EFFECTS OF PLANT GROWTH REGULATORS AND MICRONUTRIENTS ON CROP PERFORMANCE, SEED GERMINATION AND SEEDLING VIGOUR IN RICE (*ORYZA SATIVA* L.)

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(Received 5th Jul 2023; accepted 11th Oct 2023)

Abstract. The experiment was conducted during the 2019-2020 growing season at the Agricultural College and Research Institute in Thanjavur, India. The effect of plant growth regulators and micronutrients for improving seed germination of rice offsprings (*Oryza sativa* L.) were investigated. The experiment followed a randomized block design, consisting of twelve treatments and three replications. The predominant rice variety cultivated was ADT 46, known for its long, slender grain but poor germination characteristics. The treatments involved foliar spraying at 60 and 90 days after transplanting (DAT), before panicle emergence. The highest recorded plant height was achieved with the application of Cytokinin @ 50 ppm + $ZnSO_4$ @ 0.5% at 60 and 90 DAT during the main crop growth phase. Interestingly, for all other growth and yield parameters, the most favorable results were observed with the application of Naphthalene acetic acid (NAA) @ 40 ppm + $ZnSO_4$ @ 0.5%. Notably, this treatment significantly boosted the germination percentage to 98.38% when tested three months after storage, marking a 13.00% increase compared to the control. It also demonstrated the highest seedling vigor indexes I (2996) and II (792). Hence, it is recommended that the application of NAA @ 40 ppm + $ZnSO_4$ @ 0.5% foliar sprays at 60 and 90 DAT to address germination issues in rice and achieve higher yield and profit for farmers.
Keywords: *rice, offspring, germination, seedling vitality, naphthalene acetic acid (NAA), zinc*

Introduction

Rice (*Oryza sativa* L.) is a staple food for nearly half of the world's population. However, more than 90 percent of rice is consumed in Asia, where it is a staple food for the majority of the population, including 560 million people in the region. Globally, India ranks first in rice cultivation and second in production, following China. Rice is also a staple food for over 65% of India's population, contributing to more than 42% of the country's total food production. In India, rice is cultivated across 46.38 million ha, yielding 130.29 million tons with a productivity of 2809 kg ha^{-1}. In Tamil Nadu alone, rice is grown on 2.21 million ha, producing 8.07 million tons with a productivity of

http://www.aloki.hu ● ISSN 1589 1623 (Print) ● ISSN 1785 0037 (Online)
DOI: http://dx.doi.org/10.15666/aeer/2201_709719

3658 kg ha^{-1} (Government of India, 2022). The looming challenge is India’s increasing population, expected to reach 1.5 billion by 2050 (Cassen and Visaria, 1999). This demographic growth necessitates a corresponding increase in food production. Although global food availability might suffice, local production is crucial due to socio-economic and political factors. Thus, enhancing rice productivity within resource constraints remains a paramount concern.

Plant growth regulators (PGRs) may play a vital role in regulating both yield and fruit quality during growth in controlled-environment greenhouses. However, the uncertainty of results, stemming from the specificity of plant responses, can limit their use and potential benefits (Margarita et al., 2015). Plants respond to both biotic and abiotic stresses by altering their photosynthetic rates. In plants, there are five key growth hormones that interact: auxins, cytokinins, gibberellins, ethylene, and abscisic acid. The balanced interaction of these hormones is crucial, as any imbalance can activate or inhibit others.

