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Effect of ZnO Nanoparticles enhanced compost with different application methods on nutrient uptake and Grain Yield of Rice (*Oryza sativa*) Var. Inpari 32

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Abstract. A pot experiment using a randomized block design was conducted to test the effect of ZnO-nanoparticles enhanced compost (ZnO-NEC) with different application methods on nutrient uptake and grain yield of rice var. Inpari 32. The treatment tested was ZnO-NEC with various concentrations (50, 100 and 150 mg.kg^{-1}) with direct soil and foliar application methods compared with control and inorganic fertilizer treatments. All treatments using ZnO-NEC fertilizer received 50% of the recommended dose of chemical fertilizer. The results showed that the application of ZnO-NEC 100-150 mg.kg^{-1} with foliar application had leaf greenness levels (SPAD Value) and N uptake that were the same as the use of inorganic fertilizer and significantly increased the uptake of P, K and Zn compared to the use of inorganic fertilizer. The ZnO-NEC 50 mg.kg^{-1} with direct soil application ZnO-NEC 100 and 150 mg.kg^{-1} with foliar application had the same high grain yield per hill as the inorganic treatment. These treatments had an average percentage of empty grains (9.14%) and grain moisture content of 27.10%, which was lower compared to the control and inorganic treatments. These findings provide new insight into the potential for using the technology of ZnO-nanoparticles enhanced compost to reduce the use of chemical fertilizers.

1 Introduction

Sustainable agriculture is the main focus in efforts to meet global food needs while minimizing negative impacts on the environment due to the overuse of synthetic chemical fertilizers over long periods [1, 2]. Degraded ecosystems have become a severe threat to human health and civilization. The benchmark for ecosystem degradation is related to the failure of the ecosystem to retain carbon and prevent the release of various forms of nitrogen from the soil into groundwater and the atmosphere. Several efforts have been made to increase organic matter in the soil through organic soil management and the use of alternative N fertilizers, which can increase the efficiency of N use in crop cultivation [3]. However, this

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effort cannot retain carbon for a long time due to the decomposition and mineralization process of the organic material [4, 5]. In this context, research on using innovative technologies to increase agricultural productivity is becoming increasingly important.

Nanotechnology is a technology that is emerging in this decade and can become a significant application in the agricultural sector [6, 7, 8]. This technology holds excellent hope in changing conventional agricultural practices into precision and sustainable agriculture by applying balanced techniques and practices to increase crop yields by monitoring environmental variables in each situation [9]. One interesting approach is using compost enriched with zinc oxide (ZnO) nanoparticles as a source of plant nutrition, which can potentially increase nutrient uptake and agricultural yields.

Rice (*Oryza sativa* L.) is an important food crop that requires adequate nutrition for optimal growth and yield. However, using synthetic chemical fertilizers in rice cultivation has raised concerns about its negative impact on soil health and the environment. As an alternative, the use of organic compost has been proposed as an environmentally friendly and sustainable solution. In this context, ZnO nanoparticles are attracting attention as potential additives in organic compost. Zn is needed for the growth and nutritional quality of cereals, which are the staple food in several developing countries in the world, and is essential for ensuring food security and healthy eating style [10, 11].

Zinc (Zn) is essential for improving the immune system and brain function in humans and also plays an important role in enzymatic reactions and metabolic activities in plant systems [12, 13]. Zinc is required in small concentrations but is essential for several key plant physiological pathways to function normally. In plants, zinc plays a vital role as a structural constituent or regulatory cofactor of various enzymes and proteins in many critical biochemical pathways, especially those related to carbohydrate metabolism, both in photosynthesis and in the conversion of sugar into starch (starch), protein metabolism, auxin metabolism (a substance growth regulator), pollen formation, maintenance of the integrity of biological membranes, and resistance to infection by certain pathogens [14].

