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GROWTH AND YIELD RESPONSES OF SOYBEAN (*Glycine max* L.) TO ZINC OXIDE (ZnO) NANOPARTICLES FOLIAR APPLICATION

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Aim. This study was purposed to investigate the effects of zinc oxide nanoparticles (ZnO NPs) on the growth and yield performance of two soybean (*Glycine max* L.) varieties, TGX1904-6F and TGX1951-3F, under controlled experimental conditions.

Materials and methods. Zinc oxide nanoparticles were synthesized and characterized following standard protocols, and their effects were evaluated across five treatment levels (20, 40, 60, 80, and 100 ppm) in a completely randomized design with five replicates. Growth parameters, including plant height, leaf morphology, stem diameter, and branch number, were assessed alongside phenological and yield traits such as days to flowering, flower production, pod metrics, and seed weights.

The *results* revealed significant improvements in plant growth and yield metrics at intermediate ZnO NP concentrations, with enhancements observed in plant height, branch number, pod weight, and seed yield. Specifically, 60 ppm ZnO NP treatment resulted in the highest branch production, while 40 and 80 ppm treatments significantly promoted floral and pod development. Conversely, higher concentrations (100 ppm) exhibited inhibitory effects on plant height and leaf morphology, suggesting potential toxicity at elevated ZnO NP levels. Statistical analyses, including one-way ANOVA and Pearson's correlation, confirmed significant treatment effects ($P \leq 0.05$) on growth and yield parameters, highlighting the critical role of dose optimization.

Conclusions. The findings underscore the potential of ZnO NPs as a novel agricultural supplement to enhance soybean productivity while emphasizing the need for balanced application to mitigate toxicity risks. This study contributes valuable insights into sustainable farming practices, leveraging nanotechnology to optimize crop performance and address global food security challenges.

Key words: Zinc oxide nanoparticles, soybean, *Glycine max*, growth, yield.

Soybean is a globally significant crop, but its role in Africa is especially noteworthy due to its potential to address food security and improve rural livelihoods. In Sub-Saharan Africa, soybean is an important crop, with countries like Nigeria, Tanzania, and Uganda leading its production. The crop thrives across various agroecological zones, from savannas to highlands and coastal regions, making it highly adaptable to the continent's diverse climates. Recent advancements, such as the use of rhizobium bacteria inoculation, have significantly boosted soybean yields, particularly in Africa, where enhancing agricultural productivity is crucial [1]. While countries in South America, the United States, and Asia dominate global soybean production, Africa's expanding cultivation of this crop reflects its growing importance. Soybean not only offers a valuable source of protein and oil but also plays a key role in improving soil fertility, making it a vital part of sustainable farming systems across the continent [2].

Soybean's chemical composition makes it an excellent alternative to animal protein, thanks to its high protein and carbohydrate content coupled with low cholesterol levels. Beyond its direct use in food, soybean by-products are essential for animal feed, supporting broader agricultural systems. One of the most critical products of soybean is its oil, which is extracted using solvents such as hexane. The oil primarily consists of triglycerides, with minor components like phospholipids, tocopherols, and phytosterols, though these compounds are reduced during typical oil processing. Soybean oil is one of the leading edible oils globally, used in a wide array of food products, including salad dressings, margarine, and cooking oils. Nutritionally, it is rich in essential fatty acids, containing about 53% linoleic acid and 8% linolenic acid in its unhydrogenated form. Even after partial hydrogenation, it retains significant levels of linoleic acid (23%) and linolenic acid (3%). These qualities make soybean oil a highly beneficial and versatile oil, contributing both to human nutrition and the global food industry [3].

Metal and metal oxide nanoparticles, including those containing silver, gold, copper, and zinc, are increasingly recognized for their significant potential in agriculture. These nanoparticles offer remarkable opportunities to enhance crop productivity, improve stress tolerance, and increase nutrient-use efficiency. Studies have shown that the application of such nanoparticles can increase yields by up to 20%, reduce disease incidence by 50%, decrease nutrient leaching by 30%, and boost soil carbon sequestration by 15%. Moreover, these metal oxide nanoparticles act as signaling molecules, modulating various biological processes such as stress response, nutrient uptake, and hormone regulation, ultimately promoting plant health and productivity [4].

Among these metal oxide nanoparticles, zinc oxide nanoparticles (ZnO NPs) are one of the most widely utilized due to their dual role as both a micronutrient source and a stimulant for essential physiological and biochemical processes in plants. ZnO NPs positively influence growth, photosynthesis, and antioxidative defense systems. However, while ZnO NPs can promote plant health, they also have inhibitory effects on root elongation, especially at higher concentrations. For example, ZnO NPs have been shown to reduce root growth in crops like maize significantly (*Zea mays*), with more pronounced effects during the seed incubation phase as compared to the soaking stage. This indicates that ZnO NPs, depending on their concentration, can play a crucial role in determining plant health during critical growth stages [5].

