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Original paper

Research regarding the impact of cold plasma treatment applied to wheat crop seeds

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Abstract

World demand for food has an upwards trend, and in this context, solutions are sought so that each seed can benefit from optimal conditions, have a uniform rise, and produce vigorous plants that produce at the potential level. Cold plasma treatments are potentially useful in treating seeds to improve the germination quality and increase this in the early stages of vegetation. This paper aims to test the impact that cold plasma treatments have on wheat seeds and to determine the influence of treatment duration and discharge voltage on germination parameters. Applying cold plasma treatments to wheat grains had different effects depending on the time and discharge voltage used but showed a positive impact, manifested by improved germ characteristics and an increase in the biometric parameters studied. Thus, the increase in treatment duration and discharge voltage resulted in significant increases in germination parameters compared with untreated controls. Due to the results obtained, plasma can become an effective alternative method for treating seeds used in agriculture.

Keywords

Cold plasma treatment, germination rate (G).

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Introduction

With over 200 mil ha cultivated area wheat is one of main crops in the world.

The importance is given by the rate of wheat in the human nutrition and the world population growing trend, from 6 billion in 1999 to 7 billion in 2011 (FAOSTAT [1]). In the conditions of the rapid increase of food demand, it's needed to ensure the safety and security of foods, and to improve the technologies of crops, so that the level of production obtained allows the food request to be covered, while maintaining the level of the cultivated area constantly at same level.

In order to meet these requirements, particular attention must be paid to the efficiency of each component of the crop production technology flow. The first element with an impact on production is seed quality, which is why phytosanitary treatments for disease and pests protection, as well as treatments with biostimulating effect are important.

In the category of physical methods available for seed treatment, is the exposure of the seeds to the action of different fields of radiation and especially the plasma seed treatment (JIANG [2]).

The cold plasma treatment could improve the germination, promote the growth and increase the metabolic process level of wheat, leading to increased yield.

Cold plasma treatment was used for the first time for activation and decontamination of surfaces. Recently it has been applied successfully for the treatment of plant seeds. The treatment of corn seeds with plasma 50 W and 100 W at a pressure of 500 mTorr for 2.5 minutes positively influenced their germination and biometric characteristics of sprout (FILATOVA [3]).

Radish seeds harvested exposed to 9.3 GHz microwaves had higher germination compared with non-irradiated seeds. High power microwaves increased radish germination energy by 6% (RADZEVIČIUS [4]).

With the proper conditions of exposure, plasma conditioning of seeds provides a stimulating effect and could be used for pre-sowing seed stimulation. Exposure of seeds to a continuous plasma discharge is more effective than exposure to a pulsed plasma discharge. When the germination level is high the stimulating effect is more weakly expressed. (DUBINOV [5]).

The seed germination of oilseed rape was adversely affected by drought stress, but this was significantly improved by the cold plasma treatment, especially for the drought-sensitive cultivar, Zhongshuang (LING LI [6]).

The germination rate of corn seeds was also unaffected while the growth of corn plants showed slight improvement by the plasma application. Therefore, the glow discharge plasma could be applied as a pre-treatment of seeds in order to improve the cereal crops (BRAȘOVEANU [7]).

After exposing the wheat seeds to plasma treatment in a surface discharge atmospheric pressure and at room temperature, it was found that the roots and sprouts of plasma treated samples were longer and heavier than those of the untreated seeds, for all treatment durations investigated (DOBRIN [8]).

The main objective of this research was to identify the effect of the cold plasma treatment upon the winter wheat seeds, applied at the different duration and different voltages of discharges. The second objective was to establish the optimum parameters for winter wheat seeds treatment with the cold plasma.

Materials and Methods

Experiment

To test the effect of non-thermal plasma on wheat grains, carried out was an experiment with two factors where A factor is voltage with 4 graduation a_1 0 kV, a_2 13 kV, a_3 15 kV, a_4 17 kV, and Factor B during the plasma treatment b_1 t 0 min, b_2 t 5 min, b_3 t 10 min and b_4 t 15 min. The experiment was made in 4 replications and in each variant were 100 seeds.