A decrease in grain yield of up to 80% occurs due to reduced Zn levels in grain, especially in conditions of Zn deficiency [10]. This has serious implications for human health in countries dominated by cereal-based diets. Therefore, the availability of Zn needs to be increased so that the quantity and quality of crop production can be increased. Most applications of Zn are in the form of soluble Zn compounds such as $ZnCl_2$ and $ZnSO_4$. The application of this soluble Zn compound provides low Zinc Use Efficiency (ZUE) (<3.5%), which is caused by variations in soil Zn adsorption capacity [15]. Limiting factors such as pH, electrical conductivity, organic C content, lime/calcium content, and nutrient interactions also significantly reduce ZUE. Therefore, various sources, such as ZnO nanoparticles, can be used to increase the availability of Zn in the soil through Zn fertilization [16]. This research aimed to evaluate the effect of the application of compost enriched with ZnO nanoparticles with different application methods on nutrient uptake (N, P, K and Zn) and yield of rice var. Inpari 32. Considering the need for an environmentally friendly and sustainable agricultural approach, this research is expected to provide new insights into the potential use of this technology in increasing agricultural productivity while minimizing negative impacts on the environment.

2 Materials and Methods

2.1 Site and Time of Research

This research was a pot experiment carried out in a plastic house in Sidomulyo Village, Jabung, Malang Regency, from August to December 2023 with a geographical location of

7º95'68" South Latitude and 112º75'49" East Longitude. Composting was conducted in a field laboratory at the exact location, while soil and plant analysis was conducted in the Integrated Laboratory at the University of Islam Malang.

2.2 Research methodology

This research used a Randomized Block Design (RBD) consisting of eight treatments consisting of T0 (no fertilizer), T1: inorganic (NPK) fertilizer, T2: Compost+ZnO-NPs 50 mg kg^{-1} compost (ZnO-NEC 50 mg kg^{-1}), with direct application into the soil, T3: ZnO-NEC 50 mg kg^{-1} compost, with foliar spray application, T4: ZnO-NEC 100 mg kg^{-1} compost, with direct application into the soil, T5: ZnO-NEC 100 mg kg^{-1} compost, with foliar spray application, T6: ZnO-NEC 150 mg kg^{-1} compost, with direct application into the soil, and T7: ZnO-NEC 150 mg kg^{-1} compost, with foliar spray application. Each treatment was repeated five times and each replication had three plant samples.

The variables observed included N, P, K, and Zn nutrient uptake, leaf greenness levels (SPAD Value), total harvest dry weight and oven dry weight of rice plant biomass, number and length of panicles, weight per panicle, total panicle weight per clump, weight of grain per hill, oven dry weight of grain per hill, number of grains per panicle, number of empty grains per panicle, number of filled grains per panicle, and grain moisture content. The data collected was statistically analyzed for variance (F-test) at the 5% level. If the analysis results have a significant effect, continue with the 5% Tukey test to determine the differences between treatments using Minitab Vers.14.12. Microsoft Excel was employed to present the charts.

2.3 Experiment Procedure

Composting was done in a bin by preparing the compost material first. The materials used are cow dung, goat dung, spent mushroom waste, cocopeat, rice husk biochar, and rice bran in a ratio of 3:3:3:3:3:1 for these six ingredients. The organic materials are dried and ground to speed up the composting process. Next, these ingredients are mixed manually using a hoe until homogeneous. In the next stage, the compost material was put into the composting bin and added with a decomposer that was activated at a dose of 2.5 litres/500 kg. The decomposer solution is diluted with water and then sprinkled evenly over the surface of the organic matter until a moisture content of 40%. The composting process took one month. The produced compost was dried to a moisture content of 10% and ground into flour using a grinder mill. Next, the compost powder was mixed with Zn-NPs nanoparticles obtained using a top-down approach method from PT. Nanocenter Indonesia Global. The larger ZnO is milled into nanoscale particle size using ball milling [17]. The ZnO-NPs used in this research are sized 150 nm.

