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Seed Germination Enhancement via Cold Plasma Therapy: A Review

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Abstract:- Cold plasma therapy has a wide potential for seed germination. Sometimes even though moisture and temperature are in sufficient amounts seeds of many plant species fail to germinate. Thereby to increase the germination potential of such plant seeds need an exposure to stressed condition. However many times it happens that stressful conditions result in decreased growth. This limitation can be overcome by cold plasma therapy.For this treatment a non-thermal dielectric barrier discharge is used. This technique plays an important role in seed germination. However, the effect on germination rate by exposure to cold plasma varies for different species. Overall it has been observed that upon optimizing the plasma dosage an enhancement in seed germination along with increased potential germination is obtained. In this review article the cold plasma treatment for various plants and their effects are included. More focus is given on the effect on seed germination. Thus this paper shall provide a brief idea on impact of non-thermal plasma treatment on seeds.

Keywords:- *Cold Plasma Therapy, Seed Germination, Reactive Oxygen Species (ROS), Reactive Nitrogen Species (RNS), Wettability, Contact Angle.*

I. INTRODUCTION

Seeds are whole and sole material for production of plants. Better the quality of seed, better is the production. Thus it becomes necessary to select the seed that has efficient germination potential. Usually the seeds once sowed undergo dormancy and fail to germinate until favourable conditions arise. Thus, optimum conditions are required to initiate seed germination. As crops are cultivated in open land, maintaining optimum conditions is a major barrier due to unpredictable climate changes. Failure of seed germination as a result of environmental stress is a major threat. But what if we treat the seeds prior to sowing with a specific method so they survive as well as germinate irrespective of adverse environment? Striving to this concept, researchers conducted several studies using cold atmospheric pressure plasma as a technique to enhance seed germination potential as there was some evidence that cold plasma improves seed germination. According to a data estimated by Food and Agriculture Organization of the United Nation (FAO) the world population is 7.8 billion and would reach to 9.1 billion by 2050 which shall lead to about 59% to 98% increase in the food demand by 2050. According to a report by Deepak *et al*, yields of major crops such as Rice, Soybean, Maize and Wheat which accounts for two third agriculture product have been increasing at 1.0 %, 1.3%, 1.6% and 0.9% per year respectively. However, yield at this rate shall not be sufficient enough to meet the demand. The yield rate is 2.4 % less per annum in order to achieve the double global production by 2050.(Deepak K. Ray, 2013). Many researchers have advised to target the increase in crop yield rather than clearing the land for food production.(Godfray HCJ, 2010).In order to increase the crop yield, smart, efficient, and budget friendly techniques should be implemented along with traditional methods.Cold plasma technique is a unique technique applied under green biotechnology which has a direct impact on seed quality improvement (Božena ŠERÁ1, 2018). It is an alternative technique for traditional methods. This technique is different from mutation breeding, as it uses non ionized electron beams with low radiation, there is no chance for genetic risk. Many studies are carried out using this non thermal pressure plasma technique for analysing the effects on seeds, crops and their yield. This paper gives an overall outline of how cold plasma therapy improves the seed germination in various plants and how it can be used to enhance seed germination.

- *Cold plasma technology*

Cold plasma technique is also known as Non thermal plasma treatment. Plasma is the fourth state of matter constituting a major part of the universe. Upon applying heat to gas, plasma is formed. Plasma is highly energetic as it continuously release the electrons. It is available in a wide range of temperature without changing its state. Various methods can be used for generation of plasma like electric current waveform, electrode geometry and experiment geometry. Plasma does not have a specific shape. Presence of electric and magnetic fields shape it in different structures. Plasma is mainly categorized into two types: Thermal and Non thermal. This classification is on the basis of the temperature of their electrons, ions and neutrals. In thermal plasma electrons and heavy particles are in thermal equilibrium. In non-thermal plasma they are not in thermal equilibrium. In non-thermal plasma ions and neutrons are at room temperature and electrons are at high temperature. Various instruments are used for production of cold plasma like dielectric barrier discharge, plasma needle and pencil, arc plasma discharge etc.

