# Agronomic Traits of Direct-Seeded Rice (*Oryza sativa*) Applied with Microbial Inoculants and Varying Fertilizer Levels

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Submitted: 2025-02-27. Revised: 2025-05-01. Accepted: 2025-07-11.

**Abstract.** The study was conducted to evaluate the effects of microbial inoculants and nutrient levels on the agronomic performance of dry direct-seeded NSIC Rc192 rice under aerobic conditions. Six biofertilizer treatments (control, Bio-N, BioGroe, Mykovam, MykoPlus, and Avatar) were combined with four nutrient levels (0 %, 25 %, 50 %, and 75 % NP + full K). The study aimed to assess the impact of these treatments on root colonization, tiller production, and overall plant growth. Root and soil colonization at 60 days after sowing (DAS) revealed that MykoPlus facilitated more effective root colonization than other inoculants. However, unfavorable environmental conditions, including high temperatures and moisture deficits, negatively impacted tiller production and grain yield. Despite these challenges, BioGroe showed the most promise, significantly enhancing root development and tiller count, particularly when combined with 50 % NP + full K. The study concluded that microbial inoculants, especially BioGroe, can improve rice growth under aerobic conditions, but environmental factors are crucial in determining their effectiveness. Further research is needed to optimize these inoculants for varying environmental conditions to maximize their benefits in rice cultivation.

**Keywords:** direct-seeded rice; fertilizer levels; microbial *i*noculants.

**How to Cite:** Inocencio, L. G., Edaño, M. L. S., *Sta.* Cruz, P. C., Ocampo, E. T. M., & Sanchez, P. B. (2025). Agronomic Traits of Direct-Seeded Rice (*Oryza sativa*) Applied with Microbial Inoculants and Varying Fertilizer Levels. *Biosaintifika: Journal of Biology & Biology Education*, *17*(2), 187-199.

DOI: http://dx.doi.org/10.15294/biosaintifika.v17i2.24324

#### INTRODUCTION

Rice (Oryza sativa) is a major global food crop, extensively cultivated for its grain. The Food and Agriculture Organization (FAO) recognized rice as a crucial crop for global food security in 2020. Rice is grown across various ecosystems, covering approximately 150 million hectares worldwide, with 75 % of this area being irrigated, 17 % rainfed lowland, and 4% upland (Nascente et al., 2019). In Asia, upland rice constitutes approximately 6% of the total rice-growing area, with significant cultivation in countries like India and Indonesia (Saito et al., 2018). In the Ilocos Region, rice is one of the key crops cultivated under both lowland and upland conditions.

Most rice farmers utilize puddled soils combined with continuous flooding, a practice that greatly elevates methane emissions. Approximately 94% of methane released from

paddy rice production originates from developing nations, with Asia accounting for 90% of these emissions. Notably, this region experiences an annual emission increase of around 0.4 to 0.7% (Boateng et al., 2019). Drought is a significant natural disaster, not just in the Philippines, but globally, requiring urgent attention. In 2024, the Risk National Disaster Reduction Management Council (NDRRMC) reported PhP1.06 billion in damages across 17,718 hectares of crop areas in regions like Ilocos, Calabarzon, Mimaropa, Western Visayas, and Zamboanga Peninsula (Nepomuceno, 2024). Specifically, the municipalities of Solsona and Dingras in Ilocos Norte were declared under a state of calamity due to prolonged dry spells (Lazaro, 2024).

Low productivity in rice production, particularly in rainfed lowlands, remains a major concern, primarily due to drought, which results in erratic rainfall during the rice-growing season.

Aerobic rice cultivation, a technology that offers multiple benefits, including increased water efficiency, reduced labor and input requirements for seedling production, land preparation, and transplanting, along with improved nitrogen use efficiency and reduced fossil fuel inputs, is considered eco-friendly. Despite these advantages, farmers are hesitant to adopt aerobic rice farming due to its lower yield compared to paddled soil farming, a consequence of poor crop stand establishment and suboptimal nutrient management.

Microbial inoculants, which include organisms like bacteria, fungi, and other microorganisms introduced into an environment to promote plant growth or provide biocontrol, offer a promising solution (Kaminsky et al., 2019). The use of microbial inoculants can reduce dependence on chemical fertilizers (Kaur, 2022) and is considered safer as they cause less environmental harm, have more targeted activity, and are more efficient when used in smaller amounts (Shahwar et al., 2023). These inoculants have been widely adopted in various countries as alternatives to chemical fertilizers, aiming to enhance soil fertility and crop production for sustainable farming.

