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Modeling of Various Compositional Changes Occurring in the Sliced Chicken Treated with Cold Atmospheric Plasma

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ABSTRACT: Consumers demand for quality of food has triggered the need for the development of a number of non-thermal approaches to food processing, of which Cold Plasma technology has proven to be very valuable. This review and research aims to develop the models for various constituents of sliced chicken undergoing Cold Plasma treatment for increasing the self-life. The models developed are as a function of cold atmospheric plasma treatment for (0-1.5) minutes, by plotting the graph and finding the trend equation with there R^2 on M S Excel.

KEYWORDS: Cold Atmospheric Plasma, Molecular oxygen , Modeling, Ionized gas, M S Excel.

1 INTRODUCTION

1.1 Plasma Science

1.1.1 Plasma- Definition, Physics and Chemistry

In 1922, the American scientist Irving Langmuir proposed that the electrons, ions and neutrals in an ionized gas could be considered as corpuscular material entrained in some kind of fluid medium and termed this entraining medium *"plasma",* similar to the plasma, introduced by the Czech physiologist Jan Evangelista Purkinje to denote the clear fluid which remains after removal of all the corpuscular material in blood. However, it emerged that there was no "fluid medium" entraining the electrons, ions, and neutrals in an ionized gas (Bellan 2006), nevertheless the name prevailed. The term "plasma" refers to a partially or wholly ionized gas composed essentially of photons, ions and free electrons as well as atoms in their fundamental or excited states possessing a net neutral charge. The plasma possesses a net neutral charge because the number of positive charge carriers is equal to the number of negative ones (Kudra and Mujumdar 2009). Electrons and photons are usually designated as "light" species in contrast to the rest of the constituents designated as "heavy" species. Due to its unique properties plasma is often referred to as the fourth state of matter according to a scheme expressing an increase in the energy level from solid to liquid to gas and ultimately to plasma.

1.1.2 Types of Plasma

Two classes of plasma, namely thermal and NTP can be distinguished on the basis of conditions in which they are generated. This classification of plasma is based on the relative energetic levels of electrons and heavy species of the plasma. NTP (near ambient temperatures of 30-60oC) is obtained at atmospheric or reduced pressures (vacuum) and requires less power. NTPs are characterised by an electron temperature much above that of the gas (macroscopic temperature) and consequently do not present a local thermodynamic equilibrium. NTP can be generated by an electric discharge in a gas at lower pressure or using microwaves. Typical illustrations for plasma generation at atmospheric pressure include the corona discharge, Dielectric barrier discharges (DBD), Radio-frequency plasmas (RFP) and the gliding arc discharge. In contrast,

thermal plasmas are generated at higher pressures, require high power, and an almost thermal equilibrium exists between the electrons and the heavy species. Plasma generation at atmospheric pressure is of interest, both technically and industrially for the food industries because this does not require extreme conditions.

1.2 Plasma Sources

Formerly, plasma treatments were carried out under vacuum conditions, but researchers have now developed atmospheric pressure plasma system, resulting in reduced cost, increased treatment speed, and industrial applicability (Yoon and Ryu 2007; Yun et al. 2010). The ability to generate non-thermal plasma discharges at atmospheric pressure makes the decontamination process easier and less expensive (Kim et al. 2011). However, until very recently, most of the cold plasma devices available commercially were developed for research and aimed at biomedical applications. Therefore, for food applications, these devices may need to be customized or tailor made. The barrier glow discharge generated between two parallel electrodes is a widely employed NTP system. In a possible industrial scale set-up, food may be conveyed through the discharge to achieve microbial decontamination. Another configuration is the plasma pen or jet, in which a stream of gases can be directed at the object to be treated. *Biozone Scientific* has developed a new process for the generation of cold oxygen plasma (COP) by subjecting air to high- energy deep- UV light with an effective radiation spectrum between 180 nm and 270 nm. This cold gas plasma is composed of several species like negative and positive ions, free radical molecules, electron, UV-photons and ozone (Terrier et al. 2009). *Duo-Plasmaline* is a linearly extended plasma source excited using microwaves of 2.45 GHz at a pressure <1000 Pa (Petasch et al. 1997) and several other plasma treatment systems have evolved based on this principle. The *Plasmodul* is a microwave sustained low pressure plasma reactor with a modular concept based on the *Duo-Plasmaline* principle which provides an easy upscaling for industrial applications (Schulz et al.). This type of microwave excited plasma sources are well suited for large area plasma treatment (Petasch et al. 1997) and can probably be employed for surface treatment of foods or processing surfaces at industrial scale. More recently, Kim et al. (2010) developed a cold plasma jet operating at 20 kHz Alternating Current (AC) under atmospheric pressure. The most versatile feature of most of the plasma systems is the freedom to select a gas or gas mixture. Improvements in the existing plasma systems and newer equipment directed for treatment of real food systems are likely to draw attention of researchers and engineers in near future. Recently a novel approach which shows significant potential for the treatment of various foods has been reported. The approach is based on a dielectric barrier discharge with the food package in contact with high voltage electrodes. Only 40-50 W of power is needed to ionize air inside a 4 L re-sealable plastic (LDPE) bag (Klockow and Keener 2009). The high voltage process ionizes any gas within the electric field contained within the package. Ionization can generate significant amounts of reactive molecules with little increase in product surface temperature.