II. MATERIALS AND METHODS

This paper is a solely review paper, which was prepared by reviewing different research papers which were searched from the various scientific papers resources like NCBI, ELSEVIER, GOOGLE SCHOLAR, WILEY, SCIENCEDIRECT, SPRINGER, PubMed etc. Keywords used for review papers are seed germination with cold plasma treatment. We have prepared this review by selecting the papers which showed correlation between cold plasma treatment and seed germination.

III. RESULTS

➢ *Wheat*

A lot of studies were conducted for analysing the effect of cold plasma on wheat (*Triticumaestivum)* (Li Ling1, 2014)seeds. According to a study (JIANG Jiafeng, 2014)exposing the wheat seeds to a plasma discharge at different parameters such as using radiofrequency of 3x10 ⁹ MHz with a photon energy of approximately 13eV and applying series of power (60W, 80W, 100W) along with helium pressure of 150 Pa for 15 sec, a significant results were obtained when compared to untreated seeds. As per the results reported from the study, the seeds that were treated with plasma especially those at 80W showed about 6% increase in the germination potential and about 6.7% increase in germination rate as compared to the control and those treated with 60W, 100W as well. Apart from increase in growth rate, yield and germination rate; rise in weight, height, root length, chlorophyll content was observed. Also, upon stimulating wheat seeds by low temperature atmospheric plasma discharge under 500W power with a gas flow of 200ml min^{-1} at various time duration (0 to 40 min); quick germination was observed for seeds treated for 180 seconds. Also sprout acceleration was observed in wheat seeds (3 min treatment) as they had heavier shoots compared to control as well as different samples (i.e. 10-, 20-, 40 min plasma treated seeds). Also increased root/shoot (R/S) ratio was observed (Bozena Sera P. S., 2010)A study by Zahoranov´a*et al* showed that the germination increased from 70% up to 85% for control and treated samples also 12% increase in dry weight as well as water up taking capacities increased when seeds were NTP treated at radiofrequency of 14 kHz for 0-120 sec(A. Zahoranova´, 2016) Overall results from various studies show that wheat seed has increased germination potential post treatment. Overall results from various studies on wheat suggests that plasma treatment on wheat shows increased germination potential as well as germination rate.

➢ *Rice*

Rice (*Oryzasativa L*) is the most widely consumed staple food across the world. More than 4 billion people in the world's population are depending on rice and thus demand for rice production is increasing day by day. Hence there is need for increased seed germination and for which novel green agriculture technique cold plasma technique is used. For pre- treatment of rice seeds Hybrid cold plasma (HCP) was used. It will perform under atmospheric conditions and at low temperature at 27ºc and does not cause any damage to seeds. Temperature was maintained to~ 26.8ºC and base temperature was~25ºC.whcih indicates that hybrid micro corona discharge plasma was non thermal plasma.Plasma discharge will produce charged particles, reactive oxygen species(ROS) and reactive nitrogen species(RNS).Hybrid non thermal plasma has potential to remove contamination from the surface of the rice seeds and it also removes mycotoxins. Therefore cold plasma will improve wettability and germination of rice seeds. Cold plasma treatment was performed for rice with injection of Ar. Initially contact angle was 100º but it decreased as water intake increased. Rice seeds treated with Ar/plasma show no contact angle with water droplet. And time for water uptake was also decreased by 1 min.. As per the experiments whenAr with hybrid cold plasma was incorporated it will produce ROS and RNS which will decontaminate the rice seeds and energized particles of cold plasma will increase water intake which will result in enhancement of seed germination. Furthermore reactive nitrogen species like nitric oxide and Nitrate radical provide essential macronutrient to rice seed and improve seed germination. As per experiments it was concluded that non treated rice seed shows 90% of seed germination and pre-treated rice seeds shows 98% of seed germination. (Natthaporn Khamsen, 2016).

