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RESEARCH ARTICLE

Dynamic Mechanism-Based Portable Anti-Microbial Green Decontamination Station

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Abstract

Objectives: The current research is focused on development and evaluation of antimicrobial efficacy of a portable green conveyor machine. **Methods:** The conveyor belt station was fabricated and incorporated with UV-C (265nm) lamps and air plasma cluster ion generator. Antimicrobial efficacy was evaluated against bacteria (*Escherichia coli* and *Staphylococcus aureus*), fungi (*Aspergillus brasiliensis*) and bacteriophage (MS2) according to the ISO Standard No. 18593:2018 (Microbiology) at varying speeds. **Findings :** A universal portable green conveyor-based station(CoVDecon) capable of disinfecting daily use items such as; bags, lunch boxes, shoes, cloth, etc. was developed with applications in public places such as universities, airports, railway stations etc. On performing antibacterial, antifungal and antiviral studies it was observed that the machine at a speed of 3m/sec successfully eliminated 99.39% of gram-negative and gram-positive bacteria, 99% of the fungi and 99% of virus after 3 rounds. Efficacy against bacteria, fungi and virus dropped to 97%, 95% and 97% respectively on increasing the speed to 35m/sec. However, the efficacy might have reduced with an increase in speed due to less time of exposure of the test substance to UV lamps and plasma cluster ions The study indicated that the machine has successfully eliminated the test- microorganisms from contaminated object irrespective of its placement on the belt. Finally, the machine produces no toxic waste, is portable, and cost-efficient. **Novelty:** The developed conveyor tunnel uses UV and plasma cluster ions provides high efficacy antimicrobial activity by eliminating pathogens from the articles completely, including internal pockets and cavities.

Keywords: Biological decontamination; COVID19; Plasma cluster ions; UV Irradiation; CBRN decontamination

1 Introduction

The pandemic caused by Severe-acute respiratory syndrome coronavirus 2 (SARS-CoV-2), 2019, has highlighted the importance of disinfection and decontamination to healthcare professionals, first responders, and civilians throughout the world. The primary focus in a disease outbreak is minimizing the risk of cross contamination and spreading of the infection. To aid this, the ideal strategy is to restrict public contact and effectively sterilize the environment.

Multiple techniques and strategies have been implemented for early control and prevention of cross-contamination.

These strategies include the use of soap solutions, disinfectants, devices etc. for effective decontamination. However, a few major challenges need to be addressed for the effective implementation of these strategies, such as;

a. Understanding the effect of surface based cross-contamination and spreading of infection

b. Gap in the current technologies and steps to develop a disinfecting system

c. Development of no-touch solutions to minimize the risk of cross-contamination

d. Development of automated systems with high efficacy wide spectrum microbial decontamination technology

The primary focus of the current research is the development of a portable decontamination station that can be easily installed in built-in environments such as university campuses, airports, train stations, marketplaces, industrial and medical settings etc.[(1)]

Built- in environments have a wide variety of microbes inhabiting inside. This makes the individuals present in the environment susceptible to the organisms present. On contamination by an article or an individual, the dynamic of the entire environment can change, making it potentially harmful. Although hygienic practices can be used to prevent direct contamination, decontamination of articles/materials is crucial. The spread of infection in these environments are governed by factors such as accessibility of surface, rate of surface touching, environment sterilization protocols, etc.

Additionally, the threat of biological contaminants, in case of the safety of dignitaries, needs to be addressed critically[(2)]. This warrants effective decontamination of the articles before reaching the dignitaries, to ensure their safety.

Major sources of contamination are either intentional (involving the use and spreading of infectious pathogens) or non-intentional (microbial particles are spread via indirect contamination).

Indirect contamination is difficult to manage as it is not possible to detect pathogens that could have been transmitted onto various surfaces. In this scenario, spreading of the infection occurs as a result of shedding of microbial particles from asymptomatic, symptomatic carriers and contamination of articles, surfaces etc. Contamination due to hand-surface contacts is the primary source of pathogen exchange in built in environments[(3)]. Further, it has also been observed from studies, that pathogens can be trapped in the grooves and pockets of articles making it difficult to decontaminate.

The current strategies employed in the disinfection of inanimate surfaces are- Ultraviolet-C (UV-C) radiation, atmospheric plasma, alcohol and chemical based disinfectants[(4,5)].

A literature based comparative analysis of the various chemical and physical sterilization and disinfection techniques employed in various occupational and industrial settings showed the specificity of the technique with respect to the scenario and articles to be decontaminated.