Specific treatment times for targeted spore or bacterial reductions are dependent on product loading, packaging material, gas composition and package/electrode configuration. The in-package ionization process has been demonstrated in a number of common packaging materials including cardboard, glass, LDPE, HDPE, PETE, polystyrene, rubber, tygon, and others. Scale-up of the system has facilitated treatment of air filled packages with an electrode gap of up to 10 cm with rapid processing times (Keener et al. 2010).

1.3 Action of Plasma on microorganisms

1.3.1 Action on cell components and functions

The use of sterilizing properties of plasma was first introduced towards the end of 60s, patented in 1968 (Menashi 1968) and first works with plasma made from oxygen were proposed in 1989. Thereafter, considerable research has been performed on the mechanism of microbial inactivation by plasma agents. The plasma agents contribute to the lethal action by interacting with the biological material. Nelson and Berger (1989) have shown that O2 plasma could be a very efficient biocidal against bacteria. Plasma treatment can effectively inactivate a wide range of micro-organisms including spores (Kelly-Wintenberg et al. 1999; Feichtinger et al. 2003; Lee et al. 2006) and viruses (Terrier et al. 2009). Effect of plasma can be quite selective, meaning tuneable between damage to pathogenic organisms without damage to the host, or activation of different pathways in different organisms (Dobrynin et al. 2009). Low-pressure oxygen plasma has been shown to degrade lipids, proteins and DNA of cells (Mogul et al. 2003). The reactive species in plasma have been widely associated to the direct oxidative effects on the outer surface of microbial cells. As an example, commonly used oxygen and nitrogen gas plasma are excellent sources of reactive oxygen-based and nitrogen-based species, such as $O\bullet$, O^2, O^3, $OH\bullet$, $NO\bullet$, NO_2 etc.

Atomic oxygen is potentially a very effective sterilizing agent, with a chemical rate constant for oxidation at room temperature of about 106 times that of molecular oxygen (Critzer et al. 2007). These act on the unsaturated fatty acids of the lipid bilayer of the cell membrane, thereby impeding the transport of bio-molecules across it. The double bonds of

unsaturated lipids are particularly vulnerable to ozone attack (Guzel-Seydim et al. 2004). Membrane lipids are assumed to be more significantly affected by the reactive oxygen species (ROS) due to their location along the surface of bacterial cell, which allows them to be bombarded by these strong oxidizing agents (Montie et al. 2002). The proteins of the cells and the spores are equally vulnerable to the action of these species, causing denaturation and cell leakage. Oxidation of amino acids and nucleic acids may also cause changes that result in microbial death or injury (Critzer et al. 2007). Micro-organisms in plasma are exposed to an intense bombardment by the radicals most likely provoking surface lesions that the living cell cannot repair sufficiently faster. This may partially explain the observations wherein cells are in many cases destroyed very quickly. This process is termed "etching" (Pelletier 1992). The cell wall rupture has been additionally attributed by Laroussi et al., (2003) and Mendis et al., (2002) to electrostatic forces due to accumulation of charges at the outer surface of cell membranes. The morphological changes in *E. coli* cells treated with atmospheric plasma at 75W for 2 min as observed under an electron microscope by (Hong et al. 2009), clearly revealed that the treated cells had severe cytoplasmic deformations and leakage of bacterial chromosome. These observations demonstrate the loss of viability of bacterial cells after plasma treatment. An analogy between plasma and pulsed electric field has also been drawn to explain the action of plasma on the membranes (Pothakamury et al. 1995; Spilimbergo et al. 2003). It is well established that electroporation of membranes is induced by pulsed electric fields and it appears that plasma acts on similar lines inducing perforations in the membranes of micro-organisms (Sale and Hamilton 1967; Pothakamury et al. 1995; Wouters and Smelt 1997). In addition to generating pores, humid air plasma additionally provokes a marked acidification of the medium (Moreau et al. 2005; Moreau et al. 2007).

