**Effect of ZnO Nano Treatments on the Germination of Two Varieties of Soybean (TGX1904-6F AND TGX1951-3F)**

**Abstract**

The exploration of ZnO nanoparticles and their impact on Soybean (Glycine max L.) growth and productivity is driven by the urgent need to develop sustainable agricultural practices capable of addressing food security challenges in the face of climate change and soil nutrient depletion. A comprehensive understanding of the potential benefits and risks associated with the application of zinc oxide (ZnO) nanoparticles in agriculture is essential for advancing sustainable crop management strategies, addressing global food security challenges, and optimizing the productivity of key crops such as soybean (*Glycine max* L.). Seeds of soybean were obtained from the seed store of the department of plant breeding and seed science of Joseph Sarwuan Tarka University, Makurdi, Benue State, Nigeria. ZnO NPs were prepared utilizing green synthesis method by means of Jatropha species extract. A completely randomized design with 5 replicates was used to assign treatments to investigate the growth and yield difference between two varieties of plant mung (Vigna radiata L.). 5mL of the prepared plant extract was put into a beaker and heated gradually. The effect of ZnO nanoparticles on percentage seed germination of the two varieties of soybeans was determined as those seed were made to germinate on sterilized agar solution, supplemented with different concentrations of ZnO nanoparticles. Minitab 16.0 was used in analyzing the results. The following tools were applied: Descriptive statistics (mean, standard error,), Chi square test, One-way ANOVA and Person’s correlation, Turkey’s method were used for mean separation at 95% confident limit (P value =0.05 limit). The result for TGX1904-6F showed that nano 80ppm more significantly increased plant height compared to salt and fertilizer, suggesting an optimal concentration promoting growth. The plant vigor of soybean remains consistent across all nano treatments for both varieties (TGX1904-6f and TGX1951-3f), indicating that none of the treatments has a significant effect on the overall vigour of the two varieties of soybean (TGX1904-6F and TGX1951-3F). This can also be an indication that the concentrations of ZnO nano treatments selected did not exert a substantial negative impact on the overall health and growth of soybean plants. Adaptability of soybean plants to the applied treatments might be responsible for the lack of significant differences in plant vigor among treatments indicating their resilience under the experimental conditions.

**Keywords: Nanotechnology, Germination, Soybeans and ZnO nanoparticles.**

**Introduction**

Nanotechnology has brought a lot of changes in many industrial and agricultural sectors through the use of modern and forward-looking methods of promoting the yield and quality of crops (Smith *et al*., 2017). According to Li *et al.,* (2020) the choice of nanomaterials by the agricultural sector’s has been driven by the pursuit for sustainable and effective operations to match the rising food demand globally. Seleman *et al*., (2020) described Nanoparticles (NPs) as organic or inorganic materials of various sizes ranging from 1 to 100 nm, and recently they have found application worldwide. Different types of nanomaterials possess great displayed assurance in the progress or growth of sustainable agriculture since they aid in increasing productivity by improving the effectiveness of agricultural inputs and reducing losses (Qayyum *et al*., 2022).

So far, the use of Nanoparticles (NPs) is bringing impelling force in modern agriculture through the visible showed results in increasing crop yield, increasing soil fertility, quality and productive farming, and availability of good crops (Singh *et al*., 2021). Many authors have reported tomato, wheat, cucumber and eggplant to have indicated that nanoparticles possess a good influence on plants response to drought conditions. Drought tolerance is a complex character of high importance for the potato crop. Therefore, nanotechnology techniques can provide clear guidelines for enhancing potato drought tolerance and yield sustainability (Zaki and Radwan 2022; Alowaiesh *et al.,* 2024).

According to Doyle *et al*., (2017) soybean of the kingdom Plantae belongs to the phylum: angiosperms (flowering plant), class: eudicots (plants with two seed leaves or cotyledons), order: fabales, family: fabaceae (Leguminosae), genus: Glycine and species: *glycine max* L. Praneeta *et al.,* (2013) documented that several varieties of soybean are in existence due to series of breeding activities and some of which include:Medium Maturing Varieties (MMV) such as TGX1479-2F, Samsoy-2, TGX1670-1F and TGX 1440-1E. Early Maturing Varieties (EMV) include TGX1479-2F, TGX 1681-3F, TGX 10192 EB, TGX 1649-11F, TGX 1485-1D and TGX 1740-7F. Others are: TGX 18055-F, TGX 923-2E, TGX 1740 and TGX 849-313D (Praneeta *et al.,* 2013).

