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Water Dissolution of Gas-Phase Products Generated from Gliding Arc Discharge: Application to Growth of Bean Sprouts

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ABSTRACT

Using "plasma activated water" for plant growth is both lowcost and eco-friendly; this method is a way to replace existing agricultural technology. Air plasma created with an alternating current gliding arc discharge produces Nitrogen oxide (NOx) gas during discharge. The NOx gas enters the water, where it is converted into Nitrate, Peroxide, and other compounds. We named this compound-rich water "plasma activated water." We measured pH, electrical conductivity, and the concentrations of nitrate and peroxide in plasma-activated water and surface roughness of bean sprout seeds treated with Peroxide. In addition, we tested plasma-activated water as a nitrogen-fixed water for growing plants by cultivating bean sprouts. The pH of tap water was seven and its conductivity about 12 µS·cm⁻¹, but plasma activated water had a pH of three and conductivity of 17 μ S·cm⁻¹. The length and weight of the bean sprouts grown in plasma-activated water were 1.5 and 1.2 times those of the bean sprouts grown in tap water. The rootlets of the bean sprouts grown in tap water had more branches than those grown in plasma-activated water. The surface roughness of bean sprouts grown in peroxide was greater than that of bean sprouts grown in tap water. These results show that plasma activated water can enhance the growth of plants.

INTRODUCTION

In the 21st century, we face a food crisis caused by population increase, climate change, lack of agriculture, and other factors. Currently, just over one in every nine people worldwide are unable to consume enough food to conduct an active and healthy life¹. There are at least four key pathways presented by scholars as potential ways to solve the global food crisis. We focus on Pathway¹, technology for production, for study. In other words, we study the use of novel agricultural technology to increase plant production².

There are many methods to increase plant production, and here we focused on increasing the rate or total amount of growth in the same length of time. The most important factor in plant growth is nitrogen (N). Synthetic N fertilizer has played a role in increasing plant production to feed 40% of the world's population since the Harber-Bosch process was invented in the 20th century³. However, using N fertilizer causes rising levels of N₂O, a long-lasting greenhouse gas that significantly contributes to stratospheric ozone depletion and global climate change⁴⁻⁵. In addition, the Harber-Bosch process has disadvantages such as highenergy consumption. Another important factor in plant growth is H₂O₂ (Hydrogen peroxide, hereafter peroxide). Numerous recent studies have shown that reactive oxygen species (ROS), including peroxide, play a key signaling role in the achievement of major events of seed life such as germination or dormancy release. In fact, peroxide, nitric oxide, hydroxyl radicals, and superoxide radicals have been shown to accumulate during seed germination in various species. Nitrate is the most important source of mineral N for plants growing in aerobic soils⁶. Peroxide plays a crucial role as a signaling molecule in various physiological processes, including photosynthesis, respiration, and transpiration because peroxide is the most stable ROS. Thus, an increase in peroxide and therefore these physiological processes will lead to increases in crop yield and productivity⁷. Barba-Espin proposed that H₂O₂ could act as a signaling molecule at the beginning of seed germination and involve specific changes at proteomic, transcriptomic, and hormonal levels⁸. Nevertheless, there are many reports showing that the molecular mechanisms involved in this differential response remain unclear.

The purpose of our research was to use plasma to produce water-dissolved reactive oxygen and nitrogen species (RONS) to enhance plant growth. This is a lower cost, higher efficiency, and more eco-friendly method than traditional N fertilizers.

Using plasma-processing methods to accelerate seed germination and crop growth has attracted much attention. Some plasma processing methods possess many advantages such as low-temperature treatment and short processing time⁹. Most of the researcher who studies seed germination using plasma are focused on 'Seed pretreatment using cold plasma' and who study 'gliding arc plasma' apply it to decomposition of harmful organic compounds like CO₂, PFCs, Tar and so on¹⁰. However the other way, we selected gliding arc discharge (GAD) methods for our purpose, agricultural, our study is not 'pretreatment', 'cold plasma' and 'decomposition'. GAD plasma integrates non-thermal plasma and thermal plasma advantages and is more effective at lower current intensity, higher electron density, and higher injection flow rate than other types of non-thermal plasma. In addition, GAD can directly react with any gas or vapor accepting their temperature and pressure, for that reason nitrogen dioxide and peroxide are formed by dry-air GAD¹¹⁻¹². That reasons why our study selected GAD among plasma discharge methods.

