Improving growth performance of rice cultivars IR64 and Mentik Wangi in drought stress with applying nano Zn

F M Alhasan, A T Sakya* and M Rahayu

1Agrotechnology Department, Faculty of Agriculture Sebelas Maret University, Surakarta 57126, Indonesia
*Correspondence author: amaliatetrai@staff.uns.ac.id

Abstract. Increasing temperatures due to climate change caused irregular rainfall in some regions. Changes in rainfall initiate drought to increase every year and negatively impact vital human activities, including agricultural sectors. Drought stress is becoming increasingly important in agriculture because it affects changes in physiological function and leads to a severe decline in crop production. Nano Zn is one of the solutions for drought stress which has a role in enzyme activation, protein synthesis, and carbohydrate metabolism. This study aims to determine the effect of applying nano Zn on rice growth performance in drought stress conditions. The method used was a non-factorial Completely Randomized Design with analysis of variance followed by DMRT at 5% level. The levels of Zn concentration were 0; 0.5; 1; 1.5; 2 ppm Zn. Two varieties were used, ‘IR 64’ as a hybrid cultivar and ‘Mentik Wangi’ as a local cultivar in Indonesia. The variables observed were plant height (cm), number of tillers, number of leaves, biomass weight (g), and root dry weight (g). The result showed that applying nano Zn can increase rice growth due to drought stress on the various variables. The higher the nano Zn dosage, the higher plant height, number of tillers, number of leaves, biomass, and root dry weight by 3-6 %, 2-33 %, 6-23 %, 8-28 %, and 3-34 % respectively compared without Zn. These results can be considered a solution for environmental conditions affected by drought stress by paying attention to Zn concentration level.

1. Introduction
Climate change is currently occurring in various regions due to global warming. Increases in air temperature, extreme climatic conditions, and rain patterns are the effects of climate change that can affect multiple aspects, including the agricultural sector [1]. The increase in temperature in plants can increase nitrogen (N) loss, increase transpiration, decrease leaf assimilation rate, and reduce the level of water use efficiency [2, 3]. The increasing rice needs have not been met due to various environmental problems such as drought stress which impact rice production [4].

Rice plants that are exposed to drought stress show symptoms of temporary wilting and drying leaves. These symptoms indicate that the leaves cannot carry out normal metabolic activities, absorb nutrients and inhibit the formation of leaf chlorophyll [5]. Physiological changes that also occur due to drought are a decrease in PAR (photosynthetically active radiation), photosynthesis rate, and pigment degradation that can related to plant resistance [6, 7, 8].

One of the solutions related to the problem of drought-stressed plants is by giving nanoparticles Zn (Zn NPs) [9]. Zn in plants acts as a catalyst for protein formation, regulator of indoleacetic balance, and carbohydrate transformation [10, 11]. The application of Zn NPs [12] can increase fresh and dry weight and increase plant growth and yield parameters [13]. However, the exact nano Zn concentration is still unknown to obtain for optimal rice growth in drought stress therefore until now it
is still necessary to study applying Zn NPs in drought stress. Thus, this study aimed to determine the effect of nano Zn application on rice growth performance in drought stress conditions and provide benefits by contributing to nanotechnology in Indonesia in agriculture.

2. Material and methods
The study was held from September 2020 to January 2021 at the screen house Faculty of Agriculture, Sebelas Maret University, Surakarta. Planting media using andosol soil in polybags as much as 7 kg. Fertilizer application is carried out in 3 stages, namely at the time of planting, 21 days after seeding (DAS), and 35 DAS [14]. At the time of planting, the dose of manure given was 5 tons ha⁻¹ (26.25 g/polybag), urea 0.13 tons ha⁻¹ (0.65 g/polybag), KCl 0.05 tons ha⁻¹ (0.26 g/polybag), and SP 36 as much as 0.1 tons ha⁻¹ (0.525 g/polybag). At 21 HST and 35 HST, urea and KCl fertilizer were given 1/2 doses each. Zn NPs were given as foliar application twice, namely during the formation of tillers and elongation of stems. Drought stress treatment was carried out by providing water at intervals of 12 days [15] and a control treatment was irrigated every day. The method used was a non-factorial Completely Randomized Design with analysis of variance followed by DMRT at 5% level. The levels of Zn concentration were 0; 0.5; 1; 1.5; 2 ppm Zn. Two varieties were used, ‘IR64’ as a hybrid cultivar and ‘Mentik Wangi’ as a local cultivar in Indonesia.

