

Characterization of the Dielectric Barrier Discharge (DBD) Plasma Jet for Staphylococcus Aureus Bacteria Deactivation

Mohammed K. Khalaf^{1*}, Hadeel H. Alrubaye¹, Faris S. Atallah²

¹. Ministry of Science and Technology, Baghdad, Iraq.

². Department of Physics, College of Science, University of Tikrit; Tikrit, Iraq.

*Corresponding Author: Mohammed K. Khalaf

Abstract

In this paper, the insulation barrier of the dielectric-barrier discharge (DBD) plasma jet is discharged at atmospheric pressure. Where the degradation efficiency of the DBD plasma jet is evaluated on bacteria of Staphylococcus aureus with different exposure times. The influence of the flow rate and applied voltage of the argon gas used is studied and linked to the temperature and column length of the plasma jet. In addition, the Optical Emission Spectroscopy (OES) is examined to detect plasma parameters such as electron temperature and electron density. OES is also used to detect active species within a plasma column such as OI, N₂ and OH and other. The percentage of bacteria growth inhibition is increased directly with increased time of exposure from the DBD plasma jet.

Keywords: Dielectric barrier discharge, Non-thermal plasma, Bacteria inactivation. Optical Emission Spectroscopy.

Introduction

Atmospheric pressure plasma (APP) sources are increasingly used in various processing of materials and biomedical applications because of their inherent plasma stability, their capability of producing high density atomic and molecular species, electron density, low gas temperature, and good reaction chemistry [1]. Plasma jets generate plasma shafts in open space where they rotate different types of active gases [2].

Cold plasma in the atmosphere are the most important subjects or projects, the task of researchers to obtain the desired results for biomedical applications [3, 4]. Non-thermal cold plasma jet in atmospheric are able to inactivate bacteria or fungus or biomacromolecules [5, 6, 7], relying on the jet configuration and the electrical agitation [2, 8].

This work focuses predominantly on the characterization of a DBD type APP jet operated using Ar gas. In contrast to conventional plasma jets, the present work proposes a two-electrode device that can be operated at relatively low voltages. Using

OES diagnostics, various plasma parameters, such as electron density (ne), electron temperature (Te), and gas temperatures (Tg), are systematically determined to evaluate the source performance at various operating conditions. The jet plasma is studied in contact with bacteria of Staphylococcus aureus to investigate the discharge behavior and its feasibility for reactive species/radical generation for bio-applications

Experimental Processes

Plasma Jet Device

The locally home-built produced plasma jet was designed and generated in atmospheric and room temperature conditions using a pyrex tube. The length of the tube is essentially 95mm with a wall thickness of 0.90mm while the external polarity of the tube is 2mm and 3.88mm, respectively. The argon gas is also recharged at the top of the pyrex tube. The gas flow rate using flow meter is fixed at 4 liters per minute for argon (commercial grade 99.9%). The plasma jet is constructed on a double-ring structure as shown in Figure1, with two electrodes made

of aluminum with 0.1mm thickness and 11mm width. These poles have the same measurement and specification, the distance

between the electrodes is 15mm and the distance between the pipe nozzle and the tube 3 mm.

