

The logo features a green gear-like shape on the left. Inside the gear, there is a stylized rice plant with a red DNA double helix and a blue molecular structure. The year '2015' is written in small text at the bottom left of the gear.

RICE-BASED BIOSYSTEMS JOURNAL

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Philippine Rice Research Institute
Central Experiment Station
Maligaya, Science City of Muñoz, 3119 Nueva Ecija



ABOUT THE COVER

This issue features alternative strategies to enhance crop yield and soil health through the application of Silicon, peanut hulls, irradiated carrageenan, and fermented corn cob as source of fertilizer to enhance crop yield performance. It addresses the threat of fall armyworm in rice cultivation and the use of extracts from *Aratiles* leaves as a bio-insecticide against rice weevils. As the journal continues to serve its purpose in producing research studies in rice farming, it covers evaluation of farmers' practices in response to climate change and ways to address agricultural challenges. Some of the challenges include the identification of restorer lines for hybrid rice and germplasm with deep-rooting ability and root plasticity in developing new inbred rice varieties.

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Abstract

Drought stress can significantly affect plant growth and nutrient uptake, including that of phosphorus (P) by reducing its availability and mobility in the soil and the efficiency of the root system to acquire and transport phosphorus to the shoots. This study examined the contribution of root plasticity expression to P uptake under drought conditions and its contribution to increasing dry matter production under water-limited soil conditions. The two rice varieties, IR64 (irrigated lowland) and NSIC Rc 194 (IR64-Sub1, Submarino1), were grown under drought conditions and applied with various phosphorus rates (0, 40, and 80 kg ha⁻¹). The results showed that the shoot dry weight of NSIC Rc 194 responded to phosphorus application while that of IR64 did not. The root system development based on total root length also responded positively to phosphorus application in NSIC Rc 194 but not in IR64. These differences in root system development greatly contributed to their genotypic differences in water use and shoot P uptake under drought, which was higher in NSIC Rc 194 than in IR64. The results imply that the genotype with a better ability to maintain and/or increase root system development under drought has a better ability to maintain high water and P uptake to support its root growth and dry matter production. This also suggests that rice productivity under rainfed lowlands can be enhanced through the interaction of root plasticity and P fertilization.

Keywords: Drought, Dry matter production, P uptake, Root plasticity, Water use

Introduction

Rice is among the three most important grain crops in the world, along with wheat and corn (Chauhan et al., 2017). Its production worldwide has been growing steadily, especially in Asia, which accounted for 92% of the world's rice production, 60% of the global population, and 90% of global rice consumption (FAO, 2009). Rice is a luxurious user of water requiring almost 80% of the total irrigation freshwater resources (Henry et al., 2016); and thus, the availability and accessibility of water greatly influence its production.

Phosphorus, a vital macronutrient, plays a crucial role in rice plant growth and development. It is implicated in several physiological processes, including photosynthesis, respiration, and energy transfer (Khan et al., 2023; Richardson et al., 2011). However, P availability can be constrained in numerous agricultural soils due to low fertility and the impacts of climate change, such as drought and water

scarcity. As the planet continues to warm, the effects of climate change are expected to intensify (FAO, 2006; ASEAN, 2021), influencing the uptake and utilization of nutrients, including phosphorus. This can further amplify the adverse impact of drought on rice growth and yield. Drought impedes plant growth by diminishing nutrient uptake, transport, and redistribution (Anjum et al., 2017; Cramer et al., 2009), including available P. This reduction is due to decreased root growth and development, a smaller root surface area, limited phosphorus availability in the soil, and an imbalance between phosphorus and other nutrients. The soil's low P mobility leads to its deficiency, triggering various changes in plant physiology, morphology, and biochemistry. As soil moisture decreases, plant P uptake also declines (Sardans and Peñuelas, 2007).

Several studies on the interaction of phosphorus availability and drought stress on rice showed that P application can improve the growth and yield of rice under drought by increasing the activity of

antioxidant enzymes and reducing oxidative damage (Veronica et al., 2017; Ashraf et al., 2011) and water-use efficiency through enhanced photosynthesis and reduced water loss (Ahanger et al., 2016). Root plasticity, or the ability of a genotype to change its phenotype under changing soil moisture conditions, is key for rice adaptation to water stresses (Kano-Nakata et al., 2013; Bardgett et al., 2014; Suralta et al., 2018a). A genotype that exhibits more plastic root systems under drought than the others (Lynch and Brown, 2008; Kano-Nakata et al., 2011; Niones et al., 2012) will likely perform better and produce more biomass and yield under water-limited and nutrient-poor soils (Sandhu et al., 2016; Suralta et al., 2018a; Correa et al., 2019; Lucob-Agustin et al., 2020). The plant root-foraging patterns emerging from the interactions of soil nutrient distribution can significantly affect nutrient acquisition as well as plant growth and yield (Zhang et al., 2016; Suralta et al., 2018b; Helliwell et al., 2019). Under mild drought conditions, rice plants can demonstrate root plasticity through enhanced root proliferation. This process involves the development of additional lateral roots to explore a larger soil volume, thereby increasing the uptake of water and nutrients, including P (Deng et al., 2020). This response can subsequently improve the plant's growth and yield (Suralta et al., 2018a). Conversely, under severe drought conditions, rice plants may exhibit a different form of root plasticity through promoted root elongation. This involves the growth of longer and deeper roots to access nutrients and water from deeper soil layers (Farooq et al., 2012). These responses can also enhance P uptake, although this may be limited by the soil's P availability. Studies on nutrient behavior in soils, along with the morphological and physiological functions of root systems, will further aid in elucidating the mechanism of plant nutrient uptake and transport by roots. Consequently, this could provide a viable opportunity for genetically improving crop productivity in nutrient-deficient soils (Suralta et al., 2012; Zhang et al., 2020; Pandey et al., 2021).

Drought stress can significantly impact plant growth and nutrient uptake including P by reducing its availability and mobility in the soil and the root system's efficiency to acquire and transport phosphorus to the shoots. It has been hypothesized that rice plants exhibiting larger root system development under drought conditions may enhance phosphorus uptake from water-limited soil when the rate of P fertilizer application is increased. Therefore, this study investigates the role of root plasticity expression in phosphorus uptake under drought conditions and its contribution to augmenting dry matter production under water-limited soil conditions.

Materials and Methods

A pot experiment was conducted under the rain-out shelter at the Mariano Marcos State University (MMSU), Batac City, Ilocos Norte, (between 18° 03' North latitude and 120° 33' East longitude). The highest solar radiation was observed during April and May but usually extended until the start of the third quarter of the year. Data from the agrometeorological station of MMSU, Philippine Atmospheric, Geophysical and Astronomical Services Administration (PAGASA), and Philippine Council for Agriculture, Aquatic and Natural Resources Research and Development (PCAARRD) show that the average annual rainfall was 2000 mm while the average relative humidity (RH) was 82%. The average temperature and RH were 26.6°C and 81.6%, respectively. The average temperature inside the rainout shelter was 29.7°C while the RH was 71.7%.

Experimental Design and Treatments

The experiment was laid out in a split plot arranged in a completely randomized design (CRD) with four replications. The two rice varieties: IR64 (irrigated lowland) and NSIC Rc 194 (IR64-Sub1, Submarinol), were assigned as the main plots while the P treatments (0, 40, and 80 kg ha⁻¹) were assigned as subplots.

The recommended rate of P application for rice production at 40 kg ha⁻¹ was considered optimum. The experiment did not explore other recommended rates for optimum growth but rather the response or functions of roots under low P and high P.

Cultural Management Practices

The soil used has a heavy texture with 7.6 pH, low in nitrogen, moderately low in P, and sufficient in K. It was air-dried, pulverized, and sieved. After which, 4 kg soil was placed in each pot premixed with fertilizers such as ammonium sulfate (21-0-0) and muriate of potash (0-0-60) fertilizers as sources of N and K at 120 and 40 kg ha⁻¹, respectively. On the other hand, solophos (0-20-0) was used as a P source and applied at variable rates (0, 40, and 80 kg ha⁻¹). All pots were watered to set the soil moisture at 24% (v/v).

The seeds of the two rice genotypes were pre-germinated separately by soaking in water for 24 h. After 24 h, the seeds were sown in moistened tissue paper and incubated for 48 h. Then, three pre-germinated seeds of each genotype were sown in each pot (8 in x 7 in) containing soil saturated with water at 24% soil moisture content (SMC). The seedlings were thinned to one seedling per pot at 6 days after sowing (DAS).

Drought Imposition

The soil in each pot was initially saturated with water (24% SMC) and maintained at that level of SMC from the day of sowing up to 7 DAS. Thereafter, watering was withheld at 8 DAS, and the soil was allowed to dry down to 10% SMC and maintained at that level for 26 DAS. However, all plants started to wilt; thus, the target SMC for drought was adjusted from 26 to 47 DAS at 12%. Pots were weighed every afternoon using a digital balance to record the wet mass of the soil. The SMC (% by mass) in each pot was calculated as the ratio between the water mass and the air-dry mass (4 kg) of soil. Once the target SMC of 10% and subsequently 12% was reached, pots were watered to replace the amount of water lost and maintain the desired SMC.

Water Use Shoot, and Root Measurements

The water use (WU) or whole plant transpiration from 1 - 47 DAS was determined by comparing the accumulated daily water loss in a pot with a plant and another pot without a plant. The water loss in the pot without the plant served as the estimated total evaporation, while that of the pot with the plant served as the estimated total evapotranspiration. The water loss in the pot with the plant was subtracted from the water loss in the pot without a plant and the difference obtained was used as the estimated WU or transpiration of the whole plant.

Before shoot sampling, the leaf area was measured, expressed as the leaf area index (LAI), on all leaves at 40 DAS. The LAI (cm²) was estimated by multiplying the product of leaf length and width with a correction factor of 0.8.

The shoots were cut at ground level and oven-dried at 70°C for 48 h before the recording of the shoot dry weight (SDW). Thereafter, the whole root system was sampled at 49 DAS by carefully extracting the roots following Suralta et al. (2018c) method. The collected roots were gently washed free of soil in running water. Cleaned root samples were stored in FAA (formalin: acetic acid: 70% ethanol in 1:1:18 ratio by volume) solution to further measure various root components. Each root system was stained in 0.25% Coomassie Brilliant Blue R 250 aqueous solution for 48 h to get high-resolution digital photographs of the entire root system including fine lateral roots. After staining, the root samples were held gently with running water to remove excess stains.

The number of nodal (NR) roots was counted at the base of the plant. A scanner with a resolution of 300 dpi and an output format of 256 greyscales was used to take digitized images. For root length measurements, root samples were cut and evenly spread on the transparent sheets with minimal

overlapping, and then the digital images were taken using an Epson scanner (ES2200) at 300 dpi resolution. Scanned images were analyzed for total root length (TRL) using WinRhizo v. 2007d (Régent Instruments, Québec, Canada) with a pixel threshold value set at 175. The total lateral root length (TLRL) was estimated as the total length of roots with ≤ 0.2 mm in diameter (Yamauchi et al., 1987). The total nodal root length (TNRL) was computed as the difference between the total root length (TRL) and TLRL. The mean nodal root length was measured as the ratio between the total nodal root length and the number of nodal roots of each plant.

The water use efficiency was calculated as the ratio between the shoot dry weight and the total water use of each plant.

Statistical Analysis

The ANOVA for main treatment effects and their interactions were analyzed using StatExcel. The treatment mean differences were tested using Tukey's HSD at 5% level of significance. Correlations among traits were conducted using Pearson correlation analysis.

Results and Discussion

Shoot Growth and Development

There was a significant interaction between variety and phosphorus treatments on SDW, leaf area, accumulated water use, water use efficiency (WUE), and phosphorus uptake (Table 1). The SDW, leaf area, accumulated water use, WUE, and P uptake of IR64 were not affected or even significantly decreased with the increase in P application rates. On the other hand, the SDW, accumulated water use, WUE, and P uptake of NSIC Rc 194 were significantly increased at 40 kg ha⁻¹, relative to no P control. Further, an increase in P rate from 40 to 80 kg ha⁻¹, did not increase the above parameters, which either resulted in similar values with 40 kg ha⁻¹ P rate or decreased back to the level of the control. Furthermore, leaf area in NSIC Rc 194 was not affected by any of the P application rates, relative to the no P control.

Root growth and development

A significant interaction between the variety and P treatments was significant on most root traits except the number of nodal roots per plant (Table 2). The total root length (TRL), total nodal root length (TNRL), and total lateral root length (TLRL) were decreased with every P rate increment in IR64 while it was increased with P rates in NSIC Rc 194, relative to the no P control. For different root traits, the number of nodal roots per plant was not significantly affected by P rates in IR64 while it was increased in

NSIC Rc 194, especially at 80 P. The mean nodal root length (MNRL) was decreased with P rates in IR64 especially at 80P while it was increased at 40 P NSIC Rc 194, relative to the no P control.

The relationship among shoot dry weight with water use, shoot P uptake, water use efficiency, and various root lengths

The correlation values between water use and shoot dry weight (Figure 1a), shoot P uptake and shoot dry weight (Figure 1b), water use efficiency and shoot dry weight (Figure 1c), and total root length and shoot dry weight (Figure 1d) was strong, positively linear and significant both by variety and P rate factor. The correlation values between water use and shoot P uptake (Figure 2a), total root length and shoot P uptake (Figure 2b) mean nodal root length and shoot P uptake (Figure 2c), and the total lateral root length and shoot P uptake (Figure 2d) were strong, positively linear, and significant both by variety and P rate factor. Furthermore, the correlation values between mean nodal root length and water use (Figure 3a) and total root length and water use (Figure 3b) were strong, positively linear, and significant by variety and P rate factor.

Overall, the findings showed that the genotype with better root system and developmental responses to drought and greater response to P application as exhibited by NSIC Rc 194 showed greater shoot growth than IR64, a genotype with poor root growth under drought. In NSIC Rc 194, the total root length increased with the increase in P rate, especially at optimum P (40 P); thereby, increasing its water use and shoot P uptake, and consequently shoot growth (Table 1). On the other hand, in IR64, the total root length had a negative response to the increase in P which consequently showed a reduction in its

water use, shoot P uptake, and shoot biomass. The differences in root system developmental responses of the two genotypes based on total root length can be explained better with their differences in total lateral root length (Table 2).

The capacity of NSIC Rc 194 to retain greater root length can compensate for the limited water supply in the soil during drought with optimal P (40 P) by enhancing water extraction through increased root surface area (Koevoets et al., 2016; Suralta et al., 2010, 2018a). This resulted in the maintenance of a higher accumulated water use during drought (Kano-Nakata et al., 2011; Niones et al., 2012; Menge et al., 2016) as exhibited by NSIC Rc 194 in response to increased P rate. Consequently, the differences in dry matter production in response to increasing P rates under drought conditions between NSIC Rc 194 and IR64 can be attributed to variations in their root system development. NSIC Rc 194 exhibited more dry matter production than IR64, which is linked to its accelerated root growth. This growth is a result of sustained production of growth hormones and substances like cytokinins, crucial for growth and leaf functions (Fageria and Moreira, 2011). The improve growth of NSIC Rc 194 under drought conditions with varying P rates follows a well-defined mechanism for coping with low P availability stress (Ozturk et al., 2005). Root length, which is associated with P uptake (Lynch and Brown, 2008), allows for the exploration of a larger soil volume through an extensive branched root system. This system is characterized by increased production and length of lateral roots, finer roots, root hairs proliferation, and cluster roots formation (Lynch, 2005; Lambers et al., 2006). Thus, NSIC Rc 194 is an efficient responder to P rate under drought conditions, whereas IR64 is an inefficient non-responder (Gerloff, 1976).

Table 1. Shoot dry weight, leaf area, accumulated water use, water use efficiency, and shoot P uptake of IR64 and NSIC Rc 194 in response to drought conditions with various rates of phosphorus application.

Variety	Phosphorus Rate (kg ha ⁻¹)	Shoot Dry Weight, SDW (mgplant ⁻¹)	Leaf Area (cm ² plant)	Accumulated Water Use, WU (g plant ⁻¹)	Water Use Efficiency (g SDWg ⁻¹ WU)	Shoot Phosphorus Uptake (g)
IR64	0	0.15 c	21.7 a	1466.7 ab	102.4 c	0.29 c
	40	0.14 c	18.9 b	1445.0 b	95.6 c	0.23 d
	80	0.10 d	14.6 c	1441.7 b	72.2 d	0.14 e
NSIC Rc 194	0	0.20 b	21.6 a	1450.0 ab	139.7 b	0.42 b
	40	0.22 a	22.9 a	1460.0 a	153.3 a	0.53 a
	80	0.22 ab	21.8 a	1455.0 a	149.2 ab	0.46 b
Variety (A)		**	**	ns	**	**
Phosphorus (B)		**	**	**	**	**
A x B		**	**	**	**	**
CV (%)		3.9	5.2	0.2	3.9	6.0

Data are means of three replicates. ns, not significant; **, significant at P0.01.

Table 2. Number and length of roots and branching index of IR64 and NSIC Rc 194 in response to drought conditions with various rates of phosphorus application.

Variety	Phosphorous Rate (kg ha ⁻¹)	Nodal Roots (number plant ⁻¹)	Total Nodal Root Length (cm)	Total Lateral Root Length (cm)	Total Root Length (cm)	Mean Nodal Root Length (cm)	Branching Index (LRL cm ⁻¹ NRL)
IR64	0	13.0 b	143.4 c	1128.2 c	1271.6 c	11.4 bc	7.9 c
	40	11.7 b	104.9 d	1010.1 d	1114.9 d	9.1 cd	9.6 a
	80	12.0 b	90.5 e	767.0 e	857.5 e	7.8 d	8.5 b
NSIC Rc 194	0	12.0 b	144.5 c	1103.5 c	1248.1 c	12.2 b	7.6 c
	40	13.5 b	240.9 a	1540.3 a	1781.2 a	17.9 a	6.4 d
	80	17.5 a	231.4 b	1460.2 b	1691.6 b	13.2 b	6.5 d
Variety (A)		ns	**	**	**	**	**
Phosphorus (B)		ns	**	**	**	ns	ns
A x B		ns	**	**	**	**	**
CV (%)		11.1	0.8	2.4	2.2	10.3	3.3

ns, not significant; **, significant at P<0.01.

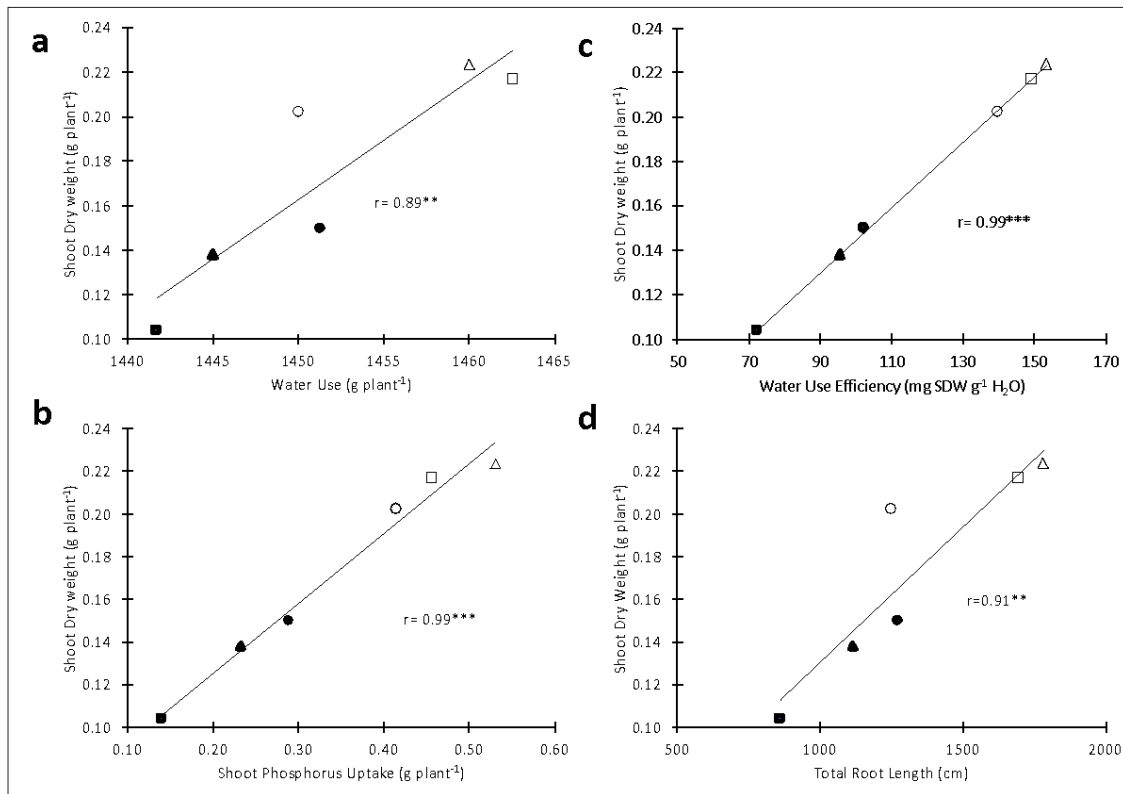


Figure 1. The relationship between water use and shoot dry weight (a) shoot P uptake and shoot dry weight (b), water use efficiency and shoot dry weight (c), and total root length and shoot dry weight of two rice varieties (IR64, shaded; NSIC Rc 194, open) under drought conditions with various rates (0, circle; 40, triangle, and 80, square) of phosphorus (kg ha⁻¹) application. Data are means of three replicates. ** and *** are significant at $p < 0.01$ and $p < 0.001$, respectively.

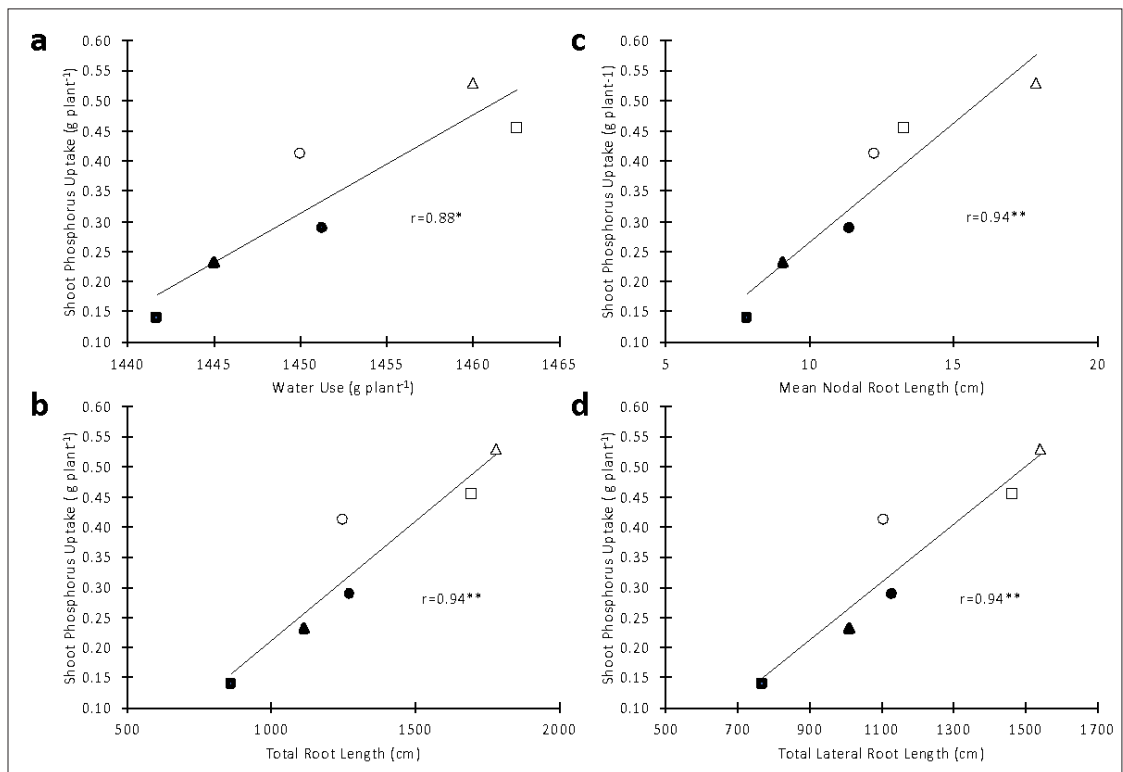


Figure 2. The relationship between water use and shoot P uptake (a) total lateral root length and shoot P uptake (b), mean nodal root length and shoot P uptake (c), and total lateral root length and shoot P uptake (d) of two rice varieties (IR64, shaded; NSIC Rc194, open) grown under drought conditions with various rates (0, circle; 40, triangle and 80, square) of phosphorus (kg ha⁻¹) application. Data are means of three replicates. ** and *** are significant at $p < 0.01$ and $p < 0.001$, respectively.

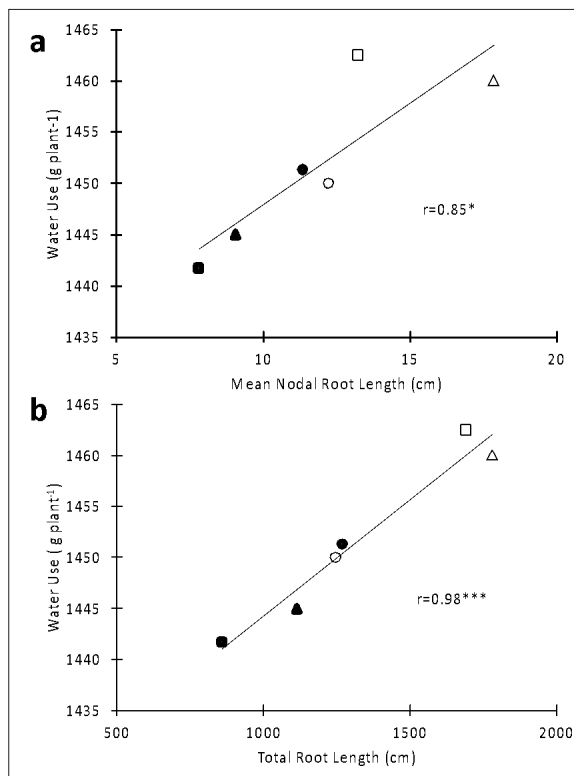


Figure 3. The relationship between mean nodal root length and water use (a), and total root length and water use (b) of two rice varieties (IR64, shaded; NSIC Rc 194, open) grown under drought conditions with various rates (0, circle; 40, triangle, and 80, square) of phosphorus (kg ha^{-1}) application. Data are means of three replicates. * and ** are significant at $p < 0.01$ and $p < 0.001$, respectively.

Interestingly, IR64 showed a reduction in shoot growth with increasing P (Table 1) possibly because high root branching in a limited soil volume may make the roots too close to each other which reduces P uptake efficiency (Lambers et al., 2006). It might be also due to nutritional imbalance such that excessive P application can induce nutritional imbalances within the plant (Lopez-Bucio et al., 2003), resulting in an antagonistic nutrient reaction limiting the nutrient availability (Ozturk et al., 2005) and impair metabolic processes by interfering with the intake of other necessary elements (Williamson et al., 2001; Lopez-Bucio et al., 2003); thus, limits shoot development and dry weight (Lynch, 2007; Lambers et al., 2006; Vance et al., 2003). Although P application promotes root growth and development (Fitter, 2002; Fageria, 2013), the excessive phosphorus application in the case of IR64 might result in tight and compact roots with restricted access to water and nutrients under drought which cannot support optimal shoot development, resulting in the reduction of its shoot dry weight.

Conclusion

This study showed that a genotype that can maintain and/or increase root system development based on total root length has a better ability to maintain high water and P uptake during drought to support root growth and dry matter production. This suggests that rice productivity under rainfed condition can be enhanced by having rice varieties with better root plasticity and enough P fertilization. Optimum P application to enhance root plasticity response and maintain dry matter production in tolerant rice genotypes to drought prone condition is 40 kg ha^{-1} . However, the timing of application must be carefully considered to maximize screening genotypes for P uptake efficiency and identifying P efficiency mechanisms at the physiological and molecular levels. There is still much to learn about the role of root development in natural systems with varying nutrient availability, and it is hoped that new understanding based on examinations of such systems will increase the ability to breed rice with high root plasticity expression and utilize P more efficiently under drought-stress conditions.

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IDENTIFICATION OF POTENTIAL HYBRID RESTORER AND MAINTAINER LINES THROUGH PHENOTYPIC AND MOLECULAR SCREENING OF SELECTED RICE GENOTYPES

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Abstract

Identifying effective restorers and maintainers is the initial steps in three-line heterosis breeding. Heterosis leads to yield increase, reproductive ability, adaptability, resistance to disease and insect, general vigour, and quality of crops (Begna, 2020); thus, exploring this phenomenon is a great advantage in crop improvement particularly in rice breeding. The phenotypic and molecular screening of F_1 testcross progenies in Philippine Rice Research Institute (PhilRice) were examined through pollen evaluation to identify potential hybrid restorer and maintainer lines. This study assessed the presence of fertility restoring genes in 93 male parents using gene specific molecular markers, PRR9-1 and SF21-5 for *Rf4* and *Rf3* genes, correspondingly. Phenotypic data, in terms of pollen fertility of the F_1 testcross progenies, shows that 19.49% of the evaluated pollen source genotypes are restorers, and 49.23% are partial restorers. Molecular characterization demonstrated that 23 of the 93 elite lines contain *Rf4*, *Rf3* or both fertility restorer genes. In the F_1 testcross progeny pollen fertility assessment, 15 genotypes were identified as restorers in the absence of *Rf4* and *Rf3* genes, indicating the significant role of modifiers in fertility restoration. This warrants further study. Genotypes possessing both *Rf4* and *Rf3* yield more robust restorers than those with just one fertility restoring gene. The identified fertility restorers, present in either one or both genes in male parents, could be used to develop new hybrids with desirable traits.

Keywords: *F₁ hybrids, Restorer lines, Fertility restoring genes, Pollen evaluation*

Introduction

Rice serves as the primary dietary staple in Asia and parts of the Pacific, with over 90% of the world's rice being produced and consumed within the Asia-Pacific region. On average, annual rice production in these five Southeast Asian countries totals 128.5 Mt, with a harvested area of 32.0 million ha. This represents approximately 17% of global rice production and 20% of the total harvested area worldwide. China and India are considered as the main producers of paddy rice worldwide. In 2022, China's paddy rice production amounted to over 208.4 Mmt, while India's production volume exceeded 196 Mmt (FAOSTAT, 2022). Ricelytics from the Philippine Rice Research Institute (PhilRice) show that the Philippines has a total rough rice production of 20.6 Mmt in 2023 from 4.82 million ha of rice area.

The utilization of modern hybrid technology in sustaining global rice production has resulted in a high increase in rice yield. Hybrid rice has a yield advantage of 15 - 20% over the conventional high yielding varieties (Virmani and Kumar, 2004). Thus, hybrid rice has been world-widely introduced in different rice farming countries.

In hybrid rice seed production, the three-line

hybrid or CGMS (cytoplasmic genetic male sterile) system combines three essential lines: a cytoplasmic male sterile (CMS) line, a maintainer line, and a restorer line carrying the restorer gene (*Rf*). These lines play an indispensable role and contribute significantly to the development of hybrid rice varieties. CMS is widely used in hybrid rice seed production and breeding programs because it produces non-functional pollen or sterile pollen caused by incompatibility of nucleus and cytoplasm, inherited maternally (Kaul, 2012). Male fertility in plants with CMS is restored by nuclear fertility restorer genes. In rice, a variable number of fertility restorer genes can completely restore the fertility of a certain CMS line. For instance, the wild abortive (WA) CMS derived from *O. sativa f. spontanea* is extensively used in commercial seed production (Virmani, 1996). Although different number of restorer genes have been proposed in various restorer lines, one or two dominant restorer alleles (*Rf3* and *Rf4*) are usually suggested to be responsible for the fertility of WA-CMS (Yao et al., 1997; Tan et al., 1998).

Initial steps in three-line hybrid breeding (Siddiq, 1996) include identifying maintainers and restorers from elite breeding lines and landraces through test crossing (Virmani, 1996) and their use in further

breeding programs. To develop stable hybrids, it is essential to select parents that consist of stable restorer and stable CMS lines. Test cross activities help identify maintainers as well as restorers. Screening fertility restoration trait is laborious and time consuming as it involves test crossing with a set of CMS lines together with the evaluation of F_1 for pollen and spikelet fertility. Molecular markers are becoming increasingly useful in enhancing the efficiency in crop improvement (Singh et al., 2014). Revathi et al. (2013) reported that molecular markers are useful tools in evaluating fertility restoration traits of a large number of germplasm or breeding lines. Most investigated fertility restoration is governed by two independent dominant major genes. The chromosomal location of *Rf* genes namely *Rf4* and *Rf3* was determined through molecular analysis in chromosome 10 and 1, respectively (Zhang et al., 1997).

This study evaluated the pollen fertility of F_1 testcross progenies from cross between six CMS lines and male parents; assessed the presence of fertility restoring genes among the selected male parents through gene specific molecular markers; and identified potential restorer and maintainer lines based on the presence of fertility restoring genes in the pollen of their F_1 testcrosses.

Materials and methods

Plant Materials

During the 2017 wet season, a panel of 93 rice genotypes, consisting of elite breeding lines without prior information about fertility restoration status, was developed using the pedigree method, in-vitro and anther-culture techniques, Marker Assisted Selection (MAS), and seed mutation. These genotypes were then crossed with six CMS lines – namely, IR58025A, PR15A, PR19, PR21A, PR27A, and PR29A – carrying the WA cytoplasm. Subsequently, 195 F_1 testcross progenies, along with their respective male parents, were transplanted with one seedling per hill and evaluated in the testcross nursery of the Plant Breeding and Biotechnology Division (PBBD) at PhilRice during the 20218 dry season.

Pollen Evaluation and Classification

Three immature panicles or boots of F_1 testcross progenies from the test cross nursery of PhilRice were obtained 1 - 3 days prior to anthesis early in the morning just before blooming and are placed in a bucket. Three spikelets were sampled from each panicle and stored temporarily in a 2-ml plastic tube containing 95% ethanol. Anthers from each spikelet representing lower, middle, and top portion of the panicles were collected using a pair of forceps and placed in a glass slide with a drop of 1% Iodine-

potassium iodide (I-KI) solution (Virmani et al., 1997). The anthers were gently crushed using the forceps to release the pollen. Anthers of each spikelet were stained with a 1% Iodine-potassium (I-KI) solution (Virmani et al., 1997).

Pollen fertility was assessed manually, with colored, well-filled, and spherical pollen grains indicating fertility. In contrast, sterile pollen grains were unstained, shrunken, and devoid of pollen (Figure 1 and Table 1). Pollen fertility was calculated and expressed as a percentage using the formula below:

$$\text{Pollen fertility \%} = \frac{\text{No of stained pollen grains}}{\text{Total no. of pollen grains examined}} \times 100$$

Potential maintainer and restorer lines were classified based on pollen fertility and spikelet fertility of experimental hybrids (Table 2). Selected restorer lines should have more than 80% pollen fertility. For the potential maintainer lines, selection was based on their ability to maintain (completely sterile testcross progenies). The pollen fertility of the potential restorer and maintainer lines was classified into four categories — maintainers (M), partial maintainers (PM), partial restorer (PR), and restorer (R) (Table 3).

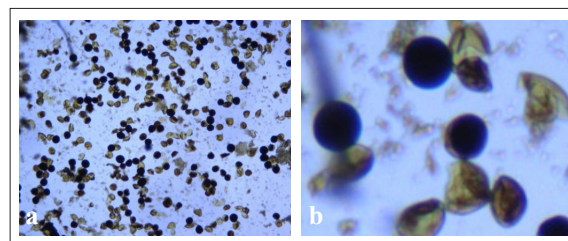


Figure 1. (a) Microscopic field (4x magnification) showing pollen grains used for evaluating and classifying pollen fertility and (b) an enlarged image (10x magnification) of the pollen grains.

Table 1. Classification of pollen based on sterility and fertility (Virmani et al., 1997).

Category	Appearance	Classification
Unstained withered sterile (UWS)		Sterile
Unstained spherical sterile (USS)		Sterile
Stained round (light) sterile (SRS)		Sterile
Stained round fertile (SRF)		Fertile

Table 2. Category based on pollen sterility (Virmani et al., 1997).

Pollen Sterility (%)	Category
100	Completely sterile (CS)
91 - 99	Sterile (S)
71 - 90	Partial sterile (PS)
31 - 70	Partial fertile (PF)
21 - 30	Fertile (F)
0 - 20	Fully fertile (FF)

Table 3. Classification of maintainer and restorer lines based on percentage pollen fertility (Virmani et al., 1997).

Category	Pollen Fertility (%)
Maintainer (M)	0 - 1
Partial maintainer (PM)	1.1 - 50
Partial restorer (PR)	50.1 - 80
Restorer (R)	>80

Marker Genotyping

Deoxyribonucleic acid (DNA) was extracted using the Genomic DNA Extraction Protocol adapted from PhilRice laboratory manual for DNA fingerprinting in hybrid rice by Perez et al. (2012). Samples from 21-day-old rice leaf of the 93 male parents were collected and placed in a properly labelled glassine bag. The collected leaf samples were ground in a 2-ml microtube using liquid nitrogen, added with a 750 µl of pre-warmed 2x detergent cetyltrimethylammonium bromide (CTAB), and 50 µl 20% sodium dodecyl sulfate (SDS). A 750-µl of chloroform was also added and centrifuged the tubes at 10,000 rpm for 30 min. Aqueous phase was then decanted into a new 1.5 ml tubes. A 600 µl cold isopropanol was poured to the solution and incubated overnight at -20°C. After incubation, the solutions were centrifuged at 10,000 rpm for 10 min. The isopropanol was decanted, and the resulting pellet was washed with 500 µl of 70% ethanol and centrifuged again at 10,000 rpm for 3 min. After discarding the alcohol, the pellets were dried overnight. Finally, the pellets were dissolved in 100 µl of TE buffer and incubated for 2 - 3 h at room temperature until fully dissolved.

Polymerase Chain Reaction (PCR) Amplification

Two gene specific markers namely PRR9-1 and SF21-5 were used to detect the presence or absence of *Rf4* and *Rf3* in 93 male parents, correspondingly. The PCR reaction was carried out in a 7.5 µl reaction containing sterile distilled water, 5x PCR buffer, 25 mM MgCl₂, 5 mM DNTPs, 10 mM Forward Primer, 10 mM Reverse Primer, 1.0 µl 1:50 Taq polymerase, and 2 µl template DNA (approximately 100 ng/µl). The PCR reaction underwent initial denaturation at

94°C (5 min), 35 cycles of final denaturation at 94°C (30 sec), primer annealing at 58°C and extension at 72°C for 1 min each. The final extension was at 72°C for 5 min and held at 4°C. The PCR products were electrophoresed for 75 min at 100 volts in 8% non-denaturing polyacrylamide gel.