At lower concentrations, ZnO NPs stimulate growth and enhance photosynthetic activity in various crops. For instance, in maize and tomatoes, ZnO NPs, in conjunction with arbuscular mycorrhizal fungi, have been shown to boost nutrient uptake, thereby improving plant resilience and overall productivity [6]. Additionally, ZnO NPs enhance a plant's antioxidative defense system, which helps mitigate oxidative stress induced by environmental challenges such as drought, salinity, and heavy metals. In tomato plants, the application of ZnO NPs increased the activities of antioxidative enzymes like superoxide dismutase (SOD) and catalase (CAT), which are vital for neutralizing reactive oxygen species (ROS). This enhancement of the plant's defense mechanisms enables greater tolerance to stress, thereby promoting growth and productivity even under suboptimal conditions [7].

In specific crops, the benefits of ZnO NPs are even more pronounced. For example, in *Gossypium hirsutum* (cotton), bioengineered ZnO NPs significantly improved growth characteristics, with plant growth increasing by 125.4% and biomass by 132.8%. Additionally, exposure to ZnO NPs led to higher chlorophyll and carotenoid content, further supporting robust plant development [8]. Similarly, in maize, biogenic ZnO NPs stimulated shoot and root biomass production while enhancing photosynthetic pigments like chlorophyll and carotenoids, which are essential for healthy plant growth [9]. In lettuce, ZnO NPs at an optimal concentration of 10 mg/kg enhanced the net photosynthetic rate, resulting in increased biomass compared to untreated plants [10]. In tomato plants, lower concentrations of ZnO NPs improved growth, chlorophyll fluorescence, and photosynthetic efficiency, further reinforcing the benefits of nanoparticle application [7].

ZnO NPs also strengthen plants' ability to combat oxidative stress. In *Pleioblastus pygmaeus*, for instance, ZnO NPs improved antioxidative defense systems, reducing heavy metal accumulation and enhancing the plant's overall tolerance to environmental stress [11]. Wheat treated with ZnO NPs under cadmium contamination showed reduced oxidative stress and an increase in SOD and peroxidase activities, enabling the plants to withstand heavy metal-induced stress more effectively [12]. Additionally, ZnO NPs enhanced zinc bioavailability in wheat, increasing zinc concentrations in roots and shoots while reducing cadmium uptake. This not only improved plant health but also mitigated toxicity risks [13]. ZnO NPs have been shown to enhance secondary metabolite production in medicinal plants. In *Stevia rebaudiana*, ZnO NPs doubled the production of steviol glycosides, a valuable compound for the sweetener industry, while also improving the plant's antioxidative response [14].

A significant challenge for soybean production is poor growth and yield, which traditional organic and inorganic fertilizers have been unable to address fully. These fertilizers not only pose environmental and health risks but are also expensive and often in limited supply. In response, metal oxide nanofertilizers, particularly zinc oxide (ZnO) nanoparticles, have emerged as effective, eco-friendly alternatives for enhancing soybean productivity.

This study set out to explore the transformative impact of zinc oxide (ZnO) nanoparticles, alongside zinc nitrate and traditional fertilizer, on two soybean (*Glycine max* L.) varieties. It focused on how these treatments influence not only vegetative growth and development but also the ultimate yield and productivity of the crop.

Materials and Methods

A. Study Area

This study was carried out in Makurdi metropolis of Benue state. Makurdi is a town within Longitude 8°30′E, 8°30′E and Latitude 7°30′N, 7°43′N. It is a 16 km radius circle, covering 804 km^2 land mass. Makurdi has an estimated population of 500,797 (The World Gazetteer, 2003). Being situated in the Lower Benue Valley, the relief of the Local Government Area (LGA) is generally low, with heights ranging between 73 meters and 167 meters above sea level. The soils of Makurdi typically 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. The wet season starts in April and lasts till October, while the dry season starts in November and lasts till March. Rainfall ranges from 775 millimeters to 1792 millimeters, with a mean annual value of 1190 millimeters. Mean Monthly Relative Humidity in Makurdi LGA varies between 43% in January to 81% in the July-August period [15]. Makurdi LGA. falls within the Guinea Savannah belt of Nigeria. The Guinea Savannah belt is a transitional vegetation zone separating the forested belt of southern Nigeria from the actual savannah of the north. A mixture of tall grasses and trees of average height characterizes it. Most of the trees are deciduous and shed their leaves during the dry season [16].