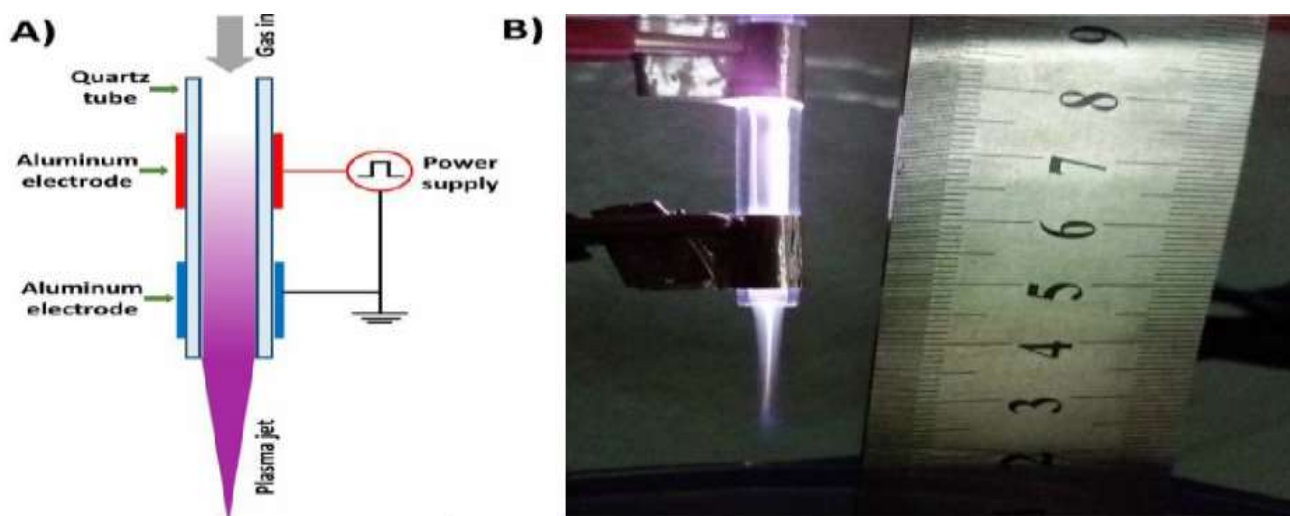


Figure 1: (A) Schematic diagram and (B) photograph of the DBD plasma- two electrodes jets

DBD plasma jet is impelled by using a home-made high voltage power supply with a frequency of 13 KHz and peak to peak voltage of 8 KV. The waveforms of the applied voltage and discharge current, as shown in Figure 2, are registered by a two-

channel using pc USB oscilloscope (Hantek6022 be, with a 20 MHz bandwidth and a 48 MS/s sampling rate) a high-voltage probe(Tektronix p6015) and a current probe(AT-C202).

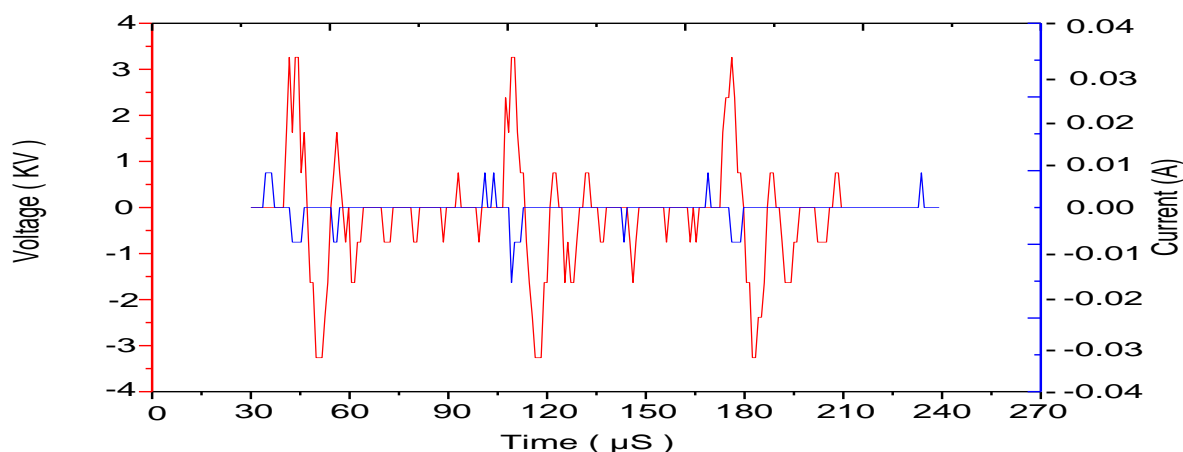


Figure 2: Waveforms of the applied voltage and discharge current

Preparation of Bacteria Suspensions

A fresh *S. aureus* colony was inoculated into 10 mL nutrient BROTH and incubated at 37°C for 24 h. Then the resultant colonies were collected. *S. aureus* colonies were washed into a sterile test tube with 10 mL phosphate buffered saline (PBS), and then the test tube was shaken to make *S. aureus* suspension equably. Finally, *S. aureus* suspension was diluted with PBS and the concentration of *S. aureus* was determined to be approximately 1×10^8 colony-forming units

(CFU) per mL This concentration is measured by spectrophotometer at 625nm, Compared to the McFarland solution. 10mL of bacterial culture were transferred to a sterilized glass petri-dish, The experiment was operated in open air under atmospheric pressure at room temperature of about 25°C. The tube nozzle was placed at the centre of the Petri dish, and 2 cm above its bottom, as shown in Fig. Three treatment time intervals of 3minutes, 5minutes and 10minutes were set, with one Petri dish placed in each

treatment. then Dilutions were made from treated samples and 10 μ l for each dilution were transferred to nutrient agar plates by using spreading technique the sample plates were incubated over night at 37°C to allow survivors to grow Plates were incubated overnight after plasma treatment.