Multiple disinfecting solutions, such as, Reactive skin decontamination lotion (RSDL), soap solutions have shown limited efficacy (with 50% chances of viable resistant pathogens) and requires high quality waste management strategies to minimize the environmental burden. Most importantly, these chemicals are harsh and generally possess corrosive properties, are unstable, have shorter shelf-life, are toxic etc. therefore, severely limiting their application[(6)].

Automated no-touch systems use UV-C irradiation and atmospheric plasma technology for effective decontamination. Although these technologies have high antimicrobial efficacy, there are a few drawbacks in the application of these technologies in the current scenario. The efficacies of UV-C based disinfecting devices are primarily governed by the intensity of the lamps and the duration of exposure. Further, the usage of atmospheric plasma technology demands the use of solutions such as hydrogen peroxide, which is vaporized to form free radicals that are highly reactive species which interact with the pathogen to disrupt them. These solutions also have shorter shelf-life and can have corrosive effects on some articles/materials[(7)].

Although, all of these strategies have high antimicrobial efficacy, they have their drawback. Ranging from toxicity and corrosive nature to long cycle times and difficulty in portability there is an urgent requirement for a device that can actively sterilize any article with high efficacy. Moreover, strategies like UV-C disinfection require the environment to be evacuated and require supervision to prevent any side effects. Also, most of the systems are neither developed indigenously nor are cost-efficient[(8)]. Finally, the portability and ease of installation of the device at prime susceptible locations such as universities, airports, bus and train stations, seaports etc. needs to be considered. A large number of articles come to these locations on a daily basis and hence time and labor required for disinfection/decontamination is a constraint. This requires an automatic system which can effectively disinfect multiple articles in less time.

The currently used decontamination technologies are only focused on the decontamination of the surface of the article/material. However, there is a susceptibility of the microbial particles to enter the pockets and grooves in the article which is difficult to detect and needs to be decontaminated to prevent spread of infection. Apart from this, the exposure time and concentration of germicidal required to kill microbes can cause toxicity to the person performing these operations [9].

To maximize the interaction and completely decontaminate the materials/articles, a conveyor belt station can be designed, which can provide high antimicrobial effect on focused articles, reduce workload significantly whilst being cost-effective and eco-friendly.

To address this problem, a high efficacy decontamination conveyor tunnel has been developed and validated. The device uses a holistic approach to effectively decontaminate an inanimate object against biological pathogens using UV-C lamps and a novel plasma cluster generator. The device is ozone-free and prevents any risk of contamination.

The novel high impact plasma cluster ion generator creates charged ions from water molecules which react with the environment to form highly reactive hydroxyl radicals that electro-statically react with the surface proteins of the pathogens and disrupt them, thereby destroying the pathogen. The significant advantages of this technology are, firstly, there are no toxic chemicals which can damage the material and secondly, there is no toxic waste generation which makes it eco-friendly. Finally, the major advantage is the ability of the developed machine to completely decontaminate the article including the grooves and pockets, which is often overlooked. This ensures minimized risk of spread of infection and transmission of biological contaminants.

Literature reviews based on various research studies has shown high antifungal, antibacterial and antiviral efficacy of plasma cluster treated microbes with effects such as cytoplasmic leakage, cell wall and membrane disruption and loss of viability [10].

The antimicrobial efficacy is further enhanced by the usage of UV-C to completely sterilize the material. It is known that UV-C at 262 nm is called the germicidal spectrum. Radiation damages the deoxyribonucleic acid (DNA) and ribonucleic acid (RNA) of a microorganism, generally affects the pyrimidine molecules [11,12].

This strategy is employed to disrupt 99% of the pathogens when passed through the conveyor belt.

2 Objective

According to the literature survey conducted prior to the invention, it was found that no machine exists that portrays high antimicrobial efficacy, while being portable and eco-friendly. Therefore, the objective of the study was to develop a portable, green decontamination station that provided high antimicrobial efficacy in a limited time exposure. While developing the device, a novel technology, plasma cluster ion technology has also been considered. Therefore, objectives of the study were as follows-

1. Identification of the suitable microbial decontamination technologies via literature review
2. Designing of the machine using software such as AutoCAD and fabricating the machine
3. Incorporating the decontamination technologies in the machine
4. Evaluation of the antimicrobial efficacy of the developed machine against bacterial, fungal and viral strains.