Gibberellins are important phytohormones that regulate many developmental processes in plants (Liu et al., 2019). They promote germination, break plant dormancy, and stimulate cell division (Holalu et al., 2021). Cytokinins also influence cell division, affecting plant growth, lateral bud formation, and organ/tissue aging. Traditionally, plant growth and development were thought to be regulated solely by five hormone groups: auxins, gibberellins, cytokinins, ethylene, and abscisic acid. For example, GA_3 application promotes cell division and elongation in rice internodes. Cytokinins have diverse roles in plant development and responses to the environment, including leaf senescence, apical dominance, chloroplast development, and regulation of cell division (Hutchison and Kieber, 2002). Micronutrients refer to essential plant nutrient elements such as boron (B), zinc (Zn), manganese (Mn), iron (Fe), copper (Cu), molybdenum (Mo), chlorine (Cl), and silicon (Si), collectively constituting less than 1% of most plant’s dry weight. As minerals break down during soil formation, micronutrients gradually become available to plants.

Abiotic stresses, such as elevated temperature and moisture stress, are key limiting factors for crop development and output (Venugopalan et al., 2022). Seed germination is influenced by both intrinsic and extrinsic factors. Water softens seed coats, enhances seed permeability, and converts stored nutrients into a soluble form for embryo nourishment. Oxygen, obtained from the soil’s air pockets, is essential for respiration, providing energy for growth. Seeds require mineral elements from the soil during growth. Seeds may remain viable for varying durations, with some exhibiting dormancy and germinating only under favorable conditions. Growth inhibitors, such as abscisic acid, induce dormancy in seeds. In direct dry-seeded rice production, poor germination and non-uniform seedling establishment pose significant challenges, particularly in water-stressed conditions (Goswami et al., 2013). To ensure food security for a growing population, it is imperative to employ sustainable methods for improving seed germination, seedling emergence, and growth, both under normal and drought conditions (Liu et al., 2015). Understanding the precise micronutrient requirements for optimal crop growth is not only crucial for fresh produce but also for seed production (Mohammadi et al., 2016). It is essential to comprehend the role of nutrient supply in seed formation and setting since mineral concentrations in plant tissues profoundly impact growth and reproductive potential (Majanbu et al., 1986). Factors such as stored protein, mRNA quality, maternal plant growth, environmental conditions during seed development, and the supply of

http://www.aloki.hu ● ISSN 1589 1623 (Print) ● ISSN 1785 0037 (Online)
DOI: http://dx.doi.org/10.15666/aeer/2201_709719

essential minerals significantly influence successful germination and seed vigor (Nonogaki et al., 2010; Rajjou et al., 2012). Seed viability and vigor are critical aspects of seed quality, ultimately affecting the success of crop cultivation. Storage conditions play a pivotal role in preserving mature seeds' quality, making it valuable for producers, retailers, and farmers to assess seed quality before sowing (Nonogaki et al., 2010). Therefore, in this study, the effect of growth regulator and micronutrient as foliar application in rice was examined for the yield, germination, and seedling vigor in rice variety [ADT(R)46] progeny.

Materials and methods

The field experiment was conducted at the Agricultural College and Research Institute, Eachangkottai, Tamil Nadu Agricultural University, Thanjavur, India, situated at coordinates 10°40'N latitude, 79°09'E longitude, and an altitude of 54.26 m. It is located in the D block of college farm. The soil was a red sandy loam in texture, medium in organic carbon (0.61%), low in available nitrogen (N; 188.1 kg ha^{-1}), moderate in available phosphorus (P_2O_5; 20.16 kg ha^{-1}) and available potassium (K_2O; 212.8 kg ha^{-1}), with a pH of 5.80 and EC of 0.221 dSm^{-1}. The soil is the red sandy loam type of soil which is suitable for both irrigated and rainfed crops. It is highly permeable soil with the top layer slightly eroded. The date of sowing in nursery on 16.09.2019 followed by transplanting in the main field on 15.10.2029 and harvested on 28.01.2020. The adopted plant spacing is 20 × 10 cm having the population density of 50 hills m^{-2}. The weather data during the experimental period were recorded from the meteorological observatory located at Agricultural College and Research Institute, Eachangkottai, Thanjavur. The mean weekly maximum and minimum temperature during the cropping period ranged from 28 to 35°C with an average of 33°C and 15 to 19°C with an average of 17.5°C respectively. The mean relative humidity (RH) during the crop growth period varied from 82.42 to 93.76% with an average of 87.31% in the morning and varied from 40.50 to 64.38% with an average of 55.69% in the afternoon. The experiment was laid out in a Randomized Block Design (RBD) with 12 treatments replicated thrice with the following treatments as mentioned in the following *Table 1*.