Recent studies have highlighted the benefits of microbial inoculants on plant growth. Seed vigor and viability, critical factors influencing seedling establishment, crop growth, productivity, can be enhanced by microbial inoculants (Dal Cortivo et al., 2020; Pellegrini et al., 2020; Mitra et al., 2021). In the Philippines, locally available microbial inoculants such as Bio-N, BioGroe, MykoPlus, MykoVam, and Avatar have been tested primarily on rice grown in paddled soil. Bio-N, containing Azospirillum lipoferum and A. brasilense, is a nitrogen-fixing microbial inoculant studied for over two decades. A review by Cruz-Hernández et al. (2022) highlights that Azospirillum species, as plant growth-promoting bacteria. enhance productivity through multiple mechanisms, including nitrogen fixation, phytohormone production, and mineral solubilization, with effectiveness influenced by environmental conditions. There is also evidence of the positive effects of Azospirillum inoculation in rice. Yasuda et al. (2022) reported that inoculation with B510 significantly improved nitrogen uptake by host plants under low nitrogen conditions and influenced the bacterial community structure by recruiting other N2-fixing bacteria even without nitrogen fertilizer. Mykovam, a fungi-based bio-

fertilizer developed by UPLB-BIOTECH, contains active organisms such as Gigaspora margarita, Glomus etunicatum, and macrocarpum (Siababa et al., 2019). Lavelah et al. (2021) found that Mykovam inoculation in mungbean intercropped with corn provided a yield advantage. MykoPlus, a biofertilizer containing various strains and species of microbes, including Endomycorrhiza fungi (Glomus spp.) and bacteria (Bacillus spp.), has been reported to have a significant impact by fostering a diverse community of beneficial microbes that revitalize soils degraded by excessive chemical use (Sarian, 2018). MykoPlus has also been shown to improve rooting, increase shoot height, enhance yields, and boost survival rates during prolonged flooding caused by typhoons in Pangasinan (Pagcaliwagan, 2015). Avatar, another biofertilizer containing Trichoderma spp., has been observed to colonize the plant root zone, enhancing fertilizer absorption and water uptake efficiency. Priyadarshani (2025) reported that Trichoderma virens increased rice plant growth, including height, tillers, leaves, and root size.

Given the advantages of aerobic direct-seeded rice in coping with the effects of climate change, its adoption over transplanted rice appears timely. However, farmers still prefer transplanted rice due to its higher yield, attributed to better crop stand establishment and more optimized nutrient management. This study aims to characterize the effects of microbial inoculants and varying nutrient levels in direct-seeded rice through pot experiments, with the goal of developing suitable microbial inoculants for field application.

## **METHODS**

The field experiment was conducted in the screen house at Ilocos Norte Research and Extension Center (INREC) at Brgy Tabug, City of Batac, Ilocos Norte. It was conducted during the dry season from January 29 to May 26, 2023. The experimental area has a coordinate of 18° 2'59.28" N and 120°33'3.61"E, 16.2 m above sea level. The soil characteristics used in the pot experiment have a heavy-textured soil, very low fertile soil with 0.33% OM, belonging to the San Fernando Soil Series. The soil was collected at the production area located at the Mariano Marcos State University Compound in Brgy. 16- Quiling Sur, City of Batac, Ilocos Norte. The test variety used was NSIC Rc192, known for its good performance under aerobic conditions.

The experimental design was a 2-factorial

Randomized Complete Block Design (RCBD) with 3 replications in a pot experiment. Four pots were allotted for the destructive sampling: two pots at the early reproductive stage for tissue analysis, plant biomass, mycorrhizal infection, and spore population; another two pots at the physiological maturity stage for tissue analysis, plant biomass, and yield and yield parameters. Factor A was assigned to six microbial inoculants: M1- control; M2-Bio-N; M3-BioGroe; M4-Mykovam; M5-MykoPlus; and M6-Avatar while Factor B was assigned to for nutrient levels: N1-0 %; N2-25 %; N3-50 %; and N4-75 % of NP + full K. Five different biofertilizers available in the Philippines used are shown their characteristics is given in Table 1. The Bio-N and BioGroe are bacterial-based biofertilizers, the Mykovam and Avatar are fungi-based, and the MykoPlus is a combination of bacteria and fungi strains of microorganisms. In addition, a Control treatment was used in which no biofertilizer was applied.

