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Construction of Cold Atmospheric Pressure Plasma system for Using in Skin Fungal Treatment

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By

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بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

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الْأَمْثَالَ لِلنَّاسِ وَاللَّهُ بِكُلِّ شَيْءٍ عَلِيمٌ)

صدق الله العلي العظيم

سورة النور (35)

Dedication

To the messengers of science and scientists who dedicated their lives to the development and prosperity of mankind.

To the first teachers, my father, and mother, who devoted their lives to my success.

To those who taught me and encouraged me, my teachers and supervisors, who did not spare us effort and energy in order to spread the message of science and progress in this generous country.

To the one who sacrificed himself for the sake of our success, my wife and children.

To everyone who stood with us and encouraged us with pure hearts, my brothers and friends...

And to all those who complete the march of science.

Dedicate this thesis to them.

Ammar Salman

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Ammar Salman

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Abstract

Fungal diseases affect human safety greatly. Some types of fungi show resistance to antibiotics when treated, and others require a long period of treatment. One of the modern and safe techniques is the use of cold plasma in biomedicine. The plasma jet system was constructed and used to treat dermatophytes, which included three common types: *Trichophyton rubrum*, *Candida albicans*, and *Candida Tropical*.

Atmospheric pressure plasma jet systems (APPJs) are designed with simple, low-cost components. A high alternating voltage generator was used, and the system consisted of a double-loop electrodes configuration, a Pyrex tube as an insulating barrier, and pure argon gas to feed the system.

The electrical properties of the system were diagnosed and the effect of gas flow rate and applied voltage on the length of the plasma jet and the temperature of the plasma was studied. Use optical emission spectroscopy to calculate electron temperature and electron density. The electron temperature was (10826.6 K) and the electron density was ($1.57 \times 10^{13} \text{ cm}^{-3}$). The results showed that the system is suitable for use in biomedical applications.

A pure isolate of fungi was obtained on solid SDA agar. Fungal cells were treated with cold plasma at 25 °C. Treatment intervals were (2, 4, 6, 8, 10, 15, 20, 25) minutes. The results showed that the growth of the fungal cells decreases with the increase in the treatment period and thus leads to the killing of the fungal cell. In the treatment of *Trichophyton rubrum* fungi, the cell lost its ability to grow after 25 minutes of plasma

treatment, but the time required to kill cells in the case of *Candida albicans* and *Candida Troprum* was shorter (15, 20) minutes, respectively. Cold plasma technology is considered a promising and safe technique that has succeeded in eliminating the fungi under investigation.

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List of Symbols and Abbreviations

α_i	Degree of Ionization
λ	Wavelength
A	Transition Probability
I	Relative Intensity
J	Angular Momentum Quantum Number
g_u	Statistical Weight
C	constant
N_e	electron density
T_e	electron temperature
N_i	Number of ions
N_n	Number of density
SE	Single Electrode
DBD	Dielectric Barrier Discharge
DC	Direct Current
DFE	Dielectric Free Electrode
E_{ion}	Ionization Energy
E_u	Energy of the Upper Level
EM	Electron Microscopy
LTE	Local Thermodynamic Equilibrium
AC	Alternating Current
APPJ	Atmospheric Pressure Plasma Jet
SDA	Sabouraud dextrose agar
OES	optical emission spectroscopy
NIST	National Institute of Standards and Technology

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1.1 Introduction

Plasma is the fourth state of matter and is described as an ionized gas that contains free charge carriers (electrons and ions). These particles (electrons and ions) generate electric and magnetic fields inside and outside the plasma [1,2]. Plasma constitutes 99% of the universe. There are many examples of plasma in nature such as the sun, stars, the earth's ionosphere, lightning, and aurora borealis. They are all in the state of plasma(natural plasma) [3,4].

The difference between the four states of matter in the strength of bonding between the particles that make up each state is as in Figure (1.1). In a solid, the bonding strength between its components is large, and the bonding strength in liquid and gas is less and weaker [5].

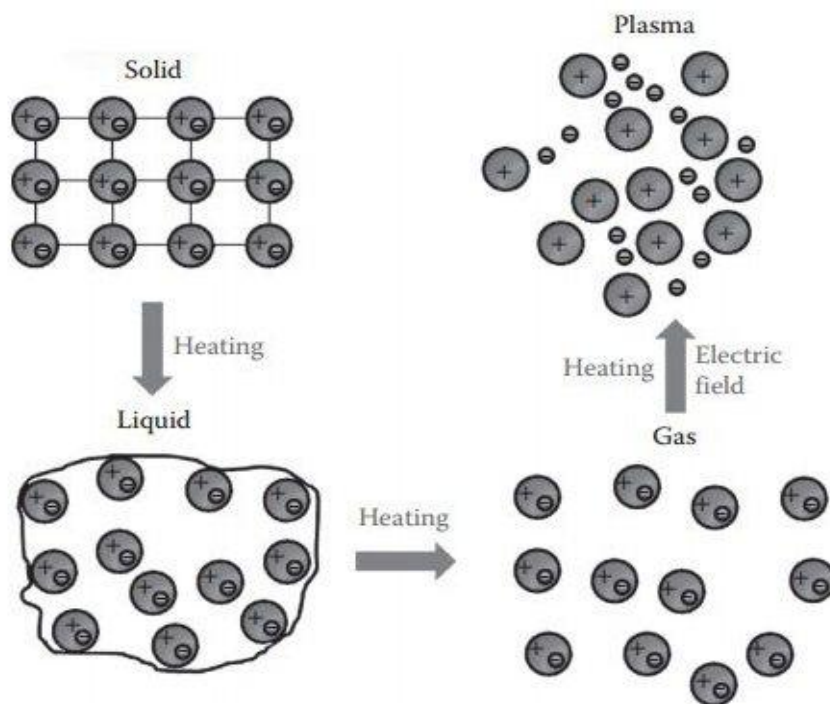


Figure (1.1): The phase change in the four states of matter with energy[6].

The solid state can be converted to the liquid state by adding energy to overcome the binding energy between molecules or atoms, which is the energy characteristic of the potential energy of fusion. Likewise, the liquid state turns gaseous by adding energy and defining the potential energy of evaporation[7]. As for the transformation into a plasma state, one way to transform is when enough energy is added to separate an electron atom from its nucleus, which is known as ionization energy[8]. The ionization is partly in the gas when the energy required for ionization is available, and this happens gradually. The transformation takes place from equivalent components in the substance to charged components [9].

It is possible to produce plasma without a wide range of pressure and temperatures. The plasma is produced when using low pressure or having standard atmospheric pressure. The use of various energy sources such as mechanical, thermal, chemical, nuclear, and radiological energy, or by applying a voltage difference, or by using laser, or by employing electromagnetic waves and these methods are used to overcome the binding energy between atoms or molecules in the gaseous medium and thus separate into a group of ions and electrons, so the plasma is a chemically active environment that combines particles and photons [10,11].

Where the plasma consists of many types of molecular, atomic and ionic, and radical species, and many reactive species types coexist in it, such as electrons, positive and negative ions, free radicals, gas atoms, and various molecules in static or excited states, as well as the quantities of electromagnetic radiation (visible light and ultraviolet photons)[12]. Free electric charges (electrons and ions) make the plasma conductive of

electricity and interact internally and respond to electromagnetic fields [13].

1.2 Plasma Ionization

Ionization energy is the energy required to convert one atom into an ion, and it is usually equal to the energy of the last electron bonding to the atom. It could be caused by heating, light, or radiation. The ratio of the ionization depends on the kinetic energy of the gas particles, i.e. it depends on the temperature of the gas, and ranges between 0 and 1[14]. The Saha equation is used to calculate the degree of ionization. Ionized gas has three types of components, which are neutral (atoms, molecules, and radicals), positive ions, and electrons. The degree of ionization is calculated according to the equation[15].

$$\alpha_i = \frac{N_i}{N_i + N_n} \quad (1.1)$$

where N_i : the density of ions , N_n : the density of neutrals .

The response of the plasma to the magnetic and electric fields is determined by the degree of ionization. When the degree of ionization is complete, close to 1 represented the hot plasma [15].

1.3 Classification of thermal plasma

Plasma is classified according to the relative temperatures between electrons, ions and neutrals, and can be divided into two main groups:

- 1- high temperature plasma (thermally equilibrium plasma)
- 2- Low temperature plasma (thermally non-equilibrium plasma) [16].

Figure (1. 2) shows the classification of thermal plasma.

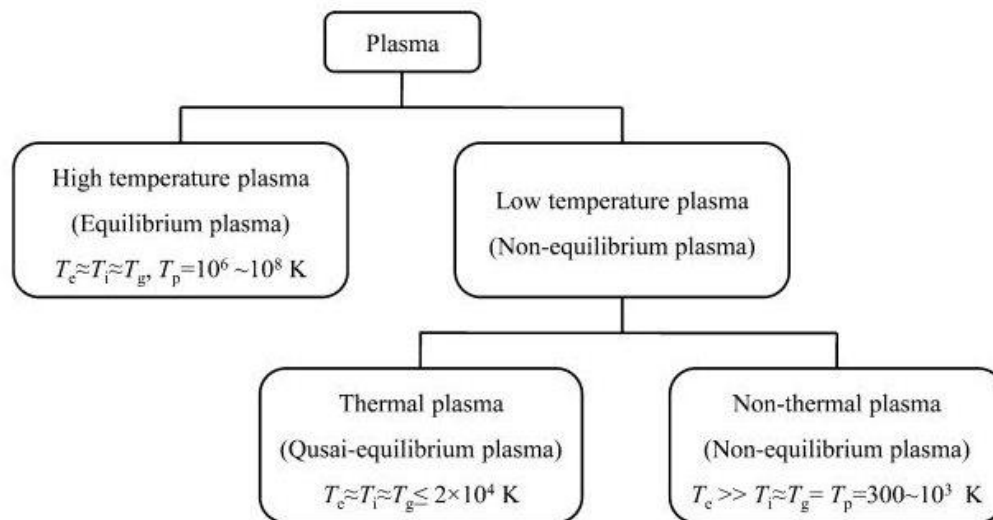


Figure (1.2) : Classification of thermal plasma [17].

1.3.1 high temperature plasma

It is a type of plasma whose temperature of the electron T_e is equal to the temperature of the ion T_i and the neutral atom T_n , they are almost identical, ($T_e \approx T_i \approx T_n = 10^6 - 10^8 \text{ K}$) [18]. This is due to the frequent and very large collisions between electrons, ions, and neutral within the plasma, which are very dense and high in temperature and have a high

degree of ionization close to 1, This type of plasma can be found in the sun, stars, the core of the earth and the solar wind[19].

1.3.2 Low temperature plasma

It is characterized by the fact that its temperature is much lower than the high temperature plasma, also the degree of its ionization and density is lower . It can be divided into two types:

- 1- thermal plasma (quasi-equilibrium plasma)
- 2- non-thermal plasma (non-equilibrium plasma).[20]

1.3.2.1 thermal plasma (Quasi-equilibrium plasma)

This type of plasma that is intermediate between the cold plasma and the hot plasma, and in which there is relative equilibrium between the temperature of the electron T_e and the temperature of the ion T_i and the neutral T_n . ($T_e \approx T_i \approx T_n \leq 2 \times 10^4$ k). This type of plasma is used in coatings, as well as in chemical and physical vapor deposition [21].

1.3.2.2 non-thermal plasma (cold plasma)

It is a plasma in which the temperature of the electron is much higher than the temperature of the ions T_i and the neutral T_n , ($T_e \gg T_i = T_n = 300 - 10^3$ k)[19]. The momentum transfer between the electrons and the heavy particles (ions and neutral) be very small and ineffective, as well as the degree of ionization is less than 1% [22,23].

For example, cold plasmas is used in environmental engineering, aviation engineering, biomedicine, analytical chemistry, and biomedical applications[24]. The cold plasma contains reactive types, charged particles, ultraviolet photons, visible light, and electric fields[25], as shown in Figure (1.3), and they have influential biological effects, so they are used in sterilization and treatment and have shown great effectiveness as an antimicrobial [26].

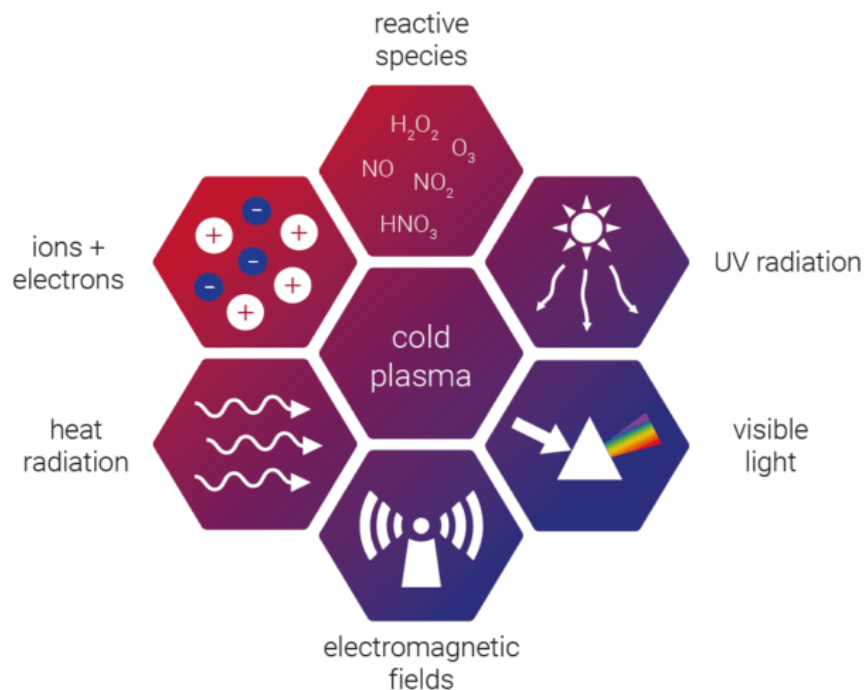


Figure (1.3) Cold plasma components[27].

The proportion of ionization in cold plasma is weak about 1%, and the density of free electrons is not as high as it is in thermal plasma ($n_e=10^{10}m^{-3}$). In cold ionized plasma, the neutral-charge interactions override multiple Coulomb interactions [27,28]. Although the collisions of electrons with electrons achieve thermodynamic equilibrium in a cold plasma, free electrons are distinguished by their high kinetic energy, as

their temperature is much higher than that of ions and neutral, but they cannot transfer their high kinetic energy to larger particles[29].

1.4 Plasma diagnostics

In Cold plasma, it is necessary to measure the electron temperature T_e and the electron density N_e . It is one of the most important parameters of the plasma. The properties of the plasma can be improved by improving the basic parameters of the plasma. These parameters are necessary to simulate the radical density as well as the speed of the plasma's interaction with the external turbulence(whether it is magnetic or electric) and Plasma response to externally applied fields[30].

The charged particles in the plasma are composed of free electrons and ions, but the total density remains in equilibrium, as the density of the number of electrons is equal to the density of the number of ions, so it is said that the plasma is a quasi-neutral gas[31,32].

the temperature of the electron is much greater than the temperature of the ion and the neutral in the cold plasma. When the temperature of the ion and the neutral is close to room temperature, the temperature of the electrons in the cold plasma is thousands of Kelvin[33]. The electrons have large kinetic energy depending on their temperature, which is a distinctive feature of the plasma. The temperature of the electron in relation to the ion energy determines the degree of ionization of the plasma. When the temperature of the electrons is lowered, the ions and electrons tend to recombine and form the neutral, thus the plasma eventually becomes a gas[34].

1.5 Plasma Production

Previously, plasma was produced in the laboratory by heating, where the gas is heated at a low pressure less than the normal atmospheric pressure until the kinetic energy of the molecules of this gas becomes sufficient to cause the ionization process and the separation of electrons and the formation of the pair (electron - ion) occurs through the non-collision process Flex between them, and the fourth state of matter is achieved[35].

Plasma is produced in several ways, and one of the most common methods is the use of electricity by electric discharge of neutral gases. This is done by passing a neutral gas between two electrodes, and then applying a high voltage difference between the electrodes under a certain pressure, where an electric discharge occurs between the two electrodes (anode and cathode) [35].

1.6 Gas discharge

Cold plasma is produced by an electrical discharge associated with the passage of gas through an electric field. When an electric current flows through a gas, you will get an ionization state in the gas molecules and its atoms. Where a sufficient potential difference will be applied between two electrodes immersed in the gas, which leads to the formation of ions and electrons after the feed gas collapses between the electrodes and leads to increased ionization processes [36]. The ionization process near the negative electrode, which accelerates electrons accelerated by an electric field. Which causes a collision with the gas atoms and this

collision will contribute to the process of excitation or ionization. This ionization of the gases is sufficient to emit energy in the form of light. The glowing discharge is a luminous plasma [37]. The glowing gas can be called the same name as the cold plasma. Glow discharge was used by the scientist Michael Faraday while studying electric discharge in gases. Gas electrodes are classified according to the shape of the electrodes, the distance between them, the source of energy, and the pressure of the gas. gas type. This phenomenon occurs in nature through lightning [38].

There are two types of visible electrical discharge:

- 1- Glowing discharge.
- 2- ARC discharge [38] .

1.7 Glowing Discharge

Glowing discharge is a type of luminous plasma because the electron's energy is large enough to produce visible light through repeated collisions. Partially ionized gas consists of several positive and negative charges in approximately equal proportions and a large number of neutral species[39]. The glow discharge is formed in a cell containing inert or molecular gases, by applying a potential difference to the electrodes, which are usually flat with a space between them inside the closed tube, and as shown in Figure (1.4) the potential difference is usually low, and so is the current in this process [40].