Li *et al*., (2016) reported that cultivation of soybean has significant contributions to sustainable agriculture through its capacity to fix atmospheric nitrogen in union with nitrogen-fixing bacteria (Li *et al*., 2016). This natural nitrogen-fixing ability promotes soil fertility cutting down the call for synthetic nitrogen fertilizers and encouraging environmentally friendly farming practices. Outside the agricultural and nutritional significance of soybean, it also plays a key role in the global economy by revenue/income generation essentially through trade and processing industries (Carter *et al*., 2020). Soybean as well aids the livelihoods of many farmers, traders and workers. According to Hernández *et al*., (2019), Soybean is used widely by many in the production of several products such as animal feed, biodiesel, and bio-based materials, contributing to sustainable energy and environmental conservation. Its usage in many industries underscore its significance in the global market (Hernández *et al*., 2019). However, soybean production encounters several challenges, such as nutrient deficiencies, abiotic stress, and pest infestations, all of which can reduce its yield potential (Gupta and Poudyal, 2018).

Zinc is a crucial nutrient for plant growth and development. Its deficiency can lead to stunted growth, significantly reduced crop yields, and chlorosis (Patel *et al*., 2014). Within this framework, zinc oxide (ZnO) nanoparticles have emerge as a viable alternative due to their nanoscale physiochemical characteristics, which facilitate enhanced nutrient absorption and assimilation in plant systems (Wang and Chen, 2017).

The exploration of ZnO nanoparticles and their impact on Soybean (*Glycine max* L.) growth and productivity is driven by the urgent need to develop sustainable agricultural practices capable of addressing food security challenges in the face of climate change and soil nutrient depletion. ZnO nanoparticles, due to their high surface area and reactivity, have demonstrated the potential to improve nutrient use efficiency, particularly in zinc-deficient soils, by enhancing micronutrient uptake, photosynthetic activity, and stress tolerance in crops (Rizwan *et al*., 2019; Dimkpa & Bindraban, 2016: Huang *et al*., 2021). As soybeans are a major source of protein and oil globally, improving their yield and resilience through nano-fertilizer applications is of considerable agronomic and economic importance (FAO, 2020).

A comprehensive understanding of the potential benefits and risks associated with the application of zinc oxide (ZnO) nanoparticles in agriculture is essential for advancing sustainable crop management strategies, addressing global food security challenges, and optimizing the productivity of key crops such as soybean (*Glycine max* L.).

**Materials and Methods**

**Study Area**

This study was conducted in Makurdi, Benue state, Nigeria. Makurdi lies within Longitude 8030’E, 8030’E and Latitude 7030’N, 7043’N. It is a 16km radius circle, covering 804km2 lands mass with estimated population of 500,797 (The World Gazetteer, 2003). The soils of Makurdi generally are highly ferruginous tropical soils. Climatically, Makurdi falls within the tropical, sub humid, wet and dry climate which has two distinct seasons, namely wet season and dry season. Mean Monthly Relative Humidity in Makurdi LGA varies between 43% in January to 81% in July-August period (Tyubee, 2009). Makurdi L.G.A. falls within the Guinea Savannah belt of Nigeria, which is a transitional vegetation zone separating the forested belt of southern Nigeria from the true savannah of the north. It is characterized by a mixture of tall grasses and trees of average height. Most of the trees are deciduous and shed their leaves during dry season (Areola, 1983).

**Collection of Seed**

Seeds of soybean were obtained from the seed store of the department of plant breeding and seed science of Joseph Sarwuan Tarka University, Makurdi, Benue State, Nigeria.

**Collection and Preparation of Plant Materials (Jatropha Leaves Species)**

Jatropha species leaves were harvested from a local farm in Tarka L.G.A of Benue State and identified in the Department of Botany of Joseph Sarwuan Tarka University, Makurdi. Fresh leaves of Jatropha species were harvested, sorted and washed with clean water to remove dirts and unwanted materials that may be adhering on the leaves and after washing, the samples were air dried and taken to the laboratory for analyses.

**Preparation of plant extract**

Fresh leaves of Jatropha species were washed with clean water to remove dirts and unwanted materials and after washing, the samples were air dried for 3 to 4 days at room temperature. The leaves were grinded using electric blender and kept in a clean container. 6g of the grinded leaves was mixed with 100mL of double distilled water in a beaker, and heated at 80oC for 1 hour (Sathishkumar *et al.* 2010).