1.3.2 Role of UV photons and charged particles

The production of UV photons of different wavelengths has been proposed to be involved in dimerizing the thymine bases of DNA including that of spores (Munakata et al. 1991). The role of UV photons in bacterial death when they are submitted to a plasma treatment was reviewed in detail by (Boudam et al. 2006). Recently, by exclusion of reactive particles and spectral fractions of UV radiation from access to the spores Roth et al., (2010) revealed that UV-C radiation is the most effective inactivation agent in the plasma. Ultraviolet (UV) photons play a less important role in atmospheric pressure glow discharge (APGD) because they are easily absorbed by gas atoms and molecules at atmospheric pressure (Vleugels et al. 2005). The role of the charged particles in the bacterial inactivation process was recently investigated by Lu *et al.* (2009). Their work revealed that the charged particles play a minor role in the inactivation process when He/N2 (3%) is used as working gas than when He/O2 (3%) is used. Also, they concluded that heat and UV play no or minor roles in the inactivation process. Similar results were earlier obtained by (Perni et al. 2007) who interplayed bacterial inactivation kinetics with optical emission spectroscopy, and identified oxygen atoms as major contributor in plasma inactivation with minor contributions from UV photons, OH radicals, singlet oxygen metastables and nitric oxide. Thus, a contradiction over the role of UV photons in plasma exists and future studies must be directed to get a clear picture.

1.3.3 Effect of process parameters

The concentrations in which the plasma agents occur in plasma depend greatly on the device set-up (reactor geometry), operating conditions (gas pressure, type, flow, frequency and power of plasma excitation) and gas composition which affect their efficacy in a process when employed. To cite an example, the destructive efficiency of various gas plasma sources and temperatures on *Bacillus spp.* spores were compared by (Hury et al. 1998). This group demonstrated that oxygen-based plasma is more efficient than pure argon plasma. Another deciding criterion is whether the substrate to be sterilized is in direct contact with the plasma (*Direct Exposure*) or located remote from it (*Remote Exposure*) (Moisan et al. 2001; Laroussi 2005; Boudam et al. 2006). If exposed remotely, the quantum of heat transmitted to a sample is reduced, the charged particles do not play a role since they recombine before reaching the sample, and many of the short-lived neutral reactive species also do not reach the sample. Since, the components of the plasma are reactive and self-quenching, with a relatively short half-life, decreased time of flight would be expected to be one of the major factors in antimicrobial efficacy in this case (Niemira and Sites 2008). By varying the process parameters involved in plasma generation, a multitude of mechanisms can be actuated which may act individually or synergistically.

Nevertheless, the details of interaction of the different plasma agents with the different components of bacterial cells or spores are currently very limited. The interactions which occur between plasma agents and biological materials, ultimately leading to sterilization are still under investigation.

1.4 How the technology works

Cool plasma produces (gaseous) activated ions, photons, electrons and free radicals, collectively termed 'plasma', that exert their effects at 30-60°C; hence the term 'cool' or 'nonthermal' (see Figure 1). While it is possible to produce cool plasmas at low pressures, atmospheric-pressure plasma are cheaper to operate and are generally more effective. The plasma can be produced in ambient air, or in gases such as oxygen, helium and argon. The choice of gas and the amplitude and frequency of the power source are used to control the density of the plasma species. The plasma system can be configured to a wide range of geometries, depending on the type of food, and can allow continuous processing.

1.5 Benefits and applications

Cool (or nonthermal) plasma has many potential applications for the food industry, including the dry disinfection of food surfaces (meat, poultry, fish, nuts, fruit and vegetables), powders (dried milk components, flour, herbs and spices) and seeds for sprouting (alfalfa and mung beans). The technology can also be used to disinfect surfaces of processing equipment and packaging materials. The gaseous atoms and ions of plasma are effective against bacteria, viruses, moulds and fungi. The plasma can penetrate cracks and crevices unlike other potential surface treatments such as ultraviolet light. Therefore, the technology functions more effectively over uneven or cracked surfaces such as those found on many foods (seeds, meats etc).