Table 1. *Treatment details*

T_1	GA_3 @ 50 ppm
T_2	Cytokinin @ 50 ppm
T_3	NAA @ 40 ppm
T_4	$ZnSO_4$ @ 0.5%
T_5	$MnSO_4$ @ 0.2%
T_6	GA_3 @ 50 ppm + $ZnSO_4$ @ 0.5%
T_7	GA_3 @ 50 ppm + $MnSO_4$ @ 0.2%
T_8	Cytokinin @ 50 ppm + $ZnSO_4$ @ 0.5%
T_9	Cytokinin @ 50 ppm + $MnSO_4$ @ 0.2%
T_{10}	NAA @ 40 ppm + $ZnSO_4$ @ 0.5%
T_{11}	NAA @ 40 ppm + $MnSO_4$ @ 0.2%
T_{12}	Control (water spray)

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The treatments are applied as a foliar spray on 60 DAT (before panicle initiation) and 90 DAT (before heading). The peculiar and popular rice variety chosen for this experiment was rice [ADT(R)46] released during the year 2002 is medium duration rice (135 days) having the issue of poor germination collected from the Tamil Nadu Rice Research Institute-Aduthurai, Tamil Nadu for the investigation. The recommended dose of phosphorus @ 50 kg P_2O_5 ha^{-1} as Di-Ammonium Phosphate (DAP) was applied before transplanting as basal, and the remaining potassium @ 50 kg K_2O ha^{-1} as Muriate of Potash (MOP), and nitrogen (150 kg ha^{-1}) as urea as per the recommendation applied in four equal splits (basal, tillering, panicle initiation and flowering) doses.

Data collection and analysis

Germination percentage

After harvesting the seeds were dried to reduce the moisture content below 12% and stored for three months after treatment with the fungicides. After storage, one hundred seeds for each treatment were placed in moist filter paper in an 11 cm diameter Petri dish. Germination count was recorded every 2 days for 8 days after sowing (DAS). The final count of germination was recorded on the 8^{th} day according to International Seed Testing Association rules (ISTA, 1985) and the number of normal seedlings was expressed as a proportion of seeds evaluated.

Seedling vigor

Root length

The ten normal seedlings were selected at random from each replication at the time of germination count and used for measuring the root length. Root length was measured from the collar region to the tip of the primary root and the mean values were expressed in centimeters.

Shoot length

The ten seedlings used for measuring root length were also used for measuring shoot length. The shoot length was measured from the collar region to the tip of the leaf and the mean values were expressed in centimeters.

Dry matter production

Ten normal seedlings from the germination test were placed in a paper cover and dried under shade for 24 h and then, kept in an oven maintained at 80 ± 2°C for 24 h. The dried seedlings were weighed and the mean values were expressed in mg $seedling^{-1}$.

Seedling vigor index-I & II

This was calculated from data on germination percentage and seedling growth and seedling dry weight according to Abdul-Baki and Anderson (1973) by the following formula:

$$\text{Seedling vigor index-I} = \text{Germination (\%)} \times \text{Mean seedling length (cm)} \quad \text{(Eq.1)}$$

$$\text{Seedling vigor index -II} = \text{Germination (\%)} \times \text{Dry matter production (mg/seedling)} \quad \text{(Eq.2)}$$

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Growth parameters

Plant height

The plant height was measured from five randomly selected hills in each treatment by measuring length from the base of the plant to the tip of the longest leaf at 120 DAT and the mean values were expressed in centimeters.