For the pot experiment, the soil samples with a clay content of 34.8 %, a silt content of 13.4%, and a sand content of 51.8%. The soils had middle soil pH (H2O) 6.4; low in organic C (1.4%) by Walkley and Black method; very low in total Kjeldahl N (0.085); high in P2O5 (18 mg kg^{-1} P) (Bray I), high in exchangeable K (0.62 cmol(+) kg^{-1}), middle in Zn (2.4 ppm) by extract DTPA and high in cation exchange capacity (32.28 cmol kg^{-1} soil). The soil samples were air-dried and ground. 120 plastic pots (diameter = 30 cm and height = 35 cm) were filled with 20 kg of dry soil. Soil samples were mixed in plastic pots. The compost dose for all ZnO-NEC treatments is the same at 100 g pot^{-1} (≈10 tons ha^{-1}). Meanwhile, the concentration of ZnO nanoparticles varies according to the treatment tested. The ZnO-NEC were added to the soil one day before planting at a rate of 100 g pot^{-1} with direct application into the soil. Two rice plants were planted in each pot. Meanwhile, for the foliar application treatment, 100 g of ZnO-NEC is divided into ten applications by dissolving 10 grams in 200 ml, then spraying it evenly on the leaves of the rice plants and the rest is poured into the planting medium. The foliar application was carried out every five days until the flowering phase. Inorganic fertilization was carried out one week after planting (WAP) for urea (100 kg ha^{-1}), superphosphate (100 kg ha^{-1}) and KCl (100 kg ha^{-1}). The second fertilization is carried out at 5 WAP only for urea fertilizer (100 kg ha^{-1}). Plant maintenance includes irrigation by maintaining a flooding height of 1 cm on the soil surface.

The nitrogen (N) content in the leaves of these crops was measured using a modified Kjeldahl's method [18]. The P and K were determined by the wet combustion method using HNO_3+$HClO_4$ solvent using a Flame photometer for K content and a UV-Spectrophotometer for P content [19]. All measurements of nutrient content in leaves were carried out 12 weeks after planting (WAP). Harvesting of rice was started at 12 WAP.

The rice plants were harvested at 17 weeks after planting WAP by cutting the panicles at each treatment first. Next, the plant is lifted slowly so the roots do not break, washed thoroughly with water, and then air-dried.

3 Results and Discussion

Treatments ZnO-NEC 50 dan 150 mg kg^{-1}, with direct application into the soil (T2, T6) and T7 (ZnO-NEC 150 mg kg^{-1} with foliar application) have the same leaf greenness level as treatment using inorganic fertilizer (T1). The N uptake of the treatment using inorganic fertilizer (T1) was the same as the T7 treatment. Treatments T3 (ZnO-NEC 50 mg kg^{-1}, with foliar application), T4 (ZnO-NEC 100 mg kg^{-1}, with direct application into the soil) and T5 (ZnO-NEC 100 mg kg^{-1} with foliar application) had higher P uptake than inorganic fertilizer treatment (T1). K and Zn uptake in treatments using inorganic fertilizer were the same as those using NEC. However, there was a tendency for T7 treatment to have higher Zn uptake than other treatments (Table 1).

Table 1. Leaf Greenness Level and N, P, K and Zn Nutrient Uptake due to ZnO Nanoparticles Enhanced Compost with Different Application Methods Compared to Control and Inorganic Fertilizer

Treatments	**Leaf Greeness Level (SPAD Value)**	**N uptake (g pot^{-1})**	**P uptake (g pot^{-1})**	**K uptake (g pot^{-1})**	**Zn uptake (mg pot^{-1})**
T0	37.78 a	2.098 a	0.147 a	0.015 a	1.351 a
T1	44.41 b	8.424 g	0.367 cd	0.041 bc	3.724 bc
T2	39.89 ab	5.362 bc	0.302 bc	0.037 bc	3.174 b
T3	39.04 a	5.887 cd	0.388 de	0.047 bc	3.568 bc
T4	38.77 a	6.429 de	0.388 de	0.048 bc	3.456 bc
T5	36.19 a	7.018 ef	0.436 e	0.051 c	3.569 bc
T6	40.24 ab	4.902 b	0.311 bc	0.033 bc	3.299 bc
T7	40.70 ab	7.826 fg	0.278 b	0.038 bc	3.815 c
HSD 5%	4.46	0.87	0.067	0.017	0.610

Remark: Means followed by different letters in the same column are statistically significantly different at HSD test, p = 0.05.