B. Materials

The materials used in the study included soybean seeds of two varieties, TGX1904-6F and TGX1951-3F, synthesized and characterized Zinc oxide nanoparticles as reported by Fayomi et al. and colleagues [17]. The ZnO NPs, the light-yellow powder with an average crystallite size of 62.49 nm, were characterized using UV/VIS, FTIR, SEM, and XRD methods. All glassware was thoroughly washed with deionized water and oven-dried before use. Deionized water was employed for all homogenization processes.

C. Collection of Seeds and Soil Samples

Soybean seeds were procured from the seed stores of the Department of Plant Breeding and Seed Science at Joseph Sarwuan Tarka University. Surface soil samples were collected from fallow land within the botanical garden of the Department of Botany at the same university. The soil samples were air-dried and sieved using a 2 mm sieve to eliminate pebbles and visible root fragments. A total of approximately 25 kg of prepared soil was used to fill forty pots for the study.

D. Experimental Design and Planting

The experiment was conducted using a completely randomized design with five replicates to evaluate the growth and yield differences between two soybean varieties (*Glycine max* L.). The two varieties were randomly assigned to treatment groups to ensure unbiased comparisons and accurate assessments of growth rates and yield production. Treatments were applied at various levels of 20, 40, 60, 80, and 100 ppm.

On September 1, 2023, four soybean seeds were manually sown at a depth of 3 cm in each pot. Following seedling establishment, the plants were thinned to two per pot to maintain uniformity.

E. Growth Parameter Determination

The growth parameters of soybean plants were assessed to evaluate their development under different treatment conditions. Plant height was measured at maturity by recording the size of five randomly selected plants from the soil surface to the top of the main stem, with results expressed in centimeters. The number of plants in each pot was determined by counting the emerging and surviving plants for each treatment, which provided insight into germination and survival rates.

Leaf production was evaluated by counting the total number of leaves per plant in each pot. To further assess leaf morphology, leaf length was measured as the distance from the base to the apex, while leaf width was measured as the distance between the two sides of the leaf lamina, excluding the petiole. Stem diameter was determined by measuring the widest part of the stem using a ruler, indicating stem robustness.

Finally, the number of branches per plant was recorded by counting the branches for each plant in every pot. These growth parameters were critical for understanding the morphological differences and responses of the soybean varieties under the applied treatment levels. Similar approaches to measuring plant growth have been reported in studies of soybeans and other crops.

F. Phenological and Yield Parameter Determination

The phenological parameters of soybean plants were evaluated to understand their reproductive development. The number of flowers per plant in each pot was counted to determine flower production. Days to flowering were recorded by counting the number of days from planting until the appearance of flowers for each plant, providing an indication of the reproductive onset.

Yield parameters were assessed to evaluate productivity. The number of pods or fruits per plant was determined by counting the pods produced by each plant in each pot. Pod length was measured using a ruler to determine the distance from the base to the tip of the pod, excluding the stalk or peduncle. To assess seed production, the number of seeds per pod was determined by counting the seeds within each pod.

Further yield assessments included measurements of pod and seed weights. Total pod weight was obtained by using a weighing scale to measure the collective weight of all pods harvested from a plant, while total seed weight was similarly determined by weighing the total seeds collected from each plant. These measurements provide critical insights into the yield potential of soybean varieties under different treatment conditions, aligning with methodologies commonly employed in crop yield studies.

G. Statistical Analysis

Minitab 16.0 was used in analyzing the results. The following tools were applied: Descriptive statistics (mean, standard error,), One-way ANOVA, and Person's correlation) Turkey's method was used to carry out the mean of separation at a 95% confidence limit (P value = 0.05 limit).

Result and Discussion

A. Effect of ZnO NP on the Growth Parameters

The application of zinc oxide nanoparticles (ZnO NPs) has emerged as an innovative approach to enhance the growth and yield of soybean (*Glycine max* L.), addressing both productivity and sustainability in agricultural practices. The findings from Tables 1 and 2 provide a comprehensive analysis of the effects of ZnO NPs on the growth parameters of two soybean varieties, TGX1904-6F and TGX1951-3F.