Tillers per plant

The number of tillers was counted and recorded from five randomly selected plants in each treatment at 120 DAT and the mean values were expressed in centimeters.

Leaf area index (LAI)

Leaf area was measured at 120 DAT using a leaf area meter (1/2 – MDL-1000, LICOR Ltd, USA), and leaf area were calculated using the formula.

Root volume

Root samples of five randomly selected hills from each treatment were taken at grain filling stage by adopting standard procedure (Karunakaran and Behera, 2015). Roots were excised from tillers and washed with 0.5 mm diameter sieve to prevent loss of fine roots during washing. By using a measuring cylinder, an increase in the water level after the immersion of roots was noted as root volume. The mean value was calculated and expressed as cc hill^{-1}.

Yield parameters and yield

Productive tillers per hill

All the panicle-bearing tillers from five randomly selected hills from each treatment were counted at harvest averaged, and expressed as numbers of productive tillers per hill.

Panicle length

From five randomly selected hills for each treatment the length of each panicle was measured from the base of the primary rachis to the topmost spikelet. The mean length of the panicle was averaged and expressed in centimeters.

Filled grains per panicle

The filled grains per panicle were counted from the five sampled panicles and their average was recorded and expressed as the number of filled grains per panicle.

Test weight

Dried 1000 seed samples were drawn randomly from each treatment were counted and weight was recorded in grams.

Grain yield

Crops were harvested manually leaving five centimeters above the ground and separated the grains and straw by threshing the separated grains were recorded as grain yield.

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Statistical analysis

The data recorded for different parameters were analyzed with the help of the analysis of variance (ANOVA) technique for a randomized block design using MSTAT-C software. The results are presented at a 5% level of significance (p = 0.05).

Results

Growth attributes

At 120 days after transplanting (DAT) the application of Cytokinin @ 50 ppm + $ZnSO_4$ @ 0.5% was recorded as significantly highest plant height (120.8 cm) and statistically on par with the application of Cytokinin @ 50 ppm (118.2 cm) and Cytokinin @ 50 ppm + $MnSO_4$ @ 0.2% (112.4), whereas the lowest plant height was recorded with the control (water spray) (90.40 cm). At 120 DAT the application of NAA @ 40 ppm + $ZnSO_4$ @ 0.5% was recorded as significantly highest tillers per hill (9.65) and statistically at par with the application of NAA @ 40 ppm (9.64) and GA_3 @ 50 ppm + $ZnSO_4$ @ 0.5% (9.48) and the lowest number of tillers per hill (6.88) was recorded with the control (water spray). At 120 DAT, the application of NAA @ 40 ppm + $ZnSO_4$ @ 0.5% was recorded significantly highest root volume (68.7 cc $hill^{-1}$) and the lowest root volume (55.9 cc $hill^{-1}$) was recorded with the control (water spray) (*Table 2*).

Table 2. *Plant height, tillers per hill, and root volume at 120 DAT as influenced by plant growth regulators, micronutrients and their combination in rice variety ADT(R) 46*

Treatment	Plant height (cm)	Tillers (no. $hill^{-1}$)	Root volume (cc $hill^{-1}$)
T_1-GA_3@50 ppm	93.96^b	7.02^b	63.3^c
T_2- Cytokinin @ 50 ppm	118.2^a	7.37^b	62.7^d
T_3-NAA @ 40 ppm	93.48^b	9.64^a	64.3^{cb}
T_4- $ZnSO_4$ @ 0.5%	90.65^b	6.91^b	64.7^{cb}
T_5- $MnSO_4$ @ 0.2%	84.86^b	7.12^b	62.8^d
T_6- GA_3 @ 50 ppm + $ZnSO_4$ @ 0.5%	95.20^b	9.48^a	65.8^b
T_7- GA_3 @ 50 ppm + $MnSO_4$ @ 0.2%	93.40^b	7.72^b	64.7^{cb}
T_8- Cytokinin @ 50 ppm + $ZnSO_4$ @ 0.5%	120.8^a	7.68^b	63.3^c
T_9- Cytokinin @ 50 ppm + $MnSO_4$ @ 0.2%	112.4^{ab}	5.30^c	65.5^{cb}
T_{10}- NAA @ 40 ppm + $ZnSO_4$ @ 0.5%	94.06^b	9.65^a	68.7^a
T_{11}-NAA @ 40 ppm + $MnSO_4$ @ 0.2%	93.60^b	9.44^a	66.0^b
T_{12}- Control (water spray)	90.40^b	6.88^b	55.9^e
SEm±	7.11	0.54	4.36
CD(P = 0.05)	20.84	1.59	1.27