➢ *Soya bean*

Soyabean (*Glycine max*) is a species of legume and it is widely grown for edible beans. It is also used as a major oilseed crop. That's why it is always in high demand. Due to the thick and impermeable outer layer its seed germination is very difficult. Various studies have indicated that enhancement of seed germination is related with water uptake which is increased by cold plasma treatment. Seeds which are immersed in cold plasma are attacked by oxygen radicals and ions which will rupture the outer surface of Soyabean seed and increase the hydrophilic ability of Soyabean seeds and improve its water uptake by decreasing contact angle. Contact angle is the angle between seed surface and water droplet. Decreased contact angle will increase water uptake and eventually enhance seed germination. Furthermore some experiments prove that cold plasma also improves activities of enzymes responsible for seed germination and enhance decomposition of nutrients present inside the seed which will increase seed reverse utilization and promote seed growth. Experimental results suggest that cold plasma with 80W shows significant results. After pre-treatment of Soyabean seeds with cold plasma germination of Soybean seeds is increased by 14.66%, water uptake was increased by 14.03% and contact angle was decreased by 26.19%. Overall it is concluded that cold plasma treatment will increase seed germination in Soyabean seeds. (Li Ling1, 2014).

➢ *Mung*

Mung (*Vigna radiate*) is alternatively known as green gram .It is mainly cultivated in East Asia. It is used in many dishes. It is rich in nutrients and antioxidants. To increase seed germination in mung seeds were pretreated with cold plasma. Seeds were treated with O_2, He and N_2 air plasma. Increased rate of germination depends on the type of the gas

used for gold plasma. Germination index of mung seeds were increased after pre-treatment i.e. by 58.3% and 48.7 % respectively plasma air and O_2.There was no significant difference in seed germination when seeds are pre-treated with He and N_2 plasma. O_2, He and N_2 plasma air will induce the H_2O_2 radicals.H_2O_2 plays an important role in enhancement of germination. There were mainly two types of results observed. First when mung seeds are incubated for 12 hrs and treated with 0.01%, 0.03%and 0.05% there was no significant increase in seed germination. Secondly when mung seeds were incubated for 48 hrs in H_2O_2 they were entirely germinated. Other positive results were observed when seeds were treated with liquid nitrogen fertilizer. Nitrogen species like $NO_3^-$and NO_2^- are generated in plasma air which is essential for seed germination. When mung seeds were treated with Liquid nitrogen fertilizer, significant germination was observed. But when the plasma treatment was for more than 15mins germination effect was restrained. From various experiments it can be concluded that interaction between reactive species generated by plasma air (nitrogen species and H_2O_2) and seeds results in enhancement of mung seed germination.(Renwu Zhou1, 2016)

- *Peanut*

Peanut (*Arachishypogaea*) commonly known as groundnut is a legume crop and widely used as edible seeds. It is also used as oilseed crop and on various commercial crops. For enhancement of seed germination cold plasma technique was used. Here seeds were treated with helium plasma discharge. Various experiments were designed for seed treatment, but the significant results were observed in the treatment with 120 W. Hypothesis indicates that cold plasma treatment will increase wettability of seed which will decrease the contact angle of peanuts. And increase the water uptake. After cold plasma treatment seed germination rate was increased by 21% and contact angle was decreased by 53%. (LI Ling, 2016)

- *Cotton Seed*

A study was conducted by Gerard J.J B. de Groot *et al*(Gerard J. J. B. de Groot, 2018)on cotton (*Gossypium sp.*) seed to analyse the stress tolerance capacities of seeds post cold plasma treatment. For this study, compressed air and argon are used as a plasma source. Post treatment the seeds were checked for cold and warm germination tests, imbibition tests, seedling tests and chilling tolerance as well. Plasma treated seeds (27 min air plasma and 81 min argon plasma treated seeds) had higher weight which indicates that plasma treatment at particular duration increases the water intake capacity (imbibition) of the seeds. Also the germination rate was observed to be doubled for 3-min air plasma and 81-min argon plasma treated seeds (on day 4 of germination) however the air plasma treated seed showed lower germination than the argon treated seeds on day 7 and 10. Thus overall it can be said that non thermal plasma treatment can enhance cotton seed germination at optimum time and dosage.