# **Establishment and Crop Maintenance of the Experimental Set-up**

The soil was sterilized in a steam process using a 200 L capacity steel drum, and a recycled can of vegetable oil with a 16 L capacity was used as shown in Figure 1a. The collected soils were put in a galvanized container and arranged inside the drum for sterilization (Figure 1b). Soil sterilization using the steam method with 4 hours of soil sterilization at 90-100 °C temperature was conducted. The sterilized soil samples were put in

a plastic pail with a 2.5-gallon capacity, with a uniform weight of 8 kg per pot. All processes in putting the soil samples inside the containers carefully to avoid contamination of the sterilized soil samples. Before microbial inoculation of the seed, surface sterilization was performed using 70 % ethanol for 60 seconds, followed by 5 % sodium hypochlorite for 10 minutes, rinsed with distilled water 10 times, and soaked for 12 hours. Inoculation for 1 hour using the different treatments. following the manufacturer's recommendations. The inoculated and noninoculated seeds were sown with five seeds per pot and irrigated immediately with 150 ml of water per pot and thinned, and kept one seedling per pot one week after emergence. Daily irrigation from germination to 60 DAS ranged from 250 ml to 300 ml/plant and increased to 600 ml/plant at 61 DAS to maturity. Irrigated in the morning and afternoon to expose the soil at approximately field capacity. Fertilizer was applied using the recommended rate of 100-7-40 kg/ha NPK and applied per pot following 0, 25, 50, and 75 % of NP + full K of the recommended rate.

The agro-climatic factors significantly impacting plant growth and development were carefully recorded. This includes daily temperature and relative humidity inside the screen house, which were measured using an Digital Electronic Thermometer. Additionally, daily data on solar radiation and rainfall were collected from the AgroMet station at MMSU (Figure 2).

**Table 1.** The profiles of microbial inoculants: Bio-N; BioGroe; Mykovam; MykoPlus and Avatar.

Microbial Inoculant	Active Organism	Mode of Action	Effect on Plant Growth
Bio-N®	Bacteria: Azospirillum lipoferum, A. brasilense	Nitrogen Fixer IAA production	Enhances shoot growth and root development of rice, corn, and vegetables Keeps plants healthy and green even during droughts and pest infestations
BioGroe ®	Bacteria: Bacillus sp.	Plant growth-promoting Rhizobacteria Produce plant hormones and provide nutrients in soluble form	Influence and promote root growth Protect plant surfaces from disease- causing microbes
Mykovam	Fungi: Gigaspora margarita, Glomus etunicatum and Gl. Macrocarpum	Facilitates the roots' absorption of water and nutrients (phosphorus)	Assist the plant in the absorption of water and nutrients.  Prevent root infection and increase plant tolerance to drought and heavy metals
MykoPlus ®	Fungi ( <i>Glomus</i> spp.) and Bacteria ( <i>Bacillus</i> spp.)	Endomycorrhiza (root-fungus symbiotic association) Nitrogen fixers, phosphorus solubilizers, and growth hormone secretors	Good seed germination, root development, better growth of plants, and increased grain and fruit yield
Avatar ®	Fungi: Trichoderma sp.	Colonize the plant root zone and increase fertilizer absorption and water uptake efficiency	Enhance plant growth



**Figure 1.** Materials used in the soil sterilization (a) A 16-liter capacity recycled vegetable oil can with a 2.5-inch diameter at the bottom and a perforated 3-inch diameter PVC pipe at the center, (b) a soil sample in a 200-liter capacity drum sterilized at 90-100 °C for 4 hours.

#### **Soil and Root Colonization**

The soil and root colonization were gathered at 60 DAS to characterize the effect of the microbial inoculants in response to varying nutrient levels in direct-seeded rice. The procedure used by Sakai et al (2024) for staining was followed, and microscopic observation was followed using the Truevision Trinocular Microscope, and the root colonization was rated using the 50 cm<sup>2</sup> gridline intersection method (Kakabouki et al. 2021).