Gas atoms are ionized by electrical discharge and electrons as well as positive ions are formed which is known as (gas breakdown). The positive ions are accelerated towards the negative electrode and a glow discharge occurs. Ions and electrons are accelerated by the applied field and get additional kinetic energy which enables them to excited atoms,

and some other atoms are excited by collision. The characteristic glare discharge light is generated as a result of these collisions[40]. Ionization collisions create new ions and electrons and lead to the formation of new pairs. The glow-discharge process is used in a number of industrial applications such as fluorescent lamps, neon discharge tubes for advertising, flat plasma display panels, laser gas generation, the microelectronics industry, and material processing technology [41].

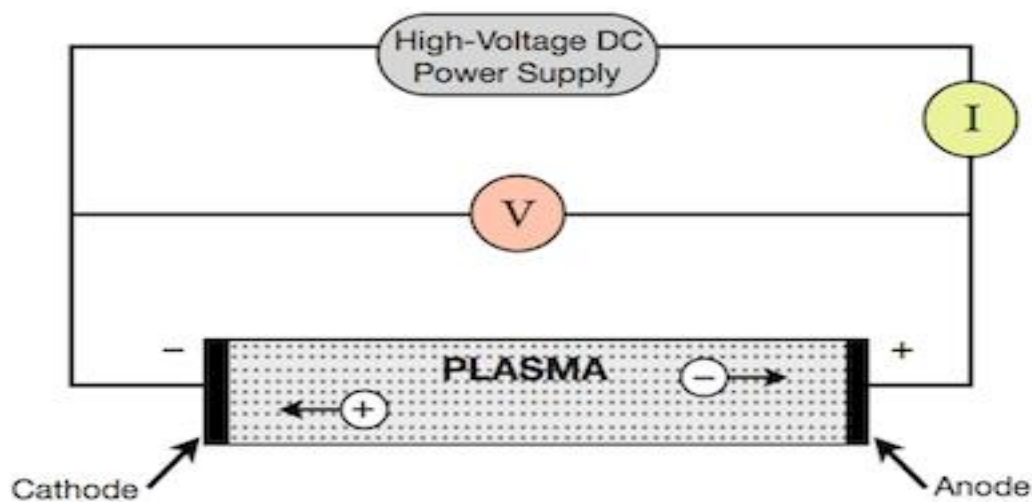


Figure (1.4): Glow discharge in a low pressure [41]

1.8 Production non-thermal plasma at atmospheric pressure

Low-pressure glow-discharge plasmas are of great use in much basic scientific research as well as in materials technologies and the microelectronics industry. Recently, the use of this type of system has decreased. The system needs to provide a vacuum tight system, which makes it very expensive and the active particles in it are relatively few. Therefore, the new direction focused on the development of plasma sources at normal atmospheric pressure. These systems are easy to manufacture, low cost, and rich in active molecules that operate under

normal atmospheric pressure. These features make it one direction for its development, and several types have been used for this purpose [42].

It will discuss the most important gas discharges using normal atmospheric pressure, its components, power supplies, electrode shape, and their applications.

1.8.1 ARC Discharge

When the electrodes acquire high energy of the current, they become hot enough and emit thermal electrons. An increase in the temperature of the electrode material occurs due to the breakdown of the gas's electrical insulator, causing a continuous discharge that leads to the flow of an electric current in a non-conductive medium such as air, causing a spark known as the electric arc[43]. This leads to a decrease in the breakdown voltage as well as the generation of a thermal field resulting from the electrons emitted from the cathode while it is hot and characterized by a high current greater than the glow discharge current and is practically used in welding and plasma cutting operations[44], as shown in the figure (1.5).

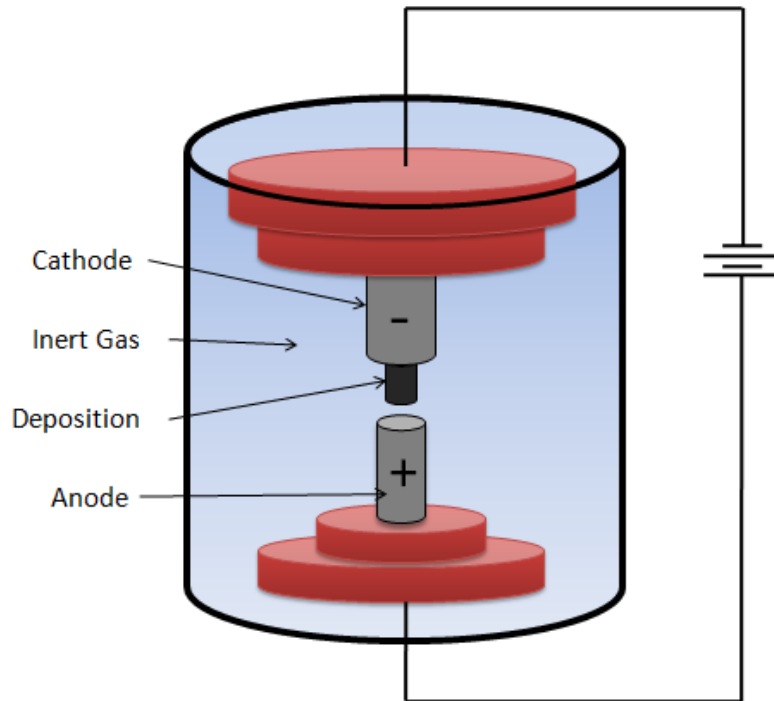


Figure (1.5) : Schematic of ARC Discharge [45].

1.8.2 Corona Discharge

It is the first design used to produce non-thermal plasma at normal atmospheric pressure. It occurs due to the collapse of the gas gap with a strong, sharply irregular electric field, meaning that it appears when using electrodes with pointed ends[46]. Distributions of irregular electric fields are formed when the size of at least one of the electrodes is much less than the distance between the electrodes and is fed by alternating current or direct current, and that the high electric field that is near the electrode and with a pointed tip is sufficient to cause a discharge in gases. The corona discharge is classified by the polarity of the electrode, the corona voltage is positive and negative.

The corona is the phenomenon of irregular discharge that does not occur in the high field region and is close to the cathode (the sharp end) towards the anode (the flat end) as shown in Figure (1.6). Which is used to collect impurities found in many industrial bad gases, water purification, ozone generators, and deposition processes[47].

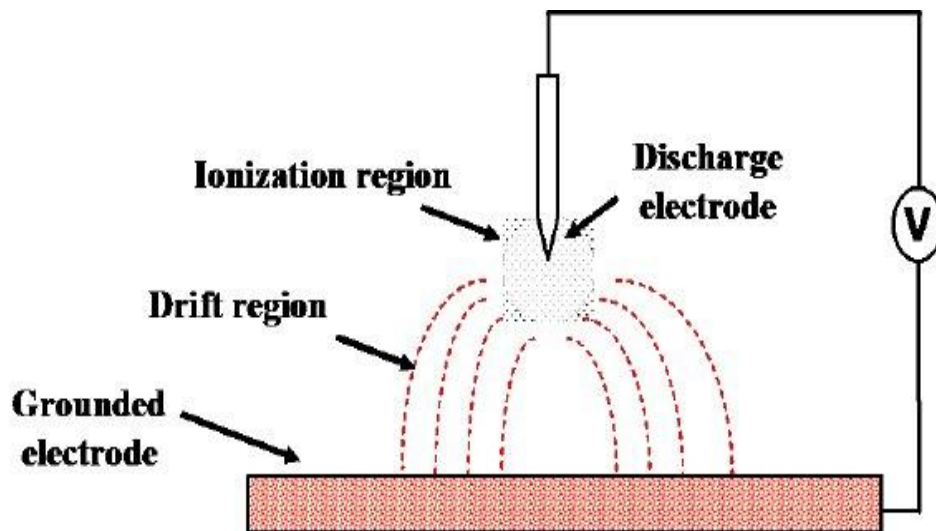
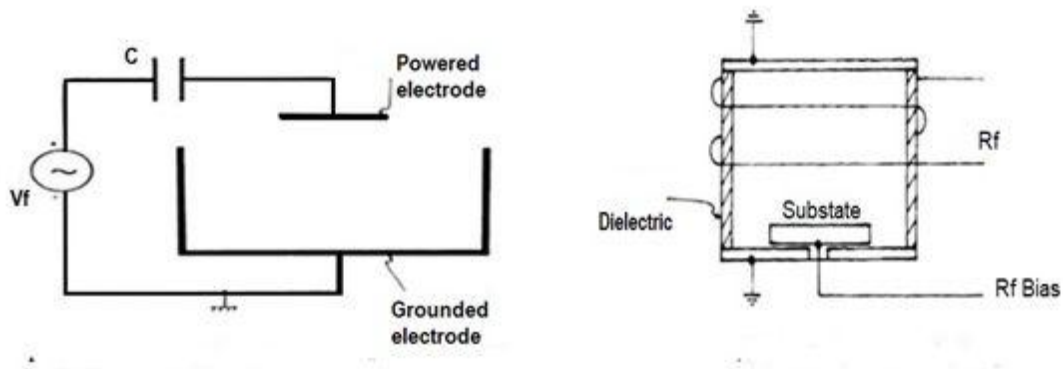


Figure (1.6) Schematic of corona discharge[47]

1.8.3 Radio Frequency Discharge

RF discharge operates at high frequencies in the range of one to 100 MHz. Is low pressure less than 10^5 Pascal used for this purpose and sometimes normal atmospheric pressure is used in some applications and this type of discharge is obtained by passing a gas in an oscillating electromagnetic field and the advantage of this type The electrodes can be kept outside the discharge tube. This feature prevents the electrode from eroding and contaminating the plasma with metallic vapor. Plasma is generated either by induction or capacitance[48]. An alternating current voltage source is provided to supply power to the electrodes through a capacitor, while the other electrode is grounded. The capacitor is charged

at a fast ionic current, which causes the voltage to drop above the plasma and drop guides the plasma in the negative half cycle, as shown in figure (1.7). This type is widely used in semiconductor manufacturing, aerospace and microelectronics [49].



(a)Capacitive coupled discharge system. (b)Inductive coupled discharge system.

Figure (1.7) Radio Frequency Discharge[49]

1.8.4 Microwave Discharge

It is an electrical discharge of gas produced by a source of electromagnetic waves at high frequencies (300 MHz to 3 GHz) without using electrodes as shown in figure (1.8). This type of discharge can be generated in a wide range of atmospheric pressure (1 mPa up to normal atmospheric pressure)[50]. to a high temperature of 105 degrees Celsius, which is a suitable method for generating high-density non-thermal plasma where the ionized gas fraction is sufficiently higher than that of other types. Microwave discharge plasmas have been used in laser generation, light sources, plasma chemistry, and analytical chemistry [51].

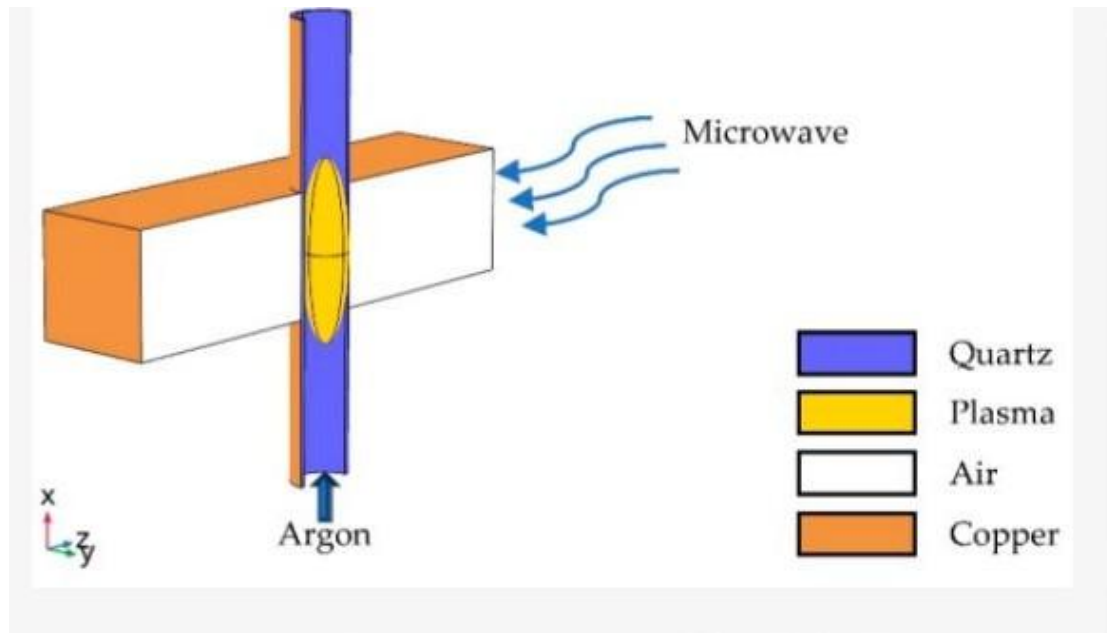


Figure (1.8) Cutaway view of the microwave Discharge[52].

1.8.5 Dielectric Barrier Discharges

Dielectric Barrier Discharge (DBD), also called (silent discharge). It operates at frequencies ranging from (50 Hz to MHz radiofrequency). It consists of two metal electrodes and the gap between the electrodes ranges from a micrometer to a few centimeters[53]. One or both electrodes are covered with an insulating material such as quartz, pyrex, or ceramic or placed between them to limit the discharge current ensuring cold plasma and avoiding a spark discharge (arc transmission). The gas passes between the two electrodes, causing the gas atoms to ionize and resulting in plasma [54].

The DBD is generated in a wide range of atmospheric pressures up to 500 kPa, and to generate it often uses an alternating current voltage source. One of the most important differences between the classical discharge and this discharge is that in the classical discharge the

electrodes are in direct contact with the discharge gas, so during the discharge process, the electrode is subjected to abrasion or corrosion. Unlike in the barrier discharge, where the electrode, gas, and discharge are isolated from each other by a dielectric barrier which prevents corrosion of the electrodes. Another fundamental difference is that the diaphragm discharge does not operate at a DC voltage, due to the dielectric capacitive coupling, which requires an alternating voltage to operate the displacement current. Figure (1.9) shows the arrangement of septum discharge. Figure (1.9 a) shows that one of the electrodes is covered with an insulating layer and (1.9 b) also shows that one of the electrodes is covered with an insulating layer. Figure (1.9 c) shows that both electrodes are separated by an insulating layer.

This type is used in a wide range of industrial and laboratory applications such as surface modification of polymers, plasma medicine, flat plasma display panels, and chemical vapor plasma deposition [56].

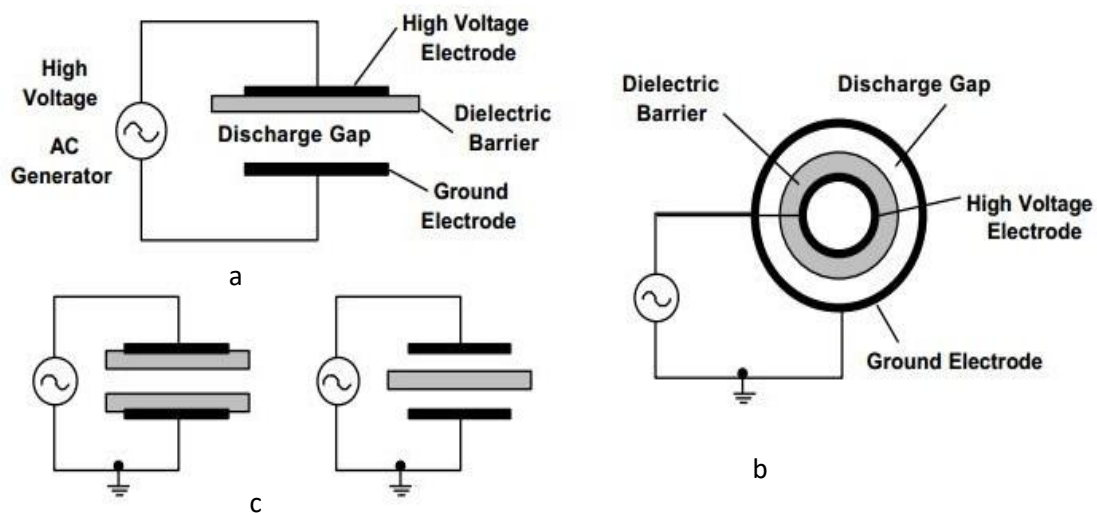


Figure (1.9) : Common DBD electrode configurations[56].

1.9 Literature Reviews

Fungal diseases affecting human safety have increased significantly. There are some types of fungi and bacteria that show resistance to antibiotics. Therefore, there is a need to use modern, more effective, and safe technologies for its elimination. Cold plasma is one of the modern and safe techniques, which is suitable for sterilization and inhibition of bacteria and fungi [57]. It will discuss some previous studies that dealt with plasma treatment and the treatment of fungi.

Daniel Dobrynin et al. (2011) used cold plasma sterilization for wounds of live mice where high concentrations of *Staphylococcus aureus* were placed at the wound site and then the bacteria were allowed to incubate in the wound area for four hours, then cold plasma treatment was used for different periods of time. The results showed a decrease of bacteria at the wound site after the first minute of treatment [58].

G. Ispary et al. (2012) Use cold argon plasma at atmospheric air pressure on chronic wounds. Treatment was determined for 2 minutes and compared with wounds without treatment. The study indicated that cold plasma is a safe, painless, and highly effective technique for reducing the bacterial load in treating wounds with high efficiency [59].

Y. F. Lee et al. (2013) A portable cold plasma device was designed for use in live skin treatment. The direct method of treatment and the indirect method were compared, and cold plasma was used at atmospheric pressure to treat the physiologically contaminated forearm in 12 healthy volunteers. The study concluded that after 30 seconds of cold plasma treatment, bacterial growth in the wound area was reduced [60].

X. Zhang et al. (2014) In this study, the leaves of plants infected with fungi were treated and the spread of infection was controlled by using cold plasma technology to treat plant diseases. It was concluded that the use of cold plasma in the treatment of fungi leads to weakening the functions of fungal cells and reducing fungal growth [61].