Then, the survival colonies were counted with the method of Colony Forming Units (CFU) was counted in order to check the efficiency of bacterial inactivation using cold plasma treatment. The killing log value (KLV) of the bacteria treated for different treating durations were calculated.

Results and Discussion

Plasma Discharges

Figure 3 shows the plasma temperature (T_g) against different flow rate; it is measure by thermometer using different flow rates at the distance of 2.3 cm from the nozzle to tube at the plasma condition of 13KHz and peak to peak voltage of 8 KV. The results shows that the gas temperature is 53°C at argon gas flow rate of 1 slm, whereas the gas temperature reached 34 °C at argon flow rate of 5 slm.

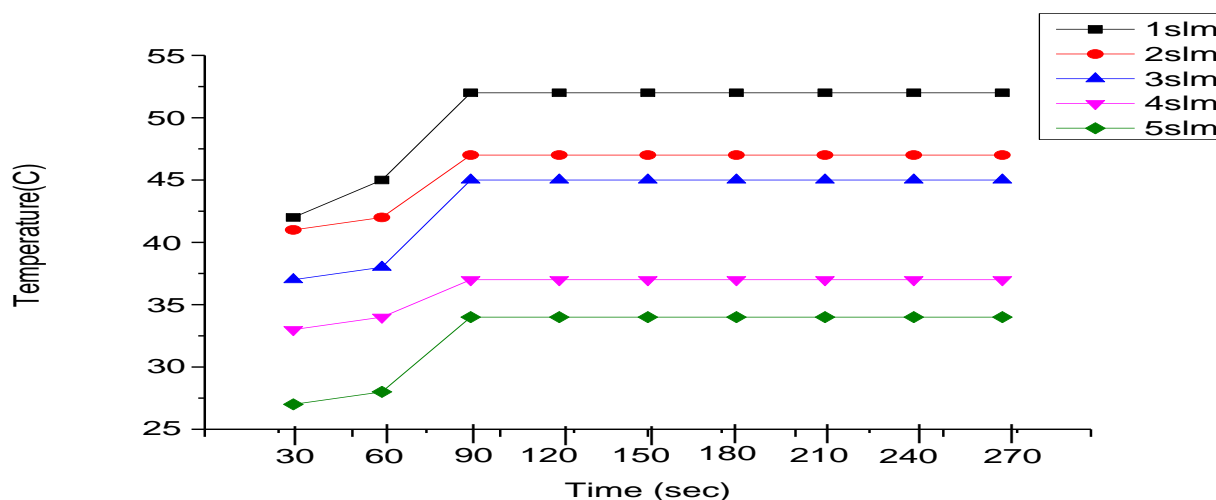


Figure 3: Plasma temperature with different flow rate

The length of the plasma column is measured from the end of the tube to the end of the plasma plume, where the length of the plasma inflator increases with the applied voltage increase. By increasing the gas flow rate, the length of the plasma column

increases up to nearly 22 cm, and then starts to decrease as the gas flow is further enhanced to 5 slm. The longest and brightest plasma observed at a pressure of 4 slm is as shown in Fig. 4.

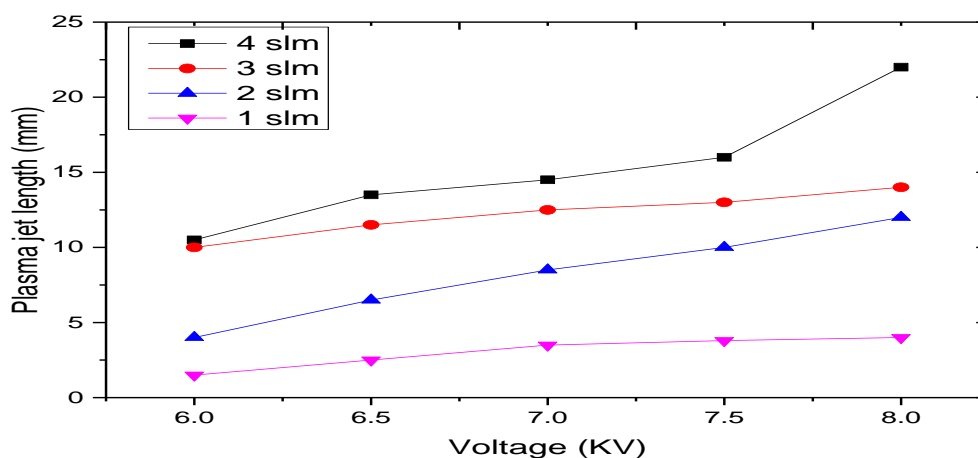


Figure 4: The length of the plasma column shows the function of the voltages with the gas flow rate

