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# Exploitation of Cold Plasma Technology for Enhancement of Seed Germination



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#### Abstract

Cold plasma is an emerging non thermal technology primarily used for microbial disinfection and surface modification. The principle of plasma surface modification is exploited in food and agriculture for enhance of seed germination and other reasons. The aim of the present review is to give some insights on cold plasma technology exploitation for enhancement of seed germination. The seed germination rate can be increased on application of cold plasma by both direct and indirect treatments. Recently, the indirect treatment through the application of plasma activated water (PAW) has drawn some attentions. The formation of reactive oxygen species and reactive nitrogen species in the plasma are mainly responsible for increase in seed germination rate. Of all those reactive species formed in the PAW, the nitrate ions severe as the fertilizer and NO radical breakdown dormancy which enhanced the seed germination rate. The synergistic effect of cold plasma can replace the traditional seed disinfection solutions and chemical seed germination enhancers.

Keywords: Seed; Germination; Cold plasma; Plasma activated water; Reactive species

# Introduction

The present human population statistic says that the population of the earth reached to about 7.6 billion. It is insufficient for the total cultivable land to meet the demand of increasing population consumption. In order to meet this demand, the only way is to increase the crop yields. Enhancing the seed germination rate and draught resistant seeds is the best way to improve crop production. The most commonly used methods to enhance the seed germination are the chemical methods (agrochemicals, fungicides, insecticides and hormones). But these types of treatments leave chemical residues which are harmful to the human health and environment. Therefore, it is necessary to develop new alternative technologies to enhance seed germination to keep up with pace of growing population. Filatova et al. [1] reported that the non thermal and electromagnetic technologies have been successfully applied to enhance the seed germination. Cold plasma is one of such technologies recently drawn interest in food and agriculture sector.

Plasma is known as the fourth state of matter, charged gas with strong electrostatic interactions. Plasma consists of neutral and excited atoms, free radicals, negative and positive ions, UV photons with net electric charge zero. Bogaerts et al. [2] reported that the plasma is divided into high temperature (local thermal equilibrium) and low temperature (non-local thermal equilibrium). The cold plasma contains low temperature particles like neutral molecules, atomic species and relatively high temperature electrons because of this the plasma is cold and does not affect the sensitive materials which comes into contact [3]. Ling et al. [4] stated that cold plasma treatment is thought to be a fast, economic and pollution-free method to improve seed performance, plant growth and ultimately plant production.

The cold plasma treatment of seeds has a synergistic effects i.e. it acts as antimicrobial agent (ability to kill wide range of microbes) and enhance the seed germination and plant growth. Till date there are many reviews on the antimicrobial effects of cold plasma treatment but only few reviews on the seed germination enhancement. The aim of the present review is to give some insights on the seed germination enhancement by the cold plasma treatment.

# Seed germination effect of cold plasma

Sirova et al. [5] reported that the seedling growth during germination involves two key steps 1) primary cell elongation of the axial part of the embryo, and 2) simultaneous or delayed

cell division in the radicle meristem. Ji et al. [6] reported that the seed germination is initiation of embryo breaking the dormant stage and always start with imbibition of water. Seed germination activity involves several physiological and biochemical changes such as protein synthesis, enzyme activation and starch metabolisms [7]. The authors have also reported that the seed germination always hindered by seed dormancy factors which is undesirable process. The cold plasma can be applied by two different ways

I. Direct treatment of seeds,

II. Indirectly treating the seeds with plasma activated water (PAW) or plasma acid.

#### **Direct treatment**

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In direct treatment method the seeds are directly placed in between the electrodes or placed under the plasma regime like in plasma jets. Direct exposure of chickpea seeds to atmospheric cold air plasma for 5 min by Mitra et al. [8] observed an overall increase in seed germination by 89.2%. The authors have reported that the increase in the seed conductivity and seed roughness after the plasma treatment is the main reason for enhancement. The increase in seed roughness or etching caused by bombardment of reactive species may be the reason for increase in hydrophilicity of seeds. Thirumdas et al. [9] reported that the radio frequency low pressure plasma treatment of brown rice increased the hydrophilicity. Plasma etching is the removal of volatile substrates removed by chemical reactions and/or physical means (ion bombardment) and which are carried away in the gas stream. An investigation carried out by Bormashenko et al. [10] on the wettability of seeds using cold air plasma decreased the contact angle was from 115° to 0°. The plasma treatment resulted in complete hydrophilicity of seeds. The water imbibition of plasma treated seeds was increased by 30% after the 12h of germination.

Dubinov et al. [11] exposed barely seeds to both continuous and pulsed glow discharge plasmas, with a pulse repetition rate of 0.5Hz and pulse duration of 150-200ms in air under 0.1-0.2Pa. The authors reported that the number of germinated seeds after the exposure was increased more than 27% after the 5th day of germination. Low pressure plasma treatment of mung beans with a radio frequency (13.56MHz) significantly enhanced the germination rate by 36.2%, radicle root length by 20% and conductivity of seeds by 102% when compared to the untreated samples.

A detailed study conducted by Ji et al. [7] on germination of coriander seeds using nitric oxide (NO) gas produced from microwave plasma torch and explained all the possible ways for increase in seed germination rate. The authors have observed 91-97% germination of seeds after treatment whereas only 60% in the case of control/untreated seeds. The treated seeds showed longer radicals and stems length compared to control seeds. The NO formed in the plasma serve as a signaling pathway, triggers activation of several biological processes and a crucial regulator for cellular activation. The authors finally concluded their argument that the NO formed in the plasma is responsible for seed germination enhancement. A similar explanation was given by Zhang et al. [12] on the synergistic effect of formation of endogenous NO radicals and breakdown of seed dormancy resulted in increase in higher germination rates.

