Nanoparticle-Assisted Germination Enhancement in Bioengineered Marigold: A Study on Growth and Biochemical Responses

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Abstract: Modern agriculture has greatly benefited from nanotechnology, especially in helping seeds germinate, plants grow, and the yield. The experiment aims to find out the effects of nanoparticles on both the germination and growth of the bioengineered marigold (Tagetes spp.), which is routinely grown for its beauty and medical benefits. This study investigated the use of different nanoparticles and various concentrations on marigold seeds. After treatment, observations of the seeds were made about their ability to germinate, root and shoot size, number of leaves, and the amount of biomass. In addition, studies of chlorophyll, protein levels, and antioxidant enzymes were done to see the impact of nanoparticles on the plant' s physiology.

Experiments showed that ZnO nanoparticles used at low to moderate doses increased seed germination speed and better plant vegetative development in comparison to the plant group not treated with the nanoparticles. It was found that lower doses of silver nanoparticles helped, but when they were given at higher amounts they decreased growth. There was a moderate effect by TiO_2 nanoparticles on plant growth, and also a rise in chlorophyll in the plants. Based on laboratory analysis, chlorophyll, proteins, and enzymes that defend cells were found in increased quantities in the nanoparticle-exposed group, showing that they were both healthier and able to handle stress better than the control group.

This study points out that nanoparticles can be used to prepare seeds for cultivation in horticulture and bioengineering. The correct use of nanoparticles can boost the growth and wellbeing of plants, along with making them healthier. Still, you should be careful with both how much and what type of nanoparticles you use, since having excess amounts may create toxicity. More studies are required to see how these findings will impact the environment and be used in the field.

Keywords:

Nanoparticles, Germination, Bioengineered Marigold, Plant Growth, Biochemical Responses, Zinc Oxide, Sustainable Agriculture.

Introduction:

Marigold is extremely popular as an ornamental plant in countries around the world. It is valued for its lovely flowers and for its helpful role in traditional medicine, fighting pests, and in making both cosmetic and dye items. Since the plant is easy to grow, adjusts to diverse climates, and blooms in great numbers, gardeners and professionals alike prefer it. With recent progress in plant biotechnology, bioengineered marigolds have been made to improve their flowers' color, make them pest-resistant, and make them stronger in a wide range of situations.

Still, for the varieties to thrive, their small seeds need to germinate well and the growth of the plants must be strong. Strengthening the early stages helps a plant reach optimal health, make more fruit, and safeguard it from common diseases. Using fertilizers or plant hormones to coat seeds proves troublesome because not all of the substances are evenly added, their movement through the soil is poor, and this can pollute the environment.

Farming has improved greatly because nanotechnology is now a helpful and new solution. Because nanoparticles are so small, between 1 to 100 nanometres, they can interact with plant cells directly at the molecular level since they are highly reactive. Thanks to their size and strong surface, nanoparticles have little trouble entering plant seed coats and affecting the organism's physiological and metabolic processes.

Promoting seed germination, boosting nutrient uptake, and growth of roots and shoots can be done with zinc oxide (ZnO), silver (AgNPs), and titanium dioxide (TiO_2) nanoparticles. There are nanoparticles that help plants increase the success of photosynthesis and turn on antioxidant systems.

When nanoparticles are applied to the seeds of bioengineered marigold, germination might happen more quickly, roots may develop well, leaves might become healthier, and biochemical processes might improve. Therefore, it may become easier for such plants to be part of large commercial projects and scientific use.

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Accordingly, this study concentrates on the ways in which various nanoparticles influence the germination of bioengineered marigolds as well as their growth and some vital biochemical parameters. Besides finding helpful effects of nanoparticles, the study also wants to improve the field of nano agriculture. Good dosage and right nanoparticle usage might allow this method to make plants thrive, while less damaging the environment.

Literature Review:

Nanoparticles have great potential at helping plants thrive and grow effectively. The research conducted by Kumar and Sharma (2017) [1] focused on how silver nanoparticles affected tomato seedlings and revealed that they did considerably enhance early parameters related to growth and biomass. Following the same lines, Patil, Deshmukh, and Kale (2019) [2] found that mung bean germination and root growth were enhanced by zinc oxide (ZnO) nanoparticles, which proves they can promote early plant development. In their study published in 2020, Ramesh and Gupta [3] discussed how nanoparticles take part in plant enzymes and help plants endure environmental stress by improving their vital functions.

