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THE IRON OXIDE (FE_2O_3NPS) AND GRAPHENE OXIDE (GONP$_S$) NANOPARTICULES PRIMING TREATMENTS ALLEVIATE THE EFFECTS OF SALINITY DURING GERMINATION OF THE SOYBEAN

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ABSTRACT

Nanoparticles containing essential plant nutrients, including micronutrients, can be effective in improving germination characteristics. In the present study, the aim was to evaluate seed germination characteristics by priming application. The study was conducted at the Field Crops Laboratory, Faculty of Agriculture, Adnan Menderes University. For this study, the germination characteristics of soybean seeds were investigated in four different priming treatments (control, Fe_2O_3NPs, Graphene oxide, Potassium humate) in both saline and non-saline conditions. The results of the study showed that Fe_2O_3NPs, graphene oxide, and potassium humate had positive and significant effects under both conditions. It was also determined that saline conditions caused a decrease in the observed properties. Among these priming treatments, almost all traits, such as germination percentage (77.19%), root length (31.74 mm), and hypocotyl length (67.01 mm), were observed in the Iron oxide NPs treatment. The study revealed a significant and positive effect of priming applications on the germination percentage and the examined characteristics in both environments.

Keywords: Soybean, Fe_2O_3 Nanoparticles, Graphene Oxide, Potassium Humate

1. INTRODUCTION

Nanofertilizers, unlike traditional fertilizers, contain nanoparticles that can provide nutrients to plants more effectively Kashyap et al. (2017), Daghan (2017). Nanosized particles are easily absorbed by plants and can be utilized more effectively. Nanofertilizers are used in agriculture to increase crop yield and nutrient utilization efficiency, while decreasing the reliance on excessive chemical fertilizers. The most important features of these fertilizers are that they contain one or more macro and micronutrients, can be applied in small amounts and frequently, and are environmentally friendly. Sharifi et al. (2016). Salinity stress is one of the most significant abiotic factors that limits crop production worldwide Efisue & Dike (2020), particularly in Mediterranean regions like Turkey. It usually affects the ribulose bisphosphate carboxylase-oxygenase activity, photosystem II light-capturing efficiency and electron transport ability Yang et al. (2006). Soybean (Glycine max L.) is one of the most important plants in the world. It is widely grown for its edible beans and as an industrial crop Teixeira et al. (2019). The seeds of the annual soybean plant, which is cultivated in summer, contain an average of 36-40% protein, 18-24% fat (including Omega-3 and similar fatty acids), vitamins B1, B2, K, and E, as well as elements such as Zn, Fe, and Ca, totaling 27%. Contains carbohydrates and 18% mineral substances Kumlay et al. (2021). Soybean, which is among the legume product group, was grown on 131 million hectares of land in the world in the 2021 production season. The country with the largest cultivated area in the world is Brazil, with 41.5 million hectares FAO (2022). Turkey's soybean production increased by 30% in the last 5 years and reached 182,000 tons in 2021. FAO (2022) The most important feature of soy is that it provides more and cheaper protein per unit area compared to other plant and animal feed sources. Soy protein is the protein closest to animal protein and has a very high biological value Tüfekçi (2019).

Priming application can accelerate the germination process in seeds, increase stress tolerance, and result in healthier seedlings. It is also important for breaking dormancy in seeds and promoting germination Arnott et al. (2021), Nile et al. (2022). It can contribute to a more efficient germination process by allowing seeds to adapt to environmental stresses more quickly. In this way, a healthier germination and more uniform plant stand formation are ensured. Priming application ensures that the seeds are more resistant to environmental stresses during the germination stage. For example, primed seeds may be better adapted to stress conditions such as drought or salinity and may exhibit better performance under such circumstances. This can give seedlings a better start in the early growth stages and allow for more robust plant development. It can promote more uniform growth among plants obtained from germinating seeds. This can help create a more organized and efficient plant stand in agricultural fields. Pawar et al. (2019).