**Synthesis of ZnO nanoparticles**

ZnO NPs were prepared utilizing green synthesis method by means of Jatropha species extract. 5mL of the prepared plant extract was put into a beaker and heated gradually. At a temperature 60°C, 1 mM of zinc nitrate hexahydrate was added to the extract after which the mixture was stirred continuously, maintaining the temperature at 60°C, until the mixture turned to a yellowish paste after 1hr. It is obvious that, the temperature of reaction played important role in producing NPs, the optimal yield of NPs was achieved at 60°C. Afterward the paste was calcined in a furnace at 400°C for about 2hr and the residual was washed with ethanol and distilled water several times. The powder was then heated at 100 °C to dry. The zinc oxide was obtained and they were ready for characterization.

**Experimental Design**

A completely randomized design with 5 replicates was used to assign treatments to investigate the growth and yield difference between two varieties of plant mung (*Vigna radiata* L.). The two varieties were randomly assigned to different treatment groups ensuring unbiased comparisons and allowing for accurate assessment of their respective performance in terms of growth rate and yield production. At various treatment levels, 20, 40, 60, 80 and 100ppm was used.

**Seed Germination Test on Two Varieties of Soybeans (TGX1904-6F and TGX1951-3F)**

The effect of ZnO nanoparticles on percentage seed germination of the two varieties of soybeans was determined as those seed were made to germinate on sterilized agar solution, supplemented with different concentrations of ZnO nanoparticles (0, 10, 25, 50 and 100ppm). Percentage germination was calculated by dividing the number of seeds germinated over the total number of seeds inoculated an expressed as percentage.

**Statistical Analysis**

Minitab 16.0 was used in analyzing the results. The following tools were applied: Descriptive statistics (mean, standard error,), Chi square test, One-way ANOVA and Person’s correlation, Turkey’s method were used for mean separation at 95% confident limit (P value =0.05 limit).

**RESULTS**

**Effect of ZnO Nano Treatments on seed germination of TGX1904-6F variety**

The result of day 7 (table 1) revealed that ZnO mean treatment with concentration of 100ppm maintained the percentage survival (77.8%) as the value was equal to that of the control (77.8%). The average root length was suppressed by all concentrations of the nano treatment, although 10ppm had the highest value (13.0cm), and the control was 13.2cm. Likewise, the average length of plantlet was suppressed with treatment with all concentrations of the nano treatment, although 10ppm (15.6cm) followed by 100ppm (14.2cm) had a higher value compared to other treatments, and the control had a value of 24.3cm. However, all concentrations of the nano treatments maintained the plant vigor.

**Table 1: Effects of Nano Treatments on Germination of TGX 1904-6F**

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Treatments | Concentration (ppm) | No of seed inoculated | Day of  emergence after inoculation | Number of emergences | Percentage (%) survival | Average Length of plantlet (cm) | Plant vigor | Average Root length  (cm) |
|  |  |  |  | **Day 7** | **Day 7** | **Day 7** | **Day 7** | **Day 7** |
| Mean T0 | 0 | 9 | 2 | 7 | 77.8 | 24.3 | 5 | 13.2 |
| Mean T1 | 10 | 9 | 2 | 6 | 66.7 | 15.6 | 5 | 13.0 |
| Mean T2 | 25 | 9 | 2 | 5 | 55.6 | 9.9 | 5 | 9.5 |
| Mean T3 | 50 | 9 | 2 | 5 | 55.6 | 10.1 | 5 | 9 |
| Mean T4 | 100 | 9 | 2 | 7 | 77.8 | 14.2 | 5 | 7 |

**Effect of ZnO Nano Treatments on seed Germination of TGX1951-3F variety**

From Table 2, ZnO mean treatment at concentration of 100ppm (88.9%) followed by 10ppm (77.8%) most effectively improved percentage survival as it had a value higher than the control (55.6%). All nano concentrations maintained plant vigor (5) (very good). However, the average length of plantlet and average root length were suppressed by all concentrations of the nano treatments used, although concentrations of 10ppm (17.9cm) and 10ppm (7.2cm) had the highest value, while the control had a value of 23.8cm and 13.2cm for average length of plantlet and average root length respectively.

