

## Water pollution removal by Non- thermal plasma jet

Mohammed Ubaid Hussein<sup>1</sup>, Rana Talb Mohsen<sup>2</sup>

<sup>1</sup>Department of biochemistry, College of Medicine, Anbar University, Anbar, Iraq.

<sup>2</sup>Department of physiology and medical physics, College of Medicine, Anbar University, Anbar, Iraq.

Corresponding Author: mmphysics361@gmail.com

### To cite this article:

Hussein,M.U.; Mohsen,R.T. Water pollution removal by Non- thermal plasma jet. Mesop. environ. j., 2016, Spicial Issue A.;86-92.

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### Abstract:

In this study used device jet plasma needle with atmospheric pressure which generates non thermal plasma jet to measure treatment potent with plasma against bacteria which contamination water, where, *Bacillus cereus* and *Escherichia coli* bacteria were inactivated with plasma at 40 sec, where bacteria respond for treatment according to bacteria type.

Bio-decontamination of water and surfaces contaminated by bacteria (*Bacillus cereus*, *Escherichia coli*) was investigated in atmospheric pressure air, in plasma needle. Electro-spraying of the treated water through the needle resulted in fast bio-decontamination, with radicals and reactive oxygen species seem to be dominant biocidal agents.

**Keywords:** non thermal plasma jet, *B. cereus*, *E. coli*, plasma needle.

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### Introduction

Plasma is defined as the fourth state of matter. It is an ionized gas containing free moving charge carriers: electrons and ions. Over 99% of the visible universe is made up of plasma [1].

Non-thermal atmospheric pressure plasmas are very effective in killing bacteria. This makes these plasmas very useful for various biological and medical applications, such as: sterilization of medical instruments, decontamination in biological warfare and air filters in hospitals [2].

The plasma needle is a type of non-thermal atmospheric glow discharge, it has a single electrode configuration and is operate by different noble gas (He-Ar), important properties of this type of plasma are that it operate at near room temperature, the plasma does not cause any thermal damage to articles it comes in contact with. This characteristic was open up the possibility to use this plasma for treatment of the heat sensitive materials. Atmospheric pressure discharge plasma is of great interest because of their low costs and simplified operation [3].

Non –equilibrium plasma at atmospheric pressure finds numerous biological and bio-medical applications thanks to their reactive nature .It has been tested on large variety of bacteria ,spores ,viruses for their sterilization and interactions of plasma with live tissue ,e.g. skin disinfection ,blood coagulation ,wound healing ,density[4,5].

In bio-decontamination by plasma, it is crucial to understand the role of various mechanisms involved.

The significant mechanisms depend on the plasma composition (gas), temperature, treated microorganisms and the environment (air, water, surfaces, etc.) [6].

*Escherichia coli* (*E. coli*) is a classic opportunistic pathogen found in hospitals. The World Health Organization professes that this bacterium is one of the primary pathogens of hospital acquired infection. *E. coli* contributing to a large percentage of nosocomial infections ranks first in the infection rate of various gram-negative nosocomial pathogens. In recent years, because of the multi-drug resistant mechanism of *E. coli*, infection incidents have occurred frequently, and the drug-resistance of the bacterium has gradually risen [7, 8].

On blood agar, the colonies appeared 3-4 mm in diameter, on MacConkey agar; the colonies were red, since this organism is a lactose fermenter [7, 8, 9].

*Bacillus subtilis* is a Gram-positive bacterium, rod-shaped and catalase-positive. *B. subtilis* cells are typically rod-shaped, and are about 4-10 micrometers ( $\mu\text{m}$ ) long and 0.25–1.0  $\mu\text{m}$  in diameter, with a cell volume of about 4.6 fL at stationary phase. As other members of the genus *Bacillus*, it can form an endospore, to survive extreme environmental conditions of temperature and desiccation. *B. subtilis* is a facultative anaerobe and had been considered as an obligate aerobe until 1998. *B. subtilis* is heavily flagellated, which gives it the ability to move quickly in liquids. *B. subtilis* has proven highly amenable to genetic manipulation, and has become widely adopted as a model organism for laboratory studies, especially of sporulation, which is a simplified example of cellular differentiation. In terms of popularity as a laboratory model organism, *B. subtilis* is often considered as the Gram- positive equivalent of *Escherichia coli*, an extensively studied Gram-negative bacterium [9, 10].