The control treatments served as a benchmark for assessing the natural growth potential of the soybean varieties. TGX1904-6F exhibited robust vertical growth with moderate branching and stem diameter, consistent with the notion that untreated conditions can support adequate growth in nutrient-sufficient soils [18]. Similarly, TGX1951-3F demonstrated comparable plant height (13.02 ± 1.73 cm) and structural stability, indicating the natural adaptability of these varieties. Fertilizer treatments enhanced stem robustness (1.54 ± 0.09 cm for TGX1951-3F) and leaf production, reflecting improvements in photosynthetic potential due to improved nutrient availability [19] have been commonly used in agriculture, and have attracted more attention for researchers. In this study, a 2-year experiment was conducted involving two Zn types (ZnO NPs and $ZnSO_4$. However, a notable reduction in plant height under fertilizer treatments underscores the limited effectiveness of conventional fertilizers in promoting vertical growth without micronutrient supplementation.

$ZnSO_4$ treatments exhibited moderate improvements in growth metrics, such as increased leaf width (3.76 ± 0.15 cm for TGX1951-3F) and stem diameter. These findings affirm the essential role of zinc in supporting structural and metabolic processes, including cell wall biosynthesis and antioxidative enzyme activities [20]. Despite these structural benefits, plant height under $ZnSO_4$ treatments remained below control levels, suggesting that while zinc supplementation aids metabolic stability, it may not fully address growth demands such as elongation and branching.

The effects of ZnO NP treatments revealed a concentration-dependent relationship with soybean growth parameters. Low concentrations (20 ppm) were particularly effective in maintaining natural growth dynamics, as seen in TGX1904-6F with plant height (12.38 ± 2.27 cm) and branch count comparable to the control. These findings align with reports highlighting the stimulatory effects of low ZnO NP concentrations on seedling vigor and metabolic efficiency [21]. Intermediate concentrations (40–60 ppm) demonstrated the most significant benefits, with TGX1951-3F producing the highest number of branches (7.00 ± 2.55) at 60 ppm. This enhancement is likely attributable to ZnO NPs' role in modulating hormonal pathways, particularly auxin and cytokinin, which are critical for lateral growth [22].

Higher ZnO NP concentrations (80–100 ppm) presented a dual effect. While stem diameter peaked, other parameters, such as plant height and leaf morphology, were adversely impacted, reflecting potential phytotoxicity. Excessive ZnO NPs are known to disrupt cellular homeostasis and induce oxidative stress, inhibiting growth and compromising overall plant performance [23] Zn oxide nanoparticles (ZnONPs).

The statistical analysis demonstrated significant effects of ZnO NPs on growth parameters, including plant height, stem diameter, and branch count, with $P \leq 0.05$. Post-hoc comparisons using least significant difference (LSD) values revealed distinct groupings, indicated by the labels "a," "b," "c," and "d," which represent statistically significant differences among treatments. Groups labeled "a" had the highest or statistically similar values, while "b," "c," and "d" denoted progressively lower values. These findings emphasize the reproducibility of observed trends and highlight the necessity of dose optimization to maximize the benefits of ZnO NPs while minimizing potential toxicity [24].

Effect of ZnO NP on the Yield Parameters

The reproductive performance of soybean (*Glycine max L.*) varieties TGX1904-6F and TGX1951-3F under various treatments, including control, fertilizer, zinc sulfate ($ZnSO_4$), and zinc oxide nanoparticles (ZnO NPs), reveals significant insights into the yield-related impacts of these inputs. Tables 3 and 4 detail how these treatments influenced parameters such as flower production, days to flowering, pod characteristics, and seed weight, underscoring the potential of ZnO NPs as a sustainable agricultural intervention.

The control treatments established benchmarks for reproductive metrics in both soybean varieties, with TGX1904-6F producing an average of 5 flowers, flowering at 55 days, and yielding pod and seed weights of 1.30 ± 0.07 g and 3.26 ± 0.36 g, respectively. Similarly, TGX1951-3F exhibited standard flowering (55.00 ± 4.95 days) and pod weights (1.30 ± 0.07 g) under untreated conditions. These observations align with studies demonstrating stable yet unimproved reproductive performance in nutrient-limited environments [25].

Fertilizer treatments accelerated flowering for both varieties (46.60 ± 1.52 days for TGX1904-6F and TGX1951-3F) and increased flower production (7.00 ± 0.71). Modest improvements in seed weights (3.58 ± 0.43 g for TGX1951-3F) suggest enhanced reproductive development driven by improved nutrient uptake and metabolic efficiency [26]. However, the limited enhancement in pod and seed weights indicates the need for micronutrient supplementation to optimize reproductive success.

$ZnSO_4$ treatments significantly improved flower production for both varieties (7.60 ± 1.34 for TGX1904-6F and TGX1951-3F) and modestly increased pod weight (1.66 ± 0.44 g for TGX1951-3F). These results underscore the pivotal role of zinc in reproductive processes, including enzyme activation, protein synthesis, and structural development [18]. However, the absence of significant gains in seed weight for TGX1904-6F indicates that $ZnSO_4$ alone may not optimize yield without precise application strategies tailored to the specific physiological requirements of the variety.