Nanofertilizers are one of the simple and cost-effective methods for reducing pests Chandrasekaran et al. (2020). Encouraging results have been observed in seed preparation, product productivity, and seed germination with the use of iron nano-oxide. Nanopreparation is a method that releases specific nutrients Sanzari et al. (2019). Increasing input costs cause farmers to aim for the easiest and fastest yield increase. In this study, it was aimed to determine the effects of nanoparticles on germination by priming soybean seeds.

2. MATERIAL AND METHOD

2.1. EXPERIMENTAL SITE

The study was conducted at Field Crops Department, University of Agriculture, Aydin, Turkey. The experiment was carried out in the randomized complete plot design with three replications. In the study, soybean seeds were carried out under 23 0C temperature and 45-50% air humidity conditions before germination.

Soybean genotype was used as a plant material. The seeds were obtained from Agricultural Research Center, Izmir, Turkey. The initial seed moisture contents were 10.0 % (on dry weight basis).

2.2. TREATMENTS

There are four applications: control, iron oxide, graphene oxide, and potassium humate.

Control (hydro-priming), soybean seeds were soaked in distilled water (dH2O) (Basra et al., 2004). A solution containing iron oxide (Fe2O3NPs) and graphene oxide (GONPs) were separately prepared with 10 mg of the application material in 100 mL of water. K humate: A solution of 0.3 g per 1 liter was prepared. For each application, 100 grams of seeds were weighed in three replicates. The amount of solution was 1.5 times fold the amount of the seeds in the box. The seeds were covered in the solution (250 ml for each solution) for 8 hours, submerged at a level that covered the seeds, and then the seeds were dried. Seeds were placed in 10*10 cm plastic box with blotting paper inside. After beginning of the germination 36 hours, 6 ml of the solution was added for each replicate.

2.3. EXAMINED CHARACTERS

In the study, germination rate, hypocotyl length, root length, root fresh weight, root dry weight, hypocotyl fresh weight, and hypocotyl dry weight values were determined according to Pour et al. (2021).

Germination percentage (GP) was estimated at five-time intervals (9th day) according to the formula GP = (seeds germinated / total seeds) x 100. Fresh weight of the hypocotyl (FHW) was determined by weighing the wet weight of the hypocotyl on a precision scale at the end of the 9th day.

Dry cotyledon weight (DCW), after determining the fresh cotyledon weight, was dried at 105°C for 24 hours and weighed on a precision scale to determine the root dry weight. In each plastic box, the length of roots (RL), in 20 germinated seed was measured at the end of the 9th day. Subsequently, the length of roots (RL) for each variety was measured with a ruler and then the average was taken. Fresh cotyledon weight (FCW), cotyledons were weighed at the end of the 9th day. Dry cotyledon weight (DCW), after determining the fresh cotyledon weight, was dried at 105°C for 24 hours and weighed on a precision scale to determine the root dry weight. Relative water content (RWC) = (Fresh Weight−Dry Weight) (Turgid Weight−Dry Weight)] ×10

JMP statistical package program was used for variance analysis of the data of the features examined in the experiment and the differences between the averages were determined with the LSD (5% and 1%) test.

Table 1

Table 1 Analysis of Variance for the Effect of Different Priming Treatments (Control, Fe2O3NPs, Graphene Oxit, Potassium Humate) on Some Morphological and Germination Parameters of Soybean Seeds Under Two Conditions (Non-Salinity and Salinity)

		Calculated F Value							
Source of Variance	**df**	**GP**	**RL**	**HL**	**FRW**	**FHW**	**DRW**	**DHW**	**RWC**
Conditions (C)	1	102.2022**	52.1158**	1882.425**	32.9146**	797.6367**	0.7885ns	243.8445**	112.3386**
Treatments (T)	3	14.1271**	5.6293**	56.1826**	8.2726**	43.2292**	59.6920**	12.9060**	27.0305**
CxT	3	0.7248ns	1.7653ns	5.2467*	6.4983**	39.1893**	4.0925*	4.4214*	3.4247*