Paschen's law can be utilized here to explain the gas pressure which is related to flow rate and the influence of voltage on the length of

the plasma column. The particle concentration in the plasma tube and jet is relatively lesser at a lower flow rate (gas

pressure), which leads to a lower collision rate between the electrons and molecules. At high flow rate, the mean free path of the electrons is too short for the electrons to gain enough energy to ionize the gas molecules. Hence, the flow rate and pressure has a most appropriate value for an electron avalanche. According to the law, the statistic breakdown voltage us is higher in both the low and high flow rate and pressure cases.

So as flow rate increases, the breakdown voltage decreases at first, and then rises. The overvoltage $U-U_s = U$ rises at first and then decreases at a fixed applied voltage, which results in that the length of the plasma column first increases and decreases afterward as the flow rate increases. Similarly, setting the flow rate at a specific value, increasing only the voltage leads to a

larger U , which results in the growth of the plasma column. In addition to that, a larger U means the electrons get more energy initially [8, 9]. Spectroscopy good instrument to calculate the electron temperature and density of argon plasma jet in range of wavelength 300-900 nm. Spectrum shows numerous peaks most of them belonged to ArI which correspond to NIST data [10,11], as shown in table 1. The emission spectra of the DBD plasma jet is measure by spectrometer (SV2100. K-MAC) with the spectra range of 300-925nm, whereas the optical fiber is located at 11mm from the edge of pyrex tube. Through OES, the active particles present within the plasma column can be obtained by OES analysis. The spectral lines of the DBD argon are shown at 13 KHz, 8 KVp-p, and 4slm as in the Figure 5.

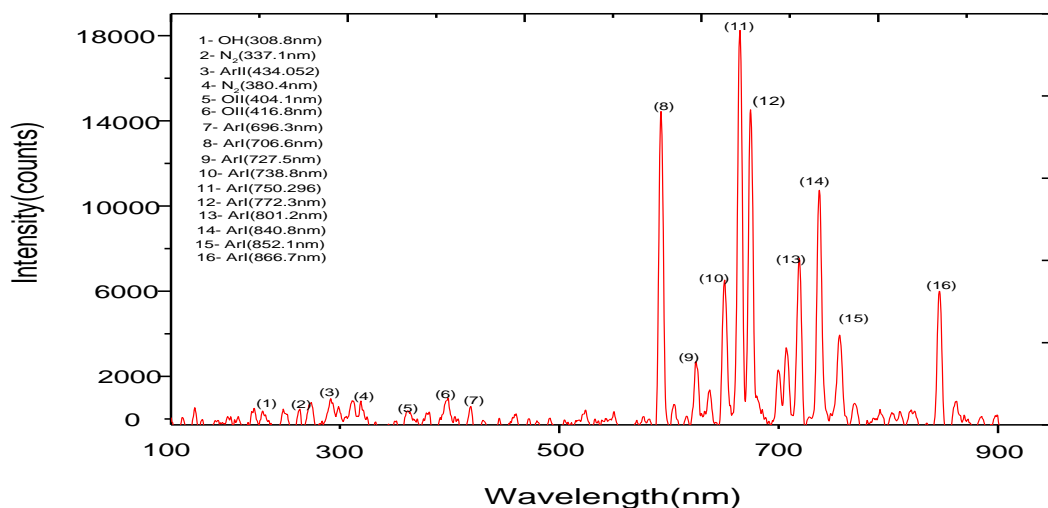


Figure 5: The emission spectra of Ar DBD plasma jet

The electron temperature (T_e) and electron density (n_e) are important parameters to prescribe the characteristics of the Argon

DBD plasma jet. Electron temperature can be determined by using a ratio method given by:

$$\frac{I_1}{I_2} = \left(\frac{\lambda_{nm,z}}{\lambda_{ki,z}} \right) \left(\frac{A_{ki,z}}{A_{nm,z}} \right) \left(\frac{g_{k,z}}{g_{n,z}} \right) e^{\left(\frac{E_{k,z} - E_{n,z}}{KT_e} \right)} \quad (1)$$

Where I_1 is the line intensity from the k_i transition and

I_2 is that from the nm transition.

