Cold Plasma: Clean Technology to Destroy Pathogenic Micro-organisms

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Abstract

Atmospheric pressure cold plasma is a promising technology in fighting pathogenic micro-organisms. In times of Covid-19 pandemic, we have decided to modify two types of cold plasma devices to study their effectiveness in the killing of pathogenic micro-organisms. Our studies have shown that both the devices are efficient in this purpose. While pencil like microwave based device can destroy *Aeromonas* bacteria and bacteriophage from 6 cm distance in 2 minutes, the larger (~ 40 cm^2) RF plasma based device could do the similar killing ability for the larger possible area in 4 minutes. OES studies revealed that both these device produce OH radicals which helped in the destruction of both bacteria and bacteriophage. With suitable modification, these devices, especially the larger area device may even be implemented for the elimination of Covid-19 affected wards of hospital without using any sensitive chemical process.

Introduction

Pathogenic micro-organisms are among the oldest known reasons for diseases in humans. In spite of great technological advancement of human race, medical science often rendered helpless in front of a pathogenic outbreak. In recent years, SARS, Ebola and ongoing Covid-19 have all shown an urgent requirement to develop novel approaches to destroy them. Traditional sterilization processes for medical products include steam, ethylene oxide (EtO), ionizing radiation (gamma or E-beam), low-temperature steam and formaldehyde, dry heat (hot air) and H_2O_2. Recent developments in sterilization technologies in the last decade focuses on the following factors: low temperature operation so that the technology is suitable for a variety of materials, shorter cycles of operation for faster sterilization, environmental friendliness, and cost reduction **[1]**. Cold plasma can be a useful tool for destroying all types of pathogenic micro-organisms **[1-3]**. Cold plasma can be used in vacuum as well as in atmospheric pressure **[4-7]**. At present, the spread of Covid-19 and other types of novel pathogens have steered the scientific community worldwide to find quick and effective solutions for the sterilization of various medical equipments as well as disinfection of residential units and hospitals. For sterilization of harmful pathogens, atmospheric pressure cold plasma based technologies has been successfully utilized for the killing of laboratory strain of bacteria (*Aeromonas*) and its bacteriophage (virus of bacteria).

Two cold plasma devices have been tweaked to suit the application of pathogenic destruction. These devices were also characterized by optical emission spectroscopic (OES) method to determine existence of important active species for destruction of pathogens.

1. Device A: Microwave Cold Plasma Jet and
2. Device B: RF - Hollow Cathode Cold Plasma Device

While device A was originally designed and used for radioactive decontamination **[5]**, we have observed that this device can also effectively destroy *Aeromonas* bacteria and bacteriophage even from 6 cm distance. It can be useful for treating infections on human body including open wounds and also sterilization of medical equipments. Device B was originally used for fabrication of carbon nanostructures **[6, 7]**. However, we found that by carefully changing the gas composition in the plasma and operating power a mere 3 & 4 minute exposure under this plasma can effectively destroy *Aeromonas* bacteria and bacteriophage respectively. These devices can help in sterilization of the pathogens.

Experimental And Device Information

Device A: Microwave Cold Plasma Jet: This pencil-like device (Figure 1 (a)) operates in low power (< 100 watts) at microwave frequency (2.45 GHz). The device has a simplistic design consisting of a stainless steel hollow outer shaft and a solid central electrode along the axis of the hollow shaft. The schematic of experimental set up is shown in the Figure 1 (c). Ar, the plasma forming gas is bubbled through a heated H_2O_2 solution to produce plasma in this case. H_2O_2 decomposes in plasma environment and produces biocidal hydroxyl and hydroproyl free radicals and also H_2O_2 molecules in excited state. The latter is a source of UV radiation which also helps in sterilization process **[8]**.

Device B: RF - Hollow Cathode Cold Plasma Device: It's 13.56 MHz RF source based hollow-cathode cold plasma device whose detailed description already been given elsewhere **[6, 7].** The actual photograph of the device is shown in Figure 1 (b). This device is targeted to produce large area cold plasma (presently ~ 40 cm^2) suitable for disinfecting larger area. The schematic of experimental set up is shown in the Figure 1 (d). Here, only Ar gas is sent through the device in a controlled manner to produce plasma. Details of the experiments are mentioned in Table 1.

Table 1: Details of experiments conducted with device A & B.