Data Analysis

Pollen fertility of the F₁ testcross progenies was presented as percentage and the corresponding male parents were categorized as M, PM, PR, and R. The male parents were also classified based on the presence or absence of the two fertility restorer genes, while the relationship to the pollen fertility of their corresponding F₁ testcross progenies were descriptively analyzed.

Results

Phenotypic Characterization

The pollen fertility of F₁ hybrids was used to classify the male parent or pollen source according to the four categories defined by Virmani et al. (1997): M at 0 - 1%, PM at 1.1 - 50%, PR at 50.1 - 80%, and R for more than 80% pollen fertility. Results showed that F₁ testcross produced by crossing CMS lines with selected rice genotypes behaved differently with regard to pollen fertility. Among 195 F₁ testcross progenies different levels of pollen sterility/fertility were observed (Table 4). Such variation in pollen fertility indicated the existence of genetic variation in respect of these reproductive traits among the genotypes.

Table 4. Number of F₁ testcross progenies categorized on the basis of pollen fertility.

Category	No. of Lines	Percentage (%)
Restorer (R)	38	19.49
Partial Restorer (PR)	96	49.23
Partial Maintainer (PM)	56	28.72
Maintainer (M)	5	2.56
Total	195	100

Results of pollen fertility evaluation shows that 38 lines (19.49%) are categorized as R and 96 lines (49.23%) as PR (Figure 2). On the other hand, only five lines were found to be maintainers or having less than 1% fertility rate.

Among the evaluated 195 F₁ testcross progenies, five male parental genotypes were identified as potential maintainers due to their pollen fertility being less than 1.0%. These genotypes include TCN-36 from the IR58025A and PR51859-TWG038-7-1-3 cross, TCN 382 from the PR19A and PR39905-H003-142-1-4-3-2-2 cross, TCN-239 and

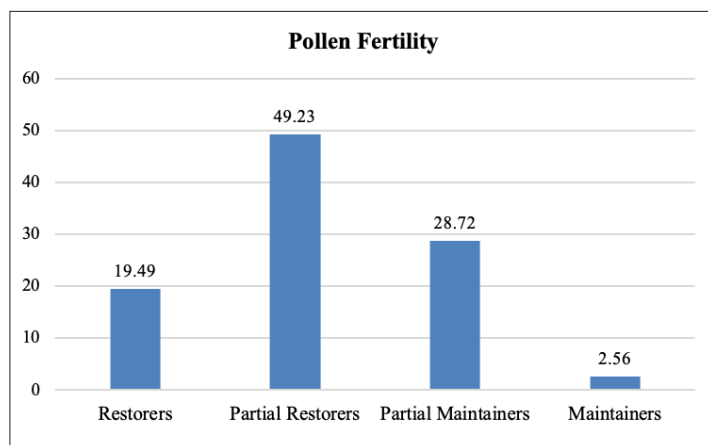


Figure 2. Percentage distribution of F₁ testcross progenies per pollen fertility category.

TCN-377 (with the same female parent) from the PR27A cross to PR39909-H007-24-2-1-1-3-2 and PR39906-H004-7-1-28-2-2-2, respectively, and TCN-147 from the PR29A and PR51858-G139-6-5-3 cross (Table 5).

Molecular Characterization

In molecular screening, gene-specific markers PRR9-1 and RMS SF21-5 were used to determine the presence of *Rf4* and *Rf3* genes, respectively. Male parents of the F₁ testcross progenies were categorized based on the presence or absence of the two fertility restorer genes. The comparison of pollen fertility among male parents based on the presence and absence of *Rf4* and *Rf3* genes were also evaluated (Figure 3).

Among the 93 elite breeding lines subjected to genotyping, 23 lines were identified as containing both fertility restorer genes, *Rf4* and *Rf3*, based on molecular screening. Of the nine genotypes with *Rf4* gene, eight lines were validated as complete restorers while four lines out of four genotypes with *Rf3* gene. Furthermore, 10 genotypes with both genes (*Rf4* and *Rf3*) are complete restorers (Tables 6 and 7). It shows that the number of restorer lines with two genes (i.e., *Rf4Rf4/Rf3Rf3*) is present as double dominant controlling the trait and has a stronger fertility restoration ability with 22.2% than the other. On the other hand, if the gene is present alone (i.e., *Rf4Rf4/rf3rf3*), pollen fertility is somewhat reduced. Finally, if the gene with weaker restoration ability (*rf4rf4/Rf3Rf3*) is present alone, it showed

Table 5. List of identified potential maintainers based on pollen fertility of F₁ testcross progenies.

TCN No.	Female	Cross Male	Pollen Fertility
TCN-36	IR58025A	PR51859-TWG038-7-1-3	0.95
TCN-382	PR19A	PR39905-H003-142-1-4-3-2-2	0.68
TCN-239	PR27A	PR39909-H007-24-2-1-1-3-2	0.46
TCN-377	PR27A	PR39906-H004-7-1-28-2-2-2	0.36
TCN-147	PR29A	PR51858-G139-6-5-3	0.00

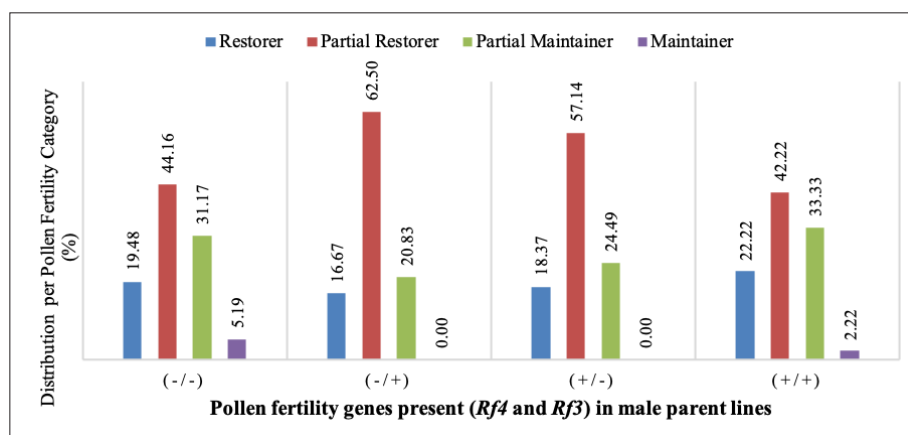


Figure 3. Comparison of pollen fertility among male parents characterized by the presence (+) or absence (-) of *Rf4* and *Rf3* genes.

Table 6. Number of male elite lines identified per category based on molecular screening for *Rf* genes *cum* pollen fertility of the F₁ testcross progenies.

<i>Rf4/Rf3</i>	Restorer	Partial Restorer	Partial Maintainer	Maintainer	Total	%Restorer	% Partial Restorer
(-/-)	15	34	24	4	77	19.5	44.2
(-/+)	4	15	5	0	24	16.7	62.5
(+/-)	9	28	12	0	49	18.4	57.1
(+/+)	10	19	15	1	45	22.2	42.2
Total	38	96	56	5	195	19.5	49.2

Table 7. Identified promising restorer lines based on F₁ testcross progeny pollen fertility evaluation and presence of *Rf* genes in male parents. (Note: TCN=testcross nursery).

TCN No.	Female	Male	F ₁ Pollen Fertility	<i>Rf</i> Gene
TCN-10	PR27A	R51847-G123-3-1-3	98.02	<i>Rf4+Rf3</i>
TCN-118	IR58025A	PR51843-G108-2-1-1	92.08	-
TCN-585	IR58025A	PR42130-M-1-B-4-1-B-1	91.76	-
TCN-175	PR15A	PR39908-H006-9-2-1-2-1-1	90.63	<i>Rf4 + Rf3</i>
TCN-92	PR27A	PR47267-G091-1-1-1	90.42	<i>Rf4 (H) + Rf3</i>
TCN-285	PR27A	PR39905-H003-142-4-1-2-2	89.84	-
TCN-193	PR21A	PR39915-H013-4-1-1-2	89.35	<i>Rf4+Rf3</i>
TCN-161	IR58025A	PR51857-G138-4-2-3	89.03	-
TCN-573	PR27A	PR 37606-42-3-3-2-2-1-B-1-2-2	88.56	<i>Rf4</i>
TCN-93	PR19A	PR47267-G091-1-1-1	88.43	<i>Rf4(H) + Rf3</i>
TCN-248	PR27A	PR39908-H006-80-2-2-2-3-1	87.97	<i>Rf4+Rf3</i>
TCN-589	PR21A	PR42130-M-4-B-B-B-B-1	87.11	<i>Rf4</i>
TCN-383	PR21A	PR39905-H003-142-1-4-3-2-2	86.68	<i>Rf4+Rf3</i>
TCN-545	PR19A	Tropical Selection	86.66	<i>Rf4</i>
TCN-255	PR15A	PR39904-H002-116-2-1-1-3	83.36	-
TCN-438	PR27A	PR45929HY-B-2396-186-154-199-254	86.12	-
TCN-498	PR29A	PR39501-12-6-71	86.01	<i>Rf3</i>
TCN-27	PR29A	PR51839-G102-3-1-1	85.16	<i>Rf3</i>
TCN-384	PR29A	PR39905-H003-142-1-4-3-2-2	84.67	<i>Rf4 + Rf3</i>
TCN-534	PR27A	PR40511-14-3-1	84.51	<i>Rf4</i>
TCN-548	PR27A	PR40496-66-1-2-5	84.43	<i>Rf3</i>
TCN-26	IR58025A	PR51839-G102-3-1-1	84.16	-
TCN-244	IR58025A	PR39909-H007-24-2-1-1-3-2	83.75	-
TCN-608	IR58025A	PR48424-Negros-IVM2011WS 45-1-6	83.63	-
TCN-60	PR15A	PR51839-G102-5-1-2	83.06	-
TCN-25	PR27A	PR51839-G102-3-1-1	83.00	-
TCN-394	PR15A	PR45886HY-B-921-95-61-85-108	81.33	-
TCN-581	IR58025A	PR37056-19-1-1-1-2-3-2-B-1-1-1	81.12	<i>Rf4</i>
TCN-588	IR58025A	PR42130-M-4-B-B-B-B-1	81.04	<i>Rf4</i>
TCN-210	IR58025A	PR40618-B024-18-1-1-2	80.97	<i>Rf4</i>
TCN-109	PR27A	PR51841-G106-2-1-2	80.78	-
TCN-107	PR15A	PR51841-G106-2-1-2	80.71	-
TCN-566	PR19A	PR37600-5-1-1-3-2-2-B-1-1-1	80.65	-
TCN-173	PR29A	PR39908-H006-2-1-3-3-1-3	80.56	<i>Rf4+Rf3</i>
TCN-401	PR19A	PR45886HY-B-921-95-63-89-110	80.50	<i>Rf3</i>
TCN-489	IR58025A	PR39501-12-6-67	80.33	<i>Rf4</i>
TCN-395	PR27A	PR45886HY-B-921-95-61-85-108	80.23	-
TCN-389	IR58025A	PR45873HY-B-501-50-23-26-43	80.00	-

the lowest restoration of pollen fertility. Virmani et al. (1997) also reported the differential reaction of rice genotypes with different CMS lines of the same cytoplasmic source.

Discussion

Identification of new genes that restore fertility in rice genotypes contributes in the development of excellent restorer lines. Furthermore, it is advantageous to look for restorer genes because phenotyping is time consuming and requires determining spikelet sterility in testcross progeny (Yao et al., 1997; Komori et al., 2003; Ahmadikhah and Karlov 2006; Ahmadikhah et al., 2007). In addition, partial restorer lines with a strong agronomic background can enhance through the transfer of fertility restorer genes without a sterile cytoplasm or extensive testcrossing with CMS lines. Revathi et al. (2013) evaluated the efficiency of tightly linked markers of *Rf3* and *Rf4* genes for fertility restoration in which 85 - 92% efficiency was identified. However, the effect of individual genes or absence of both genes through testcross with CMS were not yet studied.

Candidate gene-based markers for fertility restoration can be very useful in distinguishing restorers and non-restorers. Nas et al. (2003) demonstrated the use of molecular markers for the identification of restorer line. In a study conducted by Pranathi et al. (2016), the usefulness of RMS PRR9-1 targeting the *Rf4* gene was demonstrated, displaying a higher selection efficiency of 91% for identifying all major restorer lines. In comparison, SF21-5, which specifically targets the *Rf3* gene, exhibited a selection efficiency of 57%. These findings support the perception that a good restorer must possess *Rf4* gene alone or *Rf4* along with the *Rf3* gene, while line possessing only *Rf3* gene could not be a good restorer. However, two known restorer lines: IR66 and IR40750R, were observed to possess *Rf3* gene.

In this study, 15 genotypes were identified as restorers based on pollen fertility percentage, even in the absence of the *Rf4* and *Rf3* genes. These findings align with the results reported by Bharaj et al. (1995), which suggest that modifiers also play a role in fertility restoration. Remarkably, despite lacking the two genes responsible for restoring fertility in WA-CMS lines, the F_1 progenies still exhibited fertility restoration, including the lines categorized as restorers. Tada (2007) observed that the *Rf1* gene had varying effectiveness on BT-type CMS and partially restored pollen fertility in WA-type CMS. This implies that other fertility restoration genes may be present in the male parents of these genotypes, contributing to the fertility restoration observed in the F_1 progenies.

On the contrary, it is noteworthy that even when both *Rf4* and *Rf3* genes are present in the male parents of certain genotypes, the pollen fertility percentage of their F_1 testcrosses is not always completely restored. This outcome aligns with previous reports indicating that some well-established restorer lines (IR24, IR36, IR54, IR9761-19-1, and IR2797-105-2-2) for WA-CMS exhibit incomplete fertility restoration in CMS lines like IR17492A, which possesses WA cytoplasm (IRRI, 1986). The underlying reasons could include inhibitory genes affecting restoration in CMS lines or inter-varietal hybrid sterility (as proposed by Govinda Raj and Virmani, 1988). Modifier genes may also play a role in suppressing the expression of fertility genes, as suggested by Pande et al. (1990). Additionally, recorded variation may stem from differences in the penetrance or expressivity of pollen fertility-restoring genes across different genotypes (Umadevi et al., 2010). Environmental factors can further influence the restoration reaction. Notably, fertility restoration may vary depending on the specific CMS-restorer combination (Singh et al., 2014), as some restorers are effective only with certain CMS lines and may not restore fertility when combined with others.

Conclusion and Recommendations

The initial steps in three-line heterosis breeding involve identifying effective restorers and maintainers (Siddiq, 1996). Pollen fertility and spikelet fertility traits play a crucial role during the test cross nursery stage for distinguishing restorer and maintainer lines. Restorers identified in this study, especially those male parents carrying either or both of the two fertility restorer genes (*Rf4* and *Rf3*), can be valuable for developing new hybrids with desirable traits. Additionally, further analysis is needed for restorers from parents lacking *Rf4* or *Rf3* genes, focusing on the potential presence of other genes responsible for fertility restoration in WA CMS. The genotypes identified as maintainers through molecular and pollen sterility studies can be utilized in backcross breeding programs to create new CMS lines with desirable traits for hybrid production. Notably, this study revealed a higher number of restorers and partial restorers compared to maintainers.

In hybrid rice breeding, it is essential to perform test cross evaluations of F_1 crosses between unknown lines and the CMS line to identify maintainer and restorer lines. Consequently, hybrid rice breeding can be labor-intensive and time-consuming, spanning nearly two seasons or one year (Revathi et al., 2013). Additionally, exploring the best restorer-CMS combinations is crucial to maximize the potential utilization of male parents carrying identified fertility restoration genes.

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PHENOTYPIC DIVERSITY AND PROFILE OF ROOT ANGLE DISTRIBUTION OF SELECTED TRADITIONAL RICE GERMPLASM IN THE PHILIPPINES

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Abstract

Traditional rice varieties (TRVs) are valuable genetic resource owing to their good traits and ability to adapt to climatic changes, which are necessary in breeding programs. This study assessed the phenotypic diversity for root vertical angle distribution and profile of 54 TRVs at PhilRice Genebank. Three major clusters of TRVs were generated based on the root vertical angle (RVA). These clusters were classified as shallow-roots (C1: 4 TRVs), deep-roots (C2: 32 TRVs), and admix (C3: 16 TRVs). Majority of the TRVs have deep-rooting traits while 13 of the germplasm are shallow-rooting cultivars. TRVs exhibiting deep root growth had nodal number and length mostly distributed at a 60 - 90° growth angle. Exploring the association of abiotic stress tolerance with the deep-rooting traits of the TRVs would be useful in breeding for climate-resilient rice.

Keywords: Deep-rooting, Germplasm, Root vertical angle, Shallow-roots, Traditional rice varieties

Introduction

Rice (*Oryza sativa* L.), an important cereal, serves as a primary source of energy and nutrients for people (Zibae, 2013). Understanding the factors influencing rice production is crucial in increasing the level of production to meet the demands of a rapidly growing population. Projections suggest a 53% increase in the Asian population, driving up the demand for rice (Hossain and Fischer, 1995). This demand is expected to surge by 70% in the next three decades (Seckler and Amarasinghe, 2000). Rapid urbanization in the Philippines poses a threat, diminishing rice fields and reducing productivity (Barroga et al., 2016). The farmers must strive for increased yields to match the increasing demand (Long et al., 2015). Yet, persistent biotic and abiotic stresses continuously impede and cause significant annual losses in rice yield (Onyegbula, 2017).

Salinity, drought, and nutritional deficiencies have significant effects on rice plant development (Fahad et al., 2019). Root system plays a vital role in crop adaptation and in the maintenance of higher productivity under soil fertility and nutrient deficit conditions (Uga et al., 2009). The alternate wetting and drying (AWD) conditions can change the plant root architecture, resulting in the development of deep and shallow type rooting system. Deep roots allow plants to access water and nitrogen from deeper soil layers, helping the plant avoid deficits during times of drought or low nutrient availability (Ludlow and Muchow, 1990; de Dorlodot et al.,

2007; Rich and Watt, 2013; Uga et al., 2013a; Arai-Sanoh et al., 2014; Ramalingam et al., 2017; Lynch, 2022). It is characterized by narrower/steeper root cone and growth angles, fewer axial and lateral roots, and sparser lateral branching, facilitating deeper soil penetration (Lynch, 2013). On the other hand, shallow root is good at acquiring concentrated topsoil resources such as phosphorus, potassium, nitrogen, and ammonium (Lynch and Brown, 2001; Ahmadi et al., 2014; Kitomi et al., 2020). It can also easily capture dissolved oxygen in soil surface layers, potentially enabling it withstand waterlogged conditions (Hanzawa et al., 2013; Obara et al., 2010).

Niones et al. (2015) showed that root traits are controlled by several quantitative trait loci (QTL) or genes. Deeper rooting 1 (*DRO1*) and deeper rooting 2 (*DRO2*) genes improve root growth angle, resulting in a deeper penetration in soil (Uga et al., 2013a, 2013b). Overexpression of *DRO1* has been shown to enhance rice yield under drought conditions by facilitating access to soil moisture reserves at greater depths (Arai-Sanoh et al., 2014; Kitomi et al., 2020). In shallow-rooting rice, the key QTL is called soil surface rooting 1 (*qSOR1*), which is expressed in columella cells and induces shallow root formation, allowing for greater absorption of nutrients from the topsoil (Uga et al., 2011, 2012, 2015, 2018; Kitomi et al., 2020). There is generally a trade-off between these two root strategies, as phenotypes that promote deep rooting may negatively impact topsoil foraging and vice versa (Lynch, 2018). Understanding the root system's genetic aspects is vital in breeding programs.

This knowledge bears immense potential for boosting food sufficiency by developing stress-tolerant rice varieties, depending on their relevance to specific environmental conditions and farming techniques.

Rice germplasm is a remarkable genetic material and source of novel traits that provide potential solutions in improving the country's rice production. Understanding rice germplasm is essential in the genetic improvement of root system architecture typical of modern rice varieties (Shi and Hu, 2017). Choudhury et al. (2013) reported that cultivated traditional rice varieties (TRVs) are a good source of novel genes or traits and good grain quality. Currently, researchers focus on developing root DNA-based marker-assisted breeding technologies for precise and quick development of adapted varieties. The characterization of root system architecture provides essential insights into improving rice plants that can withstand environmental stresses. This study profiled the root angle distribution and its diversity in relation to shoot dry matter production of the TRVs germplasm collections under well-watered condition.

Materials and Methods

Plant Material

Selected 54 TRVs from Philippine Rice Research Institute (PhilRice) Genebank were used to assess root angle and its distribution under well-watered condition. The study was conducted at the PhilRice

Central Experiment Station in Nueva Ecija. The experiment was laid out in a completely randomized design (CRD) with three replications from January to March 2019.

Experimental Treatment and Cultural Management

Seeds of each genotype were soaked in water with fungicide (Benomyl benlate, 0.15% w/v) in a petri dish and incubated for 48 h at 28°C (Figure 1A). The three pre-germinated seeds of each genotype were sown in the middle of each planting stripe using bottomless seedling trays (37 x 2 planting stripes per tray) filled with garden soil pre-mixed with 0.60 g complete fertilizer (14-14-14) and 0.10 g urea (46-0-0) fertilizer (Figure 1B). The seedlings were thinned to one seedling per hill at three days after sowing (DAS). Afterward, the seedlings were cultivated in well-watered (WW) condition until 14 DAS. The 14-day seedlings were carefully extracted from the soil and rinsed with running tap water (Figure 1C). The extracted roots were temporarily stored in a plastic pack containing 95% of alcohol for further measurements.

The shoots were cut at the base of the plant and oven-dried for 48 h at 70°C, then weighed for shoot dry weight (SDW). Subsequently, the entire root system was taken off and preserved in FAA (formalin, acetic acid, and 70% ethanol in a 1:1:18 volume ratio) solution for further analysis of the root system and

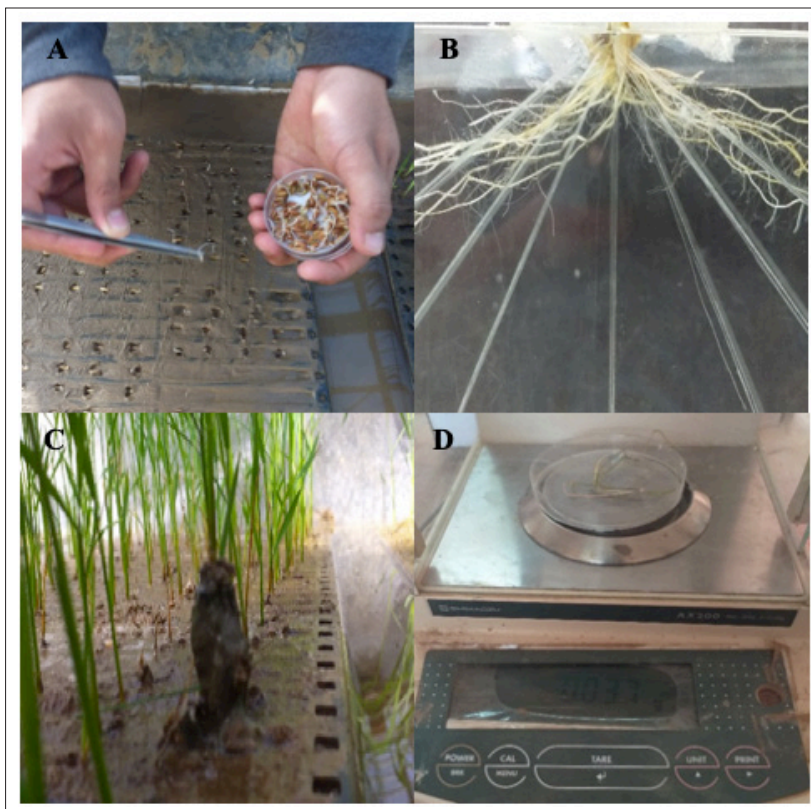


Figure 1. Treatment and cultural management of research experiments: (A) seed sowing; (B) measuring deep rooting based on number nodal roots based on angle; (C) growing of seedling in the tray; (D) weighing of shoot and root dry weight.

its root components. Digital photos were captured using a scanner set to 300 dpi and an output format of 256 greyscales. The root samples were spread out in the transparent sheets with little overlap to measure root length. An Epson scanner (ES2200) was used to capture digital photographs at a resolution of 300 dpi.

WinRHIZO v. 2007d (Regents Instruments, Québec, Canada), an image analysis system specifically designed for root measurement in different forms, was used to analyze scanned pictures for total root length (TRL) and longest nodal root length (LNRL), with a pixel threshold of 175 specified. The sum of each nodal root length was used to get the total nodal root length (TNRL). The root length per unit root dry weight, or Specific Root Length (SRL), was calculated by dividing the root dry weight by the total root length. Three distinct angles were used to count and measure the nodal root number and root angle distribution: 0 - 30°, 45 - 60°, and 60 - 90°. Deep-rooting rice can be determined by the 60 - 90° angle, while shallow-rooting rice is indicated within 0 - 30° and 45 - 60° category (Uga et al., 2012). For each angle the roots were measured and counted individually. The metric ruler was used to manually measure the shoot length (SL). The roots were oven-dried for three days at 80°C, and the root dry weight (RDW) was measured using a laboratory weighing balance (Figure 1D). The root-to-shoot ratio was calculated by dividing the dry weight of the roots by the dry weight of the plant's top (shoots).

Statistical Analysis

A Pearson product-moment correlation coefficient was computed at 0.01 level of significance (two-tailed) to assess the relationship among variables. The unrooted classification was performed using Dissimilarity Analysis and Representation for Windows (DARWin) software. Analysis of variance was performed to determine if there is a significant difference at a 5% level of significance between varieties at different parameters using SPSS version 23.

Results and Discussion

Root Vertical Angle Variation and Distribution in Rice

The root growth angle and its distribution among traditional rice varieties were classified per geographical regions in the Philippines (Table 1). Region IVA had the highest number of collected representative TRVs, contrary to regions I, VI, and XIII. All regions have at least one representative variety present at a 60 - 90° angle. Among the 54 TRVs, 39 varieties were classified as deep-rooted

(72.22%) while the remaining 15 varieties were shallow-rooted genotypes (27.78%). Region IVA had the most number (7) of collected TRVs classified as deep rooting genotypes followed by CAR (4) and Region II (4).

In this study, IR64 and CSSL 47 (with the Kasalath allele) were employed as check varieties to investigate shallow and deep root system architecture (Figure 2A and B). Notably, red rice (Figure 2C) exhibited shallow rooting characteristics, while Binnit (Figure 2D) was identified as a deep-rooted variety. The shallow-rooted IR64 displayed a low water uptake rate under water stress (Gowda et al., 2012). In contrast, CSSL 47 demonstrated the ability to promote root elongation and increase root length, enabling adaptation to drought conditions (Niones et al., 2013). Different portions of rice roots play distinct roles in soil nutrient uptake: longer roots efficiently transport water and nutrients to the soil, while shorter roots capture resources in the topsoil layer (Gu et al., 2017). Understanding phenotypic variations in root morphology can enhance crop productivity across diverse environmental conditions (Comas et al., 2013).

Classification and Frequency Distribution of Root Growth Angle

The 54 TRVs were geographically distributed in Luzon (38), Visayas (5), and Mindanao (9) based on root angle categories (Figure 3A). Luzon had 38 TRVs, comprising 12 shallow-rooted and 26 deep-rooted varieties. Collections in Visayas featured 1 shallow-rooted and 4 deep-rooted TRVs, while Mindanao collections showcased 1 shallow-rooting and 8 deep-rooting varieties. The majority of selected TRVs in the Philippines exhibited a deep rooting architecture, having a root growth angle of 60 - 90°. The root growth response of the collected TRVs is directly affected by water availability, which directly affects the root system architecture (Sariam, 2009). Hence, TRVs are well adapted and respond to different soil conditions and demonstrate an adaptable reaction to environmental stresses owing to the development of a deep root system, which is important in avoiding drought stress in rice (Uga et al., 2011). Among the collected Philippine TRVs, some exhibited a shallow root type, likely adapted to lowland areas and rainfed conditions (Kirk et al., 2013). TRV were classified into three cluster groups based on root distribution, using unrooted trees constructed from all measured traits (Figure 3B). The first group (C1) consisted of predominantly shallow-rooted TRVs, while C2 (the second cluster group) comprised deep-rooted TRVs. Cluster 3, a combination of C1 and C2, represented admix varieties.

Table 1. Classification of TRVs in the Philippines based on root angle distribution.

Region	Root angle distribution (n°)		
	0 - 45° (shallow roots)	45 - 60° (intermediate roots)	60 - 90° (deep roots)
I			(20) Kalangiking
II	(10) Burtok (11) Imelda	(22) Mimis	(46) Megamunin (12) Dugayong (23) Biniding (17) Alimoran
III		(08) Impuha (45) Aglipay (52) Balasang	(50) PSB Rc3 (51) Tuhaw
IVA		(07) 42 NA (26) Gilingan (31) Camuros (25) Binnit	(35) Pinarumpong (37) Sinaginting (44) Demorado (47) Himonay (36) Binintang (53) Nagsalay (18) Binignay
IVB		(16) Quinizon (30) Dumali (42) Tapul	(41) Balibod na Pula
V			(01) Kabulan (34) Capis (05) Ginayanggang
VI			(14) Ago-oyong
VIII		(48) Inaguyod Seln (21) Binalangahan	(49) Kinanda Inuzo (29) Baksalan
IX			(39) Maligaya (19) Morigaya (33) Kabuong
XII		(38) Halay Palawad	(40) Awot
XIII			(32) Cabting
CAR	(24) Red Rice	(02) Palawan (27) Sanip	(09) Azucena (28) Galiano (06) Inanod (15) Gobierno Puti
BARMM	(43) Cuevas		(13) Inumay (54) Kwa-Kwa
Checks		(04) IR64	(03) CSSL 47

Note: The superscript number in TRVs names is the index number of the TRVs in reference to Figure 3B.

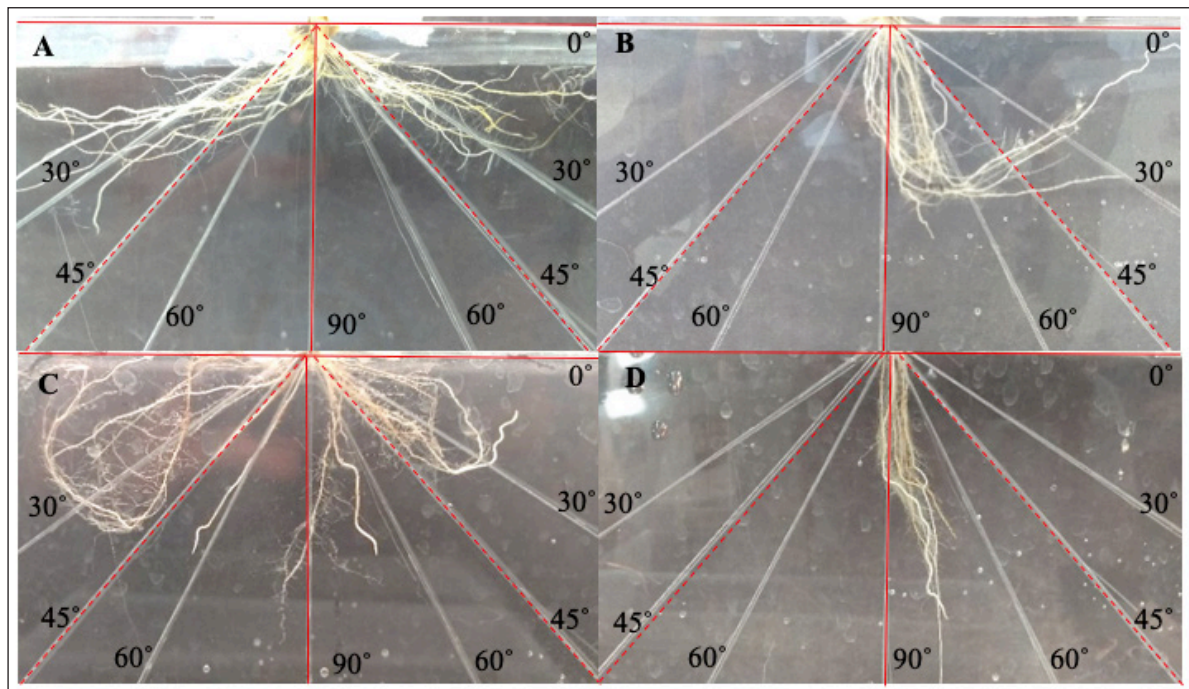


Figure 2. Root distribution based on the nodal root number positioned at each angle: (A) ⁽⁰⁴⁾IR64 (shallow); (B) ⁽⁰³⁾CSSL47 (deep); (C) ⁽²⁴⁾Red rice (shallow); (D) ⁽²⁵⁾Binnit (deep).

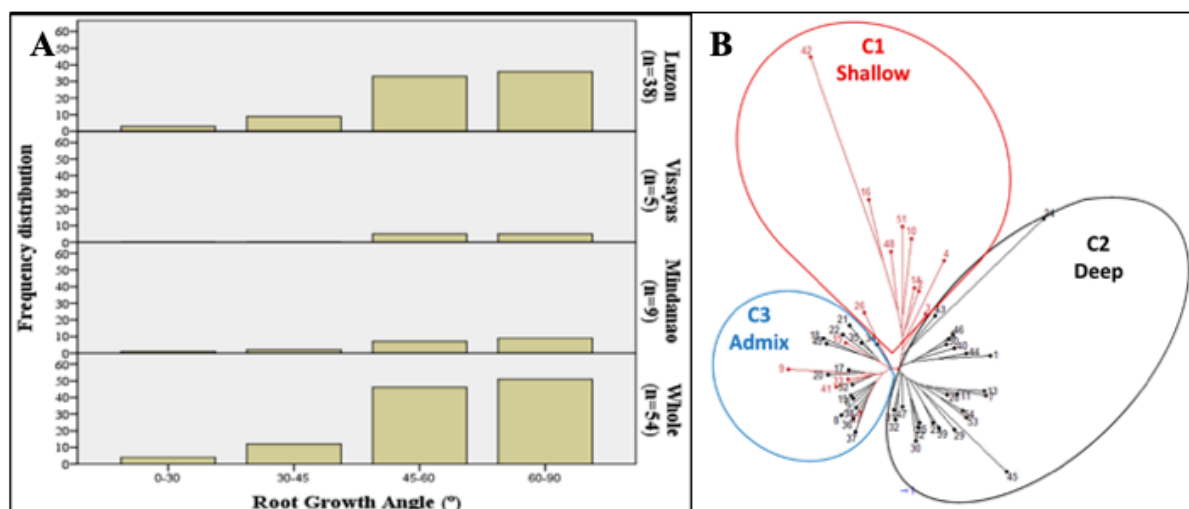


Figure 3. Root growth angle profile and distribution of the 54 rice germplasm: (A) root growth angle frequency distribution of traditional rice varieties and (B) diversity unroot tree constructed based on Nei's genetic distance of number of roots and root distribution under well-watered conditions.

Relationships of Root Traits and Shoot Length in Rice

The relationship of root vertical angle (RVA) with total root length (TRL) and total root number (TRN), and between total root number (TRN) and shoot length (SL) is shown in Figure 4. It is interesting to note the positive relationship between RVA and TRL ($r = 0.6945$), indicating a moderate positive correlation (Schober et al., 2018) (Figure 4A). The study has proven that plants classified with shallower root systems within the 0 - 30° and 30 - 45° angles exhibited significantly shorter root lengths with the majority measuring less than 35cm. Intermediate

roots situated between 45° and 60° demonstrated moderate lengths, ranging 0 - 100 cm with the majority measuring around 50 cm. The 60 - 90° category displayed markedly longer root systems, spanning from 50 - 200 cm. Ramalingam et al. (2017) demonstrated the significant and positive correlation between TRL and the portion of root with a deep angle at the maturity stage. This also implied that the longer the root length, the greater the acquisition of nutrients in the soil (Sariam, 2009).

In Figure 4B, analysis of Root Vertical Angle (RVA) and Total Root Number (TRN) data reveals significant patterns. Shallow roots within the

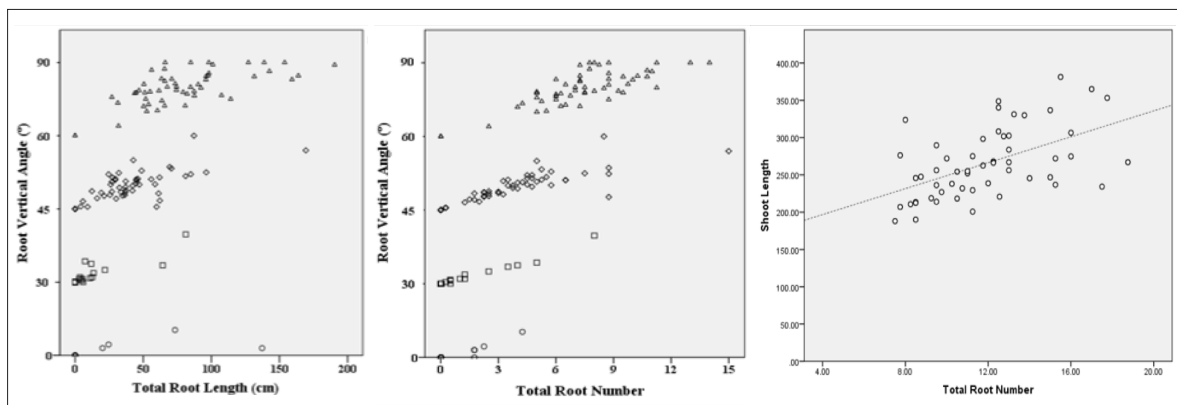


Figure 4. Relationship of root vertical angle (RVA) to (A) total root length, and (B) total root number, and (C) relationship between total root number and shoot length ($r = 0.53$). Different shapes represent the individual genotypes: open circle (O) for 0 - 30° (shallow roots), the square (□) for 30 - 45° (shallow roots), the diamond (◇) for 45 - 60° (intermediate roots), and the triangle (▲) for 60 - 90° (deep roots).

0 - 30° and 30 - 45° ranges show varying total root numbers, ranging from 0 - 3 and 0 - 6, respectively. Intermediate roots in the 45 - 60° range display 0 - 9 root numbers. Deeper roots, falling within the 60 - 90° category, exhibit a notable increase in total root number, ranging from 6 - 15. This suggests a clear trend towards increased root proliferation as the angle approaches 90°, indicating that steeper vertical root angles and deeper roots tend to prompt the development of more roots. In addition, the correlation of RVA with total root number revealed a moderate positive correlation ($r = 0.6785$), further supporting the association between angle and root quantity.

Moreover, a moderate positive correlation ($r = 0.53$) was observed between RVA and both shoot

length and TRN (Figure 4A-C), suggesting that an increase in nodal root number leads to an increase in TRL, thereby, potentially enhancing shoot growth and biomass production. This finding supports Ramalingam and Jebaraj (2013), indicating a positive relationship between root number and shoot length. Cultivating crop varieties with deep roots is one effective breeding strategy for improving yield under water-limited conditions (Lynch, 1995; Uga et al., 2013a). Furthermore, the study of Niones et al. (2012; 2015) highlighted that CSSL47, a deep rooting variety, showed greater root system development than Nipponbare (Figure 5). This superiority can be attributed to its greater capacity for promoting root elongation resulting in higher shoot dry matter and yield.