### Agronomic data

Sample plants were gathered the agronomic data such as tiller growth at 30, 60, 90 DAS, and maturity; shoot dry weight at 60 DAS and maturity; root dry weight at maturity; number of days to flower, and grain yield. The root characteristics (root diameter, total root length, L-type, and S-type lateral roots) were gathered using an Epson Perfection V850 Pro Flatbed Photo Scanner and analyzed using the WinRHIZO software. After data gathering on the root characteristics, the root oven oven-dried and weighed for the root dry weight.

#### **Data Analysis**

The data gathered was analyzed using the R-CropStat Software R version 2.11.1, following the analysis of Factorial RCBD to determine the effect of the treatments. Treatment means were compared using the Least Significant Difference (LSD).

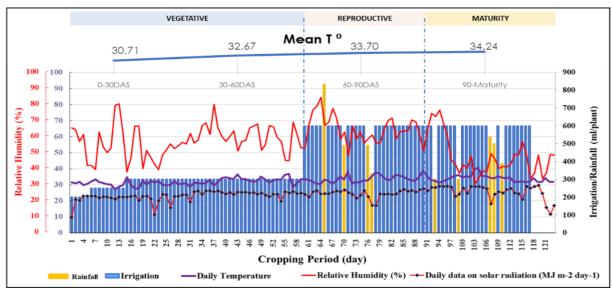
# RESULTS AND DISCUSSION

Agro-climatic Data.

Plants grown under aerobic conditions received 250-300 ml of water daily from germination to 60 days after sowing (DAS), with the amount increasing to 600 ml per plant from 61 DAS to maturity to keep soil moisture near field capacity (-33 kPa). During the reproductive stage, there were 9 instances of uncontrolled rainfall. adding 29.40-833.10 ml of water per plant per day (Figure 2). The average temperature during the first 30 DAS, 30-60 DAS, 60-90 DAS, and 90 DAS to maturity was gradually increased by 1-2 °C. During the flowering stage, temperatures ranged from 30.25 to 38.15 °C, with relative humidity between 47.50 % and 84.00 %. These unfavorable conditions. including temperatures and inconsistent water availability, led to heat stress, particularly during flowering, resulting in abnormal plant growth.

# Root and soil colonization

In evaluating soil colonization, Mykovam was the most effective fungi-based biofertilizer, increasing soil colony counts by 78 % compared to the control (Table 2). MykoPlus and Avatar showed no significant increase. A significant interaction was observed between microbial inoculation and nutrient levels, with Mykovam combined with 75 % NP and full K fertilizer showing the highest fungi colony count. For bacterial-based biofertilizers, BioGroe significantly increased colony counts by 128 %, while Bio-N also improved colonization. However, there was no significant interaction between bacterial inoculants and nutrient levels. Differences in colony growth appearance were noted 48 hours after inoculation.



**Figure 2**. Data on relative humidity (%) and daily temperature (°C) from the screen house, daily solar radiation (MJ m<sup>-2</sup> day<sup>-1</sup>), and rainfall (mm) taken from AgroMet Station throughout the growth stages of the rice planted in pots at the screen house, DS 2023.

For root colonization, effective fungal penetration was observed at 60 DAS, with MykoPlus, when combined with bacteria, promoting stronger colonization than single inoculants. This aligns with findings by Ouhaddou et al. (2024), who reported improved root colonization from dual AMF and PGPR inoculation under compost-amended Mediterranean conditions. Mycorrhizal fungi enable symbiosis by producing enzymes like

chitinase, glucanase, and polygalacturonase (Yang et al., 2023), while recent evidence highlights the importance of pectin-degrading enzymes (PMEs and PGs) in modifying host cell walls (Su, 2022). Moreover, the effectiveness of microbial inoculants in promoting root colonization and nutrient uptake is strongly influenced by soil nutrient status, with better outcomes in nutrient-deficient soils (Li et al., 2023).