Yield attributes and yield

At 120 DAT the application of NAA @ 40 ppm + $ZnSO_4$ @ 0.5% was recorded numerically highest productive tillers (294.8 no. m^{-2}) among the treatments which was 58.5 percent higher than the control and it was statistically on par with the application of NAA @ 40 ppm + $MnSO_4$ @ 0.2% (292.6 no. m^{-2}) and NAA @ 40 ppm (283.8 no. m^{-2}), GA_3 @ 50 ppm + $MnSO_4$ @ 0.2% (274.6 tillers m^{-2}) and GA_3@50 ppm (267.3 no. m^{-2}). The lowest productive tillers (185.9 no. m^{-2}) were recorded with the control. At 120 DAT

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DOI: http://dx.doi.org/10.15666/aeer/2201_709719

the application of NAA @ 40 ppm + $ZnSO_4$ @ 0.5% was recorded as significantly highest panicle length (60.8 cm) which was 32.7 percent higher than the control and it was statistically on par with the application of NAA @ 40 ppm + $MnSO_4$ @ 0.2% (59.20 cm) and NAA @ 40 ppm (58.8 cm). Similarly, the application of NAA @ 40 ppm + $ZnSO_4$ @ 0.5% was recorded significantly the highest number of filled grains per panicle (160.8 no. $panicle^{-1}$) and the lowest filed grains per panicle (101.4 no. $panicle^{-1}$) was recorded with the water spraying control treatment. At harvest the treatment NAA @ 40 ppm + $ZnSO_4$ @ 0.5% was recorded significantly highest test weight (28.21 g) and statistically on par with application of NAA @ 40 ppm (28.20 g) and GA_3 @ 50 ppm + $ZnSO_4$ @ 0.5% (28.04 g), followed by NAA @ 40 ppm + $MnSO_4$ @ 0.2% (28.00 g) and GA_3 @ 50 ppm + $MnSO_4$ @ 0.2% (26.28 g) which were on par with each other. While the lowest test weight (17.79 g) was recorded with the water spraying control treatment. The application NAA @ 40 ppm + $ZnSO_4$ @ 0.5% was recorded significantly highest grain yield (4.48 t ha^{-1}) which was 22 percent higher than the control (3.66 t ha^{-1}) and it was statistically on par with the application of NAA + $MnSO_4$ @ 0.2% (4.44 t ha^{-1}) and cytokinin @ 50 ppm + $MnSO_4$ @ 0.5 (4.39 t ha^{-1}) (*Table 3*).

Table 3. *Productive tillers per hill, panicle length, filled grains per panicle, test weight and grain yield as influenced by plant growth regulators, micronutrients and their combinations in rice variety ADT(R) 46*