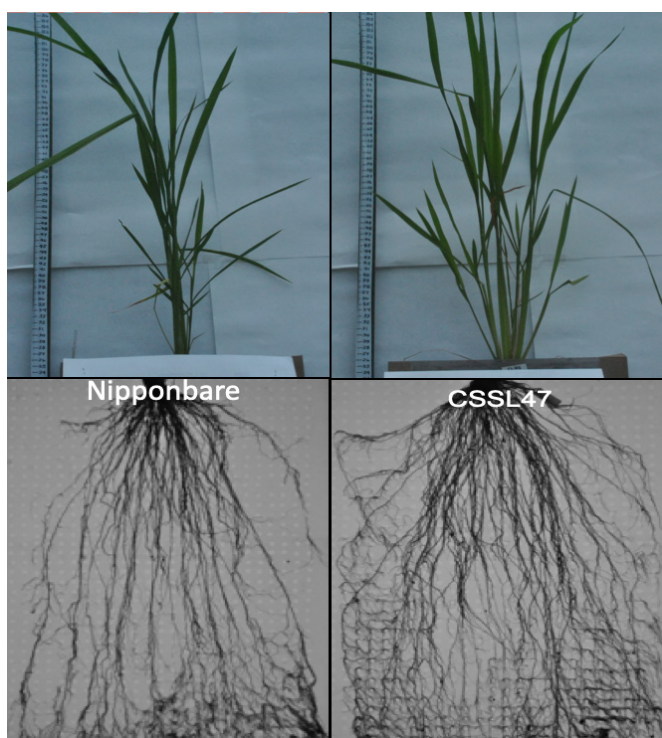


Figure 5. Shoot growth and root system development of 41-day old of Nipponbare and CSSL 47 genotype in root box experiment under soil moisture fluctuation stress. The CSSL 47, a deep rooting genotype, showed greater capacity of promoting root system resulting in higher shoot dry matter and grain yield (Niones et al., 2012; 2015).

Conclusion

The study showed a significant contribution of root traits (RVA, TRL, and TRN) to the increase in shoot growth. A profile of root angle distribution for selected TRVs was generated. TRVs with deep-rooting systems, which may exhibit good nitrogen use efficiency, and drought tolerance were classified. Among the 52 randomly selected TRVs, 39 were classified as deep-rooted, while 13 were verified as shallow-rooted. In Luzon, Sanip, Kalangiking, and Dumali accessions were identified as the most deep-rooted among the 26 TRVs. Binalangahan, Baksalan, and Ago-oyong are the deep-rooting TRVs out of four in Visayas, while Kabuong, Halay Palawad, and Morigaya ranked as the top three deep-rooted TRVs among the eight varieties in Mindanao. Findings indicate that majority of the TRV germplasm had deep-root system traits, which may significantly contribute to the increase in maintenance of biomass production. The identified deep-rooted TRVs will be used in drought stress tolerance and nitrogen use efficiency breeding programs and further tests. In addition, these TRVs will also be utilized in genome wide association (GWAS) mapping, and in exploring the physiological, genetic, and molecular mechanisms governing the identified traits. This strategy offers a more comprehensive understanding of the studied root traits, which will facilitate the implementation of the target breeding programs aimed at developing climate-resilient rice varieties in the future.

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A COMPREHENSIVE ANALYSIS OF FALL ARMYWORM, *Spodoptera frugiperda* (J. E. SMITH), FEEDING AND OVIPOSITION PREFERENCES ON WEED SPECIES IN THE PHILIPPINES

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Abstract

The polyphagous fall armyworm (FAW), *Spodoptera frugiperda*, threatens major crops in the Philippines, particularly corn, and recently expanded to rice. This study investigated FAW occurrence and weed communities in rice and non-rice ecosystems across Pampanga, Pangasinan, and Tarlac from January 2021 to October 2022. A survey identified 70 weed species in FAW-infested rice-corn areas, dominated by grasses (17 species) and broadleaves (49 species), with four sedge species also present. Five common weed species present in the monitoring sites [*Dactyloctenium aegyptium* (DTTAE), *Digitaria ciliaris* (DIGAD), *Eleusine indica* (ELEIN), *Cyperus rotundus* (CYPRO), and *Amaranthus spinosus* (AMASP)] were evaluated for FAW feeding and egg-laying preference in free-choice and no-choice tests from September 2022 to February 2023. In the no-choice test, DTTAE and ELEIN suffered the most damage at 7 h, but all plants were consumed within 24 h, with DTTAE receiving the most eggs. In the free-choice test, DTTAE, DIGAD, and ELEIN showed similar damage to corn after 24 h, suggesting their preference for FAW feeding and oviposition. Interestingly, AMASP was not favored for egg-laying. These findings suggest that weed control in rice and corn fields has the potential to reduce FAW infestation. Additionally, weed removal during fallow periods may disrupt the FAW life cycle, minimizing reinfestation risk in subsequent seasons.

Keywords: Food preference, Host plant specificity, Oviposition, Pest management, *Spodoptera frugiperda*, weed species, Philippines.

Introduction

Fall Armyworm (FAW), *Spodoptera frugiperda*, has emerged as a formidable invasive pest, displaying a distinct preference for corn while also causing damage to various major crops, including rice. In a comprehensive investigation led by Montezano et al. (2018), 353 larval host plant records of FAW were documented across 76 plant families. Among these, Poaceae (106 records), Asteraceae (31 records), and Fabaceae (31 records) stood out as primary families. Although corn remains the primary host for FAW, alternative hosts have been identified, highlighting the pest's ability to complete its life cycle on various crops. Notably, studies by Volp et al. (2022) identified sorghum and peanuts as alternate hosts, while Acharya et al. (2022) emphasized the significance of rice and potato in supporting FAW's life cycle. Subsequent research has further highlighted FAW's adaptability to complete its life cycle on crops such as peanut, sorghum, wheat (Chen et al., 2020), as well as tomato and pepper (Wu et al., 2021). Furthermore, the role of weeds as crucial alternate hosts for FAW has been emphasized in studies (Chen et al., 2023). Weeds including Napier grass (*Pennisetum purpureum*), natal grass (*Melinis repens*), and sunn hemp (*Crotalaria juncea*) were identified as significant contributors to

supporting FAW's life cycle. Agravante et al. (2022) expanded on this understanding by identifying additional weed species, including *Rottboellia cochinchinensis*, *Ipomoea triloba*, *Eleusine indica*, and *Portulaca oleracea*, as integral components of FAW's alternative host range. The intricate interplay between FAW and its diverse host plants underscores the necessity of exploring the pest's preferences and interactions within the dynamic context of agricultural ecosystems.

The first reported damage by FAW on corn in the Philippines occurred in June 2019 in Piat, Cagayan (Navasero et al., 2019). Subsequently, FAW infestations in rice were documented on May 17, 2021, in the rice seedbeds of Gonzaga, Cagayan. Recorded infestation levels ranged from 3 - 26 FAW larvae per square foot. This infestation was observed in four provinces—Cagayan, Isabela, Nueva Vizcaya, and Quirino—across 14 municipalities within Region II. FAW affected both rice seedbeds and direct-seeded rice at the seedling stage (Valdez et al., 2023a). Additionally, Valdez et al. (2023b) reported 60% FAW damage on rice seedbeds in Rapuli, Sta Ana, Cagayan, 16.67% damage in Brgy. Flourishing,

Gonzaga, Cagayan, and 60 - 90% damage at the PhilRice Central Experiment Station in Science City of Muñoz, Nueva Ecija.

As a polyphagous pest with a broad spectrum of hosts, the FAW poses a critical consideration in crop protection due to its ability to utilize weeds as alternative hosts in areas cultivated with corn and rice. Volp et al. (2022) emphasized the need for rigorous evaluation of evidence before designating a plant as a host for a given insect. They highlight that the mere presence of immature insects on a plant does not automatically classify it as a host. Thus, this study investigated the most prevalent weed species in rice and non-rice areas in Pampanga, Tarlac, and Pangasinan, which may serve as alternative hosts for FAW.

Materials and Methods

Experimental Setup and Design

The dominant weeds, identified through vegetation analysis conducted on rice and non-rice ecosystems in Pampanga, Pangasinan, and Tarlac from January 2021 to October 2022, were chosen as

test plants for subsequent experiments. The key weeds screened for host-plant specificity tests included *Dactyloctenium aegyptium* (DTTAE), *Digitaria ciliaris* (DIGAD), *Cyperus rotundus* (CYPRO), *Eleusine indica* (ELEIN), and *Amaranthus spinosus* (AMASP) (Figure 1). These selected weeds served as test plants for feeding and oviposition preference tests conducted under screenhouse conditions at PhilRice Central Experiment Station from September 2022 to February 2023. Rice and corn were used as control plants in the experimental setup.

Two distinct tests, the no-choice and free-choice tests, were implemented. In the free-choice test, plants were arranged randomly in plant boxes measuring 63 x 32 x 24 in (L x W x H) and enclosed within a screen cage. Conversely, in the no-choice test, plants were individually potted in clay pots (8 in in diameter), each enclosed within a mylar cage (24-in height and 6-in diameter) (Figure 2). The experimental subjects comprised 21-day-old rice seedlings, five selected weed species, and 14-day-old corn plants. Plants were tested in the seedling stage as findings of Valdez et al., (2023a) showed that FAW attacks in the Philippines were only observed on rice seedbeds.

<p>PANPU CYNDA DTTAE DIGAD ECHCO ELEIN ISCRU LEFCH CYPPIR CYPRO AMASP AMAVI CLERT CLEVI COMBE EPHHI ECLAL HYOFR IPOAQ IPOTR LUDLI MAACA PYLDE PHYAN POROL TRTPO</p> <p>Magalang, Pampanga</p>	<p>CYNDA DTTAE DIGAD ECHCO ECHGL ELEIN ISCRU LEFCH CYPPIR CYPRO AMASP HYOFR LUDLI PYLDE TRTPO</p> <p>Concepcion, Tarlac</p>
<p>CYNDA DTTAE DIGAD ECHCO ECHGL ELEIN ISCRU LEFCH CYPPIR CYPRO FIMMI AMASP CLERT CLEVI COMDI HYOFR IPOAQ IPOTR, LUDLI PHYAN TRTPO</p> <p>Pura, Tarlac</p>	<p>DTTAE ECHCO ELEIN LEFCH CYPPIR CYPRO FIMMI AMASP CLERT CRGOL ECLAL IPOTR, LUDLI MAACA TRTPO</p> <p>Rosales, Pangasinan</p>

Figure 1. Weed species consistently observed across monitoring areas. Grasses (in green): *Brachiaria mutica* (PANPU), *Cynodon dactylon* (CYNDA), *Dactyloctenium aegyptium* (DTTAE), *Digitaria ciliaris* (DIGAD), *Echinochloa colona* (ECHCO), *Echinochloa glabrescens* (ECHGL), *Eleusine indica* (ELEIN), *Ischaemum rugosum* (ISCRU), *Leptochloa chinensis* (LEFCH). Sedges (in red): *Cyperus iria* (CYPPIR), *Cyperus rotundus* (CYPRO), *Fimbristylis miliacea* (FIMMI). Broadleaves (in purple): *Amaranthus spinosus* (AMASP), *Amaranthus viridis* (AMAVI), *Cleome rutosperma* (CLERT), *Cleome viscosa* (CLEVI), *Commelina benghalensis* (COMBE), *Commelina diffusa* (COMDI), *Corchorus olitorius* (CRGOL), *Eclipta prostrata* (ECLAL), *Euphorbia hirta* (EPHHI), *Hedyotis corymbosa* (HYOFR), *Ipomea aquatica* (IPOAQ), *Ipomea triloba* (IPOTR), *Ludwigia hyssopifolia* (LUDLI), *Malachra capitata* (MAACA), *Phyllanthus debilis* (PYLDE), *Physalis angulate* (PHYAN), *Portulaca oleracea* (POROL), *Trianthema portulacastrum* (TRTPO).

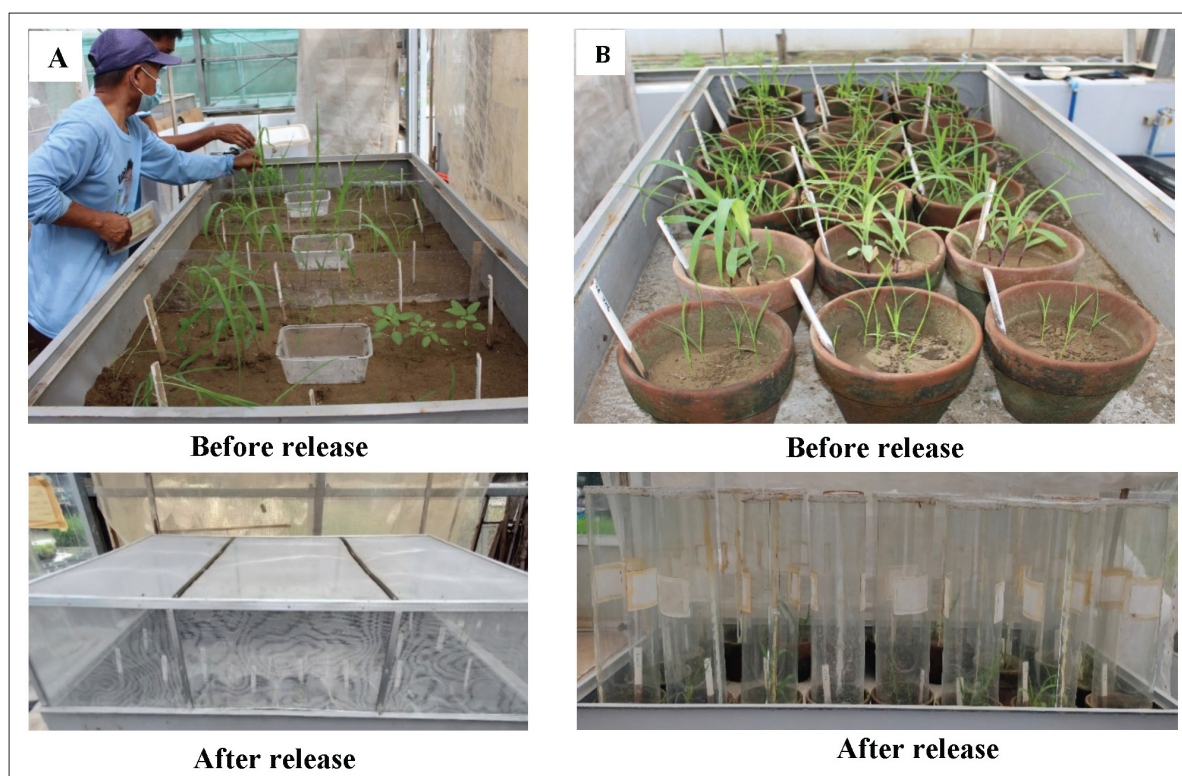


Figure 2. Feeding preferences test setups: (A) Free-choice, (B) No-choice (pre-larvae release).

This careful approach aimed to understand how FAW interacts with selected test plants, showing its preferences for feeding and oviposition under controlled conditions. To ensure robust experimental design and statistical validity, the experiments were arranged in a completely randomized design (CRD) with three replications.

Preparation of FAW Test Insects

Larvae and moths used in the experiment were reared in a laboratory setting. The initial culture originated from field-collected larvae found on corn in Magalang, Pampanga. This culture was maintained in an aerated container at a consistent room temperature of 25°C, with regulated humidity and a 12-h photoperiod. Larvae were provided with fresh corn leaves daily until reaching the pupation stage. Subsequently, pupae were transferred to an oviposition container (9.5-in diameter) lined with paper towels. Emerging moths were housed in the same container and nourished with cotton moistened with a 10% honey solution. Egg masses were gathered and then transferred to rectangular containers (12.5 x 9.5 x 4.5 inches, L x W x H) until the hatching phase.

Feeding Preference Test

In the feeding preference test, ten 3rd instar larvae were introduced per plant, and observations were carried out at 1, 3, 5, 7, 24, and 48 h post-release. Larval feeding was assessed using a rating scale: 0 (no damage), 1 (1 - 10%), 3 (11 - 20%), 5 (21 - 35%),

7 (36 - 50%) and 9 (51 - 100%) (IRRI, 2013). Two separate trials were executed, first in September 2022 and second in February 2023.

Oviposition Preference Test

To distinguish male and female FAW moths upon emergence, wing markings were used (Figure 3). Subsequently, one-day-old, lab-reared FAW adults (a single male and female pair) were released per host plant within the experimental cage (Figure 4). Egg masses on each plant were then recorded over a four-day period. The free-choice test identified the most preferred host plant for FAW oviposition, while the no-choice test pinpointed the favored oviposition site on the chosen host. Two trials were conducted, one in October 2022 and another in February 2023.

Statistical Analysis

To normalize the data for statistical analysis, an arcsine square root transformation was applied. This transformation helps ensure the data follows a normal distribution, which is a requirement for many statistical tests. Following data transformation, analysis of variance (ANOVA) was performed, while the means were differentiated through Tukey's Honest Significant Difference (HSD) test at a 5% significance level. The statistical analyses were conducted using the Statistical Tool for Agricultural Research (STAR) version 2.0.1 developed by the International Rice Research Institute (IRRI).

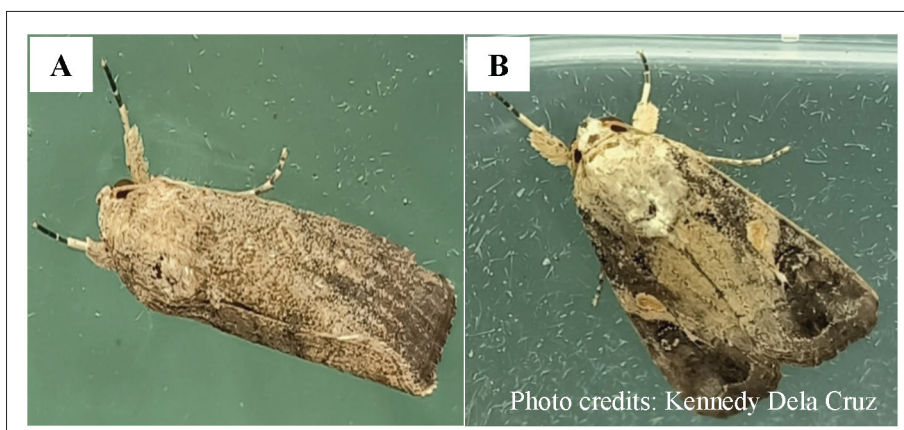


Figure 3. FAW adult identification by sex: (A) Female, and (B) Male.

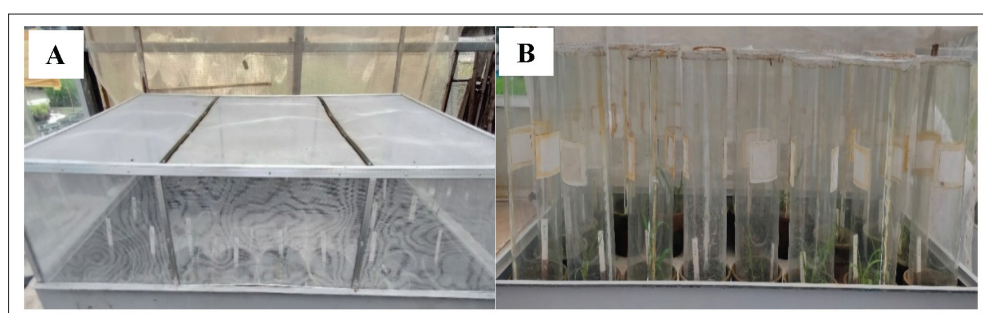


Figure 4. Oviposition preference test setup: (A) Free-choice, (B) No-choice (post-FAW release).

Results and Discussion

Feeding Preference Test

FAW larvae inflict damage on plants by consuming leaves and severing petioles or entire stems. Typically, they began feeding within one hour after release, with visible damage on preferred plants becoming apparent between 3 - 7 h. During the free-choice test in trial 1 (Figure 5), DGAD and DTTAE showed the most extensive damage within one hour of FAW release, receiving scores of 0.8 and 0.7 on a damage rating scale, respectively. At the 3-hour mark, DTTAE remained the most damaged weed species (rating; 1.4). Meanwhile, ELEIN and DTTAE received ratings of 3.0 and 2.3, respectively. By 7 h after release, DGAD, DTTAE, and ELEIN had sustained significant damage, with ratings of 3.8, 3.5, and 3.4, respectively. The trend continued at 24 h, with ELEIN, DTTAE, and DGAD scored 7.5, 7.2, and 6.3 exhibiting the most feeding damage. Notably, the damage levels on these plants remained comparable throughout the observation period. CYPRO incurred the least amount of damage among all the tested plants throughout the experiment.

In contrast to trial 1, there were no significant differences in damage ratings among the tested plants during the first 7 hours after releasing FAW larvae in trial 2. However, by the 24 h mark, a clear pattern

emerged. DGAD, ELEIN, and DTTAE sustained the most damage, with ratings of 7.1, 7.0, and 6.5, respectively. This trend continued at 48 h, with these three plants exhibiting even higher damage scores (8.5, 8.5, and 7.8, respectively). Notably, CYPRO remained relatively unscathed throughout the experiment, receiving a minimal damage rating of 0.2 (Figure 6).

Across both trials, DTTAE, DIGAD, and ELEIN consistently exhibited the highest levels of FAW damage throughout the observation period (1, 3, 5, 7, and 24 h after release) in the free-choice setup. Notably, the damage inflicted on these preferred weeds was statistically similar for both rice and corn plants up to 24 hours after releasing FAW larvae. These findings align with previous research by Agravante et al. (2022) and Nagoshi et al. (2007), which suggests that grass weeds serve as more suitable hosts for FAW compared to broadleaf plants.

Figure 7 vividly illustrates the extensive damage inflicted by FAW larvae within 48 h of release. At 48 h after release, DTTAE, ELEIN, and corn plants suffered complete defoliation (100% damage), with FAW larvae consuming both leaves and stalks. FAW feeding behavior differed on DGAD and AMASP. In these cases, FAW larvae primarily targeted leaves, leaving the stalks relatively untouched. CYPRO, on the other hand, remained largely unscathed,

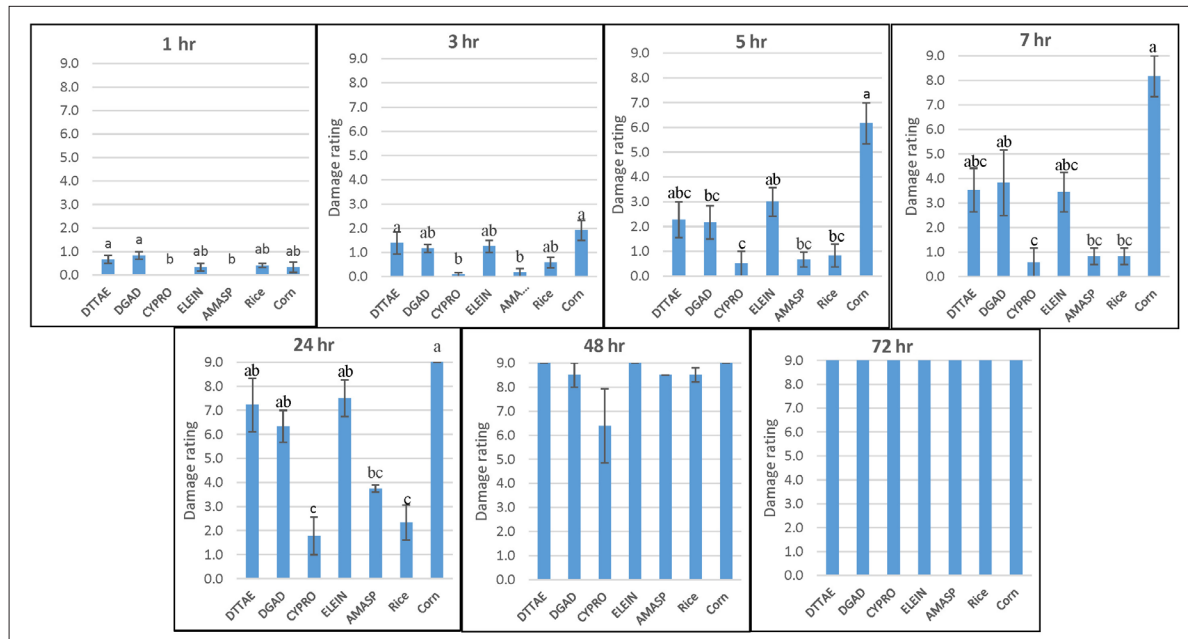


Figure 5. Feeding damage by FAW on various plants in free-choice host plant specificity test during the first trial in September 2022.

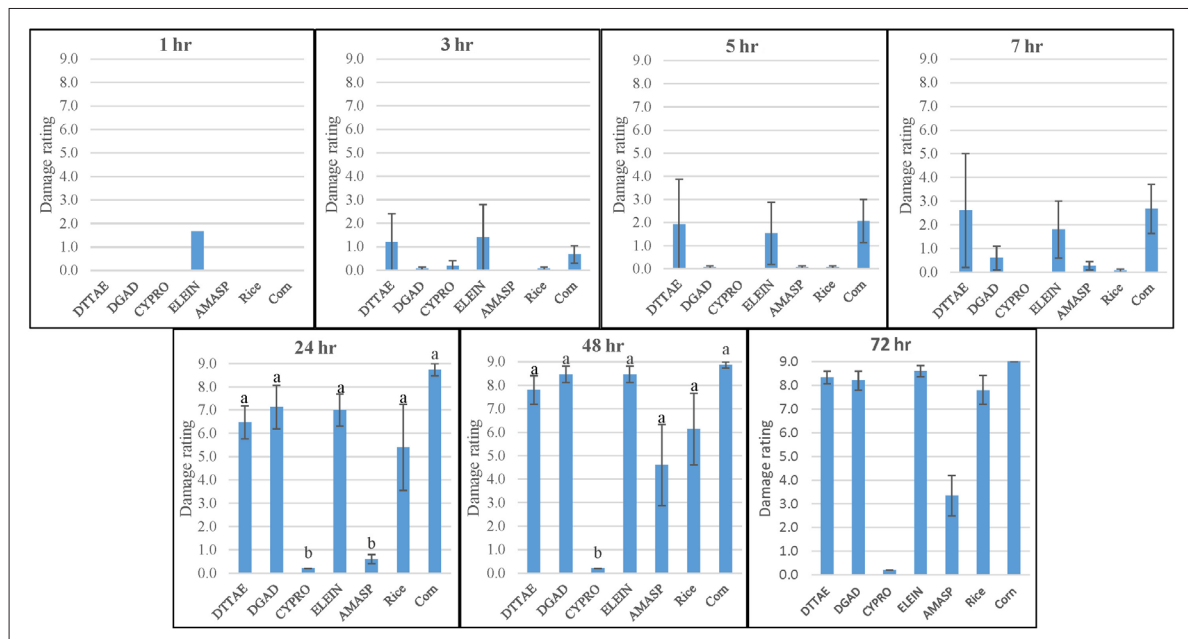


Figure 6. Feeding damage by FAW on various plants in free-choice host plant specificity test during the second trial in February 2023.

exhibiting minimal damage even after 48 h. When feeding rice, FAW primarily caused damage by severing the leaves.

The free-choice experiments, revealed a clear preference by FAW for certain plant species. This behavior aligns with the well-documented phenomenon of insect herbivore specialization. Many insect herbivores, including FAW, have evolved to focus their feeding on a limited number of host plants (Futuyma and Agrawal, 2009; Forister et al., 2012; Becerra, 2015). This specialization is often linked to the unique biochemical makeup of the

host plants. Interestingly, a study by Agravante et al. (2022) identified *Eleusine indica* (ELEIN) as one of the most favorable hosts for FAW development and reproduction. On ELEIN, FAW larvae exhibited the shortest developmental time, highest survival rates, and greatest fecundity. Notably, the study also found similar positive effects for FAW on *I. triloba* and *P. oleracea*, suggesting a potential preference for certain broadleaf plants alongside grasses.

The non-preference of FAW for *Cyperus rotundus* (CYPRO) may be attributed to its repellent and insecticidal properties. A study by Singh et al.

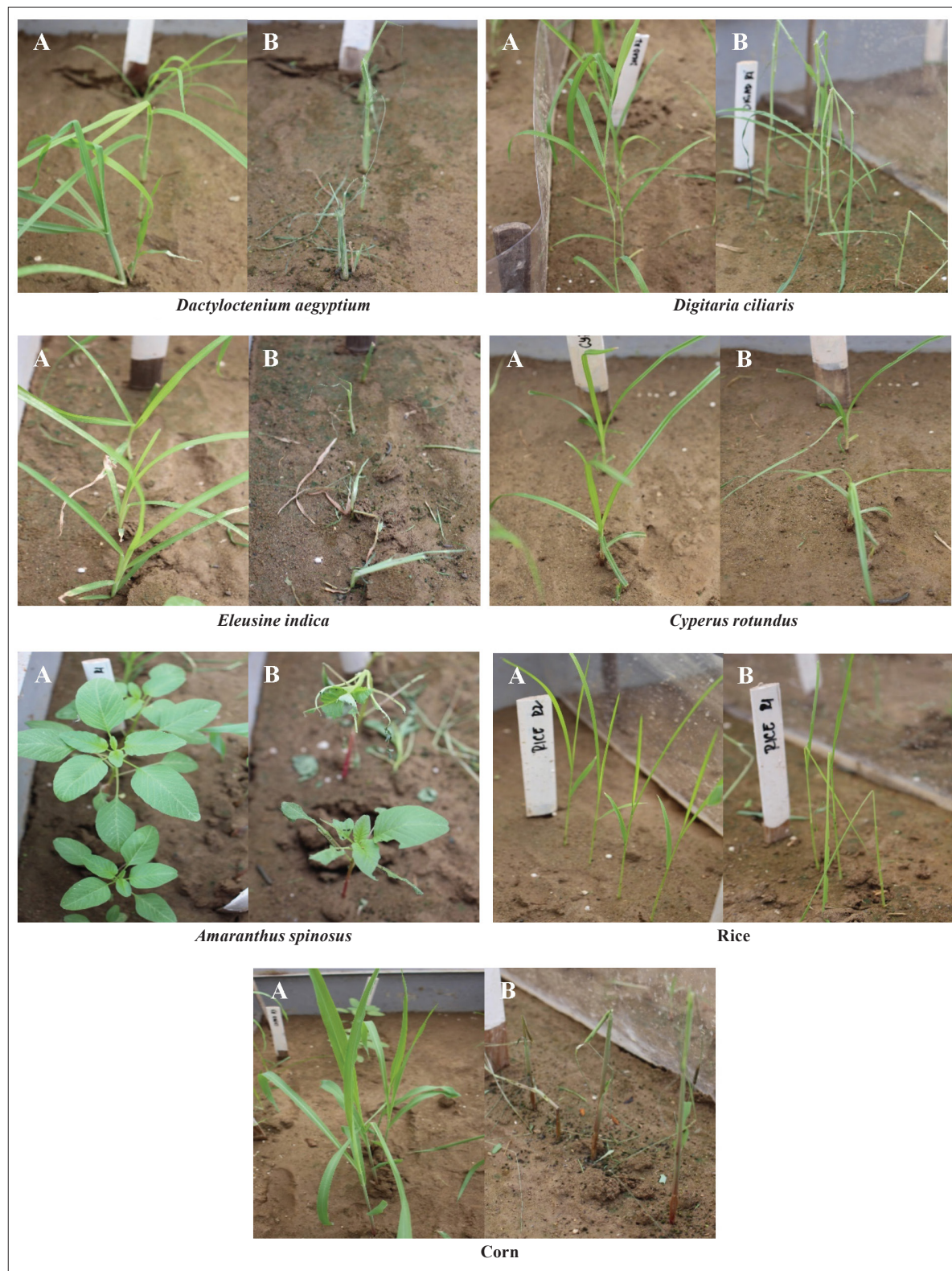


Figure 7. Free-choice setup for feeding preference assessment before release (A) and 48 hours after the release (B) of FAW larvae.

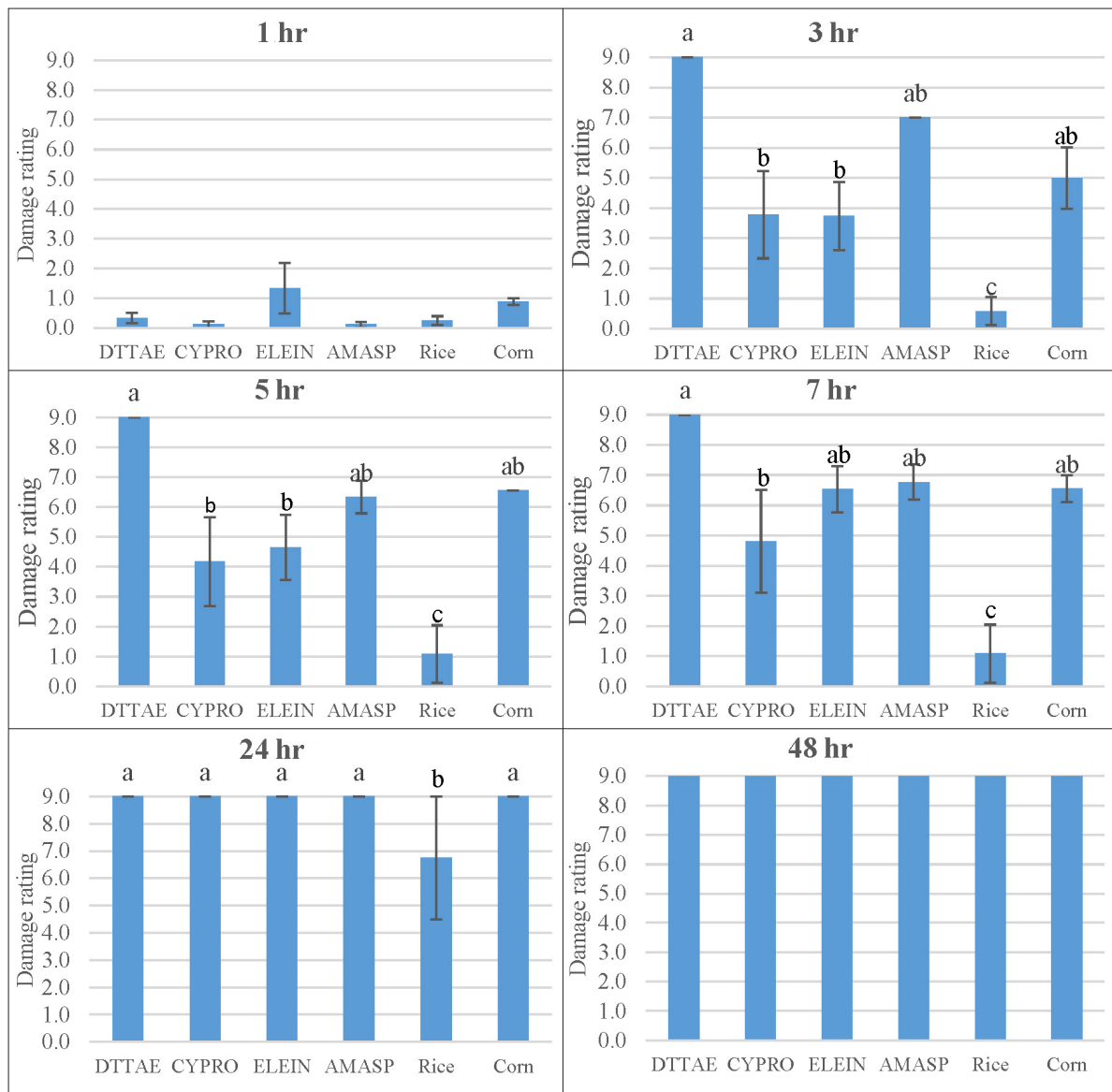


Figure 8. Feeding damage by FAW on various plants in the no-choice host plant specificity test during the first trial in September 2022.

(2009) demonstrated the effectiveness of hexane extract from *C. rotundus* tubers against *Anopheles culicifacies*, *Anopheles stephensi*, and *Culex quinquefasciatus*, even at low concentrations. Furthermore, Bañez and Castor (2011) reported that *C. rotundus* exhibited greater insecticidal activity than carbamate insecticides and comparable efficacy to organophosphates in controlling ants.

In the no-choice test of trial 1, where FAW larvae were confined to a single plant species, DTTAE and AMASP sustained the most significant damage within the first few hours after release. These plants received ratings of 9.0 and 7.0, respectively, at 3 and 5 h after release. Interestingly, when comparing damage across all weed species to corn, no significant differences were observed at 7 and 24 hours after release. This suggests FAW can adapt and feed on a

variety of weed species to a similar degree as corn within this timeframe. However, rice emerged as the least preferred host throughout the first 24 hours, consistently receiving the lowest damage ratings. By the 48-hour mark, though, feeding behavior shifted. All plants, including rice, exhibited the maximum damage rating of 9.0, indicating that FAW larvae eventually consumed significant portions of even the less-favored rice plants. (Figure 8).

