### **TEST REPORT**

Report No: AWRTCL/PRTR/ 17361/20-21 Date: 04.06.2020

CUSTOMER DETAILS	SAMPLE DETAILS	TEST DETAILS
	Sample received: 26.05.2020	
Name & Address :	Sample code no: AWRTCL/17361/20-21	Method:
Mr.Umesh Agrawal	Sample Description: INSTA PROTEX	As agreed
Watch Water INDIA ( Watch	Sample Quantity for Testing: 1 Kg Powder	between the
Water Treatment Pvt Ltd.)		Testing
B-11 SHANKAR GARDEN	Date of Analysis started : 25.05.2020	Laboratory and
VIKAS PURI,	Date of Analysis Completed: 04.06.2020	the customer
NEW DELHI - 110018, INDIA	Subcontract : Not Applicable	
·	Sample condition when received: Intact	

#### **EXECUTIVE SUMMARY:**

A project was taken up to assess Microbial decontamination on surfaces of different material like Plastic, Leather, Laminated Wood, Polished stone, Glass and Metal with induced microbial contamination using INSTA-PROTEX disinfectant solution. INSTA PROTEX disinfectant powder was tested at 1.0% solution (5.0 gr in 500 ml of Tap water) for its capability to reduce induced microbial contamination on surfaces with an exposure time of 1 minute duration. The tested disinfectant solution was found to be effective in reducing 10 different microbial species (constituting 8 bacterial species one Mold and one Yeast species) to the tune of ≥99.999% and ≥99.999% reduction for one Surrogate Virus of bacteriophage.

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#### **METHODOLOGY:**

Two similar surfaces were smeared for an area of 10 cm x 10 cm with 1 ml 24 hr old broth culture of a known microbial species. The contents were allowed to air dry for 10 minutes. One of the surfaces smear was swabbed and transferred to 10 ml of 0.9% physiological saline. Serial dilutions were made and 1 ml inoculum was plated out on selective agars. Incubation was done at 37  $^{\circ}$ C / 24-48 hr. Colonies were enumerated. **This is BEFORE TREATMENT** 

The second surface smear was exposed to 1 ml INSTA PROTEX disinfectant solution by spreading. The contents were allowed for 1 minute treatment. The treated surfaces were swabbed and contents were transferred to 10 ml of 0.9% physiological saline. Serial dilutions were made and 1 ml inoculum was plated out on selective agars. Incubation was done at 37 °C / 24-48 hr. Colonies were enumerated. **This is AFTER TREATMENT** Note: Yeast & Mold plates were incubated at 25°C/3-5 days.

% Reduction was calculated by taking microbial counts Before Treatment as reference.

The Tap water used for preparing INSTA PROTEX Solution : TDS 413 mg/L, pH: 7.40, Temperature: :  $25\,^{\circ}C$ 

# PICTURE SHOWING SWABBING PROCEDURE ON A POLISHED STONE SURFACE (WITH MICROBIAL CONTAMINATION AND INSTA PROTEX TREATMENT)



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TEST DATA: Microbial reduction with Induced microorganisms on ABS PLASTIC SURFACE

Name of Microorganism	Microbial counts Before Treatment	Microbial counts After Treatment	% Reduction
	<mark>60 s</mark>	seconds Exposure	
BACTERIA			
E.coli MTCC 68	5.0x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Pseudomonas aeruginosa MTCC 424	8.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Enterococcus faecalis MTCC 439	5.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Vibrio Cholera MTCC 3906	6.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Salmonella typhimurium MTCC 98	6.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Shigella flexneri MTCC 1457	5.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Clostridium perfringens MTCC 450	7.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Staphylococcus aureus MTCC 87	8.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
VIRUS – Bacteriophage			
MS2 phage ATCC15597B1	7.0 x 10⁵pfu/ Swab	NPFU/swab	≥99.999
MOLD			
Aspergillus niger MTCC282	6.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
YEAST			
Saccharomyces cerevisiae MTCC 2569	6.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999

TEST DATA: Microbial reduction with Induced microorganisms on LAMINATED WOOD

Name of Microorganism	Microbial counts	Microbial counts	%
_	BeforeTreatment	After Treatment	Reduction
	60 seconds Exposure		
BACTERIA			
E.coli MTCC 68	7.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Pseudomonas aeruginosa MTCC 424	9.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Enterococcus faecalis MTCC 439	6.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Vibrio Cholera MTCC 3906	8.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Salmonella typhimurium MTCC 98	5.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Shigella flexneri MTCC 1457	6.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Clostridium perfringens MTCC 450	6.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Staphylococcus aureus MTCC87	7.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
VIRUS – Bacteriophage			
MS2 phage ATCC15597B1	6.0 x 10⁵pfu/ Swab	NPFU/swab	≥99.999
MOLD			
Aspergillus niger MTCC282	6.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
YEAST			
Saccharomyces cerevisiae MTCC 2569	7.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999