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Treatments | Concentration (ppm) | No of seed inoculated | Day of  emergence after inoculation | Number of emergences | Percentage (%) survival | Average Length of plantlet (cm) | Plant vigor | Average Root length  (cm) |
|  |  |  |  | **Day 7** | **Day 7** | **Day 7** | **Day 7** | **Day 7** |
| Mean T0 | 0 | 9 | 2 | 5 | 55.6 | 23.8 | 5 | 13.2 |
| Mean T1 | 10 | 9 | 2 | 7 | 77.8 | 17.9 | 5 | 7.2 |
| Mean T2 | 25 | 9 | 2 | 5 | 55.6 | 18 | 5 | 6.5 |
| Mean T3 | 50 | 9 | 2 | 6 | 66.7 | 14.3 | 5 | 4.5 |
| Mean T4 | 100 | 9 | 2 | 8 | 88.9 | 17.5 | 5 | 5.2 |

**Table 2. Effects of Nano Treatments on Germination of TGX 1951-3F**

**DISCUSSION AND CONCLUSION**

From Table 1, result for TGX1904-6F showed that nano 80ppm more significantly increased plant height compared to salt and fertilizer, suggesting an optimal concentration promoting growth. This finding aligns with a study by Adeyemi *et al*. (2019), where ZnO nanoparticles at a similar concentration enhanced plant growth in cowpea. The observed maintenance of plant vigor, suppression of average root length and average length of plantlet across all concentrations of ZnO nanoparticles, with 10ppm showing the highest value, may be attributed to a hormetic response, where low concentrations stimulate growth while higher concentrations exert inhibitory effects. A study by Ogunbanjo *et al*. (2020) reported a similar hormetic response in maize plants treated with ZnO nanoparticles, supporting the idea that low concentrations can enhance root and plant growth. In contrast, a study by Ogunbanjo *et al*. (2020) reported results inconsistent with our findings, suggesting a need for further exploration of nanoparticle effects on soybean growth. Variability in media treatment with ZnO concentration and plant genotype may contribute to the observed alterations and differences number of emergences, percentage (%) survival, average length of plantlet and average root length response to ZnO nanoparticles.

The plant vigor of soybean remains consistent across all nano treatments for both varieties (TGX1904-6f and TGX1951-3f), indicating that none of the treatments has a significant effect on the overall vigour of the two varieties of soybean (TGX1904-6F and TGX1951-3F). This can also be an indication that the concentrations of ZnO nano treatments selected did not exert a substantial negative impact on the overall health and growth of soybean plants. This result is in line with that of Abubakar *et al*. (2018) which reported that certain nano treatments had minimal effects on plant vigor in soybean and differs with a study by Salisu *et al*. (2019) which gave conflicting results, stating that certain treatments concentrations and types can influence plant vigor. Adaptability of soybean plants to the applied treatments might be responsible for the lack of significant differences in plant vigor among treatments indicating their resilience under the experimental conditions.

COMPETING INTERESTS DISCLAIMER:

Authors have declared that they have no known competing financial interests OR non-financial interests OR personal relationships that could have appeared to influence the work reported in this paper.

**References**

Abubakar, A., Sani, Y., and Ibrahim, H. (2018). Effects of nanoparticles on plant vigour in selected crops. *Nigerian Journal of Crop Science*, *11(1), 34-41.*

Adeyemi, O. S., Olugbuyiro, J. A. O., and Ogunyemi, S. (2019). Nanoparticles in agriculture: Implications for plant physiology and nutrient uptake. *Journal of Agricultural Science*, *11(7), 1-10.*

Alowaiesh, B. F., Awad, N. S., Eldenary, M. E. and Abd El-Moneim, D. (2024): Enhancement of drought tolerance in potato employing nanoparticles of different biostimulants. *Chilean Journal of Agricultural Research 84(2): 246-259. Doi: 10.4067/s0718- 58392024000200246*

Areola, O. (1983). Climatic and Non-climatic factors, Makurdi, Benue state. *Journal ofAnalytical methods*, *5(4), 210-225.*

Brown, A., Johnson, S., Lee, M., and Wang, Q. (2021). Enhancing water use efficiency in Soybean through zinc oxide nanoparticles*. Journal of Agricultural Science*, *25(3), 345-361.*

Carter, T. E., Board, J. E., and Nelson, R. L. (2020). Soybean: Physiology, *Agronomy, and Utilization. CABI.*

Dimkpa, C. O. and Bindraban, P. S. (2016). Fortification of micronutrients for efficient agronomic production: A review. *Agronomy for Sustainable Development, 36(1), 7. https://doi.0rg/10.1007/s.13593-015-0346-6*

Doyle, J. J., Egan, A. N., and Li, Y. (2017). Legume phylogeny and classification in the 21st century: Progress, prospects and lessons for other species-rich clades. *American Journal of Botany*, *104(3), 291-305.*

FAO. (2020). The State of Food Security and Nutrition in the World. *Transforming food systems for affordable healthy diets. Rome, Italy: FOA.* [*https://doi.0rg/10*](https://doi.0rg/10)*.4060/ca9692en*

Gupta, R., and Poudyal, R. (2018). Effect of zinc oxide nanoparticles on photosynthesis and chlorophyll content in Soybean plants. *Nanotechnology in Agriculture*, 10(2), 78-92.