3 Methodology

3.1 Fabrication of machine

3.1.1 Structural design of the conveyor tunnel

The tunnel chamber is made up of stainless steel (SS) 304 grade. The conveyor at the base is made of SS covered with a thick rubber sheet. The chamber is designed to decontaminate microbial pathogens from all sides as demonstrated in Figure 1. The chamber is equipped with a UV-C (265nm) lamp on all the walls including the base to destroy bacteria, viruses, and fungus [13]. The plasma cluster ion generator is installed at the base of the chamber as represented in Figure 1. The plasma cluster is spread by a suitable arrangement of nozzles as the material passes through the chamber. A conveyor is used for the easy movement of articles from the entry point to the exit end.

The internal dimensions of the tunnel were kept in a way to ensure the installation of multiple units at places expecting large footfall. The entire system is modular and portable. It has four lockable wheels on all corners to ensure easy mobility. One of the main advantages of this design is the dome-shaped structure at the top and bottom, without any corners making it easy to clean if required.

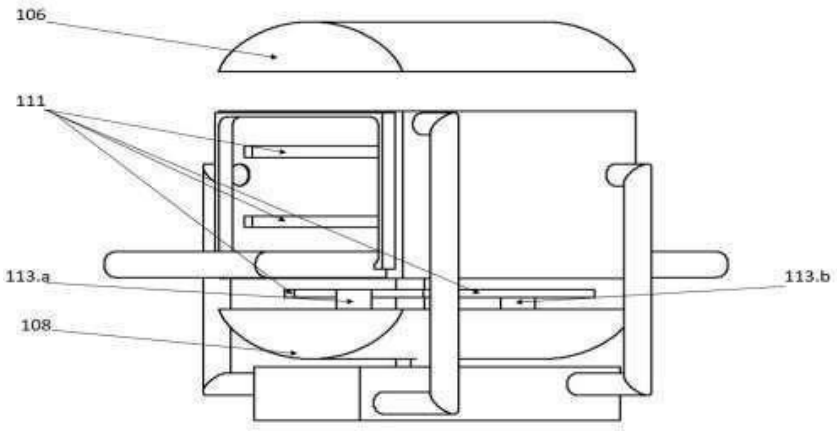

Fig 1. Isometric representation of the decontamination conveyor belt station-illustrates the UV-C tube light (111) the upper dome and the lower dome (106,108). A pipe opening (113a) for lower ionized air carrier pipe-1 and a pipe opening (113b) for lower ionized air carrier pipe- 2. Both the pipe openings (113.a and 113.b) are fitted with spraying systems such as a nozzle for spraying ionized plasma cluster ions.

3.1.2 Working mechanism of the conveyor tunnel

The use of a microcontroller (Arduino Mega 2560 Rev30) is involved in the automation of the tunnel. There are color- based indicators (red, orange, and green) which indicate the user about the operation of the machine. When an article enters the chamber, the decontaminating plasma cluster ions and UV-C light act on the article that passes at an approximate speed of 35m/sec.

3.2 Materials

Sabouraud Dextrose Agar (SDA), Soybean casein digest agar, Tryptone soy agar (TSA), and Luria broth were purchased from Sigma-Aldrich, Germany. All the solutions were prepared in MQ water (Milli-Q grade derived from Millipore water system model Elix 3, Millipore Corp., USA). All other chemicals used were of analytical grade and used as received without further purification.

3.3 Method

3.3.1Evaluation of the antimicrobial efficacy of CoVDecon

All the anti-microbial studies were conducted by Apex testing and research laboratory, New Delhi, India (ISO 45001:2018 certified laboratory). ISO 18593:2018 guidelines were used for the evaluation of the antimicrobial efficacy of the CoVDecon. Prior to the start of the experiment, UVC lights were kept on for 15 minutes followed by the starting of the plasma generator[14,15].