Similar to trial 1, the no-choice test in trial 2 revealed a preference for certain plants early on. ELEIN, DTTAE, and DGAD sustained the most significant damage at 1 and 3 h after releasing FAW larva, with the highest rating being 4.5. Interestingly, feeding patterns shifted at the 7 h mark. ELEIN and DTTAE maintained relatively high damage ratings (4.5 and 2.1, respectively), but CYPRO emerged as

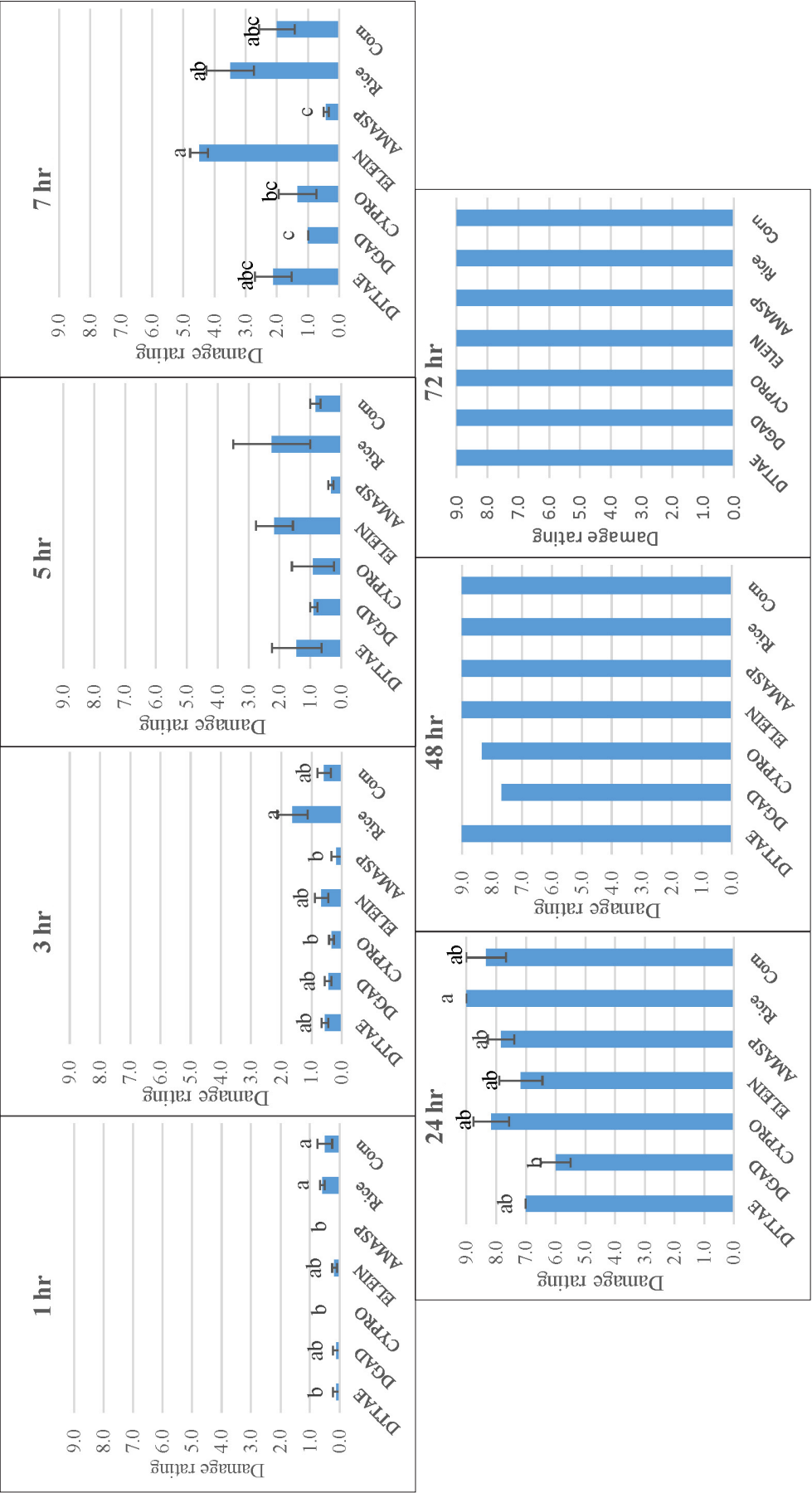


Figure 9. Feeding damage by FAW on various plants in no-choice host plant specificity test during the second trial in February 2023.

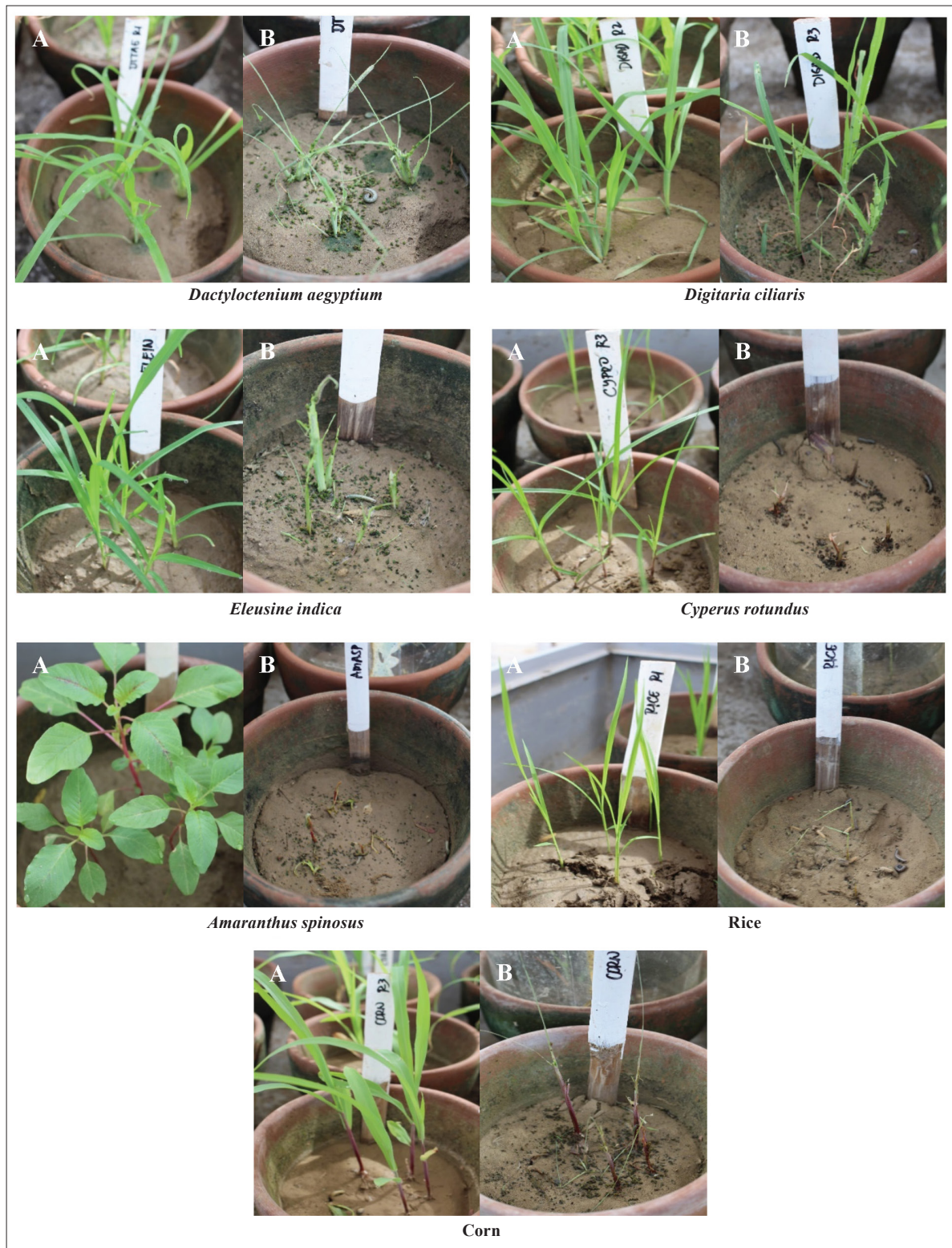


Figure 10. No-choice test setup for FAW feeding preference: (A) before FAW larvae release, (B) 48 h after the release.

a less preferred host receiving a rating of only 1.3. Conversely, DGAD and AMASP exhibited the least damage at this point. Throughout the remaining observation period (24 and 48 h), no significant differences were observed in damage ratings between any of the weed species, corn, or rice (Figure 9). This suggests that FAW larvae can adapt and consume all these plants to a similar extent within a day after release.

The findings from the free-choice test in trial 2 differed slightly. Here, CYPRO remained the least preferred host throughout the 48 h, with consistently lower damage compared to other plants. However, it is important to note that in the no-choice setup, where FAW larvae were confined to a single plant species, the damage inflicted on CYPRO was not statistically different from that on corn, rice, or any of other weeds at any point between 3 and 48 h after release. This suggest that FAW can eventually consume even less-favored plants like CYPRO if no other options are available.

Figure 10 shows that FAW consumed leaves and stems of all plants tested. In the no-choice set up, all

plants tested had 50 - 100% damage 48 h after release and 100 % damage 72 h after release.

Oviposition Preference Test

FAW females preferentially depositing their eggs on the underside of leaves or on plant stems (Figure 13). This behavior likely helps to camouflage the eggs and protect them from predators and environmental factors. In the first trial of the free-choice setup, FAW deposited eggs on DTTAE, DGAD, ELEIN, rice, and corn. ELEIN received the highest number of egg

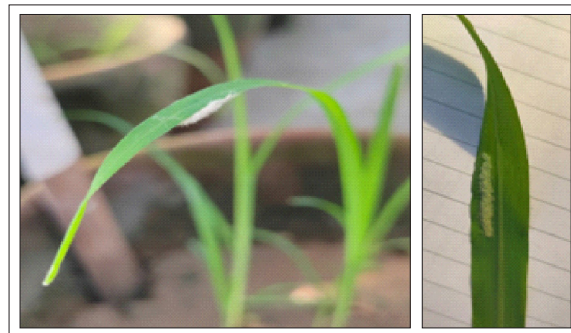


Figure 13. FAW eggs laid on the underside of a leaf.

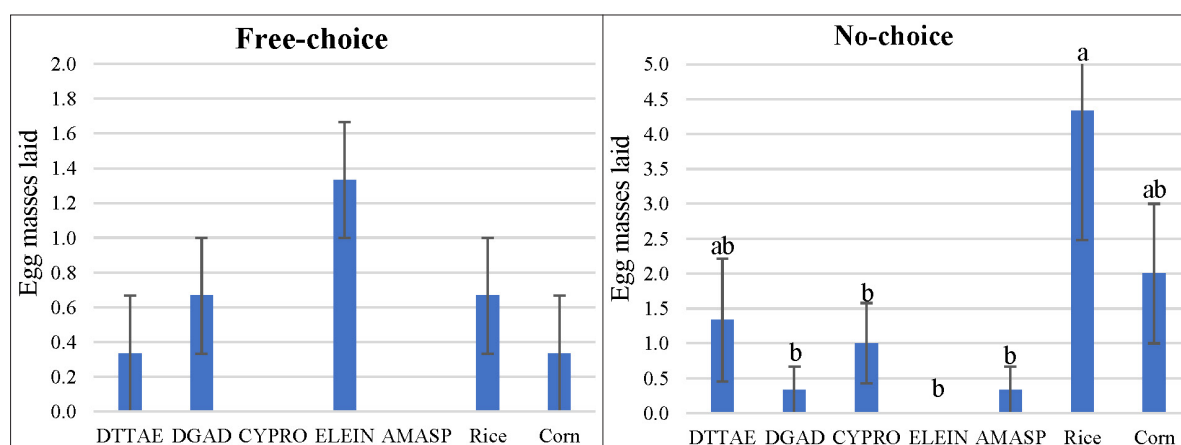


Figure 11. Average number of FAW egg masses laid on each plant in the free-choice and no-choice setups during the oviposition preference experiment, first trial.

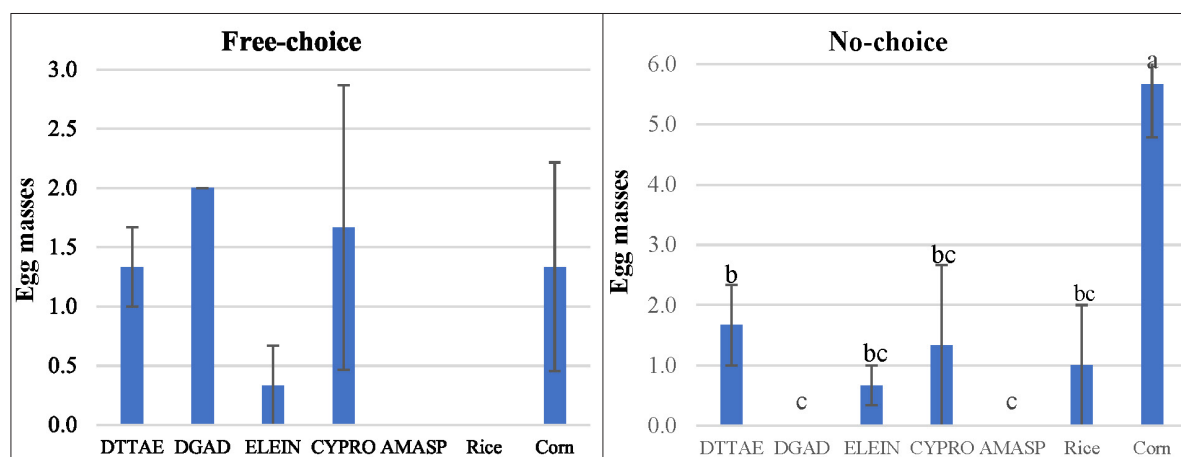


Figure 12. Average number of FAW egg masses laid on each plant in the free-choice and no-choice setups during the oviposition experiment, second trial.

masses, averaging 1.3 per plant. This was followed by DGAD and rice with an average of 0.7 egg masses each. Corn received a lower number of egg masses (0.3 per plant), while CYPRO and AMASP were not targeted for egg deposition (Figure 11). In the second trial, DGAD emerged as the most favored oviposition site, with an average of 2.0 egg masses per plant. CYPRO and DTTAE followed closely behind with 1.7 and 1.3 egg masses, respectively. Similar to trial 1, AMASP and rice were not selected for egg-laying (Figure 12).

The findings from the no-choice tests, where FAW females had only one plant species available for egg-laying, differed somewhat from the free-choice results.

In the first trial of the no-choice setup, DTTAE was the most favored weed species for egg-laying, with an average of 1.3 egg masses per plant. However, surprisingly, rice received a significantly higher number of egg masses (4.3 on average), despite not being a preferred choice in the free-choice setup. Interestingly, ELEIN, the most favored plant in trial 1 free-choice test, received no egg masses in the no-choice setup (Figure 11).

Similar to trial 1, DTTAE displayed a relatively high number of egg masses (1.7 on average). However, in this trial CYPRO, which was not targeted in the free-choice test, also received a notable number of egg masses (1.3). ELEIN again received a lower number of egg masses (0.7) compared to the free-choice setup, and DGAD along with AMASP did not receive any egg masses (Figure 12).

In the free-choice setup, ELEIN had the highest number of egg masses in the first trial while DGAD had the highest in the second trial. Notably, FAW avoided laying eggs on AMASP in both trials. In a no-choice setup among weed species, DTTAE had the highest number of egg masses.

While several plant species have been identified as potential hosts for FAW, not all provide equally suitable conditions for successful development and reproduction. The study by Montezano et al. (2018) listed DTTAE, DGAD, CYPRO, ELEIN, and AMASP as plants that FAW can feed on. However, as Volp et al. (2022) pointed out, a true FAW host requires attraction of egg-laying females, successful larval development, and adult emergence for reproduction. The findings of this study provide valuable insights into FAW host suitability. Notably, DTTAE, DGAD, and ELEIN emerged as the most preferred choices for both feeding and egg-laying in the free-choice tests. This suggests they can be classified as putative host plants for FAW, allowing for larval development but potentially not optimal for full lifecycle success. These results align with Agravante et al. (2022), who found that grass

weeds like *Rottboellia cochinchinensis* and *Eleusine indica* supported shorter development times, higher larval survival rates, and greater egg production in FAW compared to broadleaf weeds like *Ipomoea triloba*, *Portulaca oleracea*, *Corchorus olitorius*, and *Synedrella*. This highlights the importance of considering factors beyond just feeding preference when evaluating host suitability for FAW.

Conclusion

This study investigated FAW feeding preference and host suitability for various plant species. The free-choice experiments revealed a clear preference for certain weeds. *Digitaria ciliaris* (DGAD), *Dactyloctenium aegyptium* (DTTAE), and *Eleusine indica* (ELEIN) consistently showed the most significant damage over 24 h period in both trials. This preference extended to egg-laying behavior, as these same weed species attracted the highest number of eggs in the free-choice oviposition tests.

The no-choice setup yielded interesting insights. While DTTAE remained attractive for egg-laying, *Cyperus rotundus* (CYPRO), a less preferred choice in the free-choice scenario, also received a notable number of eggs. This suggests that FAW females may lay eggs on less favorable hosts if preferred options are unavailable.

Overall, the findings suggest that DGAD, DTTAE, and ELEIN can be classified as putative host plants for FAW. These weeds support larval development, but their suitability for complete lifecycle success (including egg production) remains unclear. Further studies are needed to investigate how these plants influence FAW development time, survival rates, and fecundity compared to known optimal hosts.

This study revealed a preference by FAW for feeding and egg-laying on DGAD, DTTAE, and ELEIN, mirroring their attraction to corn. The widespread presence of these weeds suggests they may serve as alternative hosts, particularly during fallow periods. However, further research is needed to determine if these weeds fully support the FAW lifecycle.

The potential for these weeds to provide FAW with food and egg-laying sites during the absence of corn and rice crops highlights the complexity of FAW management. This underscores the importance of integrated pest management (IPM) strategies that target FAW populations throughout the year, including fallow periods.

The presence of alternate host plants plays a critical role in FAW development and outbreak potential, especially when its primary host, corn, is absent (Chen et al., 2023). These alternative hosts allow FAW populations to persist (as a residual population)

even without corn. This residual population can then rapidly increase when suitable environmental conditions become favorable for FAW growth.

Furthermore, the broad host range of FAW facilitates its migration to other crops, posing a significant challenge for pest management strategies. The close proximity of various crops and their associated weed communities, especially those commonly found with corn, might even influence FAW host preferences when its primary host is absent. This potential shift in preference highlights the importance of managing weed populations that can serve as alternative food sources for FAW, in order to mitigate its population growth and development.

Acknowledgment

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EFFECT OF PEANUT HULLS ON SOIL SALINITY AND GROWTH OF RICE (*Oryza sativa* L.)

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Abstract

Soil salinization is one of the major constraints in rice production. Although there are existing methods that have proven the amelioration of salt-affected soils, inadequate resources pose restrictions to local farmers. This study determined the potential of peanut hulls in decreasing soil salinity and improving rice growth. A pot experiment was conducted consisting of three treatments with 15 pots each. Soils were simulated using various amount of salts such as sodium chloride, calcium chloride, and sodium sulfate. Crops were grown for 29 days. Soil salinity and growth parameters were measured and subjected to statistical analyses. Results indicated that soil salinity varied significantly due to a decrease in electrical conductivity (EC) over time. Instances of crop death were also observed in control groups. All treatments exhibited significant differences in growth parameters. The treatment that incorporated peanut hulls demonstrated the most substantial growth, followed by the treatment with vermicast, and then the control group. This indicates that the introduction of peanut hulls significantly influenced plant height, leaf count, and leaf color. The data suggest that peanut hulls have had a positive impact on both the properties of the soil and the growth of the crops. Therefore, peanut hulls show promise as an affordable and sustainable alternative for managing soil salinity and cultivating rice. This study lays the groundwork for more in-depth exploration of soil properties and the condition of rice crops at maturity.

Keywords: *Arachis hypogaea*, *Oryza sativa*, Organic matter, Soil amendment, Salinization

Introduction

Soil salinity is one of the most devastating environmental stresses that causes major reduction in prime cultivated area and crop productivity and quality (Ashraf and Shahbaz, 2013 as cited in Shrivastava and Kumar, 2015). This problem arises when the total amount of salts that accumulate in the root zone is high enough to affect plant growth negatively. Excess soluble salts in the root zone restrict plant roots from withdrawing water from the surrounding soil, effectively reducing the amount of water that is available to the plant (USDA, Natural Resources Conservation Service, 2002 as cited in Aderoju and Festus, 2016). In countries with hot and dry climates, the soil often has high salinity and low potential for agriculture. Despite these challenging conditions, crops in these regions are typically cultivated using irrigation methods (Glick et al., 2007 as cited in Ouda et al., 2018).

In 2011, the Philippine Rice Research Institute (PhilRice) identified salinity as a prevalent issue hindering rice production across most of the country. Coastal lands impacted by salinity are estimated to be around 0.4 M ha, with half of this area (0.2 M ha) being severely affected by salt. In the regions of Bicol and Cagayan Valley, it is reported that approximately

70,000 ha of rice fields are potentially at risk from saltwater intrusion.

The International Rice Research Institute (IRRI, 1997, as cited by De Young et al., 2012) classified rice as a crop that is sensitive to salinity. According to Grattan et al. (2002) as cited in Reddy et al. (2017), salinity delays heading in rice, which negatively affects a number of yield components. It is expected that over extraction of groundwater in coastal areas for household and agriculture uses and the rising sea level brought by global warming will increase the problem of salinization throughout the country (Asio et al., 2009). It is also expected that by 2050, more than 50% of the world's arable land will be salinized and a 50% significant increase in rice grain yields must be acquired to fulfill the food supply requirements of the projected population increase (Shrivastava and Kumar, 2015).

In the study of Fall et al. (2018), improvement of salt-affected fields can be achieved in various ways such as leaching, chemical remediation, and phytoremediation, but these have been impractical to farmers due to the lack of resources. Leaching is referred to as the removal of substances through the use of aqueous solutions such as rain, mist, and fog (Tukey Jr, 2003). A typical method used to remove

water-soluble chemicals from trash or soil. Although effective, it still possesses a negative impact on the soil in such a manner that all components are washed out from the surface. Soil remediation, on the other hand, is the process of reusing contaminated land by removing the contaminant around the soil (Pandit et al., 2023). Soil remediation could be done by using chemical reagents (chemical remediation) or by the use of plants and technology (phytoremediation). These may be effective but are highly costly and take time before the implementation of those processes is complete. Furthermore, soil remediation activities have a major effect on the quantity of hazardous waste a facility produces (Gunaratne et al., 2020).

In line with the designed paradigm (Figure 1), this study determined whether peanut hulls could reduce the salt content of the soil and improve rice growth as indicated by plant height, number of leaves, leaf color, and dry weight.

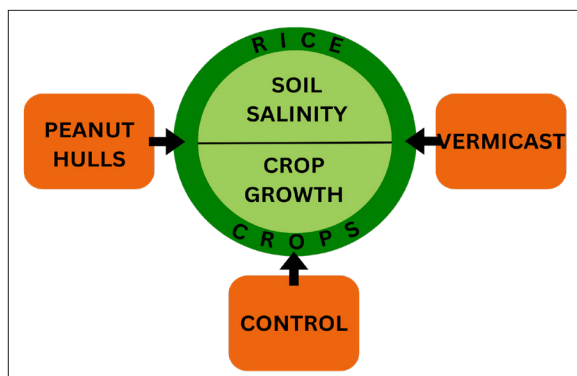


Figure 1. An illustration of soil salinity scenario using different methods affecting rice growth.

Rice is the most salt-sensitive cereal crop with a threshold of 3 dSm⁻¹ for most cultivated varieties (Grattan et al., 2002). Razzaq et al. (2019) stated that rice crops exhibit an abrupt response against the damaging effects of high salt accumulation in salt-affected soils. Salinity affects the crop in terms of height, weight, and number of grains. Moreover, changes brought about by salt stress impede the growth stages of rice and lead to yield losses.

Previous studies have shown an approach on this growing problem through incorporating organic materials into the soil (Fall et al., 2017; Omidi et al., 2019; Win et al., 2019). Abd Elrahman et al. (2012) added that the physical, chemical, and biological properties of soil in salt-affected areas are improved by the application of organic matter which then enhances plant growth and development. Subsequently, Hasanuzzaman et al. (2014) considered peanut hulls after decomposition served as a viable alternative source of organic fertilizer. Peanut hulls are the external shell of peanut (*Arachis hypogaea* L.). The peanut shells are usually thrown after

peanut processing and cleaning. To avoid the massive disposal of peanut hulls in landfills, these have been repurposed for various uses (Nalluri and Karri, 2018).

Fall et al. (2018), reported that peanut hulls were effective in reducing soil salinity together with other organic matter as it contains calcium, which replaces the ions affecting the soil salinity thus, reducing the ionization levels in the soil. Its application in *Senegalia senegal*, *Vachellia seyal*, and *Prosopis juliflora* plants improved their soil chemical properties, one of which is the Cation-exchange Capacity (CEC), and soil Electrical Conductivity (EC) where a decreasing trend was found. CEC is defined as the concentration of negative charges that can absorb and retain exchangeable cations, which is referred to as a reliable indicator of soil productivity. EC is a common measure of salinity and determined by the ability of the suspension to conduct electricity between two electrodes. The greater the salt concentration, the greater the EC level (Corwin and Yemoto, 2020).

Peanut hulls could not only reduce soil salinity but also increase the nutrient content in the soil such as carbon, nitrogen, phosphorus, and calcium structure (Fall et al., 2018). In a study conducted by Nalluri and Karri (2018), peanut hulls were used as an alternative to chemical fertilizers using a pot system. The results have shown to be an effective alternative as it enhances the growth and yield of vegetable plants. In Senegal, significant effects were observed in millet and corn plants affected by salinity (Proger, 2008 as cited in Fall et al., 2018). To date, there have been reports regarding the application of peanut hulls in paddy fields but in the form of biochar. These reports have cited beneficial effects on rice productivity and soil amelioration (Yao et al., 2021; Jin et al., 2022).

About 35,000 mt of peanuts are harvested each year in the country. However, this crop receives a small amount of attention for research and development (Palomar, 1998 as cited in Macatangay et al., 2012). Likewise, Mojiri et al. (2011) noted how its effects have never been scientifically reported for recycling and sustainable use. The crop's effect on soil's physical, chemical, and biological characteristics remained less prioritized.

The results of this study can contribute to the existing knowledge about the efficacy of peanut hulls in the agricultural sector. This can help local rice farmers in increasing productivity and provide an alternative way on how their crops can thrive and improve under saline conditions. Investigating its potential in soil will also be beneficial in reducing and maximizing the amount of waste being produced in factories. However, it is beyond the scope of this study to examine the potential effect of peanut hulls in large-scale rice fields as the experiment was done in pots.

Materials and Methods

Research Design and Experimental Setups

The pot experiment was carried out on an open area (i.e., rooftop) and followed a standard experimental design. The setup comprised three treatments with 15 pots each: (1) saline soil as the control, (2) saline soil with peanut hull, and (3) saline soil with vermicast.

Preparation of Soil Conditioners

Soil Simulation. The loam soil was purchased from an online shop in Taguig, Metro Manila. As suggested by Gupta et al. (2012) cited in Dheeravathu et al. (2018), 3.3 g NaCl, 0.9 g CaCl₂, and 0.6 g Na₂SO₄ are required for 1 L of 8 dSm⁻¹ EC water to saturate 2.5 kg of soil. These quantities were computed salt requirements for the desired saline water level. The salts required for the simulation were purchased from Alysons' Chemical Enterprises, Inc. An amount of 35 kg of soil was used to fill 45 pots, resulting in 46.2 g NaCl, 12.6 g CaCl₂, and 8.4 g Na₂SO₄, dissolved in 14 L water. The prepared saline water and soil were combined and transferred to the respective pots sized 5.5" in height and diameter without drainage holes (holes were covered with black trash bags). These were watered daily for 7 days with 175 mL of water to ensure uniform salt distribution with the soil before planting the seeds.

Treatment. Peanut hulls were crushed through the use of a kitchen grinding machine until grinded in small sized particles and sieved thoroughly until no chunks passed through. For vermicast, it was purchased at the same online shop in Taguig, Metro Manila from where the soil was bought. These were incorporated into respective treatment groups with 7.5 t ha⁻¹ (45 g pot⁻¹) recommended rate (Bako et al., 2022) per pot after the 7-day soil simulation.

Preparation and Management of Crop

NSIC 2016 Rc 470 or Salinas 25 was provided by the Philippine Rice Research Institute (PhilRice) and grown for 29 days. The seeds were pre-germinated by soaking for 24 h and another 24 h incubation. Five seeds were planted in every pot using the wet direct-seeding method used by IRRI's protocol. Watering was regulated after two weeks using 300 mL maintaining 2.5 cm water level above soil surface.

Assessment of Soil Salinity

Soil salinity was measured prior to application of treatments for three weeks. A soil salinity tester purchased from Shen Zhen YIERI Technology Co., Ltd was used every week to measure EC (μS/cm) levels.

Assessment of Plant Characteristics

In determining the growth parameters, data was measured at vegetative stage according to the following parameters: plant height, number of leaves, leaf color, and dry weight. Plant height was measured from the soil surface to tip of the highest leaf from 2 to 4 weeks using a standard tape measure. The number of leaves were counted. The leaf color chart acquired at PhilRice was used in quantitative measurement by assigning a numerical value per shade of color green. The highest leaf was used as reference for charting. A value of 2 (yellow-green) implies low concentration of nitrogen and a value of 5 (dark-green) implies high concentration of nitrogen. The shoot and root parts of the crop were oven dried at 80°C for 15 h to obtain the dry weight along with the root-shoot ratio computed using the following formula:

Due to the death of few plants in negative and positive control, seven randomly selected plants from each treatment were used in analyzing leaf color and dry weight. This is to properly test and interpret the remaining data based on statistics.

Statistical Analysis

Data analysis was performed using Jamovi Statistical Software. The means and standard deviations per parameter were calculated. Assessment between the difference of the obtained initial and final soil salinity was checked with the student's t-test. The final measurement of soil salinity and each growth parameter were subjected to either parametric and non-parametric tests, determined by the Shapiro-Wilk Test under tests for normality and Levene's Test under tests for equal variances to reveal significant effects among and between treatment groups. Parametric Tests involved Fisher's and Welch's One-way ANOVA followed by the Tukey Post-Hoc Test and Games-Howell Post-Hoc Test, respectively. On the contrary, the non-parametric tests involved Kruskal-Wallis Test along with Dwass-Steel-Critchlow-Fligner Pairwise Comparisons.

Results and Discussion

The following results were obtained according to different parameters. A 5% level of significance was utilized throughout the analysis.

Table 1 shows an overview of the various parametric and non-parametric statistical tests used in the analysis. Each treatment was determined through testing for normality and equal variances.

In Table 2, the control shows the highest average EC of 2,321 μS/cm (SD = 192.9) while the vermicast treated pots obtained the least at 2,214 μS/cm (SD =

Table 1. Overview of the statistical test.

Parameters	Test for Normality	Test for Equal Variances	Statistical Tests
Initial and Final soil salinity	normally distributed	-	Paired-Samples t-test
Final soil salinity	normally distributed	unequal variances	Welch's One-Way ANOVA, Games-Howell Post-Hoc Test
Plant height	not normally distributed	unequal variances	Kruskal-Wallis Test, Dwass-Steel-Critchlow-Fligner Pairwise Comparisons
Number of leaves	normally distributed	equal variances	Fisher's One-Way ANOVA, Tukey Post-Hoc
Leaf color	not normally distributed	unequal variances	Kruskal-Wallis Test, Dwass-Steel-Critchlow-Fligner Pairwise Comparisons
Dry weight			
Root	not normally distributed	equal variances	Kruskal-Wallis Test
Shoot	normally distributed	equal variances	Welch's One-Way ANOVA
R/S Ratio	normally distributed	unequal variances	Fisher's One-Way ANOVA

Table 2. Electrical Conductivity (EC) of soil under different treatments.

Treatment	Initial and Final Soil Salinity	Initial Soil Salinity	Final Soil Salinity	Soil Salinity Reduction
Control	-	2321	1455a	866
Peanut hulls	-	2275	933b	1342
Vermicast	-	2214	1153c	1061
P	<.001*	-	<.001*	-

Means with the same letter within a column are not statistically different at 5% level. * = significant at 5% level

99.1). For the final soil salinity, the control had the highest average EC of 1,455 $\mu\text{S}/\text{cm}$ (SD = 359.4). Peanut hull-treated pots had the lowest average at 933 $\mu\text{S}/\text{cm}$ (SD = 161.9) and observed to have a greater salinity reduction.

Initial and Final Soil Salinity. The treatments applied led to a notable reduction in soil salinity. All treatments resulted in a decrease in EC within 3 weeks. As reported by Machado and Serralheiro in 2017, this reduction can be attributed to irrigation and fertilization practices. Varying EC values per week were also observed. Gong (2022) explained how these were affected by irrigation, fertilizer application, unalterable soil minerals, climate, and soil texture. In their study, Wang et al. (2014) investigated the use of green waste compost. They highlighted the capacity of organic matter to speed up the leaching of Na^+ in saline soils, which resulted in a decrease in EC and soil salinity.

Final Soil Salinity. Final soil salinity measurements of the treatments applied varied from one another. This demonstrates the extent of the decrease in EC values observed during the final measurement stage and the disparity in EC values among the different treatments. Significant differences between treatments were observed, which implies that peanut hulls produced a significant contribution

in the decline of soil salinity. Consistent with the results of Fall et al. (2018), the application of peanut hulls increased the amount of exchangeable cations, specifically Ca^{2+} , K^+ and Mg^{2+} , which restricts the entry of Na^+ into the soil's exchange complex. It then emphasized the positive contribution of peanut hulls in terms of CEC in decreasing soil salinity. Their results exhibited high CEC with the incorporation of peanut hulls and led to a high rate of organic matter mineralization.

Seven pots from the control treatment (Figure 2) and one pot from the vermicast treatment (Figure 3) had dead plants during the experiment process. During germination, which is an early growth stage, plants exhibited a higher sensitivity to salt stress than they did in later stages (Machado and Serralheiro, 2017). Symptoms of salt injury such as drying of leaves, white leaf tips, and stunted growth, which align with PhilRice's observations, were noted during the growth period. Queensland Government (2013), reported that osmosis occurs within the system; water may flow from the plant roots back into the soil if excessive salt level is in the soil water, which then dehydrates the plant causing yield loss or plant death. Lazcano and Domínguez (2011) observed that the impact of vermicast treatment on plants is not consistent and can vary over time. This inconsistency in the effects



Figure 2. Crop death on the negative control.



Figure 3. Crop death on vermicast treatment.

of vermicast can sometimes result in stunted growth or even death of the plants. The degree to which these effects vary depends on several factors, including the cultivation system used, the specific characteristics of the vermicast, its production process, and the age of the crop being treated.

Plant Height. Table 3 presents the lowest mean plant height was 14.6 cm (SD = 14.22) in the control while the highest was at 30.4 cm in peanut hulls treatment (SD = 4.04). Among these treatments, peanut hulls had a notable advantage in plant height. Peanut hulls-treated pots had taller plants compared to other treatments. Plant height varied among treatments and manifested a more positive effect. Significant differences between treatments were also observed. The substantial difference between the control and peanut hull treatment indicated how peanut hulls affect plant height. This observation was similar to Torkashvand et al. (2014) on peanut shell compost decreasing the C/N ratio and abundant

nutrients in the compost that improved plant height. The C/N ratio refers to the amount of carbon relative to the amount of nitrogen available. Decreased C/N ratio corresponds to a prompt release of nitrogen into the soil for the plant to make use (Brust, 2019). Improved plant height in rice determines the grain yield and quality (Yu et al., 2020) that is vital for farmers and consumers.

Number of Leaves. The lowest number (Table 3) of leaves was 5.87 in the control (SD = 6.63) while the highest was 16.53 (SD = 6.88) in peanut hulls treatment. Among the treatments, peanut hulls had more leaves, which are capable of producing more leaves than the other treatments. Significant differences were observed from treatments between the control and peanut hulls, as well as between the peanut hulls and vermicast. Results imply that peanut hulls with significant amount of nitrogen in soil can greatly influence the number of leaves produced. Nitrogen is an essential element that significantly influences plant growth and development, as it is a constituent of vital compounds that regulate these processes (Shrestha et al., 2020). This assertion is corroborated by the study conducted by Mahboub Khomami et al. (2019), which demonstrated that the application of peanut shell compost, a rich source of nitrogen, resulted in an enhancement of growth parameters, including an increase in leaf count. Hua et al. (2021) have indicated that the yield of rice crops is intrinsically linked to the efficiency of photosynthesis. Therefore, practices that increase leaf count and, consequently, the photosynthetic capacity of the plant, can potentially lead to an increase in rice yield.

Table 3. Growth parameters of rice crop under different treatments.

Treatment	Plant Height (cm)	Number of Leaves	Leaf Color	Dry Weight (g)		R/S Ratio
				Root	Shoot	
Negative control	14.6a	5.87a	2.57a	0.0786	0.1286	0.6332
Peanut hulls	30.4b	16.53b	4.14bc	0.1571	0.3243	0.4277
Vermicast	23.7a	9.47a	2.86ac	0.0629	0.12	0.6144
P	<.001*	<.001*	0.010*	0.429ns	0.050ns	0.323ns

Means with the same letter(s) within a column are not significantly different at 5% level. * = significant at 5% level, ns = not significant

Leaf Color. As seen in Table 3, the control displayed a lowest average leaf color value of 2.57 (SD = 0.535). The peanut hulls, on the contrary, displayed the highest average leaf color value of 4.14 (SD = 1.069). Among the treatments, peanut hulls had a darker shade of green, which means these have higher concentration of nitrogen than other treatments. Leaf color varies among treatments and the recommended shade of green identifies a greater concentration of nitrogen. Peanut hulls increased nitrogen intake in plants as it increases substrate availability for microbial activities (Fall et al., 2018). The organic matter is mineralized, which makes more nutrients available to plants. Increase in nitrogen rates would result in darker shades of green, while inadequate nitrogen causes chlorosis or yellowing of plants (Hou et al., 2020). Results implied that peanut hulls can increase nutrients in the soil and thereby expressed in the dark green color of leaves. Hence, applying peanut hulls can increase nitrogen content in the soil (Win et al., 2019).

Dry Weight. The growth of plants, in terms of dry weight, is influenced by several factors including root dry weight (RDW), shoot dry weight (SDW), and the root/shoot (R/S) ratio, as presented in Table 3. Khosh Kholg Sima et al. (2012) conducted a study on barley and found that salt stress negatively impacted RDW. This was attributed to the high osmotic potential of the soil solution, which reduced the plant's ability to absorb water and nutrients. Similarly, the SDW of paddy cultivars was affected by salinity, as observed by Puvanitha and Mahendran (2017). The detrimental effects were primarily due to the toxicity of sodium chloride (NaCl) and its interference with nutrient uptake. These findings underscore the significant impact of environmental stressors, such as salinity, on key growth parameters of plants. The R/S ratio, which did not yield desirable values in this study, is an indicator of a plant's health status. As Bláha (2019) noted, a higher R/S ratio typically corresponds to enhanced nutrient absorption and vice versa. However, Price and Munns (2017) found that conditions of low water or nutrient availability often result in a higher R/S ratio, seemingly contradicting the previous assertion.

This apparent contradiction was addressed by Suzuki (1983), who explained that the dry matter distribution among plant organs is subject to variation depending on the growth stages and environmental conditions, such as temperature. Therefore, the R/S ratios observed in this study are not static but will continue to change as the crops grow and encounter different environmental conditions. This suggests that the current data does not provide sufficient evidence to conclusively determine the effect of peanut hulls on the R/S ratio.

On the overall impact of peanut hulls on growth parameters of rice, it was consistently shown that plant height, number of leaves, and leaf color had substantial differences between the control and peanut hulls. There were differences between peanut hulls and vermicast treatment but no significant differences between the control and vermicast. Positive impacts of peanut-shell-based amendments were noticed in crown daisies and the improvement of the said growth parameters could be attributed mainly to macro and micronutrients brought in the soil (Nazir et al., 2021).