S. A. Ouf, et al (2015), five fungi isolates were taken and treated with cold plasma at atmospheric pressure, and electron microscopy showed electrical perforation in cell walls and inside cells, and the infection was cured by 91.7% in the case of guinea pigs, after cold plasma treatment for two minutes. The roots generate NO and excited nitrogen molecules as these reactive species interact with the fungal cell and inhibit its activity inside the laboratory and in the infected guinea pig skin. The results were promising and good when compared to the traditional anti-fungal drug [62].

L. Kordas, et al (2015), fungi were studied for colonization of winter wheat grains, and the potential effect of cold plasma on fungi was determined. 200 samples were taken and divided in half, a section was treated with cold plasma and another section was traditionally treated with 0.5% of sodium hypochlorite. The results showed that the grains treated with cold plasma For a period of 10 seconds, reduced the number of fungal colonies formed on the grains, and the results were good with regard to the basic values related to seed quality and on the initial growth of winter wheat treated by cold plasma [63].

B. G. Dasan, et al. (2016) In this study, cold plasma technology was used to treat fungi contaminated with corn kernels. The plasma treatment intervals were 1 to 5 minutes. The results showed a decrease in the rate of fungal contamination after 5 minutes. She indicated changes in

the surface of the fungal spores and the loss of their integrity, which led to their death. The study indicated that cold plasma has a good effect in improving the safety and quality of processed wheat grains [64].

P. Thonglor, *et al.* (2017), cold plasma jet treatment was performed on bread molds contaminated with types of fungi, and the results showed that the appropriate treatment time is 20 minutes in treating fungi, and the most effective in inhibiting the growth of fungal cell and fungi. The cytoskeleton implanted in the bread mold was destroyed, and the study showed the possibility of using plasma in the optimal time and energy condition, in addition to the possibility of applying this technology to bread making in the future [65].

Y. Devi, *et al.* (2017) in this study, the effect of cold plasma on the growth of fungi was studied, where they were free of toxic substances, and the treatment was carried out on fungi artificially dumped with peanut material at energy levels in different time periods. The treated fungus was inhibited and the rate of growth of fungi was observed after treatment. The cold plasma was considered a successful alternative to disinfecting foods, including peanuts, due to its strong ability to inhibit the activity of microbes. The treatment effectiveness depends on the type of gas and the type of plasma device used. The effectiveness of the treatment was observed on the rupture of fungi starch after treatment[66].

M. Medeiros, *et al.* (2018) experiment was conducted using fungal samples that were treated with cold plasma and for different periods and for a period of six days. The samples were stored after treatment at 25 degrees Celsius for six days and there was no fungal growth in the treatments for 15 to 20 minutes after six days while there was fungal growth. Fewer treatment times of 10 and 12 minutes

compared to lesser treatment periods. It was found that the plasma treatments used were effective in inactivating the fungi under study. Cold plasma may be a promising green method that will be applied in the future to microorganisms present in grains and other products when stored [67].

S. Hosseini, et al. (2018) in this paper, the cold oxygen plasma was used to disrupt the spread of fungi on the saffron plant, and different types of reactive plasma were produced and were examined by means of optical emission spectroscopy. The best exposure time in treating the fungi was evaluated at 15 minutes and the cold plasma had no adverse effect on the treated saffron nor on the color, aroma, and flavor of saffron [68].

Y. Veremii, et al. (2019) used cold plasma to disinfect the surfaces of pine seeds from fungi spores with different exposure times, and then germination was done in pale dishes and at room temperature. Estimation of seed plants and the effect of cold plasma treatment and compared with their counterparts. The results obtained through treatment are that cold plasma is a promising technique. In improving germination and in sterilizing and disinfecting seeds according to the data, cold plasma will be a disinfectant for the surface of different seeds with great effectiveness[69].

M. He, et al. (2019), in this study the efficacy of cold plasma in the treatment of fungi and the effect of inhibition of plasma therapy were tested in vitro and in vivo, where cold plasma increased the possibility of cure, in vivo achieved the best antifungal effect after four minutes treatment. The increase in the treatment periods will cause biological tissue damage and give the opposite of the desired result, as increasing

the treatment periods leads to an exacerbation of the infection compared to a total of four minutes [70].

N. Misra, et al. (2019) the research concluded that cold plasma is a promising technique and a successful alternative to reduce fungi on foodstuffs, including grains, spices, fruits, vegetables, meat, etc. It is an alternative to traditional methods of ultraviolet light, gamma irradiation, and pulsed light at a relatively lower cost. Studies indicate that cold plasma is a fast, effective, and economically feasible technique for eliminating the effect of mycotoxins, compared to its counterparts of temperature, chemotherapy, and ultraviolet rays[71].

L. Ott, et al. (2020) study demonstrates the use of cold plasma, atmospheric pressure, using a high voltage applied to the air, to generate reactive oxygen, and types of nitrogen to sterilize types of fungi. Conducting an electron microscopy scan and the results showed that I can aromatize fungal germs and toxins in minutes by using cold plasma, using air and the fungal toxins have become non-toxic after a 20-minute treatment period, thus proving the importance of using cold plasma in sterilizing materials from fungi and germs[72].

A. Alizadeh, et al. (2020), new preventive methods were investigated against fungi that intend on different food products and can destroy large quantities of food and spoil it. The new green techniques were reviewed with different types of ionizing radiation and non-specific, cold plasma, pulsed light, ultrasound, and pulsed electric field. The high pressure waves and the study concluded that these technologies have significantly reduced the types of fungi, their destruction, the decomposition of fungi toxins and their elimination, and the destruction of the structure was extremely promising as well as for the fungi toxins,

as well as the techniques provided health and safety features for food products and for the consumer, these technologies have a great potential in preventing the growth of fungi causing diseases and spoilage of fungal toxins[73].

A. Ghorashi, et al. (2020) the aim of the research is to obtain a cold plasma device with the best performance to disrupt the fungi from the surface of pistachios, and three types of devices are used to treat the fungi on the surfaces of pistachios. The study succeeded in finding the best performance to reduce fungi in practice using argon as the main gas after a treatment period of 20 minutes The fungi were reduced on the surface of the pistachio nuts, and the color, smell, and texture were changed in general, which appears to be a promising and safe technique for getting rid of fungi for nuts [74].

1.10 aim of the study

This study aims to:

1. Construct a non-thermal plasma jet system Dielectric Barrier Discharge(DBD) type.
2. Study of the effect of applied voltage and flow rate of argon gas on the length of the plasma jet and the temperature of the plasma jet.
3. Analysis of spectral emission of the plasma jet and determine the reactive species.
4. Distinguish the optimal use of the system by finding the basic plasma parameters(electron temperature and electron density).
5. Assess the efficiency of the plasma jet device inactivation of fungi and confirm the inhibition process after treating fungi and determining the optimal duration of plasma treatment.

2.1 Introduction

This chapter focuses on the properties of the electrical discharge and its dependence on the shape of the electrodes. The power source and gas pressure are discussed. The components of Atmospheric Pressure Plasma Jets (APPJs) are also discussed in this study. As well as the interaction of reactive species in cold plasma in biomedical applications and the method of calculating electron temperature and density by optical emission spectroscopy.

2.2 Atmospheric Pressure Plasma Jets

Atmospheric Pressure Plasma Jets (APPJs) are a promising source of plasma, providing cold plasma close to room temperature, controllable and reproducible under normal conditions and not limited to electrodes as in normal electricity, as well as being able to generate Different types of active species. It consists of an insulating tube made of pyrex, quartz, or ceramic tube, with an electrode around the tube or inside the tube. One of the noble gases (argon, helium, or nitrogen) is introduced into the insulating tube and sometimes mixed with a very small percentage of oxygen to increase the reactive species[75]. A sinusoidal or pulsed voltage (for some kilovolts) is applied at a variable frequency (several hundreds of Hz to gigahertz). This process does not require the use of a vacuum chamber, as it operates at normal atmospheric pressure, allowing the generation of plasma at a low cost. Today, plasma jets are increasingly used in material processing and biomedical applications such as surface modification, etching, and nano-deposition, as well as medical

applications in sterilization, decontamination, wound healing, and cancer cell killing [76].

Delivering cold plasma through a flexible tube can be very useful in endoscopic applications in medicine such as treating colorectal, dental, and pancreatic cancer[65]. Therefore, the development of suitable plasma sources for in vivo therapies has been the subject of extensive research and this type of plasma has been of interest because it provides an open-air conduction medium, reactive plasma species, as radicals, positive and negative ions, and ultraviolet radiation at atmospheric pressure and normal temperature of a target located a few centimeters from the end of the tube in which ionization occurs. [77].

2.3 Types of Atmospheric Pressure Plasma Jets Configurations

Non-thermal Atmospheric Pressure Plasma Jets are classified into four types: Dielectric Free Electrode (DFE) Jets, Single Electrode (SE) Jets, Dielectric Barrier Discharge (DBD) Jets, Dielectric Barrier Discharge-Like (DBD Like) Jets [77].

2.3.1 Dielectric- Free Electrode Jets

Dielectric Free Dielectric (DFE) jets are one of the oldest types of APPJs. It is powered by an RF power source. It consists of an internal electrode connected to the power supply and a grounded external electrode. One of the noble gases and one of the reactive gases such as Oxygen are introduced into the annular space between the poles[78]. To

prevent overheating, cooling is done by water. Depending on the RF power and its power (between 50 and 500 watts), the temperature of the plasma jet can range from 50 to 300 degrees Celsius. As shown in Figure (2.1). This type of plasma jet is not suitable for medical and biological applications due to its relatively high temperature, but it can be used in materials processing as long as these materials are not sensitive to high temperatures [79].

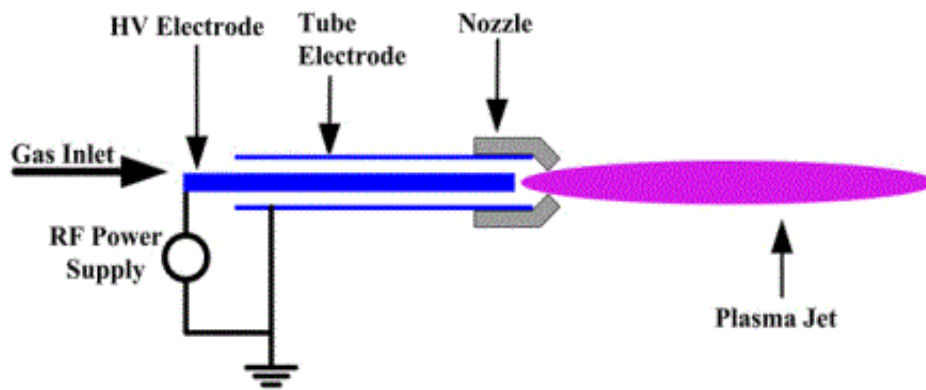


Figure (2.1) Schematic of a dielectric - free electrode (DFE) jet [78]

2.3.2 Single Electrode Jets

Single electrode (SE) jets are powered by an AC, AC, or RF generator, as shown in Fig. (2.2) A and B. It consists of an insulating tube and electrode connected to the power supply and SE jets fed to one or two types of noble gases[78]. With sometimes reactive gases, which are not suitable for medical applications due to the danger of arcing, SE aircraft has developed a plasma jet consisting of a single hollow electrode connected to the power source as shown in Figure (2.2) C where a resistor and a capacitor connected to a hollow electrode are used to control In the amount of voltage and current discharge this type of jet can be used in

biomedical applications where the plasma jet can be touched without any damage, so SE jets are used, especially in dentistry [79].

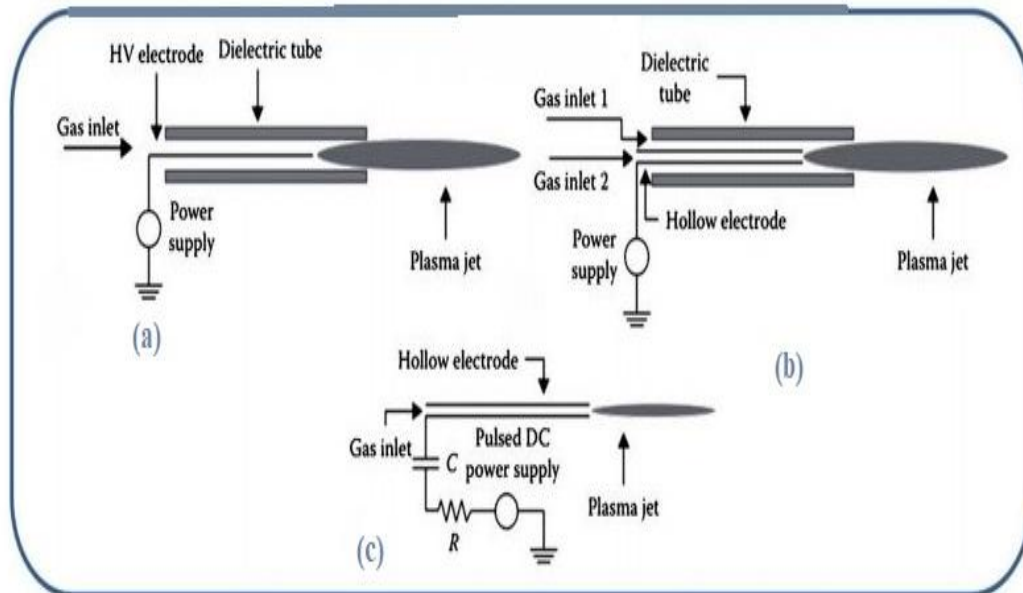


Figure (2.2) : Schematic representation of a Single Electrode jet [80].

2.3.3 Dielectric Barrier Discharge-Like Jets

Dielectric Barrier Discharge-like (DBD-like) jets, as shown in Fig. (2.3). In which the discharge occurs between the high voltage electrode and the object to be treated, usually using two types of noble gas with different mixing ratios and one of the types of reactive gases can be used for this purpose, and it works using different sources such as an AC, DC, or RF, and if it is from an insulating tube and an electrode annular[78]. This type of plasma jet is used in plasma medicine applications when the organism to be treated is large-scale cells and

tissues. These devices should be used with caution due to the risk of bending [80].

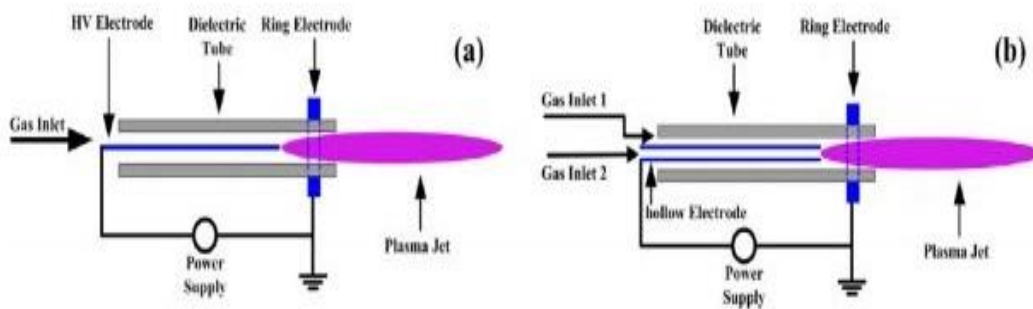


Figure 2.3: Schematic representation of an DBD-like jet [78].

2.3.4 Dielectric Barrier Discharge Jets

The generation of a Dielectric Barrier Discharge (DBD) jet is carried out by means of a high voltage alternating current. Whereas, DBD jets, as shown in Fig. (2.4), can be generated by a high-voltage AC power source or by a pulsed DC power source. As shown in Fig. (2.4) (a), the DBD jet consists of an insulating tube around which there are double metal ring electrodes at the outer side of the tube and these electrodes are connected to power. The temperature of the plasma gas is close to room temperature, so it was used in this study. Noble gases are used to feed the insulating tube in Figure (2.4) (b), the flow consists of one annular electrode and the other electrode is the body. In Fig. (2.4) (c), the flux consists of two dielectric tubes (inner and outer) and two electrodes (ring and pin). Its design enhances electric field along with plasma jetting. The high electric field along the plasma jet is suitable for generating long plasma jets and active chemical elements. Figure (2.4) (d), the plasma

flow consists of two insulating tubes with an electrode, so the discharge inside the tube is also attenuated [78]. The advantages of DBD jets are that the temperature of the cold plasma jet remains close to room temperature because the energy density is low in reactive species. There are also no bending hazards for plasma-treated specimens. The two advantages are very important for many applications especially in biomedical applications [81].