3.3.1.1 Evaluation of the antibacterial efficacy of the conveyor belt. The antibacterial activity of the station was evaluated against two bacterial species *Staphylococcus aureus* (NCIM 2127) and *Escherichia coli* (NCIM 5346) with the protocol of Beck et al. with slight modifications[16]. Briefly, a solution of 2% Luria broth was prepared in MQ water, autoclaved, and cooled down to room temperature. This was followed by inoculation with bacteria using loop wire and then incubating for 24 hours at 37°C. The absorption was measured at 625 nm. Then, tenfold serial dilutions were performed to prepare a control sample and the rest was stored for future. One ml of the prepared microbial culture (approx. 10^5cfu/ml) of both the gram- positive and gram-negative bacterial strains was poured onto sterile wooden boards of size 10cm^2 and air-dried at room temperature. Contaminated wooden boards were kept at a predefined location inside the tunnel. The machine was turned on and the wooden board was passed to the conveyor at different speeds shown in Table 2. After completing the cycle inside the tunnel, wooden boards were taken out from the station and swab samples were collected. Immediately, the collected swab samples were diluted and plated out on specific marked Soybean casein digest agar (SDA) media plates and incubated for 24 h at 37°C in an incubator (Bacteriological Incubator, Atico Medical, New Delhi, India). The colonies were counted after the incubation period to observe the reduction of microbial load. Bacterial growth reduction(R) was then calculated from the formula given below in

Equation (1).

$$R = 100 \times B - A/B \quad (1)$$

Where;

B=No. of colonies in the blank; and A=No. of colonies in a sample.

3.3.1.2 Evaluation of the antiviral efficacy of the conveyor belt. CoVDecon was tested against Bacteriophage MS2 (*Emesvirus zinderi*) (*Escherichia coli* ATCC 15597B1) according to the manufacturer and Beck et al. protocol with slight modifications[16]. The culture suspension was prepared from a 24-hour hold stock culture. After that, 1 ml (approx.105 pfu/ml) of the above culture suspension was applied on the surface of 10cm^2 area of sterile wooden board and dried at room temperature. A swab sample was collected at an area of 10cm^2 from the contaminated wooden board and was used as the control sample. The samples were kept at a predefined location inside the tunnel and passed at different conveyor speeds, as reported in Table 4. After completion of the cycle, wooden boards were taken out from the station and swab samples were collected. Immediately, the collected swab samples were diluted and plated out on specific marked Escherichia coli (ATCC700891) culture in Tryptone soy agar (TSA) plates and incubated for 24 hour at 37 °C in an incubator (Bacteriological Incubator, Atico Medical, New Delhi, India). The colonies were counted after the incubation period to observe the reduction of microbial load. Bacterial growth reduction (R) was then calculated from the formula given in Equation (1).

3.3.1.3 Evaluation of the antifungal efficacy of the conveyor belt. Briefly, a suspension of fungal spores (*Aspergillus* species (MTCC 1344)) was prepared by adding 10 ml of a sterile 0.5% saline solution containing 0.05% of a non-fungicidal wetting agent to a 7–10-day old culture already prepared in a lab. From this stock culture, an inoculum was gently scraped with a platinum wire and added to the saline solution. This suspension of fungal culture was shaken vigorously to break up clumps of spores, if any, followed by filtering through a thin layer of sterile cotton. The inoculum sample for testing was adjusted using a colony counter (Petroff-Hausser, Hausser Scientific, Pennsylvania USA) to contain 5×10^6 fungal cells/ml on the day of use by appropriate dilution of stock suspension with saline solution. The fungal suspension (approx.5×10^6fungal cells/ml) of 1 ml of each was poured onto a sterile wooden board of size 10cm^2 and air-dried at room temperature inside a closed system. Contaminated wooden boards were kept at a predefined location inside the tunnel at different conveyor speeds as mentioned in Table 2. After completing the cycle of the contaminated wooden blocks inside the tunnel, swabs were taken from the artificially contaminated 25cm^2 areas of wooden boards for determining the number of cells/ml. Immediately, the collected swab samples were diluted and plated out on specifically marked SDA media plates and incubated for 120 hours at 25 °C in an incubator (Bacteriological Incubator, Atico Medical, New Delhi, India). After 120 hours, each plate was observed visually for fungal growth. The colonies were counted after the incubation period to observe the reduction of fungal load[17].

4 Results and Discussion

4.1 Results

4.1.1 Antibacterial efficacy of the conveyor belt

The results of the antibacterial activity are represented in the Figure 2(a to d) and summarized in Table 1.