Cfu:Colony forming units, Pfu:Plaque forming units, <10 cfu: NVC/Swab:No viable colony/Swab , <10 pfu = NPFU/Swab: No plaque forming unit /Swab

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**TEST DATA: Microbial reduction with Induced microorganisms on POLISHED STONE** 

Name of Microorganism	Microbial counts	Microbial counts	%
	BeforeTreatment	After Treatment	Reduction
	60 seconds Exposure		
BACTERIA			
E.coli MTCC 68	7.0x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Pseudomonas aeruginosa MTCC 424	5.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Enterococcus faecalis MTCC 439	8.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Vibrio Cholera MTCC 3906	7.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Salmonella typhimurium MTCC 98	9.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Shigella flexneri MTCC 1457	9.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Clostridium perfringens MTCC 450	8x10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
Staphylococcus aureus MTCC 87	8.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
VIRUS – Bacteriophage			
MS2 phage ATCC15597B1	7.0 x 10⁵pfu/ Swab	NPFU/ Swab	≥99.999
MOLD			
Aspergillus niger MTCC282	5.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999
YEAST			
Saccharomyces cerevisiae MTCC 2569	7.0 x 10 <sup>6</sup> cfu/ Swab	NVC/swab	≥99.9999

**TEST DATA: Microbial reduction with Induced microorganisms on LEATHER** 

Name of Microorganism	Microbial counts BeforeTreatment	Microbial counts After Treatment	% Reduction
	60 seconds Exposure		House
BACTERIA		•	
E.coli MTCC 68	8.0x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Pseudomonas aeruginosa MTCC 424	6.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Enterococcus faecalis MTCC 439	7.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Vibrio Cholera MTCC 3906	8.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Salmonella typhimurium MTCC 98	5.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Shigella flexneri MTCC 1457	7.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Clostridium perfringens MTCC 450	6x10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Staphylococcus aureus MTCC 87	7.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
VIRUS - Bacteriophage			
MS2 phage ATCC15597B1	9.0 x 10 <sup>6</sup> pfu/ Swab	NPFU/ Swab	≥99.999
MOLD			
Aspergillus niger MTCC282	6.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
YEAST			
Saccharomyces cerevisiae MTCC 2569	7.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999

Cfu:Colony forming units, Pfu:Plaque forming units, <10 cfu: NVC/Swab:No viable colony/Swab , <10 pfu = NPFU/Swab: No plaque forming unit /Swab

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GSTIN: 29AAHCAO185G1ZP

TEST DATA: Microbial reduction with Induced microorganisms on GLASS SURFACE

Name of Microorganism	Microbial counts BeforeTreatment	Microbial counts After Treatment	% Reduction
		seconds Exposure	Reduction
BACTERIA			
E.coli MTCC 68	5.0x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Pseudomonas aeruginosa MTCC 424	6.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Enterococcus faecalis MTCC 439	7.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Vibrio Cholera MTCC 3906	6.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Salmonella typhimurium MTCC 98	7.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Shigella flexneri MTCC 1457	8.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Clostridium perfringens MTCC 450	8 x10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Staphylococcus aureus MTCC 87	8.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
VIRUS – Bacteriophage			
MS2 phage ATCC15597B1	8.0 x 10⁵pfu/ Swab	NPFU/ Swab	≥99.999
MOLD			
Aspergillus niger MTCC282	7.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
YEAST			
Saccharomyces cerevisiae MTCC 2569	7.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999

TEST DATA: Microbial reduction with Induced microorganisms on METAL SURFACE

Name of Microorganism	Microbial counts	Microbial counts	%
	BeforeTreatment	After Treatment	Reduction
	60 \$	seconds Exposure	
BACTERIA			
E.coli MTCC 68	9.0x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Pseudomonas aeruginosa MTCC 424	8.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Enterococcus faecalis MTCC 439	8.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Vibrio Cholera MTCC 3906	5.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Salmonella typhimuriumMTCC 98	5.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Shigella flexneri MTCC 1457	8.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Clostridium perfringens MTCC 450	7x10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
Staphylococcus aureus MTCC 87	7.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
VIRUS - Bacteriophage			
MS2 phage ATCC15597B1	7.0 x 10⁵pfu/ Swab	NPFU/ Swab	≥99.999
MOLD			
Aspergillus niger MTCC282	6.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999
YEAST			
Saccharomyces cerevisiae MTCC 2569	7.0 x 10 <sup>6</sup> cfu/ Swab	NVC/ Swab	≥99.9999