IV. DISCUSSION

Preceding studies on various plants shows that cold plasma treatment is an efficient technique to enhance seed germination. The principle cause for seed germination is reactive oxygen species (ROS) and reactive nitrogen species (RNS) which are produced by cold atmospheric pressure plasma.RNS are essential for plant growth. It will cause acidification of the outer surface of seeds which will degrade the waxy layer of seed and increase water absorbance thus seed germination will increases.ROS will function as signalling molecule required for seed germination. In addition to this various experiments demonstrate that plasma air will decrease the contact angle of the seed surface and water droplet, which increases water uptake and results in increased potential of seed germination. Numerous studies are conducted on other plants to check the effect of cold atmospheric pressure plasma on seed germination. A study was conducted on soyabean with Diaporthe/Phomopsis fungal infection by Maria et al (María Cecilia Pérez Pizá, 2018). The experiment concluded that plasma frequency of 50 Hz will improve seed health by reducing Diaporthe/Phomopsis infection and ROS will evoke oxidation of lipids in seed coat and promote water uptake. Another study was conducted on pea by Tibor et al (Tibor Stola´rik1, 2015)and they concluded that plasma treatment will rupture the wax layer on pea, increase water and nutrient uptake. It will increase after 120 sec of pre-treatment and inhibit >300 seconds inhibit growth. Taieb et al (Taieb Tounekti1, 2018) conducted studies on coffee beans. Plasma treatment of 50W for 240 seconds for 10 days will double the germination. Jiafeng et al (Jiafeng JIANG, 2018) performed experiments on tomatoes. Plasma treatment of 80 W increases $N_2$and P absorption. Germination rate increases up to13%. Vlasta et al (Černák1, 2017)perform experiments for cucumber and capsicum. After 20 seconds of plasma treatment germination rate increased to 96% but after 30 seconds it will decrease to 93% due to thermal damage. In capsicum after 4 seconds of plasma treatment germination rate increases up to 89% and after 15 seconds decreases to 34% due to thermal damage. Moreover on many other plants satisfactory effects of plasma treatment observed i.e. in radish (A.L. MIHAI1, 2014), quinoa (A. Gómez-Ramírez1, 2017)), oat (Božena Šerá, 2010)(Bozena Sera P. S., 2010) lentil (Edward Bormashenko1), chickpea (Zimmermann, 2014) etc. However contradictory results were obtained for oats as less germination was observed(Bozena Sera P. S., 2010).

V. CONCLUSION

Cold atmospheric pressure plasma proved to be an effective technique for seed germination without any requirement of genetic manipulation. Number of studies was conducted to confirm positive effect of cold plasma on seed germination. Cold plasma treatment could be cost effective and proves to be a promising technique for seed germination.As discussed in the results, this treatment resulted in an increase in germination efficiency which lead to increased production yield. Various gases can be used as a plasma source. It has been observed that different plasma

gas has different effect on seed germination, thus due to availability / possibility of having broad list of plasma source such as inert gases and combination of gases such as heliox, further studies can be performed so that more accurate results can be obtained(Clotilde Hoffmann, 2013). Furthermore, studies using various plasma source on each seed species should be conducted to optimize the process so as to obtain the efficient and uniform production. As increase in the crop yield is obtained, this technique shall help in fulfilling the demand due to increasing population and also the track of double global production by 2050.

REFERENCES

[1]. A. Gómez-Ramírez1, 2. C.-S.-E. (2017). Surface chemistry and germination improvement of Quinoa seedssubjected to plasma activation. *Scientific Reports*.

[2]. A. Zahoranova´, M. H. (2016). Effect of Cold Atmospheric Pressure Plasma on the Wheat Seedlings Vigor and on the Inactivation of Microorganisms on the Seeds Surface. *Plasma Chemistry and Plasma Processing*, 397–414.

[3]. A.L. MIHAI1, D. D. (2014). POSITIVE EFFECT OF NON-THERMAL PLASMA TREATMENT ON RADISH SEEDS. *Research gate*, 1110-1117.