The result shows that the average percentage elimination of both the bacterial strains was 99.9%. In the first round, *Escherichia coli* contaminated wooden boards, running at a speed of 3m/sec, was decontaminated with an efficacy of 89%. Interestingly, in the second round, the decontamination efficacy drastically improved to 95.8%. In the case of Staphylococcus aureus, in the first round, at minimum speed of 3m/sec, 88% efficacy was obtained. The efficacy jumped to 95%, in the second round, when the conveyor was running at the same speed of 3m/sec. It was interesting to note that, when the wooden board was placed for the third round with the same speed (3m/sec), the decontamination efficacy rose to 99% against both the strains. Similarly, if the article was held for 15 minutes, 99.9% of the bacteria were eliminated. Following the previous procedure, the contaminated wooden board was again kept on the conveyor with an optimum speed of 35m/sec. For the same, the hold time of 15 minutes showed the best result with 99.9% decontamination of the bacteria. At the speed of 35m/sec, the average elimination of *Escherichia Coli* was 78.3% and *Staphylococcus aureus* was 78.9% in the first round. In the second round, the average elimination of *Escherichia coli* and *Staphylococcus aureus* was 86.3% and 85.8% respectively. A third round was performed and the average elimination of *Escherichia coli* was 97.3% and *Staphylococcus aureus* was 96.9%.

Fig 2. (a) Presence of *Escherichia coli* on the plate after taking a swab from the contaminated wooden board before passing it through CoVDecon. (b) After passing contaminated wooden boards through CoVDecon. (c) After passing the contaminated wooden board through CoVDecon. (d) Presence of the *Staphylococcus aureus* on the contaminated wooden boards before passing it through CoVDecon

Table 1. Percentage elimination of the test organism *Escherichia coli*and *Staphylococcus aureus* after passing the contaminated wooden board through CoVDecon

S.No.	Parameters	Results (Percentage Elimination)	Test Organisms	Test Method
	Microbiological Parameter	-	-	-
	UV Disinfection	-	-	-
I.	**Sterilization technique (Swab test)**	-	-	-
(a)	Hold Time 15 minutes (middle section)	99.9	*Escherichia coli*	In House Method Reference no. ISO 18593
(b)	At min speed (3m/s) of a belt (1st round)	89.4	*Escherichia coli*	In House Method Reference no. ISO 18593
(c)	At min speed (3m/s) of a belt (2nd round)	95.8	*Escherichia coli*	In House Method Reference no. ISO 18593
(d)	At min speed (3m/s) of a belt (3rd round)	99.9	*Escherichia coli*	In House Method Reference no. ISO 18593
(e)	At optimum speed (35m/s) of a belt (1st round)	78.3	*Escherichia coli*	In House Method Reference no. ISO 18593
(f)	At optimum speed (35m/s) of a belt (2nd round)	86.3	*Escherichia coli*	In House Method Reference no. ISO 18593
(g)	At an optimum speed of (35m/s) belt (3rd round)	97.3	*Escherichia coli*	In House Method Reference no. ISO 18593
II.	**Sterilization technique (Swab test)**		-	-
(a)	Hold Time 15 minutes (middle section)	99.9	*Staphylococcus aureus*	In House Method ISO 18593
(b)	At min speed (3m/s) of a belt (1st round)	88.9	*Staphylococcus aureus*	In House Method ISO 18593
(c)	At min speed (3m/s) of a belt (2nd round)	95.4	*Staphylococcus aureus*	In House Method ISO 18593
(d)	At min speed (3m/s) of a belt (3rd round)	99.9	*Staphylococcus aureus*	In House Method ISO 18593
(e)	At optimum speed (35m/s) of a belt (1st round	78.9	*Staphylococcus aureus*	In House Method ISO 18593
(f)	At optimum speed (35m/s) of a belt (2nd round)	85.8	*Staphylococcus aureus*	In House Method ISO 18593
(g)	At an optimum speed of (35m/s) belt (3rd round)	96.9	*Staphylococcus aureus*	In House Method ISO 18593

4.1.2 Antiviral efficacy of the conveyor belt

The viral elimination percentage was 99.9%. The contaminated wooden boards were tested under the station at a minimum speed of 3m/sec. In the first round it was capable to eliminate 80.2% of the overall viral load. In the second round, the average elimination was 91.6%. In the third round CoVDecon eliminates an average 99.9% of viral load.

Next, the speed of the conveyor was increased to 35m/sec in which the first- round average elimination was 65.6%. Whereas the second round was able to eliminate an average of 80.1% and in the third-round average elimination was 97.1%. Table 2 and Figure 3 provide a summary of the antiviral efficacy of the machine.