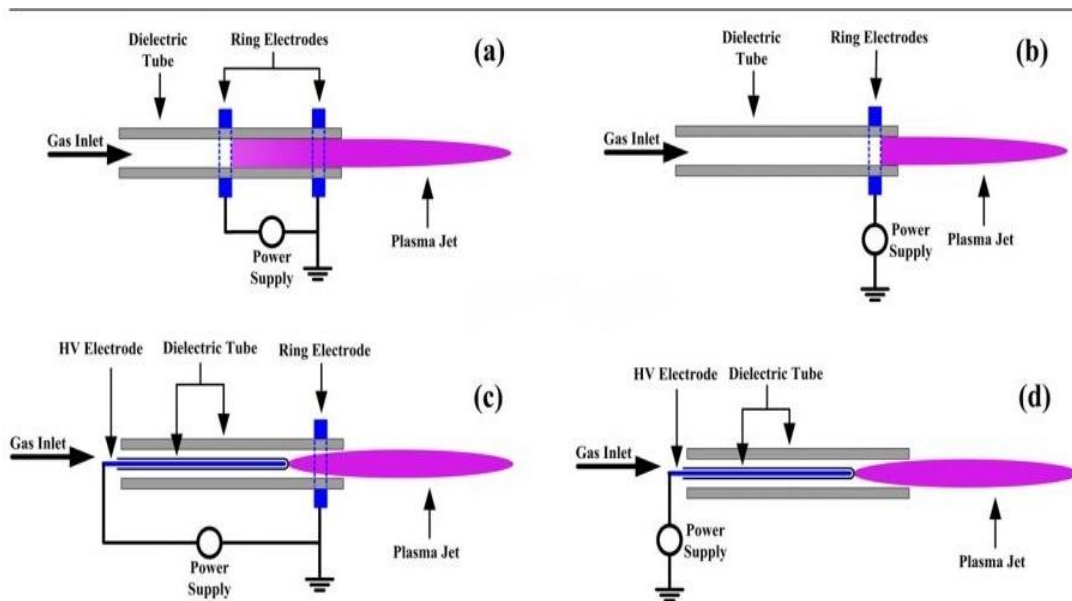


Figure 2.4: Schematic representation of an DBD jet [78]

2.4 Application of Non-Thermal Plasma Jet

There are important advantages of the atmospheric pressure plasma jet, including that it produces a uniform, homogeneous and stable discharge at atmospheric pressure, and the ionized gas from the plasma jet exits through a small nozzle with different ranges of temperature. These advantages enable the plasma generated from the jet to hit the target accurately and at a moderate temperature close to room

temperature on thin surfaces without tissue damage. And in some applications with a high temperature of more than 300°C and a wide nozzle allowing it to treat hard surfaces by directing it as per the demand and application [82,83].

Plasma jet is widely used in material processing. It is also used for thin-film deposition of silicon dioxide using plasma and chemical vapor deposition. Plasma is also widely used in biomedical applications, as it has the advantage of being able to be produced at room temperature. Cold plasma treatment has a degree of control over the treatment area and a good effect because it will show a clear effect between the area that has been exposed to the plasma from the area that has not been exposed to the plasma[83,84].

Delivering cold plasma through a flexible tube can be very useful in endoscopic applications in medicine such as treating colorectal, dental, and pancreatic cancer[65]. Therefore, the development of suitable plasma sources for in vivo therapies has been the subject of extensive research and this type of plasma has been of interest because it provides an open-air conduction medium, reactive plasma species, i.e. radicals, positive and negative ions. and ultraviolet radiation at atmospheric pressure and normal temperature of a target located a few centimeters from the end of the tube in which ionization occurs. The plasma jet machine is designed, built, and operated using a homemade power source suitable for various uses such as biomedicine and medicine [77].

Due to the high precision of the plasma, the plasma treatment process is done by dumping the plasma directly onto the cancerous cells without causing any damage to the surrounding healthy cells[84]. Another important application of the plasma jet device is its use in the

disinfection of materials containing biological and chemical contaminants, in the removal of radionuclides from surfaces and equipment, as well as in the more effective disinfection of large industrial parts from solvents, as well as in the sterilization of dental equipment, surgical equipment, and textile floors. for hospitals [85].

2.5 Reactive Species

When electrons are accelerated at high energy and collide with gas atoms and molecules in different types of reactive plasma, resulting in ionization, excitation, and separation of gas molecules[27]. To control the quality and quantity of the optimum species in the plasma as well as the frequency and voltage applied, Reactive Oxygen Species (ROS) contain species such as O atoms, OH (hydroxyl radicals), and O₃ atoms. While Reactive Nitrogen Species RNS have types like N, NO, and NO₂[86].

The roots are the main component in the sterilization process and can sterilize a large area contaminated with toxic biological agents. Ozone water and free radicals are an effective way to eliminate germs and pathogens and can be used by cells to defend against invading pathogens and for a short period to convert them into stable molecules. Short exposure times do not negatively affect vital cells unlike bacteria and pathogens[87]. Reactive species appear inside and outside Cells at different concentrations, places, and times work to modulate cell signaling pathways that affect immune responses, invading cell death, and proliferation[88].

2.6 Fungal infections of the skin

Fungal skin infections is a skin infection caused by a family of fungi, and it is one of the most common pathogens of human skin. A fungal infection occurs in humans when an invading fungus spreads in an area of the body and the immune system is too large to deal with it and results in different types of fungi such as Trichophyton, Candida, Microsporum, and Epidermophyton [89].

Eczema, mycosis fungoides, athlete's foot, ringworm and jock itch are common in humans, with studies showing that 25% of people have some type of skin fungi. Skin places are preferred for fungi because they are moist and the right temperature. The fungal infection of the skin spreads on the outer surface of the skin, the inner tissues are rarely affected, the symptoms are discoloration, itching and sometimes smell, and the treatment period takes a long time to heal with antifungal drugs [90].

Fungi can live in air, soil, water, and plants. There are also some fungi that live naturally in humans. Like many microbes, there are beneficial fungi and harmful fungi. When harmful fungi invade the body, they can be difficult to kill, as they can survive in the environment and re-infect the person trying to improve, but they increase and pose a threat to human health when immunity is weak and may cause death [91].

2.6.1 Trichophyton Rubrum

Trichophyton Rubrum is an anthropophilic fungus that is considered the most common dermatophyte fungus in humans. Often causing chronic infections of the skin, nails and, rarely, the scalp, this fungus is usually caused the disease to the athlete's foot, ringworm, and jock itch. The growth rate of the fungus is slow to moderately rapid[92]. The fungal tissue is waxy, smooth, or cottony. The outward appearance of the colony is white to yellowish-beige or red-violet. While the lower face of the colony is colored pale yellow, brown, or reddish-brown. Among the group of dermatophytes, this fungus is the most pathogenic for fungal nail infections [93].

2.6.2 Candida Albicans

Candida Albicans is the most famous fungi associated with humans, it is considered as the most fungus that affects human health and colonizes the human skin and mucous membranes naturally, but it is defined as an opportunistic fungus that invades the skin and mucous membranes when the body's immunity decreases, its symptoms are limited to itching and redness of the skin may move to more dangerous places in the mouth[94]. They are initially diagnosed according to the shape of their colonies on the solid medium SDA, which is characterized by being smooth in texture, cream in color, and mucous. Clinically diagnosed by the affected skin. In agar and note the shape of the middle [95].

2.6.3 Candida Tropical

Candida Tropical is a yeast fungus. When it's grown on agar it appears as a creamy white colony and the individual fungal cells are spherical and resemble baker's yeast. About 10 percent of all systemic fungal infections are caused by Tropical Candida, which is one of the most common colonies and pathogens[96]. which are found especially on human skin, in the digestive tract as well as in the female urogenital system. It can be transmitted between health care workers and patients, especially in environments such as hospitals, and is a type of fungi resistant to antifungals [97].

2.7 Determination of electron temperature and density by optical emission spectroscopy

Electron temperature (T_e) is one of the important parameters to describe the properties of the APPJ jet. The measurement is made using the Boltzmann diagram method. The Boltzmann diagram method is a simple and widely used spectroscopic method using the relative densities of two or more line spectra with a relatively large energy difference.

T_e can be evaluated from the Boltzmann plot method using this relationship [98]:

$$\ln \left(\frac{\lambda I}{A g_u} \right) = - \frac{E_u}{K_B T_e} + C \quad (2.1)$$

where I : the relative density of the emitted line, λ : the wavelength, g_u : the statistic weight of the upper plane that can be calculated from the total number of angular momentum quantity J by $g_u = 2J + 1$, A is the

transition probability λ (the probability per second that the atom is in a state The higher level emits in a random direction and is unexcited to the lower level state), k_B : Boltzmann's constant (1.38×10^{-23} J/K), T : temperature in K, C constant and E_u : upper level energy. A histogram plotted for different values of $\ln A_g \lambda I$ versus higher level energies E_u , gives a straight line with a slope. (ionic) emitted by the plasma flow using the Saha-Boltzmann equation which is given as [99]:

$$n_e = 6.04 \times 10^{21} \frac{I^a \lambda^a g^i A^i}{I^i \lambda^i g^a A^a} T^{\frac{3}{2}} \times \exp \left[\frac{E^a - E_{ion} - E^i}{k_B T_e} \right] \quad (2.2)$$

The upper indices a and i denote the neutral particle and the singly charged ion, respectively. E_{ion} is the ionization energy of a neutral particle. The values for E_u , A , E_{ion} , λ , and g_u can be obtained from the National Institute of Standards and Technology (NIST) atomic spectroscopic database [99].

3.1 Introduction

In this chapter, a full explanation of how to create a cold plasma system under atmospheric pressure will be presented. The details of the items used in building the system as well as the electrical measuring devices that were used to measure the voltage and current applied in the system will be presented. Also, thermal and optical devices were used to measure plasma jet temperature and emission spectrometry.

3.2 Components of the DBD system

3.2.1 High voltage AC power supply

To generate plasma in the laboratory, a potential difference is applied to a gas under a certain pressure such that the energy is sufficient to excite the gas atoms and lead to ionization. To build the DBD system, we need a high-voltage alternating electric generator, as shown in Figure (3.1). This device delivers high voltages (0 to 12) kV and at variable frequencies between (0 to 120) kHz. The upper electrode of a cold atmospheric plasma jet system is connected to an alternating power source to generate the plasma jet.



Figure (3.1) A high voltage AC power supply

3.2.1 Pyrex tube

The atmospheric pressure plasma used in this work is a DBD type. The elements of the system are a pyrex tube used as a barrier discharge. Its dimensions are 95 mm in length and 0.9 mm in wall thickness. The inner and outer diameters are 3.2 mm and 5 mm, respectively, as shown in Figure (3.2), where different diameters were used, but the pyrex tube (B) was chosen to obtain the best jet length for it.

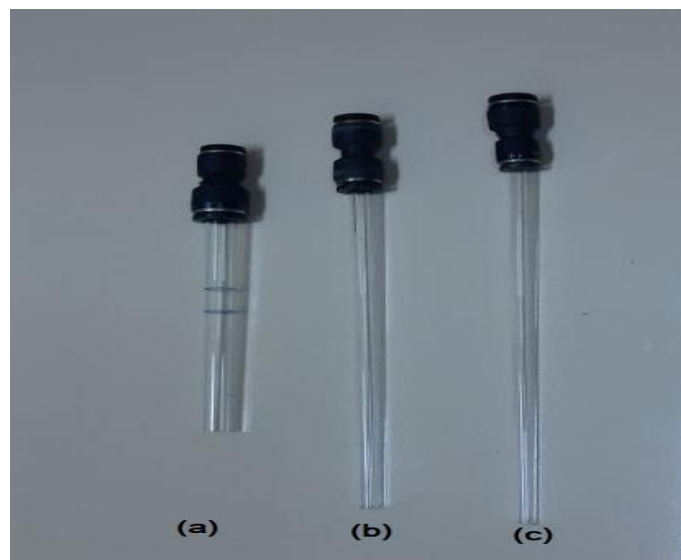


Figure (3.2) Photograph of the Pyrex tubes.

3.2.3 The ring electrodes

The double ring was used to build a plasma jet system using circular electrodes made of thin aluminum wrapped around an outer Pyrex tube. The ring electrodes are 0.1 mm thick and 10 mm wide. The distance between the electrodes was 12 mm and the distance between the ground electrode and the orifice of the Pyrex tube was 3 mm, and it was found that it is the best measurement experimentally and does not cause sparks. The upper electrode is connected to the high voltage AC power source and the lower electrode is connected to the ground (as shown in Figure (3.3)).

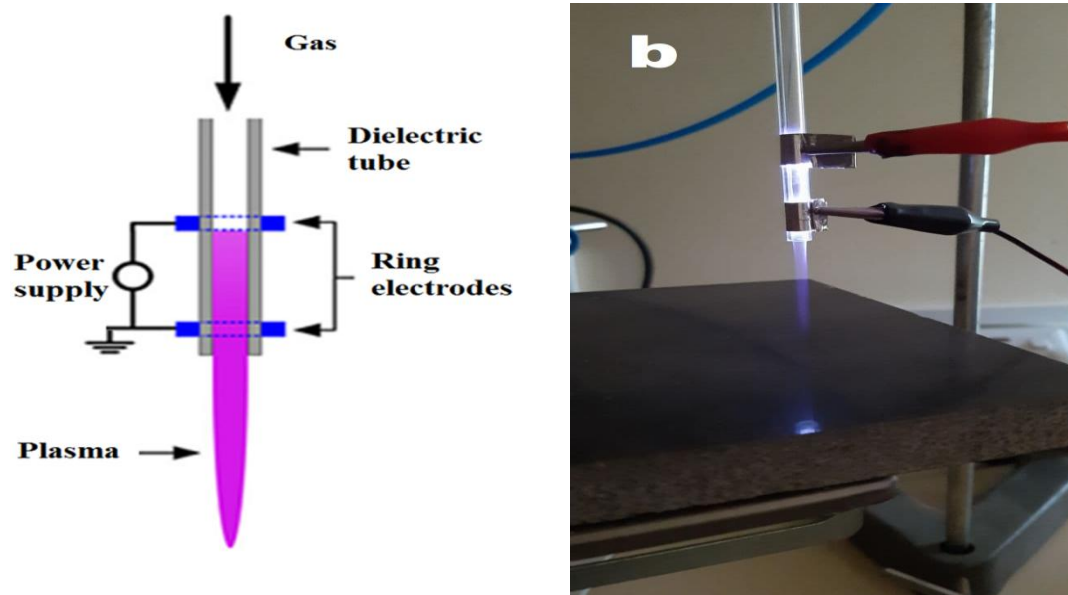


Figure (3.3) : Schematic of the DBD plasma jet. (a) Ring to plate electrode configuration. (b) Photograph of the Pyrex tube with A ring electrodes .

3.2.4 Gas Flow Controller

Commercial argon gas (99.999%) is fed to the top of the Pyrex tube by a flexible plastic tube. The flow meter is used to control the amount of argon gas flowing through the flexible plastic tube and is

connected by a tight lock to the pyrex tube, where it is used in a gradient from 1 to 6 liters/minute as shown in Figure (3.4).



Figure (3.4) flow meter .

3.3 Measurement of plasma jet System Parameter

3.3.1 Measurement of the Electrical Characteristics

An electric oscilloscope was used, an electronic measuring device that allows an electrical signaling device to be shown and plotted in the form of a two-dimensional graph, usually with two inputs. Use an oscilloscope type (PeckTech 1265) where one or two signals can be drawn on the screen.

The oscilloscope input is connected to a high-voltage probe (PD-28). The voltage is divided from 1 to 1000V and the bandwidth is 75MHz. The upper part of the high voltage probe is touched by the high voltage and frequency wire. The oscilloscope will be connected to a computer to show the applied voltage waveforms as shown in Figure (3.5)



Figure (3.5) : Photograph of high voltage probe.

The input of the other oscilloscope is connected to a secondary coil used to measure the current in the wire at a high frequency. The wire will pass through the secondary coil. The voltage transformer is to counter an alternating magnetic field in a coil inside the circuit. The coil is wrapped around an iron core. The magnetic field of the other coil is spread in the oscilloscope as shown in figure (3.7).

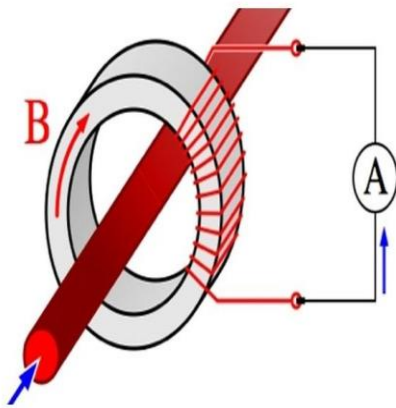


Figure (3.6) Schematic and photograph of a coil current .

3.3.2 Plasma jet length measurement

The plasma jet length, defined as the distance between the tip of the plasma jet and the edge of the Pyrex tube, or the visible length of the plasma jet in ambient air. The plasma jet length, is one of the properties of cold plasma studied in this work. The length of the plasma jet was measured with a metric ruler from the tip end of the tube, at a different gas flow rate and applied voltage as shown in figure (3.7).

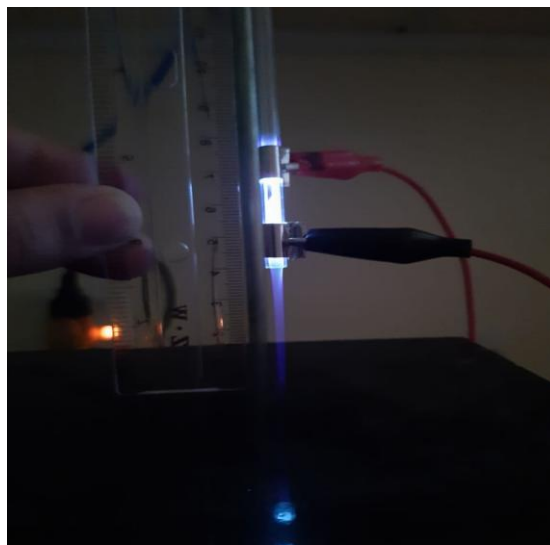


Figure (3.7) : Plasma jet length measurement.

3.3.3 Plasma jet temperature measurement

The temperature of the plasma jet as mentioned earlier is one of the important parameters that determine the scope of its applications. The temperature can be measured using a mercury thermometer in laboratories where the heat-sensitive part is located at different distances from the end of the tube nozzle at different gas flow rates.

3.3.4 Measurement of the Optical Diagnostics

The emission spectra of the plasma jet were measured by a UV-visible near-infrared spectrometer with a wavelength range of 200 nm to 1500 nm as showing in Fig. (3.8). The spectrometer was connected to a fiber-optic cable to record the spectral emission o avoid scattering the plasma radiation, a lens was used to collect the radiation emitted by the plasma. The lens was placed at a distance of 5 cm from the end of the jet to focus the rays. The spectral emission lines data and their lengths were recorded by a spectrometer connected to a laptop computer and then plotted.



Figure (3.8): photograph of Optical Emission Spectra (OES)

Figures (3.9 and 3.10) present a schematic diagram, illustration, and a photograph of a downstream DBD cold plasma jet system.

