm numbers of viable viral particles from test sample at Time = t Min

According to the standard, an antiviral product is determined to have antiviral effectiveness when the antiviral activity (R) is 2.0 or more.

Percent reductions are determined by comparing the sample after the contact time to the untreated laminate control after the contact. Reporting of percent reduction is not indicated by the test method but is provided by NFSU as additional information.

Percent reduction is translated into log reduction by the following:

90% reduction = 1 log reduction; i.e. Ct 10 increased to Ct 11 is a 1 log reduction

99% reduction = 2 log reduction; i.e. Ct 10 increased to Ct 12 is a 2 log reduction

99.9% reduction = 3 log reduction; i.e. Ct 10 increased to Ct 13 is a 3 log reduction

99.99% reduction = 4 log reduction; i.e. Ct 10 increased to Ct 14 is a 4 log reduction

99.999% reduction = 5 log reduction; i.e. Ct 10 increased to Ct 15 is a 5 log reduction

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Analysis Reviewed and Approved by

19/7/2021

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