RONS generated by the plasma can be used to induce desirable changes in a broad spectrum of developmental and physiological processes in plants, including increasing resistance to stress and diseases, modifying seed coat structures, increasing the permeability of seed coats, and stimulating seed germination and seedling growth¹³. There is evidence that RONS are generated by plasma. Reactions that occur include OH is formed through electron impact dissociation (reaction 1), dissociative excitation transfers to H₂O from N₂(A) (2), photodissociation of water (3), a charge exchange of incident positive ions on the liquid surface (4), two hydroxyl radicals react to form H₂O₂ in the gas phase (5), and H₂O₂ is formed through the reaction of two aqueous hydroxyl radicals (6). The most important pathway for OH is Formation via liquid phase reactions is the formation of peroxynitrous acid is unstable and decomposes through reactions (8) and (9), resulting in the formation of OH⁻¹⁴.

$$e^{-} + H_2O \rightarrow OH^{-} + H^{-} + e^{-}$$
 (1)

$$N_2(A) + H_2O \rightarrow OH^{\cdot} + H^{\cdot} + N_2(X)$$
(2)

$$hv + H_2O_{aq} \rightarrow H_{aq}^{\cdot} + OH_{aq}^{\cdot}$$
 (3)

$$2H_2O_{aq} + M^+ \rightarrow M + H_3O^+_{aq} + OH^-_{aq}$$
(4)

$$OH' + OH' + M \rightarrow H_2O_2 + M$$
(5)

$$OH_{aq}^{\cdot} + OH_{aq}^{\cdot} \rightarrow H_2O_{2aq}$$
(6)

$$NO_2^- + H_2O_2 + H^+ \rightarrow O = NOOH + H_2O$$
(7)

$$O = NOOH \rightarrow NO_2^{\circ} + OH^{\circ} \sim 30\%$$
(8)

$$O = NOOH \rightarrow HNO_3 \rightarrow NO_3^- + H^+ + \sim 70\%$$
 (9)

In this study, we injected air discharged by a gliding arc plasma into ordinary tap water. We expected that RONS would be generated in the water. We named this water "plasma activated water" (PAW).

MATERIALS AND METHODS

Gliding arc discharge plasma was used to treat tap water, which was then used to grow to bean sprouts. We selected bean sprouts because they grow quickly and are less affected by the external environment than are other plants. We grouped PAW treatments according to the plasma discharge time.

Plasma system

Schematic of the experimental equipment setup is shown in Fig. 1. The plasma system used in this experiment was an alternating current GAD. A cone-shaped stainless steel electrode was placed inside a circular cylinder electrode as shown in Fig. 1A. Air, as a plasma-forming gas, was injected through four tangential inlets at the cylindrical electrode to create vortex flows in the space formed between two electrodes. High voltage from a transformer was applied to the electrodes and control of the applied voltage was accomplished by regulating the primary voltage of the transformer using a voltage adjustor (SLIDAC, Daelimec, Korea). We measured the discharge voltage and current waveforms using a digital oscilloscope (Tektronix DPO4054B, 2.5 GHz; Tektronix, Beaverton, OR, USA), a high voltage probe

(Tektronix P6015A), and a current probe (Tektronix TCP 303). Fig. 2. Shows typical waveforms of alternating current GAD.



Figure 1. (A) Schematic of the plasma discharge process for making plasma activated water. (B, C) the electrode in discharge device and (D) Image of gliding arc discharge.



Figure 2. Voltage and current waveforms of gliding arc discharge

In the rotating gliding arc reactor shown in Fig. 1B, a formed arc is initially elongated by the energetic effect of swirling flows. When the arc voltage reaches a peak, a breakdown occurs in the space between two electrodes, creating a new current path and a new arc attachment, which in turn results in a sudden drop of the arc voltage. The position of the restrike breakdown on the electrodes is related to the potential distribution in the space formed by the two electrodes, which strongly depends on the ionization conditions, thickness, and temperature of the gaseous space. Fig. 1D. Shows an image of the GAD during operation, creating a rotating gliding plasma at an airflow rate of 15 L·min⁻¹.