The results of this research showed that reducing the dose of NPK fertilizer by 50% and adding compost enriched with 150 mg ZnO nanoparticles per kg through foliar application was able to equal the nutrient uptake of N, K and Zn from inorganic fertilizer treatment. ZnO-NPs undergo quick dissolution/transformation upon release into the environment [20, 21]. This rapid change will accelerate the uptake of ZnO nutrients into plant tissue to play a role in plant metabolism. Zinc is an essential element in photosynthesis and protein synthesis. By improving the availability of zinc in the soil, ZnO nanoparticles can help rice plants absorb nutrients more efficiently, which supports vegetative growth and root development [22].

Moreover, zinc can be included in proteins and other macromolecules. Zinc is an element found in proteins and functions as a structural, functional, or regulatory cofactor for numerous enzymes [23]. A high Zn uptake in T7 treatment reduced plant P uptake (Table 1). P-Zn antagonism existed in the roots of the plants. If plants absorb Zn in high enough amounts, plant P uptake can be reduced [22, 24].

Table 2. The number, length, and weight of panicles due to ZnO Nanoparticles Enhanced Compost with Different Application Methods Compared to Control (T0) and Inorganic Fertilizer (T1).

Treatments	Observed parameter				
	number of panicles	length of panicles (cm)	weight per panicle (g)	panicles weight per hill (g)	number of grains per panicle
T0	14.11 a	19.43	3.79	46.30 A	116.62 a
T1	27.67 b	19.76	4.59	118.28 C	178.04 d
T2	25.00 b	21.30	4.52	94.30 b	155.60 bc
T3	24.78 b	21.26	4.52	90.11 b	157.93 bc
T4	25.67 b	20.54	4.16	90.96 b	141.85 b
T5	27.33 b	21.41	4.45	88.94 b	158.25 bc
T6	25.78 b	21.21	4.50	97.59 b	150.64 b
T7	25.44 b	20.90	4.46	111.40 C	170.26 cd
HSD 5%	3.01	ns	ns	14.67	15.20

Remark: Means followed by different letters in the same column are statistically significantly different at the HSD test, p = 0.05.

The inorganic fertilizer treatment (T1) had the same number of panicles as the ZnO-NEC treatment, and all panicles were higher than those of the control. All treatments had the same panicle length and weight per panicle, while the highest panicle weight per hill and the number of grains per panicle were found in treatment T7 (ZnO-NEC 150 mg kg^{-1}, with foliar application) (Table 2).

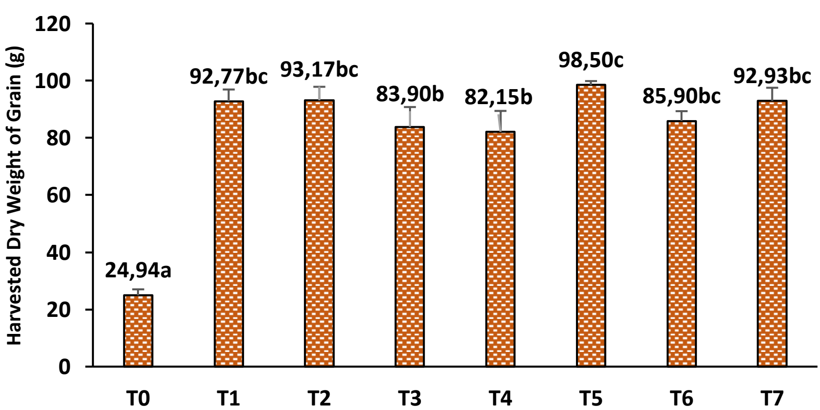

Figure 1. Effect of Various Treatments of Nanoparticles ZnO Enhanced with Different Application Methods on the Harvested Dry Weight of Grain Compost Compared to Control (no fertilizer, T0) and Inorganic Fertilizer (T1), Based on the Results of HSD Test (P<0.05). (Remark: HSD 5% =12.34)