Treatment	Productive tillers (no. m^{-2})	Panicle length (cm)	Filled grains (no. $panicle^{-1}$)	Test weight (g)	Grain yield (t ha^{-1})
T_1-GA_3@50 ppm	267.3^{a}	45.8^{cb}	145.8^{ab}	25.58^{ab}	4.33^{ab}
T_2- Cytokinin @ 50 ppm	235.4^{ab}	28.4^{d}	128.4^{cb}	22.53^{ab}	4.18^{ab}
T_3-NAA @ 40 ppm	283.8^{a}	58.8^{ab}	158.8^{a}	28.20^{a}	4.35^{ab}
T_4- $ZnSO_4$ @ 0.5%	266.2^{a}	45.2^{cbd}	145.2^{ab}	25.47^{ab}	4.28^{ab}
T_5- $MnSO_4$ @ 0.2%	247.5^{a}	35.0^{d}	135.0^{ab}	23.68^{ab}	4.37^{ab}
T_6- GA_3 @ 50 ppm + $ZnSO_4$ @ 0.5%	245.3^{ab}	53.8^{ab}	153.8^{ab}	28.04^{a}	4.36^{ab}
T_7- GA_3 @ 50 ppm + $MnSO_4$ @ 0.2%	274.6^{a}	49.8^{ab}	149.8^{ab}	26.28^{ab}	4.38^{ab}
T_8- Cytokinin @ 50 ppm + $ZnSO_4$ @ 0.5%	265.8^{a}	45.0^{cbd}	145.0^{ab}	25.44^{ab}	4.28^{ab}
T_9- Cytokinin @ 50 ppm + $MnSO_4$ @ 0.2%	249.3^{a}	36.0^{cbd}	136.0^{ab}	23.86^{ab}	4.39^{ab}
T_{10}- NAA @ 40 ppm + $ZnSO_4$ @ 0.5%	294.8^{a}	60.8^{a}	160.8^{a}	28.21^{a}	4.48^{a}
T_{11}-NAA @ 40 ppm + $MnSO_4$ @ 0.5%	292.6^{a}	59.2^{a}	159.2^{a}	28.00^{a}	4.44^{ab}
T_{12}- Control (water spray)	185.9^{b}	45.8^{cb}	101.4^{c}	17.79^{cb}	3.66^{b}
SEm±	17.72	3.23	9.73	1.71	0.29
CD(P = 0.05)	51.96	9.48	28.54	5.00	0.80

Germination percentage

Application of NAA @ 40 ppm + $ZnSO_4$ @ 0.5% were recorded significantly highest germination percentage (98.38%) which was 13 percent higher than the control (85.20%) and it was statistically at par with the application of NAA @ 40 ppm (96.98%) (*Fig. 1*).

Seedling vigor index I and II

The application of NAA @ 40 ppm + $ZnSO_4$ @ 0.5% was recorded as significantly highest seedling vigor index I (2996) and statistically on par with the application of

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Cytokinin @ 50 ppm + $ZnSO_4$ @ 0.5% (2805) and the lowest vigor index I (2235) was recorded with the control (*Fig. 2*). Similarly, the highest seedling vigor index II (792) was recorded with the application of NAA @ 40 ppm + $ZnSO_4$ @ 0.5% and the least vigor index II (581) was found with control treatment sprayed with water alone (*Fig. 3*).

Discussion

Growth attributes

The results are in agreement with that of Zahir et al. (2001) indicated that an exogenous supply of Cytokinin or its precursors in the root zone could improve the growth and yield of treated plants by improving cell division, chloroplast biogenesis, vascular differentiation and shoot differentiation. Anzer Alam and Manoj Kumar (2015) in their experiments also stated that the application of Zinc (Zn) was found to increase the growth, yield attributes, and yield of the crop due to increased nutrient uptake. The results are in agreement with that of Zahir et al. (1999) indicated that the application of auxins has increased the number of tillers per plant as exogenous application of auxin can increase the translocation rate. Basuchaudhuri (2016) also indicated that NAA application influences the life cycle of rice by the metabolic process to manifest beneficially through the translocation of assimilates from source to sink. Thus, it increased the number of tillers per plant significantly. Ghoneim (2016) also mentioned in their experiment Zn application significantly increased the tillers number over control due to increased nutrient uptake. The results were in agreement with that of Yang Liu (2012) has reported that NAA alone and in combination with GA_3 has increased the leaf area index and dry matter production in rice**.** Anzer Alam and Manoj Kumar (2015) in their experiments also stated that the application of Zinc (Zn) was found to increase the growth of the rice crop due to increased nutrient uptake. The results were in agreement with that of Basuchaudhuri (2016) who indicated that root length, root volume, and root weight (dry and fresh) were found to markedly increase by application of NAA as applied in the spray-influenced life cycle of rice by metabolic process to manifest beneficially through translocation of assimilates from source to sink.