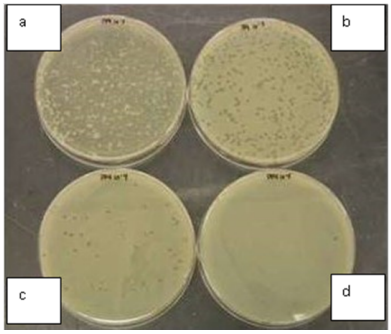

Fig 3. a) Presence of Bacteriophage MS2 on the plate before passing it through CoVDecon conveyor tunnel. (b) Presence of Bacteriophage MS2 after the plate was passed through the CoVDecon conveyor tunnel with a speed of 3m/sec in the first round. (c) Presence of BacteriophageMS2 on the plate after passing it through CovDecon with a speed of 3m/sec in the second round. (d) Presence of Bacteriophage MS2 after the plate was passed through CovDecon with a speed of 3m/sec in the third round.

Table 2. Percentage elimination of the MS2 Phage after decontamination by CoVDecon

S.No.	Parameters	Results (Percentage Elimination)	Test Organisms	Test Method
	Microbiological Parameter	-	-	-
I.	**Sterilization technique (Swab test)**	-	-	-
(a)	Hold Time 15 minutes (middle section)	99.9	Bacteriophage MS2	In House Method ISO 18593
(b)	At min speed (3m/s) of a belt (1st round)	80.2	Bacteriophage MS2	In House Method ISO 18593
(c)	At min speed (3m/s) of a belt (2nd round)	91.6	Bacteriophage MS2	In House Method ISO 18593
(d)	At min speed (3m/s) of a belt (3rd round)	99.9	Bacteriophage MS2	In House Method ISO 18593
(e)	At optimum speed (35m/s) of a belt (1st round)	65.6	Bacteriophage MS2	In House Method ISO 18593
(f)	At optimum speed (35m/s) of a belt (2nd round)	80.1	Bacteriophage MS2	In House Method ISO 18593
(g)	At an optimum speed of (35m/s) belt (3rd round)	97.1	Bacteriophage MS2	In House Method ISO 18593

4.1.3 Antifungal efficacy of the conveyor belt

The fungal inhibition activity of CoVDecon was investigated by inhibiting the growth of *Aspergillus brasiliensis*. The results of antifungal activity are shown in Figure 4 and summarized in Table 3.

While fungal growth was observed on the control plate (a), no fungal growth could be seen on the contaminated wooden board (b), which was passed through CoVDecon. Thus, the results confirm efficient decontamination activity of CoVDecon against Aspergillus species. The anti-fungal test was conducted by placing the contaminated wooden board in the middle of the conveyor for 15 minutes and the decontamination efficacy was found to be 99.9%. When the board was placed inside the

Fig 4. (a) Growth of the *Aspergillus brasiliensis* on the plate contaminated by the swab taken from the contaminated wooden board before passing it through CoVDecon . (b) Absence of the fungi on the plate after the contaminated wooden board was passed under the CoVDecon.

Table 3. Percentage elimination of the test organism *Aspergillus Brasiliensis* through an in house method bypassing contaminated wooden board under the CoVDecon

S.No.	Parameters	Results (Percentage Elimination)	Test Organisms	Test Method
	Microbiological Parameter	-	-	-
	UV Disinfection	-	-	-
I.	**Sterilization technique (Swab test)**	-	-	-
(a)	Hold Time 15 minutes (middle section)	99.9	*Aspergillus brasiliensis*	In House Method ISO 18593
(b)	At min speed (3m/s) of a belt (1st round)	84.2	*Aspergillus brasiliensis*	In House Method ISO 18593
(c)	At min speed (3m/s) of a belt (2nd round)	93.7	*Aspergillus brasiliensis*	In House Method ISO 18593
(d)	At min speed (3m/s) of a belt (3rd round)	99.9	*Aspergillus brasiliensis*	In House Method ISO 18593
(e)	At optimum speed (35m/s) of a belt (1st round)	73.5	*Aspergillus brasiliensis*	In House Method ISO 18593
(f)	At optimum speed (35m/s) of a belt (2nd round)	83.9	*Aspergillus brasiliensis*	In House Method ISO 18593
(g)	At an optimum speed of (35m/s) belt (3rd round)	95.2	*Aspergillus brasiliensis*	In House Method ISO 18593

conveyor at a minimum speed of 3m/sec in the first round, 84.2% of the fungal spores were found to be eliminated. In the second round, the average elimination was found to be 93.7%. In the third round, the conveyor eliminates showed antifungal efficacy of 99.9%.

Then, the speed of the conveyor was increased to 35m/sec, in which, the first-round average elimination was 73.5%, whereas, in the second round and third round the average elimination increased to 83.9% and 95.2% respectively.