Application of ZnO-NEC 50 and 150 mg kg^{-1} compost directly into the soil (T2 and T6) and foliar application of ZnO-NEC 100-150 mg kg^{-1} compost (T5 and T7) have yields of harvested dry grain weight which was high and statistically not significantly different from inorganic treatment (T1) (Fig. 1). Treatments T2, T3, T4, and T7 had the same total harvested dry plant weight as treatment T1 (Fig.2). However, the quality of the grain as measured by the percentage of filled grains and empty grains and the grain moisture content of the ZnO-NEC treatment was better than that of the inorganic fertilizer treatment and control (T0) (Fig. 3 and 4).

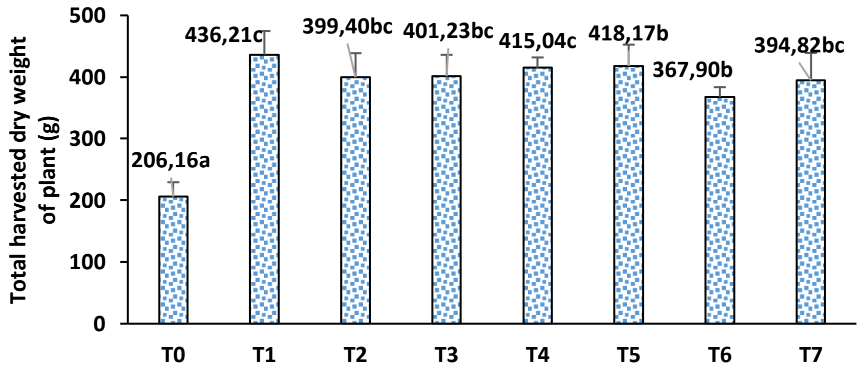

Figure 2. Effect of Various Treatments of Nanoparticles ZnO Enhanced Compost with Different Application Methods on the Total Harvested Dry Weight of Plant Compared to Control (no fertilizer (T0) and Inorganic Fertilizer (T1), Based on the Results of HSD Test (P<0.05). (Remark: HSD 5% =45.94).

ZnO-NPs can penetrate different plant tissues through the stomata and cuticles of plant leaves and root hair cells. This allows for enhancing plant growth and yield [25, 26]. For plants, zinc (Zn) is a vital mineral ingredient that is needed for several physiological functions, including the synthesis of proteins, chlorophyll, and carbohydrates [27, 28]. ZnO-NPs have several advantages over conventional ZnO fertilizers, including their small size, high specific surface area, ease of dissolution, and diffusion. These attributes enable plants to absorb Zn produced by ZnO-NPs quickly, satisfying their nutritional demands and stimulating growth and development [29, 30].