The variables observed were plant height (cm), number of tillers, number of leaves, biomass (g), and root dry weight (g). Number of leaves, tillers and plant height was observed in 11 WAP (weeks after planting). Biomass was measured by cutting all parts of the plant, and the root dry weight was measured by weighing the roots. Then oven it at 65°C for 72 hours to achieve a constant weight [16].

3. Result and discussion

3.1. Plant height

Table 1. Plant height, number of tillers, and number of leaves rice plant under normal and drought condition with the application of Zn NPs on 11 WAP.

| Treatments                            | Plant height (cm) | Number of tillers | Number of leaves |
|---------------------------------------|-------------------|-------------------|-----------------|
| IR64 + 0 ppm non-stressed             | 97.00 cde         | 16.00 bcd         | 69.50 bcde      |
| IR64 + 0 ppm stress                   | 76.75 a           | 10.25 a           | 44.25 a         |
| IR64 + 0.5 ppm non-stressed           | 105.3 e           | 18.50 d           | 84.25 e         |
| IR64 + 0.5 ppm stress                 | 79.00 a           | 13.25 abcd        | 53.00 abc       |
| IR64 + 1 ppm non-stressed             | 95.00 e           | 14.50 abcd        | 66.25 bcde      |
| IR64 + 1 ppm stress                   | 77.50 a           | 13.25 abcd        | 52.00 ab        |
| IR64 + 1.5 ppm non-stressed           | 100.0 bcde        | 17.00 cd          | 75.25 de        |
| IR64 + 1.5 ppm stress                 | 81.00 a           | 13.50 abcd        | 54.40 abcd      |
| IR64 + 2 ppm non-stressed             | 94.75 de          | 17.00 cd          | 74.25 cde       |
| IR64 + 2 ppm stress                   | 79.00 a           | 10.50 ab          | 43.00 a         |
| Mentik Wangi + 0 ppm non-stressed     | 104.0 bcde        | 14.75 abcd        | 58.75 abcd      |
| Mentik Wangi + 0 ppm stress           | 81.75 ab          | 11.75 abc         | 48.00 ab        |
| Mentik Wangi + 0.5 ppm non-stressed   | 90.50 abcd        | 11.25 ab          | 42.50 a         |
| Mentik Wangi + 0.5 ppm stress         | 80.75 a           | 11.50 abc         | 42.50 a         |
| Mentik Wangi + 1 ppm non-stressed     | 105.3 e           | 14.00 abcd        | 55.25 abcd      |
| Mentik Wangi + 1 ppm stress           | 77.25 a           | 15.00 abcd        | 51.00 ab        |
| Mentik Wangi + 1.5 ppm non-stressed   | 98.50 de          | 16.00 bcd         | 61.75 abcd      |
| Mentik Wangi + 1.5 ppm stress         | 79.50 a           | 14.25 abcd        | 55.75 abcd      |
| Mentik Wangi + 2 ppm non-stressed     | 107.5 e           | 17.50 d           | 67.00 bcde      |
| Mentik Wangi + 2 ppm stressed         | 84.25 abc         | 15.57 abcd        | 58.25 abcd      |

Note: The number followed by the same letter in one column shows that it is not significantly different at the 5% level of DMRT
The height and low of the plant stems can be influenced by the traits or characteristics of various varieties that affect yield and external factors such as climate or others [17]. Table 1 shows that IR64 under stress conditions in all treatments had a significant decrease in height plant with a range of 16-20% compared to without Zn under stress conditions. Likewise, Mentik Wangi decreased by 19-26%. Water deficit in plants can affect the vegetative growth of plants [18]. Water plays a role in carrying out various plant processes such as regulating cell turgidity to carry out the mechanism of movement of the stomatal organs and cell formation and filling [7].

Plant height in drought-stressed conditions was highest in Zn NPs 2 ppm applications, and there was a 6% increase compared to without Zn applications. However, IR64 with the same application and concentration did not show the highest plant height because one of the plants in the treatment had a disease resulting in a decrease in the resulting average value. That also applies to other parameters.