The Cold plasma reactor

The seeds were treated by non-thermal plasma using a surface discharge reactor, at atmospheric pressure and room temperature. The set-up is illustrated in Figure 1.

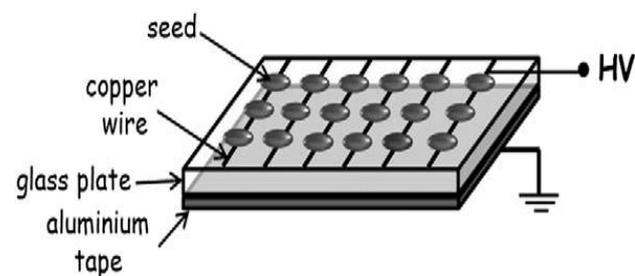


Figure 1. Experimental set-up: HV – a.c. high voltage transformer, high voltage electrode – array of 13 copper wires (diameter 100 μ m, length 44 mm, distance between adjacent wires 6 mm), ground electrode – aluminum tape (length 90 mm, width 48 mm), glass plate (length 120 mm, width 90 mm, thickness 1.5 mm), and seeds – *Triticum aestivum*.

The plasma reactor consists of two electrodes placed on both sides of a glass plate (length 120 mm, width 90 mm, thickness 1.5 mm). The high voltage electrode is an array of 13 copper wires, each wire having a diameter of 100 μ m and length of 44 mm. The distance between adjacent wires was 6 mm. The ground electrode was an aluminum tape (length 90 mm, width 48 mm). The seeds were distributed uniformly on each wire. The plasma reactor was placed in a rectangular case and air was flown with a rate of 1 L/min. The discharge was generated in AC mode using a high

voltage transformer which provides a sinusoidal voltage at 50 Hz frequency. The discharge voltage was measured by a high voltage probe (P6015A, Tektronix, USA). The current was measured with a shunt resistor of 3 Ω connected in a series with the ground electrode. The total charge dissipated in the discharge was measured with a non-inductive capacitor of 1.5 μF placed instead of the shunt resistor. The discharge characteristics were monitored by a digital oscilloscope (DPO 2024, Tektronix, USA). The average power dissipated in the discharge was calculated by the Lissajous method.

Seeds for testing

The wheat (*Triticum aestivum*) seeds used for the test were from Pedro variety, the production of year 2017, Thousand Grain Weight 43,52, grain specific weight 72,6. For testing the germination was used “Towel Germination Test” method (TGL) in accordance with the standard SR1634/1999. (ISTA [9, 17]). The samples were incubated at 20°C and at 80% humidity in the Caloris ITU 150 vegetation chamber.

- a. Germination rate The germination rate $G = Ng \cdot 100 / Nt$, Ng is the number of germinated seeds, is the total number of seeds tested ISTA [10, 11].
- b. Speed of germination was calculated by formula $SG = \sum_{i=1}^N \frac{n_i}{d_i}$ where, n_i = number of germinated seeds in the day i, d_i = number of days from start testing (day 1, day 2...day i) [3, 12, 13, 14].
- c. Mean germination Time $MGT = \sum_{i=1}^N \frac{n_i d_i}{N}$ where n = number of germinated seeds, d = number of days, N = total number of days of monitoring test [3, 12, 13, 14].
- d. Mean daily germination $MDG = Ng / N$,

Peak Value (PV) is the day when recorded the highest number of seed germinated, calculated by the following

$$PV = \frac{\max_x n_x}{d_x}$$

formula given by $PV = \frac{\max_x n_x}{d_x}$, n_x = the maximum number of seed germinated in one day, d_x = the day when was recorded the maxim number of germinated seeds in one day.

- e. Germination Value $GV = PV \cdot MDG$ [12]

Statistical analysis

Data was subject to analysis of variance using XL STAT application under Excel.

The data were compared using Least Significant Difference (LSD) for 5%, 1% and 0,1%.

Results and Discussions

The effect of plasma treatments applied to wheat grains vary depending on discharge voltage and treatment time. We present the impact in which the factor A is the voltage of discharge and factor B is the duration of the discharge on the parameters of germination characterization that have been studied.