Error	16	10.919	8.0159	2.894	0.000641	0.03845	7.095e-7	0.000204	1.3874
$LSD_{0,05\ (C)}$		2.84	2.42	1.455	0.0211	0.1688	Ns	0.0105	1.0128
$LSD_{0,05\ (T)}$		4.009	3.44	2.068	0.0295	0.2384	0.00084	0.0168	1.4340
$LSD_{0,05\ (CxT)}$		ns	ns	2.911	0.0422	0.3376	0.0014	0.0244	2.0277

GP: Germination percentage, FHW: Fresh weight of hypocotyl, DHW: Dry weight of hypocotyl, FRW: Fresh weight of roots, DRW: Dry weight of roots, RL: Length of roots, FCW: fresh cotyledon weight, DCW: Dry cotyledon weight, RWC: Relative water content, HL: length of hypocotyl. Treatments; Graphene oxide, Distilled water, K-humate, Iron oxide. (The mean square of error was given)

Table 2

Table 2 The Effects of Different Priming Treatments (Control, Fe2O3NPs, Graphene Oxit, Potassium Humate) on Some Morphological and Germination Parameters of Soybean Seeds Under Two Conditions (Non-Salinity and Salinity)

Priming Treatments	Control (non-salinity)								NaCl (salinity 3 dS m^{-1})							
	GP (%)	RL (mm)	HL (mm)	FRW (g)	FHW (g)	DRW (g)	DHW (g)	RWC (%)	GP (%)	RL (mm)	HL (mm)	FRW (g)	FHW (g)	DRW (g)	DHW (g)	RWC (%)
Control	65.41	26.53	51.45	0.05	0.91	0.002	0.13	15.30	50.80	15.78	23.51	0.03	0.12	0.00	0.05	7.58
Graphene oxide NPs	73.71	28.10	58.85	0.07	2.64	0.00	0.17	16.41	59.59	17.95	29.09	0.04	0.17	0.01	0.10	12.03
K-humate	73.03	27.25	60.84	0.10	3.07	0.01	0.20	16.24	62.73	23.29	32.76	0.04	0.15	0.01	0.07	11.68
Iron oxide NPs	77.19	31.74	67.01	0.17	3.07	0.01	0.18	19.40	61.67	23.23	32.27	0.04	0.20	0.01	0.08	15.67

Conditions (C), Priming Treatments (T)

The mean values of length of hypocotyl ranged between 23.51-67.01 mm. The highest mean (67.01 mm) was obtained from iron oxide NPs treatment under non-salinity conditions, while the lowest mean (23.51 mm) was obtained from control treatment under salinity conditions. Positive effects of graphene oxide NPs, K-humate and iron oxide NPs were observed under saline conditions.

3. RESULTS AND DISCUSSIONS

ANOVA analysis results and LSD values are presented in Table 1. While Conditions*Treatments interaction was found significant on Length of hypocotyl, Fresh weight of roots, Dry weight of roots, Dry weight of hypocotyl, Relative water content traits, it was found insignificant on Germination percentage, Length of roots traits. However, the effect of conditions on all traits except Dry weight of roots was found significant. The effect of treatments on all traits examined was found significant. Poor seed quality caused the germination rate to be lower than expected. Germination rate was found to be higher under salt-free conditions.

Table 2 shows the mean values of the examined traits. Germination percentage values ranged between 65.41- 77.19% under unsaline conditions and 50.80-62.73% under saline conditions. However, it was determined that priming treatment was effective on germination percentage and the effect of Iron oxide NPs had the highest (77.19%) germination percentage under saline conditions. Although germination percentage decreased under saline conditions, it was observed that K-humate and iron oxide NPs were effective under saline conditions and increased the germination percentage. Nanoparticles can activate anti-stress activities in plants Taran et al.

(2016). As a result of priming applications, it was found that the breakdown of storage substances in the seed and activation of enzymes promoted germination and seedling development Pour et al. (2021), Basaran et al. (2019).

The germination-promoting effect of priming is reported to be associated with a variety of biochemical, cellular, and molecular events, including increased enzyme and respiratory activity, RNA, DNA, and protein synthesis Bray (1995). In mung bean, a faster seedling emergence resulting from priming treatments resulted in up to 45% increase in total yield Rashid et al. (2004).