4.2 Comparative analysis of results with existing technologies

An extensive literature survey was conducted to determine the complete antimicrobial effectivity of the various existing decontaminating and disinfecting technologies used in a built-in environment. Primarily, chemical disinfectants and sterilizing agents were reviewed, and it was found that bleaching agents showed 99% effectivity only after an exposure time of 10 mins against gram-positive and gram-negative strains. This was followed by hydrogen peroxide-based agents such as Prevail™, surfactant and quaternary ammonium compound based agent like HLDL™ etc. Moreover, it was concluded from the study that efficacy was directly proportional to the concentration and the holding time of the chemical disinfectants with the materials [18]. Glutaraldehyde at a concentration of >2% when combined with isopropyl alcohol at 26% concentration, required a holding time of 15 mins and chlorine derivatives-based compounds such as hypochlorous acid requires a holding time of 15 mins [19]. Some devices developed such as micro aerosol decontamination technology (PAEROSOL), which uses atomized table salt, took an exposure time of 30 mins to exhibit 99.99% antibacterial and antiviral efficacy [20]. No-touch decontamination technologies

such as Altapure High-Level disinfection cabinet (Mequonm WI) which use peracetic acid and hydrogen peroxide aerosols, along with a UV-C box required an exposure time of 21 mins to completely disinfect the object as compared to the general 10 minutes taken by UV-C chambers [21,22]. An automatic disinfection system was also developed to be used at airports, train/bus stations that use TUV bulbs and non-foaming soap solution system but was not tested extensively. Table 4 provides a brief description of the current technologies and strategies employed for sterilization purposes [23].

Table 4. Current technologies and strategies employed for sterilization of different articles, their advantages and disadvantages [4,7,23]

Techniques	Articles used on	Advantages	Disadvantages
Chemical			
1. Alcohol	Devices (computers, ECG machine etc.), equipment (stethoscopes), tools (scissors, scalpel) etc.	Rapid bactericidal, fungicidal and virucidal activity	Inflammable, evaporates rapidly, short exposure time, non-sporicidal
2. Chlorine derivatives	Equipments(PPE kits)	Broad spectrum antimicrobial	Decomposition, corrosive, cytotoxic, release of toxic chlorine gas
3.Gluteraldehyde	Endoscopic equipments, dialyzers, metal and plastic equipemnts	Excellent biocidal properties; non-corrosive to some equipments such as (rubber, plastic etc.)	Needs to be activated by alkalinizing agents to become sporicidal, short shelf-life, toxic above 2%
4. Peroxides	Medical equipment	Non-toxic, high antimicrobial property	Short-term stability
Physical			
1. Steam	Medical devices, glass equipment, packaging materials	Non-toxic, rapid antimicrobial activity, active penetration for enhanced antimicrobial activity	Potential for burning, can destroy heat sensitive equipments; cannot be used on electronic gadgets as moisture can cause damage
1. Hydrogen gas plasma	Most devices and equipment	Less cycle time, can be used for heat sensitive equipment, non-toxic, user-friendly	Hydrogen peroxide may be toxic at higher concentrations, cannot be used on linens, paper , liquids etc.
2. UV radiation	Most equipment and devices	Highly effective antimicrobial activity	Efficiency dependent on intensity, placement of lamp

5 Discussion

Currently, the potential of an outbreak of severely infectious and deadly diseases is a global concern. It is of critical importance to find disinfection methods and equipment that can be used to avoid cross-contamination and disease transmission.

Many strategies exist to decontaminate open rooms or closed environments. But as mentioned above, they cannot be used for complete decontamination of articles in built-in environments such as airports, stations, universities which are the prime locations of cross-contamination. Although use of UV sterilization techniques has been proven to be beneficial, a major drawback is the inability of the UV rays to reach the pockets and cavities on the articles. Similarly, even hydrogen peroxide vapors are unable to reach confined spaces and pockets on the articles, thus, reducing the decontamination efficacy. Further, the chemical disinfectants and ozone sterilizers have corrosive properties and could potentially damage the material, especially sensitive materials. This limits the versatility and applicability of these technologies in multiple scenarios.

Further, it has been observed that varied range of UV energy is required for the effective decontamination of biological contaminants. UV-C has been observed to have the ability to reduce viral load at ~ 120 secs. However, some contaminants such as non-enveloped viruses cannot be decontaminated by UVC irradiation. Therefore, the use of only UVC is not the most feasible solution [24].

