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29/4/2021

Report

Experiment 8-23/4/21 C-polar treated cellulose/polyester textile material

Objective: Evaluate the C-polar treated cellulose+polyester material for antiviral effect on SARS-CoV-2 according ISO 18184.

Tests:

A) Verification of toxicity by cell sensitivity to virus
B) Determination of anti-SARS-CoV-2 activity of the material

Samples:

a. 6% C-polar cellulose + polyester
b. Control cellulose + polyester

SARS-CoV-2 isolate:

hCoV-19/Czech Republic/NRL_6632_2/2020

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A)

Control toxicity test to exclude possibility that test specimen can reduce cell sensitivity to virus. The difference between titer from untreated specimen and treated specimen should not be bigger than 0.5 log_{10}.

Protocol:

Textile samples 1×1 cm in triplicate were washed in 10ml DMEM complete medium by vortexing for 5 times for 5 seconds. 5ml of media was transferred to another tube and 50µl of SARS-CoV-2 (10e5 IU/mL) was added and mixture was incubated 30 min at room temperature. After incubation, 200µl was removed and titered by plaque assay in 24-well plate in DMEM complete using 10-fold dilution. Virus with cells (250 000/well) was briefly gently mixed and incubated for 4 hours at 37°C in CO_2 incubator. After that, 0.4 ml of 3% carboxymethylcellulose was added. Plates were incubated for 5 days. After incubation, cells were washed, stained with Naphthol blue black dye, washed with water and dried. Plaques were counted as pfu/ml, expressed in log_{10}, compared to log_{10} pfu/ml of untreated control and log_{10} differentials in virus titer were determined.

Results:

Sample	SARS-CoV-2 titer [pfu/ml]	Log SARS-CoV-2 titer [pfu/ml]	Differential of log pfu/ml to control
Control cellulose+polyester	453333	5.66	-
6% C-polar cellulose+polyester	440000	5.64	0.02

Conclusion:

Samples do not reduce cell sensitivity to virus.

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B) Determination of anti-SARS-CoV-2 activity of the materials

50 µL of SARS-CoV-2 (10e5 IU/mL) was dropped to 1×1 cm squares of textiles (treated and control) in triplicate. Textiles were incubated for 5 and 30 minutes at room temperature covered with glass. After incubation, textiles were transferred into 5mL DMEM complete, vortexed 5-times for 5 s and 200 µl of media was transferred on VERO-E6 cells and titer of remaining virus was determined by plaque assay. Virus with cells (250 000/well) was briefly gently mixed and incubated for 4 hours at 37°C in CO_2 incubator in 24-well plate. After that, 0.4 ml of 3% carboxymethylcellulose was added. Plates were incubated for 5 days. After incubation, cells were washed, stained with Naphthol blue black dye, washed with water and dried. Plaques were counted and titer was expressed as pfu/ml and virus yield reduction was expressed in percentage.

Results:

	5 minutes exposure		30 minutes exposure	
Sample	Titer of recovered virus pfu/ml	Virus yield reduction [%]	Titer of recovered virus pfu/ml	Virus yield reduction [%]
Control cellulose+polyester	236667	-	255000	-
6% C-polar cellulose+polyester	5167	98	1000	99.6

Conclusion:

Cellulose+polyester with 6% C-polar eliminated on average 99.6% of SARS-CoV-2 (in one of the biological replicate we achieve 100% virus elimination) after 30 minutes exposure.

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