Cool plasma has several advantages over competing preservation technologies such as irradiation, chemical sterilization (e.g. ethylene oxide) or disinfection treatments (e.g. chlorine). These can have detrimental sensory effects on treated food products or they are being phased out because of safety concerns. Although cool plasma technology is not yet used commercially on a large scale in the food industry, it has been successfully used for decades in many other industries, and is readily scaleable.

2 MATERIALS AND METHODS

[26]Sliced chicken were purchased from local market (Benha, Qaliobia governorate, Egypt). All samples were transported to our laboratory food irradiation unit, Nuclear Research Center in ice-box (0°C) and surveyed for microbiological counts for counts of total bacteria, psychrophilic bacteria, spore forming bacteria, total molds and yeasts. Then, sliced chicken samples were packed in tightly sealed polyethylene pouches and divided into seven groups and stored in freezing till irradiation treatments.

2.1 Gamma irradiation treatments Four bags from each of sliced chicken were gamma irradiated at 0, 2, 4, and 6 kGy doses using cobalt-60 gamma chamber (1.367 kGy/h) in Cyclotron Project, Nuclear Research Center Atomic Energy Authority, Inshas, Cairo, Egypt. After irradiation, all samples were stored at 4±1°C.

2.2 Plasma treatments *Character of exposure machine*

The plasma generator consisted of a negative dc source, a Blumlein-type pulse-forming network (E-PFN), and a dynamic spark gap switch. A triggered spark gap switch was used as a closing switch of E-PFN. E-PFN had four stages of LC ladder, which were composed of 5 nF of capacitor and 3 μH of inductor. The characteristic impedance ($2\sqrt{L/C}$) and the pulse width ($2N\sqrt{LC}$) of E-PFN, calculated from capacitance (C) and inductance (L) of the LC ladder, and number (N) of LC ladder stages were approximately 49 Ω and 1.0 μs, respectively.

A charging resistance value of 50 kΩ was chosen in the present case which corresponds to a charging RC time constant of 1 ms, which is 40 times faster compared to the repetition rate of the pulse. A schematic of the pulsed atmospheric-pressure plasma jet (PAPPJ) device for generating high voltage pulsed, cold atmospheric plasma jets is shown in Figure 1. The high voltage (HV) wire electrode, which is made of a copper wire, is inserted into a hollow barrel of a syringe. The distance between the tip of the HV electrode and the nozzle is 0.5 cm. When HV pulsed, DC voltage (amplitudes up to 25 kV, repetition rate up to 25 Hz), was applied to the HV electrode and helium gas was injected into the hollow barrel. This device was made using medical syringe (made out of an insulating material cylinder). The gas was fed into the system via flow meter. The applied voltage to and the discharge current through the discharge chamber were measured using a voltage divider (Homemade), which was connected between the two electrodes, and a current monitor, which can be located upon returning to the ground. The signals from the voltage divider and the current monitor were recorded in a digitizing

oscilloscope (Lecroy, USA) with a 200-MHz bandwidth. The high voltage pulses are applied between the needle electrode positioned inside a dielectric cylinder (a simple medical syringe) and a metal ring placed on the exterior of this cylinder. In order to obtain electric discharges at atmospheric pressure, a high voltage pulses (tens of kV) which have limited duration (hundreds of nanoseconds) and are repeated (tens of pulses per second), in addition to an inert gas (argon) is introduced in the cylinder.

The gas flows were in the range 0.5-10 l/min. The discharge takes place between the metallic needle top and a metallic ring fit on the outer surface of the syringe. Under optimal conditions, plasma is emitted as centimeter-long jets, just millimeters in diameter or even smaller. The working gases are supplied by high-pressure cylinders. Gas pressure regulators are used to reduce the pressure of gases to a workable level. Then, gas flow controllers deliver the gases with the desired flow. For voltage amplitudes of 15-18 kV, the plasma jet is very weak. The plasma jet disappears for voltage amplitudes lower than 15 kV. When argon is injected from the gas inlet and high voltage pulses, 26 kV voltages is applied to the electrode, the plasma jet is generated and a plasma plume reaching length of 21 mm is launched through the end of the tube and in the surrounding air. The length of the plasma plume can be adjusted by the gas flow rate and the applied voltage. Three bags from each of sliced chicken were exposured to plasma at 0.5, 1.0 and 1.5 min in Plasma Physics and Nuclear Fusion Department, Nuclear Research Center, Atomic Energy Authority, Inshas, Cairo, Egypt. After the exposure time of plasma, all samples were stored at 4±1°C.