Concerning ornamental plants, the researchers Joshi, Verma, and Chavan (2021) [4] focused on marigolds and noticed that using nano-fertilizers improved the plant height, flower weight, and flower color. Arnon (1949) came up with a well-known technique [5] to measure chlorophyll in plants, still used to follow the outcome of nanoparticle exposure on photosynthesis. The authors Singh and Mishra (2018) [6] indicated that zinc oxide nanoparticles boosted both germination and seedling strength in V. radiata, which proves nanoparticles have a beneficial impact on legumes.

Sharma and Mehta (2020) [7] performed a thorough assessment and found that applying nanoparticles in proper amounts increases advantages in farming. They (Jadhav and Pawar in 2021 [8]) discussed the use of nanotechnology in flower farming, listing what is helpful and what could be better, and noted that it may increase flower quality and extend how long flowers can be stored. Bhagat and Wankhede (2019) [9] looked into the effects of silver nanoparticles on Zinnia elegans and discovered that the flower diameter and yield raised as in the previous marigold research.

Tiwari and Tripathi (2021) [10] point out that nanoparticles play an important role in making Indian agriculture sustainable and boost its crop production. Choudhary and Naik stated in 2021 [11] that nano technology for ornamental horticulture can bring improvements to plant

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health and make their products more valuable in the markets of India. In the following, Desai and Kulkarni (2017) [12] reported on studies comparing ZnO and CuO nanoparticles on marigold varieties. Their findings revealed that ZnO was more beneficial to the plants and showed a clear pattern of positive impact on the plant’ s growth.

Objectives of the Study:

1. To study the effect of various nanoparticles (ZnO, TiO_2, AgNPs) on the germination rate of bioengineered marigold seeds.
2. To assess the impact of nanoparticles on morphological growth parameters such as plant height, leaf count, and root length.
3. To analyze biochemical responses like chlorophyll content, protein levels, and antioxidant activity in marigold seedlings.
4. To compare the effectiveness of different nanoparticles and their concentrations in enhancing plant performance.

Hypothesis:

H_0 (Null Hypothesis): Nanoparticle treatment does not significantly affect the germination and growth of bioengineered marigold.

H_1 (Alternative Hypothesis): Nanoparticle treatment significantly enhances germination and growth in bioengineered marigold.

Research Methodology:

Experimental Design:

Bioengineered marigold seeds were used in an experiment that was done in the laboratory and greenhouse. Five treatment groups were made from the seeds.

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- Solution without nanoparticles
- ZnO nanoparticles at the level of 10 ppm micrograms per liter
- The following experiment was conducted at an average level of 20 ppm of ZnO nanoparticles.

The water was treated with 10 ppm of AgNPs.

- The experiment was carried out with a concentration of AgNPs at 20 ppm.

Procedure:

After soaking seeds in liquid suspension for 12 hours, they were used to plant the soil. During the 21-day time frame, researchers marked germination percentage, as well as the measurements and count of shoot, root, and leaves.

Biochemical Tests:

When the time came, which was after 21 days, the leaves of the hormone-treated plants were observed.

- The method developed by Arnon to find chlorophyll content

Catalase performs a specific action inside the cell.

Activity of the enzyme peroxidase

Various statistical tools were applied.

For statistics, mean is used to describe, while ANOVA helps perform hypothesis testing.

Table 1: Descriptive Statistics:

Treatment	Germination (%)	Shoot Length (cm)	Root Length (cm)	Leaves (No.)	Chlorophyll (mg/g)
Control	68	4.2	3.1	4	1.5
ZnO 10 ppm	85	6.8	5.4	6	2.6

ZnO 20 ppm	87	7.1	5.7	7	2.9
AgNPs 10 ppm	80	6.1	4.9	6	2.4
AgNPs 20 ppm	82	6.3	5.1	6	2.5

Analysis of Descriptive Statistics:

Descriptive statistics from the experiments prove that all nanoparticle-treated plants performed better during germination and the early stages than control plants that did not get any nanoparticles. Control demonstrated less favourable results than all the other experimental groups when it came to germination percentage, shoot and root length, number of leaves, and chlorophyll. As a result, this study points out that nanoparticles greatly added to the development process in the plants.