PAW analysis

Our study analyzed several chemical properties of water after plasma treatment: pH, electrical conductivity (EC), and concentrations of nitrate (NO₃⁻) and peroxide. We measured pH and EC using a pH and EC meter (LAQUA, Horiba, Japan). The meter was dipped into a sample of treated water and allowed to rest for the recommended amount of time. We measured the generation of nitrate and nitrate at different flow rates before the air was dissolved in the water. The concentrations of nitrate and peroxide were measured using test strips (Vacu-vials, K-6933, K-5543, CHEMetrics, Midland, VA, USA). The test strips were analyzed by color indication. We produced new PAW every day because the stability of peroxide and nitrate in water stored in plastic containers is poor¹⁵. We analyzed the chemical properties of the water on five (1st, 3rd, 5th, 7th. 9th) of the 10 days of the experiment and reported the minimum, average, and maximum values.

Bean Sprouts

Bean sprout seeds (Glycine max, 50g) were placed in a plastic container with a lid (Fig. 1A.). There were six replicates per plasma discharge treatment time: control (untreated tap water) and PAW with 1, 3, 5, 10, and 15 min discharge time. All bean sprouts were given the appropriate tap water or PAW every day, water at intervals of about four hours. The temperature of the room was 24°C and light were blocked by the container lid. Bean sprout samples were measured and weighed after 5, 7, and 10 days. After 10 days, we measured length, weight, and length of rootlets and took photographs of the bean sprouts. Reported data represent the maximum, average, and minimum length of 10 bean sprouts and weight of 30 bean sprouts per treatment group.

Finally, we measured the roughness of the bean sprouts' surfaces. There are many different roughness parameters in use, but R_a is the most common. We watered the bean sprouts with water containing 3-ppm peroxide for three days and then measured the roughness of the surface of the seeds using a 3D optical surface profiler (NV-2400, Nano System). This was to determine the effects of peroxide on the seed surface, as surface roughness could influence germination.

RESULTS AND DISCUSSION



The concentrations of nitrogen oxide and nitrite decreased when the flow rate increased (Fig. 3.).

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Although the total concentration is highest (about 3500 ppm) at 2 L·min⁻¹, we selected 10 L·min⁻¹ because the faster flow rate helps the water to circulate, which allows ions to mix more easily. This result shows that nitrogen compounds are generated through GAD. In Fig. 3. The average total concentration of nitrate and nitrate is about 600 ppm. However, the graph is shown in Fig. 4. Indicates that the concentration of nitrate in water treated for 15 minutes is about 97 ppm, so we know that approximately of the 16% nitrite and nitrate were retained in the water. It is because the solubility of nitrite is 0.0098 g/100 mL and nitrate is hydrolysis, but we selected flow rate condition 101 ppm, the effect of nitrate solubility is insignificant. It is caused by the influence of nitrite with low solubility, the water retained nitrogen oxide only 16 %.



Figure 4. (A) Nitrate and (B) peroxide of plasma activated water

PAW analysis

Decreased the pH with an increase in plasma discharge time (Fig. 5A.). Tap water had an average pH of 7.34, while PAW treated for 15 minutes had an average pH of 4.37. This result indicates that some acidic compounds are formed in the plasma system. We believe that these the acid compounds include nitrate¹⁴. Electrical conductivity is increased with treatment time; this is shown in Fig. 5B. The average Electrical conductivity was 12.49 μ S·cm⁻¹ in tap water and 15.54 μ S·cm⁻¹ in PAW treated for 15 min. This is because more air is ionized over time, so more ionization compounds dissolve in the water.



Figure 5. (A) pH and (B) electrical conductivity of plasma activated water

Fig. 4A. Shows that more nitrate is generated with a longer treatment time. Tap water has no nitrate, but the concentration of nitrate increased to 105 ppm with 15 min of treatment. This indicates that nitrate was formed in water through plasma discharge, and therefore we expected that PAW would affect plant growth.

As above, the concentration of peroxide increased with treatment time (Fig. 4B.). The concentration of peroxide in tap water is 0 ppm. However, after 15 min of plasma discharge, the concentration of peroxide increased to 3.45 ppm. The pH and Electrical conductivity results indicate that discharging air with gliding arc plasma generates compounds that help plant growth.

However, in agriculture, the excessive concentration of nitrate and peroxide is the disadvantage of plant growth, pH is also a cause, our study able to identify it through the bean sprouts results.

Bean sprouts results

The graph in Fig. 6. Shows the changes in bean sprout length and weight with increasing treatment time. Both measurements were greatest with 1 minute of treatment time, with an average length of about 22 cm and weight of about 32 g, respectively 1.5 and 1.2 times those of the bean sprouts grown in tap water.