## Material and Methods

### Experiment setup

#### Plasma torch

Plasma needle designed with diameter 1mm from interior, this needle constitutes cylindrical tube made from glass material with length 100mm interior this glass tube, put other cylindrical tube made from iron material with external diameter 2.7 mm ,this tube connect to anode from high voltage power supply about 9.6kV peak to peak, applied power was lasting of electrical discharge which calculated from simultaneous values of voltage and current about 15 watt and applied frequency 33kHz ,it through pass argon gas where discharge between electrode and space through needle hole where plasma generation outside from hole, figure (1) shows plasma needle.



Figure (1): plasma needle

**Method:**

These samples were collected from water, sterilization tests were performed by drop from water sample and culturing bacteria before and after plasma needle treatment, while show growth of normal water bacteria (e.g *E.coli* bacteria ) on cultures from taken on control sample cultured a large quantity of bacteria obtained from were cultured on artificial media (MacConkey agar, Blood agar) to obtain bacteria colonies, carried out investigation of effects of plasma needle on bacterial cultures to quantify the extent of sterilization and determine possible factors responsible[11]. The prepared culture plates were treated by plasma needle and they are then incubated for (24h).

**Results and Discussion**

**1. Influence of argon gas flow rate on the Bacteria deactivation**

Optimum conditions was selected Voltage (1750 volt), frequency (15 kHz), distance (2.5cm), time (10, 20, 30, 40, 50) sec, and flow rates ( 1, 2, 3, 4, 5) l/min, by using plasma needle for killing bacteria that inhabit in water.

The initial study compared the individual susceptibility of two micro-organisms belonging to different species *E. coli* bacteria as shown in Figure (2), and *Bacillus subtilis* bacteria as shown in Figure (3).