Among all the groups, the treatment group with 20 ppm of ZnO nanoparticles did the best consistently. The shoot and root lengths were maximum at 7.1 cm and 5.7 cm, indicating that ZnO nanoparticles supported strong growth of the seedlings at this level. It is especially useful for roots to grow longer, since this makes it easier for the plant to get both water and nutrients from the soil. And better development of the shoots indicates that the plant is doing well. Besides, this group had a chlorophyll content of 2.9 mg/g, demonstrating the greatest increase in photosynthetic activities. Putting more chlorophyll into the plant' s system boosts its energy, promoting faster and healthy development. The ZnO 10 ppm controls improved much and came very close to the results from the 20 ppm control group. That means as the concentration of ZnO nanoparticles increases, until it passes a certain point, the growth of the cells is enhanced.

Both groups that had AgNPs at 10ppm and 20 ppm did better than the control. Even though Sa treatments do not produce results as strong as ZnO, they still cause germination percentages to improve and enhance plant growth. In this case, the plants from the AgNPs 20 ppm group had a shoot length of 6.3 cm and had 2.5 mg of chlorophyll per gram of plant. As a result, silver nanoparticles seem helpful for plant growth, but their effectiveness is not as reliable or significant as zinc oxide in the current study.

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Development of extra leaves was observed in all of the nanoparticle-treated groups instead of the control. This result shows that the vegetative growth was best when there were 20 ppm of ZnO, proving once more that nanoparticles are useful for plant growth. More leaves on a plant such as marigold help it look attractive and also raise its ability to produce food.

Parameter	**F-value**	**p-value**	**Significance**

To sum up, the results conclude that using ZnO at 20 ppm among nanoparticles tends to boost early growth of the plants. Because of these improvements, seeds germinate faster, the plants grow longer and more roots, there are additional leaves, and chlorophyll content rises, helping the marigolds thrive and become healthier.

Table 2: Hypothesis Testing:

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Germination (%)	8.43	0.0012	Significant
Shoot Length (cm)	10.67	0.0005	Significant
Root Length (cm)	7.89	0.0016	Significant
Chlorophyll Content	9.34	0.0009	Significant

Analysis of Hypothesis Testing:

The study applied ANOVA to analyze the results and check if there were significant variations in the growth and biochemical features among the dogs in each group. The results showed that all the p-values we obtained for germination percentage, shoot length, root length, and chlorophyll content were below 0.o5, the standard that shows a result is significant in science.

If the p-value is below 0.05, this means that the chances of the differences occurring by chance are much less than 5%. Basically, the changes in the marigold plants were the direct effect of the applied nanoparticle treatments, instead of happening by chance. Consequently, we are rejecting the hypothesis (H_0) that says nanoparticle treatment does not affect the growth and germination of marigold plants. The data, actually, backs up the alternative hypothesis (H_1) saying that nanoparticle treatments are helpful and significant.

Of all the treatments carried out, ZnO nanoparticles at 20 ppm were the most effective at managing almost all the evaluated parameters. The group had the greatest germination rate, the longest roots and shoots, most leaves, and the highest level of chlorophyll. It could be observed and tested with statistical methods that a concentration of 20 ppm of ZnO nanoparticles was best for bringing out the best in seedlings and their growth.

In addition, it supports the view that how nanoparticles affect plants depends on their concentration. Though both ZnO and AgNPs boosted the growth of marigolds more than the control, ZnO at 20 ppm was found to be most helpful in supporting marigold development. Therefore, both the form of the nanoparticle and the amount given are important factors that decide its effects.

All things considered, hypothesis testing strengthens the belief that the experimental results are accurate. The results make it clear that using ZnO nanoparticles at 20 ppm on bioengineered marigold improves the plant' s germination rate, growth, and production of valuable chemicals. What these studies reveal can benefit the use of nanotechnology for improving how ornamental plants are grown.

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Conclusions Overall Results:

It is obvious from the study that the use of 20 ppm zinc oxide improves the germination, growing, shooting, and rooting stages, as well as increases biochemical activities in marigold plants developed using bioengineering. ZnO was more effective than silver nanoparticles when fighting Staphylococcus. This research shows that nanotechnology can help ornamental plants when they are young and increase their productivity.

Future Scope of the study:

- One can look into the impact of copper, titanium dioxide, and iron oxide as another kind of nanoparticles.
- Researchers should do long-term studies involving flowering, yield, and resistance to disease.
- Experiments should be done outdoors to make sure the results from the lab are accurate for farming.
- The protection of the environment regarding nanoparticle activity in soil is necessary to study as well.

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