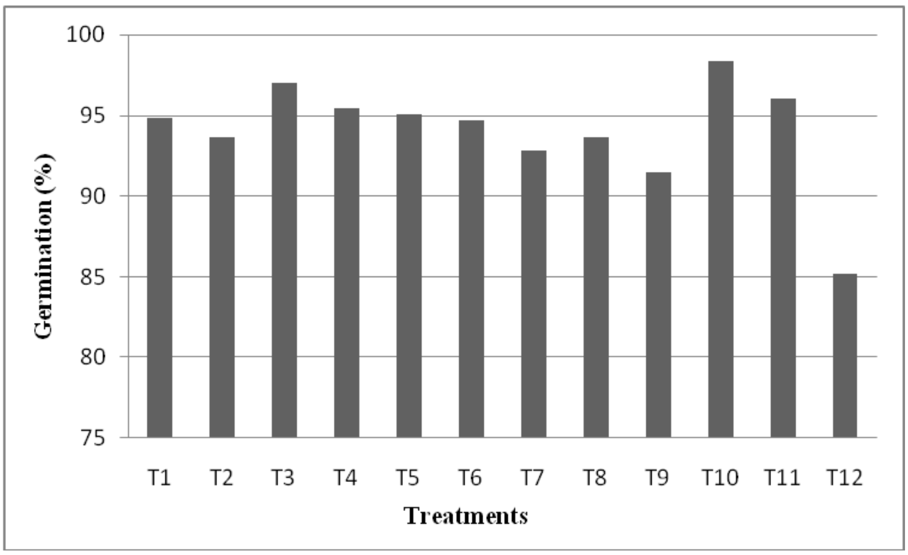

Figure 1. *Germination percentage as influenced by plant growth regulators, micronutrients and their combination in rice variety ADT(R) 46*

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DOI: http://dx.doi.org/10.15666/aeer/2201_709719

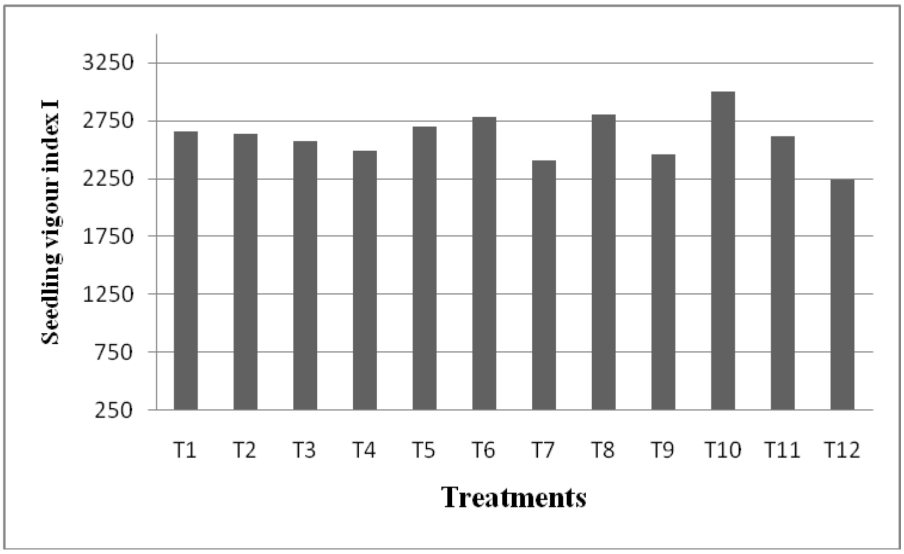

Figure 2. *Seedling vigor index I as influenced by plant growth regulators, micronutrients and their combination in rice variety ADT(R) 46*