Cfu:Colony forming units, Pfu:Plaque forming units, <10 cfu: NVC/Swab:No viable colony/Swab , <10 pfu = NPFU/Swab: No plaque forming unit /Swab

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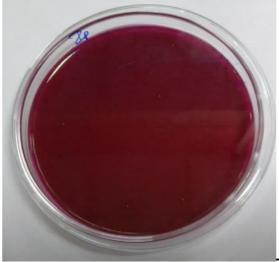
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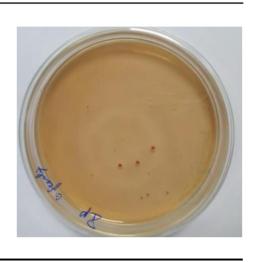
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G STIN: 29 AAHCAO 1 85G I ZP

#### COLONY CHARACTERISTICS OF MICROROGANISMS USED FOR THE STUDIES

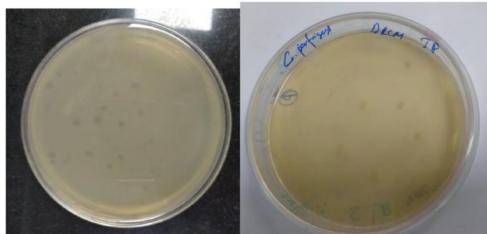
### E.coli Enterococcus faecalis





MS2 phage

Clostridium perfringens



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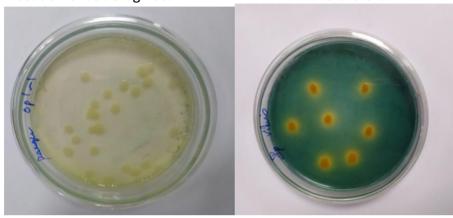
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Pseudomonas aeruginosa



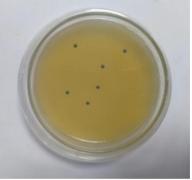


Aspergillus niger

Saccharomyces cerevisiae

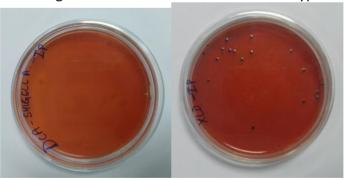
Staphylococcus aureus





Shigella flexneri

Salmonella typhimurium



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#### **RESULTS & CONCLUSION:**

The test data obtained from various tests conducted using 1.0% solution of INSTA PROTEX disinfectant to decontaminate the different surfaces having contamination of know microbial cultures reveals that INSTA PROTEX is effective in 60 seconds exposure to bring about at least 99.999% reduction of MS2 phage surrogate virus and at least 99.9999% reduction of bacteria, Yeast and Mold species.

#### MICROBIOLOGICAL MEDIA USED

Name of Microorganism	Growth Media used
BACTERIA	
E.coli MTCC 68	M Endo agar medium
Pseudomonas aeruginosa MTCC 424	Cetrimide agar medium
Enterococcus faecalis MTCC 439	Slanetz Bartely agar medium
Vibrio Cholera MTCC 3906	Thiosulphate Citrate Bile slats sucrose agar medium
Salmonella typhimurium MTCC 98	Xylose Lysine Dextrose agar medium
Shigella flexneri MTCC 1457	Deoxycholate Citrate agar
Clostridium perfringens MTCC 450	Differential reinforced clostridial agar
Staphylococcus aureus MTCC 87	Baired parker agar
VIRUS – Bacteriophage	
MS2 phage ATCC15597B1	Tryptone Soya agar
MOLD	
Aspergillus niger MTCC282	Chloramphenicol Yeast Glucose agar
YEAST	
Saccharomyces cerevisiae MTCC 2569	Chloramphenicol Yeast Glucose agar

#### **IMPORTANT**

**ANALYTICAL METHODS:** Standard Methods from IS, APHA and USEPA published documents.

**CHEMICALS:** All chemicals used are Analytical grade.

LAB EQIPMENT: All equipment used, as applicable, are calibrated by NABL accredited laboratories

WATER: Reagent grade water

MICROBIAL CULTURES: MTCC and ATCC standard cultures

Dr S.MURALIDHARA RAO Head – Laboratory

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00----- End of the Test Report ----- 00

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