2.3 Gamma irradiation treatments[26]

Four bags from each of sliced chicken were gamma irradiated at 0, 2, 4, and 6 kGy doses using cobalt-60 gamma chamber (1.367 kGy/h) in Cyclotron Project, Nuclear Research Center Atomic Energy Authority, Inshas, Cairo, Egypt. After irradiation, all samples were stored at 4±1°C.

2.4 Microbial analysis[26]

Colony forming units for total bacterial count were counted by plating on plate count agar medium and incubated at 30°C for three to five days (APHA, 1992). Total molds and yeasts were counted on oxytetracycline glucose yeast extract agar medium according to Oxoid, (1998). psychrophilic and spore forming bacteria count according to (FDA, 2002).

2.5 Statistical analysis[26]

The statistical evaluation of the mean data was compared using one-way analysis of variance (ANOVA) according to Zar (1984). The chosen level of significance was P≤ 0.05.

3 RESULT AND DISCUSSION

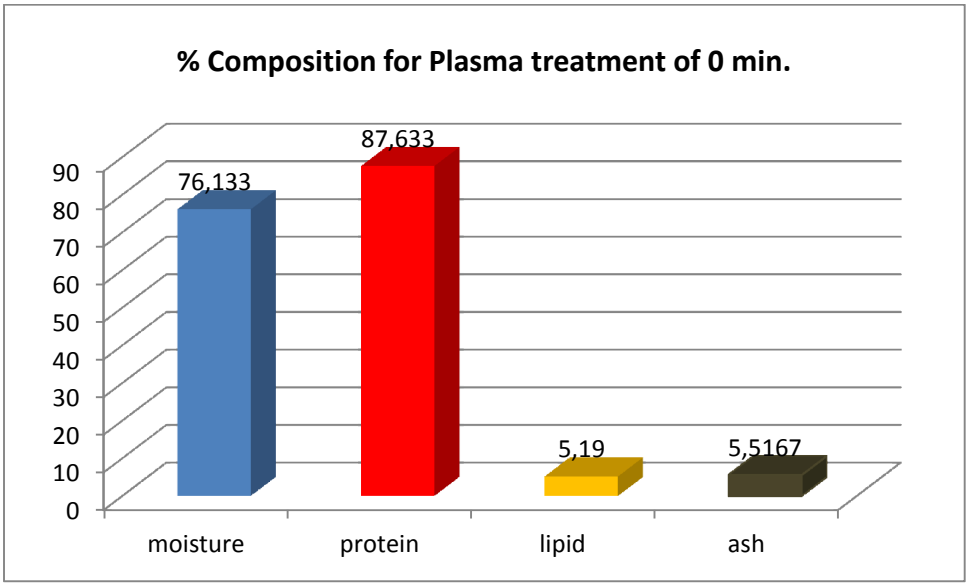

Fig 1. Graph for % composition of sliced chicken treated with CAP for 0 minute.

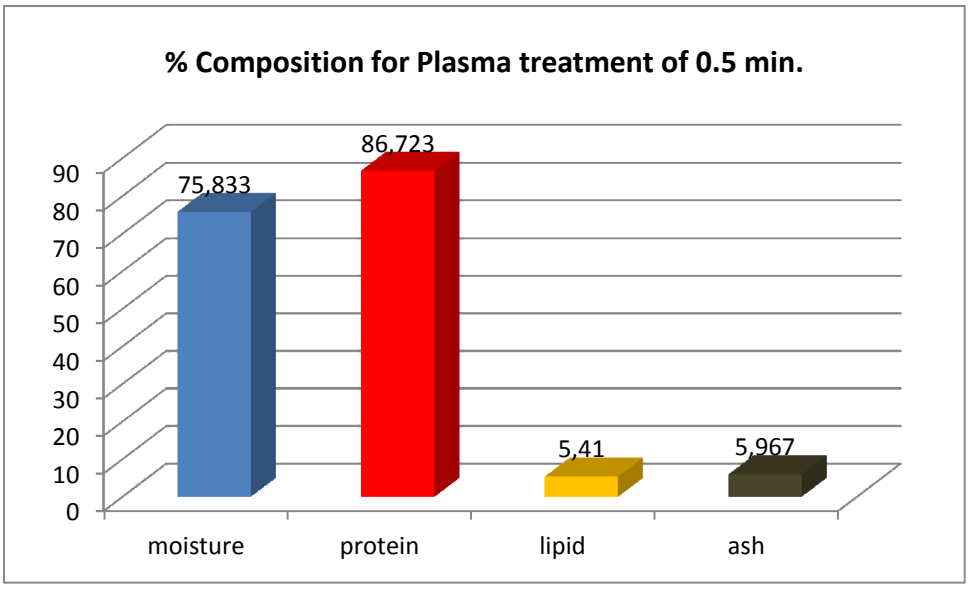

Fig 2. Graph for % composition of sliced chicken treated with CAP for 0.5 minute.