In conditions without stress, the highest plant height was in 2 ppm Zn in the Mentik Wangi cultivar. However, it had not shown a significant difference without the Zn application. Giving Zn NPs 2 ppm can increase plant height because Zn plays a role in forming the auxin, which is allocated to the height of the plant itself [19]. Zn on plant height correlates with membrane activity and cell elongation under drought stress conditions [20]. Furthermore, the application of Zn NPs increased the content of chlorophyll which increased the photosynthetic process in plants [21].

3.2. Number of tillers
One of the factors in plants that affect the number of tillers is water [22]. Table 1 shows that the IR64 in stress conditions with applying Zn decreased the number of tillers significantly with a range of 15-34% compared to without Zn in normal conditions. Whereas in Mentik Wangi was a decrease in the range of 3-22% and an increase at 2 ppm Zn NPs of 6%. The formation of tillers is influenced by several factors, one of which is the availability of water so drought was the main factor in decreased the number of tillers in most treatments [22].

The highest number of tillers on drought stress was showed in 1.5 ppm Zn NPs applying in IR64 and 2 ppm Zn NPs in Mentik Wangi. Applying those treatments increase 32% and 33% in IR 64 and Mentik Wangi respectively. The results of the least number of tillers in drought-stressed conditions were obtained in cultivar IR64 without Zn NPs. In conditions without stress, the highest number of tillers were obtained at the level of 0.5 ppm Zn NPs cultivar IR64 which is not significantly different from the without Zn NPs. The application of Zn affects plant processes such as protein synthesis, carbohydrate metabolism, and the formation of the hormone auxin [11].

3.3. Number of leaves
Leaves are plant organs that produce carbohydrates that are produced from photosynthesis [23] because they contain chlorophyll, which is needed by plants in the photosynthetic process [24]. Table 1 shows IR64 under stress conditions in all treatments significantly reduced the number of leaves with a range of 22-38% compared to without Zn in normal/unstressed conditions. This result is the same as Mentik Wangi, which has decreased by 1-28%. The application of Zn NPs in stress conditions has not given the number of leaves that are close to the normal conditions. Water is the factor that most influences the growth rate of plants therefore the plant that lacks water will disrupt metabolism [8], [25].

Mentik Wangi with 2 ppm Zn NPs and IR64 with 1.5 ppm Zn NPs on drought conditions gave the highest number of leaves. Spraying those concentrations increased the number of leaves by 21% and 23% respectively, compared to without Zn application in drought conditions. IR64 without Zn in drought conditions gave the least number of leaves. The condition without stress gave the highest number of tillers at the level of 0.5 ppm Zn NPs of IR64 cultivar. The number of tillers was not significantly different from the level of 0 ppm Zn. Zn becomes a cofactor in plant growth, including the number of leaves, because it acts as an auxin enzyme activator [26]. That can be a solution for
drought condition [27] because Zn found in the soil accumulates in the root tissue and is channeled through the xylem to the shoots [26].

3.4. Biomass
Biomass is the entire weight of the shoot that has gone through the oven process and represents the net CO₂ assimilation and photosynthetic assimilation during the plant growth process [28]. Table 2 shows that rice cultivar IR64 under stress conditions of 2 ppm Zn NPs decreased the biomass significantly by 18% compared to treatment at the 0 ppm Zn NPs level without drought-stressed conditions. Spraying of 0.5; 1; 1.5 ppm Zn NPs shows a 1-4% increase in biomass. Cultivar Mentik Wangi decreased in all treatments in drought conditions with a range of 26-38%. The physiological response of plants in drought stress will reduce the transpiration rate by closing the stomata so that it can inhibit the exchange of CO₂ and O₂ from plant tissues with the atmosphere [26]. Then reduce of net CO₂ assimilation accumulation and result in decreasing dry weight [3, 29].

The heaviest biomass in drought-stressed conditions was showed on cultivar IR64 1.5 ppm Zn NPs and 1 ppm Zn NPs in cultivar Mentik Wangi with an increase of 27% and 28%, respectively. In non-drought conditions, the result of 2 ppm Zn NPs of Mentik Wangi cultivar was the heaviest biomass. Zn application will increase the N absorption rate to increase the dry biomass [30]. Biomass of the 0 ppm Zn Mentik Wangi that gives the smallest is thought to be due to low N absorption. The increase in N levels will directly increase the biomass, carbohydrates, and leaf chlorophyll [31]. An increase in biomass was also reported in a study of Zn NPs which applied to saline-stressed sunflower plants, the addition of Zn NPs increased 13% biomass compared to without Zn application [32].