The decreasing of length after 1 minute of treatment time is due to an oversupply of N. This is because when plants receive too much N, they become more attractive to insects and diseases. Excess N can also cause excessive growth and reduce the strength of the stems¹⁶, then ultimately lead to poor growth of the plants. N increases chlorophyll production by creating larger leaf structures to allow for a bigger surface area for the photosynthesizing pigment. Having too much N can cause fast and excessive foliage growth because energy originally intended for the growth of reproductive organs and roots may be re-directed toward leaf growth. The longer the plasma treatment time, the lower the pH, which can also affect the growth rate. Electrolyzed acidic water shortens the length of bean sprouts¹⁷.

The difference in weight seems to be a lot of the data of graph, but the difference in the actual values is not large. However, it has the highest value at 1 minute, which is considered to be an appropriate nutrient in our study.

This study compared images of the image of the 1-minute-treatment and control groups. Fig.7. Is a comparison of the total length of the bean sprouts and Fig. 8. is a comparison of the length of the rootlets. In Fig. 7. We can see that treated bean sprouts are longer than control bean sprouts on days 5, 7, and 10. To make it easier to see the differences, we showed the black arrows that indicate the difference in the length of bean sprouts.



Figure 6. Bean sprout (A) length and (B) weight on the 10th day of growth.

In addition, the density of treated rootlets is lower than that of the control, as seen in Fig. 8. It can be seen that the roots of bean sprouts in PAW are about twice as abundant as the roots of

bean sprouts in tap water. This shows that PAW has more nutrients for plants, so it seems that plants do not need to make as many rootlets to absorb nutrients.



Figure 7. Morphology of bean sprouts grown in tap water or plasma activated water with plasma discharged for 1 min. (A) 5 days, (B) 7 days, (C) 10 days. Black arrows indicate average length.



Figure 8. Morphology of rootlets of bean sprouts grown in (A) tap water and (B) treated plasma activated water 1min.

The differences in bean sprout surfaces when the bean sprouts were treated with either tap water or a peroxide solution. The larger the color variation, the greater the degree of roughness. In this case, more red dots show that the surface is rougher.

In Fig. 9. The R_a value of roughness of seeds grown in peroxide for 3 days is 0.98 µm, which is higher than the roughness of bean sprouts grown in tap water. Therefore, we infer that the seeds grown in peroxide are stressed by exogenous H_2O_2 , as even with the naked eye one can see that the external stress applied to the seeds caused the surface to wrinkle and shrink.



Figure 9. The roughness of bean sprout surfaces after 3 days of treatment with (A) tap water (B) H₂O₂ condition (3 ppm).

In many studies, seeds grown in peroxide had enhanced stress tolerance during post-priming germination¹⁸. It is speculated that H_2O_2 treatments that involve seed soaking or the use of foliar spray induce low levels of oxidative stress and that ROS induce the accumulation of latent defense proteins, resulting in the generation of primed states and in improved stress responses¹⁹.

CONCLUSIONS



In this study, we investigated the growth of bean sprouts grown in water treated with gliding arc plasma discharge. Analysis showed that the nitrogen and peroxide generated by air plasma played a critical role in the growing process.

Our study concludes that the water dissolution produced by GAD helped the bean sprouts grow. Increasing the concentrations of nitrate and nitrite helps plant growth, but if the N concentration is excessive, the results did not help plant growth. We concluded that the concentration of nitrate and the pH after 1 minute of treatment was best in our experiments. Before it can be used as a fertilizer applied to actual cultivation, further research on the plasma treatment time and subsequent nutrient development in crops are needed²⁰.

The effects of hydrogen peroxide on seed germination have been illustrated in numerous studies; however, the exact mechanism of action remains unknown. Therefore, further studies should be conducted to clarify the causal relationship.

This "seed priming" is more attractive than chemical treatments applied to plants in field conditions¹⁸. In our study, we did not focus on priming seeds; our method is an alternate pretreatment using a simple plasma discharge, which is also a way of providing more nutrients to plants without added fertilizer.

Plasma treatment could improve germination, promote growth, and increase the physiological level (Overall physiological health) of plants, leading to increased yield²¹. Our study shows the feasibility and advantages of applying GAD plasma on plant growth and in producing nitrogen-fixed water. It is low cost, high efficiency, and a more eco-friendly method than traditional N fertilizer. However, the treatment time and the concentrations of nutrients should be controlled, to an extent depending on the plants involved, and further research on the mechanisms and components of GAD plasma treatment that were not included in this study should be performed. We expect these results to help address or prevent the "global food crisis."

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