The ZnO NP treatments exhibited marked concentration-dependent effects on both varieties. Low concentrations (20 ppm) delayed flowering for TGX1904-6F (57.20 ± 1.30 days) and TGX1951-3F (57.20 ± 1.30 days) but improved seed weights slightly for TGX1904-6F (3.72 ± 0.33 g). This suggests that low ZnO NP concentrations prioritize seed quality over flowering intensity or pod weight, a trend consistent with findings linking ZnO NPs to improved nutrient allocation at lower doses [24].

Intermediate concentrations (40–80 ppm) produced the most significant improvements,

Table 1. **Effect of ZnO Nanoparticles Treatment on the Growth Parameters of Soybean TGX1904-6F**

Treatment	**No. of plants**	**No. of leaves**	**Leaf width (cm)**	**Plant height (cm)**	**Leaf length (cm)**	**No. of branches**	**Stem diameter (cm)**
Control	3.00 ± 0.00	7.60 ± 2.30	3.64 ± 0.65	13.02 ± 1.73^{a}	5.00 ± 0.71^{bc}	3.60 ± 0.89	0.98 ± 0.08^{d}
Fertilizer	2.20 ± 0.84	8.60 ± 1.14	3.56 ± 0.36	5.60 ± 0.55^{d}	3.00 ± 0.71^{d}	3.60 ± 0.89	1.54 ± 0.09^{ab}
$ZnSO_4$ Salt	2.40 ± 0.89	7.80 ± 1.10	3.76 ± 0.15	8.20 ± 0.45^{c}	3.96 ± 0.05^{cd}	4.00 ± 0.71	1.60 ± 0.20^{a}
ZnO NP 20 ppm	3.00 ± 0.00	8.40 ± 2.07	3.62 ± 0.16	12.38 ± 2.27^{a}	5.16 ± 0.88^{bc}	3.00 ± 0.00	1.32 ± 0.41^{bc}
ZnO NP 40 ppm	2.40 ± 0.55	6.60 ± 0.55	3.54 ± 0.51	12.80 ± 1.10^{a}	6.40 ± 2.30^{ab}	3.40 ± 1.14	0.92 ± 0.08^{d}
ZnO NP 60 ppm	2.80 ± 0.45	6.00 ± 1.58	3.78 ± 0.13	10.40 ± 1.52^{b}	7.00 ± 2.55^{a}	3.00 ± 0.71	1.18 ± 0.29^{cd}
ZnO NP 80 ppm	3.00 ± 0.00	8.40 ± 0.55	3.60 ± 0.34	8.00 ± 1.22^{c}	4.56 ± 0.52^{cd}	4.40 ± 0.55	1.58 ± 0.13^{ab}
ZnO NP 100 ppm	2.20 ± 0.84	7.80 ± 0.84	3.54 ± 0.51	8.60 ± 1.14^{c}	4.14 ± 0.48^{cd}	4.20 ± 0.84	1.68 ± 0.18^{a}
p-value	0.10	0.07	0.96	0.00	0.00	0.06	0.00
LSD				1.76	1.71		0.28

Table 2. **Effect of ZnO Nanoparticles treatment on the Growth Parameters of Soybean TGX1951-3F**