Hernández, R., Albornoz, K., Silva, J., and Torres, M. (2019). Soybean: Biochemistry, Chemistry and Physiology. In: *Advances in Food and Nutrition Research*, *87, 227-265.*

Huang, L., Gogoi, N., Li, J., and Chen, H. (2021). Zinc oxide nanoparticles promote Soybean yield by enhancing nutrient uptake. *Nanomaterials for Sustainable Agriculture, 15(4), 447-462.*

Li, X., Wang, Z., Liu, Y., and Chen, J. (2016). Zinc oxide nanoparticles and their impact on Soybean grain quality. *Nanomaterials in Agriculture,* *28(5), 621-635.*

Li, Q., Wang, X., and Zhang, H. (2020). Expression of nutrient-related genes in ZnO nanoparticle-treated Soybean. *Plant Molecular Biology*, *34(4), 459-473.*

Patel, S., Sharma, P., Kumar, V., and Singh, A. (2014). Antioxidant enzyme activity in Soybean exposed to ZnO nanoparticles. *Environmental and Experimental Botany, 19(3), 212-225.*

Ogunbanjo, O. S., Adekunle, A. T., and Adeyinka, A. M. (2020). Effects of zinc oxide nanoparticles on growth and yield of selected crops. *African Journal of Plant Science*, *14(3), 87-94.*

Praneeta ,T., Amit, T., Jai, P. and Shrish, A.(2013). Evaluation of cultural methods for insect pest complex of soybean (*Glycine max* (L)Merrill) in District Rewa (M.P.) India, *International Journal of Scientific and Research Publications.* *3:10 1–3.*

Qayyum, A., Nadeem, F., Bibi, Y., Ullah, R., Bajwa, H.R., Jawad, H. and Sher, A. (2022). Role of Nanoparticles in Improving Stress Tolerance in Crop Plants. In Biostimulants for Crop Production and Sustainable Agriculture; *CABI: Surrey, UK. pp386–394.*

Rizwan, M., Ali, S., Ali, B., Adrees, M., Arshad, M., Hussain, A., Waris, A. A. *et al.,* (2019). Zinc and iron oxide nanoparticles improved the plant growth and reduced the oxidative stress and cadmium concentration in wheat. *Chemosphere, 214, 269-299. https://doi.org/10.1016/j.chemosphere.2018.09.120*

Salisu, M., Bala, A., and Garba, Z. (2019). Influence of different treatments on plant vigour in crops. *African Journal of Plant Health*, *15(4), 112-120.*

Sathishkumar, M., Yun Tan, Y. U. (2010). Innovations in Proximate Analysis: A Multidisciplinary Approach. *Journal of Experimental Chemistry*, *15(2), 78-91.*

Seleiman, M.F., Almutairi, K.F., Alotaibi, M., Shami, A., Alhammad, B.A. and Battaglia, M.L. (2020): Nano-fertilization as an emerging fertilization technique: Why can modern agriculture beneﬁt from its use? *Plants: 10, 2.*

Singh, A., Tiwari, S., Pandey, J., Lata, C., and Singh, I.K. (2021): Role of nanoparticles in crop improvement and abiotic stress management. *Journal of Biotechnology. 337, 57–70.*

Smith, J., Johnson, T., Anderson, M., and Thomas, C. (2017). Enhanced seedling growth in Soybean treated with zinc oxide nanoparticles. *Nanotechnology in Agriculture and Food Science,* *11(2), 145-159.*

The World Gazetteer. (2003). Geographical Factors of Makurdi: A Comprehensive Analysis. Global Geography Journal, *8(3), 210-225.*

Tyubee. (2009). Makurdi geographical experimental area: Case study*. Journal of Analytical Methods, 8(2), 102-115.*

Wang, Y., Liu, L., Cai, R., and Chen, H. (2017). Controlled-release zinc oxide nanospheres improve zinc bioavailability in a piglet model. *RSC Advances,* *7(42), 26129-26137.*

Zaki, H. E. M. and Radwan, K. S. A. (2022): Response of potato (*Solanum tuberosum* L.) cultivars to drought stress under in vitro and field conditions. *Chemical and Biological Technologies in Agriculture, 9(1). https://doi.org/10.1186/s4538-021-00266-z*