**Table 2**. Colony count of fungi and bacteria in the soil, and % mycorrhization (colonization of mycorrhiza) in roots of direct-seeded rice as affected by microbial inoculation and nutrient levels in a pot experiment in Ilocos Norte, DS 2023.

levels in a pot experiment in nocos Norte, DS 2025.						
Treatment	•	t (CFU/ml) in the	* % Mycorrhization			
		soil	(hyphae and vesicles) in			
	Fungi	Bacteria	the root			
Microbial Inoculation	**	*	**			
No microbial inoculants	1,761.25 b	2,012.50 b	0.71 b			
Bio-N	-	3,512.50 ab	-			
BioGroe	-	4,591.25 a	-			
Mykovam	3,136.25 a	-	1.40 b			
MykoPlus	1,779.58 b	Not tested	2.92 a			
Avatar	1,492.50 c	-	2.63 a			
Nutrient Level	ns	ns	ns			
No fertilizer	2,210.00	2,318.89	1.65			
25 % of NP + full K	1,948.33	3,891.67	2.26			
50 % of NP + full K	1,853.75	3,661.00	1.88			
75 % of NP + full K	2,157.50	3,616.67	1.88			
Inoculation x N level	*	ns	ns			

<sup>\*\*-</sup> significant at 1 % level

ns- not significant

<sup>\*-</sup> significant at 5 % level

The means marked with the same letters within a column are not significantly different 1% and 5% levels of the Least Significant Difference Test.

#### Tiller Development.

Between 30 and 60 DAS, microbial inoculants, including MykoPlus, Bio-N, BioGroe, and Avatar, significantly increased the number of tillers and shoot dry weight (Table 3). However, this effect was not sustained until maturity. The decline in growth effects toward maturity was likely due to high temperatures and water stress, which can limit water uptake and reduce cell division, factors known to impair plant development under heat stress, as reviewed by Ahmad et al. (2022). Studies by Suralta et al. (2017) and Datta et al. (2023) have shown that microbial inoculants like PGPR strains in BioGroe

and *Trichoderma spp.* in Avatar can enhance tiller development under optimal conditions. From 60 to 90 DAS, increased temperatures and water stress required higher irrigation. Rainfall occurring at 65 DAS, which promoted additional tillering by prolonging the vegetative phase, aligns with the review by Kim et al. (2024), which emphasized that waterlogging caused by rainfall during the vegetative stage can delay phenological development in cereal crops.

Higher nutrient levels at 90 DAS also led to a significant increase in tillers, in line with the result of the study of Tiwari & Pandey (2018) on barley plants. At maturity, the number of tillers per hill doubled regardless of treatment, suggesting that heat stress during the reproductive stage may have induced additional tillering, as observed by Xu et al. (2022), where heat stress led to reallocating photosynthates to new tillers due to spikelet sterility.



Figure 3a. The overall view of the pot experiment was taken 55 days after sowing.



**Figure 3b.** View of the treatments of the study. Showing the effect of the growth response of the rice plant at 55 days after sowing. The treatments include Factor A (M1- no microbial inoculants, M2- Bio-N, M3- BioGroe, M4-Mykovam, M5-MykoPlus, M6-Avatar), and Factor B (N1- No fertilizer, N2- 25 % of NP + full K, N3-50 % of NP + full K, N4-75 % of NP + full K).

**Table 3.** Number of tillers per plant of direct seeded rice as affected by microbial inoculation and nutrient levels in a pot experiment in Ilocos Norte, DS 2023.

Treatments	Number Of Tillers				
	30DAS	60DAS	90DAS	Maturity	
Microbial Inoculation	**	**	ns	ns	
No microbial inoculants	1.04 b	4.98 b	9.00	14.33	
Bio-N	1.14 b	5.23 b	8.75	16.00	
BioGroe	1.09 b	5.81 b	9.50	15.17	
Mykovam	1.17 b	4.86 b	10.58	17.58	
MykoPlus	1.58 a	7.29 a	9.92	14.75	
Avatar	1.04 b	5.15 b	9.00	15.08	
Nutrient Level	ns	ns	**	**	
No fertilizer	1.21	5.29	8.33 c	12.89 b	
25 % of NP + full K	1.2	5.49	9.33 b	14.22 b	
50 % of NP + full K	1.14	5.65	9.89 b	17.22 a	
75 % of NP + full K	1.16	5.79	10.28 a	17.61 a	
Inoculation x N level	ns	ns	*	ns	

<sup>\*\*-</sup> significant at 1 % level

ns- not significant

The means marked with the same letters within a column are not significantly different 1% and 5% levels of the Least Significant Difference Test.