Vermicast had improved various soil properties including CEC, through increased organic matter, nutrient contents, soil aeration, and soil enzyme action (Bziouech et al., 2022). These factors had lowered soil salinity and mitigated its harmful effects. Tomatoes grown in their study have achieved maximum plant growth in vermicast substrates amidst salt stress. The beneficial effects on both soil and plant development observed in this study align with the findings of previous research. However, the impact of vermicast and the control on growth parameters did not show a significant enough difference to conclusively determine the potential of vermicast. In a related study conducted by Dollison (2023), vermicast was utilized as an organic soil enhancer for NSIC Rc 160 (Tubigan 14), and its effects were compared with those of inorganic fertilizers commonly used in agriculture. Although no significant difference was found between the application of organic and inorganic fertilizers, the study acknowledged that vermicast could potentially serve as a promising alternative for sustainable agricultural production.

Proliferation of algae, predominantly in pots containing peanut hulls, was also observed in the experiment, which can be attributed to the high nutrient content of the hulls. As described by Ohadi et al. (2022), algae thrive in environments with excessive levels of nitrogen and phosphorus, such as rice fields. While the presence of algae may initially interfere with the growth of rice seedlings, it could potentially be beneficial in later stages of rice development due to their nitrogen-fixing capacity. This observation aligns with the findings of Saito and Watanabe (1978), who reported similar occurrences in both fertilized and unfertilized rice plots. This suggests a complex interaction between algae, nutrient availability, and rice growth that warrants further investigation.

The results of this study underscore the potential of peanut hulls as a valuable resource in rice production. Zhou et al. (2023) have articulated the concept of transforming crop waste into a means of enhancing soil and plant health, thereby contributing to agricultural sustainability. This approach not only promotes resource efficiency but also offers a solution to mitigate plant stressors. The effectiveness of this

strategy has been demonstrated in this study through the successful utilization of peanut hulls.

The results of this study were derived under controlled conditions, yet certain limitations were encountered in the experimental setup and data collection process. The methodology employed pots without drainage holes, which were lined with black trash bags to prevent leakage. Despite these precautions, water run-off was observed in several pots, indicating a potential flaw in the setup.

The measurement of soil salinity was intended to be carried out over a four-week period, but was unfortunately limited to 3 weeks due to a malfunction in the measuring tool. Consequently, the study was unable to thoroughly examine and analyze the effects of each treatment on the condition of the rice crops and soil at later stages.

Furthermore, the simulated salinized soil did not meet the EC threshold of 3,000 $\mu\text{S}/\text{cm}$, which is considered detrimental to rice. The maximum recorded reading was only 2,580 $\mu\text{S}/\text{cm}$, classifying the soil as only slightly saline. This discrepancy suggests that the simulated conditions may not have accurately represented the harsh conditions under which salinity-stressed rice crops typically grow. These limitations should be taken into account when interpreting the findings of this study.

Conclusion and Recommendations

The study demonstrated the efficacy of peanut hulls in reducing soil salinity and enhancing crop growth. A significant reduction in EC by -1342 $\mu\text{S}/\text{cm}$ was observed, indicating a strong correlation between the application of peanut hulls as a soil conditioner and the decrease in EC levels in salt-affected soils. Moreover, the use of peanut hulls had a positive impact on crop growth parameters. The effectiveness of peanut hulls, a low-cost and sustainable by-product, suggests their potential as an alternative soil conditioner for crops in saline-prone paddy fields.

However, further research is needed to fully realize the potential of peanut hulls in salt-affected soils. It is recommended to monitor the crops until maturity to accurately determine the yield and other growth parameters under conditions of high soil salinity. A comprehensive soil analysis, both pre- and post-planting, is suggested to identify the specific nutrient contents in the soil that could contribute to the reduction or increase of soil salinity. Additional data on soil parameters such as pH and NPK contents, should also be considered. To validate the findings of this study, it is recommended to replicate the methods under both controlled and field conditions.

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SOIL FERTILITY, PEST DYNAMICS, DISEASE INCIDENCE, AND PERFORMANCE OF SOYBEAN APPLIED WITH RADIATED CARRAGEENAN

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Abstract

This study evaluated the performance of soybean varieties, pest dynamics, and disease incidence under different types of carrageenan applications. A split-plot design with two factors and three replications was employed. Growth and yield performance were assessed using analysis of variance and Least Significant Difference tests. Soil fertility dynamics were measured by analyzing pH, organic matter, phosphorus (P), and potassium (K) levels. Additionally, disease incidence, pest dynamics, and return on investment (ROI) were evaluated. Among the tested varieties, IPB 96-27-23 showed the best agronomic performance and responded positively to carrageenan application. Generally, soil pH, P, and K decreased during the vegetative stage but increased at maturity, while soil organic matter increased during the vegetative stage and declined at harvest. Disease and pest incidence were minimal throughout the study, with no significant impact on soybean yield, although slightly higher incidences were observed in soybeans treated with irradiated carrageenan. The highest gross income was achieved by IPB 96-27-23 using commercial fertilizer combined with irradiated carrageenan, while the highest ROI was obtained with commercial fertilizer alone. This study demonstrates that while the application of carrageenan to IPB 96-27-23 variety enhances yield and soil fertility, it is less profitable compared with conventional fertilization methods.

Keywords: Irradiation. Profitability, Soybean production, Supplementation, Yield increase

Introduction

Soybean (*Glycine max* L.) Merrill, is a versatile leguminous crop that is increasingly integrated into rice-based farming systems due to its agronomic and economic benefits. Originating from East Asia, soybeans are renowned for their high protein content and valuable oil, making them a vital crop for food, feed, and industrial applications. When intercropped with rice, soybeans offer several advantages, including enhanced soil fertility through nitrogen fixation (Giller, 2001), efficient land utilization, and diversification of farm income.

Soybeans thrive in well-drained soils with a neutral pH, conditions often found in fields previously cultivated with rice. The crop has a relatively short growth cycle of 3 - 4 months, allowing for efficient rotation with rice. Intercropping or rotating soybeans with rice can improve soil structure, reduce pest and disease incidence, and increase overall productivity of the farming system.

According to Rathorea et al. (2009), any improvement in the agricultural system that results in higher production should reduce the negative environmental impact of agriculture and enhance the sustainability of the system. Previous studies

conducted to soybean assessing the effect of carrageenan or seaweeds extracts gained positive results but using it in irradiated form is a research gap, which this study had addressed.

Magsino et al. (2008) claimed that carrageenan-derived polysaccharide from seaweeds treated to break its particle down into very small pieces enhances growth as foliar organic fertilizer.

Although several trials were conducted in the country and in the region on the effect of irradiated carrageenan on rice, its effect in the growth and yield of soybean crop has not been tested. Furthermore, its effect on soil fertility, pest dynamics, and disease incidence is not yet documented in soybean. This study determined the soil fertility, pest dynamics, disease incidence, and agronomic performance of soybean varieties applied with irradiated carrageenan as foliar fertilizer.

Materials and Methods

The following materials and equipment were used in the study: seeds of soybean varieties (IPB -96-27-23, PSB Sy-6 or Tiwala 8, and Manchuria), radiated carrageenan, red seaweeds, blender, molasses, labeling materials, soil sample bags, pails,

sacks, spade, bolo, knife, paper, plastics, weighing scale, digital balance, motor-drawn plow and harrow, sprinkler, drums, complete fertilizer (14-14-14), Muriate of Potash (0-0-60), sprayer, moisture meter, and rhizobia inoculants.

Experimental Design and Treatments

Experimental treatments. The experiment had two factors. Factor A was the soybean varieties including A₁ - IBP-96-27-23, A₂ - PSB Sy-6 or Tiwala 8, and A₃ - Manchuria. Factor B was represented by the kinds of carrageenan and the commercial granular fertilizer including B₁ - Recommended rate of commercial granular fertilizer (CGF), B₂ - RR of commercial granular fertilizer (CGF) plus RR of commercial radiated carrageenan (CRC), B₃ - RR of commercial granular fertilizer (CGF) plus RR of radiated carrageenan (RC), B₄ - RR of radiated carrageenan (RC) and B₅ - RR of granular fertilizer plus RR of fermented seaweed extract (FSE).

Experimental design and field lay-out. The study was laid out in split-plot design with three blocks each representing a replication. The main plot was represented by the different soybean varieties and the subplots were the different types of carrageenan in combination with commercial granular fertilizer. The area was divided into three for the main plots spaced at 1 m from each other. Each main plot was further sub-divided randomly into five subplots with a spacing of 50 cm from each other. Fifteen subplots for each block were made: each having a dimension of 4 x 5 m.

Cultural Management Practices

Selection of experimental area. Department of Agriculture-Capiz Research Outreach Station in Astorga, Dumarao was inspected to determine the appropriate area for the experiment. Two criteria were considered in the selection: (1) uniformity of the area in terms of slope and fertility gradient and (2) an area with at least 1,470 m².

Land preparation. The study was conducted in a 1,470 m² experimental area. The area was cleared from unnecessary vegetation by plowing and harrowing twice using a hand tractor with one week interval between operations. Weeds were manually removed and cleared. Field was furrowed immediately prior to planting.

Securing the planting materials and inoculants. The soybean seeds of the different varieties were secured from the Department of Agriculture-Western Visayas Integrated Agricultural and Research Center (DA-WESVIARC), Hamungaya, Jaro, Iloilo City and in Luzon, while seed inoculants were secured from DA-Regional Soils Laboratory.

Seed inoculation. Seed inoculants were procured from DA Regional Soils Laboratory and were applied before planting by mixing it with the seeds at a ratio of one pack (100 g) per 20 kg seeds. During seed inoculation, 10 kg of seeds were placed in a basin and were moistened with a glass of water. The inoculants were poured and mixed thoroughly under the shade after which inoculated seeds were sown following treatments.

Planting of soybean. Soybean seeds were planted on the furrows following an east-west direction at 50 cm apart at a depth of 5 cm with two seeds per hill. The seeds were dropped 5 cm apart or 50 cm per row by 5 cm per hill. The seeds were then covered with soil. Two border rows in each side of the plot were established.

Replanting and thinning. A week after germination, the missing hill was replanted following the recommended practices for soybeans. Similarly, some plots were thinned with extra growing plants leaving one seedling per hill two weeks after the emergence of the plants. Extra plots were made simultaneously planted with different varieties outside the experimental area and were used for planting of missing hills. The less healthy and undesirable plants were removed to have a uniform growth and number of plants per hill.

Fertilizer application. Commercial granular fertilizer was applied uniformly in every treatment through broadcast method based on the recommended rate per hectare and treatments.

Preparation of fermented seaweed extract (FSE). Red seaweeds (*Kappaphycus leverezeii*) were purchased from a reliable source, weighed and crushed using blender, boiled for 45 min, and cooled. It was then added with water and molasses and was fermented for 15 days. The ratio of the materials was 1 kg crushed seaweed, 5 L water, and 2 L molasses. After 15 days of fermentation, it was harvested by separating the juice from the solid materials placed in a clean container and stored in a cool dry place defined by DA-RAFIS (2017).

Application of different kinds of carrageenan. The different kinds of carrageenan were applied thrice to the soybean crop at 20, 40, and 60 days after planting (DAP) at the rate of 20 mL/L water depending on the treatments. The liquid forms of carrageenan were applied as foliar spray.

Analysis of different kinds of carrageenan. After acquisition of irradiated carrageenan, commercial carrageenan, and fermented seaweed extract, 100 mL sample for each kind of carrageenan were brought to the DA Regional Soils Laboratory 6 DA-WESVIARC and Capiz State University Pontevedra Campus for nutrient analysis.

Water management. Plants were watered during dry months when the soil moisture was insufficient to support crop growth. The same amount of water was applied for every experimental unit.

Weed management. Inter row cultivation and hilling-up were practiced 15 days after emergence. Hand weeding was also employed to control weeds whenever necessary.

Insect and disease management. Prevalence of insect pests and disease was monitored and recorded biweekly until harvest. Clean culture was also practiced within the experimental sites.

Harvesting and post-harvest operation. The pods were picked when these turned brown or approximately 85 - 90 days after planting. The pods were collected in separate containers with proper labels. Soybeans were threshed through the de-podding method that separates the grains from the pods using the hands. The seeds were properly cleaned by winnowing and sun-drying for 24 h prior to weighing.

Collection and Preparation of Soil Samples

Fifteen composite samples, representing the total number of experimental units, were collected. Approximately half a kilogram of sub-samples was taken from each plot at a depth of 20 cm before planting, during the vegetative and reproductive stages (45 DAP), and after harvesting. The soil samples were air-dried for one week, occasionally crushed during the drying process, and finally passed through a 2-mm mesh sieve. Following these preparations, the soil samples were placed inside labeled plastic bags and submitted to the DA Region VI for analysis of soil pH, organic matter (%), available P (ppm), and exchangeable K (ppm).

Data Gathered

Growth parameters of soybeans. Thirty sample plants were randomly selected from the inner rows of each plot to serve as representative specimens. Measurements of various growth parameters were taken from these sample plants.

Plant height (cm). The height parameter was measured from the base to the tip of the tallest part of the plant two weeks before harvest. This measurement was obtained by dividing the total plant height by the number of sample plants within the plot.

Fresh biomass ($t\ ha^{-1}$). Soybean plants were randomly selected and uprooted to determine their biomass. Soil debris was properly removed before weighing the plants. This procedure was carried out when the plants were in full maturity, either before leaf fall or two weeks before harvest.

Number of nodules per plant. Ten soybean plants were randomly selected from the outer rows, out of the 30 sample plants, and uprooted to determine the number of nodules per plant. Soil debris was carefully removed, and the nodules per plant were counted. This assessment was conducted after the flowering stage during active nodulation.

Yield parameters of soybeans. Thirty sample pods were randomly selected from the 20 sample plants from each plot.

1000-seed weight (g). Randomly selected 1,000 soybean seeds from each plot, then dried under the sun to achieve 14% moisture content (MC) and assessed using a moisture meter. Weight was measured using a weighing scale.

Yield ($t\ ha^{-1}$). All the grains harvested from sample plants in the harvestable area were collected, dried to achieve 14% MC, and assessed using a moisture meter. Subsequently, these were placed in separate containers with proper labels and weighed in kilograms using a weighing scale. The yield was then expressed in tons per hectare ($t\ ha^{-1}$) using the following formula:

$$Yield\ t\ ha^{-1} = \frac{Yield\ per\ sub-plot\ (kg)}{1000\ kg} \times \frac{(100-MC)}{86} \times \frac{10,000\ m^2\ ha^{-1}}{Sub-plot\ area\ (in\ m^2)}$$

Disease Incidence. Disease incidence was assessed by counting the number of plants infected by the disease within a plot. Data were collected at 10, 30, 50, and 70 DAP for the soybean crop. The percent incidence was calculated using the following formula:

$$Pest\ damage\ incidence\ (\%) = \frac{No.\ of\ damaged\ plants}{No.\ of\ sample\ plants} \times 100$$

Insect pest dynamics. Insect pests, natural enemies, and other insects affecting soybeans in the experimental area were monitored every two weeks to assess their population. This monitoring involved collecting specimens using insect-catching nets and handpicking in the morning. Collected insects were then brought to the laboratory for sorting and identification based on reference materials. Damage to plants in each plot was also recorded.

Soil Fertility Dynamics. The data on soil organic matter (SOM), soil pH, available P (ppm), and exchangeable K (ppm) were obtained from the results of soil analysis.

Computation for economics of production. The profitability of different soybean varieties, as affected by various treatments, was assessed using the return on investment (ROI). ROI is calculated by dividing the

net income by the total investment and multiplying the result by 100. Net income is obtained by subtracting total expenses from the gross income derived from sales of clean grains, based on the prevailing market price of soybeans. The total investment represents the entire amount used to establish the study. The formula for ROI is as follows:

$$ROI (\%) = \frac{\text{Net Income}}{\text{Total Investment}} \times 100$$

The growth and yield data collected from the study were subjected to the analysis of variance (ANOVA) using F-test for split-plot design and the results were interpreted at 1 and 5% levels of significance. Significant differences between and among the factors studied was determined using Least Significant Difference (LSD test).

Results and Discussion

Growth Components of Soybean Crop

Plant Height (cm)

Varietal performance. The mean height of soybean plants at harvest, as influenced by the different types of carrageenan applied, is presented in Table 1. It is evident that the varieties significantly affected this parameter.

Among the three varieties, PSB Sy-6 exhibited the tallest plants with a mean height of 167.60 cm, followed by the Manchuria variety with a mean height of 125.40 cm. These varieties significantly differed from the shortest variety, IPB 96-27-23, which had a mean height of 66.03 cm. This result contrasts with DA-WESVIARC (2017), which reported heights ranging from 38.77 - 44.77 cm for these three varieties, indicating they were relatively shorter than those in the present study. According to DA-WESVIARC (2013), soybean varieties vary in their seasonal adaptability and reaction to climatic conditions. Short-day length plants, in particular, exhibit excessive vegetative growth during longer day lengths, a characteristic attributed to the genetic makeup of each variety.

Effect of different kinds of carrageenan. The analysis of variance for this growth component revealed no significant difference among the various kinds of carrageenan applied, with mean heights ranging 127.11 (commercial granular fertilizer and radiated carrageenan) - 130.55 cm (commercial granular fertilizer). This suggests that regardless of the type of carrageenan used, whether supplemented or not with the recommended rate of commercial granular fertilizer, soybean plants exhibited comparable heights. These findings align with the observations made by Rathorea et al. (2009), who reported that soybean was not significantly affected by foliar applications of seaweed extract up to a 5% concentration. However, in contrast, higher concentrations did significantly impact plant height. Interestingly, this result diverges from the study by Gil (2019), which found that the use of radiated carrageenan in rice resulted in thicker and taller plants.

Interaction effect. Analysis of variance for this parameter shows no significant interaction effect between variety and kinds of carrageenan applied. This implies that the effects of different varieties of soybean on plant height was not dependent on different kinds of carrageenan applied.

Fresh Biomass (t ha⁻¹)

Varietal performance. The mean fresh biomass of the soybean varieties did not significantly differ among the varieties (Table 2). The data indicate that the IPB 96-27-23 variety had the highest mean fresh biomass at 11.60 t ha⁻¹, compared with PSB Sy-6 and Manchuria, both of which had a mean fresh biomass of 10.53 t ha⁻¹. Despite these differences, the fresh biomass of the three varieties was statistically comparable.

Effect of different kinds of carrageenan. The type of carrageenan applied to soybean significantly influenced the fresh biomass (Table 2). Data reveal that commercial granular fertilizer (CGF) application combined with either commercial radiated carrageenan (CRC) or radiated carrageenan (RC) yielded the highest fresh biomass weights with means

Table 1. Mean plant height (cm) of soybean varieties applied with commercial granular fertilizer and kinds of carrageenan.

Soybean Varieties	Kinds of Carrageenan and Commercial Granular Fertilizer					Mean*
	CGF	CGF +CRC	CGF + RC	RC	CGF + FSE	
IPB 96-27-23	92.33	94.00	93.33	91.00	92.67	92.67c
PSB Sy-6	175.00	169.67	161.33	170.00	162.00	167.60a
Manchuria	124.33	127.67	126.67	123.33	125.00	125.40b
Mean ^{ns}	130.55	130.45	127.11	128.11	126.56	128.56

cv (a) = 10.25%; cv (b) = 7.54 %; * = significant at 5% level; ns = not significant; means followed by a common letter are not significantly different at the 5% level by LSD. Institute of Plant Breeding (IPB), Philippine Seed Board Soybean (PSB Sy), commercial granular fertilizer (CGF), radiated carrageenan (RC), commercial radiated carrageenan (CRC), fermented seaweed extract (FSE).

Table 2. Mean fresh biomass (t ha^{-1}) of different soybean varieties applied with commercial granular fertilizer and kinds of carrageenan.

Soybean Varieties	Kinds of Carrageenan and Commercial Granular Fertilizer					Mean ^{ns}
	CGF	CGF + CRC	CGF + RC	RC	CGF + FSE	
IPB-96-27-23	11.33	13.77	14.00	7.77	11.57	11.69
PSB Sy-6	9.77	12.67	12.00	7.33	10.90	10.53
Manchuria	9.77	12.00	12.23	7.77	10.90	10.53
Mean*	10.29 ^b	12.81 ^a	12.74 ^a	7.62 ^c	11.12 ^b	10.92

cv (a) = 10.41%; cv (b) = 8.41 %; * = significant at 5% level; ns = not significant; means followed by a common letter are not significantly different at the 5% level by LSD. Institute of Plant Breeding (IPB), Philippine Seed Board Soybean (PSB Sy), commercial granular fertilizer (CGF), radiated carrageenan (RC), commercial radiated carrageenan (CRC), fermented seaweed extract (FSE).

of 12.81 t ha^{-1} and 12.74 t ha^{-1} , respectively. This was followed by soybeans treated with CGF plus fermented seaweed extracts (FSE) (11.12 t ha^{-1}) and CGF alone (10.29 t ha^{-1}), both of which had comparable fresh biomass weights. In contrast, soybeans treated with RC alone had the lowest fresh biomass with a mean of 7.62 t ha^{-1} .

These findings indicate that the application of carrageenan can influence soybean fresh biomass, supporting Gil's (2019)'s statement that carrageenan develops substances that can improve the overall growth and development of plants. Additionally, it confirms Naeem et al.'s (2015) claim that the application of gamma rays irradiated carrageenan to *Catharanthus roseus* (L.) improved its performance, enhancing leaf yield by 29.2% and 35.4% and herbage yield by 32.5% and 37.4%. The lower biomass observed with RC alone can be attributed to the absence of additional nutrient supplements, as it was applied without CGF.

Interaction effect. The analysis of variance data indicates that there was no significant interaction effect between the two factors (soybean varieties and types of carrageenan applied) on fresh biomass. This suggests that these two factors independently influenced the fresh biomass of soybean plants.

Number of Nodules Per Plant

Varietal performance. Table 3 presents the nodules per plant for different soybean varieties. Data indicate that the Manchuria variety produced the highest number of nodules per plant with a mean of 73. This was followed by the PSB Sy-6, which had a mean of 71, and the IPB 96-27-23, with the lowest number of nodules per plant at 63. However, the data also reveal that the number of nodules produced by the three varieties is comparable, suggesting that they produce a similar quantity of nodules.

Effect of different kinds of carrageenan. The application of different kinds of carrageenan to soybean did not significantly influence the number of nodules per plant (Table 3). The highest nodule count per plant was observed with the use of RC, which had a mean of 71. Following this, CGF combined with

CRC had a mean of 70 while both CGF and CGF plus RC had the same mean of 69. In contrast, the lowest number of nodules per plant (mean of 67) was produced by the combination of CGF and fermented seaweed extract (FSE).

Interestingly, regardless of the type of carrageenan applied, with or without inorganic fertilizer, the number of nodules remained comparable. However, this finding contradicts the results reported by Tandon and Dubey (2015), who found that soybeans treated with Biozyme crop-plus spray at 500 mL ha^{-1} (a seaweed-based stimulant) along with half of the recommended NPK produced fewer nodules, averaging 35.086 nodules per plant.

Interaction effect. Result shows that there was no relationship observed between different kinds of carrageenan and variety in the production of nodules per plant of soybean. This implies that the two factors acted independently from each other on their effect on the number of nodules per plant of soybean.

1,000 Seed-weight (g)

Varietal performance. Table 4 presents the mean 1,000 seed weight of soybean plants at harvest, influenced by different varieties. The use of various varieties significantly affected the weight of 1,000 seeds. Comparing the 1,000 seed weights across varieties, IPB 96-27-23 produced the heaviest seeds with a mean of 148.20 g, significantly different from Manchuria's seed weight (mean of 113.60 g). Additionally, the 1,000 seed weight of PSB Sy-6 (104.20 g) is comparable with that of Manchuria.

In a study by DA-WESVIARC (2013), IPB 96-27-23 consistently exhibited the heaviest 1,000 seed weight at 133.33 g. However, results also showed that PSB Sy-6 had a 1,000 seed weight of 130 g, heavier than Manchuria's 123.33 g.

On the other hand, Magsino et al. (2016) obtained comparable results, with the Manchuria variety producing a heavier 1,000 seed weight (mean of 114.9 g) compared with PSB Sy-6 (mean of 106.8 g). These findings suggest that IPB 96-27-23 is a promising variety for local cultivation due to its larger seeds.

Table 3. Mean number of nodules per plant of different soybean varieties applied with commercial granular fertilizer and carrageenan.

Soybean Varieties	Kinds of Carrageenan and Commercial Granular Fertilizer					Mean ^{ns}
	CGF	CGF + CRC	CGF + RC	RC	CGF + FSE	
IPB-96-27-23	64	64	64	63	61	63
PSB Sy- 6	67	69	70	78	70	71
Manchuria	76	76	73	71	71	73
Mean ^{ns}	69	70	69	71	67	69

cv (a) = 19.20% ; cv (b) = 6.12 % ; * = significant at 5% level; ns = not significant; means followed by a common letter are not significantly different at the 5% level by LSD. Institute of Plant Breeding (IPB), Philippine Seed Board Soybean (PSB Sy), commercial granular fertilizer (CGF), radiated carrageenan (RC), commercial radiated carrageenan (CRC), fermented seaweed extract (FSE).

Table 4. Mean 1,000 seed weight (g) of different soybean varieties applied with commercial granular fertilizer and carrageenan.

Soybean Varieties	Kinds of Carrageenan and Commercial Granular Fertilizer					Mean*
	CGF	CGF + CRC	CGF + RC	RC	CGF + FSE	
IPB 96-27-23	140.00	161.33	161.00	129.00	149.67	148.20a
PSB Sy- 6	96.33	116.33	115.33	86.00	107.00	104.20b
Manchuria	113.67	124.33	125.00	97.00	108.00	113.60b
MEAN*	116.67c	134.00a	133.78a	104.00d	121.56b	122.00

cv (a) = 10.05%; cv (b) = 3.53 %; * = significant at 5% level; ns = not significant; means followed by a common letter are not significantly different at the 5% level by LSD. Institute of Plant Breeding (IPB), Philippine Seed Board Soybean (PSB SY), commercial granular fertilizer (CGF), radiated carrageenan (RC), commercial radiated carrageenan (CRC), fermented seaweed extract (FSE).

In Bakal et al.'s study (2017), soybean seeds reached a mean weight of 145.8 g during double cropping seasons. Shanmugasundaram (1991) also reported that promising soybean varieties could achieve at least 72 g of 1,000 seed weight, while other varieties had a maximum of only 91 g. The observed variations in values and trends may be attributed to differences in agro-climatic conditions, soil fertility status, fertilization methods, and nutrient uptake capabilities.

Effect of different kinds of carrageenan. The mean 1,000 seed weight was influenced by the application of different kinds of carrageenan. The analysis of variance reveals that soybeans treated with CGF combined with CRC had the highest mean 1,000 seed weight at 134 g. Additionally, soybean plants treated with CGF plus RC exhibited a comparable mean of 133.78 g. Conversely, a lighter 1,000 seed weight was observed when CGF was combined with FSE, resulting in a mean of 121.56 g. The lowest 1,000 seed weight (mean of 116.67 g) occurred in plants treated with CGF alone. Notably, the lightest 1,000 seed-weight (mean of 104 g) was from soybeans treated solely with RC.

These findings align with Rathorea et al.'s (2009) study, which demonstrated that soybeans could produce a 1,000 seed-weight of 146.48 g when treated with 15% *K alvarezii* seaweed extract. Rathorea further elaborated that the extract enhanced all growth parameters measured for soybeans compared to no application. Similarly, Nicavera (2021) emphasized that carrageenan sources like seaweeds effectively increase rice yield in Western

Visayas, especially when irradiated or broken down into shorter fragments for better plant absorption. Ahmad et al. (2019) also cited that gamma-irradiated carrageenan outperforms the un-irradiated form due to reduced molecular weight and specific structural changes. This is evident in our results, where RC-treated plots and CRC recorded heavier 1,000 seed weights than those treated with FSE.

Interaction effect. There was no significant interaction effect observed between soybean varieties and the different types of carrageenan applied. This indicates that soybean varieties and carrageenan types acted independently in their effects on soybeans, specifically regarding 1,000 seed weight.

Dry Grain Yield (t ha⁻¹)

Varietal performance. The dry grain yield significantly varies among the different soybean varieties used. IPB 96-27-23 obtained the highest yield at 2.91 t ha⁻¹, significantly different from the yield of PSB SY 6 (2.44 t ha⁻¹), which was comparable with Manchuria (mean dry grain yield of 2.41 t ha⁻¹) (Table 5). These variations suggest that each variety possesses distinct varietal characteristics, contributing to the significant differences in dry grain yield. These findings align with the results reported by DOST-PCAARRD (2002), which indicated that planting PSB Sy-6 can yield between 1.0 and 4.1 t ha⁻¹. Notably, the two new soybean varieties—IPB 96-27-23 (with a much higher yield) and Manchuria—could potentially match the yield of PSB Sy-6.

In contrast, DA-WESVIARC's trial (2017) found that IPB 96-27-23 produced the lowest yield (mean

Table 5. Mean dry grain yield (t ha^{-1}) of different soybean varieties applied with commercial granular fertilizer and carrageenan.

Soybean Varieties	Kinds of Carrageenan and Commercial Granular Fertilizer					Mean*
	CGF	CGF + CRC	CGF + RC	RC	CGF + FSE	
IPB 96-27-23	2.90	3.23	3.30	2.10	3.03	2.91a
PSB Sy- 6	2.50	2.83	2.90	1.53	2.43	2.44b
Manchuria	2.17	2.9	2.87	1.70	2.40	2.41b
Mean*	2.52b	2.99a	3.02a	1.78c	2.62b	2.59

cv (a) = 9.34%; cv (b) = 5.40 % ; * = significant at 5% level; ns = not significant; means followed by a common letter are not significantly different at the 5% level by LSD. Institute of Plant Breeding (IPB), Philippine Seed Board Soybean (PSB Sy) commercial granular fertilizer (CGF), radiated carrageenan (RC), commercial radiated carrageenan (CRC), fermented seaweed extract (FSE).

of 1.75 t ha^{-1}), followed by PSB Sy-6 (mean yield of 2.12 t ha^{-1}). Manchuria exhibited the highest yield (mean of 2.5 t ha^{-1}). Similarly, Magsino et al. (2016) reported that PSB Sy-6 had a higher mean yield of 2.0 t ha^{-1} compared with Manchuria (mean yield of 1.2 t ha^{-1}). The observed variations in yield may be attributed to differences in agro-climatic conditions, soil fertility status, fertilization methods, and nutrient uptake capabilities

Effect of different kinds of carrageenan. The mean dry grain yield (t ha^{-1}) was influenced by the application of different types of carrageenan. Soybeans treated with CGF plus RC yielded the highest (3.02 t ha^{-1}) and were comparable with those treated with CGF plus CRC (mean of 2.99 t ha^{-1}). Soybeans treated with CGF plus fermented seaweed extract (FSE) (2.59 t ha^{-1}) and those treated with CGF alone (2.52 t ha^{-1}) exhibited similar yields.

In contrast, the lowest yield was observed in soybeans treated solely with RC, yielding a mean of 1.78 t ha^{-1} . These results differ from Rathorea et al.'s (2009) findings, where the highest grain yield occurred with 15% seaweed extract application, followed by 12.5% seaweed extract (resulting in 57% and 46% increases, respectively, compared with the control with no application). Our study further indicates that soybean yield improves when treated with a combination of commercial granular fertilizer and various forms of carrageenan, whether radiated or fresh seaweed extract. Notably, the highest yields

were obtained with the combination of CGF and RC, regardless of whether it was in commercial form or not.

These findings highlight the effectiveness of carrageenan sources in increasing yield, particularly when irradiated and broken down into shorter fragments for better plant absorption, as suggested by Gil (2019). Ahmad et al. (2019) also emphasized that gamma-irradiated carrageenan outperforms the un-irradiated form due to reduced molecular weight and specific structural changes. Similarly, our results align with Gatan's (2020) report, which demonstrated an 88% - 102.9% yield increase in mungbean with varying dosages and frequencies of RC application.

Interaction effect. No significant interaction effect was noted between the two factors. This implies that the two factors acted independently on their effects on dry grain yield of soybean.

Soil Fertility Dynamics

Soil pH

Effect of varieties. Figure 1 shows the results of soil pH analysis in plots where soybean varieties were treated with different types of carrageenan at three intervals: before planting, 45 days after planting, and after harvest. Initially, the soil pH ranged from 5.74 - 5.99, indicating a moderately acidic condition. Notably, during the vegetative stage, soil pH decreased in the three varieties: PSB Sy-6 (4.82), Manchuria (5.06), and IPB 6-27-23 (5.00). However,

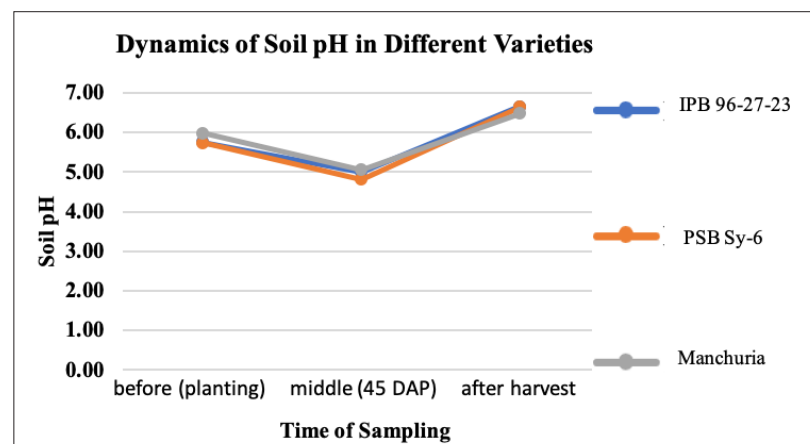


Figure 1. Dynamics of soil pH in plots with soybean varieties treated using carrageenan types and commercial granular fertilizer combinations.

as the soybeans progressed toward harvest, soil pH exhibited an increasing trend across all three varieties ranging from 6.48 - 6.66.

The acidic nature of the soil at the study's outset aligns with Tang et al.'s (2002) observation that nitrate leaching through soil profiles contributes significantly to soil acidification. Legumes, including soybeans, have the capacity to take up nitrate (NO_3^-), and the extent to which soil NO_3^- affects legume nodulation and NO_2 fixation is evident, especially considering the research area's prior soybean cultivation. A decline in soil pH incurred during the vegetative stage. Lofton and Arnall (2017) proposed that leguminous plants, like soybeans, reach their maximum biomass levels during full bloom, coinciding with high nodulation activity. As the crop accumulates biomass, nutrients are taken up, leading to the release of hydrogen ions by soybean roots—a process that tends to acidify the rhizosphere.

Furthermore, rhizobia activity during nodulation enhances nitrogen availability, which, according to Chen et al. (2023), contributes to a significant decrease in soil pH due to nitrogen addition.

Additionally, during the reproductive stage of soybeans, soil microbial activity—especially rhizobia—increases, correlating with soil pH changes, as noted by Slattery et al. (2001). As soybean plants accumulate biomass and microbial activity around 45 DAP, all soybean varieties exhibit decreasing soil pH from planting until the reproductive stage, followed by an upward trend leading up to harvest.

Effect of different kinds of carrageenan and commercial granular fertilizer. Figure 2 shows the results of soil pH applying different kinds of carrageenan plus commercial granular fertilizer to soybean crop. At the start of planting soil pH ranged from 5.61 (CGF plus RC) - 6.01 (CGF) alone. During vegetative stage, there was decreasing trend seen in soil pH of all plots applied with CGF plus RC (5.10), CGF alone (4.97), RC (4.96), CGF plus RC (4.96), CGF (4.93), and CGF plus FSE (4.87). From the onset of the reproductive stage up to maturity, soil

pH value went up in all treatment plots. This result agrees with a previous result, which was explained in Figure 2 that during vegetative stage of soybean, soil pH decrease was affected by hydrogen ion accumulation as a result of previous crop planted and biomass accumulation. Thus, this further magnified the acidity of soil because treatments were applied from the start of vegetative stage. The increase of soil pH during reproductive stage up to harvest conforms to Tayangona et al. (2017) that soil pH was affected by foliar application of irradiated carrageenan at the end of a 3-month period applied in banana. This only means that radiated carrageenan or seaweed-based bio stimulants may increase soil pH when applied to soybean crop same with banana from the reproductive stage onwards (Tayangona et al., 2017).

Soil Organic Matter (%)

Effect of varieties. Figure 3 presents the results of soil organic matter (%) in plots where soybean varieties were treated with different types of carrageenan at three intervals: before planting, 45 DAT, and after harvest. Overall, soil organic matter exhibited an increasing trend across all varieties during the vegetative stage, with the Manchuria variety showing the highest increase (0.18%). However, as the reproductive stage commenced, soil organic matter declined in all soybean varieties.

The initial increase in soil organic matter during the first half of the growth period can be attributed to the ongoing decomposition of organic matter residues resulting from field operations, particularly cultivation and other tilling activities. Additionally, this trend may relate to soybean's biological nitrogen fixation process, where atmospheric nitrogen is utilized by plants, leaving residual organic matter unutilized. Consequently, soil organic matter content remains high during the vegetative stage.

The subsequent decline in organic matter is likely due to reduced decaying organisms or an accelerated rate of decay. Decomposition and mineralization become contributing factors during the second half of the growth period.

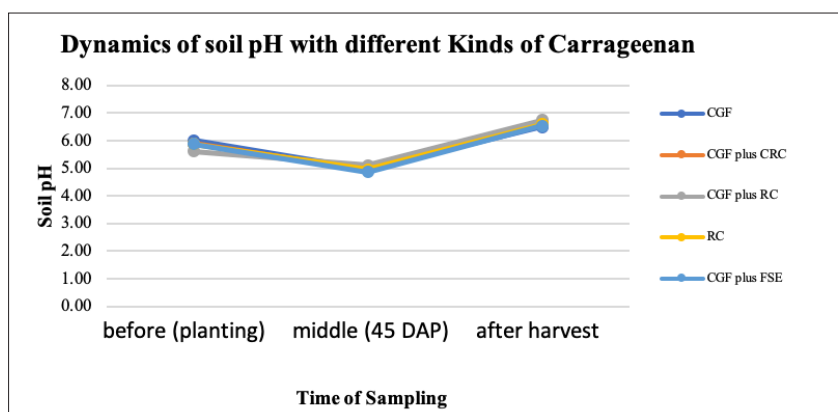


Figure 2. Dynamics of soil pH in plots with carrageenan types and commercial granular fertilizer combinations applied to soybean.

Effect of different kinds of carrageenan and commercial granular fertilizer. Figure 4 illustrates the impact of different types of carrageenan applied to soybeans on soil organic matter. During the vegetative stage, all carrageenan and fertilizer combinations led to an increase in soil organic matter across all plots. Notably, the highest increase occurred in soybeans treated with CGF plus FSE, resulting in a remarkable 177% increase.

However, from the second sampling period until harvest, a consistent trend of decreasing soil organic matter was observed in all plots. This decline during the later growth stages may be attributed to the accumulation of soil microbial populations in treatment plots where seaweed and carrageenan extracts were applied. These microbial activities likely contributed to the reduction in available soil organic matter. In summary, the initial increase in soil organic matter during the vegetative stage could be linked to the decomposition of organic matter following cultivation. Conversely, the subsequent decrease during the reproductive stage is likely due to mineralization processes.