Analyzing the germination rate (G) Fig 1, it is found that it varies between 83% in the untreated control and reaches a maximum of 88.8% at a_3b_1 . Analyzing the influence of the voltage of discharge (A Factor) on the rate of germination, it was found that all treatments determined the G increase, but these significant increases occurred only at the voltages of 15 and 17 kV. For the same time of treatment, increasing the voltage of discharge caused increases in germination rates, these increases are not significant for one voltage of discharge of 13 kV for all times of the treatments; for 5 min duration of treatment at 17 kV the increase was distinctly significant, and for all other durations of discharge was significantly.

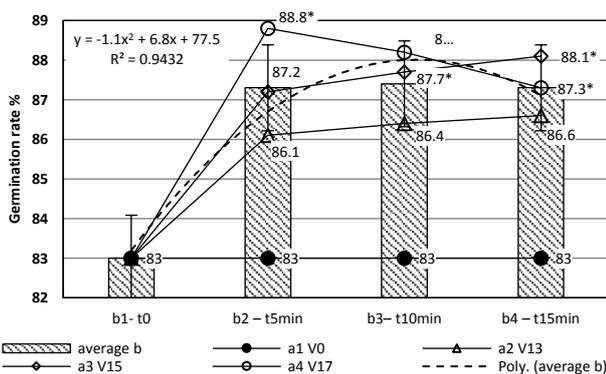


Figure 1.a. Germination rate (for same voltage of discharge)

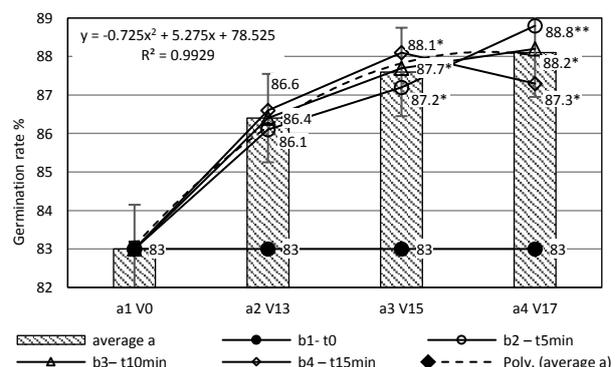


Figure 1.b. Germination rate (for same time of treatment)

JIANG [17] showed that the application of cold plasma treatments led to an increase in the germination rate, proportional to the discharge power, with significant values

being obtained at powers higher than 80 W. Applying cold plasma treatment with low power may be insufficient to determine quantitative effects on seeds.

Increasing the duration of treatment (Factor B) determined a significant increase in germination rate. For each discharge voltage, increasing the discharge time

caused significant increases in germination rate, except for the discharge at 13 kV when the increases were not significant.

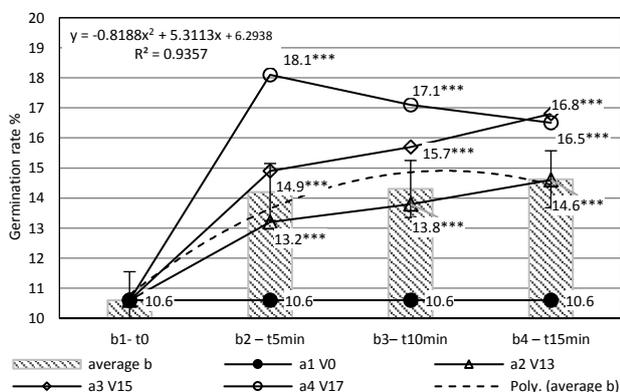


Figure 2.a. Speed of germination (for same voltage of discharge BxA)