Length of roots averages ranged between 26.53-31.74 mm under non-saline conditions and 15.78-23.29 mm under saline conditions. The highest root length (31.74 mm) was obtained from the iron oxide NPs treatment under saline conditions, while the lowest root length (15.78 mm) was obtained from the control treatment under saline conditions. K-humate and iron oxide NPs were found to be effective and increased root length under saline conditions. In previous studies, it was observed that root length values increased with priming Doğrusöz et al. (2022), Pour et al. (2021) and Hao et al. (2016). Under stress conditions, rapid root and shoot emergence as a result of priming application allows stronger seedling development, drought resistance increases, plants flower in a shorter time, reach harvest maturity and yield increases Lee-suskoon et al. (1998).

The mean values of hypocotyl length ranged between 23.51-67.01 mm. The highest mean (67.01 mm) was obtained from iron oxide NPs treatment under non-saline conditions, while the lowest mean (23.51 mm) was obtained from control treatment under saline conditions. The positive effect of graphene oxide NPs, K-humate and iron oxide NPs was observed under saline conditions. Seedlings emerging from primed seeds show rapid and vigorous root development Danneberger et al. (1992).

The mean values of hypocotyl fresh weight ranged between 0.03-0.17 g. The highest value (0.17 g) was obtained from iron oxide NPs treatment under saline conditions, while the lowest value (0.03 g) was measured from the control treatment under saline conditions. It was revealed that K-humate and iron oxide NPs treatment increased hypocotyl free weight under saline conditions.

The average values of fresh weight of hypocotyl vary between 0.91-0.20 g. While the highest value (3.07 g) was obtained from iron oxide NPs and K-humate applications under non-salinity conditions, the lowest value (0.12 g) was measured from the control application under saline conditions. It has been demonstrated that the application of K-humate and iron oxide NPs under salt-free conditions increases the fresh weight of hypocotyl. Soybeans treated with iron oxide nanoparticles increase in chlorophyll levels without traces of toxicity. This results in It was concluded that it may have an impact on the biochemical and enzymatic efficiency of various reactions of photosynthesis Ghafariyan et al. (2013).

The average values of dry weight of roots vary between 0.00-0.01 g. It has been measured that the effect of conditions on this is not significant, only the effect of the applications is significant. It has been observed that out-of-control practices have a significant positive impact.

The average values of dry weight of hypocotyl vary between 0.05-0.20 g. While the highest value (0.20 g) was obtained from the K-humate application under non-salinity conditions, the lowest value (0.05 g) was measured from the control application under saline conditions. It has been demonstrated that the application of graphene oxide NPs, K-humate and iron oxide NPs under salt-free conditions increases the dry weight of hypocotyl. Under salty conditions, the highest value was obtained from graphene oxide NPs application.

The mean values of relative water content ranged between 7.58-19.40. The highest value (19.40) was obtained from iron oxide NPs treatment under non-salinity conditions, while the lowest value (7.58) was measured from the control treatment under saline conditions. It was revealed that iron oxide NPs application increased the Relative water content under both conditions.

4. CONCLUSION

High yield in crop production depends on the germination and emergence of the seed used in a short time and at a high rate. In order to eliminate the problems that may occur during germination and emergence depending on both environmental and genetic factors and seed structure, and to obtain sufficient plant emergence and yield, seeds are generally subjected to various applications called priming before sowing. Priming increases the rate and speed of germination and emergence in many plant species, especially in unfavourable conditions such as salinity, and allows the desired plant emergence to be achieved in a short time. Rapid root and shoot emergence as a result of priming allows for stronger seedling development and increases yield. On the other hand, there are many factors affecting the success of priming applications. Therefore, knowing the advantages and disadvantages of priming application techniques; in addition to this, revealing the factors affecting priming applications will contribute to the planning and execution of priming application studies in a healthier way.

CONFLICT OF INTERESTS

None.

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