Available Soil Phosphorus (ppm)

Effect of varieties. Figure 5 depicts the trend of available soil phosphorus (ppm) in plots where soybean varieties were treated with different types of carrageenan at three intervals: before planting, 45 DAP, and after harvest. The three soybean varieties exhibited a similar pattern: P levels decreased as the crop reached the reproductive stage but eventually increased at maturity.

These results suggest that, across all three varieties, soybeans generally take up more phosphorus during the vegetative stage than during the reproductive stage. Specifically, PSB Sy-6 exhibited lower P uptake than IPB 96-27-23 and Manchuria. This phenomenon aligns with findings by Bender et al. (2015), who observed that nutrient accumulation in soybeans occurs in three distinct phases: a slow acquisition rate during the initial 30 DAP, maximum nutrient uptake between R2 (full bloom) and R5 (beginning seed), and reduced nutrient accumulation during late reproductive growth (seed maturation). Notably, P uptake in soybeans is evenly distributed between vegetative and seed-filling growth phases. Comparing the accumulated phosphorus among the three varieties, IPB 96-27-23 accumulated more P than the other two varieties, resulting in a heavier 1,000 seed weight (Table 4) and higher dry grain yield (Table 5).

Effect of different kinds of carrageenan and commercial granular fertilizer. Various combinations of carrageenan and fertilizer applied to soybean exhibited different trends of P (Figure 6). Plots treated with RC alone showed a decreasing trend in P levels from the vegetative stage to the onset of the reproductive stage, continuing to decrease until harvest. In contrast, plots treated with CGF, CGF plus CRC, CGF plus RC, and CGF plus FSE showed an initial decrease in P during the vegetative stage, followed by an increase from the reproductive stage to harvest.

This suggests that the P required by soybean during its growth period (vegetative to seed formation

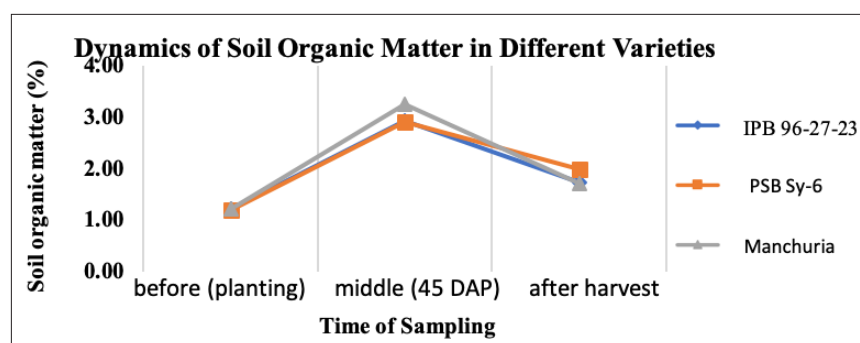


Figure 3. Dynamics of soil organic matter (%) in plots with soybean varieties treated using various carrageenan types and commercial granular fertilizer combinations.

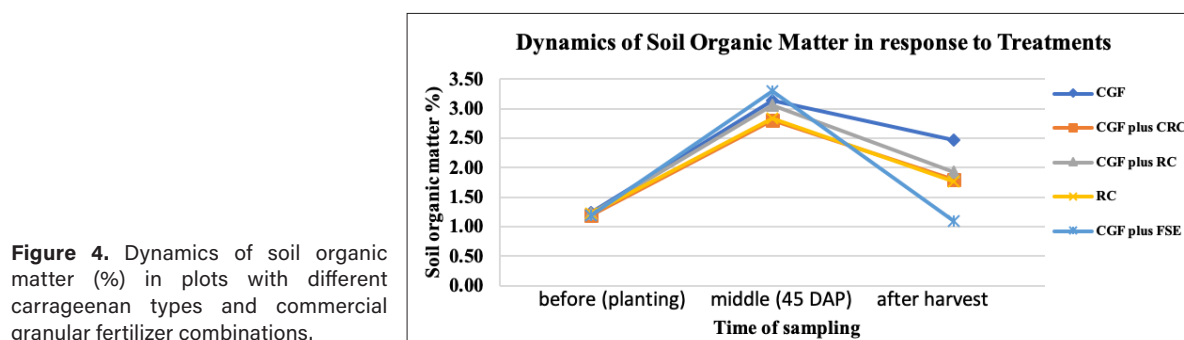


Figure 4. Dynamics of soil organic matter (%) in plots with different carrageenan types and commercial granular fertilizer combinations.

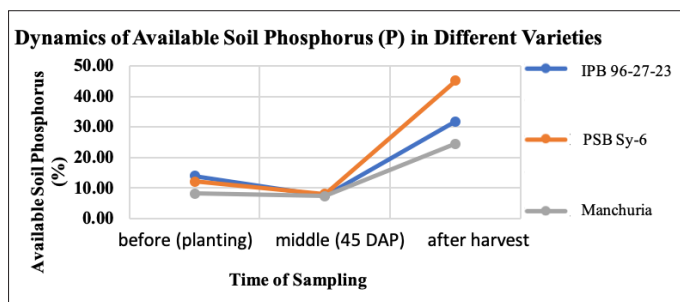
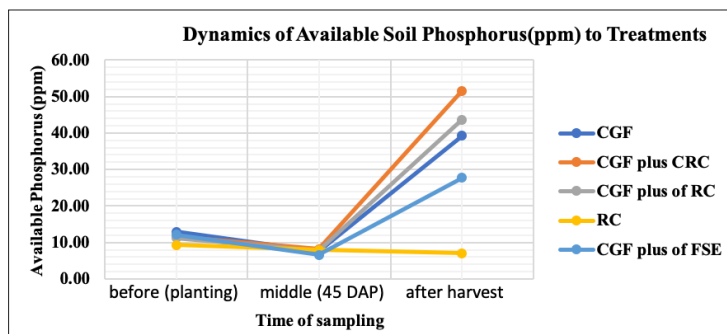


Figure 5. Dynamics of available soil P in plots with soybean varieties treated using carrageenan types and commercial granular fertilizer combinations.

Figure 6. Dynamics of available soil P in plots with soybean varieties treated using carrageenan types and commercial granular fertilizer combinations.



stages) was adequately supplied by CGF, as indicated by the chemical analysis of the soil. The remaining P was utilized as the soybean matured, minimizing nutrient uptake. The observed behavior of P is related to the chemical composition of the applied treatments. The chemical analysis showed that FSE contains 117 ppm of P, CRC contains 27 ppm, and RC contains 17 ppm. These supplements to CGF likely increased the soil P content, explaining the observed results during the reproductive stage to harvest.

Bongalos et al. (2019) noted that seaweed and seaweed extracts activate beneficial microorganisms, releasing soil-conditioning substances that transform unavailable P into forms readily absorbed by plants. Ahmad et al. (2019) further stated that gamma-irradiated carrageenan is more effective than the non-irradiated form due to its reduced molecular weight and structural changes, which could account for the increased P availability in plots treated with RC and CRC compared to FSE during the reproductive stage to harvest. However, plots treated with RC alone showed a decrease in P, likely due to insufficient P supply without the supplementation of CGF.

Exchangeable Soil Potassium (ppm)

Effect of varieties. Figure 7 presents the K dynamics in plots grown with different soybean varieties applied with irradiated carrageenan. All plots planted with soybean varieties IPB 96-27-23, PSB Sy-6, and Manchuria exhibited a similar pattern: K levels rapidly decreased from the start of the experiment until the vegetative stage, then slightly increased from the onset of the reproductive stage

to harvest. Cheruiyot (2020) explained that in most plants, K uptake accelerates during the vegetative phase, peaks in nutrient accumulation in the foliage, and decreases in the late stage.

Effect of different kinds of carrageenan and commercial granular fertilizer. The K dynamics in plots applied with various carrageenan treatments were similar to those observed in the variety plots (Figure 8). K levels rapidly decreased from the start of the experiment until the reproductive stage, then slightly increased up to harvest. This indicates that K consumption is higher from the early vegetative stage to the late vegetative stage, suggesting that K fertilizer should be applied early, and slow-release fertilizers should be applied at the basal stage. The data also show that the next crop could benefit from the remaining K in the soil, eliminating the need for additional K application in the next cropping season.

The behavior of K is also related to the chemical analysis of the applied treatments. FSE contains 1,041 ppm of K, CRC contains 497 ppm, and RC contains 322 ppm, contributing to the soil K levels from the reproductive stage to harvest. The availability of K in plots treated with CGF plus CRC, RC, and FSE was lower than in plots treated with CGF alone, likely because the K from foliar applications of carrageenan and seaweed extracts was more readily absorbed by plants than the K from CGF, resulting in more K retention in the soil. However, plots treated with RC alone showed a continuous decrease in K from start to harvest, possibly due to a K deficiency without supplementation.

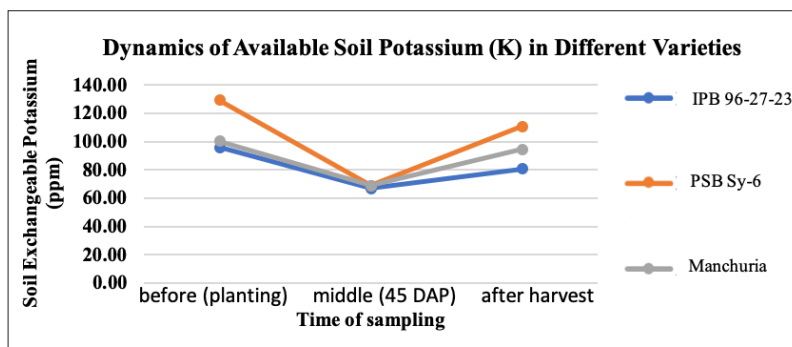
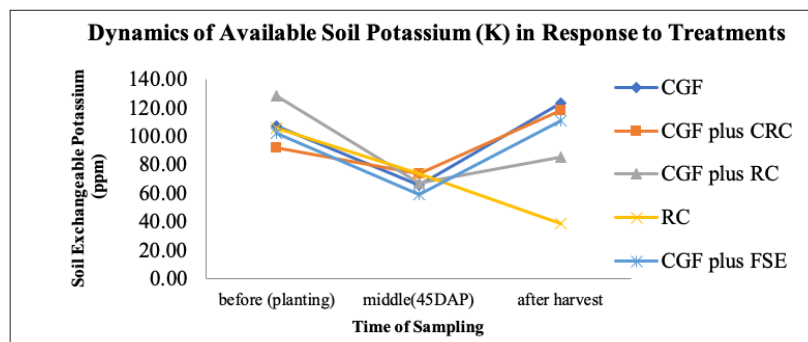


Figure 7. Dynamics of exchangeable soil potassium (ppm) in plots with soybean varieties treated using various carrageenan types and commercial granular fertilizer combinations.

Figure 8. Dynamics of exchangeable soil potassium (ppm) in plots with soybean varieties treated using carrageenan types and commercial granular fertilizer combinations.



Dynamics of Insect Pest Associated with Soybean Varieties as Influenced by Kinds of Carrageenan and Commercial Granular Fertilizer

IPB 96-27-23

Table 6 shows the average insect pest incidence (%) per treatment for various insect pests found in IPB 96-27-23, influenced by different kinds of carrageenan and commercial granular fertilizer combinations. Five insect pest species were associated with this variety: bean leafroller (larva) (*Urbanus proteus* L.), leaf miner (*Odontota horni*), Dectes stem borer (*Dectes texus*), and cutworm (*Spodoptera litura*). All insect pest damage occurred during the vegetative stage and decreased up to the reproductive stage. Higher damage was observed from bean leafroller (larva) in plots applied with commercial granular fertilizer plus fermented seaweed extract. Notable damage from leaf miner was observed in plots applied with CGF plus CRC, but in a small percentage. Stem borer only attacked plots applied with CGF plus FSE, and cutworm attacked only plots applied with CGF.

PSB SY-6

Table 7 shows the average insect pest incidence (%) per treatment of various insect pests found in PSB Sy-6, influenced by different carrageenan and fertilizer combinations. Four insect species were associated with this variety: bean leafroller (*Urbanus proteus* L.), leaf miner (*Odontota horni*), Dectes stem borer (*Dectes texus*), and tussock moth (*Orgyia leucostigma*).

Insect pest damage was observed during the vegetative stage, except for stink bug damage, which occurred during the reproductive stage. The highest damage was caused by the bean leafroller (larva) in plots treated with CGF plus FSE. Bean leafroller damage was present in all treatments, while leaf miner damage was seen in all plots except those treated with RC alone. Stem borer only attacked plots treated with CGF plus RC and CGF alone, and tussock moth only attacked plots treated with CGF plus RC, though incidence was minimal.

Manchuria

Table 8 shows the average insect pest incidence (%) per treatment of various insect pests found in Manchuria, influenced by different carrageenan and fertilizer combinations. Five insect species were associated with this variety: bean leafroller (*Urbanus proteus* L.), leaf miner (*Odontota horni*), Dectes stem borer (*Dectes texus*), stink bug (*Pentatomidae*), and tussock moth.

As in the previous variety, insect pest damage occurred during the vegetative stage, except for stink bug damage, which occurred during the reproductive stage. The highest damage was caused by the stink bug in plots treated with CGF plus CRC. Bean leafroller damage was seen in all treatments, while stink bug damage was observed in all plots except those treated with CGF alone. Leaf miner damage was noted in plots treated with CGF, CGF plus FSE, and RC. Stem borer only attacked plots treated with CGF and CGF plus RC, while tussock moth only attacked plots treated with CGF plus RC and RC alone.

Table 6. Insect pest incidence (%) in plots planted with IPB 96-27-23 as influenced by kinds of carrageenan and commercial granular fertilizer combination.

Classification	Average Insect Pest Incidence (%)					Mode of F Feeding	Crop Stages
	CGF	CGF +CRC	CGF + RC	RC	CGF + FSE		
Bean leafroller (<i>Urbanus proteus</i> L.)	4.0	3.0	5.7	0.6	0.9	Defoliator	Vegetative stage
Leaf miner (<i>Odontotahorni</i>)	0.0	1.9	0.1	0.0	0.0	Mining	Vegetative to flowering stage
Dectes stem borer (<i>Dectes texus</i>)	0.0	0.0	0.0	0.0	1.4	Borer	Vegetative to reproductive stage
Cutworm (<i>Spodoptera litura</i>)	1.3	0.0	0.0	0.0	0.0	Defoliator	Vvegetative stage

Note: Data was collected through physical examination of soybean crop conducted every 2 weeks starting the fourth week after planting.

Table 7. Insect pest incidence (%) in plots planted with PSB Sy-6 as influenced by kinds of carrageenan and commercial granular fertilizer combination.

Classification	Average Insect Pest Incidence (%)					Mode of Feeding	Crop Stages
	CGF	CGF+ CRC	CGF+ RC	RC	CGF+ FSE		
Bean leafroller (<i>Urbanus proteus</i> L.)	2.0	1.4	0.9	1.0	3.3	Defoliator	Vegetative to flowering stage
Leaf miner (<i>Odontota horni</i>)	1.2	11.3	0.2	0.0	1.3	Mining	Vegetative stage
Dectes stem borer (<i>Dectes texus</i>)	0.3	0.0	0.6	0.0	0.0	Borer	Vegetative to reproductive stage
Tussock moth (<i>Orygialeuco stigma</i>)	0.0	0.0	0.3	0.0	0.0	Defoliator	Vegetative stage

Note: Data was collected through physical examination of soybean crop conducted every 2 weeks starting the fourth week after planting.

Table 8. Insect pest incidence (%) in plots planted with Manchuria as influenced by kinds of carrageenan and commercial granular fertilizer combination.

Classification	Average Insect Pest Incidence (%)					Mode of Feeding	Crop Stages
	CGF	CGF + CRC	CGF + RC	RC	CGF + FSE		
Bean leafroller (<i>Urbanus proteus</i> L.)	0.6	2.1	3.2	0.7	1.9	Defoliator	Vegetative to flowering stage
Leaf miner (<i>Odontota horni</i>)	0..3	0.0	0.0	0.3	0.3	Defoliator	Vegetative stage
Stink bug (green and brown) (<i>Halyomorpha</i> and <i>Pentatomidae</i>)	0.0	3.5	2.8	2.0	2.5	Plant sucker	Reproductive stage
Dectes stem borer (<i>Dectes texus</i>)	0.3	0.0	0.6	0.0	0.0	Borer	Vegetative to reproductive stage
Tussock moth (<i>Orygialeuco stigma</i>)	0.0	0.0	0.5	0.3	0.0	Defoliator	Vegetative stage

Note: Data was collected through physical examination of soybean crop conducted every 2 weeks starting the 4th week after planting.

Insect Pest Dynamics

Insect pests in the experimental area were monitored every two weeks by collecting them with insect-catching nets and handpicking. Collected insects were brought to the laboratory for sorting and identification. Figure 9 shows that the highest total number of insects collected was the green stink bug (*Pentatomidae*), seen prominently during the 8th to 10th week of collection or from the start of the reproductive stage. The next highest total number was bean leafroller, prominently seen from the 4th week, with a slight decrease during the 6th week. Leaf miner was observed only during the 6th and 8th weeks of collection. Other notable insect pests collected include Tussock moth, seen during the 4th week (vegetative stage), and a variety of other pests

in smaller numbers. Major pests with significant numbers included stink bugs (green and brown), leaf miner, and bean leaf roller, as cited by DOST-PCAARRD (2002).

Disease Incidence

Effect of Varieties. Table 9 shows the disease incidence (%) in plots planted with soybean varieties applied with different kinds of carrageenan and CGF combinations. Diseases affecting soybean included leaf blight, root and stem rot, *Cercospora* leaf spot, and powdery mildew. The highest leaf blight incidence was in IPB 96-27-23 (9.40%), followed by Manchuria (1.37%), with the lowest in PSB Sy-6 (0.86%). Root and stem rot incidence was similar across varieties, ranging from 1.86% - 1.04%. PSB Sy-6 had the

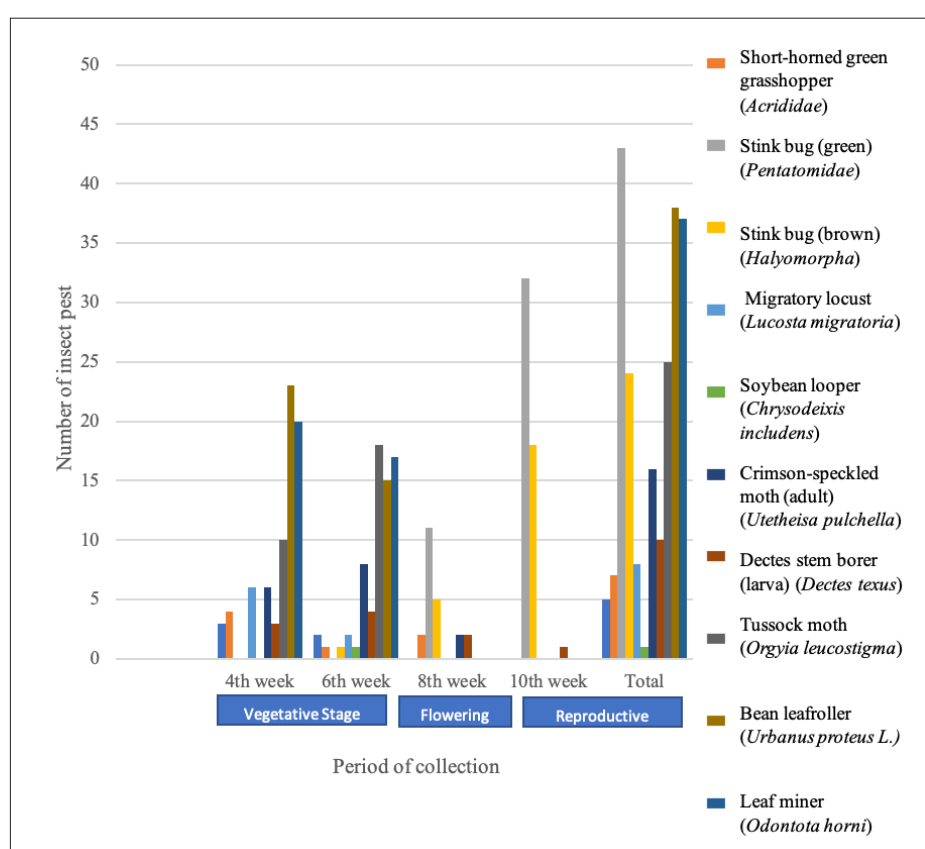


Figure 9. Number of insect pests gathered in the research area.

Table 9. Disease incidence (%) in plots planted with soybean varieties applied with different kinds of carrageenan and commercial granular fertilizer combination.

Soybean Varieties	Diseases and Causal Organism			
	Bacterial blight (<i>Pseudomonas syringae</i>) pv. <i>Glycinea</i>	Root and stem rot (<i>Phytophthora sojae</i>)	<i>Cercospora</i> leaf spot (<i>Cercosporakichii</i>)	Powdery mildew (<i>Microsphaeradiffusa</i>)
IPB-96-27-23	9.40	1.86	0.00	0.20
PSB Sy-6	0.86	1.42	3.68	1.99
Manchuria	1.37	1.04	0.81	0.81

Note: Institute of Plant Breeding (IPB), Philippine Seed Board Soybean (PSB Sy)

highest incidence of *Cercospora* leaf spot (3.68%), followed by Manchuria (0.81%), with no incidence in IPB 96-27-23. For powdery mildew, IPB 96-27-23 had the lowest incidence (0.20%), Manchuria had 0.81%, and PSB Sy-6 had the highest (1.99%). This indicates that IPB 96-27-23 is more susceptible to leaf blight and root and stem rot, while PSB Sy-6 is more susceptible to powdery mildew and *Cercospora* leaf spot.

Effect of different kinds of carrageenan and commercial granular fertilizer. Table 10 shows the incidence of four diseases affecting soybean with different carrageenan and CGF treatments. The highest leaf blight incidence was in soybean applied with CGF (3.66%), and the lowest was with CGF plus CRC. For root and stem rot, the lowest incidence was with CGF plus FSE, and the highest was with CGF only (1.64%). CGF-only treatment had the highest incidence of *Cercospora* leaf spot (1.16%), while the lowest was with CGF plus RC (0.80%). For powdery mildew, the highest incidence was with CGF plus FSE (1.78%), and the lowest was with CGF plus irradiated

carrageenan (RC) (0.13%). This suggests that while CGF alone results in higher disease incidence, combining it with FSE or RC reduces disease incidence, supporting findings by Jayaraman et al. (2011) and Duarte et al. (2018) regarding the benefits of seaweed extracts in enhancing disease resistance.

Economics of Production

Gross income (PhP ha⁻¹). Table 11 presents the gross income (PhP ha⁻¹) from soybean applied with different kinds of carrageenan on a hectare basis. IPB 96-27-23 with CGF plus radiated carrageenan (RC) achieved the highest gross income (PhP 132,000.00 ha⁻¹). The lowest gross income was from PSB Sy-6 applied with RC only (PhP 60,000.00 ha⁻¹).

Total expenses (PhP ha⁻¹). Table 12 shows the total expenses (PhP ha⁻¹) for soybean production with different carrageenan applications. The production cost for all varieties was PhP45,992.00 ha⁻¹ with CGF plus RC and CGF plus CRC. Radiated carrageenan (RC) alone had the lowest production cost (PhP36,268.00 ha⁻¹).

Table 10. Disease incidence (%) in plots applied with different kinds of carrageenan and commercial granular fertilizer combination.

Kinds of Carrageenan and Fertilizer Combinations	Major Diseases and Causal Organism			
	Bacterial blight (<i>Pseudomonas syringae</i>) pv. <i>Glycinea</i>	Root and stem rot (<i>Phytophthora sojae</i>)	<i>Cercospora</i> leaf spot (<i>Cercosporakichii</i>)	Powdery mildew (<i>Microsphaeradiffusa</i>)
CGF	3.66	1.64	1.16	0.53
CGF plus CRC	1.34	0.11	0.84	0.81
CGF plus RC	2.99	1.43	0.80	0.13
RC	2.02	0.43	0.94	0.65
CGF plus FSE	3.51	0.05	1.00	1.78

Commercial granular fertilizer (CGF), radiated carrageenan (RC), commercial radiated carrageenan (CRC), fermented seaweed extract (FSE).

Table 11. Gross income (PhP ha⁻¹) of growing soybean varieties using different kinds of carrageenan.

Soybean Varieties	Kinds of Carrageenan and Commercial Granular Fertilizer and Granular Fertilizer				
	CGF	CGF + CRC	CGF + RC	RC	CGF + FSE
IPB 96-27-23	116,000.00	128,000.00	132,000.00	84,000.00	12,200.00
PSB Sy-6	100,000.00	112,000.00	116,000.00	60,000.00	97,200.00
Manchuria	88,000.00	116,000.00	112,000.00	68,000.00	96,000.00

Institute of Plant Breeding (IPB), Philippine Seed Board Soybean (PSB Sy), commercial granular fertilizer (CGF), radiated carrageenan (RC), commercial radiated carrageenan (CRC), fermented seaweed extract (FSE).

Table 12. Total expenses (PhP ha⁻¹) of growing soybean varieties using different kinds of carrageenan.

Soybean Varieties	Kinds of Carrageenan and Commercial Granular Fertilizer				
	CGF	CGF + CRC	CGF + RC	RC	CGF + FSE
IPB 96-27-23	39,704.00	45,992.00	45,992.00	36,268.00	44,001.50
PSB Sy-6	39,704.00	45,992.00	45,992.00	36,268.00	44,001.50
Manchuria	39,704.00	45,992.00	45,992.00	36,268.00	44,001.50

Institute of Plant Breeding (IPB), Philippine Seed Board Soybean (PSB Sy), commercial granular fertilizer (CGF), radiated carrageenan (RC), commercial radiated carrageenan (CRC), fermented seaweed extract (FSE).

Net income (PhP ha⁻¹). Table 13 indicates that the highest net income (PhP 96,400.00 ha⁻¹) was from IPB 96-27-23 applied with CGF plus RC, while the lowest (PhP 28,900.00 ha⁻¹) was from PSB Sy-6 with RC only.

Return on investment (%ROI ha⁻¹). The computed return on investment (ROI) is presented in Table 14. It was found that IPB 96-27-23 applied with CGF alone recorded the highest ROI of 211.57%, while the

lowest ROI (79.68%) was obtained from PSB Sy-6 applied with RC only.

This indicates that using IPB 96-27-23 with CGF is the most profitable treatment combination. These findings contradict the claim by Abad et al. (2014) that the use of irradiated carrageenan can increase yield and profitability by about 30% in crops like peanut and mungbean, suggesting that this effect does not extend to soybeans.

Table 13. Net income (PhP ha⁻¹) of growing soybean varieties using different kinds of carrageenan.

Soybean Varieties	Kinds of Carrageenan and Commercial Granular Fertilizer				
	CGF	CGF + CRC	CGF + RC	RC	CGF + FSE
IPB 96-27-23	84,000.00	92,400.00	96,400.00	52,900.00	85,600.00
PSB Sy-6	68,000.00	76,400.00	80,400.00	28,900.00	61,600.00
Manchuria	56,000.00	80,400.00	76,400.00	36,900.00	60,400.00

Institute of Plant Breeding (IPB), Philippine Seed Board Soybean (PSB Sy), Commercial granular fertilizer (CGF), Radiated carrageenan (RC), Commercial radiated carrageenan (CRC), Fermented seaweed extract (FSE).

Table 14. Return on investments ROI (%) of growing soybean varieties using different kinds of carrageenan.

Soybean Varieties	Kinds of Carrageenan and Commercial Granular Fertilizer				
	CGF	CGF + CRC	CGF + RC	RC	CGF + FSE
IPB 96-27-23	211.57	200.90	209.60	145.86	194.54
PSB Sy-6	171.27	166.12	174.81	79.68	140.00
Manchuria	141.04	174.81	166.12	101.74	137.27

Institute of Plant Breeding (IPB), Philippine Seed Board Soybean (PSB Sy), commercial granular fertilizer (CGF), radiated carrageenan (RC), commercial radiated carrageenan (CRC), fermented seaweed extract (FSE).

Conclusion

Different kinds of carrageenan affected various soybean varieties, influencing fresh biomass, number of pods per plant, 1,000-seed weight, and grain yield, but not the number of nodules per plant, number of seeds per plant, and pod length. Agronomic parameters such as plant height, 1000-seed weight, and dry grain yield varied. IPB 96-27-23 performed best with irradiated carrageenan application. There was no significant interaction effect between soybean varieties and different kinds of carrageenan. Disease and pest damage incidence were minimal and did not affect total yield, although irradiated carrageenan slightly increased pest incidence. Varietal effects on disease susceptibility and resistance were observed. Soil pH decreased during the vegetative stage but increased at maturity, and soil organic matter increased during the vegetative stage but declined at harvest. P and K levels decreased during the vegetative stage but increased up to harvest in all plots with irradiated carrageenan, fermented seaweed extract, and CGF combinations. The highest gross and net incomes were from IPB 96-27-23 with CGF plus irradiated carrageenan. The highest ROI was from CGF alone.

Recommendations

It is recommended to plant IPB 96-27-23 for high yield and ROI, even without carrageenan, but with CGF application. In the long term, irradiated carrageenan can be recommended for improving soil fertility and nutrient uptake, along with P and K fertilizers as basal applications for maximum utilization. For lowest pest and disease incidence, irradiated carrageenan or seaweed extract alone is not reliable; varietal selection is crucial. Further studies should explore different rates, preparations, and concentrations of irradiated carrageenan, other seaweed extract products, forms or kinds, and other soybean varieties.

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INSECTICIDAL ACTIVITY OF *ARATILES* (*Muntingia calabura* L.) LEAVES AGAINST THE RICE WEEVIL (*Sitophilus oryzae* L.)

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ABSTRACT

The rice weevil is one of the most serious stored cereal pests in the world. Chemical-based insecticides have been used to eradicate this insect, but this very action is also the main cause of the constantly emerging health and environment risks, leading the interest to natural compounds. *Aratiles* on the other hand, is a fruit-bearing tree known to grow fast and thrive well even in polluted areas. This study evaluated the insecticidal activity of *aratiles* against the adult rice weevil. *Aratiles* leaves were dried, macerated, and processed for extraction. Filter paper discs placed in petri dishes were utilized for the bioassay, in which 20 insects were placed per replicate and corresponding treatments were applied. Solutions of 25, 50, and 75% *aratiles* extract were formulated for the treatment groups, 10 ppm of chemical Malathion was used for the positive control and distilled water for the negative control. Based on 1.5 h of exposure time, the results showed that the highest average value of rice weevil mortality, 76.15%, was obtained utilizing the 75% concentration of *aratiles* extracts. This was followed by 73.05% at 25% concentration and 65.10% at 50% concentration. Conversely, the mortality rate for both the positive and negative controls was 0.03%. These results point to the extract from *aratiles*' potential as an insecticide that works well against rice weevils, underscoring the plant's potential for use in sustainable pest management techniques.

Keywords: Plant-derived insecticides, Pest mortality assessment, Plant-derived insecticides, Pest eradication strategies, Sustainable pest control

Introduction

Cereal grains are a vital component of human diet and play a very important role in the human civilization as billions of people consume rice for survival (Awika, 2011). Rice is one of the most important crops and is also considered a staple food for more than two billion people due to the minerals, vitamins, fibers, and carbohydrates it contains. (Akhtar et al., 2015). However, pest infestation remains a significant concern in stored grains, particularly in developing countries like the Philippines (Hodges et al., 1996). As one of Asia's leading rice producers, the Philippines' tropical climate fosters the proliferation of major pests that target stored grains. Birds, fungi, and rodents are among the primary culprits contributing to grain loss in the region (Francisco et al., 2009).

One of the most widespread and destructive pests of cereal grains and related products is the rice weevil, or *Sitophilus oryzae* (Koehler, 2022; Majd-Marani et al, 2023). It feeds and develops on stored grain (Akhtar et al., 2017). These insects cause severe damage to rice and reduce its weight, quality, commercial value, and seed viability, making it unfit for human consumption (Madrid et al., 1990). These conditions further reduce the selling power of food grains, which results in huge losses for farmers (Kumar and Kalita, 2017).

In light of that, attempts have been made to eliminate these stored grain insects. The cheapest and most efficient way of controlling stored-grain insect pests is the use of fumigants (Snelson, 1987). Fumigants need to be biologically active, highly volatile enough to be eliminated by aeration, not absorbed by grain, non-flammable, and non-corrosive. Currently, there are not many chemicals that adhere to all these requirements and can be used as fumigants.

In addition, the excessive use of conventional chemical insecticides has resulted in several serious problems, e.g., resistance to the chemical insecticides, elimination of economically beneficial insects, persistence in the environment, toxicity to humans and wildlife, and a higher cost of crop production (Khan and Selman, 1988). Numerous insects and mites have developed cross and multiple resistance, enabling them to withstand nearly all pesticides used for their control (Metcalf, 1980). This recognition of pesticides' harmful effects has led to the exploration of alternative management strategies that are less invasive. One such strategy is the utilization of natural pesticides, particularly those derived from plants (Isman, 2006).

Natural insecticides, which possess a diverse array of biological properties effective against stored

grain pests, have been developed and are extensively utilized. Compounds derived from plants have proven successful in preventing pest infestations in stored products. The insect-repellent, toxic, antifeedant, and growth-inhibiting properties of this bioinsecticide disrupt the activities of insect pests (Chowański et al., 2016). Secondary plant metabolites such as organic acids, alkaloids, terpenoids, saponins, and glycoalkaloids serve as insecticides, especially as protectants for food ingredients. These plant-derived chemicals may act as toxins, chemosterilants, antifeedants, repellents, or attractants (Hikal et al., 2017).

Although evidence supports the benefits and potential of plant-based pesticides, their use in agriculture in the industrialized world is limited due to stringent registration requirements, resulting in few commercial opportunities for new botanical products. Nevertheless, the scientific literature on the bioactivity of plant derivatives against arthropod pests is expanding. In Africa, many small-scale farmers protect stored foodstuffs from pests by incorporating various plant materials (Jembere et al., 1995; Poswal and Akpa 1991; Talukder and Howse 1995). Recent research has been driven by the use of these traditional substances to establish a scientific foundation for their efficacy, active ingredients, and application methods (Weaver et al., 1991; Regnault-Roger et al., 1993; Schmidt and Streloke 1994).

Muntingia calabura, known locally as *aratiles* in the Philippines, is an evergreen tree that grows rapidly and is noted to flourish in Manila's polluted environment (Sarojini and Mounika, 2018). The tree's prevalence in the country underscores the need for further research into its potential community benefits. *Aratiles* has been shown to exhibit various pharmacological properties, including antibacterial (Sufian et al., 2013; Sibi et al., 2012), antifungal (Rajesh et al., 2014), antioxidant (Ragasa et al., 2015), and anti-inflammatory effects (Preethi et al., 2012). Additionally, the insecticidal potential of *aratiles* extracts is gaining interest. Studies indicate that its leaves contain flavonoids, chalcones, terpenoids, and phenolic compounds, making it a viable candidate for pest management programs (Preethi et al., 2012).

In this context, a study by Aarthi et al. (2021) demonstrated that *aratiles* leaf extract exhibits significant larvicidal activity against the filarial vector *Culex quinquefasciatus*, with both chloroform and ethanol extracts causing mortality in mosquito larvae. Furthermore, Mendoza et al. (2019) found that the ethanolic extract from fresh *aratiles* leaves has potential larvicidal effects on mosquito larvae. Bandeira et al. (2013) also discovered that extracts from the flowers and fruits of *aratiles* show insecticidal activity against the diamondback

moth, *Plutella xylostella*. These findings suggest that *aratiles* possess strong insecticidal properties, making it a promising candidate for inclusion in pest management strategies.

This study evaluated the insecticidal efficacy of ethanolic extracts from *aratiles* leaves against rice weevils. Weevil mortality was measured at 1.5 h post-exposure to various extract concentrations to assess effectiveness and detect any significant differences.

This research could provide substantial benefits to farmers, particularly rice growers, by safeguarding the quality of their harvests and assisting commercial entities in protecting stored grain products from rice weevil infestations. It has the potential to reduce costs and minimize resource wastage significantly. Moreover, this study will enrich the existing literature on rice weevils and the insecticidal properties of the *aratiles* plant, which is currently limited.

Materials and Methods

Collection and Preparation of Plant Leaf Extracts

Aratiles leaves of 5 kg were collected from multiple trees in a compound along the C5 Service Road in Valenzuela City, Philippines. These materials were sorted to separate ones with fresh quality from damaged or blackish colored leaves. Every 150 g of fresh leaves were washed thoroughly with normal tap water to remove the dirt adhering to it and then placed in nylon bags that were hanged.

Extracting the samples adhered to the protocols outlined by Surjowardojo et al. (2014), with some modifications. The leaves were shade-dried for three weeks in a setup placed under a roof that allowed open-air circulation without direct sunlight exposure. Once the leaves attained a brittle texture, their dry weight was recorded. The leaves were then ground using a mortar and pestle and subsequently powdered using a blender. The resulting powder was weighed and stored in an airtight glass jar.

The sample was subjected to ethanol extraction at the University of the Philippines Manila (UPM). Although methanol has been shown to extract greater quantities of secondary metabolites and lower molecular weight polyphenols, ethanol is preferred due to its safety and suitability for human use (Dai and Mumper, 2010).

Preparation of Test Insects

Four hundred live adult rice weevils were collected from the Karuhatan public market in Valenzuela City, following the methodology of Acero (2014). They were housed in a plastic container filled with rice, white flour, yeast, and wheat corn

flour, and covered with a fine mesh cloth to provide sustenance and oxygen when not in use for the study. The insects were isolated from any other eradicates and insecticides. The test insects utilized in this study were authenticated by the Department of Natural Sciences at San Beda University, Manila.

Application of the Aratiles Leaves Ethanolic Extract

The experiment was conducted using a completely randomized design (CRD) with four replications to evaluate the effect of *aratiles* leaves ethanolic extracts on rice weevils. Filter paper discs were placed at the bottom of disposable petri dishes, onto which 20 rice weevils were introduced. The *aratiles* leaves ethanolic extract was diluted in distilled water to create different concentrations: 25% for treatment 1, 50% for treatment 2, and 75% for treatment 3. Each filter paper received 1 mL of the corresponding *aratiles* leaves ethanolic extract solution for the treatment groups, distilled water for the negative control, and Malathion for the positive control, administered using a syringe. A new syringe was used for each concentration. The four replicates of each treatment were compared with the negative control and a 10 ppm Malathion standard, following the methodology from Acero (2019).

Data Collection and Analysis

The number of dead insects was tallied 1.5 h post-treatment application. Mortality was determined by the lack of response to a gentle prod with a glass rod (Su, 1977). Insect behavior was also monitored.