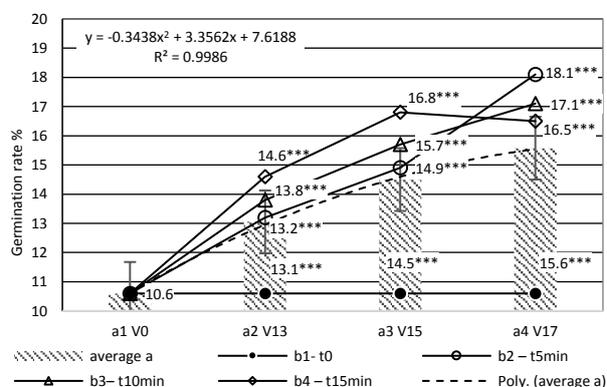


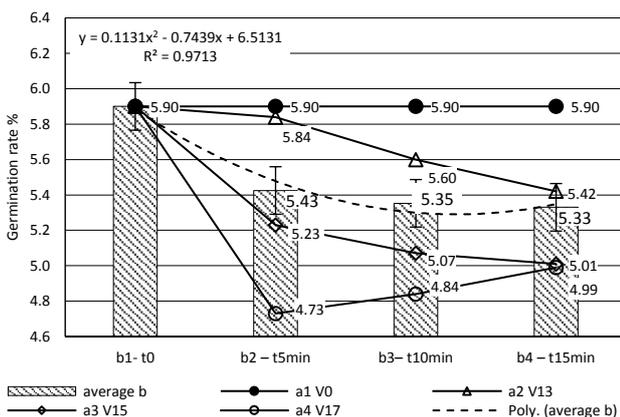
Figure 2.b. Speed of germination (for same time of treatment Ax B)

The results regarding the speed of germination (SG) are centralized in fig 2, where we can see that SG was varied between 10,6 for non treated seeds up to 18,6 at variant a4b2. The increase of the voltage of discharge was followed by increasing of SG. For the same duration of discharge the increasing of voltage was recording the very significant increases of SG. The increase of discharge time caused increases of SG. For the same voltage of discharges,

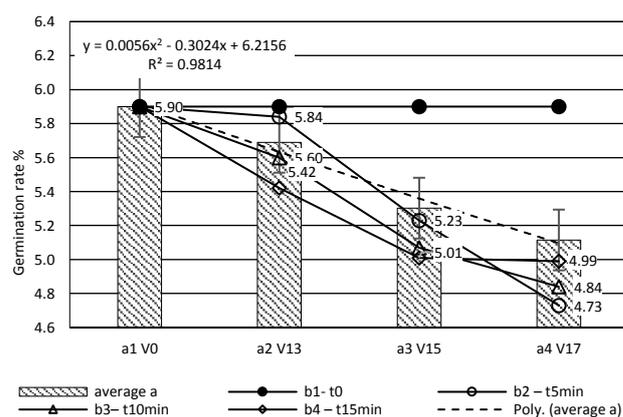
increasing the durations of treatment was obtained by the very significant increases of SG.

SHAPIRA [20] showed that the application of cold plasma treatments to alfalfa, clover and beans had the effect of increasing the speed of germination, this increase was more pronounced under drought conditions, the results being explained by the increase in the permeability of the skin under the influence of treatments.

Mean germination Time (for same voltage of discharge)



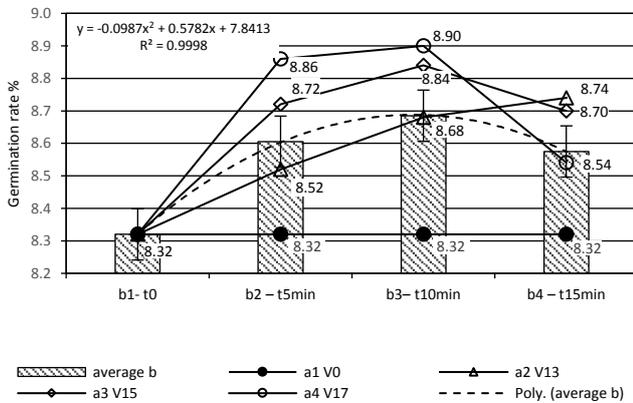
Mean germination Time (for same time of treatment)