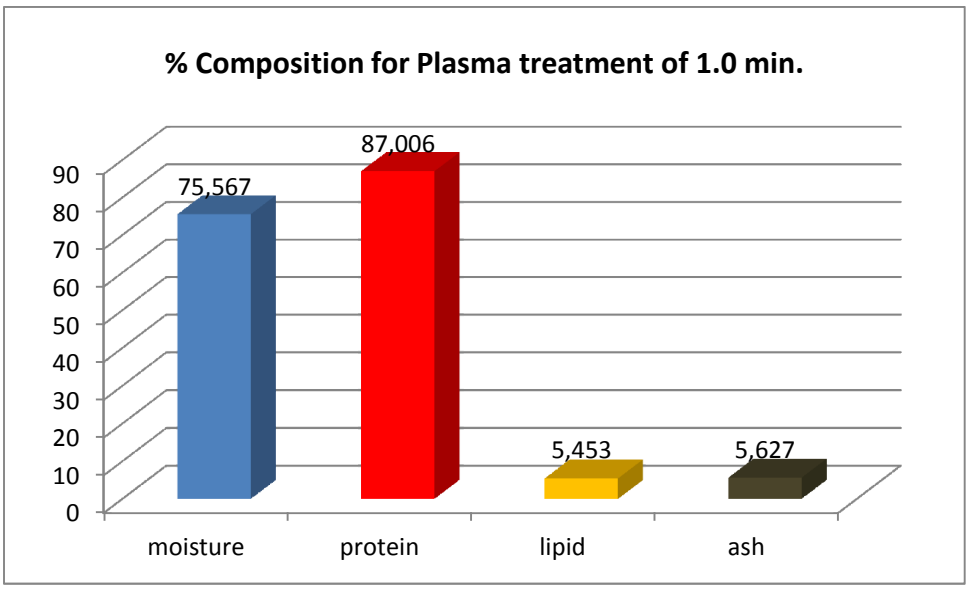

Fig 3. Graph for % composition of sliced chicken treated with CAP for 1.0 minute.

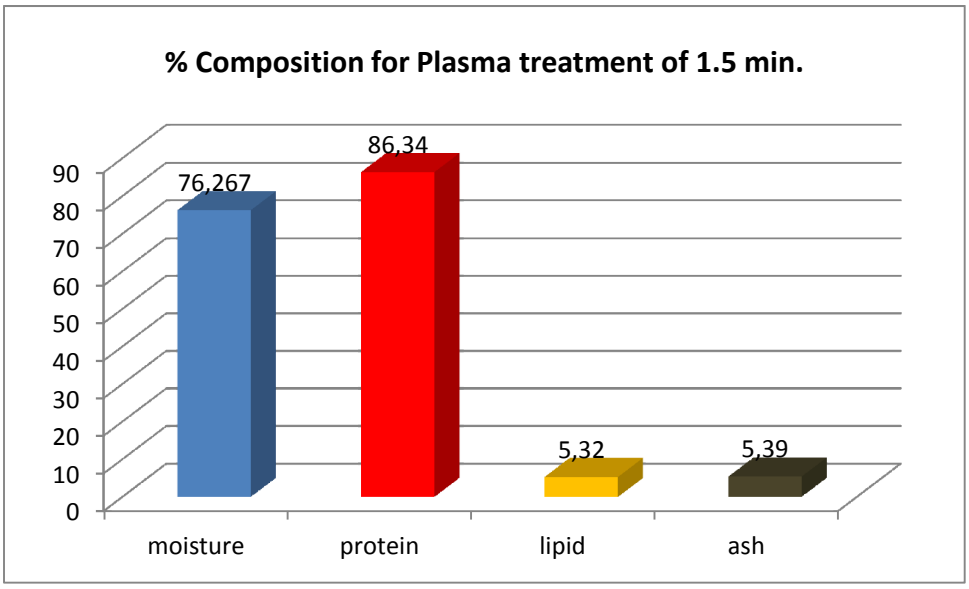

Fig 4. Graph for % composition of sliced chicken treated with CAP for 1.5 minute.