The normality of the collected data was assessed using the Shapiro-Wilk Test. Upon failing to meet the criteria for normal distribution, the data were analyzed using the Kruskal-Wallis non-parametric test to evaluate significant differences in mortality counts among rice weevil groups treated with various concentrations of *aratiles* ethanolic extract, and both negative and positive controls. Subsequent pairwise comparisons were conducted using the Mann-Whitney U Test as a post hoc analysis. All statistical tests were performed using Microsoft Excel.

Results and Discussion

Figure 1 exhibits the percent mortality of the rice weevils under the treatments after 1.5 h. The results illustrated that the highest average value of mortality of the rice weevil, 76.15%, was obtained using the 75% concentration of *aratiles* extract, while there was only 0.03% mortality for both the negative (water) and positive controls (Malathion).

When these mortalities were subjected to the Kruskal-Wallis test, a p-value of $p = 0.006$ was obtained indicating a significant difference in the treatments in terms of the mortality counts of the rice weevils. Consequently, it can be said that the concentrations influenced the mortality counts.

Following a post hoc analysis using the Mann-Whitney U-test, it was shown that there was no significant difference in death rates between the positive (malathion) and negative (water) control groups ($U = 8$, $p > 0.05$, $U_{crit} = 0$). Significant

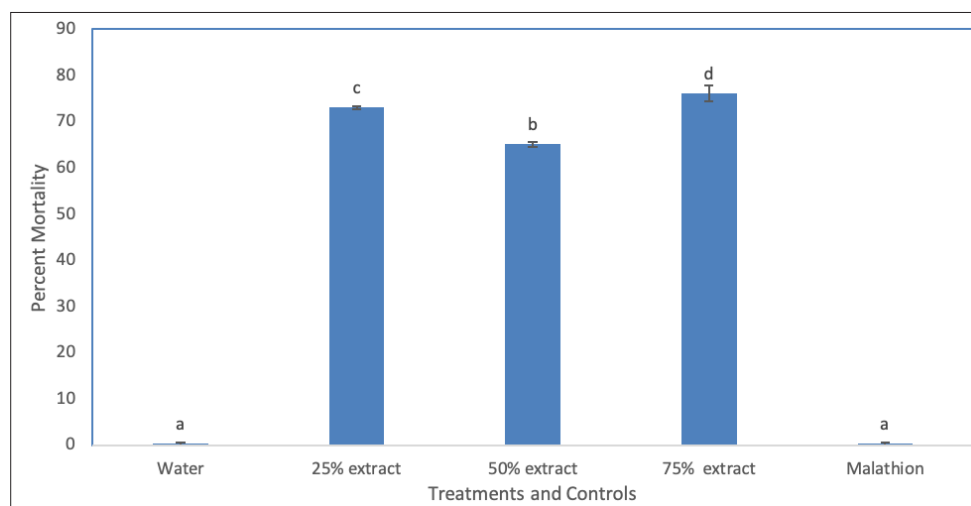


Figure 1. Percent mortality of the rice weevil under different treatments and controls.

Note: Treatment groups with the same letter do not exhibit statistically significant differences at 5% level of significance, as determined by Pairwise Mann-Whitney U Test. Different letters indicate significant differences between treatments. Error bars represent the standard error (SE) of the mean.

differences were observed between the two control groups and the various concentrations of *aratiles* extract. The comparison of *aratiles* extract concentrations at 25% and 50% ($U = -31$, $p > 0.05$, $U_{crit} = 0$), 25% and 75% ($U = -39.5$, $p > 0.05$, $U_{crit} = 0$), and 50% and 75% ($U = -39.5$, $p > 0.05$, $U_{crit} = 0$) revealed notable variations in mortality percentages. Notably, the highest percent mortality was recorded at the 75% concentration. Duke (2017) posits that even in optimal conditions, only a minimal fraction of pesticides reach their molecular targets in pests. Factors such as the target insect species, its life cycle, the insecticide's mode of action, and application method can influence an insecticide's effectiveness. The results also indicate that the extracts surpassed both positive and negative controls in terms of mortality rates.

The observed mortality in rice weevils post-exposure to different concentrations of *aratiles* extracts could be linked to the presence of phytochemicals such as flavonoids, chalcones, terpenoids, and phenolic compounds, which have been identified to possess insecticidal properties (Preethi et al., 2012). Additionally, tannin compounds found in *aratiles*, known for their insecticidal activities (Mohamad Yusof et al., 2013), may have contributed to the weevils' mortality.

Ngozi et al. (2009) provided evidence that higher doses of flavonoids disrupt the normal physiological functions of insects. These phytochemicals alter certain metabolic pathways within organisms. The impact of flavonoids on transhydrogenation, NADH oxidase, and succinate dehydrogenase reactions indicates that compounds with similar properties may be effective in controlling insect populations by modifying mitochondrial enzyme components. This research corroborates the findings of the current study, which demonstrate that rice weevils are particularly vulnerable to mortality upon exposure to *aratiles* ethanolic extract.

Interestingly, the positive control exhibited no significant effect on the mortality of rice weevils. Past research has indicated that the excessive use of insecticides to manage stored-grain pests in tropical regions has frequently led to insecticide resistance (Champ and Dyte, 1976; Kay and Collins, 1987; Subramanyam and Hagstrum, 1996, as cited in Pereira et al., 2009). Rahim and Ong (1991, as cited in Odeyemi et al., 2010) reported from a 1985 Malaysian survey that stored-grain pests, including those from the rice weevil family *Curculionidae*, had developed resistance to Malathion. Furthermore, the adult rice weevils used in this study were collected from a public market without a controlled environment, unlike in the study by Al-Harbi et al. (2021), where

all adult rice weevils were less than two weeks old and maintained under controlled conditions prior to testing, resulting in a 100% mortality rate.

Conclusion

The findings highlight the efficacy of ethanolic extract from *aratiles* leaves as an insecticide. Specifically, a 75% extract concentration resulted in the highest average mortality rate for rice weevils, reaching 76.15% just 1.5 h after treatment. This starkly contrasts with the negligible 0.03% mortality rate observed in both the positive (Malathion) and negative (water) controls. This study provides valuable insights into the potential of *aratiles* leaf ethanolic extract as a safe, natural pesticide for rice weevils. Further research is necessary to fully understand the repellent properties of *aratiles* against rice weevils. Given the resistance of Curculionids to Malathion, future studies should include other commercially available insecticides as positive controls to enhance the validity of results. Additionally, investigating other parts of the *aratiles* plant, such as stems and roots, for their insecticidal properties is advisable. Exploring various solvents and extraction methods, including Soxhlet extraction, and different bioassay techniques like fumigation could yield further insights. Moreover, assessing the effectiveness of *aratiles* extract across different growth stages of rice weevils and its insecticidal activity against other significant rice pests warrants investigation.

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SILICON DEPOSITION AND EFFECTS OF FERTILIZATION ON THE YIELD OF THREE RICE VARIETIES

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Abstract

Other sources of nutrients can improve plant growth and development in rice. It is necessary to develop alternate strategies to ensure crops provide competitive yields and at the same time maintain the long-term ecological balance of the soil ecosystem. The use of silicon fertilizers in agriculture is considered an alternative approach to enhance crop yield and soil health. This study used scanning electron microscopy (SEM) and energy dispersive x-ray (EDX) analysis to evaluate the effectiveness of silicon fertilizer on enhancing the rice yield and silicon deposition on the adaxial leaf surface of NSIC Rc 160, NSIC Rc 600, and NSIC Rc 512. Silicon treatments include 0 t ha⁻¹, 1 t ha⁻¹ and 2 t ha⁻¹ Si. SEM analysis of NSIC Rc 512 with 2 t ha⁻¹ Si application rate obtained the highest silicon (12.76% Si by weight) concentration. In NSIC Rc 160, plants treated with 1 t ha⁻¹ Si had the highest silicon content (10.32% Si by weight). On the other hand, NSIC Rc 600 obtained its highest silicon content at 0 t ha⁻¹ Si (8.93 Si by weight). Regardless of varieties, higher grain yield was obtained in 1 t ha⁻¹ Si (NSIC Rc 160, 8.17 kg/plot; NSIC Rc 512, 8.70 kg/plot; and NSIC Rc 600, 8.75 kg/plot). The enhanced Si content and the increase in grain yield demonstrate the potential of Si fertilizer on improving rice production. However, further studies on the evaluation of the effectiveness of silicon fertilizer on different soil types is necessary.

Keywords: *Silicon fertilizers, Silicon deposition, SEM-EDX analysis, Rice, Yield*

Introduction

For more than three billion people worldwide, rice is one of the most significant cereal crops, serving as their primary source of calories and staple food (Datta et al., 2017). In the Philippines, rice production produces approximately 3% of the world in both lowland transplanted paddies and upland rainfed direct seeded areas (Haefele et al., 2014). According to Prakash (2010), rice yields are decelerating in post green revolution mainly because of the imbalance in fertilizer use, soil degradation, type of cropping system practiced, and lack of suitable rice genotypes for low moisture adaptability and disease resistance. Considering rice is a rigorous feeder crop, effective nutritional management is necessary to increase rice yield (Thind et al., 2012). As rice is a silicon accumulator, adequate consideration should be given to silicon nutrition for the productive and healthy plant growth, especially rice (Rao et al., 2017).

Silicon (Si) is the second most abundant element in the soil after oxygen, but not yet classified as an essential nutrient and still not recognized as an important component for most plants. As a consequence of its poor solubility, silicon in soil is limited while being abundant in the earth's crust (Lindsay 1979). Plants can only absorb Si in the form of soluble monosilicic acid (H₂SiO₄), a non-charged molecule, which plays a significant role in wide variety of plants for beneficial roles in growth,

production, yield and plant resistance to biotic and abiotic stress. According to Ma and Takahashi (2002), adequate Si uptake can boost the tolerance of agronomic crops, especially rice. Furthermore, Si has long been recognized as particularly important to rice in the family *Gramineae* (Cai et al., 2008).

Metal toxicity, salinity, drought, and temperature stresses can be alleviated by Si application in a study by Currie and Perry (2007). Toxicities from excessive level of manganese (Mn), cadmium (Cd), aluminum (Al), and zinc (Zn) are alleviated by Si through several mechanisms: (1) accumulation of metal as silicate, (2) reduction in lipid peroxidation, (3) increased activity of enzymatic (e.g., superoxide dismutase) and non-enzymatic antioxidants (e.g., ascorbate), and (4) increased release of phenolics. In addition, Si can also make other nutrients available for plant uptake like phosphorus. Si fertilization can increase the P content of plants in terrestrial systems by increasing P availability in the soil as suggested in the study of Schaller et al. (2019). Despite the growing interest Si availability in terrestrial soil systems, there remains limited information regarding its interactions with P availability. Some studies suggest that Si fertilization in terrestrial systems can enhance plant P content by potentially increasing P availability. However, our understanding of how Si specifically interferes with P mobilization in soils remains incomplete, with only a few sorption experiments using pure minerals and limited research on Si's effects on P dynamics.

Si also enhances plant resistance to insect pests by thickening plant tissue in the culm's cell wall, making it less digestible or even causing substantial harm to feeding insects' mandibles (Savant et al., 1997; Massey and Hartley, 2006). Conversely, Si deficiency renders plants more susceptible to insect feeding, negatively impacting crop yield and quality. The use of Si as a pest control agent has shown promise across various plant species, as it can modify plant defense responses and mechanically impair herbivorous insects.

Studies investigating the impact of Si on rice grain quality and nutrition have garnered significant attention. Notably, Si application during the reproductive stage of rice plants led to a remarkable 30% increase in grain yield (Ma et al., 1989). Furthermore, research by Zhang et al. (2007) demonstrated that Si fertilizer enhances both milling and eating qualities of rice by increasing head rice yield and improving the breakdown value in Rapid Viscosity Analysis (RVA) profiles. Additionally, Liu et al. (2017) found that Si fertilizer positively influences the nutritional composition of rice, elevating mineral element concentrations, protein content, and certain amino acids in both brown and milled rice.

Silicon (Si) offers manifold advantages, including its role in promoting essential growth and productive development of rice crops (Rao et al., 2017). As global environmental changes unfold, the significance of Si will continue to rise, especially as crop production strives for greater efficiency and sustainability (Chinnasami et al., 1978). Consequently, there is a pressing need to explore effective and ecologically sound alternative methods to enhance rice productivity. Si amendment emerges as a promising avenue for achieving this goal. This study investigated the impact of Si fertilization on yield enhancement across different rice cultivars.

Materials and Methods

Collection, Processing and Characterization of Soil Samples

Soil was sieved through a 5 mm screen, homogenized, and allowed to air-dry. Composite samples were taken from each processed bulk soil for chemical characterization (nitrogen, phosphorus, potassium) and pH (1:1, soil: water suspension).

Treatments

The 3 x 3 complete factorial treatment structure was arranged in a randomized complete block design (RCBD) with four replicates. The study employed nine treatment combinations of the two factors. Variety (V): V1 - NSIC Rc 160, V2 - NSIC Rc 512,

and V3 - NSIC Rc 600. Silicon treatment (S): S1 = 0 t ha⁻¹ Si, S2 = 1 t ha⁻¹ Si, and S3 = 2 t ha⁻¹ Si. Silicon fertilizer (silica-25%, Alkaline-40% and magnesium 2%) was incorporated into the soil during the first harrowing.

Silicon Mapping Using Scanning Electron Microscopy (SEM) and Energy Dispersive X-ray (EDX) Analysis

Leaf samples were collected at reproductive stage then six small sections of leaves were cut for microscopic characterization of Si deposition. Scanning Electron Microscopy (FlexSEM1000, Hitachi, Tokyo, Japan) microanalysis and mapping were used to determine Si deposition in the adaxial leaf surface of rice while Energy Dispersive X-ray spectrometers (Quantax 70/80, Bruker Nano GmbH, Berlin, Germany) were used to analyze relative silicon content.

Two replications for each treatment were analyzed. Under SEM, the magnification of samples' images was set to 200x and 2,000x, and 5,000x system operation at voltage of 5.0 kV. Focus and brightness were adjusted to obtain clear and good quality images. After generation of SEM images, EDX was set to scan samples. System operation was set at voltage 20.0 kV. Silicon picks were proportionally quantified according to leaf carbon (C), oxygen (O), aluminum (Al), sulfur (S), chlorine (Cl), potassium (K), and calcium (Ca) contents. Similarly, scanning electron microscopy coupled to EDX microanalysis mapping were conducted to determine Si content and deposition on leaf surface. Sample numbers of pixels were multiplied by a corresponding scale value and were summed to give the overall Si content. Silicon content was associated with a green color in this study.

Crop Management Practices

Land preparation followed the PhilRice Palay Check System (2022) with final leveling and layout completed a day before transplanting. Seedbed was prepared using wet-bed following the standard procedures. Twenty-one-day-old seedlings were transplanted in a 4.5 x 23 m plot with a 20 x 20 cm spacing. Replanting occurred a week after transplanting. The fertilizer rate of 120N - 60P₂O₅ - 60K₂O was applied 10 days after transplanting, at tillering stage, early panicle initiation, and a week after early panicle initiation. Management of pests, water, and other cultural practices was based on PhilRice Palay Check System. Data on plant height, grain yield and yield components (number of panicles per m², number of spikelet per panicle and 1000-grain weight) were recorded at physiological maturity.

Soil Chemical Characteristics

Soil samples were collected from Block 4, Lot 2 at PhilRice CES Experimental fields. The results of the soil chemical characteristics were identified. Soil samples had low concentration (0.1 - 0.2 % N) of nitrogen, medium (5 - 10 ppm) level of soil available phosphorus, and low concentration (<0.20) of exchangeable potassium. The soil pH was moderately alkaline (7.9 - 8.4).

Results and Discussion

Scanning Electron Microscopy (SEM) and Energy Dispersive X-ray (EDX) analysis

Silica deposition of NSIC Rc 160, NSIC Rc 512, and NSIC Rc 600 using SEM has been best magnified at 2,000x (Figure 1). The images generated show

silica bodies on the adaxial leaf surface of the rice plants, including the stomata. According to Heckman (2013), Si is absorbed as monosilicic acid (H_4SiO_4) by the plants and translocated through the xylem until it deposits under the cuticle and in intercellular gaps. Silica tends to be deposited as a 2.5 μ thick layer immediately beneath the thin cuticle layer, forming a Cuticle-Silicon double layer (Rao et al., 2017). The location and the mechanical strength of this Cuticle-Silicon double layer help to maintain erect leaves, minimize transpiration and protects the rice plant from fungal diseases and insect pests (Savant et al., 1996).

Figure 2 and 3 show the silicon map (in green color) and the graph of the energy dispersive X-ray (EDX) microanalysis of the adaxial leaf surface of NSIC Rc 160, NSIC Rc 600, and NSIC Rc 512 with

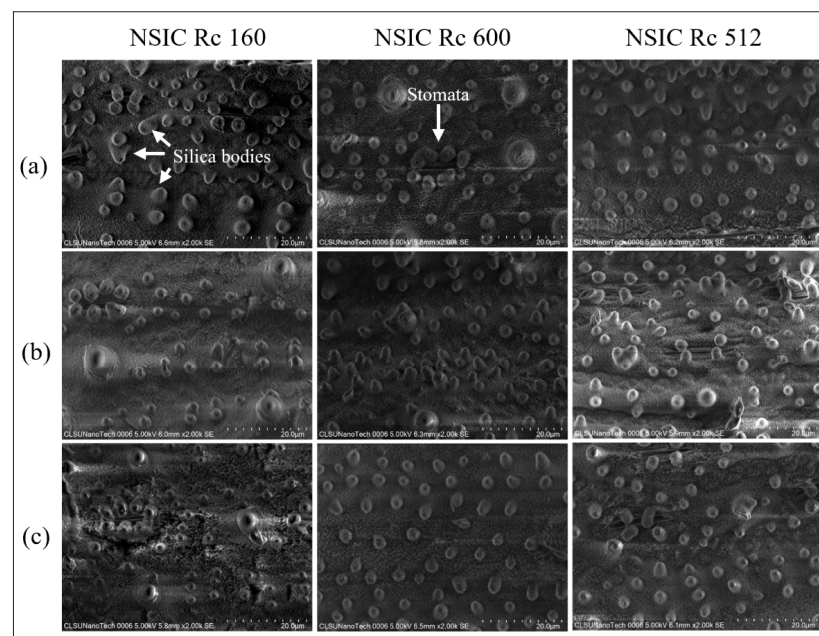


Figure 1. Silicon deposition on the adaxial leaf surface of NSIC Rc 160, NSIC Rc 600 and NSIC Rc 512 at 2,000x magnification with different silicon fertilization treatments; (a) 0 t ha⁻¹, (b) 1 t ha⁻¹, and (c) 2 t ha⁻¹ Si application.

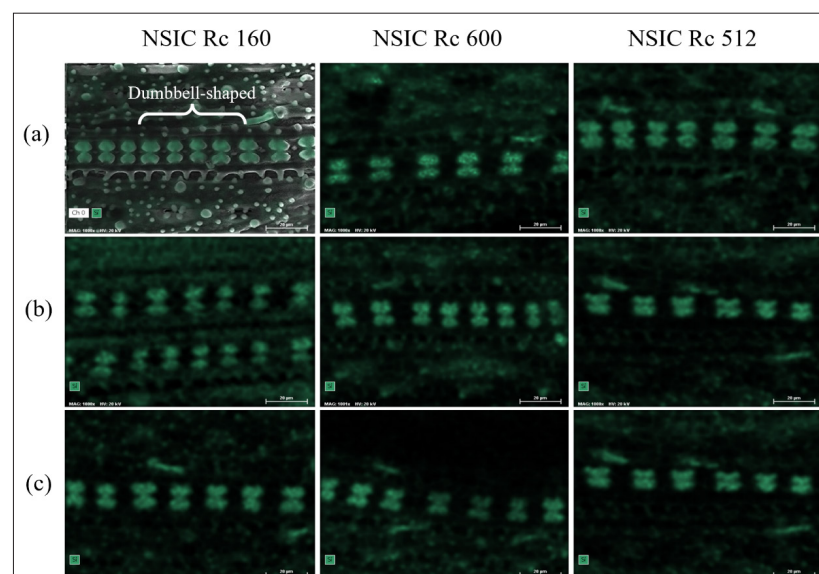


Figure 2. Silicon mapping conducted on the adaxial leaf surface of NSIC Rc 160, NSIC Rc 600, and NSIC Rc 512 using SEM-EDX. The study investigated different silicon fertilization treatments; (a) 0 t ha⁻¹, (b) 1 t ha⁻¹, and (c) 2 t ha⁻¹ Si application. Notably, silicon deposition is visualized in green color.

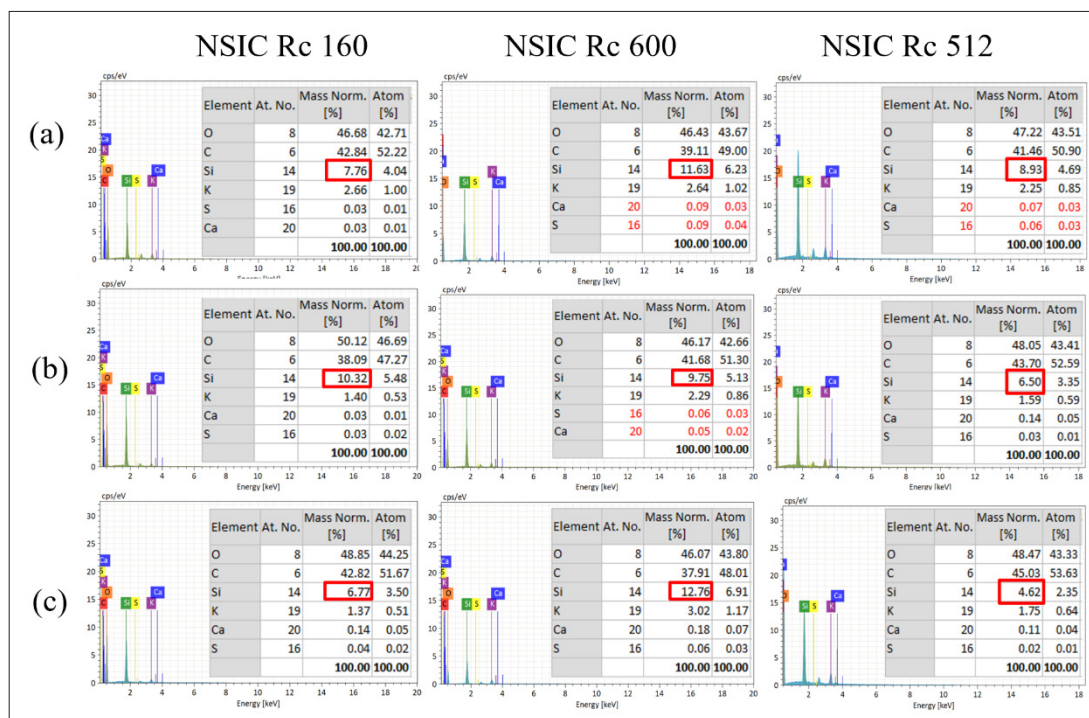


Figure 3. Micro-analysis conducted on adaxial leaf surface of NSIC Rc 160, NSIC Rc 600, and NSIC Rc 512 using SEM-EDX. The study investigated different silicon fertilization treatments: (a) 0 t ha⁻¹, (b) 1 t ha⁻¹, and (c) 2 t ha⁻¹ Si application.

different silicon treatments. According to Abed-Ashtiani et al. (2012), SEM/EDX observations of leaf samples from the adaxial surfaces of rice revealed Dumbbell-shaped or ladder-like silica cells, along with small scattered silica cells (Figure 2a).

In NSIC Rc 160, plots without Si application resulted in a 7.76% Si by weight. Highest concentration of silicon by weight (10.32%) was recorded in plots with 1 t ha⁻¹ Si application. Lowest silicon concentration (6.77% Si by weight) was obtained by NSIC Rc 160 with 2 t ha⁻¹ Si application. NSIC Rc 600 with 0 t ha⁻¹, 1 t ha⁻¹, and 2 t ha⁻¹ Si applications had 11.63, 9.75, 12.76% Si by weight, respectively. NSIC Rc 512 without Si application had 8.93% Si by weight, which is higher than treatments 1 t ha⁻¹ and 2 t ha⁻¹ Si application with 4.62% and 6.50%, respectively. All values are %Si relative to other elements (%C, %O, %S, %Ca, and %K).

The corresponding EDX spectra compared with the SEM images demonstrated a significant difference in silicon content between silicon treated and non-treated plants. Si-treated plants contained more silicon than non-treated ones (Abed-Ashtiani et al., 2012).

Grain Yield

Figure 4 shows the grain yield of different varieties with different silicon fertilization treatments. NSIC Rc 160 with 1 t ha⁻¹ Si rate had the highest grain yield (8.17 kg/plot), followed by 0 t ha⁻¹

and 2 t ha⁻¹ Si treatments with 7.12 and 7.27 kg/plot, respectively. NSIC Rc 600 with 2 t ha⁻¹, 1 t ha⁻¹, and without Si application produced 7.77 kg/plot, 8.75 kg/plot, and 6.62 kg/plot, respectively. Meanwhile, NSIC Rc 512 produced more yield among varieties. Application of 1 t ha⁻¹ Si resulted in a higher grain yield of 8.70 kg/plot, while treatments with 2 t ha⁻¹ and 0 t ha⁻¹ Si yielded 7.98 kg/plot and 7.24 kg/plot, respectively. However, grain yields of the varieties are not statistically different. These results could be due to variety differences, planting season, growing conditions, timing of Si application, and response of varieties to the Si treatments. According to Tamai and Ma (2008), the impact of silicon on plant growth becomes more pronounced under stress conditions, but it may not be readily discernible under non-stressed conditions. In soil culture experiments including pot and field studies, applying Si fertilizers at varying levels remains the primary method for assessing the effects of silicon. Notably, even in the absence of silicate fertilizers, plants in the control treatment can still access a significant reservoir of Si from the soil. Consequently, evaluating the effects of Si application in field experiments may have limitations. Nevertheless, this study represents the initial attempt to evaluate Si utilization under PhilRice environmental conditions.

The Si content of rice can reach up to 10% of the dry weight of the shoot, making it a Si-accumulating species (Tamai and Ma, 2003). Si-uptake ability by rice roots during a period of 24 h was much higher

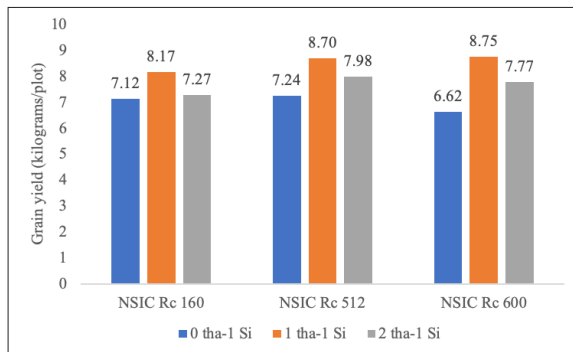


Figure 4. Grain yield of different varieties with different silicon fertilization treatments.

than other gramineous species (wheat, triticale, sorghum, rye, maize, and barley). However, there are significant differences in the silicon concentration of different plant species. Understanding genotypic differences is crucial because they affect the amount of Si that accumulates (Swain and Rout, 2018).

Chaiwong et al.'s (2021) study highlighted the beneficial effects of Si application, which led to an increase in rice grain yield and a reduction in the lodging index by approximately 13.7%. Additionally, Si treatment resulted in higher spikelet numbers, increased percentage of filled grains, and enhanced straw dry weight compared to the absence of Si application. Notably, Si application significantly improved panicle characteristics, ultimately contributing to increased grain yield (Kim et al., 2012).

The application of Si fertilizers has been shown to enhance growth parameters, increase yield, improve yield attributes, and enhance rice grain quality (Ahmad et al., 2013). Si plays a crucial role in grain development and appears to significantly impact grain quality (Bhaskaran, 2014). Notably, introducing Si during the reproductive stage of rice plants led to a remarkable 30% increase in grain yield (Ma et al., 1989). Additionally, Prabhu et al. (2001) reported a 20 - 40% increase in grain weight when Si levels were raised from 0 - 800 kg/hm² SiO₂. Research findings by Rao et al. (2017) indicated that rice crops can uptake Si within the range of 230 - 470 kg ha⁻¹. However, the response of rice to Si application varies due to the specific application conditions (Pati et al., 2016). The observed changes in grain yield may be attributed to rice growth characteristics and the stimulating effects of Si under both biotic and abiotic stress conditions.

In Asian countries, particularly South Korea, the use of Si fertilizers in paddy soils for rice cultivation has been well-established (Liang et al., 2015). Since the 1960s, South Korea has conducted extensive research on the impact of Si on rice growth and productivity (Kim and Choi, 2002; Lee et al., 2005). Initially, grounded wollastonite served as the first Si fertilizer in Korea from 1963 to 1990. Subsequently, blast furnace slag—a by-product of the iron and steel industries—became another significant source of Si fertilizer. Supported by government funding since 1996, farms have received this Si fertilizer at four-year intervals. As a result of successive Si fertilizer applications, the average Si content in soil increased from 75 mg SiO₂ kg⁻¹ in the 1970s - 118 mg SiO₂ kg⁻¹ in 2003. South Korea has extensively studied the beneficial effects of Si on rice yield. For instance, a 26-year field experiment conducted from 1975 to 2000 involved Si fertilization at 1.5 Mg ha⁻¹ in clay loam paddy fields. Compared to NPK treatment, the average rice yield consistently increased over five-year intervals with Si fertilizer applied at 1.5 Mg ha⁻¹ (Kim and Choi, 2002). It is important to note that the recommended Si application rate in Korea (200-300 kg ha⁻¹) differs from that in the Philippines due to variations in soil and climate conditions.

Conclusion and Recommendations

In this study, varietal differences were observed in response to Si fertilization. Using SEM-EDX analysis on the adaxial leaf surface, the highest silicon content (10.32% by weight) was observed in NSIC Rc 160 plots treated with 1 t ha⁻¹ Si. Conversely, NSIC Rc 600 and NSIC Rc 512 exhibited their highest silicon content at 2 t ha⁻¹ Si (12.76%) and 0 t ha⁻¹ Si (8.93% by weight), respectively. Regarding grain yield, the highest yield was obtained in plots treated with 1 t ha⁻¹ Si, regardless of rice varieties: NSIC Rc 160 (8.17 kg/plot), NSIC Rc 512 (8.70 kg/plot), and NSIC Rc 600 (8.75 kg/plot). Notably, even with 1 t ha⁻¹ of Si treatment, yields were superior to those without Si. However, given that Si was initially applied in this field, significant yield differences may take more time to manifest, similar to findings from studies conducted in Korea. Therefore, this study recommends further field trials across different varieties and seasons in a single location to thoroughly evaluate the effectiveness of Si application in rice.

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YIELD PERFORMANCE OF GINGER APPLIED WITH DIFFERENT LEVELS OF FERMENTED CORN COB SOLUTION AS POTASSIUM SOURCE

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Abstract

Intercropping ginger (*Zingiber officinale*), locally known as “luya,” with rice provides agronomic and economic benefits, including enhanced soil fertility, efficient land utilization, and increased income for farmers. Ginger cultivation requires substantial nutrient input, particularly potassium, with recommended application rates of 180-180-255 kg N P₂O₅ K₂O per hectare. This highlights ginger’s sensitivity to potassium levels. This study, conducted at the Department of Agriculture, Capiz Research Outreach Station in Astorga, Dumarao, Capiz, aimed to assess ginger yield performance using fermented corn cob solution (FCCS) as a potassium source. A Randomized Complete Block Design (RCBD) with four replications was employed, analyzing differences among tested factors using the Least Significant Difference (LSD) test. The results revealed that combining 6,300 kg of commercial organic fertilizer with varying rates of FCCS (22,000 L, 33,000 L, and 44,000 L per hectare) produced yields comparable with using 88.36 kg of Muriate of Potash as the potassium source. Notably, the 22,000 L rate of fermented corn cob solution emerged as the most profitable organic potassium source among the evaluated FCCS rates, with the production cost incurred per kilogram of ginger being PhP 29.60. These findings suggest that FCCS can be an effective and economically viable alternative to commercial potassium fertilizers in ginger-rice intercropping systems, promoting sustainable agricultural practices and optimizing resource use.

Keywords: Commercial organic fertilizer (COF) plus potassium source, Fermented corn cob solution (FCCS), Ginger

Introduction

Ginger (*Zingiber officinale*) is a rhizomatous herbaceous perennial plant that is commonly known for its aromatic and pungent root. Ginger (*luya*) can be effectively intercropped with rice. Originating in Southeast Asia, ginger has been widely cultivated in tropical and subtropical regions around the world. It thrives in well-drained, loamy soils with high organic matter content, conditions often found in rice paddies after the rice harvest.

Intercropping ginger with rice can enhance soil fertility, optimize land use, and provide additional income to farmers. The cultivation of ginger alongside rice can also help in pest and disease management, as the strong aroma of ginger can repel certain pests that typically affect rice crops. Moreover, ginger has a relatively short growing cycle of about 8 - 10 months, which complements the rice growing season and allows for efficient rotation and intercropping strategies.

Incorporating ginger into rice-based farming systems aligns with sustainable agricultural practices, promoting biodiversity and improving the resilience of crop production systems. The integration of ginger and rice cultivation is a promising approach to achieving higher productivity and sustainability in agro-ecosystems.

In the Philippine agriculture, the National Organic Agriculture Program (NOAP, 2016), spearheaded by the Department of Agriculture (DA), aims to foster sustainable agriculture while benefiting the health of Filipinos.

Extracts and active constituents from ginger exhibit potent antioxidant, anti-inflammatory, anti-mutagenic, antimicrobial, and potential anti-cancer properties (Rahmani et al., 2014). Ginger is a staple in many dishes, and its economic value is significant (DA-RFO VI, 2012). However, ginger’s nutrient requirements are substantial. In a study by Azizah et al. (2021), yield, plant biomass, potassium (K) uptake, and soil exchangeable K were strongly influenced by K application rates, with the highest impact observed at 300 kg of K ha⁻¹. This underscores ginger’s sensitivity to potassium levels.

Fertilizer application is essential for achieving good yields in ginger production. While commercial synthetic fertilizers are commonly used by growers, they conflict with organic agriculture practices. Fortunately, farms have abundant sources of organic fertilizer in the form of farm waste, including rice straw, rice hulls, corn stalks, and corn cobs.

Corn farmers in the region could consider planting ginger as a main crop or alternate crop. Corn cobs, a byproduct of corn plants, contain potassium (average

of 4.8 g kg⁻¹ DM) and phosphorus (average of 0.7 g kg⁻¹ DM). Heuzé et al. (2016) reported that corn cob dry matter alone could supply significant potassium and other minerals if used as organic plant fertilizer.

Furthermore, soaking corn cobs in water for several weeks to release their potassium content has shown promising results as a cost-effective fertilizer, leading to increased harvest yields (Cascaro, 2012). This study aims to evaluate the yield performance of ginger using fermented corn cob solution (FCCS) as a potassium source.

Materials and Methods

Project Site

The experimental site is a plain upland located at the Department of Agriculture-Research Outreach Station in Astorga, Dumarao, Capiz. Situated within an upland corn area of Dumarao, the site has coordinates 110°15'47" north and 122°48'18" east, with an elevation of 123 feet above sea level. The soil in the area is predominantly clay loam to heavy clay, and it receives an average annual rainfall of 3,021 mm.

Established in 1982, the station accommodates various crops, including cassava, corn, sweet potato, soybean, peanut, banana, fruit trees, and other vegetables, covering an area of approximately 4.5 ha. For this study, we selected an area previously planted with sweet potato, considering two main criteria: (1) uniformity in terms of slope and fertility gradient, and (2) an area with at least 408 m² to accommodate the research.

Experimental Design

The experiment was laid out in a randomized complete block design (RCBD) with four replications. The area was divided into 20 plots spaced 1 m from each other. Plot size was 3 m long x 4 meter wide at 100 x 30 cm plant spacing (Figure 1).

Treatments

The study comprised five treatments involving a combination of commercial organic fertilizer and varying rates of corn cob solution as a potassium source, as shown in Table 1:

- T1 – 6,300 kg commercial organic fertilizer + 11,000 L fermented corn cob solution per hectare (81.87-187.22-29.7 NPK ha⁻¹).
- T2 – 6,300 kg commercial organic fertilizer + 22,000 L fermented corn cob solution per hectare (83.74 -187.44-37.4 kg NPK ha⁻¹).
- T3 – 6,300 kg commercial organic fertilizer + 33,000 L fermented corn cob solution per hectare (85.61-187.66-45.1 kg NPK ha⁻¹).
- T4 – 6,300 kg commercial organic fertilizer + 44,000 L fermented corn cob solution per hectare (87.48-187.88-52.8 kg NPK ha⁻¹).
- T5 – 6,300 kg commercial organic fertilizer + 88.36 kg Muriate of Potash per hectare (80-187-75 kg NPK ha⁻¹).

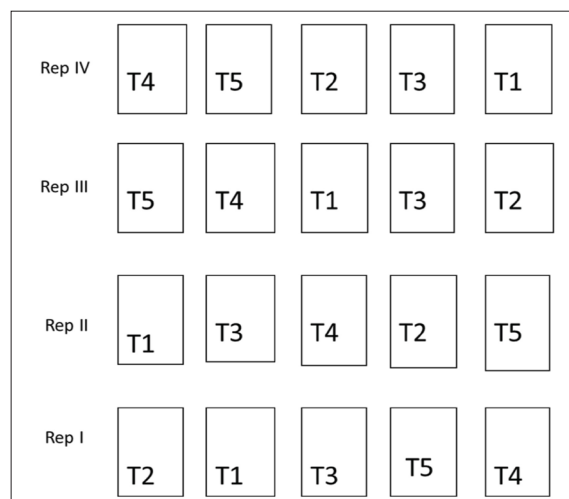


Figure 1. Experimental lay-out laid in RCBD with four replications.

Table 1. Timing and amount of fertilizer applied per treatment, May 2017 to March 2018.

Treatment	Time of Application	Age of the Crop	Fertilizer Source	Amount (kg ha ⁻¹)	Amount (bags ha ⁻¹)
T1 6,300 kg COF + 11,000 L FCCS ha ⁻¹ (81.87-187.22-29.7 NPK ha ⁻¹)	Basal	0 DAP	COF	3,150	63
	Side dressing	30 DAP	COF	3,150	63
			FCCS	3,667 l	
		60 DAP	FCCS	3,667 l	
T2 6,300 kg COF + 22,000 L FCCS ha ⁻¹ (83.74 -187.44-37.4 kg NPK ha ⁻¹)	Basal	0 DAP	COF	3,150	63
	Side dressing	30 DAP	COF	3,150	63
			FCCS	7,333 l	
		60 DAP	FCCS	7,333 l	
T3 6,300 kg COF + 33,000 L FCCS ha ⁻¹ (85.61-187.66-45.1 kg NPK ha ⁻¹)	Basal	0 DAP	COF	3,150	63
	Side dressing	30 DAP	COF	3,150	63
			FCCS	11,000 l	
		60 DAP	FCCS	11,000 l	
T4 6,300 kg COF + 44,000 L FCCS ha ⁻¹ (87.48-187.88-52.8 kg NPK ha ⁻¹)	Basal	0 DAP	COF	3,150	63
	Side dressing	30 DAP	COF	3,150	63
			FCCS	14,667 l	
		60 DAP	FCCS	14,667 l	
T5 6,300 kg COF + 88.36 kg MOP ha ⁻¹ (80-187-75 kg NPK ha ⁻¹)	Basal	0 DAP	COF	3,150	63
	Side dressing		MOP	44.18	0.88
		30 DAP	COF	3,150	63
			MOP	44.18	0.88

Note: DAP-Days after Planting, FCCS-Fermented corn cob solutions, MOP-Muriate of Potash, NPK ha⁻¹-Nitrogen, Phosphorus, Potassium per hectare, COF-Commercial Organic Fertilizer

Laboratory Analysis of soil nutrient requirement

Table 2. Soil Analysis results of the area (based on the result given by Department of Agriculture VI-Regional Disease and Diagnostic Laboratory as nutrient requirement for ginger per hectare).