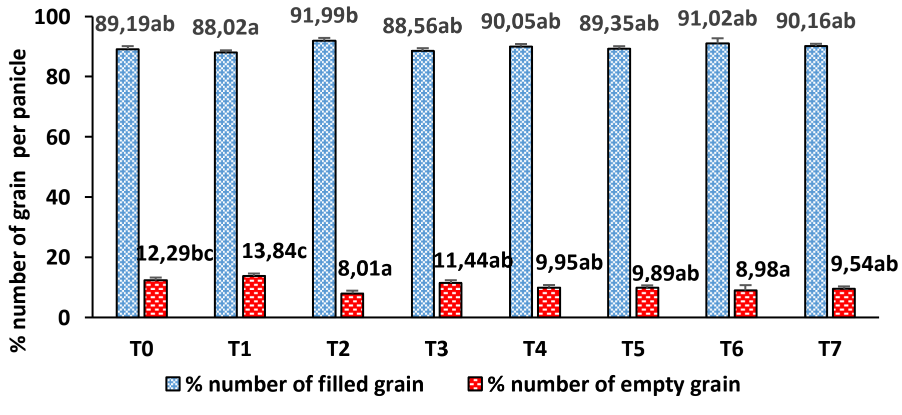

Figure 3. Effect of Various Treatments of Nanoparticles ZnO Enhanced Compost with Different Application Methods on the Harvested Dry Weight of Grain Compared to Control (no fertilizer, T0) and Inorganic Fertilizer (T1), Based on the Results of HSD Test (P<0.05). (Remark: HSD 5% =3.11)

According to Nazir et al. (2024) [31], ZnO nanoparticles spray at 25 mg/l concentration improved morphological, physiological, biochemical, and enzymatic activity during wheat crop growth. Better morphology and physiology of plant roots will increase water absorption capacity. Strong stems will support better plants, reduce susceptibility to physical damage and thus produce better-quality rice grains.

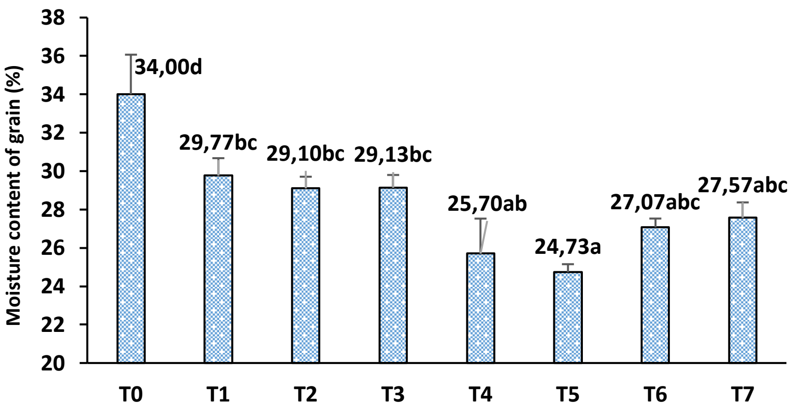

Figure 4. Effect of Various Treatments of Nanoparticles ZnO Enhanced Compost with Different Application Methods on the Moisture Content of Grain Compared to Control (no fertilizer, T0) and Inorganic Fertilizer (T1), Based on the Results of HSD Test ($P<0.05$). (Remark: HSD 5% =3.45)

The moisture content of dry harvested grain rice yields is one of the grain quality parameters. The lower the water content of the grain, the higher the quality of the grain. Grain quality is determined by the availability and nutrient uptake of rice plants [32]. Application of compost enriched with 100-150 mg kg^{-1} ZnO nanoparticles produced grain with low water content and lower than the control treatment (T0) and inorganic fertilizer (T1). Nutrient uptake of N, P, K and Zn in the compost+100 mg kg^{-1} treatment, with foliar application provided optimum nutrient uptake for rice plants (Table 1).

4 Conclusion

Application of ZnO nanoparticles enhanced compost (ZnO-NEC) has a significant impact on the nutrient uptake, rice yield and quality of grain compared to control (no treatment). The current research shows that addition of ZnO-nanoparticles 100-150 mg kg^{-1} compost with foliar application has the potential to reduce 50% of inorganic fertilization dose. This treatment provides higher P, K and Zn nutrient uptake compared to the use of inorganic fertilizer, but the N uptake of this treatment is the same as the treatment using inorganic fertilizer. The yield of harvested dry grain weight also equal to the treatment using 100% inorganic fertilizer. Grain quality as measured by the percentage of empty grain and moisture content of harvested dry grain in the ZnO-NEC of 100-150 mg kg^{-1} compost was lower than when using 100% inorganic fertilizer. These research results suggest that the use of ZnO-NEC can be used as an alternative fertilizer to reduce the use of chemical fertilizers in environmentally friendly and sustainable rice cultivation.

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