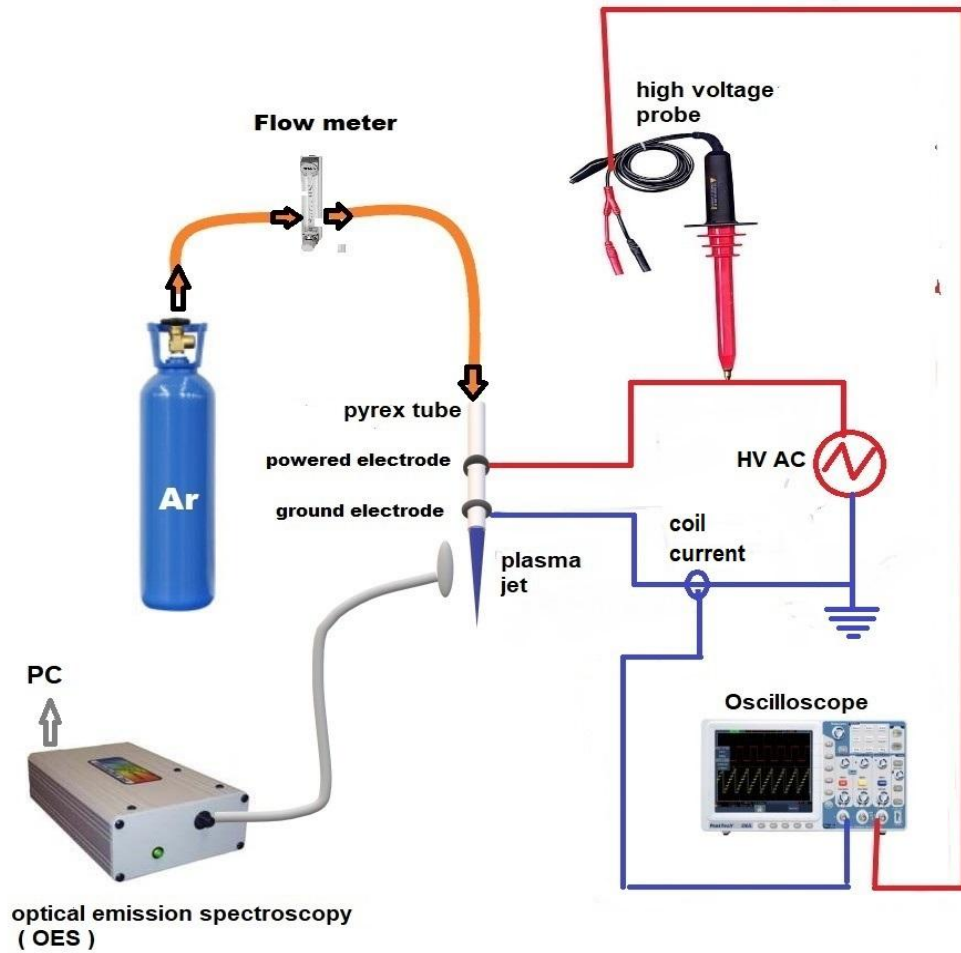


Figure 3.9: Schematic of built DBD plasma jet system (double ring electrodes structure).

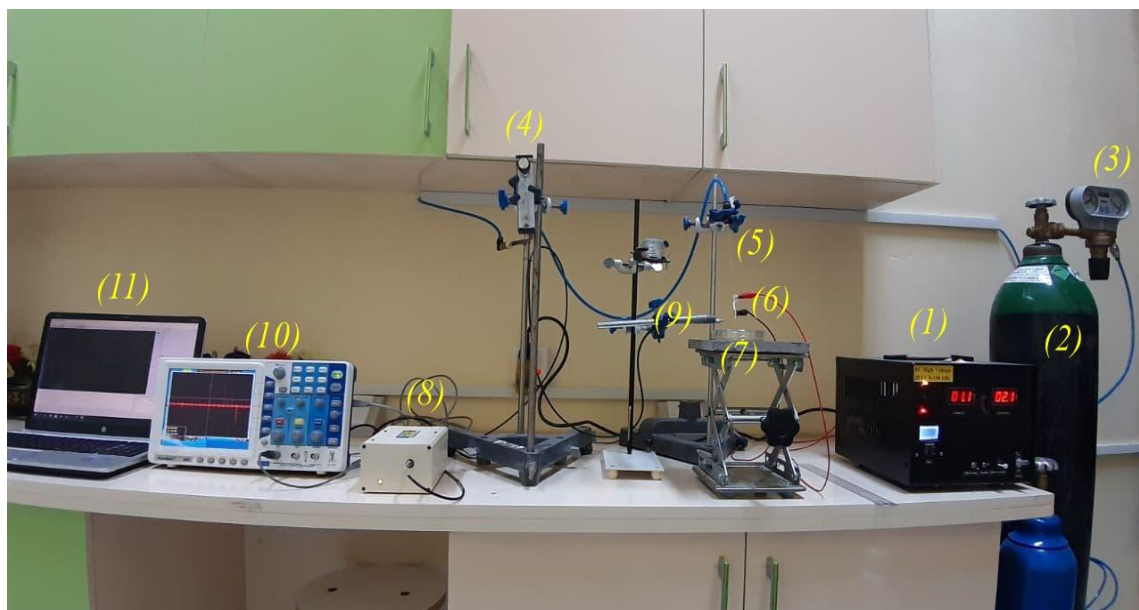


Figure 3.10: Photograph of a DBD plasma jet system.

- (1) Home-made power supply (2) Argon gas (3) Regulator (4) Flow meter (5) Pyrex tube
(6) A ring electrodes (7) Petri dish with sample (8) OES (9) Optical fiber with collimator
(10) Oscilloscope (11) PC.

4.1 Introduction

In this chapter, results such as electrical, thermal, and optical properties of the cold plasma jet system are presented and discussed. The effect of the electrical properties, the amount of voltage and the appropriate frequency for the work of the system, and their effect on the jet length and temperature, were studied. The effect of gas flow rate on the length of the plasma jet and its temperature was also studied. The electron temperature and electron density of the plasma jet were calculated. The appropriate jetting length was found for treatment. The treatment for skin fungi was done by cold plasma for different periods of time and to find the time needed to completely eliminate the fungi.

4.2 Electrical Characteristics

For ionization to occur, an inert gas is passed inside an insulating tube and then an electric field is applied to it so that a certain minimum voltage is applied. This rise slowly leads to an electric discharge in the gas that produces a characteristic light depending on the type of gas used and thus ionization of the gas occurs and plasma is obtained, the parameters adopted for the production of plasma are 4 liters/min, applied voltage 9 kV and frequency 60 kHz on The upper electrode is connected but the lower electrode is connected to the ground. The high voltage was measured by a probe connected to the input No. 1 of the oscilloscope, and the current was measured by a secondary coil connected to the input No. 2 of the oscilloscope, then the oscilloscope screen was connected to a computer and the electrical waveforms were obtained as shown in Figure (4.1).

The use of argon plasma leads in a variable sinusoidal pulse whose values increase with increasing the applied voltage, then the signal decrease before the applied voltage reaches the peak value[98]. This occurs due to the accumulation of negative charge on the Pyrex tube around the negative electrode and then the discharge occurs when the pulse decreases and in this way will avoid the risk of electrical arcing and is an important advantage of the DBD plasma jet system [99].

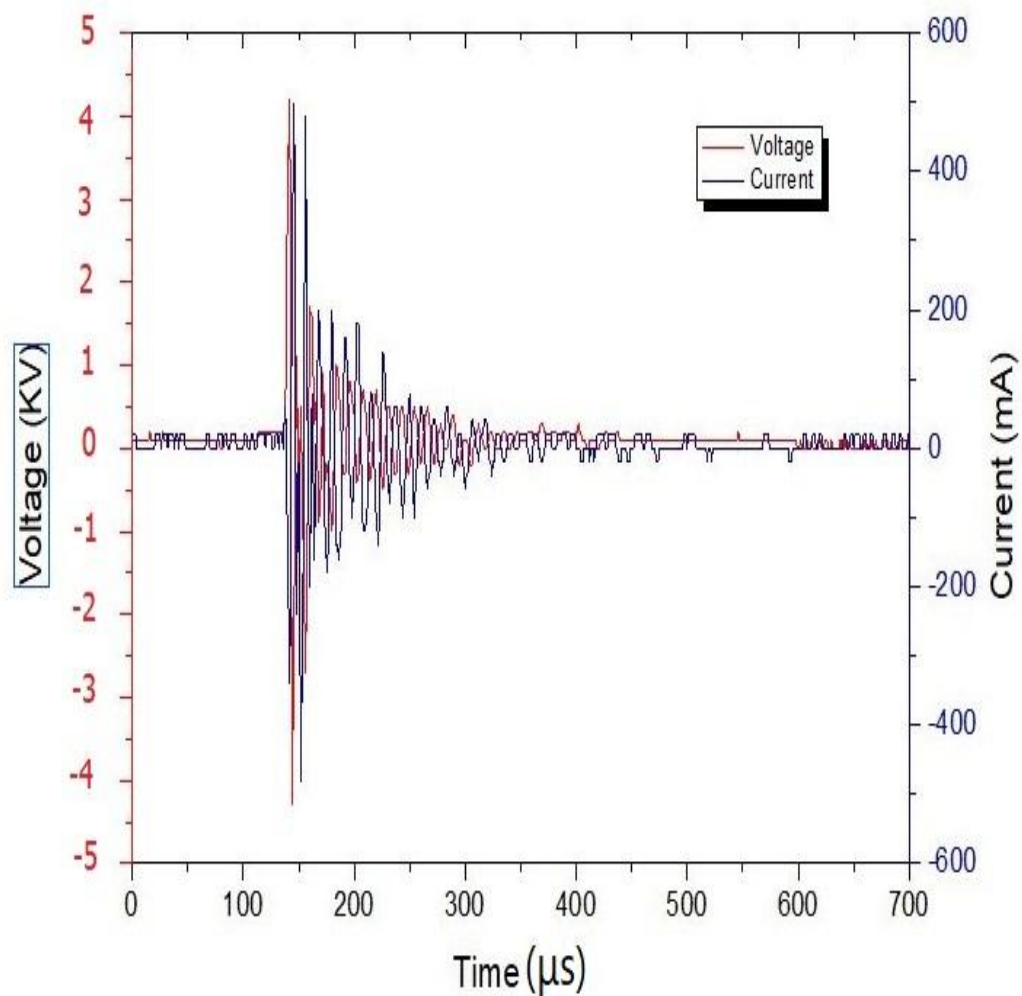


Figure (4.1): Waveforms of the applied high voltage and discharge current.

4.3 Length of the argon plasma jet.

It was found practically after several attempts to operate the system by changing the applied voltage, frequency, and gas flow rate, that the best length of plasma argon jetting is 41 mm at 25 ° C at applied voltage, 11 kV, frequency 60 kHz, and flow rate of 2 liters/min as shown In Figure (4.2).

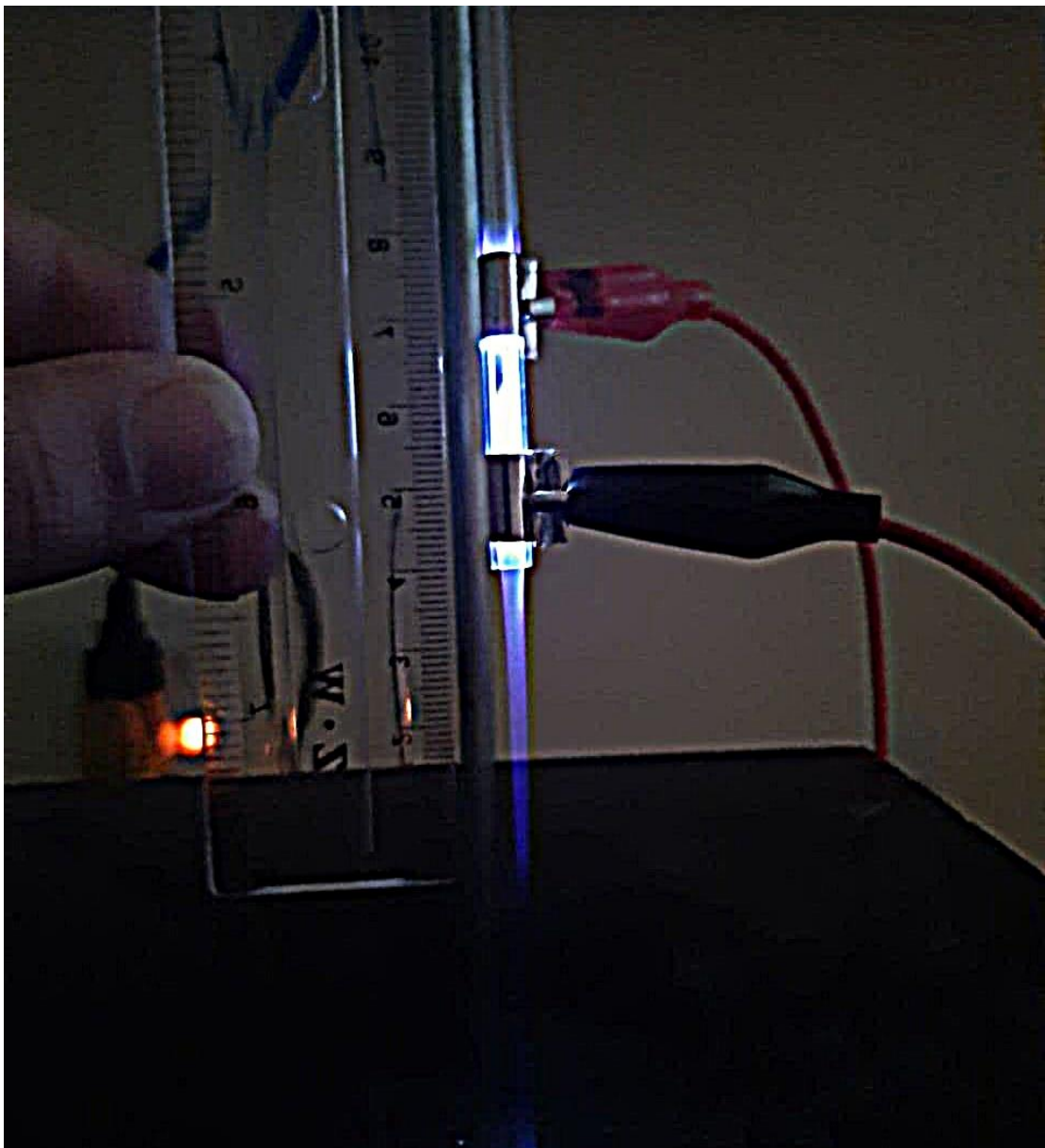


Figure (4.2) : plasma jet length

Figure (4.3) shows the optically measured plasma flow length. It is concluded from the figure that the length of the jet plasma varies with the argon flow rate at a constant input voltage. As the flow rate increases (from 1 L/min to 2 L/min) the length of the plasma flow will increase and we will get the longest plasma flow length of 4.2 cm at 2 L/min when 11 kV voltage is applied. When the argon flow rate is increased (from 2 to 6 L/min), the length of the plasma flow decreases with the increase in the flow rate due to the random increase and decrease in the ionization rate of atoms and (disturbing ionization) will occur.

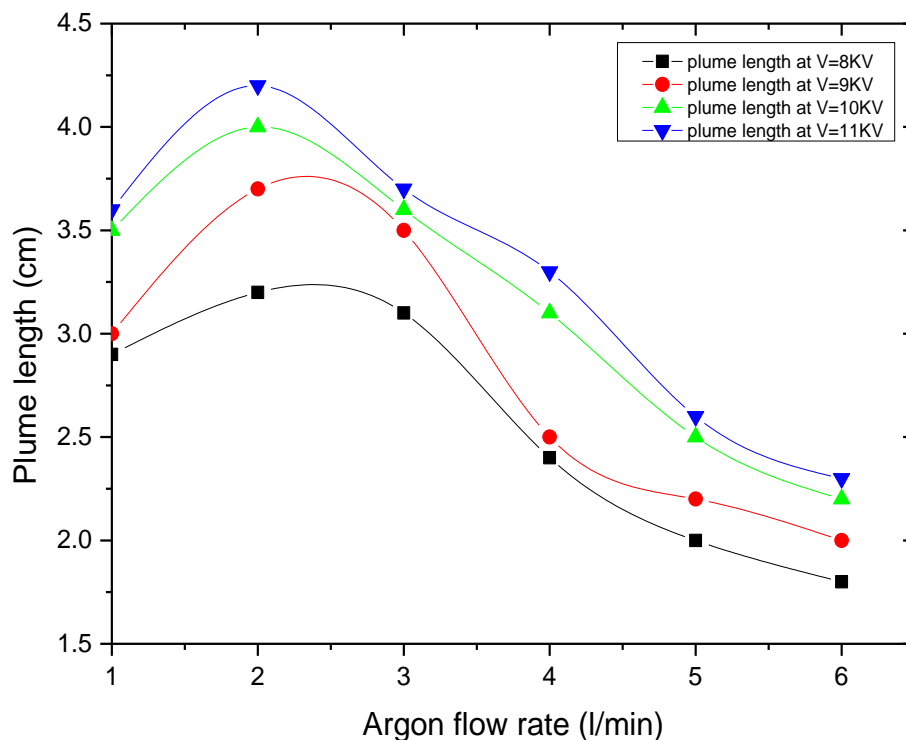


Figure (4.3). The plasma jet length with flow rate and applied voltage.

It is also concluded from Figure (4.3) that the length of the plasma flow increases with the increase of the applied voltage at a constant flow rate.

The longest plasma flow occurs at an applied voltage of 11 kV with a constant flow rate. The increase in the plasma jet length occurs with an increase in the applied voltage because the electric field between the two electrodes increases, and so does the speed and energy of the electron drift, which leads to an increase in the degree of ionization of argon [100,101].