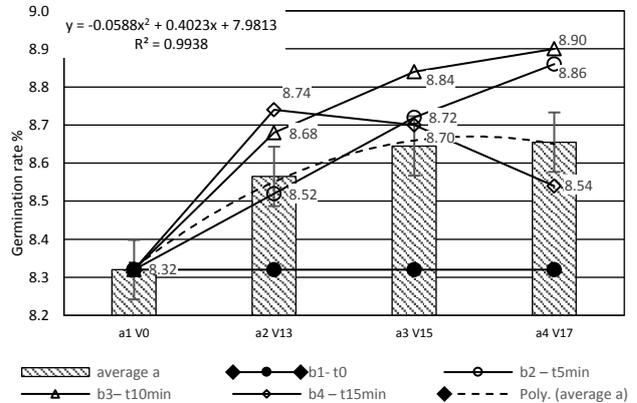
Mean germination Time (MGT) recorded a variation from 5,9 to control variants up to 4,8 at a3b1. Compared with the control, the plasma treatments resulted in a decrease in MGT. Increasing the discharge voltage caused a decrease in MGT, significant for 13 kV and very significant for 17 kV. Increasing the duration of treatment

resulted in very significant decreases in MGT compared to untreated control. For 13 kV, the increase in treatment duration resulted in a reduction in MGT compared to the untreated control, and for the 17 kV voltage, the increase in treatment duration led to a slight increase.

Mean daily germination (for same voltage of discharge)



Mean daily germination (for same time of treatment)

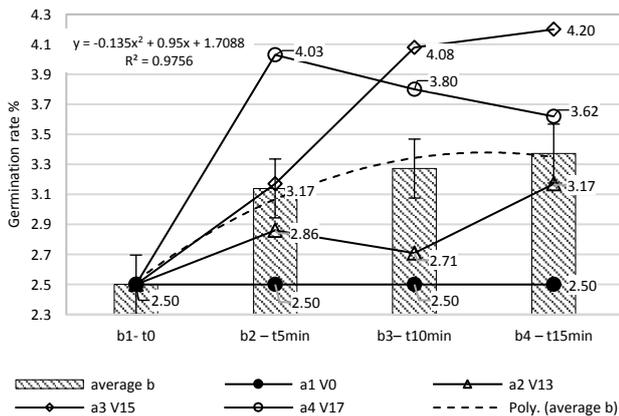


In the research carried out by (LI LONG [21]) on soybean, it has been shown that increasing the discharge power by increasing the treatment time or the discharge power resulted in an increase in MGT.

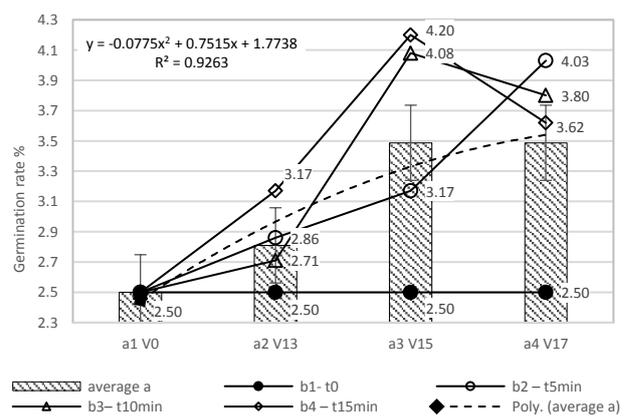
The mean daily germination MDG showed an increase from 8,3 at untreated, up to a maxim value of 8,9 at variant a₃b₁. The increase in A Factor's download voltage

has led to a significant increase in the MDG. Increasing the duration of B Factor treatment led to increases that are significant only at 15 min. By analysing the MDG for the same discharge voltage, it was found that increasing treatment duration at 13 kV did not cause significant increases, and for the 17 kV voltage, the increase in treatment duration led to a reduction in the MDG.

Peak value (for same voltage of discharge)



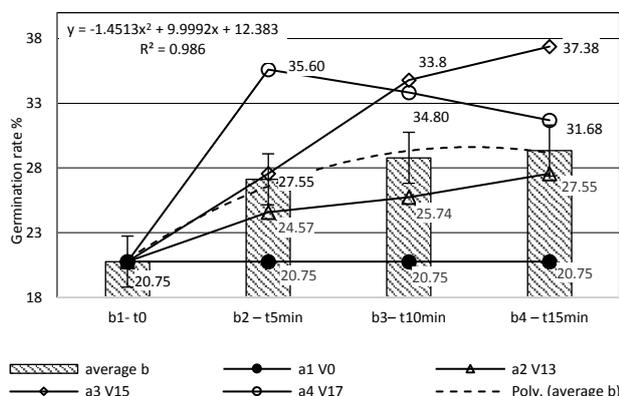
Peak value (for same time of treatment)