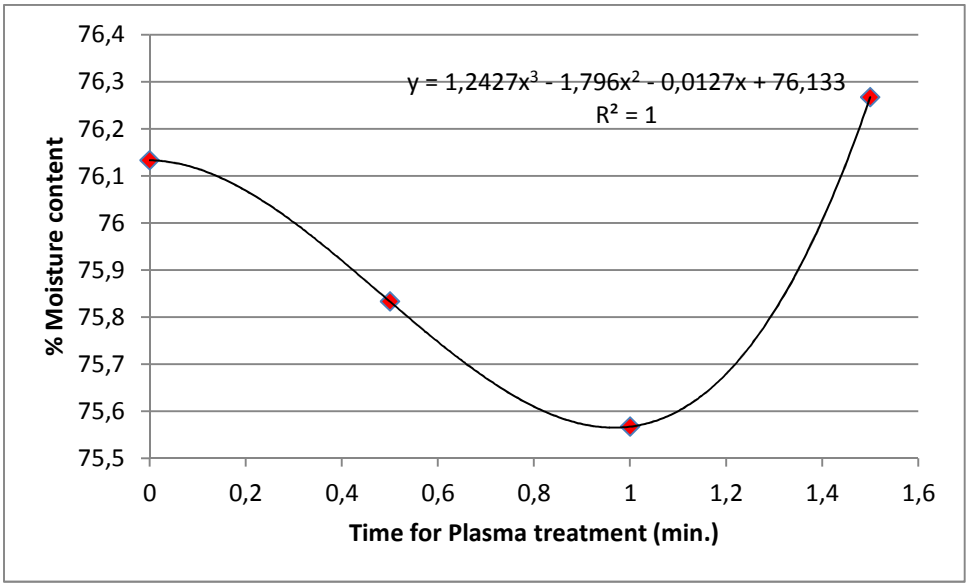

Fig 5. Graph showing the trend line and the modeled equation with R^2 for change in moisture content with the treatment time.

From the above graph one can say that decreases from 0-1 min treatment and then it increases till 1.5 minutes.

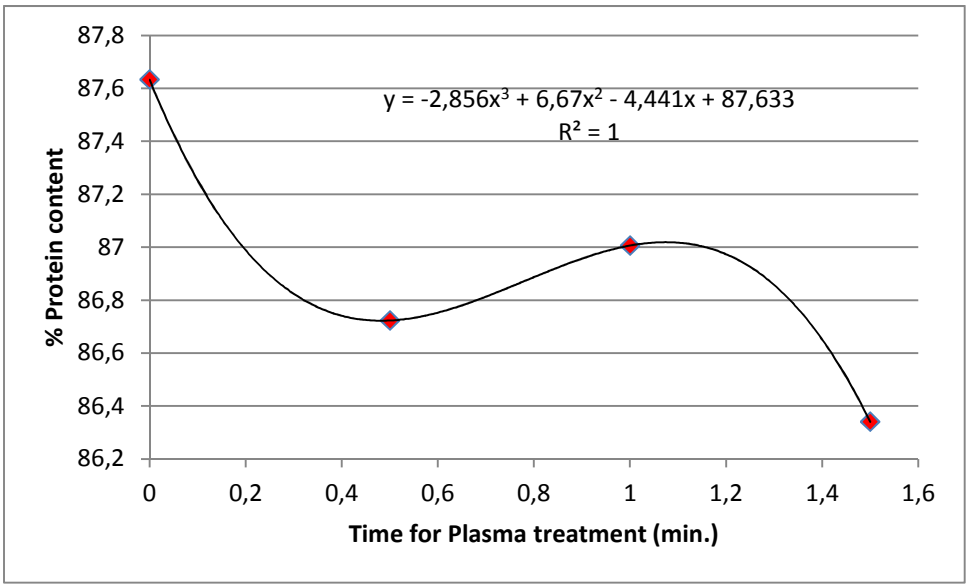

Fig 6. Graph showing the trend line and the modeled equation with R^2 for change in Protein content with the treatment time.

From the above graph one can say that Protein content decreases from 0-0.5 minutes then increases from 0.5-1.1 then decreases from 1.1- 1.5 minutes.