On use of plasma-based systems, using gases such as oxygen, argon etc. the reduction in colony forming units (CFU) was observed within 1hr. Further, studies by Ekem et al., 2011 has shown that the plasma can inactivate 3.5 CFU log units of S. aureus within 12 mins of treatment. It has also been observed in a study conducted by Lukes, et. al., 2012, that plasma activated water takes 30 mins to reduce log CFU units to <2.1 [25].

Evaluation of results highlights the fact that although strategies and technologies are equipped for decontamination, most of the studies conducted were focused on surface decontamination. This implies that if the contaminants enter the internal pockets or cavities of the articles, these techniques will be incapable of eliminating the contaminants. Further, on comparing the data from the literature survey with the results obtained by the developed conveyor belt, it is evident that the developed machine is a more efficient, cost effective and eco-friendly alternative. The technologies equipped in the CoVDecon machine can effectively decontaminate the entire article completely including any pockets or grooves in the article. This provides a holistic approach for effective decontamination of any article irrespective of shape of the article. This is evident from the fact that the CoVDecon machine shows 99.99% efficacy within 3 rounds at a constant speed of 3m/s and within a holding time of just 15 minutes.

The results revealed that regardless of the experimental environment used, a strong ion inhibitory effect on the survival of free bacteria was observed. The anti-bacterial efficacy against both Gram-positive (*S. aureus*) and Gram-negative (*E. coli*) was found to be extremely effective at a 3m/sec with a 99% elimination after the third round. Similarly, in case of fungal species, the machine was able to successfully decontaminate the article with 99% efficacy after the third round. Similar results were observed against MS2 bacteriophage as well. However, it was observed that the anti-microbial efficacy reduced to 97.1%, 95.2% and 96.9% against virus, fungi and bacteria respectively. The decrease in efficacy could be the result of a shorter exposure time to plasma ion and UV radiation in case of 35m/sec. Exposure time plays a significant role in this scenario, as can be observed from the results wherein, a 15-minute hold time of the article inside the machine showed a high antimicrobial efficacy at 99.9%. Therefore, 3 rounds at 3m/sec could be identified as the ideal speed for appropriate exposure. The concept of decontaminating an article' surface using UV and plasma cluster ions and targeting not just the surface but also the pockets and grooves has been demonstrated in the current study. The novel triple shield technology is capable of successfully decontaminating various microbial contaminants with extremely high efficacy.

6 Conclusion

The developed equipment 'CoVDecon' conveyor tunnel shows immense prospect in considerably reducing indirect transmission and cross-contamination of potentially infectious and deadly pathogens. The strategy of a two-pronged approach of UVC irradiation combined with plasma cluster ions ensures successful decontamination of biological agents and thus, also add as precaution against future threats both intentional and unintentional. According to the evaluation conducted, the developed machine has a 99% efficacy against bacterial, fungal and viral strains within just 3 rounds run at an average speed of 3m/sec. This means that the machine portrays greater efficacy than the current devices available in the market. The developed equipment ensures negligible environmental toxicity, with high antimicrobial efficacy making it a suitable product to be installed by healthcare facilities, airports, train stations, hotels, industries etc. The added benefit is the universality and the ability to decontaminate not just bacterial pathogens, but also viral and fungal pathogens without corrosive or degradation of the object. Finally, waste management are also insignificant as only water is used and can be managed easily. Finally, most devices designed have been theorized for full antimicrobial efficacy but the machine mentioned in the current research has been evaluated and validated for high antimicrobial efficacy.

Comparative studies were conducted only theoretically and via extensive literature review because the machine was developed as a time-sensitive emergency solution against Coronavirus disease 19 (COVID-19). Future studies can incorporate rigorous studies against multiple decontamination systems available commercially to develop higher efficacy systems with multiple prospects.

Further strategies and technologies can be evaluated and the developed machine can be improved to provide a universal solution against a multitude of threats. The future scope should be focused on a universal machine to mitigate CBRN threats.

Abbreviations

SARS-CoV2- Severe acute respiratory syndrome coronavirus 2; UV-C- Ultraviolet-C; DNA- Deoxyribonucleic acid; RNA- Ribonucleic acid; RSDL- Reactive skin decontamination lotion; SDA- Sabouraud Dextrose Agar; TSA- Tryptone soy agar; MQ - Milli-Q grade; SS- Stainless steel; Pfu- Plaque forming units; CFU- Colony forming units; COVID-19- Coronavirus disease 19

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