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NTP: Non-Thermal Plasma, HP: Hybrid Plasma, ROS: Reactive Oxygen Species

Abbreviations

HP: Hybrid Plasma, NTP: Non-Thermal Plasma, ROS: Reactive Oxygen Species FWM: Free Water Molecules

Research Article

Confined Water for Passive Generation and Accumulation of Non-Thermal Plasma

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Abstract

Introduction: In a recent publication, we described the production method for Non-Thermal Plasma (NTP) without added external energy. Bulk water contains 12-19% free water molecules. Once filtered through glass into a sealed confined jar, free water molecules reacted to produce an new form of NTP, a gas/water combination, Hybrid-Plasma (HP). This report describes a second production method for HP.

Methods: We added 800mL distilled water to a 4000mL acrylic cylinder. Inside the cylinder, an uncovered 500mL collection jar equipped with an ion counter and hygrometer was placed on a platform above the water line. When the acrylic cylinder was sealed with a cover, it formed the confined chamber similar to the water through glass method.

Results: After 24 hours, maximum readings were registered on the ion counter and hygrometer inside the open jar confined in the chamber. The uncovered collection jar was sealed with a plastic cover and placed on a shelf for observation (n=6). Initially, the ion counters (n=6) registered a mean ion count of 1443 and a mean hygrometer reading of 93%; both readings gradually declined until day 6.

Discussion: We produced HP by a second method with the same properties as HP produced by the original water-through-glass method. Analysis of the HP indicated the presence of Hydroxide/OH- and H_2O_2/Hydrogen Peroxide.

Conclusions: We discovered how to separate highly kinetic free water molecules in a confined chamber to produce stable NTP. Hybrid plasma was accumulated, analyzed and used in experiments to determine the effects it has on living systems.

Introduction

The production methods for Non-Thermal Plasma (NTP) have been the same since Langmuir's [1] research a century ago. Today, NTP continues to be produced from electricity introduced to a gas by the dielectric or corona discharge method that excites molecules in a low-level ionization reaction. In a recently published article, we detailed a new method for NTP production without added external energy [2]. The original production method used an underwater sealed glass jar, with pore size (8-12Å) [3] as a molecular filter. The jar also accumulated free water molecules (2.75Å) from distilled water along an hydrostatic/osmotic pressure gradient. Free Water Molecules (FWM) are self-diffusing water molecules that once unencumbered by bulk water do not experience restriction and exhibit high kinetic energy for an ionization reaction. FWM were first calculated in 2013 by NV Penkov [4]. His calculations gave us the insight to hypothesize the mechanism for the ionization reaction and subsequent production of NTP without added energy. Penkov reported that the fraction of free water molecules in bulk water at room temperature ranged from 12% to 19%. Penkov's FWM findings supports our hypothesis that we filtered FWM's through and into the glass jar from distilled bulk water. These small molecules were able to pass through glass pores into the underwater confined vessel by hydrostatic and osmotic pressure. Confined kinetic FWM's reacted with each other and produced Hybrid-Plasma (HP), a new form of NTP. HP, an invisible mixture of water and gas, was determined by experiments resulting in maximum readings on the hygrometers and ion counters inside the sealed jars. This report details another method to produce NTP without added energy. The confined kinetic chamber is an environment in which free water molecules from distilled bulk water diffused into the air-filled confined space above the water line. These kinetic molecules, when liberated into the chamber reacted with other FWM's to produce HP. Analysis with hygrometers and Ion counters confirmed the presence of ionization. Conventionally produced plasma, by the di-electric and corona discharge methods resulted in NTP containing reactive oxygen species (ROS). Similar to conventional NTP, hybrid plasma contains ROS, hydroxide and hydrogen peroxide. Conventionally produced NTP exists on a transient basis making it difficult to harness, apply and determine its effects. HP is produced by ongoing confined molecular action so the effects from external energy (electricity) are removed from the experiment's results.

Methods

Figure 1A: Components of confined chamber

Figure 1B: Collection jar inside covered chamber

Figure 1C: Covered jar removed from chamber.

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A 4500ml clear, acrylic cylinder container with a tight-fitting cover served as the confined kinetic chamber. We added 800ml of distilled water to the bottom of the chamber. An uncovered 500ml jar (Ball canning jar) served as the collection vessel, it was outfitted with a mini-ion counter and mini-hygrometer was staged above the water line. The chamber was sealed with a tightfitting cover (Figure 1A, 1B).

After 24 hours, the instruments readings in the collection jar indicated that a reaction had occurred. The chamber cover was removed and a plastic cover placed on the collection jar. The collection jars (n=6) were then placed on a remote shelf (Figure 1C) for recording ionization levels and absolute humidity for the duration of the experiment.

