

# Challenges in the enhancement of seed germination by non-thermal plasmas

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**Abstract:** A review of several research works has been done and the principal properties of non-thermal plasma technologies applied to enhance the seed germination are here also depicted. The importance of hydroxyl radicals ( $\bullet\text{OH}$ ) to accelerate dormancy of seeds and growth of radicles is also described such as the influence of superoxide ( $\text{O}_2\bullet^-$ ) to increase the growth of the plant. Preliminary results obtained from the interaction of a double dielectric barrier discharge on endangered Mexican native Zea Maize are presented as well.

**Keywords:** cold plasma, seed germination, hydrophilic properties, 2DBD.

## 1. Introduction

To improve crop production innovative methods and techniques have to be to guarantee the food demand that increases according to the growth of the population (i.e. 10 billion inhabitants in Earth by 2050). Additionally, climate change, scarcity of water, toxicity and deficiency of minerals in soils induces stress conditions on seeds inhibiting germination and normal growth of plants. Our interest to study a native Zea Maize, in particular a carrot yellow race, is because it is an endangered species, it is being replaced by hybrid maize [1].

Non-thermal technologies have been widely used since numerous years ago and represent a solution to meet the food-product safety while diminishing non desired effects. For example UV light has been used since 1900s as germicide [2].

Recent technologies as ozone generators have successfully applied as antimicrobial agent and to preserve fruits, vegetables and grains [3,4].

Concerning non-thermal plasma techniques they also could be seen as a possible solution to enhance the growth of seeds of different plants species; this technique is quite attractive because some inherent properties of plasma processes like activation of very active radicals (e.g.  $\bullet\text{OH}$ ,  $\text{O}_2\bullet^-$ ) and chemical species ( $\text{O}_3$ ,  $\text{H}_2\text{O}_2$ ) at low temperature (i.e. near ambient temperature).

The principal properties of non-thermal plasmas applied in the enhancement of seed germination are depicted in Fig.1.

Cold plasma has been studied to decontaminate seeds; encouraging results have been shown by exterminating *E. Coli*, spores of *B. cereus*, *G. stearothermophilus* and *B. atrophaeus*, and even little insects such as aphids [5,6].

Concerning the enhancement of seed germination, non-thermal plasma techniques also presents good results [7-

9]; generally the germination rate and the yield of germinations seeds have been enhanced with these techniques.

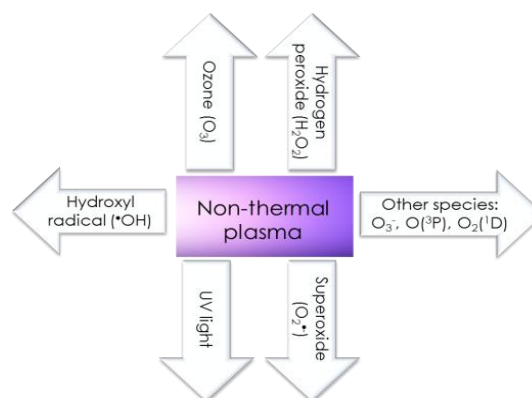


Fig 1. Non-thermal plasmas are an interesting option to enhance seeds' growth, they combine several processes such as ozonification and UV lightning, both used to remove pest and mycotoxins. Especially hydroxyl radical is characteristic to enhance germination.

And finally, the influence of cold plasmas on plants growing is also promising; as an example an investigation of a dielectric barrier discharge on radish seeds showed a growth enhancement of sprouts more than 3 times [10].

### 1.1 Importance of Reactive Oxygen Species.

Reactive oxygen species (ROS), such as  $\text{H}_2\text{O}_2$ ,  $\bullet\text{OH}$  and  $\text{O}_2\bullet^-$ , are well known as regulators of immunity and growth of plants [11], but in particular the role of radical  $\bullet\text{OH}$  is really amazing, because it is the most potent yet short lived ( $10^{-9}$  s) of the ROS [12].

It has been found that  $\bullet\text{OH}$  breaks dormancy of seed reducing the after-ripening, especially if this radical is present in the endosperm cap, because it allows making

the cap weaker and the radicle easily punctures the endosperm cap [13].

Furthermore, the influence of  $\bullet\text{OH}$  on plants elongation and even in the breakdown of plants to recycle into soil organic matter, has been proved [14,15]. Radicals  $\text{O}_2^-$  also play a strong role on plants growth [16].