#### **Root Growth**

Root analysis of direct-seeded rice at maturity using the WinRHIZO revealed varying effects of microbial inoculants and nutrient levels on root morphology, including root diameter, total root length, and L-type and S-type lateral root (LR) lengths (Table 4). According to De Bauw et al. (2020), S-type roots are vital for phosphorus uptake under moist conditions, while L-type roots are more important during drought for acquiring both water and phosphorus. Watanabe et al. (2020) found that S-type roots maintain root hydraulic conductivity, whereas L-type roots, though structurally adaptable, have higher resistance and a limited role in water uptake. The study highlighted that microbial inoculation significantly influenced root development in rice, with BioGroe notably enhancing L-type lateral root length and producing the highest number of S-type lateral roots, resulting in the longest total root length, especially at 50 % NP + full K. This aligns with Naher et al. (2016), who found that mixed strains of Bacillus, Burkholderia, and Sphingomonas improved agronomic performance when combined with 50% NP + full K. The

bacteria facilitated nutrient uptake through organic acid production, aiding phosphorus solubilization and mobilization. Conversely, Avatar, containing Trichoderma sp., showed the lowest total root length but supported root growth under nutrient-limiting conditions, similar to findings by Singh et al. (2019). Mykovam enhanced root growth at higher nutrient levels, consistent with Hoseinzade et al. (2016), who observed significant growth improvements in rice with full nitrogen and phosphorus fertilizers when inoculated with AMF. The study underscores the variability in mycorrhizal responses depending on environmental factors like nutrient levels and the specific associations between plants and fungi, as noted by Pelegrino et al. (2015).

Root growth at the bottom of the pots, forming a waistline along the pot base, indicates continuous development in sync with aboveground growth. This aligns with findings by Li et al. (2022), who reported that improved root traits in modern rice cultivars enhanced both root and shoot biomass. Similarly, Sandhu et al. (2017) found that greater root biomass and nodal root number at early stages were linked to increased shoot dry matter and grain yield under alternate wetting and drying. These results support the view that enhanced root development promotes aboveground productivity.

<sup>\*-</sup> significant at 5 % level

**Table 4.** Root analysis from Win*RHIZO* software, such as root diameter, total root length, L-type, and S-type root length of direct seeded rice as affected by microbial inoculation and nutrient levels in a pot experiment in Ilocos Norte, DS 2023.

Treatment	Root Diameter (mm)	Total Root Length (cm)	L-type Root length (cm)	S-type Root Length (cm)	
Microbial Inoculation	ns	ns	*	ns	
No microbial inoculants	0.17	66,734.90	12,517.62 bc	42,453.60	
Bio-N	0.18	73,860.60	15,205.83 b	44,060.28	
BioGroe	0.17	84,197.95	16,853.87 a	52,462.98	
Mykovam	0.17	81,299.08	15,951.72 ab	51,266.08	
MykoPlus	0.17	67,526.08	13,238.87 bc	41,625.13	
Avatar	0.17	61,979.78	11,786.44 c	38,806.25	
Nutrient Level	*	ns	ns	ns	
No fertilizer	0.16 c	74,716.13	13,945.50	48,222.75	
25 % of NP + full K	0.18 a	68,949.52	14,819.87	39,654.48	
50 % of NP + full K	0.17 b	70,662.27	13,651.75	44,446.75	
75 % of NP + full K	0.17 b	76,071.00	14,619.11	48,125.55	
Inoculation x N level	**	*	ns	*	
CV %	9.66	5.93	3.74	3.05	

<sup>\*\*-</sup> significant at 1 % level

The means marked with the same letters within a column are not significantly different 1% and 5% level of the Least Significant Difference Test.

# **Shoot Dry Weight**

At 60 DAS, microbial inoculants significantly increased shoot dry weight, with MykoPlus showing a 36 % increase over the non-inoculated treatment, comparable to the effects of BioGroe, Bio-N, and Avatar (Table 5). The interaction between microbial inoculants and

nutrient levels was significant, with Bio-N and BioGroe treatments leading to higher biomass at specific nutrient levels, particularly 25 % and 75 % NP + full K. Mykovam and Avatar also enhanced biomass but at different nutrient levels, with Avatar being more effective at lower levels. In contrast, at maturity, there were no significant effects of microbial inoculation or nutrient levels on shoot dry weight, although BioGroe, Mykovam, and MykoPlus still showed a slight increase of 5-16 % over the control.