Table 1: values of transition probability, upper energy level and statistical weight that used to calculate T_e

Wavelength (nm)		A, (S ⁻¹)		g		E, (eV)	
ArI	ArII	A _{ki}	A _{nm}	g _{ki}	g _m	E _{ki}	E _{nm}
750.296	434.052	4.45x10 ⁷	1.17x10 ⁸	3	6	11.8280	16.6438

The electron density can be determined by using

Saha- Boltzmann equation, given by [8].

$$n_e = \frac{6.04 \times 10^{21} I_2^*}{I_{Z+1}^*} T_e^{\frac{3}{2}} \exp \left[\frac{-E_{k,Z+1} + E_{k,Z} - x_Z}{k_B T_e} \right] \text{cm}^{-3} \dots\dots\dots (2)$$

The electron density was estimated to be

$n_e = 1.88 \times 10^{13} \text{cm}^{-3}$ and electron temperature $T_e = 1.67 \text{eV}$.

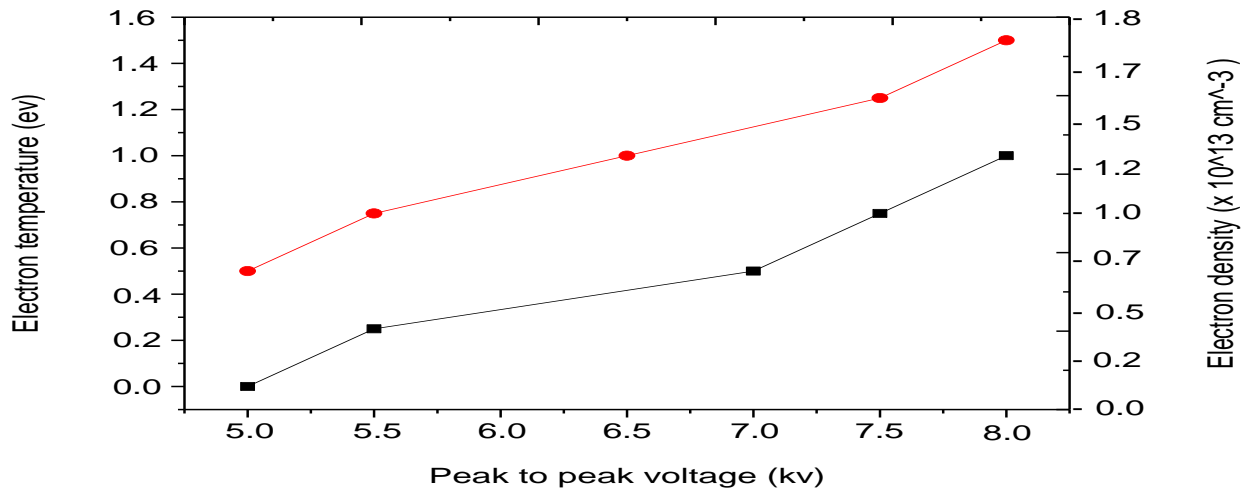


Figure 6: Electron density and electron temperature as a function of peak to peak applied voltage

Survival Images of *S. aureus*

The images of the surviving bacteria after treatment are shown in Fig. 7, it is seen that the numbers of the surviving bacteria decreased sharply as treating time increased. At a treating time of 10 min. almost all the bacteria were killed. Figure 7 shows the bacterial percentage of inhibition with a

different time exposure of (DBD) plasma jet. It was observed in this figure that the size of inhibition percentage was permanently larger than the diameter of the (DBD) plasma jet. From these results, we believe that the main reasons for the inactivation of bacteria were due to the reactive species of oxygen and nitrogen (RONS) which formed from the surrounding air [2, 5].