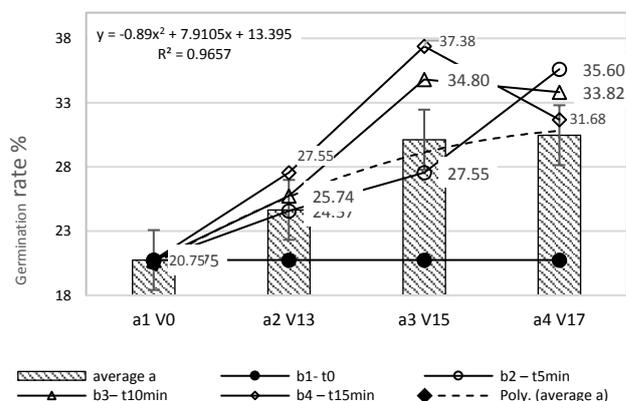
The Peak value (PV) of wheat grain after plasma treatment increased from 2.5 to control and up to 4,2 in the a₂b₃ variant. Increase in withdrawal pressure and duration of treatment resulted in very significant increases in PV

compared to the untreated control. These increases are correlated with the treatment time for treatment voltages of 13 and 15 kV, and at a voltage of 17 kV there was a decrease.

Germination rate (for same voltage of discharge)



Germination rate (for same time of tretment)



Germination value (GV) ranged between 20.8 on the control variant and 37.4 on the a2b3 variant. The increase in discharge rate and duration of treatment resulted in very significant increases in GV. Increasing the duration of treatment led to increases in GV for 13 kV and 15 kV and resulted in decreases in GV with an increase in treatment duration at 17 kV.

Conclusions

The application of cold plasma treatments to wheat seeds has led to an improvement in germination monitoring parameters.

The increase of the discharge voltage was determined in all cases by the increase of germination parameters, and the increase of the duration of treatment at the low discharge volumes had an stimulating effect, while at high voltages had an inhibitory effect.

In the case of increasing the time of application of cold plasma treatment at discharge voltages above 17 kV there were decreases in the values of the tested parameters, but also in these cases there were significant increases compared to the untreated control.

The results recorded create the premises for use of this treatment for improving seed germination and the growth indicators in earlier stage of wheat, and probably will be followed by uniform emergence in the field.

References

1. <http://www.fao.org/faostat/en/?#search/world%20population>
2. JIANG, JIAFENG & HE, XIN & LI, LING & LI, JIANGANG & SHAO, HANLIANG & XU, QILAI & YE, RENHONG & DONG, YUANHUA. (2013). Effect of Cold Plasma Treatment on Seed Germination and Growth of Wheat. *Plasma Science and Technology*. 16. 10.1088/1009-0630/16/1/12.
3. FILATOVA I, AZHARONOK V, LUSHKEVICH V, ZHUKOVSKY A, GADZHIEVA G, SPASIĆ K, ŽIVKOVIĆ S, PUAČ N, LAZOVIĆ S, MALOVIĆ G and PETROVIĆ Z. LJ; Plasma seeds treatment as a promising technique for seed germination improvement; 31st ICPIG, July 14-19, 2013, Granada, Spain.
4. RADZEVIČIUS A, SAKALAIUSKIENĖ S, DAGYS M, SIMNIŠKIS R, KARKLELIENĖ R, BOBINAS C, DUCHOVSKIS P, The effect of strong microwave electric field radiation on: (1) vegetable seed germination and seedling growth rate, *Zemdirbyste-Agriculture*, vol. 100, no. 2 (2013), p. 179-184, ISSN 1392-3196 / e-ISSN 2335-8947, DOI 10.13080/z-a.2013.100.023, www.zemdirbyste-agriculture.lt/wp.../06/100_2_str23.pdf
5. DUBINOV, ALEXANDER & LAZARENKO, E.R. & SELEMIR, V.D. (2000). Effect of glow discharge air plasma on grain crops seed. *Plasma Science, IEEE Transactions on*. 28. 180-183. 10.1109/27.842898.