Treatment	**No. of plants**	**No. of leaves**	**Leaf width (cm)**	**Plant height (cm)**	**Leaf length (cm)**	**No. of branches**	**Stem diameter (cm)**
Control	3.00 ± 0.00	7.60 ± 2.30	3.64 ± 0.65	13.02 ± 1.73^{a}	5.00 ± 0.71^{bc}	3.60 ± 0.89	0.98 ± 0.08^{d}
Fertilizer	2.20 ± 0.84	8.60 ± 1.14	3.56 ± 0.36	5.60 ± 0.55^{d}	3.00 ± 0.71^{d}	3.60 ± 0.89	1.54 ± 0.09^{ab}
$ZnSO_4$ Salt	2.40 ± 0.89	7.80 ± 1.10	3.76 ± 0.15	8.20 ± 0.45^{c}	3.96 ± 0.05^{cd}	4.00 ± 0.71	1.60 ± 0.20^{a}
ZnO NP 20 ppm	3.00 ± 0.00	8.40 ± 2.07	3.62 ± 0.16	12.38 ± 2.27^{a}	5.16 ± 0.88^{bc}	3.00 ± 0.00	1.32 ± 0.41^{bc}
ZnO NP 40 ppm	2.40 ± 0.55	6.60 ± 0.55	3.54 ± 0.51	12.80 ± 1.10^{a}	6.40 ± 2.30^{ab}	3.40 ± 1.14	0.92 ± 0.08^{d}
ZnO NP 60 ppm	2.80 ± 0.45	6.00 ± 1.58	3.78 ± 0.13	10.40 ± 1.52^{b}	7.00 ± 2.55^{a}	3.00 ± 0.71	1.18 ± 0.29^{cd}
ZnO NP 80 ppm	3.00 ± 0.00	8.40 ± 0.55	3.60 ± 0.34	8.00 ± 1.22^{c}	4.56 ± 0.52^{cd}	4.40 ± 0.55	1.58 ± 0.13^{ab}
ZnO NP 100 ppm	2.20 ± 0.84	7.80 ± 0.84	3.54 ± 0.51	8.60 ± 1.14^{c}	4.14 ± 0.48^{cd}	4.20 ± 0.84	1.68 ± 0.18^{a}
p-value	0.10	0.07	0.96	0.00	0.00	0.06	0.00
LSD				1.76	1.71		0.28

with TGX1904-6F achieving the highest flower production (8.80 ± 1.64 at 80 ppm) and TGX1951-3F exhibiting enhanced pod weights (1.80 ± 0.28 g at 40 ppm). These results reflect the stimulatory effects of ZnO NPs on auxin and cytokinin regulation, which promote reproductive organ development and enhance yield potential [27].

At high concentrations (100 ppm), yield metrics declined significantly for both varieties, with TGX1904-6F showing reduced pod weight (0.92 ± 0.08 g) and TGX1951-3F

Table 3. **Effect of ZnO NPs treatment on the yield of TGX1904-6F**

Treatment	**No. of flowers**	**Days to flowering**	**Pods per plant**	**Pod length (g)**	**Seeds per pod**	**Pod weight (g)**	**Seed weight (g)**
Control	5.00 ± 0.71^{d}	55.00 ± 4.95^{ab}	5.00 ± 0.71	4.78 ± 0.19	3.20 ± 0.45^{a}	1.30 ± 0.07^{cd}	3.26 ± 0.36
Fertilizer	7.00 ± 0.71^{abcd}	46.60 ± 1.52^{d}	5.40 ± 0.89	4.92 ± 0.28	3.00 ± 0.00^{a}	1.50 ± 0.29^{bc}	3.58 ± 0.43
$ZnSO_4$ Salt	7.60 ± 1.34^{abc}	50.20 ± 4.38^{bcd}	5.20 ± 0.84	4.64 ± 0.11	2.20 ± 0.45^{b}	1.66 ± 0.44^{ab}	3.32 ± 0.43
ZnO NP 20 ppm	6.40 ± 1.34^{bcd}	57.20 ± 1.30^{a}	5.00 ± 1.41	4.58 ± 0.40	3.00 ± 0.00^{a}	1.14 ± 0.11^{de}	3.72 ± 0.33
ZnO NP 40 ppm	7.80 ± 3.11^{ab}	52.80 ± 6.22^{ab}	5.60 ± 0.89	4.58 ± 0.63	3.00 ± 0.00^{a}	1.80 ± 0.28^{a}	3.32 ± 0.54
ZnO NP 60 ppm	5.60 ± 1.95^{cd}	49.60 ± 5.73^{bcd}	4.40 ± 1.52	4.86 ± 0.19	3.00 ± 0.00^{a}	1.06 ± 0.17^{de}	3.50 ± 0.62
ZnO NP 80 ppm	8.80 ± 1.64^{a}	52.20 ± 4.92^{abc}	5.20 ± 0.45	4.68 ± 0.41	2.20 ± 0.45^{b}	1.48 ± 0.11^{bc}	3.26 ± 0.46
ZnO NP 100 ppm	7.60 ± 1.14^{abc}	47.20 ± 1.10^{cd}	4.40 ± 0.55	4.62 ± 0.70	3.00 ± 0.00^{a}	0.92 ± 0.08^{e}	3.64 ± 0.54
p-value	0.02	0.00	0.46	0.84	0.00	0.00	0.63
LSD	2.14	5.48			0.35	0.30	

Note: ($P < 0.05$) indicates a significant effect or difference.