[4]. Bozena Sera, M. S. (2018). Non-thermal plasma treatment as a new biotechnology in relation to seeds, dry fruits, and grains. *Plasma Science and Technology*.

[5]. Bozena Sera, P. S. (2010). Influence of plasma treatment on wheat and oat germination and early growth. *IEEE Transactions on Plasma Science*, 2963-2968.

[6]. Božena Šerá, P. Š. (2010). Influence of Plasma Treatment on Wheat and Oat Germination and Early Growth. *IEEE TRANSACTIONS ON PLASMA SCIENCE*, 2963-2968.

[7]. Božena ŠERÁ1, 2. a. (2018). Non-thermal plasma treatment as a new biotechnology in relation to seeds, dry fruits,and grains. *Plasma Science and Technology*.

[8]. Černák1, V. Š. (2017). Atmospheric pressure plasma treatment of agricultural seeds of cucumber (Cucumis sativus L.) and pepper(Capsicum annuum L.) with effect on reduction of diseasesand germination improvement. *plasma process and polymerase*.

[9]. Clotilde Hoffmann, C. B. (2013, Oct 1). Cold Atmospheric Plasma: methods of production and application in dentistry and oncology. *Medical Gas Research*.

[10]. Deepak K. Ray, N. D. (2013). Yield Trends Are Insufficient to Double Global Crop Production by 2050. *PLOS ONE, 8*(6).

[11]. Edward Bormashenko1, R. G. (n.d.). Cold Radiofrequency Plasma Treatment Modifies Wettability and Germination.

[12]. Gerard J. J. B. de Groot, A. H.-P. (2018). Cold plasma treatment for cotton seed germination improvement. *Nature/ Scientific Reports*.

[13]. Godfray HCJ, B. J. (2010). Food security: The challenge of feeding 9 billion people. *Science*, 812–818.

[14]. Hugo Valin, R. D. (2014). The future of food demand: understanding differences in global economic models. *Agricultural Economics, 45*, 51–67.

[15]. Jiafeng JIANG, J. L. (2018). Effect of cold plasma treatment on seedling growth and nutrient absorption of tomato. *Plasma Science and Technology*.

[16]. JIANG Jiafeng, H. X. (2014). Effect of Cold Plasma Treatment on Seed Germination and Growth. *Plasma Science and Technology,*, 54-58.

[17]. LI Ling, L. J. (2016). Improving Seed Germination and Peanut Yields by Cold Plasma Treatment. *Plasma Science and Technology*.

[18]. Li Ling1, 4. J. (2014). Effects of cold plasma treatment on seed germination and seedling growth of soyabean. *SCIENTIFIC REPORTS*, 1-7.

[19]. María Cecilia Pérez Pizá, L. P. (2018). Effects of non–thermal plasmas on seed-borne Diaporthe/Phomopsis complex and germination parameters of soybean seeds. *Innovative Food Science and Emerging Technologies*.

[20]. Natthaporn Khamsen, †. D. (2016). Rice (Oryza sativa L.) Seed Sterilization and Germination enhancement via atmospheric hybrid nonthermal discharge plasma. *ACS Applied Materials & Interfaces*, 19268−19275.

[21]. Renwu Zhou1, 2. R. (2016). Effects of Atmospheric-PressureN2, He, Air, and O2 Microplasmas onMung Bean Seed Germination and. *Scientific Reports*.

[22]. Taieb Tounekti1, Z.-u.-I. M. (2018). Non-Thermal Dielectric Barrier Discharge (DBD)plasma affects germination of coffee and grape seeds. *AIP publishing*.

[23]. Tibor Stola´rik1, M. H. (2015). Effect of Low-Temperature Plasma on the Structureof Seeds, Growth and Metabolism of EndogenousPhytohormones in Pea (Pisum sativum L.). *Plasma Chem Plasma Process*, 659-676.

[24]. Zimmermann, A. M.-F. (2014). Inactivation of Surface-Borne Microorganisms and IncreasedGermination of Seed Specimen by Cold Atmospheric Plasma. *Food Bioprocess Technol*.