It is important to note that radicals earlier described are generated in nature on intracellular plasma membranes of plants or during their decomposition, but under stress conditions the radicals' concentration varies and the wrong concentration of ROS could lead to non-germinating seeds and even to an irreversible damage, the concept of an "oxidative window" is then used [17].

Therefore plasmas technologies could be efficiently used when the concentrations of ROS are under this oxidative window.

An additional influence of ROS formed on non-thermal plasmas discharges on seed properties, is the hydrophilicity [18].

Remarkably the plasmas could also give hydrophobic properties to seeds in order to support cold and extremely wet soil, or to augment their storage time. Fluorocarbon plasmas were investigated to deposit chemically and thermally inert (Teflon-like) macromolecules, such as  $\text{CF}_3$ ,  $\text{CF}_2$ ,  $\text{C}^*\text{HF-CH}_2$ , and  $\text{C}^*\text{-CF}_2$  functionalities without affecting the germination [8].

The effect of radicals on germination and radicles growth is resumed on Fig. 2.

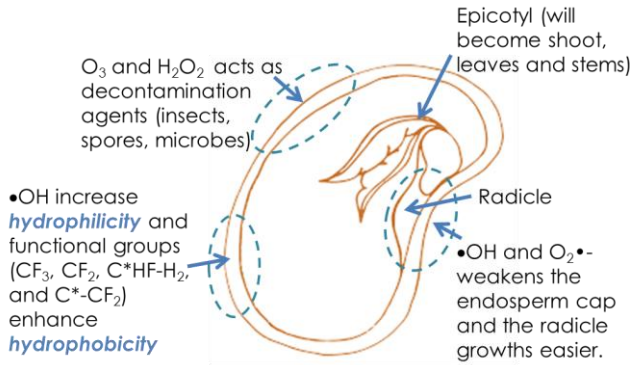


Fig.2. Influence of plasma chemical species in seeds.

## 2. Experimental set-up and preliminary results.

In this paper, a simple and inexpensive topology for an AC bipolar pulsed generator is proposed, which is able to work in a dielectric barrier discharge. The principal characteristics of the ac bipolar pulsed power supply are briefly described according to Fig. 3. The AC bipolar pulsed power supply is based in a current source, push-pull, and parallel-resonance inverter. The basic topology of this converter includes a push-pull stage composed by two transistors  $Q_1$  and  $Q_2$ , (IXFN80N50), which are controlled by a high speed double-ended PWM controller; a serial inductor ( $L_{in}$ ) directly connected to the center tap

of transformer ( $T_1$ ), and a resonant network (capacitor  $C_r$  and inductance  $L_r$  of the primary of the transformer) [19,20]. The properties of this converter are suitable for the purposes pursued here, specifically in high-frequency non-linear current discharges in a Double Dielectric Barrier Reactor (2DBD).

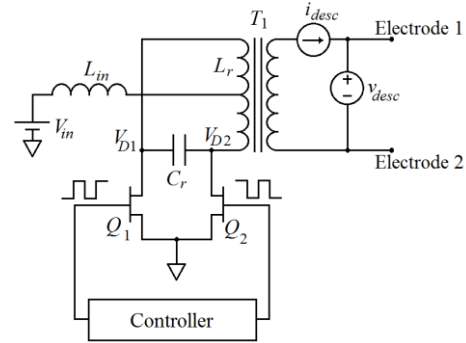


Fig. 3. Configuration of the 2DBD pulsed power source.

The 2DBD reactor was used to treat seeds of Zea Maize; the reactor consists in a quartz tube of 28mm diameter and 300mm length, outside the quartz tube, a grounded mesh electrode is wrapped. An additional dielectric made of alumina is inserted in the quartz tube. The seeds are uniformly distributed between both dielectric walls and treated in the plasma generated with 25V ( $V_{in}$ ), 20 watts, during 3 minutes under atmospheric pressure and 2 LPM of He gas (see Fig. 4).