4.4 Plasma Jet Temperature

The temperature of the plasma jet is an important property and one of the main plasma parameters in determining the appropriate application. To measure the temperature, the tip of the thermometer is placed at the end of the plasma flow and it waits for a period of time from 1 to 2 minutes until the system is thermally equilibrated as shown in Figure (4.4).

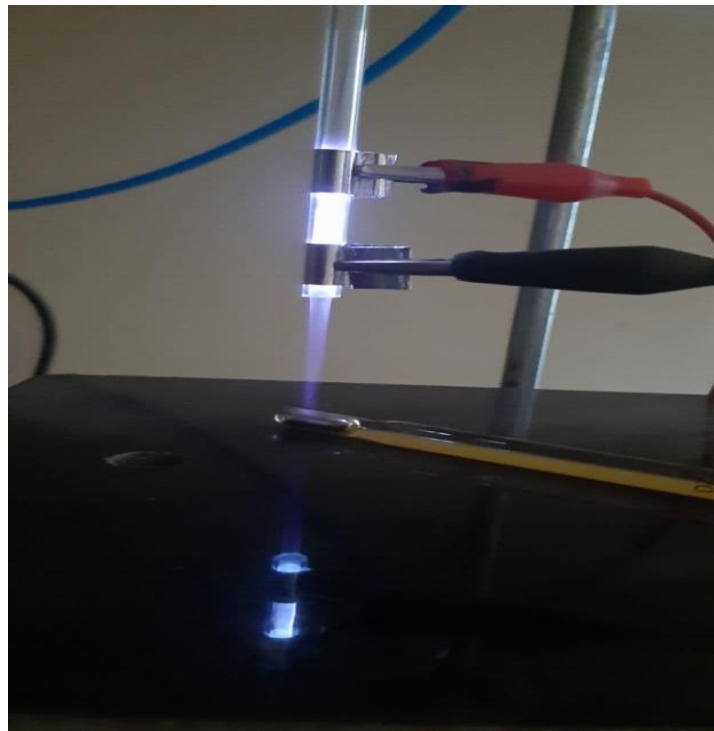


Figure (4.4) Plasma jet temperature measurement using a mercury thermometer.

The plasma temperature decreases when the argon flow rate is increased from (1 liter/min to 6 liter/min), as shown in Fig. (4.5). When argon gas passes at a speed of 6 liters per minute, this behavior is explained by the increase in pressure after the glow and cooling near room temperature. It was concluded from the figure that the temperature increases with an increase in the applied voltage as a result of the increase in the collision. It was found that the lowest plasma temperature is 22°C with a flow of 6 L/min and 8 kV when the laboratory temperature is 20°C.

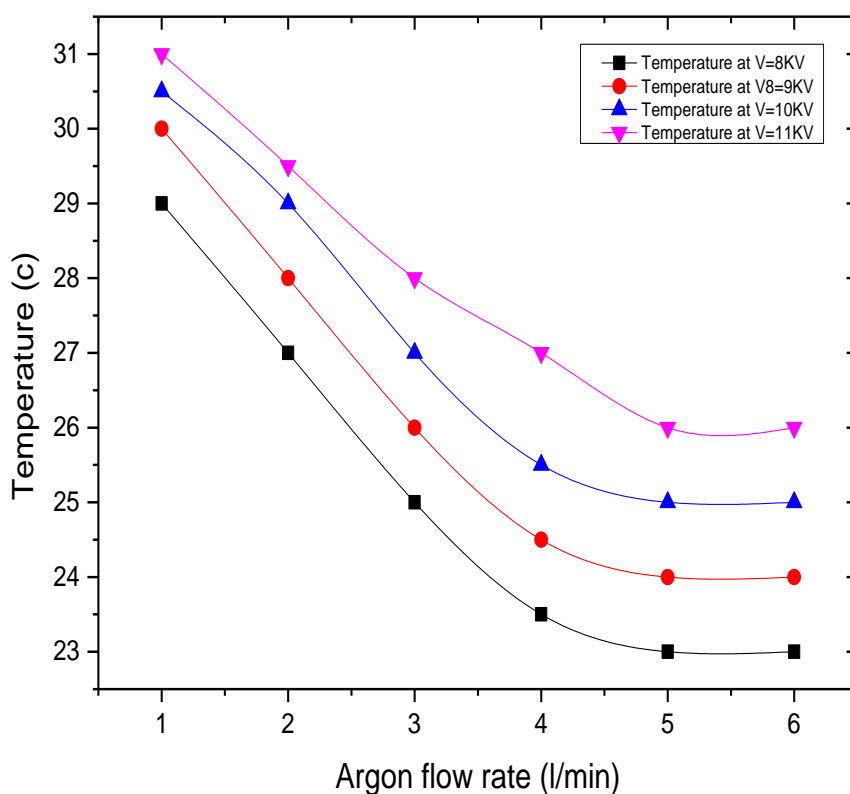


FIGURE (4.5) : The plasma jet temperature at different argon flow rates and applied voltage.

4.5 Optical Emission Spectroscopy Diagnostics.

The spectral emission of the argon plasma jet was analyzed using Optical Emission Spectroscopy (OES). OES is a suitable method for determining the reactive species that used in the cold plasma to treat the fungi. The factors influencing the emission are the voltage difference, frequency, jet length, and argon flow rate that will be discussed to know their effects before treatment and it is illustrated by the argon plasma emission spectra shown in Figures (4.6), (4.7), (4.8) and (4.9).

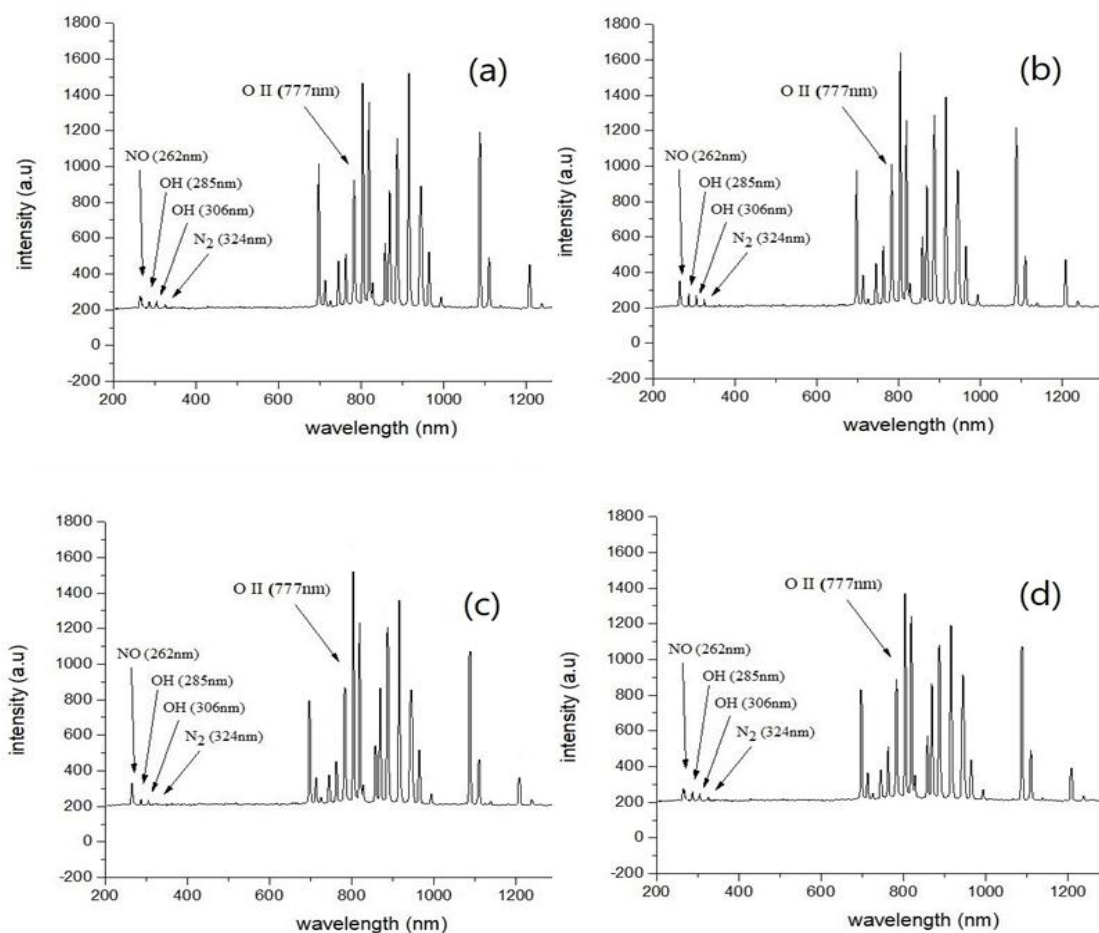


FIGURE (4.6) : The relationship between current frequency and spectral intensity at a voltage of 10 kV and a flow rate of 2 liters per minute, where (a) at 50 kHz, (b) at 60 kHz, (c) at 70 kHz and (d) at 80 kHz.

Table (4.1) shows the intensity values of the reactive species at frequencies (50, 60, 70 and 80) kHz.

Frequency KHz	intensity (a.u)				
	NO (262nm)	OH (285nm)	OH (306nm)	N2 (324nm)	O II 777nm
50	240	220	225	220	750
60	340	280	270	250	990
70	340	235	225	210	850
80	280	250	240	220	890

By comparing the density values of the reactive species in Table 4.1, it will be noted that the best treatment frequency is 60 kHz because the plasma jet, in this case, contains more reactive species and compounds that we need in the treatment process such as (OH, NO, N₂, O) explains the presence of these radicals. The argon jet has interacted with the surrounding air and produced these beneficial radicals that are used in biomedical treatment.

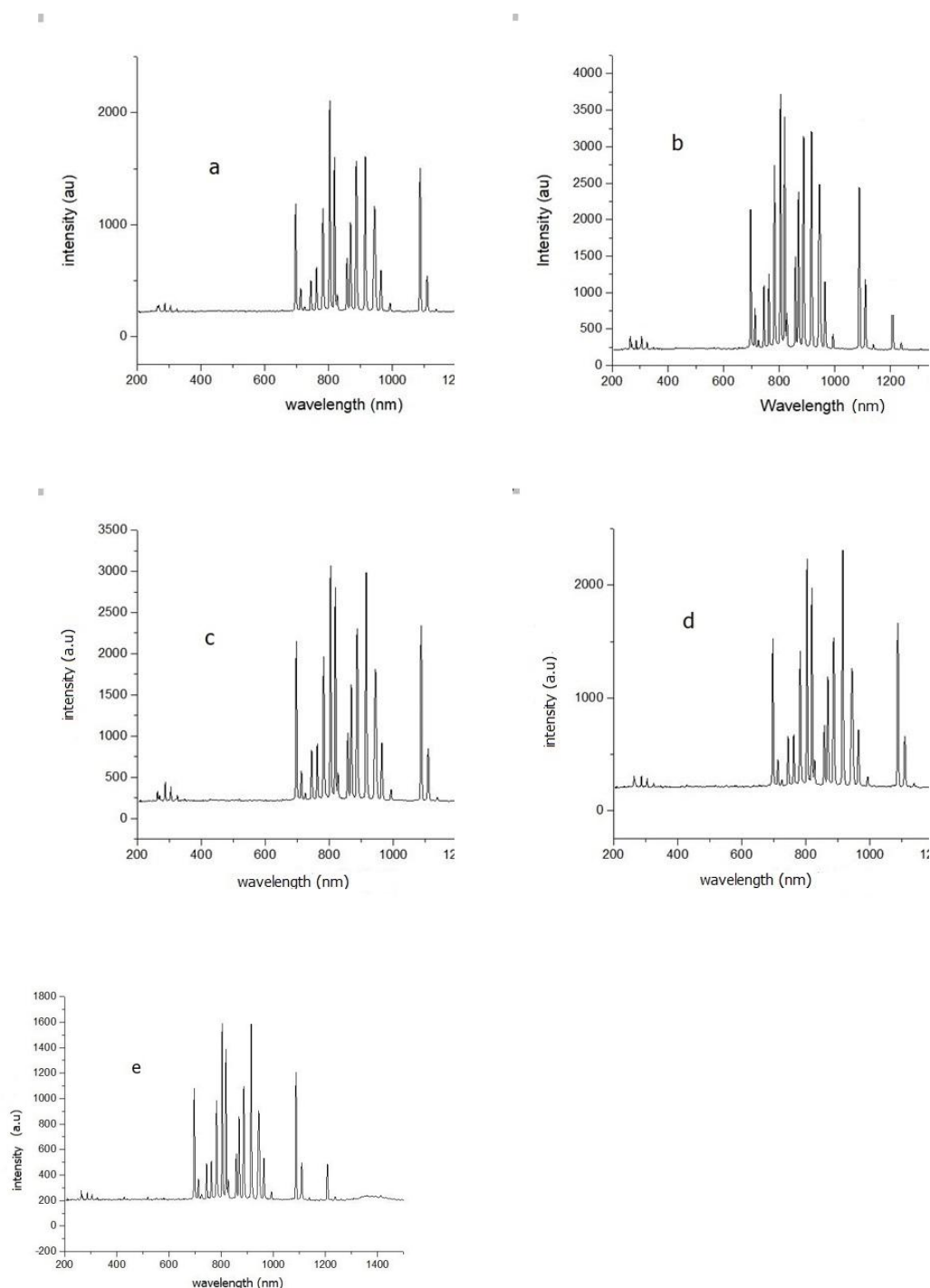


FIGURE (4.7) : The relationship between current frequency and spectral intensity at a voltage of 10 kV, freq=60Hz and a flow rate of 2 liters per minute, where (a) with distance 1 cm from the edge of the tube , (b) distance from edge 2cm ,(c) distance from edge 3cm ,(d) distance from edge 4cm and (e) distance from edge 5cm.

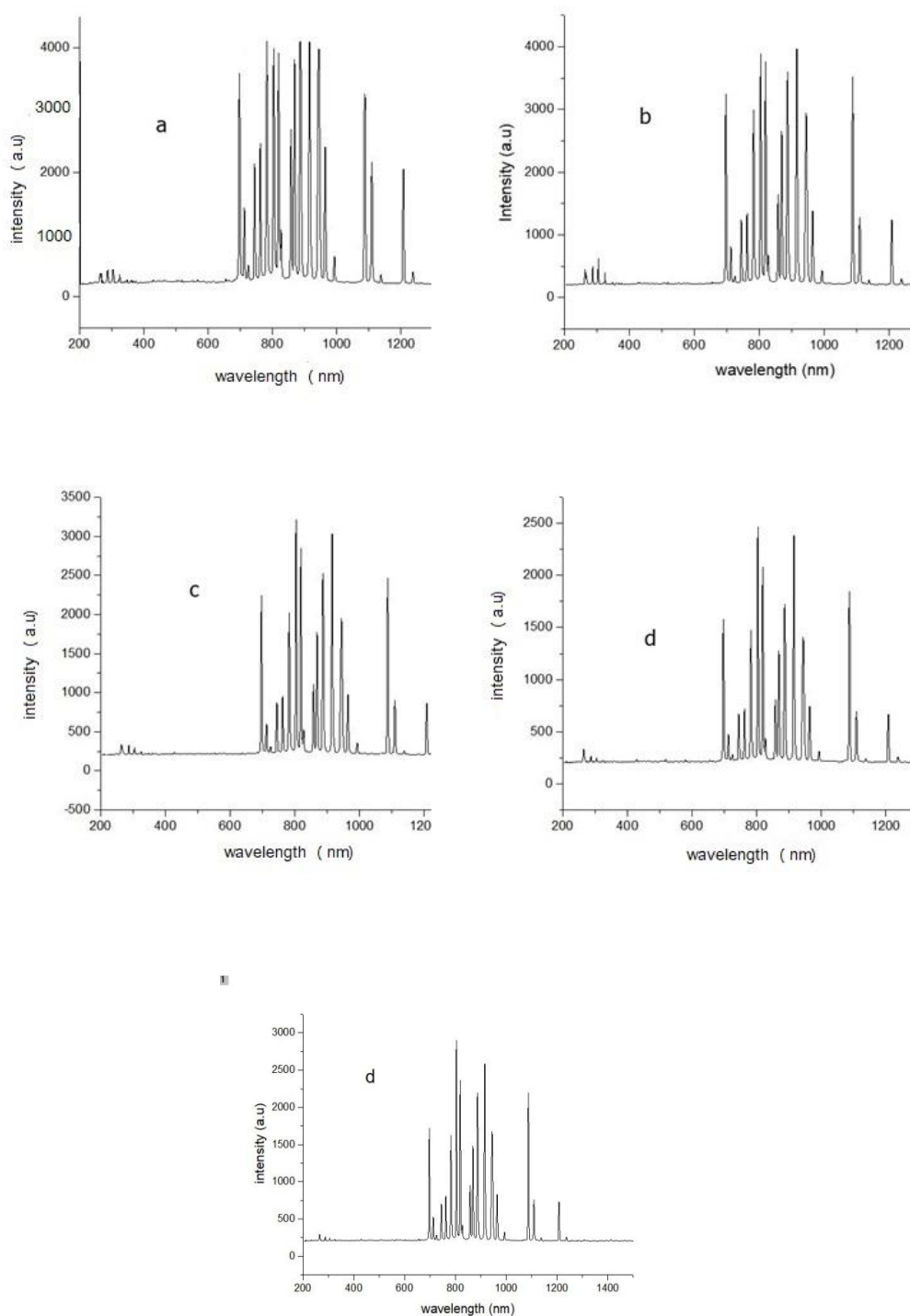


FIGURE (4.8) : The relationship between current frequency and spectral intensity at a voltage of 10 kV, freq=60Hz and a flow rate of 4 liters per minute, where (a) with distance 1 cm from the edge of the tube , (b) distance from edge 2cm ,(c) distance from edge 3cm ,(d) distance from edge 4cm and (e) distance from edge 5cm.