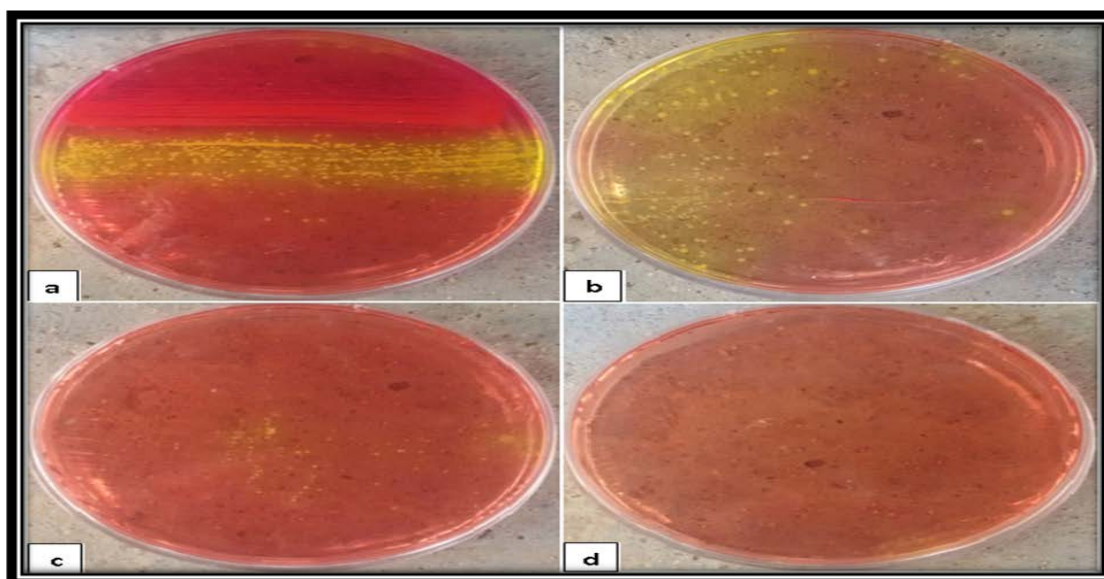


Fig.(2): *E.coli* bacteria , a=10sec, b=20sec, c=30sec, d=40sec

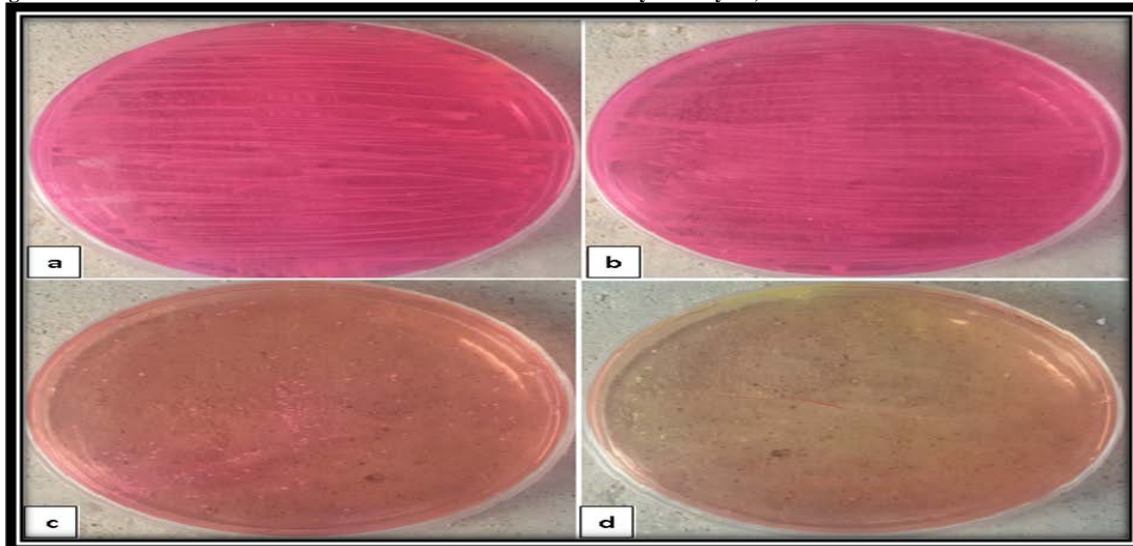


Fig.(3): Bacillus subtilis bacteria, a=10sec, b=20sec, c=30sec, d=40sec

Figures (4) & (5) show the relation between the survival percentage for bacteria and the relationship between the Reduction percentages as a function of flow rate respectively.

The effects of the increasing gas flow rate and high speed particle discharge penetrating through the outer structure of the bacteria may play a dominant role during the inactivation of the bacteria caused by plasma needle. If bacteria are treated with increase gas flow rate, the cell membrane’s structure and electric charges distribution over the cell membrane can be destroyed. In addition with the penetrating effect of the high speed particle discharge the outer structure of bacteria, namely cell wall and cell membrane of culture form, exosporium and coating of the spore, could be destroyed and cytoplasm would be released, which would cause the death of the bacteria. Because the outer structure of the spore was tighter than that of the vegetative form, the vegetative form could be broken by plasma and the spore could not be broken but only left with cuts by plasma needle [12]. *Bacillus subtilis* bacteria more sensitive to the plasma needle treatment than the *E.coli* bacteria; at the operating condition that applied in this work, the partial inactivation time for the *Bacillus subtilis* bacteria greater than *E. coli* at time (30sec) of same conditions [12]. While complete inactivation time for two bacteria at 40 sec of same conditions.

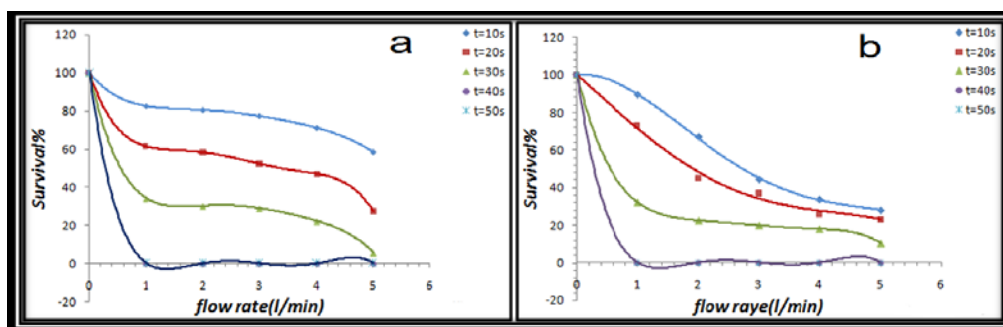


Fig.(4):The relationship between the Survival% as a function of flow rate (a: Bacillus subtilis , b: E.coli)