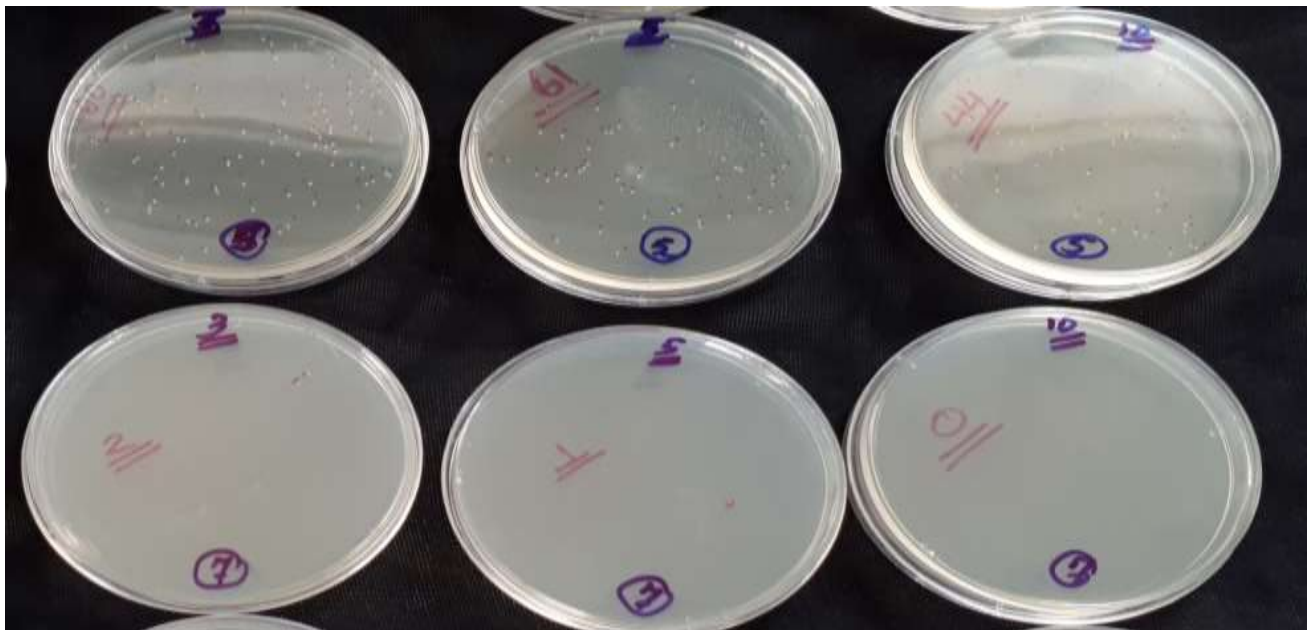


Figure 7: photographs staphylococcus aureus samples on Ar in Petri dishes; 3 min, 5 min, and 10 min

The relationship between the percentages of inhibition percentage with different treatment time can be shown in (Figure

8). Where it is seen that the area of growth inhibition percentage increased with increased the treatment time.

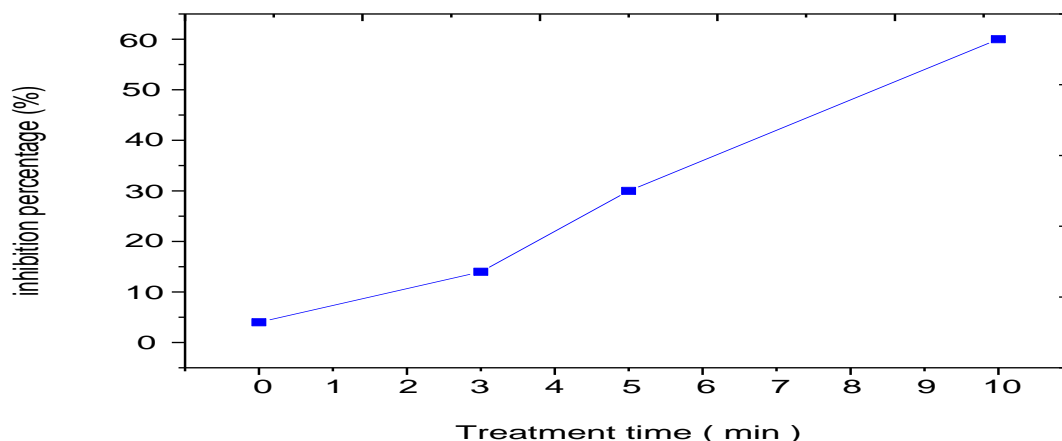


Figure 8: Inhibition percentage as a function of treatment time

Conclusions

Our experimental results strongly suggest that the APPJ originally realized with the double dielectric electrode configuration for DBD is essentially evaluated against staphylococcus aureus bacteria with different time exposure. The argon gas plasma works in atmospheric conditions as well as room temperature, to kill bacteria Staphylococcus

aureus, and the high efficiency plasma jet argon affects bacteria at different times.

Acknowledgments

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