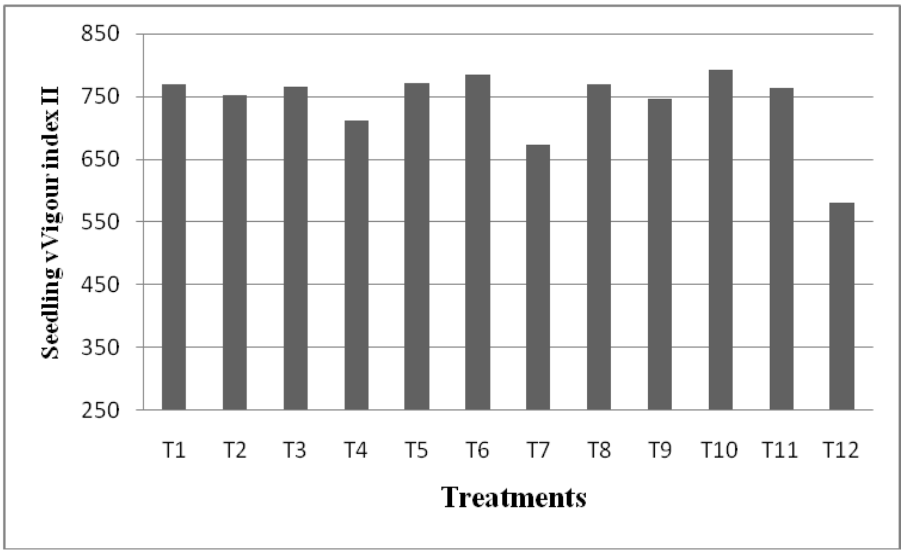

Figure 3. *Seedling vigor index II as influenced by plant growth regulators, micronutrients and their combination in rice variety ADT(R) 46*

Yield attributes and yield

The results were in agreement with that of Adam and Jahan (2011) reported that the application of NAA and its combinations had increased the number of productive tillers per plant, panicle length, number of filled grains, and yield of rice due to increased assimilative area and as a result more photosynthesis throughout the growth period. Yang (2012) has reported that NAA alone and in combination with GA_3 has increased the number of productive tillers in rice by increasing the tillering bud growth. The results were in agreement with that of Ghoneim (2016) reported that the application of zinc increased the number of panicles per m^{-2}, 1000 grain weight, and percentage of filled grains in rice due to improved leaf remobilization of Zn during grain filling.

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DOI: http://dx.doi.org/10.15666/aeer/2201_709719

Bakhsh et al. (2012) also indicated that the application of naphthalene acetic acid was found very effective alone and in combination, improved the yield and yield components of rice considerably due to increased water uptake by the plants.

Germination percentage, seedling vigor I and II

Similar results were reported by the application NAA was not toxic and promoted growth of sweet corn and cowpea seedling reported by Somtrakoon and Kruatrachue (2014). They also further reported that the application of NAA increased dry weight of shoot in cowpea crop.

Conclusion

Hence from this study, it can be concluded that the NAA @ 40 ppm + $ZnSO_4$ @ 0.5% application as two sprays before panicle initiation and flowering at 60 and 90 DAT can be recommended to the farmers for higher yield and seed producers for good quality seeds to overcome the problem of germination in rice for getting higher yield and profit.

Acknowledgements. The authors acknowledge the Dean of the Agricultural College and Research Institute, Eachangkottai, Tamil Nadu, India, for providing all necessary support for this study.

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DOI: http://dx.doi.org/10.15666/aeer/2201_709719