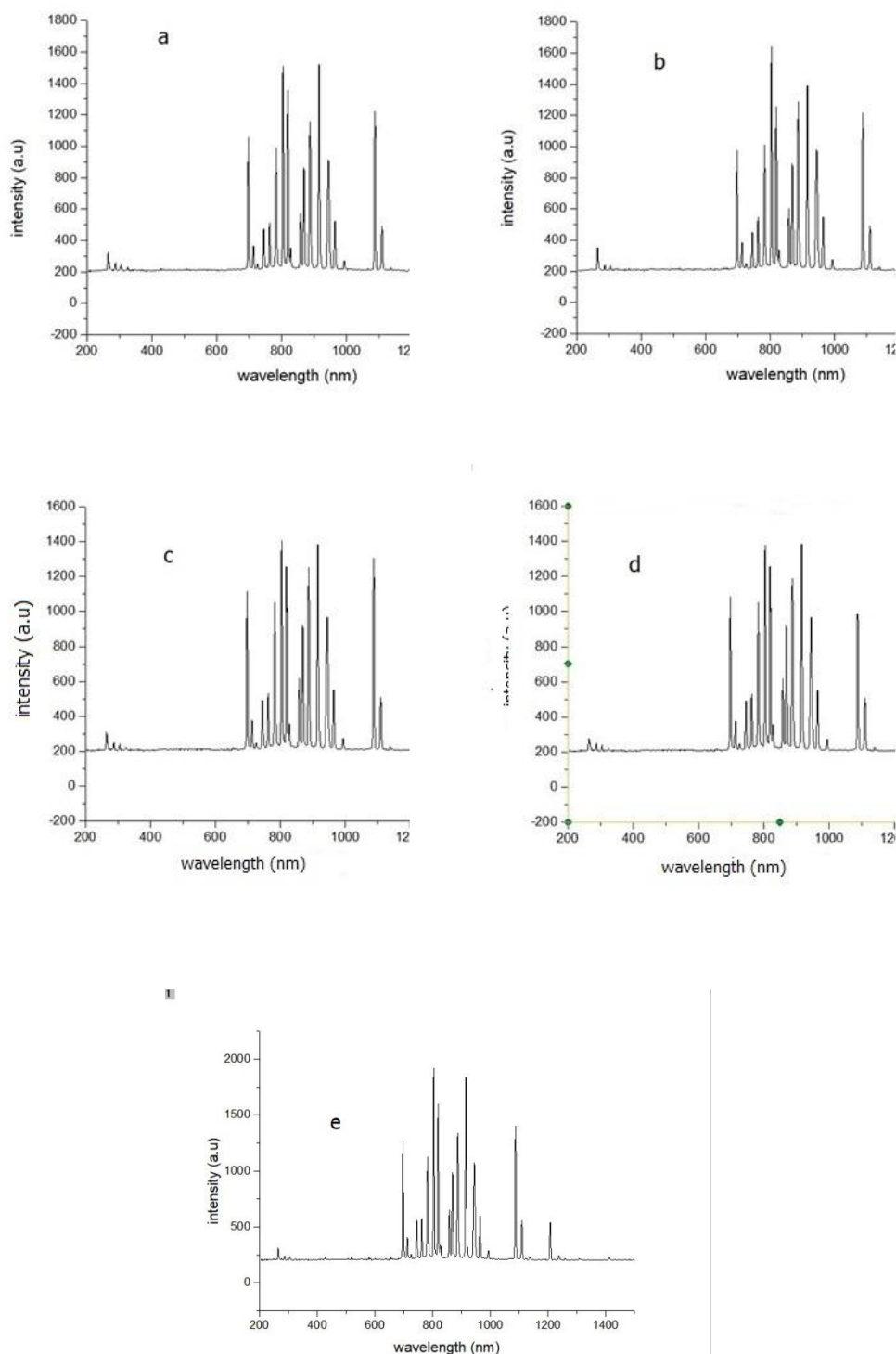


FIGURE (4.9) : The relationship between current frequency and spectral intensity at a voltage of 10 kV, freq=60Hz and a flow rate of 6 liters per minute, where (a) with distance 1 cm from the edge of the tube , (b) distance from edge 2cm ,(c) distance from edge 3cm ,(d) distance from edge 4cm and (e) distance from edge 5cm.

Table (4.2) shows the intensity values of the reactive species with flow rate (2, 4 and 6) l/min at distance from edge (1,2,3,4 and 5) cm

Flow rate	intensity (a.u)				
	NO (262nm)	OH (285nm)	OH (306nm)	N2 (324nm)	O II 777nm
2 l/min at 1 cm	230	250	230	220	1100
2 l/min at 2 cm	400	320	390	300	2750
2 l/min at 3 cm	250	320	300	250	2000
2 l/min at 4 cm	300	300	280	220	1460
2 l/min at 5 cm	280	260	250	220	1000
4 l/min at 1 cm	400	500	490	300	2600
4 l/min at 2 cm	480	510	600	350	2800
4 l/min at 3 cm	300	280	270	220	1800
4 l/min at 4 cm	290	260	250	220	1400
4 l/min at 5 cm	260	250	240	220	1400
6 l/min at 1 cm	500	550	500	300	2500
6 l/min at 2 cm	400	370	250	220	2000
6 l/min at 3 cm	380	300	250	220	2000
6 l/min at 4 cm	300	250	250	220	1800
6 l/min at 5 cm	280	240	240	220	1100

Looking to the table (4.2), we can conclude that the best curing length is 2 cm from the edge of the insulating tube because it contains the plasma components [excited atoms and molecules, reactive species (O^{+2} , OH, NO, etc.)] At the right intensity, it will also make us safe from electric field problems if we get closer. The best flow rate of argon gas compared to the rest of the cases is 4 liter/minute because it contains a

rich percentage of treated plasma elements, as well as it is economically appropriate. As for treatment with a distance greater than the length of the jet, it turns out that the elements in the plasma will still be present, but in weak proportions.

4.6 Diagnosis of Electron Temperature and Density

The active species can be obtained by analyzing its emission spectrum. Also, the diagnostics of plasma parameters (T_e and n_e) are estimated by employing the emission spectrum. Figure (4.11) shows the emission spectrum lines of argon plasma jets at system conditions of applied voltage 10 kV, flow rate 4 l/min, and a frequency of 60 kHz under plasma temperature is 22°C. Figure (4.10) shows the emission spectrum line and their wavelength for the argon and some other reactive species. These lines were obtained from the OES. The species were identified by comparing the data that we got from the OES with the data from NIST [102]. A series of spectral lines can be noticed in the argon plasma jet as shown in Figure (4.10). The emission spectrum lines appear in the UV-Vis-NIR regions. The spectral lines also contain hydrogen oxide (OH), excited molecular nitrogen (N₂), and ionic oxygen (OII) which was observed in the emission spectrum of the argon plasma jet.

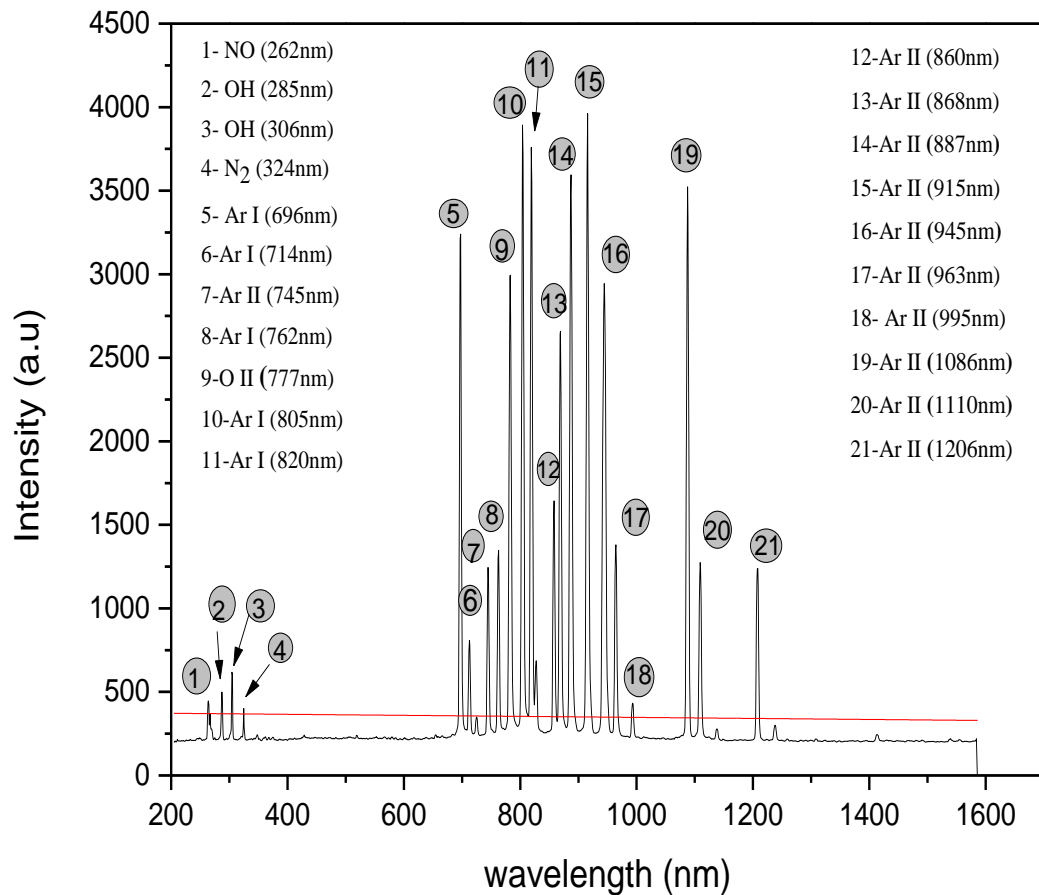


FIGURE (4.10) . Emission spectrum lines of argon DBD plasma jet.

Boltzmann plot method is used to calculate the T_e for argon plasma jets. Five distinct spectral lines (696.543, 714.704, 762.89, 805.33, and 860.578) nm are considered to determine the T_e . The relative intensity was taken from OES. The T_e for argon plasma jet was evaluated using the Boltzmann plot method by the relation [98].

$$\ln\left(\frac{\lambda I}{A g_u}\right) = -\frac{E_u}{K_B T_e} + C \quad (2.1)$$

where I is the relative intensity of the emitted line, λ is the wavelength, K_B is the Boltzmann constant (1.38×10^{-23} J/K), (T_e) is the

electron temperature in K, C is a constant and E_u is the energy of the upper level. A graph is plotted for different values of $\ln\left(\frac{\lambda I}{Ag_u}\right)$ versus the energies of the upper level E_u give a straight line with a slope of (-1.0709) which corresponding to T_e of 10826.6 K (see figure 4.11). The argon spectral lines parameters A, g_u, E_u can be obtained from NIST [98,100] (see table 4.3).

Table (4.3). Parameters of atomic argon lines used for the Boltzmann plot method to calculate T_e .

Species	Wavelength λ (nm)	Intensity (a.u)	A (S^{-1})	g_u	E_u (cm^{-1})	E_u (ev)	$\ln\left(\frac{\lambda I}{Ag_u}\right)$
Ar I	696.543	3240.2	6.40E+06	3	107496.416 6	13.28261776	1.012681241
Ar I	714.704	808.9	6.30E+05	3	107131.708 6	14.95258081	2.363873583
Ar I	762.89	1347.2	2.90E+05	3	120600.890 5	14.71087456	2.392291893
Ar I	805.33	3893.7	8.60E+05	3	118651.395	14.74251726	1.693385134
Ar I	860.578	1642.9	1.04E+06	4	118906.611	13.32783572	0.056323748

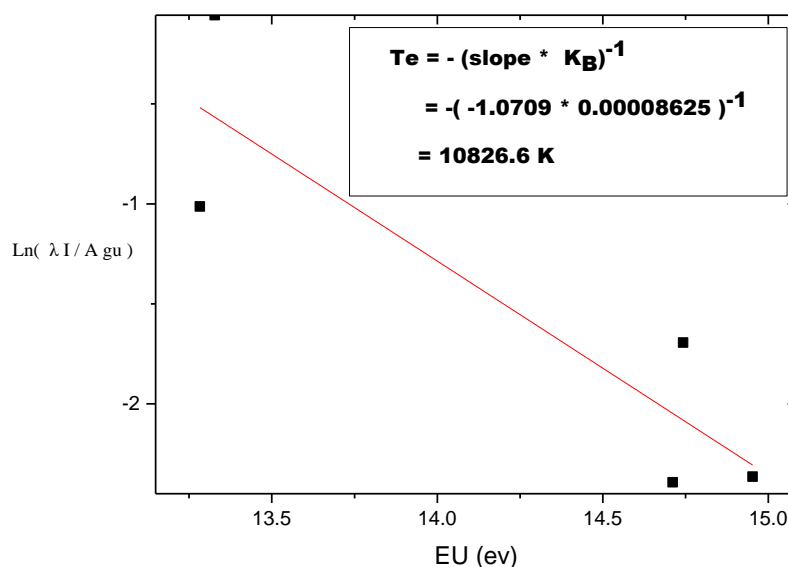


FIGURE 4.11 Boltzmann plot for atomic argon spectral lines

The calculation of electron density n_e can be determined from the neutral particle (atomic) and the singly charged ion (ionic) spectral lines emitted from the plasma jet using the Saha-Boltzmann equation which is given by [99]:

$$n_e = 6.04 \times 10^{21} \frac{I^a \lambda^a g^i A^i}{I^i \lambda^i g^a A^a} T^{\frac{3}{2}} \times \exp \left[\frac{E^a - E_{ion} - E^i}{k_B T_e} \right] \quad (2.2)$$

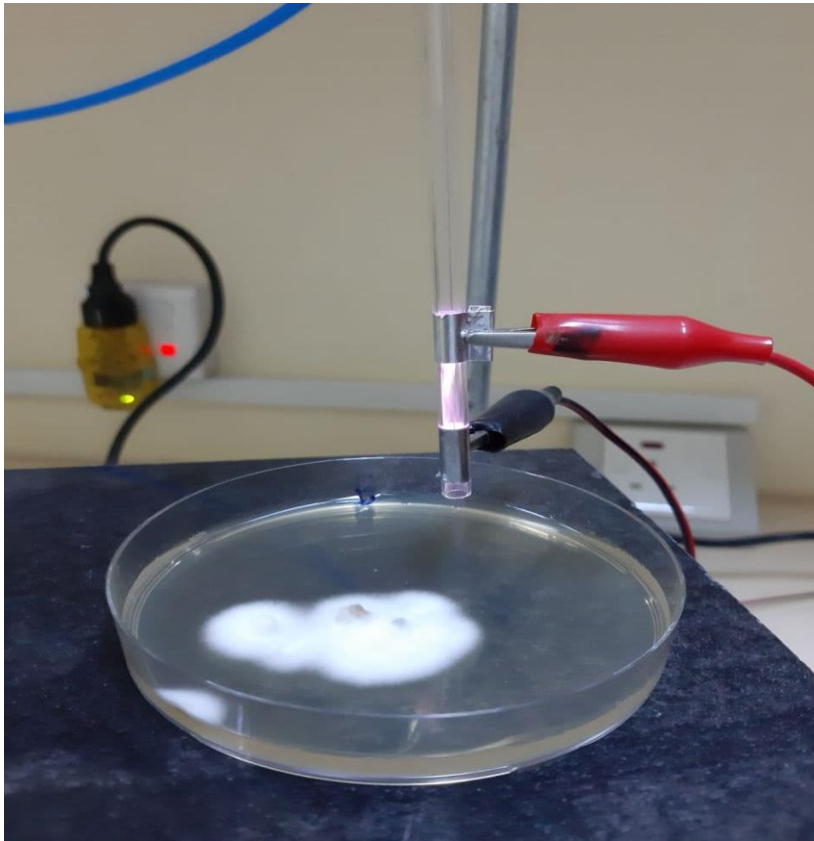
The upper parameters refer to the neutral particle and the singly charged ion, respectively. E_{ion} is the ionization energy of a neutral particle. The values for E_u , A , E_{ion} , and g_u can be obtained from the National Institute of Standards and Technology (NIST) atomic spectroscopic database [100].

The electron density n_e can be determined using equation (2) , by applying the first wavelength 696.54 nm, intensity and wavelength of the last 860.57 nm , and its intensity, the electron density of the cold plasma was obtained , it is equal to $1.57 \times 10^{13} \text{ cm}^{-3}$.

4.7 Treatment of *Trichophyton Rubrum* Samples

The pure cultured media of *Trichophyton rubrum* were cultured in petritic dishes containing Sabouraud Dextrose Agar (SDA), then placed in an incubator at 30°C for 7 days to grow, then the dishes were extracted and treated with plasma jet at 10 kV voltage, 60 kHz frequency, and flow rate. For argon gas 4 liters/min and a distance from the edge of 2 cm and for different periods of time (2, 4, 6, 8, 10, 15, 20, 25) minutes as shown in Figure (4.12), a piece is taken from the place of treatment and planted in Petritic dishes containing SDA agar, and

compared to a sample not exposed to plasma jets which is placed for 7 days in the incubator at 30°C.



Figure(4.12) : A petri dish containing fungi treated with cold plasma

There is significant difference between the untreated and the treated samples of *Trichophyton rubrum* with cold plasma in their ability to grow and form spores, as 33 % of their growth is lost after a period of 2 minutes of treatment, 50 % after 4 minutes, 60 % after 6 minutes, 75 % after 8 minutes, 85 % after 10 minutes, 90 % after 15 minutes, 95 % after 20 minutes, and the fungal cell lost its ability to grow after 25 minutes of plasma treatment as shown in figure (4.13).