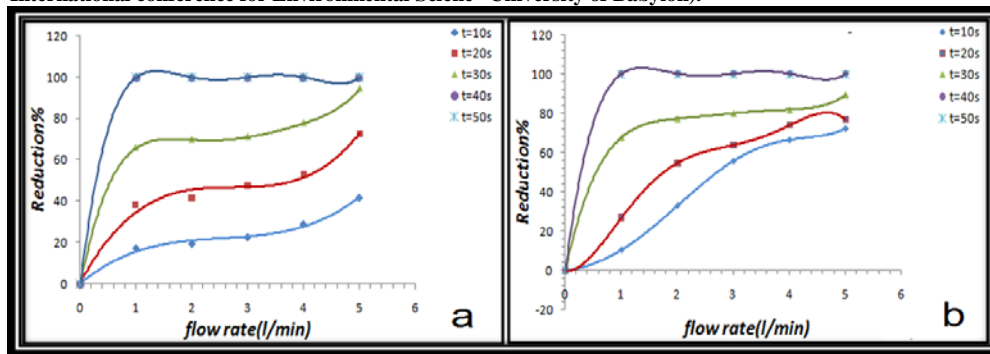


Fig.(5): The relationship between the Reduction% as a function of flow rate: (a: *Bacillus subtilis*, b: *E.coli* )

From results, one can show that; *E.coli*, *Bacillus subtilis* bacteria can be inactivated by exposed to the plasma needle for a period of time. The inactivation increases with treatment time increasing, the inactivation depends on the plasma needle system operating conditions such as applied voltage, gas flow rate and treatment time.

**2. Studying the influence of applied voltage on the Bacteria deactivation**

In this section, the effects of the plasma needle treatment on *E.coli* and *Bacillus subtilis* bacteria were studied according to conditions. The plasma needle was generated under different applied voltages and times. The bacteria were reduction in different percentages depending on the experiments conditions. The reduction percentage as a function of the plasma needle applied voltages for different conditions was presented in figures (6) & (7). The results, show that the reduction percentage increases with the increasing of treatment time for all applied voltages.

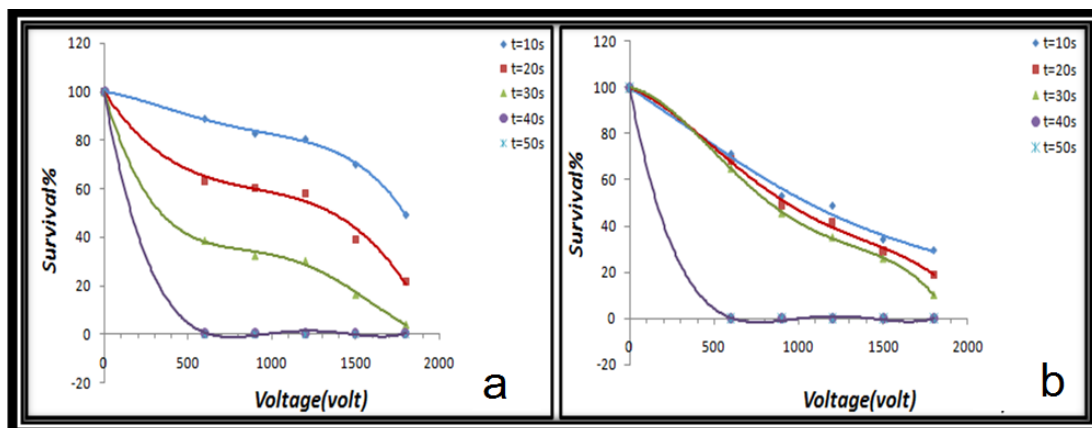


Fig.(6): The relationship between the Survival% as a function of Voltage: (a: *Bacillus subtilis*, b: *E.coli*)