Sampling

Samples were taken for analysis using the experimental components seen in (Figure 2A). For the confined kinetic chamber production method, uncovered glass headspace vials (n=4) were placed inside a 200ml beaker (Figure 2B). The beaker with the vials was placed into the confined kinetic chamber for 24-48 hours. When instruments readings indicated an ionization reaction, each sample vial was sealed using a crimping tool. (Figure 2B) shows the confined chamber HP sample collection method, which was labelled BR. Similar sampling vials (n=4) were prepared using the water through glass method to sample and compare with the confined chamber method and were labeled UW (Figure 2C). A third group of sealed vials prepared in the ambient environment served as a control, labeled Ctrl.

Figure 2A: Sampling Components **Figure 2B:** Sample vials inside covered chamber **Figure 2C:** Sample vials underwater method.

Results

After 24 hours, the ion count (IC = 104 ions/cm^3) in the collection jars (n=6) showed a mean IC=1443 and a mean hygrometer reading of 93%. Over six days of observation, the instrumented jars registered a progressive decrease in the ion counts from mean 1443 to 74. Hygrometer readings declined similarly falling from a mean of 93% to 56% on day 6. (Figure 3) graphically presents the data collected for IC (3A) and humidity (3B) over six days.

Analysis of Hybrid Plasma

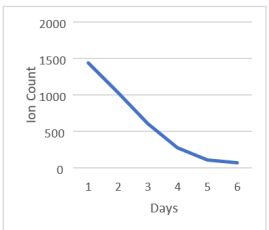

Figure 3A: Average ion counts over 6 days.

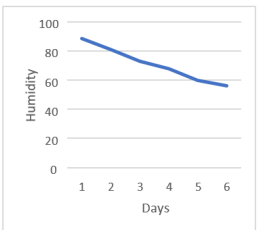

Figure 3B: Average humidity level over 6 days.

Spectral Analysis of Hybrid Plasma sample vials from both production methods indicated the confined reaction produced the ROS OH-/Hydroxide. Each sample was analyzed by means of headspace (GC-MS) in negative ion mode. The headspace sampler oven temperature was held at 45°C, the loop was held at 55°C, and the transfer line was held at 60°C. Vial equilibration time was 5 minutes and injection duration was 0.5 minutes. The gas chromatograph oven was held at 110°C and helium flowed at a rate of 1.1 ml/min. The inlet temperature was operated in the split less mode at 250°C. The MS transfer line was held at 250°C and the ion source and quadrupole temperatures were at 150°C. Negative ion scanning at m/z 17 revealed the presence of hydroxide ions in each sample (Figure 4).

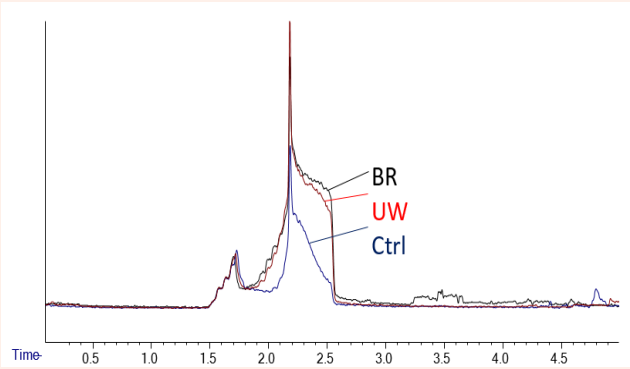

Figure 4: The response for m/z 17, representing hydroxide ions in each sample was measured in triplicate. Peak areas for BR, UW, and Ctrl were compared. The blank control sample (Ctrl) had a peak area of 16.1. The peak areas for the BR and UW samples were roughly twice that of the control, with values of 32.0 and 32.6, respectively. An overlay of the three sample groups shows their relative peak responses.

Testing for additional ROS indicated the presence of H_2O_2. We used a hydrogen peroxide test strip and found hydrogen peroxide at 50ppm in the HP sample. HP is a hybrid of gas and water so the test strip was able to register and exhibit color change after 24 hours in the gasous atmosphere inside the confined space.