Instrument	Micro-organisms	Gas Composition	Net Power (Watt)	Exposure Time (Min.)	Distance from plasma (cm)
Device A	*Aeromonas*	10 LPM Argon bubbled through 11% H_2O_2 solution, heated at 75^0C.	50	1	4.5
				2	6
	Bacteriophage	Same as above	50	2	6
Device B	*Aeromonas*	Argon at 18 LPM	60	3	1
	Bacteriophage	Argon at 18 LPM	90	4	2

Results & Discussion

Figure 2 shows the results of cold plasma treatment on the laboratory strain of *Aeromonas* bacteria and bacteriophage virus by both the devices. Standard control sample for bacteria is shown in Figure 2(a). From Figure 2 (b) it can be seen that 2 minutes of plasma treatment form device A can effectively destroy *Aeromonas* even from 6 cm distance (3rd quadrant in Figure 2 (b)) while similar type of result can be obtained in device B (Figure 2 (c)) after 3 minutes of Ar plasma treatment. Figures 2(d) and 2(f) shows control sets for bacteriophage for device A and device B respectively. It is to be mentioned that, virus was spotted on top of a bacterial 'lawn'. The appearance of a clearing means that the virus is active and bacterial cell growth in the area indicates that the virus in inactive. Figure 2(e) and 2(g) shows the effect of cold plasma treatment on bacteriophage by device A and B respectively with the aforementioned experimental parameters. Thus, device A was able to contain growth of bacteriophage from 6 cm distance by 2 minutes of plasma treatment and similar result is obtained in device B with 4 minutes of treatment from 2 cm distance.

It is to be mentioned that bacteriophage has the outer protective layer made of protein similar to Covid-19 and hence more resistant to external sanitizing agents. This virus has also been reported as stronger than coronavirus **[9]**. Since these devices are capable of destroying this bacteriophage, it might be also effective for destroying Covid-19 type of viruses. In the present scenario, both the devices can be useful for sterilization of gloves, masks and other reusable medical tools.

Typical OES spectra recorded during operation of both the devices are shown in Figures 3(a) and (b). Figure 3(a) exhibits typical characteristic emission lines of the plasma species during operation of Device A. Here, Ar gas was bubbled through the 11% H_2O_2 solution to carry H_2O_2 vapour in plasma resulting in the occurrence of characteristic OH peek at 281 & 309 nm **[10]**. This OH radical plays an important role in sterilization process **[8**]. Apart from that, Characteristic H_α line at 656 nm and Ar I and OI lines can also be seen in the spectra. A remarkable fact to notice in the plasma is that although Ar is the major constituent of the plasma, OI line is the strongest among all. Oxygen can get incorporated in the plasma from H_2O_2 and the atmosphere as well. Although the intensity of the OI line does not imply increased dissociation of H_2O_2, the presence of OI, H_α and OH lines indicates effectiveness of this device for destruction of pathogens. Figure 3(b) represents typical characteristic emission lines from plasma during operation of Device B. An interesting fact to note that, in spite of absence of H_2O_2 in plasma, characteristic emission lines of OH, O I are observed along with NH. Different molecular bands of N_2 and NO are also observed here along with Ar I lines. Device B is actually helping in dissociation of atmospheric moisture and air resulting in emission of the above bands.

Obtained results clearly indicate that both these devices can be successfully used to contain the spread of pathogenic micro-organism like Covid-19.

Conclusion

Our findings clearly indicate that both the devices are capable of destruction of pathogenic *Aeromonas* bacteria & its bacteriophage. It is seen that 3 to 4 minutes exposure is sufficient for the killing of bacteria and bacteriophage. It is therefore anticipated that the devices surely have the capabilility of destruction of Covid-19 and similar strain of viruses successfully. The devices show ability of rapid disinfection as compared to presently available plasma sterilization technologies.

Declarations

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Competing Interests: We declare no competing interests.