Table 2. Biomass (g) of rice plant under normal and drought condition with the application of Zn NPs on 11 WAP.

| Treatments                  | Drought Stress Condition (g) |
|-----------------------------|-------------------------------|
|                            | Non-stressed | Stressed   |
| IR64 + 0 ppm               | 27.15 abc    | 22.34 a    |
| IR64 + 0.5 ppm             | 37.95 cde    | 27.53 abcd |
| IR64 + 1 ppm               | 33.42 abcd   | 27.71 abcd |
| IR64 + 1.5 ppm             | 35.13 abcd   | 28.31 abcd |
| IR64 + 2 ppm               | 33.74 abcd   | 22.20 a    |
| Mentik Wangi + 0 ppm       | 38.33 cde    | 21.84 a    |
| Mentik Wangi + 0.5 ppm     | 30.30 abde   | 27.70 abed |
| Mentik Wangi + 1 ppm       | 41.41 de     | 28.05 abed |
| Mentik Wangi + 1.5 ppm     | 36.83 bcde   | 25.14 abc  |
| Mentik Wangi + 2 ppm       | 42.77 e      | 23.59 a    |
| Means                      | 35.70        | 25.44      |

Note: The number followed by the same letter in rows and columns shows that they are not significantly different at the 5% level of DMRT

3.5. Root dry weight
Root dry weight indicates the ability of the plant to absorb water by taking into account the tolerance level, which is proportional to the root dry weight [33]. Table 3 shows that the IR64 on stress conditions 0.5; and 2 ppm Zn NPs, there was a significant decrease in the root dry weight as much as 5-17% compared to the 0 ppm Zn NPs treatment without drought stress conditions. While giving 1; and 1.5 ppm Zn NPs increase a 3-31%. Whereas, Mentik Wangi decreased in all treatments with a range of 31-52% compared to the conditions without giving Zn in normal condition. The availability of sufficient water will absorb nutrients from the roots due to the loose soil texture [34]. The process of cell division and the rate of transpiration are directly proportional to water availability in plants [25].
In normal conditions, 0.5 ppm Zn NPs application on IR64 showed the heaviest root weight and showed an increase of 150% compared to the condition without Zn application. Whereas, in the Mentik Wangi cultivar the increase in Zn concentration did not show a significant difference in root weight without Zn application. The dry weight root of the Mentik Wangi cultivar 0.5 ppm Zn NPs and at IR64 1 ppm Zn NPs increased 18% and 43% respectively compared without Zn application, although it did not show a significant difference (Table 3). The increase in the root system, both dry weight, and root length were also shown in the application of Zn to tomatoes that were in stressed-drought condition [35].

Cultivar IR64 0 ppm Zn and 2 ppm Zn NPs in Mentik Wangi cultivar gave the most negligible root dry weight under stress conditions. As stated by Juliati [36], giving Zn NPs does not always significantly affect plant growth. However, this study showed an increase in the dry weight of IR64 rice roots with Zn 0.5; 1; and 1; 5 ppm respectively, 3%, 43%, and 13%.

### Table 3. Root dry weight (g) of rice plant under normal and drought condition with the application of Zn NPs on 11 WAP.