6. LING LI, JIANGANG LI, MINCHONG SHEN, CHUNLEI ZHANG, YUANHUA DONG, Cold plasma treatment enhances oilseed rape seed germination under drought stress; *Scientific Reports*, 2015/08/12/online - 5 - 13033 <https://www.nature.com/articles/srep13033>
7. BRAȘOVEANU M, NEMȚANU M R, SURDU-BOB C, KARACA G, ERPER I; Effect of glow discharge plasma on germination and fungal load of some cereal seeds, *Romanian Reports in Physics*, vol. 67, no. 2, p. 617-624, 2015.
8. DOBRIN D, MAGUREANU M, MANDACHE N B, IONITA MD, The effect of non-thermal plasma treatment on wheat germination and early growth, *Innovative food science and emerging technologies* 29 (2015), 255-260.
9. ISTA, International rules for seed testing. *Seed science and Technology*, 21, 1999, pp. 288.
10. ISTA Method Validation for Seed, Testing <https://www.seedtest.org/upload/cms/user/ISTAMethodValidationforSeedTesting-V1.01.pdf>
11. ISTA Accreditation Standard for Seed Testing and Seed Sampling <https://www.seedtest.org/upload/cms/user/ISTAMethodValidationforSeedTesting-V1.01.pdf>
12. F.J. CZABATOR, Germination value: An index combining speed and completeness of pine seed germination, *Forest Science*, 8, 386, 395 (1962).
13. TUDOR V, ASĂNICĂ A, TEODORESCU RI, GIDEA M, TANASESCU C, TUDOR AD, TIU JV, Germination capacity of some *Lycium barbarum* L. and *Lycium chinense* Mill. biotypes seeds, *Romanian Biotechnological Letters*, vol. 22, no. 1, 2017, 12191.
14. K.C. GAIROLA, A.R. NAUTIYAL, A.K. DWIVEDI, Effect of Temperatures and Germination Media on Seed Germination of *Jatropha Curcas* Linn., *Advances in Bioresearch*, 2 (2), 66, 71 (2011).
15. FALKENSTEIN, Z., & COOGAN, J.J. (1997). Microdischarge behaviour in the silent discharge of nitrogen – oxygen and water – air mixtures. *Journal of Physics D: Applied Physics*, 30, 817-825.
16. <https://www.uaf.edu/files/ces/publications.../FGV-00249.pdf> Procedures for the Wet Paper Towel Germination Test.
17. JIANG JIAFENG, HE XIN LI JIANGANG, SHAO HANLIANG, XU QILAI, YE RENHONG, DONG YUANHUA, Effect of Cold Plasma Treatment on Seed Germination and Growth of Wheat, *Plasma Science and Technology*, vol. 16, no. 1, Jan. 2014.
18. ŠERÁ, B., ŠPATENKA, P., ŠERY, M., VRCHOTOVÁ, N. & HRUŠKOVÁ, I. Influence of plasma treatment on wheat and oat germination and early growth. *IEEE Transactions on Plasma Science* 38, 2963-2968 (2010).
19. HUANG, M.J. Physiological effect of plasma on wheat seed germination. *J. Shanxi Agricultural Science* 38, 22-25 (2010).
20. YEKATERINA SHAPIRA, EDWARD BORMASHENKO, ELYASHIV DRORI, Plasma treatment of seeds, its influence on wetting and germination of seeds, <https://www.ariel.ac.il/sites/conf/mmt/mmt-2016/Service%20files/papers/32-45.pdf>
21. LING L, JIAFENG J, JIANGANG L, et al. Effects of cold plasma treatment on seed germination and seedling growth of soybean. *Sci Rep.* 2014; 4:5859. Published 2014 Jul 31. doi:10.1038/srep05859
22. SERÁ, B., GAJDVÁ, I., ŠERY, M. & ŠPATENKA, P. New physicochemical treatment method of poppy seeds for agriculture and food industries. *Plasma Sci. Technol.* 15, 935-938 (2013).