Total Nitrogen(%N) Requirement	Total Phosphorous (% P ₂ O ₅) Requirement	Total Potassium (% K ₂ O) Requirement
80	0	75

Table 3. Laboratory analysis of commercial organic fertilizer (COF).

Total Nitrogen(%N) Kjeldahl Method	Total Phosphorous (% P ₂ O ₅)	Total Potassium (% K ₂ O)
1.27	2.97	0.349

Table 4. Laboratory analysis of Fermented Corn Cob Solution (FCCS).

Total Nitrogen(%N) Kjeldahl Method	Total Phosphorous (% P ₂ O ₅)	Total Potassium (% K ₂ O)
0.017	0.002	0.07

According to DA-Regional soils Laboratory 6 computation: Every 100 L of fermented corn cob solution this amount of nutrients are present.

Table 5. Total nutrient applied in ginger by treatment 2 per hectare.

Treatments	Total Nitrogen (N) kg ha ⁻¹	Total Phosphorous (P ₂ O ₅) (kg ha ⁻¹)	Total Potassium (K ₂ O) (kg ha ⁻¹)
Treatment 2 COF 6,299 kg ha ⁻¹	80	187	22
22,000 L ha ⁻¹ fermented corn cob solution	3.74	0.44	15.4
Total	83.74	187.44	37.4

Cultural Management Practices

Land preparation. The experimental area was cleared from unnecessary vegetation. Land was prepared by plowing and harrowing twice using a hand tractor with one week interval between operations. Study area was furrowed immediately prior to planting.

Selection of seed rhizome. The seed rhizomes were sorted before planting. Rhizome that is large, shiny, free from spots or marks, bud or eye injury was selected for planting. The planting materials used were native ginger varieties from the locality.

Planting of ginger. Planting materials were prepared and placed in a cool area using coconut or banana leaves. A good piece with ample buds was selected. The planting materials were cut into pieces, each containing 3 - 4 sprouts, and placed in a seed bed with a height of 20 cm and a width of 1 m (the length varied based on the area). Next, they were arranged in rows with a spacing of 2 cm between each piece. Compost and water were applied immediately after planting.

The ginger was transferred to the experimental plot once the sprouts had reached a length of 1 - 2 cm. Rhizomes were planted in furrows following an east-west direction, with one rhizome per hill at a planting distance of 100 cm x 30 cm and a depth of 4 - 6 cm. The total plant population was 33,333 seed pieces per hectare. To serve as mulching material, the newly planted ginger was covered with coconut leaves.

Preparation and application of corn cob solution. The corn cobs were collected from the surrounding corn area of DA-Research Outreach Station Astorga, Dumarao, Capiz.

The collected corn cobs were soaked in a plastic drum at a ratio of 1 kg of corn cob per 25 L of water. Soaking lasted for 4 weeks and was supplemented with 3 L of molasses in each preparation. Subsequently, the solution derived from the soaked corn cobs was drenched and applied to the ginger crop with the quantity adjusted based on the specific treatment requirements (Cascaro, 2012).

Fertilizer application. The application of fertilizer was based on the recommended rate per hectare per treatment using organic fertilizer, fermented corn cob solution, and inorganic fertilizer.

Water management. Plants were watered especially during dry months when the soil moisture was not sufficient to support crop growth.

Pest control. Inter-row cultivation was practiced 10 - 15 days after emergence, followed by hilling-up at 15 - 25 days after emergence. Regrowth weeds were manually controlled using hand tools throughout the ginger's growing period. Insect and disease prevalence were monitored, and good agricultural practices were applied at the experimental site.

Harvesting and post-harvest operation. The ginger was harvested 11 months after planting. Ginger was uprooted and the harvested rhizome was cleaned in water then air dried.

Yield. All the rhizomes taken from plants in the harvestable area were collected with marketable and non-marketable rhizomes separated. The total weight of the harvested ginger was divided by the number of hills per subplot. The rhizomes were air-dried for 2 h, placed in separate containers, properly labeled, and weighed in kilograms using a weighing scale. The final measurement was expressed in ha⁻¹ using the formula:

$$\text{Yield (tha}^{-1}\text{)} = \frac{\text{Yield per sub-plot}}{1,000 \text{ kg}} \times \frac{10,000 \text{ m}^2 \text{ (ha)}}{\text{Sub-plot area (m}^2\text{)}}$$

The yield data collected from each study was subjected to the analysis of variance using F-test for RCBD and the results were interpreted at 5% level of significance.

Data on production costs and return from sales were recorded. Profitability by treatment was determined using the return of investment (ROI) analysis:

$$\text{ROI (\%)} = \frac{\text{Net Income}}{\text{Total Investment}} \times 100$$

The profitability of the various treatments was computed using the return of investment analysis.

This was determined by dividing the net income with the total investment. The net income was obtained by subtracting the total expenses from the gross income derived from sales of cleaned ginger based on prevailing price. The total investment was taken as the total amount used as capital to establish the study.

Results and Discussions

The effect of fermented corn cob solution to ginger production

Table 6 shows that there were no significant differences on the yield of ginger when applied with 6,300 kg commercial organic fertilizer (COF) + 22,000 L of fermented corn cob solution (FCCS)/ha; 6,300 kg COF + 33,000 L FCCS ha⁻¹; and 6,300 kg COF + 44,000 L FCCS ha⁻¹ compared with the ginger applied with 6,300 kg COF + 88.36 kg of Muriate of Potash (control treatment). However, a significant difference on yield was observed in ginger applied with 6,300 kg COF + 11,000 L FCCS ha⁻¹.

The yields were: 9.24 t ha⁻¹ in 6,299 kg COF + MOP; 8.92 t ha⁻¹ in 6,300 kg COF + 22,000 L FCCS ha⁻¹ and 6,300 kg COF + 44,000 L FCCS ha⁻¹, respectively; 8.71 t ha⁻¹ in 6,300 kg COF + 33,000 L FCCS ha⁻¹; and 6.83 t ha⁻¹ in 6,300 kg COF + 11,000 L FCCS ha⁻¹.

This supports the findings of Budiastuti et al. (2023a), which indicate that FCCS can serve as an organic source of potassium in ginger production. Corn cob, as an organic fertilizer, met the standards with an organic carbon content of 62.21% and organic matter of 85.71%, along with a total potassium content of 2.17%, ranking it in the high category. These results align strongly with Ocampo's study (2013), which demonstrated that using corn cob as a potassium fertilizer can increase corn yield up to 10-15 t ha⁻¹ for open-pollinated corn and 20 t ha⁻¹ for hybrid corn.

The study also revealed that an extract from corn cob soaked in water for 4 - 5 weeks could be used as a foliar fertilizer, resulting in significant yield gains. Additionally, Budiastuti et al., (2023a) emphasized the correlation between phosphate and potassium uptake and plant biomass and root length. Given that ginger is a root crop, there is potential to benefit from the fermented corn cob solution.

Similar findings were observed in Budiastuti et al.'s (2023b) study on soybean, in which corn cobs promoted growth and yield. Furthermore, De Leon (2014) conducted research in Luzon, comparing corn cob rates of 0, 5, 10, 15, and 20 t ha⁻¹. The grain yield from plots using corn cobs did not significantly differ from commercially potassium-fertilized plots. Specifically, applying 6,300 kg of COF + FCCS at

rates of 22,000, 33,000, and 44,000 L resulted in a yield comparable to ginger treated with 6,300 kg of COF + 88.36 kg of MOP ha⁻¹. The study confirmed that the application of corn cob solution could be a substitute for MOP (Cascaro, 2012).

Furthermore, soil analysis and nutrient requirements for soybean (Table 2) already satisfied the total nutrient needs applied in ginger (Table 5). These conclusions are based on laboratory analyses of commercial organic fertilizer (COF) detailed in Tables 3 and 4.

Table 6. Mean yield of ginger applied with commercial organic fertilizer + different rates of Fermented Corn Cob Solution (FCCS) as source of potassium, Capiz ROS, Astorga, Dumarao, Capiz, May 2017 to March 2018.

Treatments	Mean Yield (t ha ⁻¹) ^{1/}
T1- 6,300 kg COF + 11,000 L FCCS ha ⁻¹	6.83 ^b
T2- 6,300 kg COF + 22,000 L FCCS ha ⁻¹	8.92 ^a
T3 - 6,300 kg COF + 33,000 L FCCS ha ⁻¹	8.71 ^a
T4 - 6,300 kg COF + 44,000 L FCCS ha ⁻¹	8.92 ^a
T5 - 6,300 kg COF + 88.36 kg MOP ha ⁻¹ (Control)	9.24 ^a
Mean Yield (t ha ⁻¹)	8.57
F-value	19.87
Pr(>F)	0.0000
C.V. (%)	5.23

Means with the same letter superscript are not significantly difference with each other

Economics of ginger production using different rates of fermented corn cob solution

Results of the economic analysis of ginger production per hectare using different rates of corn cob solution as source of potassium in comparison to inorganic source using MOP is shown in Table 7 and Table 8.

Gross income. The results showed that the highest gross income was obtained from ginger treated with 6,300 kg of commercial organic fertilizer (COF) combined with 88.36 kg of MOP resulting in PhP462,000.00 per hectare. Following closely were the treatments of ginger with 6,300 kg COF + 22,000 L of FCCS and 6,300 kg COF + 44,000 L FCCS ha⁻¹, yielding PhP446,000.00 ha⁻¹ and PhP435,500.00 ha⁻¹, respectively. The lowest gross income of PhP341,500.00 was observed in ginger treated with 6,300 kg COF + 11,000 L FCCS ha⁻¹.

Total expenses. Highest expense was obtained in ginger applied with 6,300 kg COF + 44,000 L FCCS ha⁻¹ (PhP269,900.00) or 2.58% higher than the ginger applied with 6,300 COF + 88.36 kg MOP ha⁻¹ (PhP263,121.00) while lowest expense of PhP262,125.00 was obtained from ginger applied with 6,300 kg COF + 11,000 L FCCS ha⁻¹ with 0.38%

Table 7. Economic analysis of ginger production applied with COF + different rates of FCCS as source of potassium, May 2017 to March 2018.

Parameter	Treatments				
	6,300 kg COF + 11,000 L FCCS ha ⁻¹	6,300 kg COF + 22,000 L FCCS ha ⁻¹	6,300 kg COF + 33,000 L FCCS ha ⁻¹	6,300 kg COF + 44,000 L FCCS ha ⁻¹	6,300 kg COF + kg 88.36 kg MOP ha ⁻¹
Yield of Ginger (t ha ⁻¹)	6.83	8.92	8.71	8.92	9.24
Farm gate Price (PhP kg ⁻¹)	50.0	50.0	50.0	50.0	50.0
Gross Income (PhP ha ⁻¹)	341,500.0	446,000.0	435,500.0	446,000.0	462,000.0
Total Expenses (PhP ha ⁻¹)	262,125.0	264,050.0	265,975.0	269,900.0	263,121.0
Net Income (PhP ha ⁻¹)	79,375.0	181,950.0	169,525.0	176,100.0	198,879.0
ROI (%)	30.28	68.91	63.74	65.25	75.58
Cost/kg of Ginger	38.38	29.60	30.54	30.26	28.48

Table 8. Cost and return analysis per hectare of ginger production.

Particular	T1 6,300 kg COF + 11,000 L FCCS ha ⁻¹	T2 6,300 kg COF + 22,000 L FCCS ha ⁻¹	T3 6,300 kg COF + 33,000 L FCCS ha ⁻¹	T4 6,300 kg COF + 44,000 L FCCS ha ⁻¹	T5 6,300 kg COF + 88.36 kg MOP ha ⁻¹
I. Income					
Yield of ginger (t ha ⁻¹)	6.83	8.92	8.71	8.92	9.24
Price (PhP kg ⁻¹)	50.0	50.0	50.0	50.0	50.0
Gross income (PhP ha ⁻¹)	341,500.0	446,000.0	435,500.0	446,000.0	462,000.0
II. Production Cost					
Material cost					
Ginger planting materials	167,000.0	167,000.0	167,000.0	167,000.0	167,000.0
COF	44,100.0	44,100.0	44,100.0	44,100.0	44,100.0
MOP					2,121.0
Total material cost	211,100.0	211,100.0	211,100.0	211,100.0	213,221.0
Labor cost					
Land preparation. (first plowing)	2,100.0	2,100.0	2,100.0	2,100.0	2,100.0
Second plowing and harrowing	800.0	800.0	800.0	800.0	800.0
Planting	3,000.0	3,000.0	3,000.0	3,000.0	3,000.0
Fermented corn cob preparation	1,925.0	3,850.0	5,775.0	7,700.0	-
Fermented corn cob solution application	1,200.0	1,200.0	1,200.0	1,200.0	-
Fertilizer application (basal and top-dressing)	6,000.0	6,000.0	6,000.0	8,000.0	8,000.0
Weeding	32,000.0	32,000.0	32,000.0	32,000.0	32,000.0
Harvesting	4,000.0	4,000.0	4,000.0	4,000.0	4,000.0
Total labor cost	51,025.0	52,950.0	54,875.0	58,800.0	49,900.0
Total production cost	262,125.0	264,050.0	265,975.0	269,900.0	263,121.0
III. Net income	79,375.0	181,950.0	169,525.0	176,100.0	198,879.0
ROI	30.28	68.91	63.74	65.25	75.58

COF= PhP350/bag; MOP = P1,200/bag

difference over the crop applied with 6,300 COF + 88.36 kg MOP ha⁻¹.

Net income. The highest net income was achieved from ginger treated with 6,300 kg of COF combined with 88.36 kg of MOP ha⁻¹ resulting in PhP198,879.00 income per hectare. Conversely, the lowest net income of PhP79,375.00 ha⁻¹ was observed in ginger treated with 6,300 kg COF + 11,000 L of FCCS ha⁻¹.

Return on investment (ROI). Ginger applied with 6,300 kg COF + 88.36 kg MOP ha⁻¹ with 75.58% recorded the highest ROI. This was followed by ginger applied with 6,300 kg COF + 22,000 L FCCS ha⁻¹ with 68.91%; 6,300 kg COF + 44,000 L FCCS ha⁻¹ with 65.25%; and ginger applied with 6,300 kg COF + 33,000 L FCCS ha⁻¹ with 63.74%. The lowest ROI was from ginger applied with 6,300 kg COF + 11,000 L FCCS ha⁻¹ with 30.28%. This showed that the application of 6,300 kg COF + 88.36 kg MOP in ginger production obtained the highest profit. However, using FCCS as an organic source of potassium at the rate of 22,000 L FCCS ha⁻¹ is more profitable among the fermented corn cob solution rates evaluated.

Cost/kg of ginger. The cost incurred per kilogram of ginger produced were as follows: PhP28.48 using MOP; PhP29.60 from 22,000 L FCCS; PhP30.26 from 44,000 L FCCS; PhP30.54 from 33,000 L FCCS; and PhP38.38 from 11,000.00 L FCCS.

Conclusion

The application of 6,300 kg commercial organic fertilizer in combination with different rates of fermented corn cob solution of 22,000, 33,000, and 44,000 L per hectare obtained a comparable yield with 88.36 kg Muriate of Potash as source of potassium in ginger production. The rate of 22,000 L fermented corn cob solution as organic source of potassium is more profitable to use among different rates evaluated.

Recommendations

Farmers can utilize corn cobs from the surrounding corn farm area, and fermentation can be applied using the following steps:

1. Collect corn cobs
2. Soak in a plastic drum:
 - Place the collected corn cobs in a plastic drum.
 - Maintain a ratio of 1 kg of corn cob per 25 L of water.
 - Allow the corn cobs to soak for at least 4 weeks.
3. Add molasses:
 - During soaking, add 3 L of molasses to the solution.
4. Drench and apply to ginger crop:
 - Once the fermentation is complete, drench the solution onto the ginger crop.
 - Apply the solution at a rate of 22,000 L per hectare.
 - Time the applications as follows:
 - 30 Days After Planting (DAP): Apply 7,333 L of FCCS
 - 60 DAP: Apply another 7,333 L of FCCS
 - 90 DAP: Apply a final 7,333 L of FCCS

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A REVIEW OF CLIMATE CHANGE ADAPTATION LITERATURE IN AGRICULTURE: AIMS, THEMES, AND A FOCUS ON DECISION-ORIENTED SCHOLARSHIP

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Abstract

This paper conducts a comprehensive review of literature on climate change adaptation within the agricultural sector. The primary objective of this review is to elucidate the diverse and complex measures farmers employ in response to climate change, which could inform the development of more effective and pertinent policies. The paper commences with definitions of key terms relevant to this research, including ‘adaptation,’ ‘adaptive capacity,’ and ‘vulnerability,’ while also clarifying the interconnections between these concepts. Subsequently, it outlines the objectives and predominant themes within climate change adaptation scholarship pertaining to agriculture and highlights significant research gaps. Finally, it proposes areas for future investigation.

Keywords: *Climate change adaptation, Climate change, Decision-making processes, Climate change in rice, Climate change adaptation review*

Introduction

Agriculture is amongst the worst-impacted sectors by climate change-related drought (Panda et al. 2013). Agricultural drought, defined as the inadequacy of soil moisture needed for plant growth is projected to be more frequent in the climate change regime (IPCC, 2012). In general, drought relates to more frequent occurrences of El Niño (IPCC, 2012). With global warming, scientists believe that the frequency of strong El Niño events will also increase (Cai et al., 2018). Given this scenario, agricultural crops will be significantly affected. Yield decreases due to drought have been reported globally for different crops such as wheat, maize, rice (Wang et al., 2015), and corn (Lindoso et al., 2014).

In rice, there are several documented massive yield losses due to drought. For example, the Chhatisgarh State in India had almost complete crop failure brought about by the 2002 drought (Pandey et al., 2007). In Thailand, the 1998 - 1999 drought resulted in USD 290 million losses for the farming sector. These effects are extremely worrying when considering that rice is being grown in more than 190 million hectares in more than 100 countries (FAOSTAT, 2019). This means millions of people depend on rice either as a source of livelihood or energy. It should also be noted that the majority of rice farmers live in dire poverty (Hilson and Van Bockstael, 2012; Hoang et al., 2015; IPCC, 2012; Salami et al., 2017) and have to deal with multifarious concerns such as dealing with cutthroat arrangements with traders, lack of access to credit and

extension, and debt repayment (Arida, 2009). Climate change-related drought is expected to amplify these existing pressures.

This review aimed to advance decision-oriented scholarship in relation to climate change adaptation in agriculture. This overall direction sets it apart from other reviews such as the one by Grigorieva et al. (2023), which focus on identifying climate adaptive-mechanisms. While identifying climate adaptive mechanisms is practical, there have already been a number of similar studies in the past as will be mentioned in the succeeding sections of this paper. The assumption is that understanding how farmers make adaptation decisions may lead to crafting policies that are relevant and more effective; therefore, may have lasting impacts for resource-poor farmers.

The next section of this review defines climate change adaptation and presents its dominant framing in the agriculture sector (Section 2). It is followed by a discussion of the interrelations between adaptation, vulnerability, and adaptive capacity. The discussion of the goals of climate change adaptation studies follows. After which, the scholarship on decision-making processes in the agriculture sector is presented.

Defining climate change adaptation

Adaptation refers to any “processes people use to reduce the adverse effects of climate on their livelihood and well-being, and take advantage

of new opportunities provided by their changing environment” (Jones, 2010). In this review, adaptation may take the form of rice farmers planting early maturing rice varieties or planting other crops in times of drought, pumping water from the ground, or taking on off farm work to compensate for income loss.

In climate change adaptation, the dominant view is that it is a technical or technological issue (Resurreccion et al., 2008). In the rice sector, this framing is particularly apparent. This is made evident by different research initiatives by the International Rice Research Institute (IRRI) such as breeding drought-resistant rice varieties (Tonini and Cabrera, 2011), and developing water management technologies (Cabangon et al., 2011). Locally, in the Philippines, this same agenda is echoed by PhilRice, which is the lead agency for rice research and development in the Philippines (e.g., Manigbas et al., 2014; Orge et al., 2019; Samoy-Pascual et al., 2021). In PhilRice’s *Strategic Plan (2017 to 2022)* there is a general reference on developing rice varieties that would be able to respond to different climatic stresses (PhilRice, 2016). In the Department of Agriculture in the Philippines, the Adaptation and Mitigation Initiatives in Agriculture Report notes that the first six years of implementation of the Department’s Action Plan on climate change solely focused on promoting organic agriculture. However, the focus on technology fails to understand the complexities of climate change, which is enabled or constrained by social and economic factors (Resurreccion et al., 2008).

Adaptation, adaptive capacity, and vulnerability

The centrality of the social and economic factors can never be overemphasised in the climate change adaptation discourse. In fact, these very same factors figure prominently when discussing about the triumvirate concepts of adaptation, adaptive capacity, and vulnerability relative to the impacts of climate change. Although these three concepts may be interpreted differently across various disciplines or discourses, within the context of climate change, these are closely interrelated (Smit and Wandel, 2006).

There is agreement, with slight variations, amongst authors that adaptive capacity is synonymous with coping ability, resilience, and adaptability (Engle, 2011; Füssel and Klein, 2006; Tompkins and Adger, 2004). A formal definition forwarded by Smit et al. (2000) states that adaptive capacity is the “potential or capability of a system to adapt to (to alter to better suit) climatic stimuli” (pp 239). Adaptive capacity is context-specific. Context is understood with reference to the community that is the focus of

adaptation. For instance, a community of farmers that has access to farmers network or strong support from the household is expected to have higher adaptive capacity than one that does not have it, considering social capital is amongst the determinants of adaptive capacity (Arunrat et al., 2017; Chen et al., 2014). Adaptive capacity is also determined by a broad range of factors such as the level of technology available in the area, economic factors, political influence, and information sources (Hamdy et al., 1998; Handmer et al., 1999; Kelly and Adger, 2000; Tóth, 1999; Watts and Bohle, 1993).

Vulnerability is a concept that has multifarious conceptualisations. Adger (1999), in emphasising the social aspects of climate change, advances the concept of Social Vulnerability. He refers to it as “the exposure of groups or individuals to stress as a result of the impacts of climate change and related climate extremes” (pp 252). A key feature of his work is the differentiation of individual and collective vulnerabilities. Individual vulnerabilities may be determined by a person’s wealth or economic status in general while collective vulnerability is determined by inequality and institutional arrangements. Inequality can have some positive outcomes. An example given by Adger (1999) is if a wealthy man finances the operations of an irrigation system that benefits not just his farm but also that of others, then that is something good. Hence, what should be investigated is not just inequality per se but the nature of inequality, i.e., if it constrains people to adapt or to reduce their vulnerability.

There are two dominant views on vulnerability. First, is the view that the poor are the most vulnerable (Adger, 1999; Apotsos et al., 2016; Beck, 1992). In the context of this paper, this view is understood in terms of the inability to have access to resources that will help cushion the impacts of climate change on livelihoods. The second view is that vulnerability [to climate change impacts] is socially differentiated (Adger, 1999; Adger, 2001, 2003; McNeeley and Lazrus, 2014). This means that the vulnerability of a community will differ depending on social context. This view is similar to the one forwarded by the United Nations Framework Convention on Climate Change (UNFCCC) and the Intergovernmental Panel on Climate Change (IPCC). One interesting view in relation to the social differentiation of climate change impacts is that it is more important to look at the vulnerability of poor communities, not of poor countries (Kates, 2000). This is not difficult to understand as, for instance, the interests of a poor country as a whole do not always represent the interests of its poor communities. There certainly are many social and political dynamics within a poor country that may not always favour the interests of its poor communities.

Having explained what adaptation, adaptive capacity, and vulnerability mean, the discussion turns to explaining their interrelatedness. Smit and Wandel (2006,) explain: “adaptations are manifestations of adaptive capacity, and they represent ways of reducing vulnerability.” For example, in the context of drought-prone rice-farming communities, a community with access to irrigation and drought-tolerant rice varieties (examples of indicators of high adaptive capacity), means that they have a fighting stance against drought. Hence, it follows that this community may be able to adapt to the impacts of drought on their livelihood. This means as well that the community’s vulnerability to drought impacts is greatly reduced because of its high adaptive capacity. In general, high adaptive capacity means reduced vulnerability, which, consequently, means high chance of adapting to climate change impacts.

Goals and major themes in climate change adaptation research

Goals of climate change adaptation research

According to Smit and Wandel (2006) there are four major aims of climate change adaptation studies. The first are studies that identify adaptation options to the modelled impacts of climate change (e.g., Ceci et al., 2021; Cobon et al., 2016; Harrison et al., 2015; Kelkar et al., 2008; Laureta et al., 2021). Downing (2012) refers to this group of scholarship as the “what if” type of studies. These studies are hypothetical in nature and do not investigate adaptation empirically. The second are studies that identify options or measures for a certain system; the usual output is a “suite of possible adaptations” (Smit and Wandel, 2006). These suites of possible adaptations are then evaluated using different criteria such as cost, doability, and effectiveness (e.g., Ame et al., 2014; Georgopoulou et al., 2017; Nigussie et al., 2018). The third group of studies focuses on evaluating the relative adaptive capacity and vulnerability of countries to the impacts of climate change. Usually, the route of analysis is to compare countries, regions, or communities using a set of criteria or indices (e.g., Acosta-Michlik and Espaldon, 2008; Almaden et al., 2020; Varadan and Kumar, 2015; Varela et al., 2022; Weis et al., 2016).

The fourth group of studies focuses on practical forms of adaptation. The difference of this group of scholarship with the second is that this group tackles decision-making processes; the second group does not. Specifically, this fourth group of studies aims to capture adaptive decision-making processes in relation to the changing conditions in their respective communities. This fourth goal has received much less research attention. Fujisawa et al. (2015) note that adaptation decision-making processes of

farmers to climate change are “poorly understood” (p 1) although it should be noted that there is an increasing scholarship on this focus in recent years (e.g., Alauddin and Sarker, 2014; Comoe and Siegrist, 2015; Khanal et al., 2018). As will be argued further below, decision-making processes of farmers are not well understood and there is a longstanding call for more decision-oriented scholarship in climate change adaptation research (Willows et al., 2003).

Themes in climate change adaptation research in agriculture

Following the section above, which focuses on the general goals of climate change adaptation scholarship, this section discusses the themes of climate change adaptation studies specific to the agriculture sector. There are four major themes that can be noted. The first are studies that deal with the perceptions, attitudes, and knowledge of farmers in relation to climate change and climate change adaptation (Onyeneke et al., 2021; Sánchez-Cortés and Chavero, 2011; Soriano et al., 2017). This body of scholarship looks at how climate change impacts on the livelihood of farmers based on their experiences. Typically, the investigation involves asking farmers to narrate the changes in the usual weather pattern in the past X number of years. These observations will then be compared with records from the weather bureau (e.g., Evangelista et al., 2016). Amongst the key findings of these studies are associations of climate change to increases in temperature, increase in rainfall variability, and increase in the occurrence of extreme climatic events such as drought and flood (Kelkar et al., 2008; Lasco et al., 2016; Thomas et al., 2007; Uy et al., 2011).

Another group of scholarship focuses on maladaptation and barriers to adaptation. In the rice sector, the study of Warner et al. (2015) is the only one found to have dealt with barriers to adaptation. The authors interrogated the complexities surrounding farmers’ decision to solely plant rice and ignore other crops. Rice forms a significant part of the farmers’ culture and identity. Hence, the authors recommended that future studies may consider looking into identity as a barrier to climate change adaptation, i.e., to what extent are the farmers willing to negotiate their identities relative to climate change adaptation. Outside the rice sector, Nielsen and Reenberg (2010) note how culture hinders community members in a village in Burkina Faso from practising the four key livelihood strategies that would help them adapt to drought: “labour migration, working for development projects, gardening, and the engagement of women in economic activities” (pp 142). In the same article, Nielsen and Reenberg (2010) report that usually at the local level, there is an increasing recognition of the influence of factors such as “class, gender, and

culture” (pp 142) on the decision to accept or reject an adaptive practice.

In the Philippines, there are some studies that tackle barriers to adaptation (Cuevas, 2017; Peñalba et al., 2012; Ravago et al., 2017). Amongst these studies is the one by Peñalba et al. (2012) in which they documented that local communities have an attitude of dependence that the government or a “supreme being” will provide. This finding on the belief of “supreme being” was not elaborated, but in Philippine studies this can be equated to religiosity. If seen in this context, Peñalba et al. (2012)’s finding stands in stark contrast with the many Philippine studies on climate change that have documented “praying” (Acosta-Michlik and Espaldon, 2008) or belief in God (Evangelista et al., 2016) as amongst the key factors that enhance adaptive capacities of Filipino farmers to climate change. As for scholarship on maladaptation, there are studies that scrutinise how combinations of adaptive practice result in increased emissions. That is, it results in worsening the greenhouse gas emissions issue (e.g., Tan, 2013). Another study documents the potential social conflict issues in taking on an adaptive practice. Snorek et al. (2014) document how one’s adaptive practice reduces the adaptive capacity of another, a phenomenon which they call “divergent adaptation” (pp 371). The case study shows how expansion of land area for agriculture reduces adaptation in pastoral areas.

The third type of scholarship pertains to studies that tackle factors that enhance adaptive capacity of farmers to climate change (Asfaw et al., 2016; Below et al., 2012; Saroar and Routray, 2015). This theme is widely explored both at the global and local levels. Amongst the major work under this theme are studies on social capital (with contrasting findings e.g., see Chen et al. (2014) and Paul et al. (2016); and institutions (Goldman and Riosmena, 2013; Yates, 2012).

In general work on social capital gravitates around how farmers benefit from their membership in social groups such as in agricultural associations (Below et al., 2012) and accessing information on adaptation measures (Chen et al., 2014). Scholars report the highly positive impact of having strong social capital on enhancing adaptive capacity of farmers with a few exceptions such as the work of Paul et al. (2016). These authors weigh in on the overall impact of social capital in enhancing adaptation. One of Paul et al.’s (2016) conclusions is that whilst social capital is good at fuelling up community action, it may also be detrimental to private adaptation. For instance, the government dictating activities to adapt to climate might alter patterns of private adaptation. They also

raised the point that there is a possibility that those who may be able to rely on their communities for certain adaptive practices may not be enticed to take on individual adaptive practices. On the other hand, studies on institutions focus on the importance of having strong and reliable institutions that can help farmers cope with the impacts of climate change. Amongst the roles that formal institutions can contribute in enhancing adaptive capacity are helping facilitate social learning processes (Raymond and Robinson, 2013) and developing and promoting adaptation measures (Wang et al., 2013). Some scholars also highlight that institutions may also constrain adaptation (Wise et al., 2014) such as by way of controlling adaptation projects and the flow of information resulting in uneven interventions amongst communities needing assistance (Yates, 2012).

The fourth type are those that deal with the determinants of adaptation and/or the intent to adapt. These studies use quantitative survey methods to test for the relationship between determinants and adaptation outcomes (Le Dang et al., 2014; Masud et al., 2017; Truelove et al., 2015). This type of studies is discussed in more detail in the succeeding section.

The discussion above shows that there is an impressive breadth of scholarship available on climate change adaptation in the agriculture sector. In hindsight, a closer scrutiny of the literature would lead one to conclude that decision-making processes of farmers are an uncharted territory in climate change adaptation research. This is not to say, however, that there are no studies on decision-making processes in this area. There is, in fact, a growing body of scholarship in this area in recent years, which will be discussed in the next section. The term decision-making process, in this paper, refers to how decisions are arrived at in making adaptive or non-adaptive choices. This term also pays attention to the complexities and dynamics of the process (Downing, 2012) brought about by different interacting elements such as farmers’ values (O’Brien and Wolf, 2010).

Decision-making process also refers to how farmers negotiate aspects of their lives as farmers such as their practices and overall knowledge in farming as they encounter climate change-related drought (Eriksen et al., 2015). At the level of policy, understanding decision-making processes can substantively inform programs aimed at assisting farmers to cope with climate change. Theoretically, understanding decision-making processes widens and deepens discourses on climate change

adaptation as it raises questions that interrogate how decisions are made at different levels.

Zeroing in on adaptation decision-making scholarship in the agriculture sector

As noted above, in the agriculture sector, there is a growing focus on scholarship on decision-making processes in relation to climate change adaptation in recent years. In the general climate change adaptation literature, however, focus on decision-making processes had taken off more than a decade ago (e.g., Grothmann and Patt, 2005).

At this point, it might be good to ask: why is there a need to bring in decision-oriented studies in the climate change adaptation discourse? First it should be noted that in the climate change adaptation discourse in the agriculture sector, there is dominance of the economics and technological lenses. Whilst they are both useful and have shown their robustness in understanding adaptation, there remain some gaps that need to be addressed. For instance, the economic lens may have difficulty explaining how farmers refused to take on more profitable adaptation options. Another example is the study where farmers refused to plant other crops aside from rice even though rice proved to be non-economical because of drought - how can the technological lens explain this? It is argued in this paper that bringing in a new lens such as one on decision-making may help explain these gaps.

Another reason is that there is a need for climate change studies to have an instrumental aim, and that is to explore ways on how to better assist rice farmers in the event of drought. With that aim, it pays to note that adaptation is about making choices and making choices is all about making decisions. In the agriculture sector, this can take the form of deciding to postpone or not cultivating a particular crop such as rice, use technologies such as a drought-tolerant rice variety, or take on livelihood outside the agriculture domain. Scrutinising the ways in which farmers decide on their adaptive (or non-adaptive) options is a step closer to coming up with interventions that are both relevant and impactful.

Building on from the reasons cited above, decision-oriented studies on climate change adaptation are important with respect to the shift in research priorities within the climate change domain. Because of the increasing likelihood of 2°C global warming, the focus of research has shifted from mitigation to adaptation (Wise et al., 2014). In the context of rice farming, this becomes crucially important as rice is extremely sensitive to climatic changes. For instance, beyond 35°C, chance of spikelet sterility is high, which means that there will be unfilled grains, and, eventually, this will be translated to low yield.

Consequently, this may worsen the poverty situation in rice-farming communities, i.e., from transient poverty to chronic poverty (Morduch, 1994) in some cases.

Considering the shift from mitigation to adaptation, it pays to highlight that within adaptation scholarship, there has also been a shift as to how it is viewed. Before the dominant view was the predict-and-provide approach, i.e., “adaptation is the provision for a prediction of future impacts” (Downing, 2012). This view never suffices in understanding dynamic processes such as adaptation. Hence, the way adaptation is viewed has shifted to the emphasis on highlighting its being dynamic and complex. This complexity and dynamism are due to multiple perspectives and value systems of entities involved. Values, for instance, are key to determining what is “worth preserving and achieving” when people think of what successful adaptation means (O’Brien and Wolf, 2010).

The studies on farmers’ decision to adapt are drawn from different disciplines, and in this section, they are grouped into two. There is the research on determinants of adaptation and preferred adaptive practices (e.g., Arunrat et al., 2017; Khanal et al., 2018). Then there is the psychological approach that looks at intentions and highlights that intentions do not always translate into actions (e.g., Le Dang et al., 2014; Zeweld et al., 2017).

The first group of scholarship has thus far identified factors that determine adaptation and preferred adaptive practices (e.g., Alauddin and Sarker, 2014); identified combinations of factors that can facilitate adaptation (e.g., Arunrat et al., 2017); presented adaptation scenarios based on relationships of variables and on some assumptions (e.g., Khanal et al., 2018). In general, these types of studies are useful for rapid assessments that are oftentimes useful in policymaking.

The second group of scholarship also touches on some of the themes covered by the first but they are more focused on bringing in psychological (Le Dang et al., 2014) and socio-psychological (Truelove et al., 2015; Zeweld et al., 2017) lenses in looking at adaptation. Under this group, there are plenty of disagreements noted. For instance, the studies of Zeweld et al. (2017) and Niles et al. (2016) disagree on the view concerning how intent to adapt translates to actual practice. Zeweld et al. (2017) note that carrying out the intention is expected to follow once intent has formed. Niles et al. (2016) argue that this is not always the case, and that there is a “disconnect” between intent and actual behaviour (pp 278). Some differences on the degree of importance accorded to risk perception and its relationship to the likelihood

of having some intent to adapt are noted between Le Dang et al. (2014) and Truelove et al. (2015). Le Dang et al. (2014, p 11) emphasise that farmers who “perceive higher risks of climate change” are more likely to have some intention to adapt. For Truelove et al. (2015), risk perception is also important, but it is just among the secondary predictors of adaptation intent.

The most important are beliefs relating to efficacy. Le Dang et al. (2014) also stress that farmers are likely to adapt if there is pressure from others. This body of scholarship has strong roots in psychology. Amongst its key achievements in relation to decisions on climate change adaptation are its capacity to predict intent to adapt using certain parameters and models (e.g., socio-psychological model in Truelove et al. (2015) and identify factors that constrain adaptation decisions (Truelove et al., 2015). Additionally, these studies have shown their usefulness in identifying some characteristics of those who are likely to adapt (e.g., Le Dang et al., 2014) to climate change, in looking at the effects of some interventions with respect to the likelihood of a farmer to take on adaptive behaviour (e.g., Le Dang et al., 2014), and in estimating support by farmers (e.g., willingness to pay) for adaptation mechanisms that are being proposed (Masud et al., 2017).

Carefully scrutinising the scholarship on decision-making presented above, the gap is on studies that deal with the processes of decision-making. That is, how does one move from one decision to another. The complexities of decision-making remain inadequately covered. It should also be noted that none of the studies mentioned above focused on rice-farming communities. In this paper I make a case for rice-farming communities given the relative importance of rice, both as a source of energy to more than half of the world’s population and as a source of livelihood for millions of rice farmers.

Conclusion

This paper presented an extensive review of literature on climate change adaptation. Amongst the major observations is that the climate change adaptation scholarship in the agriculture sector is dominated by studies that use the economic and technological lenses. The paucity of scholarship on the complexities of decision-making processes has been highlighted. It has been argued that a focus on decision-making scholarship in the climate change adaptation in the agriculture sector is a step closer to realising policies that may better assist farmers to cope with drought.

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