| Treatments          | Drought Stress Condition (g) |
|---------------------|-----------------------------|
|                     | Non-stressed | Stressed    |
| IR64 + 0 ppm        | 12.17 ab     | 11.14 ab    |
| IR64 + 0.5 ppm      | 30.45 d      | 11.49 ab    |
| IR64 + 1 ppm        | 17.68 abc    | 15.89 abc   |
| IR64 + 1.5 ppm      | 14.51 abc    | 12.57 abc   |
| IR64 + 2 ppm        | 17.63 abc    | 10.05 a     |
| Mentik Wangi + 0 ppm| 23.90 cd     | 13.84 abc   |
| Mentik Wangi + 0.5 ppm| 20.94 bcd    | 16.30 abc   |
| Mentik Wangi + 1 ppm| 21.87 bcd    | 12.83 ab    |
| Mentik Wangi + 1.5 ppm| 29.50 d      | 12.53 ab    |
| Mentik Wangi + 2 ppm| 23.93 cd     | 11.34 ab    |
| Means               | 21.26        | 12.80       |
[11] Broadley M, Brown P, Cakmak I, Renge L and Zhao F 2012 Function of nutrients: micronutrients Marschner's mineral nutrition of higher plants ed Marschner P (Massachusetts: Academic Press) chapter 7 pp 191-248
[12] Sadak M S and Bakry B A 2020 Bulletin of the National Research Centre 44 1–12
[13] Attaya A K G, Genaidy E A E and Zahran H A 2018 Bioscience Rsrch 15 1528–41
[14] Sukristiyonubowo, Jamil A and Hastono D S 2013 Budi daya padi pada sawah bukaan baru (Jakarta: IAARD Press) p 33
[15] Gumisa V 2017 Impact of gradual drought stress vegetative growth phase in chili pepper (Palembang: Sriwijaya University)
[16] Fischer K S, Lafitte R, Fukai S, Atlin G and Hardy B 2003 Breeding rice for drought-prone environments (Los Baños: International Rice Research Institute) p 98
[17] Jiang T, Liu J, Gao Y, Sun Z, Chen S, Yao N, Ma H, Feng H, Yu Q and He J 2020 Agricultural Water Management 232 106066
[18] Suprihatno B, Daradjat A A, Satoto S E B, Widiarta I N, Setyono A, Indrasari S D, Lesmana O S and Semiring H 2009 Deskripsi varietas padi (Jakarta: Badan Penelitian dan Pengembangan Pertanian Departemen Pertanian)
[19] Cakmak I 2008 Plant and Soil 302 1-17
[20] Cakmak I and Engels C 1999 Role of mineral nutrients in photosynthesis and yield formation ed Rengel Z (New York: The Haworth Press) p 168
[21] Adrees M, Khan Z S, Hafeez M, Rizwan M, Hussain K, Asrar M, Alyemeni M N, Wijaya L and Ali S 2021 Ecotoxicology and Environmental Safety 208 1–9
[22] Fageria N K 2007 J. of Plant Nutrition 30 843–79
[23] Wirastmaja I W 2017 Bahan ajar zat pengatur tumbuh auksin dan cara penggunaannya dalam bidang pertanian (Bali: Fakultas Pertanian UNUD)
[24] Pertivi L, Suprapto A dan Farid N 2020 Proc. Seminar Nasional Riset Teknologi Terapan (Magelang: Universitas Tidar)
[25] Bachelet D and Gay C A 1993 Ecological Modelling 65 71–93
[26] Noulas C, Tziouvalekas M and Karyotis T 2018 J. of Trace Elements in Med. and Bio. 49 252–60
[27] Mori A, Kirk G J D, Lee J S, Morete M J, Nanda A K, Johnson-Beebou S E and Wissuwa M 2016 Frontiers in Plant Science 6 1160
[28] Faizal R, Raden S and Sigit S 2017 J. Agritrop 15 162–80.
[29] Liu F, Jensen C R and Andersen M N 2004 Field Crops Research 86 1–13
[30] Hosseiny Y and Maftoun M 2008. J. of Agr. Sci. and Tech. 10 307–16
[31] Sulaiman F, Suwignyo R A and Hasmeda M 2014 J. Lahan Suboptimal 3 145–51
[32] Torabian S, Zahedi M and Khoshgoftar A H 2016 J. of Plant Nutrition 39 172–80
[33] Winangsih, Prihastanti E and Parman S 2013 Buletin Anatomi Dan Fisiologi 21 19–25
[34] Isnawan B H, Kurwasit N, Supangkat G and Ediyono S 2017 Saintis 9 181–91
[35] Sakya A T, Sulistyaningsih E, Indradewa D and Purwanto B H 2017 J. of Soil Sci. and Agroclimatology 13 74–80
[36] Juliati S 2008 J. Hortikultura 18 408–19

Acknowledgment
The author would like to thank all Faculty of Agriculture UNS stakeholders who have helped with this research.