Table 4. **Effect of ZnO Nano Treatment on the Yield of TGX1951-3F**

Treatment	**No. of flowers**	**Days to flowering**	**Pods per plant**	**Pod length (g)**	**Seeds per pod**	**Pod weight (g)**	**Seed weight (g)**
Control	5.00 ± 0.71^{d}	55.00 ± 4.95^{ab}	5.00 ± 0.71	4.78 ± 0.19	3.20 ± 0.45^{a}	1.30 ± 0.07^{cd}	3.26 ± 0.36
Fertilizer	7.00 ± 0.71^{abcd}	46.60 ± 1.52^{d}	5.40 ± 0.89	4.92 ± 0.28	3.00 ± 0.00^{a}	1.50 ± 0.29^{bc}	3.58 ± 0.43
$ZnSO_4$ Salt	7.60 ± 1.34^{abc}	50.20 ± 4.38^{bcd}	5.20 ± 0.84	4.64 ± 0.11	2.20 ± 0.45^{b}	1.66 ± 0.44^{ab}	3.32 ± 0.43
ZnO NP 20 ppm	6.40 ± 1.34^{bcd}	57.20 ± 1.30^{a}	5.00 ± 1.41	4.58 ± 0.40	3.00 ± 0.00^{a}	1.14 ± 0.11^{de}	3.72 ± 0.33
ZnO NP 40 ppm	7.80 ± 3.11^{ab}	52.80 ± 6.22^{ab}	5.60 ± 0.89	4.58 ± 0.63	3.00 ± 0.00^{a}	1.80 ± 0.28^{a}	3.32 ± 0.54
ZnO NP 60 ppm	5.60 ± 1.95^{cd}	49.60 ± 5.73^{bcd}	4.40 ± 1.52	4.86 ± 0.19	3.00 ± 0.00^{a}	1.06 ± 0.17^{de}	3.50 ± 0.62
ZnO NP 80 ppm	8.80 ± 1.64^{a}	52.20 ± 4.92^{abc}	5.20 ± 0.45	4.68 ± 0.41	2.20 ± 0.45^{b}	1.48 ± 0.11^{bc}	3.26 ± 0.46
ZnO NP 100 ppm	7.60 ± 1.14^{abc}	47.20 ± 1.10^{cd}	4.40 ± 0.55	4.62 ± 0.70	3.00 ± 0.00^{a}	0.92 ± 0.08^{e}	3.64 ± 0.54
p-value	0.02	0.00	0.46	0.84	0.00	0.00	0.63
LSD	2.14	5.48			0.35	0.30	

Note: ($P < 0.05$) indicates a significant effect or difference.

exhibiting diminished flower production. This trend aligns with reports that excessive ZnO NPs induce oxidative stress and cellular disruption, negatively impacting reproductive processes [28].

The statistical analyses confirmed the significant effects of ZnO NP treatments on flower production ($P \leq 0.02$), days to flowering ($P \leq 0.05$), and pod and seed weights ($P \leq 0.05$), validating the reproducibility of the results. The least significant difference (LSD) values further highlighted these variations, with labels “a,” “b,” “c,” and “d,” denoting statistically distinct groups. Groups labeled "a" had the highest or statistically similar values, while “b,” “c,” and “d,” represented progressively lower means. These findings underscore the importance of dose optimization to maximize the benefits of ZnO NPs while minimizing toxicity risks.

The use of zinc oxide nanoparticles (ZnO NPs) modulates key physiological and biochemical pathways, supporting the development of sustainable agricultural practices [18]. ZnO NPs positively influence soybean growth by enhancing photosynthetic efficiency, stem robustness, and branch formation, particularly at low to moderate concentrations (20–60 ppm). These effects are attributed to improved nutrient uptake and the regulation of key growth hormones, such as auxins and cytokinins, which drive lateral growth [27]. Foliar applications of ZnO NPs have shown significant improvements in seed yield and antioxidant enzyme activity [29]. These findings underscore the importance of tailoring ZnO NP applications to optimize benefits.

The efficacy of ZnO NPs is highly concentration-dependent. Low to moderate concentrations enhance growth metrics, while excessive concentrations (80–100 ppm) induce phytotoxicity, disrupting cellular homeostasis and increasing oxidative stress [30]. The morphological properties of ZnO NPs, such as particle size, further influence their impact. For example, spherical ZnO NPs (38 nm) demonstrate enhanced uptake efficiency and reduced toxicity compared to more extensive or irregularly shaped particles [28].

The reproductive performance of soybean varieties TGX1904-6F and TGX1951-3F highlights the transformative potential of ZnO NPs. Under control conditions, both varieties showed stable yet suboptimal reproductive metrics. Fertilizer treatments improved flower production and accelerated flowering, although gains in pod and seed weights remained modest, suggesting the need for micronutrient interventions [31]. $ZnSO_4$ treatments further improved flower production and pod weight, emphasizing zinc’s role in enzymatic and structural processes, but were insufficient to maximize seed weight without precise application strategies [32].