Discussion

This study describes a second method for the production of a new form of non-thermal plasma without added external energy. Both production methods, water through glass and the confined kinetic chamber described in this report, used the kinetic energy from FWM reaction to achieve activation energy and ionization. An ionization energy of 2.18 × 10-18 joule (13.6 electron volts) is required to force the electron from its lowest energy level entirely out of the atom [5]. This passively occurring ionization phenomena has been observed by past scientific efforts. The auto-ionization of water is the most documented reaction that does not require external energy. Spray electrification first described by Lenard (1892), is another way to produce ionization without added electricity. Recently a team of researchers [6] demonstrated that a strong electric field generated at the interface of water and air in a microdroplett, seems to activate water molecules, forming various reactive oxygen species. These species are unstable molecular fragments that can quickly react with other molecules to yield hydrogen peroxide. Similarly, analysis of HP produced without external electric input contained elevated levels of reactive oxygen species H_2O_2 and OH-/Hydroxide. Both of our methods used to produce HP required the confinement of the molecular reaction for HP production. The natural kinetics of water molecules are known to increase by confinement. Studies have shown that confinement leads to amplified reactivity's in bimolecular reactions, stabilization of otherwise reactive species, and limitation in motions that create new stereo chemical arrangements [7]. In our experiments, the increased reaction rate can be attributed to confinement of kinetic FWM that resulted in ionized gas, stable NTP containing ROS. HP as a collective ongoing reaction explains how free water molecules, once confined in the jars, did not escape. If a collection jar was opened to exchange instruments, the ionization levels dropped slightly then rebounded after the cover was replaced. FWM's continued to fuel the ionization reaction that decreased as the HP exhausted its fuel in the course of the experiment. This was determined by the progressive decrease in the average ionized readings, from 1443 to 74 (X 10^3 ion/cm^3) and average humidity levels from 93% to 56% over a six-day period. These findings support the hypothesis that confined FWM were the natural energy source for production of stable relatively long-lived HP containing reactive oxygen species.

Limitations

Ion levels will vary inside the confined chamber. Each collection jar had different rates of accumulation. If the cover of the chamber was removed for very short intervals, the ion levels dropped by up to 80%. Ion levels rebounded rapidly back to previous levels when the chamber cover was back in place. The acrylic container acting as the chamber must have a tight-locking cover to properly confine the reaction. For best results, we allowed the ionization count to begin before placing the collection jars on the stage inside

the chamber. Metal covers on 500ml jars inhibited plasma production; plastic covers must be used. Magnetic adhesion is likely the cause for inhibited HP production due to magnetic nature of water molecules. The ion counters are cleaned before use. Batteries in the ion counters are replaced at the first low-battery indicator or signs of malfunction. Before experiments, ion counters were calibrated to zero, according to instructions. The water in the chamber loses its potency over multiple uses, reducing the chamber's ability to produce HP with a higher ion count. Distilled water must be changed periodically in the chamber to maintain its ability to produce strong non-thermal HP. Larger 19-qt and 32-qt storage containers with gasket locking covers also served as chambers, they allowed for multiple collection jars at the same time. The smaller acrylic cylinder reactors were better suited for these experiments by producing a reaction at a faster rate one jar at a time in a smaller confined area.

Future applications

Due to the stable nature of HP and stable ROS, experiments using HP for biological and chemical determinations can be performed for extended periods without the effects of electricity. Others have identified non-thermal plasma and its hydroxide content as a possible treatment to fight coronaviruses [8]. There are many on-going studies about the role ROS plays in cellular function regarding health and longevity. Studies suggest that modulation of ROS at the cellular level will eliminate cellular dysfunction and disease [9]. HP is a novel NTP with a shelf life that could, in the future, be used to modulate positive ROS at the cellular level to improve health and longevity in humans [10].

Conclusions

This report describes a second production method for HP from confined water. We added 800ml distilled water to a 4000ml acrylic cylinder. Inside the cylinder an uncovered 500ml collection jar equipped with an ion counter and hygrometer was placed on a platform above the water line. When the acrylic cylinder was sealed with a cover it created a confined kinetic chamber. After 24 hours, maximum readings were registered on the ion counter and hygrometer inside the open jar confined in chamber. The uncovered collection jar was sealed with a plastic cover and placed on a shelf for observation (n=6). Initially, the ion counters (n=6) registered a mean ion count of 1443 and a mean hygrometer reading of 93%; both readings gradually declined until day 6. We produced HP by a second method with the same properties as HP produced by the original water-through-glass method. Analysis of the HP indicated the presence of Hydroxide/OH- and H_2O_2/Hydrogen Peroxide. We discovered how to separate highly kinetic free water molecules in a confined chamber to produce stable NTP. Hybrid plasma was accumulated, analyzed and can be used in experiments to determine its effects on living systems. Studies suggest NTP can be used to modulate ROS at the cellular level. On-going experiments indicate HP has potential ROS modulating effects at the cellular level [11].

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