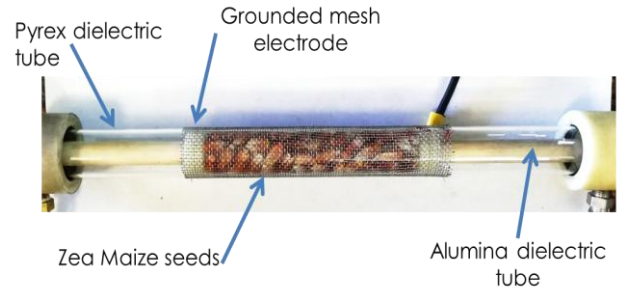


Fig 4. 2DBD reactor and distribution of Zea Maize seeds

At our laboratory, humidity reach a water vapor concentration of 1% v/v, under these conditions the continuous input of key radicals is assured, as can be seen in Fig. 5. The calculation of these radicals was obtained by only considering the discharge head, where active species are formed during the first  $1 \times 10^{-8}$  s. An electric field of  $120 \text{ kV} \cdot \text{cm}^{-1}$  ( $\approx 600 \text{ Td}$ ), an initial electron density of  $1 \text{ cm}^{-3}$  and sixteen chemical species ( $\text{N}_2$ ,  $\text{O}_2$ ,  $\text{H}_2\text{O}$ ,  $\text{O}_3$ ,  $\text{O}^\bullet$ ,  $\text{N}^\bullet$ ,  $\bullet\text{OH}$ ,  $\text{H}^\bullet$ ,  $\text{e}^-$ ,  $\text{O}_2(\text{a}^1\Delta\text{g})$ ,  $\text{O}(\text{D})$ ,  $\text{N}_2(\text{A})$ ,  $\text{O}^{\bullet 2}$ ,  $\text{O}^-$ ,  $\text{O}^{+2}$ ,  $\text{H}^\bullet$ ) were taken into account. Chemical reactions were taken from [21].

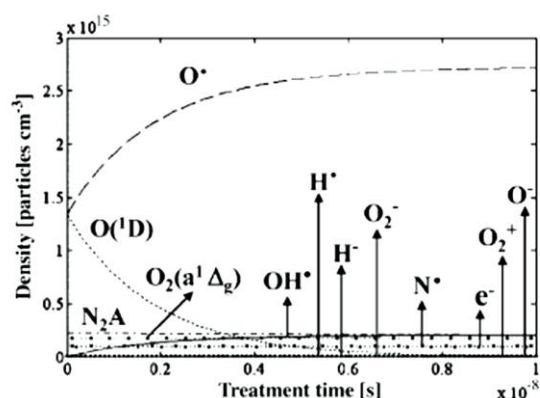


Fig 5. Radicals formed in a 2DBD reactor.

The contact angle of a 7 $\mu$ L water drop was calculated on seeds before and after the plasma treatment. In Fig. 6 the change in contact angle could be appreciated, it changes from 111° to 58°. Therefore the wetting properties of the Zea maize seed increased with 2DBD.

This may have a direct impact on the acceleration of germination and posterior growth of the plant; therefore additional work has to be done in that phase.

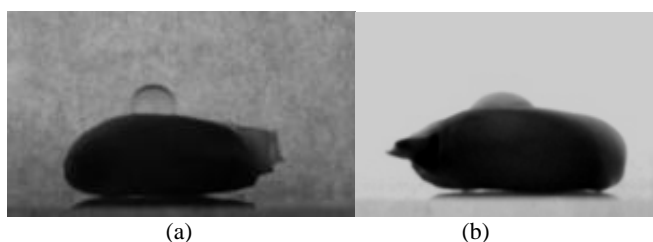


Fig 6. Change in contact angle on Zea maize seeds, (a) before the plasma treatment, and (b) after the plasma treatment.

### 3. Challenges of non-thermal plasmas technologies

Results obtained by numerous researchers show the feasibility of non-thermal plasma technology to enhance germination and the growing of plants in a world where seeds are subjected more and more to stress situations.

However, additional research has to be done to deeper understand the role of other radicals formed in the plasma, such as the radical O•, which concentration is higher than that of •OH. The study of those radicals could be supported by optical emission spectroscopy study of the plasma discharge.

Additionally, the investigation of the influence of different concentrations of ROS has to be done; it is worth to note that •OH reduces the need of the after-ripening whilst increasing the concentration of H<sub>2</sub>O<sub>2</sub> inhibits germination [22], or elevated concentrations of •OH can lead to cell death [23].

As a final observation, the use of plasma technology has to take into account its impact on environment by

reducing noxious chemicals and the energetic requirements.

The use of 2DBD reactors achieves the seed germination enhancement with only 4kJ of energy. No expensive inert atmosphere or reduced pressure equipment is required.

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