**Table 5.** Shoot, root, panicle, and total dry weight at 60 DAS and maturity, number of days to flower, and grain yield/plant of direct-seeded rice as affected by microbial inoculation and nutrient levels in a pot experiment in Ilocos Norte, DS 2023.

Treatment	Dry Weight At 60das (G/Plant)	•	Dry Weight At Maturity (G/Plant)			Grain Yield/plant (g/plant)
	Shoot	Shoot Root		Total	(DAS)	(C1 )
Microbial Inoculation	**	ns	*	ns	*	ns
No inoculants	3.46 b	25.08	4.02 b	61.26	87.33 ab	29.49
Bio-N	3.58 ab	25.17	4.47 ab	61.92	87.17 b	29.61
BioGroe	3.78 ab	26.30	4.54 ab	65.76	87.92 ab	32.25
Mykovam	3.17 b	29.11	5.48 a	68.44	86.50 b	31.18
MykoPlus	4.70 a	26.23	4.55 ab	63.77	89.00 a	30.32
Avatar	3.58 ab	24.82	4.08 ab	62.82	86.50 b	31.25
Nutrient Level	ns	ns	ns	ns	ns	*
No fertilizer	3.43	25.64	4.22	58.92	87.56	26.39 b
25 % of NP + full K	3.79	25.68	4.39	62.05	87.67	29.31 ab
50 % of NP + full K	3.66	26.83	4.78	67.24	87.00	32.95 a
75 % of NP + full K	3.96	26.33	4.69	67.77	87.39	34.08 a
Inoculation x N level	*	ns	ns	ns	ns	ns

<sup>\*\*-</sup> significant at 1 % level

<sup>\*-</sup> significant at 5 % level

ns- not significant

<sup>\*-</sup> significant at 5 % level

ns- not significant

The means marked with the same letters within a column are not significantly different 1% and 5% levels of Least Significant Difference Test.

# Heat stress induced by high temperature and water deficit affects yield

Although the pot experiment was maintained near traditional field capacity, the observed soil moisture potential of -33 kPa likely reflected a moisture deficit. Krueger and Ochsner (2024) suggest that field capacity is closer to -7.4 kPa. meaning the observed conditions may have caused water stress. This is supported by dos Santos et al. (2018), who found that rice growth declined between -46 and -56 kPa, with notable reductions in biomass, leaf area, and water use efficiency. Thus, the soil conditions may have imposed water stress, contributing to abnormal rice growth in the screen house. High temperatures from 30-60 DAS (32.7 °C), 60-90 DAS (33.7 °C), and 90-maturity (34.2 °C) further stressed the plants, extending the vegetative stage. Similar observations Sanwong et al. (2023) noted increased tillers overlapping into the reproductive stage. Water constraints during the vegetative stage delay panicle initiation, while drought from germination to flowering disrupts development in upland rice. Champness et al. (2023) found that increased soil moisture deficits delayed PI by up to 14 days in aerobic rice. Similarly, Waheed et al. (2023) reported that vegetative-stage drought significantly reduced key growth traits in Basmati rice, indicating delayed development and yield

Rice development, particularly during its reproductive stages, is adversely affected when temperatures exceed the optimal range of 25-30 °C. Shrestha et al. (2022) observed that temperatures above 28 °C disrupt physiological functions and elevate evapotranspiration rates, which can lead to moisture stress. Hu et al. (2021) found that temperatures exceeding 35 °C during panicle initiation and flowering impair pollen viability and hinder anther dehiscence, resulting in reduced spikelet fertility. Additionally, Zhang et al. (2018) reported that heat stress hampers pollen tube growth by disrupting auxin balance, further contributing to sterility. These findings align with the present study's observation that prolonged exposure to high temperatures, specifically 27 days above 30 °C and 9 days exceeding 35 °C during the crucial 30-60 DAS period and reproductive phase, leads to increased spikelet sterility, particularly under aerobic

cultivation.

Ultimately, the study resulted in high spikelet sterility and lower grain yield (Table 5). Microbial inoculant selection was based on the formula by Won et al. (2020), considering the sink capacity based on NSIC Rc192 seed size (1000-grain weight). Although the effect of microbial inoculation was not significant, plants applied with BioGroe performed numerically higher than those applied with Bio-N, and Avatar, and outperformed those with Mykovam MykoPlus. Mykovam-treated plants showed more tillers and shoot dry weight, but fewer paniclebearing tillers, due to insufficient photosynthates to allow grain filling. Plants with the highest nutrient levels had the most spikelets per plant and the highest grain yield. Increased nitrogen fertilization significantly enhanced grain yield by expanding source and sink capacity (Fageria and Santos 2015).