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Figures

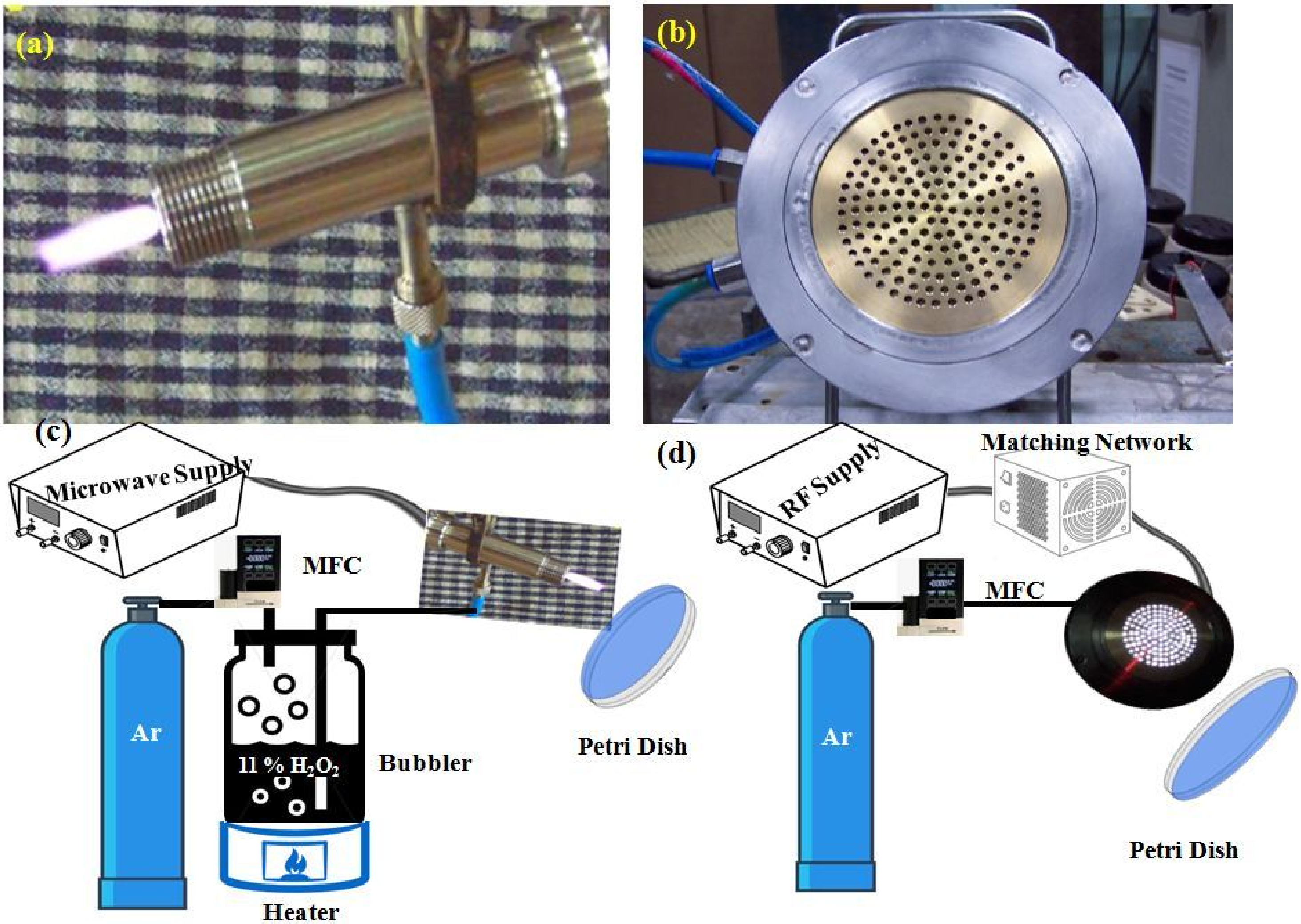

Figure 1

Photograph of (a) device A, (b) device B, schematic of experimental set-up with (c) device A and (d) Device B.