Violleau et al. [13] reported that the oxygen plasma treatment of corn seeds increased the germination rate and higher yields. Another study conducted by Puligundla et al. [14] on effect of corona discharge plasma jet on sprouting of rapeseeds. Exposure of rapeseeds for 1 min increased the germination rate by 7.7% compared to untreated seeds. The authors have reported that exposure of seeds resulted in scarification; formation of deep longitudinal cracks on the seed surface, increase in surface area attributing to increase in surface energy. Sera et al. [15] studied the effect of cold plasma on seed germination of oat corns and wheat at 500W at different time durations. The authors have observed that plasma treatment did not affect the germination of oats but there is slight increase in germination rate in wheat. Similar increase in the germination rate of rice was observed after the treatment with static magnetic fields [16]. Table 1 report on the different seeds enhancement caused by cold plasma.

Table 1: Key findings of different studies conducted on the seed germination rate using cold plasma technology.

Seeds	Plasma Source	Feed Gas	Key Findings	References
Rapeseed	DBD plasma	Argon	No change in the germination rate. Increase in seed viability was observed	Schnabel et al. [23]
Реа	Diffuse coplanar surface barrier discharge	Air	Increase in germination percentage, root length, shoot length, seed vigor	Stolarik et al. [24]
Herbaceous plant	DBD plasma	Air	Permeability of the seeds was improved significantly, acceleration of seed germination and seedling emergence	Jiayun et al. [25]
Radish sprout	atmospheric pressure plasma torch	Oxygen	Enhancement of the germination and lengths of the stem and root of plants are observed after seeding	Hayashi et al. [26]

Tomato	Arc discharge Plasma	Air	In pot experiments the yield is increased by 20.7%. Sprouting rate after the treatment is 32.75%, whereas the untreated was only 4.75%	Meiqiang et al. [27]
Wheat	Atmospheric pressure surface discharge	Air	Plasma had little effect on germination rate but the distribution of roots was shifted towards higher lengths as compared to untreated samples	Dobrin et al. [11]
Rapeseed	Radio frequency discharge	Helium	Significant improvement in germination rate by 6.25% in drought sensitive variety, and 4.44% in drought tolerant variety	Ling et al. [4]
Soybean	Radio frequency discharge	Helium	Germination and vigor indices significantly increased by 14.66% and 63.33%, respectively. Water uptake improved by 14.03%	Ling et al. [28]
Mung beans	Radio frequency capacitively coupled plasma	Air	Increase in following parameters- germination rate by 36.2%,radicle root length by 20% and conductivity of seeds by 102%	Sadhu et al. 2017
Coriander seeds	DBD	Nitrogen	After 7 days of germination there is 90% of seed germination compared to 40-60% for control seeds	Ji et al. [7]

# **Indirect treatment**

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In this type of treatments the seeds are treated with plasma activated water (PAW). The PAW is generated by application of cold plasma on the water surface or underneath water using different plasma sources. Shen et al. [17] reported that based on the working gas, discharge type and chemical composition of plasma several reactions are initiated in the water resulting in primary and secondary reactive species changing the physicochemical properties of water. The important reactive species formed in the PAW are reactive oxygen species (ROS) like atomic oxygen, singlet oxygen, superoxide, hydroxyl radicals and reactive nitrogen species (RNS) like peroxynitrite, nitric oxide, nitrates and nitrite ions. The formation of these reactive species attributes some important change in properties of water are pH, oxidation-reduction potential, conductivity, H2O2concentration, nitrites and nitrates concentration.

Naumova et al. [18] conducted a study on underwater electric front type discharge to generate PAW and treatment of PAW on rye seeds. The rye seeds treatment with PAW for 5min increased the germinability by 50% and the number of seeds germinated. Along with the hormones, the ROS and NO radicals participate in several signalling pathways involved in the seed germination [19]. The studies related to germination suggested that the ROS including superoxide, hydroxyl radicals, hydrogen peroxide and atomic oxygen are responsible for seed germination [20]. An investigation carried out by Ji et al. [6] on the seed germination of spinach observed a slight increase in germination rate after treating with DBD N2 plasma. The authors have also reported increase in level of GA3 hormone and mRNA expression of an amylolytic enzyme related gene expression in seeds. Sirova et al. [5] reported that the reactive nitrogen species (RNS) particularly NO can break the seed dormancy and enhance the seed germination.



Figure 1: Response surface and contour plot showing the interactive effect of glycerol concentration on the nitrilase activity.

A study conducted by Sivachandiran & Khacef [21] on the effect of PAW (generated using cylindrical double DBD (Cyl-DBD) reactor and tap water on seed germination. The authors have observed 60% and 100% germination rate when seeds treated with PAW-15 and PAW-30 respectively along with improved seedling growth whereas only 40% with tap water. They have stated that the increase in nitrate ion concentration is the main reason for obtaining higher germination rates. Takaki et al. [22] reported that the nitrate species formed in PAW served as a fertilizer which resulted in enhanced growth. The activation of tap water with the atmospheric plasma jet increased the germination rate of lentil seeds to 80% compared to 30% of untreated seeds (Table 1) [23-28].

## Conclusion

It can be concluded from the reports of several researchers that the seed germination rate enhancement is achieved from cold plasma technology. The plasma etching or scratching effect on the seed coat increased the hydrophilicity of seeds. Few authors have reported that few reactive species formed in the plasma has the ability to breakdown the seed dormancy and increased the seed germination rate. But a detailed studied on the formation of different reactive species formed in the plasma needs to be carried out. Cold plasma can be considered as the alternative technology for enhancement of seed germination rate and plant yield.

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