The study presents a novel approach by integrating microbial inoculants with reduced chemical fertilizer inputs in aerobic rice cultivation, a method still underutilized due to challenges in establishment and yield. It evaluates the combined effects of five microbial inoculants (Bio-N, BioGroe, Mykovam, MykoPlus, and Avatar) with varying fertilizer levels under waterlimited and high-temperature conditions. Using WinRHIZO imaging, the study provides precise root architecture analysis, particularly of L-type and S-type lateral roots, offering valuable insights into root and soil colonization. A key finding is the potential of BioGroe combined with 50 percent NP plus full K to maintain growth while reducing synthetic fertilizer use.

Scientifically, the research enhances understanding of root-microbe interactions in aerobic systems, showing how specific inoculants influence root colonization and nutrient uptake under stress. It provides empirical evidence on inoculant performance across nutrient levels, supporting microbial screening under abiotic conditions. The study also demonstrates the potential to reduce chemical inputs without compromising yield, contributing to sustainable crop production and improved soil health.

From a societal perspective, the findings promote sustainable agriculture by supporting low-input, climate-resilient systems and addressing the environmental impact of excessive fertilizer use. In drought-prone areas like Ilocos Norte, microbial amendments can improve water and nutrient use efficiency, aiding climate change adaptation. This approach empowers smallholder

farmers by lowering input costs and enhancing productivity through accessible, eco-friendly solutions, ultimately contributing to improved rice production and food security in stress-prone environments.

#### **CONCLUSION**

The study demonstrated that specific microbial inoculants, particularly BioGroe, significantly improved root development, tiller production, and shoot biomass in direct-seeded rice, especially when combined with 50% NP + full K fertilizer. While environmental stress limited grain yield, the findings suggest that microbial inoculants can enhance plant growth and reduce fertilizer dependence, contributing to more sustainable and efficient rice production under aerobic conditions.

The research underscores the need for further studies under field conditions to optimize microbial inoculant use and nutrient management, contributing to the broader field of sustainable agriculture by identifying strategies to improve crop productivity and resilience under stress conditions.

#### **ACKNOWLEDGEMENT**

The researchers extend their sincere gratitude to the DA-RFO I Ilocos Norte Research & Experiment Center (DA-INREC) Batac Station, the Philippine Rice Research Institute (PhilRice) Batac Branch, the Crop Biotechnology Center at PhilRice-Central Experimental Station, the National Tobacco Administration (NTA) Batac Station, and the MMSU Research Directorate-Soil and Water Laboratory for their invaluable support and for providing the facilities necessary to conduct this study. Special acknowledgment is given to the National Bioenergy Research and Innovation Center (NBERIC) team for their assistance and for granting access to their laboratory and equipment. The researchers also express their deepest appreciation to the Department of Science and Technology -Accelerated Science and Technology Human (DOST-Resource Development Program ASTHRDP) for their financial support for this research.

# INFORMED CONSENT STATEMENT

All experimental procedures involving live animals were conducted in accordance with ethical standards, including those outlined in the Declaration of Helsinki.

## **AUTHOR CONTRIBUTION STATEMENT**

LGI conceptualized the study, led the experimental design, conducted the fieldwork, collected and analyzed the data, and prepared the initial manuscript draft. MLSE, guided the methodology and statistical analysis, contributed to manuscript revision interpretation of results. PCSC supervised the scientific framing of the research, assisted in data validation, and reviewed and edited the manuscript. ETMO contributed to data analysis, literature review, and editing of the manuscript. PBS advised on soil and microbial aspects of the study and reviewed the final draft for accuracy and clarity. All authors read and approved the final version of the manuscript.

#### CONFLICT OF INTEREST STATEMENT

The authors declare that they have no conflict of interest regarding the publication of this paper.

# USE OF ARTIFICIAL INTELLIGENCE (AI)-ASSISTED TECHNOLOGY

The authors declare that no artificial intelligence (AI) tools were used in the generation, analysis, or writing of this manuscript. All aspects of the research, including data collection, interpretation, and manuscript preparation, were carried out entirely by the authors without the assistance of AI-based technologies

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