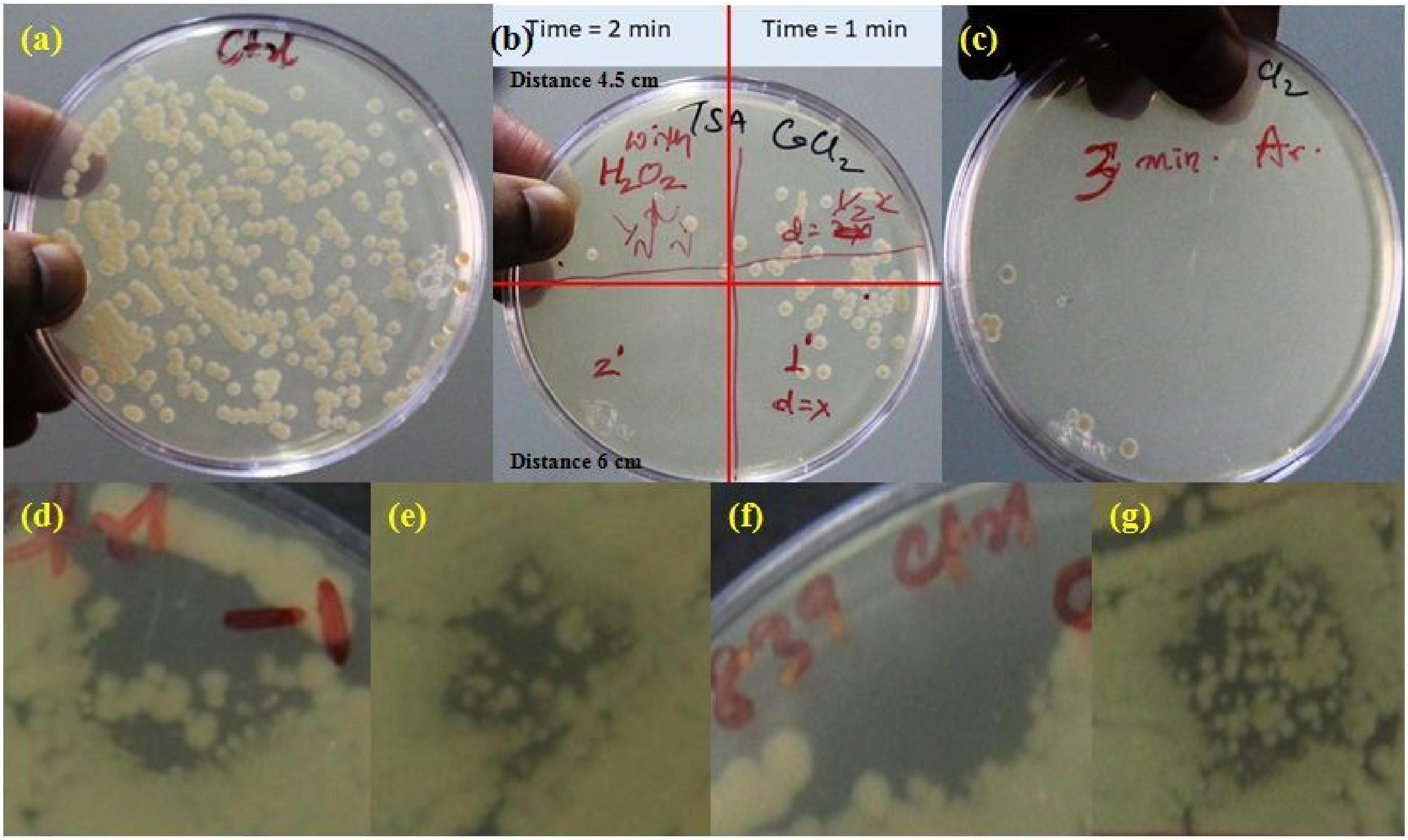

Figure 2

(a) Standard control sample for Aeromonas bacteria, (b) & (c) Growth after 24 hour incubation following cold plasma treatment with device A & device B respectively (d) Standard control sample for bacteriophage for device A, (e) Growth after 24 hour incubation following cold plasma treatment results with device A on bacteriophage, (f) Standard control sample for bacteriophage for device B, (g) Growth after 24 hour incubation following cold plasma treatment with device B on bacteriophage.

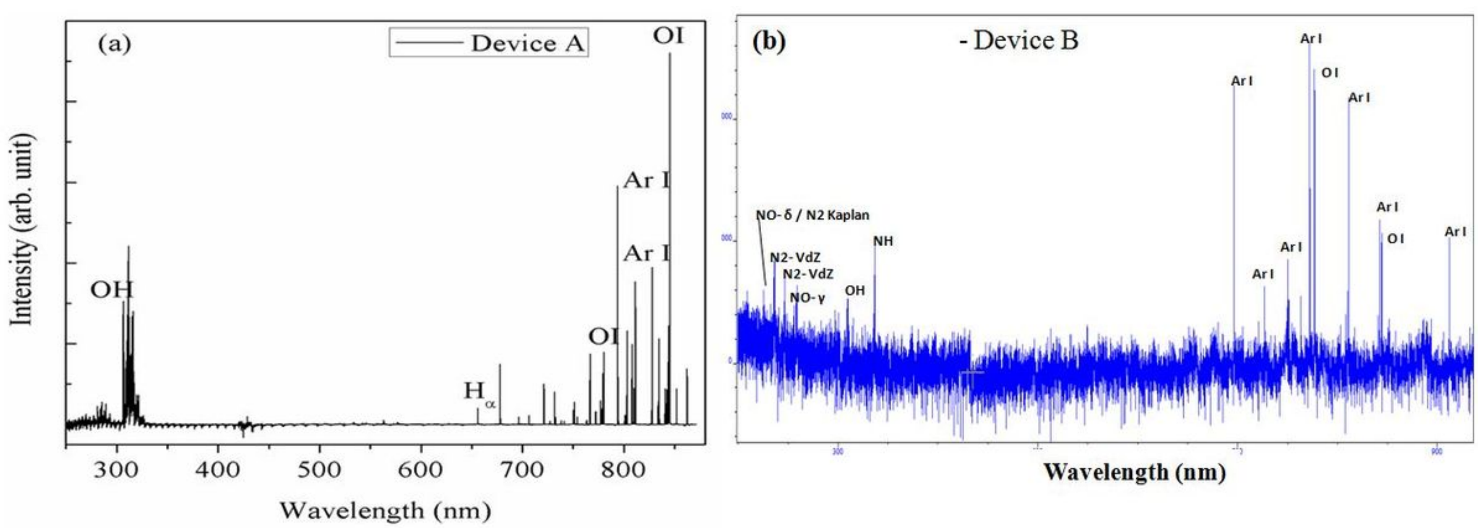

Figure 3

OES signal recorded during operation of (a) Device A and (b) Device B.