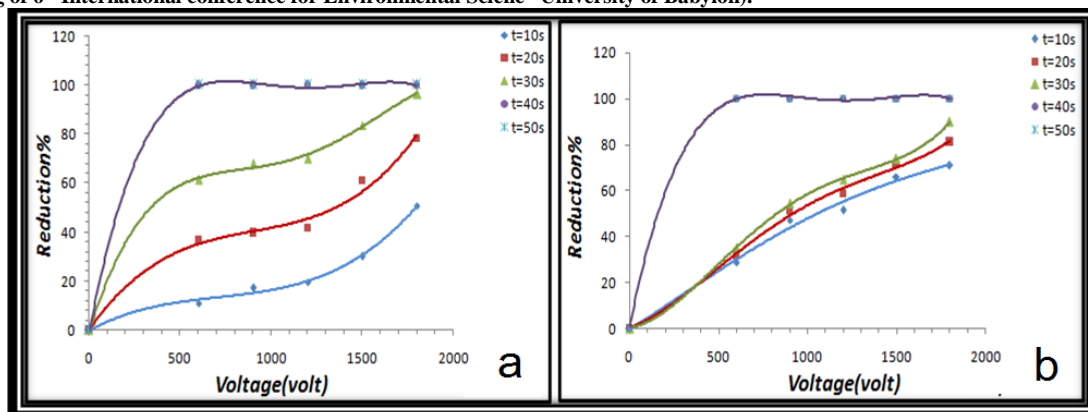


Fig.(7): The relationship between the Reduction% as a function of Voltage: (a: *Bacillus subtilis*, b: *E.coli*)

This was because the interaction between bacteria and reaction species was almost complete within 40 sec. This indicate to applied voltage height produced in high degree for gas ionization, hereby increases density various reaction species which was reactive agents to reduction bacteria cells [13].

The previous results showed that Gram negative bacteria are more resistant to plasma needle than Positive negative bacteria .This is related to the difference in the structure of the cell wall for both Gram negative bacteria and Gram positive bacteria .The cell wall of both Gram negative cells and Gram positive cells contains peptidoglycan ,which is responsible for protecting the cell .The peptidoglycan layer of the Gram negative cell wall is very thin and often comprises only 10% or less of the cell wall ,while the Gram positive cell wall has a peptidoglycan layer ,approximately comprising 90% of the cell wall ,thus providing these bacteria with higher strength and rigidity ,making then harder to be sterilize [9].

The specific mechanism for the plasma effect on epithelial cells is similarly unclear. Cold plasma produces long living ( $O_3, NO, HO_2, H_2O_2$ ) and short lived ( $OH, O$  electronically excited) neutral particles and charged particles (ions and electrons ). All of these could be toxic to cells ,induce low levels of cell membrane damage and potentially change intercellular signaling pathways .Specific plasmas can be created to produce either neutrals or charged particles in order to elucidate the critical mechanism. Charged particles can play a very significant role in the rupture of the outer membrane of bacterial cells. The electrostatic force caused by charge accumulation on the outer surface of the cell membrane could overcome the tensile strength of the membrane and cause its rupture [13, 14].

## Conclusions

The influence of argon gas flow rate on the bacteria deactivation, the effects of the increasing gas flow rate and high speed particle discharge penetrating through the outer structure of the bacteria may play a dominant role during the inactivation of the bacteria caused by plasma needle.

*Bacillus subtilis* bacteria more sensitive to the plasma needle treatment than the *E.coli* bacteria; at the operating condition that applied in this work, the partial inactivation time for the *Bacillus subtilis* bacteria greater than *E. coli* bacteria at time (30sec) of same conditions . While complete inactivation time for two bacteria at 40 sec of same conditions.

## Acknowledgment

I would like to express special words of thanks with deepest appreciation of college medicine of Anbar University and Clinical Laboratories in microbiology branch, also the Staff working in these Laboratories.



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