ZnO NPs exhibited significant concentration-dependent effects, with intermediate concentrations (40–80 ppm) achieving the highest flower production and pod weights. These benefits are linked to the stimulatory effects of ZnO NPs on reproductive hormone pathways, including auxin and cytokinin regulation [24]. However, high concentrations (100 ppm) caused oxidative stress, reducing reproductive success, consistent with studies demonstrating the negative impacts of nanoparticle overuse [33].

ZnO NPs enhance soybean resilience to abiotic stresses by improving antioxidant defenses and osmolyte accumulation, as demonstrated under drought and arsenic stress conditions [29]. Their integration with biofertilizers further amplifies benefits, enhancing nodulation and increasing unsaturated fatty acids in soybeans, which contribute to improved crop quality and yield [34].

Conclusions

This work showed the effects of zinc oxide (ZnO) nanoparticles on the growth and yield of soybean (*Glycine max* L.), providing valuable insights into the potential benefits of employing nanotechnology in agriculture. The findings demonstrate that ZnO nanoparticle applications can significantly enhance growth parameters such as plant height, leaf area, and overall yield metrics, establishing their potential as a viable agricultural supplement. These results are particularly encouraging in the context of increasing global food demands and the need for sustainable intensification of farming systems.

The concentration-dependent effects observed in this study underscore the importance of optimizing nanoparticle dosages to maximize benefits while minimizing potential toxicity or environmental risks. While the benefits of ZnO nanoparticles are apparent, further research is essential to unravel the precise biochemical and physiological mechanisms underlying these enhancements. Additionally, studies addressing the long-term environmental impacts of ZnO nanoparticles, including their interactions with soil microbiota, nutrient cycles, and non-target organisms, are imperative for ensuring safe and sustainable implementation.

Conflict of Interest

The authors declare that they have no conflict of interest.

Author Contributions

All the authors planned the work, contributed to the article's conception, manuscript article writing, and editing. They all contributed to seeing the experiment done. All authors contributed to the reading, revision, and approval of the submitted version.

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ВПЛИВ ЛИСТКОВОЇ ОБРОБКИ НАНОЧАСТКАМИ ОКСИДУ ЦИНКУ (ZnO) НА РІСТ І ВРОЖАЙНІСТЬ СОЇ (*Glycine max* L.)

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Мета. Дослідження було спрямовано на вивчення впливу наночасток оксиду цинку (ZnO NPs) на ріст та продуктивність двох сортів сої (*Glycine max* L.), TGX1904-6F та TGX1951-3F, за контрольованих експериментальних умов.

Матеріали та методи. Наночастки оксиду цинку були синтезовані та охарактеризовані відповідно до стандартних протоколів, а їхній вплив оцінювали на п'яти рівнях оброблення (20, 40, 60, 80 та 100 ppm) у повністю рандомізованому плануванні з п’ятьма повтореннями. Параметри росту, включаючи висоту рослин, морфологію листків, діаметр стебла та кількість гілок, аналізували разом із фенологічними та врожайними характеристиками, такими як кількість днів до цвітіння, утворення квітів, параметри бобів та вага насіння.

Результати. Результати показали значне покращення росту рослин та показників врожайності за середніх концентрацій наночасток ZnO. Спостерігали збільшення висоти рослин, кількості гілок, ваги бобів та врожайності насіння. Зокрема, оброблення 60 ppm ZnO NP забезпечило найбільшу кількість гілок, тоді як оброблення 40 та 80 ppm значно сприяло розвитку квітів та бобів. Водночас вищі концентрації (100 ppm) мали інгібувальний вплив на висоту рослин і морфологію листків, що вказує на можливу токсичність при підвищених рівнях ZnO NP. Статистичні аналізи, включаючи однофакторний дисперсійний аналіз (ANOVA) та кореляцію за Пірсоном, підтвердили значний вплив оброблень ($P \le 0.05$) на параметри росту та врожайності, підкреслюючи важливість оптимізації дозування. Отримані дані свідчать про перспективність використання наночасток ZnO як нового аграрного додатку для підвищення продуктивності сої, водночас акцентуючи на необхідності збалансованого застосування для зменшення ризиків токсичності.

Висовок. Це дслідження надає цінну інформіцію сталих агарних практик, використовуючи нанотехнології для оптимізації продуктивності культур та вирішення глобальних проблем продовольчої безпеки.

Ключові слова: наночастинки оксиду цинку, соя, *Glycine max*, ріст, урожайність.