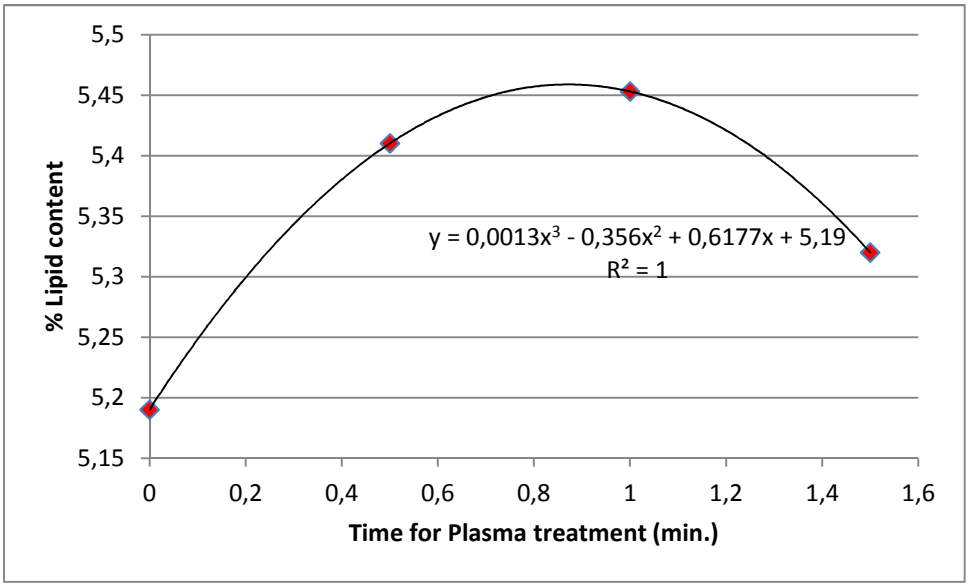

Fig 7. Graph showing the trend line and the modeled equation with R^2 for change in Lipid content with the treatment time.

From the above graph it can be concluded that the Lipid content increases from 0-0.9 minutes and then fall down till 1.5 minutes.

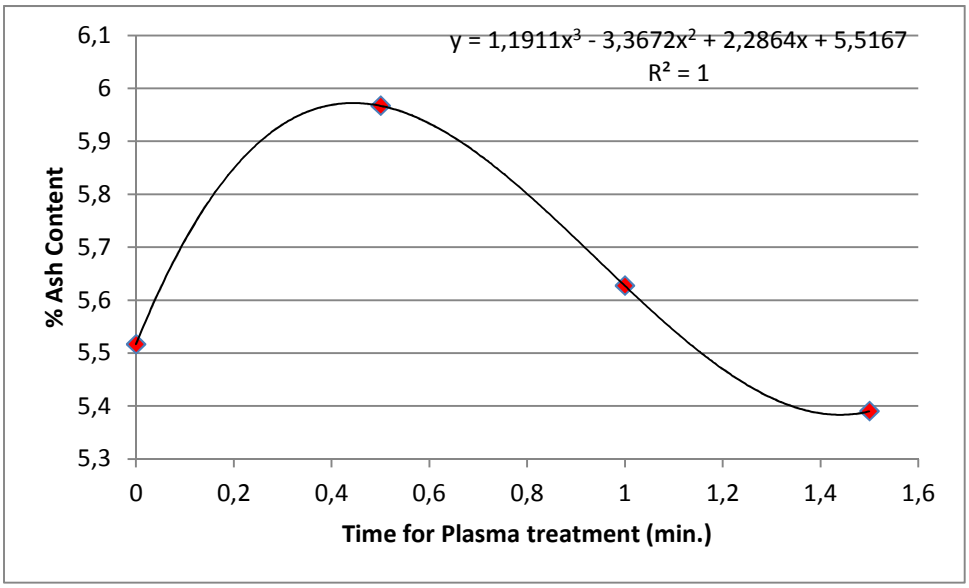

Fig 8. Graph showing the trend line and the modeled equation with R^2 for change in Ash content with the treatment time.

From the above graph it can be concluded that Ash content increases from 0-0.4 minutes of treatment then decreases till 1.5 minutes.

4 CONCLUSION

The effect of Cold Atmospheric Plasma various time interval on the chemical composition of sliced chicken was studied and the data and From above graphs, it could be noticed that the moisture, total protein, lipid and ash contents were tends to change with the CAP treatment of sliced chicken respectively. From the above graphs and model prepared one can make out the changes in compositions of sliced chicken within the plasma treatment of (0-1.5) minutes of the same.

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