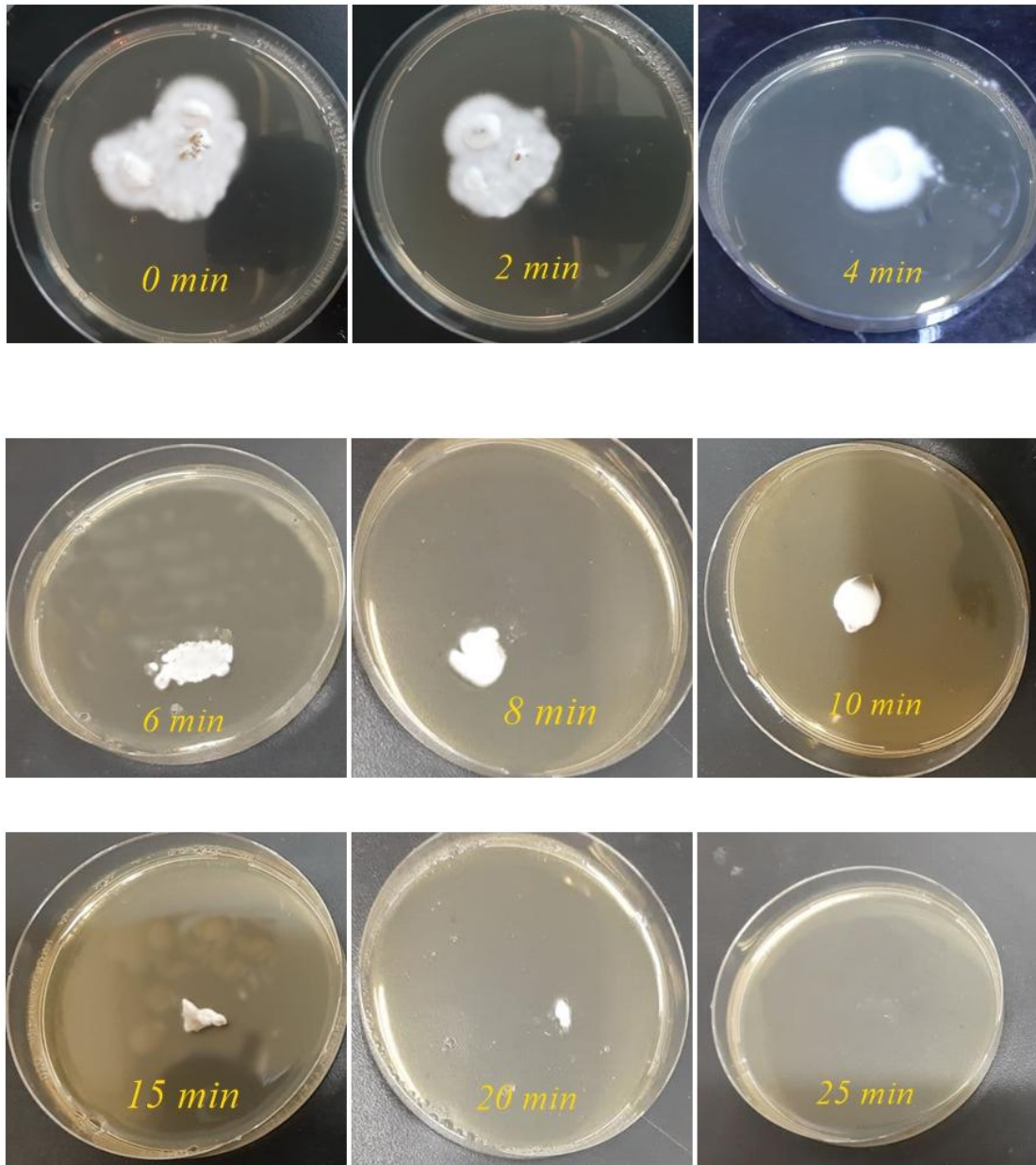


Figure (4.13) shows the growth of *Trichophyton Rubrum* after treatment with cold plasma for different periods.

4.8 Treatment of Candida Albicans Samples

Pure fungal media of candida albicans is cultured in petri dishes containing SDA agar, and then placed in an incubator at a temperature of 30 Celsius for a period of 14 days, the dishes were extracted and treated with the plasma plume at different time (2, 4, 6, 8, 10, 15) minutes, it is cultured in petritic dishes containing SDA agar, and compared with a sample that was not exposed to plasma jet, which its cultured for a period of 14 days in a special incubator at a temperature of 30 Celsius. It is found that there is difference between the untreated samples and that are treated with cold plasma in their ability to grow and form spores, as 30 % of their growth is lost after a period of 2 minutes of treatment, 55 % after 4 minutes, 74 % after 6 minutes , 86 % after 8 minutes, 95 % after 10 minutes, and the fungal cell lost its ability to grow after 15 minutes of plasma treatment as shown in figure (4. 14).

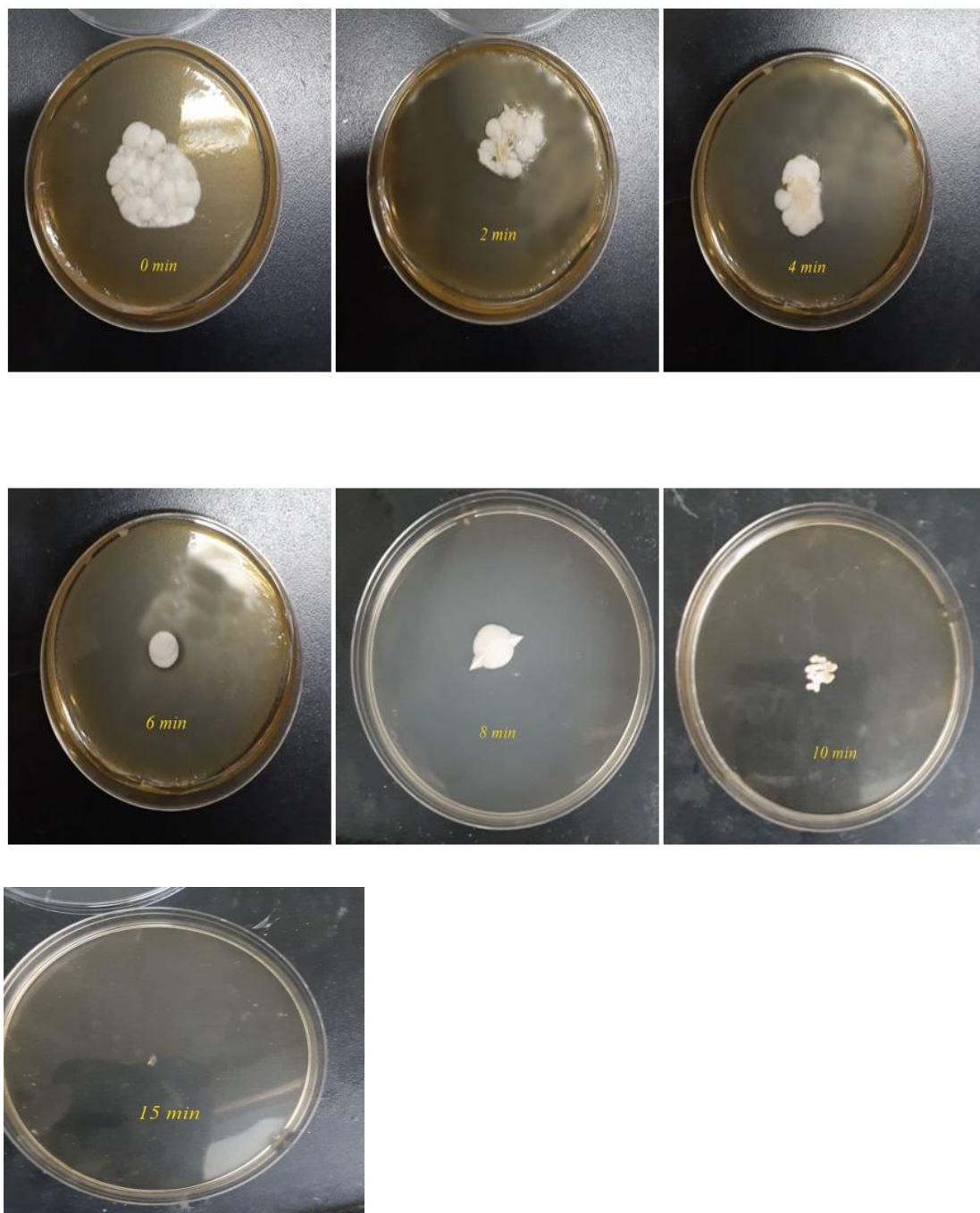


Figure (4.14) shows the growth of *Candida Albicans* after treatment with cold plasma for different periods.

4.9 Treatment of Candida Tropicalis Samples

Pure fungal media of candida tropicalis is cultured in petri dishes containing SDA agar, and then placed in an incubator at a temperature of 30 Celsius for a period of 14 days, then the dishes are extracted and treated with the plasma plume at different periods of time (2, 4, 6, 8, 10, 15,20) minutes, it is cultured in petritic dishes containing SDA agar, and compared with a sample that was not exposed to plasma jet, which is cultured for a period of 14 days in a special incubator at a temperature of 30 Celsius. It is found that there is significant difference between the untreated samples and that are treated with cold plasma in their ability to grow and form spores, as 31 % of their growth was lost after a period of 2 minutes of treatment, 53 % after 4 minutes, 70 % after 6 minutes , 81 % after 8 minutes, 89 % after 10 minutes, 93% after 15 minutes and the fungal cell lost its ability to grow after 20 minutes of plasma treatment ,as shown in figure (4.15).

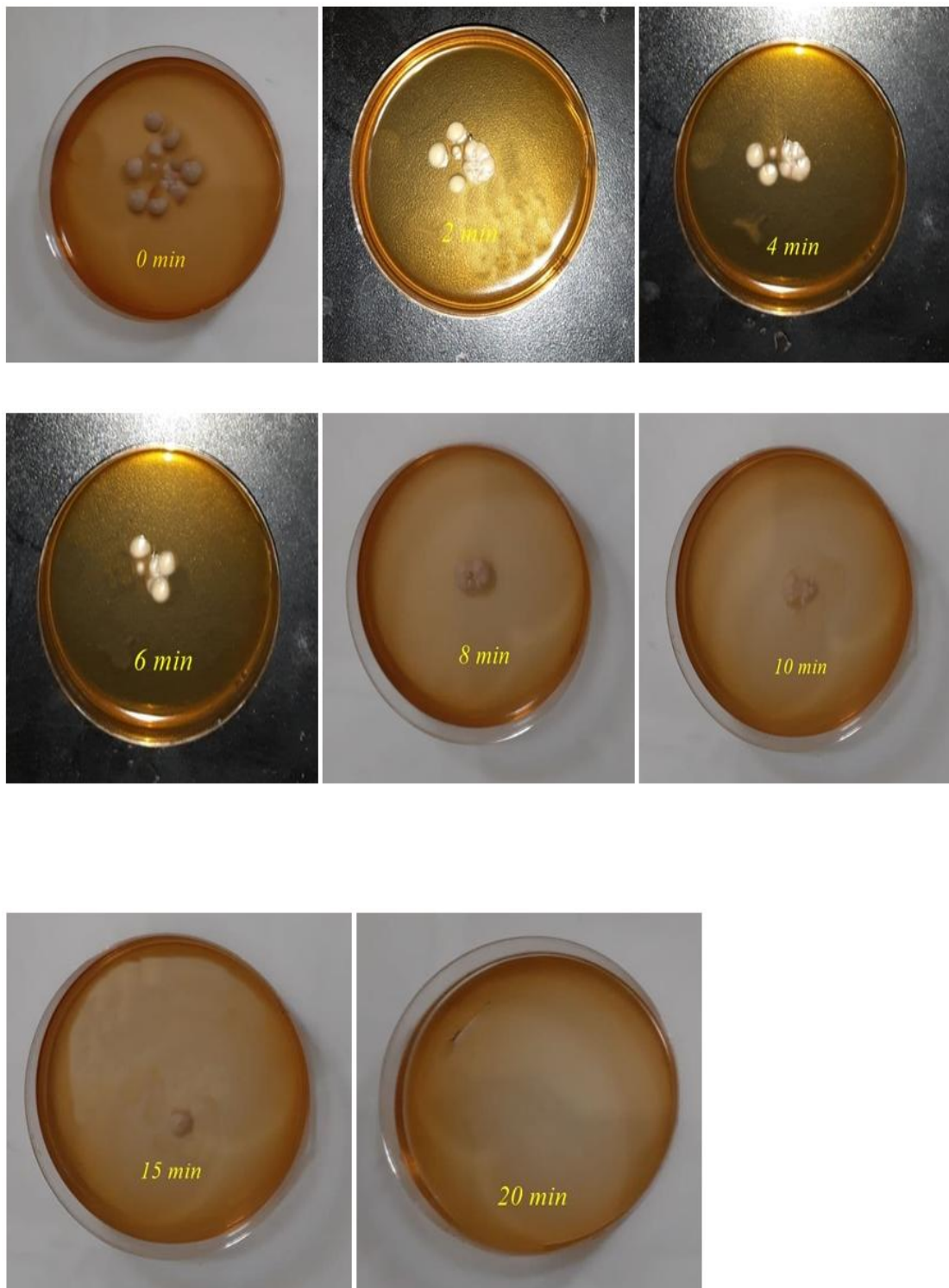


Figure (4.15) shows the growth of *Candida Tropicalis* after treatment with cold plasma for different periods.

4.10 Conclusions

1. Atmospheric Pressure Plasma Jet (double - ring electrode) was built.
2. The optimum jet length of the system was obtained at a voltage of 11 kV, when the frequency was 60 kHz and the flow rate was 2 liters/min.
3. The length of the plasma column depends on the applied voltage and the gas flow rate.
- 4 . The jet temperature increase with the increase in the applied voltage and decreases with the increasing the gas rate.
- 5 . The temperature of the generated plasma jet was close to room temperature (25°C), which make it suitable for biomedical applications.
- 6 . The plasma Electron temperature and electron density were calculated and they found to be within the range that suitable for biomedical applications.
- 7 . The appropriate treatment time was very effective in inhibiting the fungi. It is limited to (15-25) minutes.
- 8- Cold plasma treatment of fungi has proven to be a promising and safe technique.

4.11 Future work

1. Construction a non-thermal plasma jet array system and study the characteristics of the proposed system.
2. Design and construct an atmospheric pressure plasma jet system by using Nitrogen gas and study the electrical and optical properties of the system.
3. Design and construct an atmospheric pressure plasma jet system using a radio frequency (RF) power source and a He/O₂ gas mixture.
4. Utilizing the cold atmospheric plasma in treat the plant leaves that infected with fungi.

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الخلاصة

الأمراض الفطرية تؤثر على سلامة الإنسان بشكل كبير . بعض أنواع الفطريات تبدي مقاومة للمضادات الحيوية عند العلاج و البعض الآخر يحتاج الى فترة طويلة للعلاج . واحدة من التقنيات الحديثة والأمنة استخدام البلازما الباردة في الطب. تم بناء منظومة البلازما النفاث واستخدمت لعلاج بعض أنواع فطريات الجلد ، والتي تضمنت ثلاثة أنواع شائعة : الشعروية الحمراء و المبيضة البيضاء والمبيضة الاستوائية.

تم تصميم نظام نفاث البلازما للضغط الجوي (APPJs) بمكونات بسيطة ومنخفضة التكلفة. استخدم مولد جهد عالي متناوب وكان النظام عبارة عن تكوين أقطاب كهربائية ذات حلقة مزدوجة ، استخدم أنبوب Pyrex كحاجز عازل ، كما استخدم غاز الأرجون النقي لتغذية المنظومة .

تم تشخيص الخصائص الكهربائية للمنظومة و دراسة تأثير معدل تدفق الغاز والجهد المطبق على طول نفاث البلازما ودرجة حرارة البلازما. استخدم التحليل الطيفي للانبعاش الضوئية لحساب درجة حرارة الإلكترون وكثافة الإلكترون. وكانت درجة حرارة الإلكترون (K 10826.6) وكثافة الإلكترون (1.57×10^{13} سم⁻³). اظهرت النتائج أن النظام مناسب للاستخدام في التطبيقات الطبية .

تم الحصول على عزلة نقية من الفطريات تم إنمائها على أجار SDA الصلب. عولجت الخلايا الفطرية بالبلازما الباردة عند 25 درجة مئوية. كانت فترات العلاج تتراوح بين (2 ، 4 ، 6 ، 8 ، 10 ، 15 ، 20 ، 25) دقيقة. أظهرت النتائج أن نمو الخلايا الفطرية يتناقص مع زيادة فترة العلاج ويؤدي بالتالي إلى قتل الخلية الفطرية. في علاج فطريات الشعروية الحمراء ، فقدت الخلية قدرتها على النمو بعد

25 دقيقة من العلاج بالبلازما ، لكن الوقت اللازم لقتل الخلايا في حالة المبيضة البيضاء و المبيضة الاستوائية كان اقصر (15، 20) دقيقة على التوالي.من خلال النتائج نرى ان البلازما الباردة تقنية واعدة للقضاء على الفطريات قيد البحث .



جمهورية العراق

وزارة التعليم العالي والبحث العلمي

جامعة كربلاء

كلية العلوم / قسم الفيزياء

بناء منظومة بلازما الضغط الجوي الباردة لاستخدامها في

معالجة فطريات الجلد

رسالة مقدمة الى

مجلس كلية العلوم - جامعة كربلاء

كجزء من استكمال متطلبات نيل شهادة الماجستير

في علوم الفيزياء

تقدم بها

عمار سلمان محمد حسين

بكالوريوس علوم فيزياء (٢٠٠٦) / جامعة بابل

اشرف

أ.د فاضل خدام فليفل

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