

Volume 12 • Issue 2 • August 2024





PhilippineRiceResearch Institute Central Experiment Station Maligaya, Science City of Muñoz, 3119 Nueva Edja



ABOUT THE COVER

This issue delves into the crucial role of nutrient application, encompassing macro and trace elements, in enhancing crop yields. It investigates innovative farming methods aimed at increasing farmer profitability, while prioritizing food safety. Expect to explore a spectrum of agricultural advancements that not only contribute to higher yields and financial success but also ensure the safety of food consumption.

Rice-based Biosystems Journal

A bi-annual publication of Philippine Rice Research Institute

ISSN 2960-3692

Instructions on manuscript submission are found on pages 95-99. For more information, please visit www.philrice.gov.ph/grants/call-publication/.



VOLUME 12 · ISSUE 2 · AUGUST 2024

Published by:



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

EDITORIAL TEAM

EDITOR-IN-CHIEF

NORVIE L. MANIGBAS

Scientist III and Chief Science Research Specialist, PhilRice

 $nlmanigbas@philrice.gov.ph \bullet norviem@yahoo.com \bullet norviemanigbas@gmail.com$

EDITORIAL BOARD

CHARISMA LOVE B. GADO-GONZALES Editor, PhilRice

CONSORCIA E. REAÑO Statistical Editor, University of the Philippines Los Baños (UPLB)

EDITORIAL BOARD

JOHN C. DE LEON Chair, Plant Breeder and Executive Director, PhilRice

VETHAIYA T. BALASUBRAMANIAN Agronomist and Soil Scientist, Tami Nadu, India

CALIXTO M. PROTACIO Professor, Institute of Crop Science, College of Agriculture and Food Science, UPLB

EUFEMIO T. RASCO JR. Academician, National Academy of Science and Technology (NAST)

RODOLFO C. UNDAN Professor of Agricultural Engineering and former President, Central Luzon State University (CLSU)

> LUIS REY I. VELASCO Professor of Entomology, UPLB

RUBEN L. VILLAREAL Academician, NAST

MARILOU J. ANG-LOPEZ Assistant Professor VII, School of Technology, University of the Philippines Visayas (UPV)

SANKALP U. BHOSALE Deputy Platform Leader Rice Breeding Innovations Lead, Product Development & Varietal replacement International Rice Research Institute

ROLANDO T. CRUZ Agronomist, Los Baños, Laguna. Philippines

EVELYN F. DELFIN Agronomist, Los Baños, Laguna. Philippines GLENN B. GREGORIO Professor Institute of Crop Science College of Agriculture and Food Science, UP Los Baños Center Director Southeast Asian Regional Center Graduate Study and Research in Agriculture (SEAMEO-SEARCA) Los Baños, Laguna, Philippines

JOSE E. HERNANDEZ Professor, Institute of Crop Science, College of Agriculture and Food Science, UPLB

Academician, NAST

YOICHIRO KATO Assistant Professor Faculty of Agriculture, The University of Tokyo, Tokyo, Japan

TANGUY LAFARGE Crop Physiologist, Centre de cooperation internationale en recherche agronomique pour le developpement (CIRAD), Montpellier, France

> FELINO P. LANSIGAN Professor of Statistics and Dean, College of Arts and Sciences, UPLB

> > **CEZAR P. MAMARIL** Soil Scientist and Rice Farmer

MILAGROS R. MARTINEZ-GOSS Professor Emeritus, Institute of Biological Sciences, UPLB

> TEODORO C. MENDOZA Professor, Institute of Crop Science, College of Agriculture and Food Science, UPLB

> > AGNES C. ROLA Academician, NAST and Professor, College of Public Affairs, UPLB

INVITED REVIEWERS

ROSALY V. MANAOIS Scientist I and Senior Science Research Specialist, PhilRice

RICARDO F. ORGE Scientist III and Chief Science Specialist, PhilRice, Retired

ELMER G. BAUTISTA Scientist I and Supervising Science Research Specialist, PhilRice

> **EVELYN M. VALDEZ** Senior Science Research Specialist, PhilRice

NENITA V. DESAMERO

Chief Science Research Specialist, PhilRice

PRODUCTION TEAM

JEREMHACE V. DEL PILAR Editorial Assistant, PhilRice

> CARLO G. DACUMOS Layout and Design, PhilRice

> > STANLEY OMAR PB. SAMONTE Associate Professor in Rice Breeding Texas A&M AgriLife Research, USA

GENARO O. SAN VALENTIN Soil Scientist and Senior Consultant, PhilRice

POMPE C. STA CRUZ Director and Professor, Institute of Crop Science, College of Agriculture and Food Science, UPLB

BERNARDO D. TADEO President and CEO, Full Advantage Phils. International, Inc.

EMMANUEL R. TIONGCO Plant Pathologist and Senior Consultant, PhilRice

GURDEV S. KHUSH Member, US National Academy of Sciences Adjunct Professor Emeritus University of California-Davis Former Head, Plant Breeding, Genetics, and Biotechnology International Rice Research Institute

LEE TARPLEY Professor, Plant Physiology Texas A&M AgriLife Research Center at Beaumont

MAYUMI YOSHIMOTO Senior Principal Scientist Climate Change Adaptation Research Institute for Agro-environmental Sciences National Agriculture and Food Research Organization

> **GIHWAN YI** PROFESSOR Director, Department of Agro-Industry Kyungpook National University

JAYVEE C. KITMA Scientist I and Senior Science Research Specialist, PhilRice

RIZA A. RAMOS Scientist I and Supervising Science Research Specialist, PhilRice

JESUSA C. BELTRAN Scientist I and Chief Science Research Specialist, PhilRice

MANUEL JOSE C. REGALADO Scientist II and Chief Science Research Specialist, PhilRice, Retired

> NORVIE L. MANIGBAS Scientist III and Chief Science Specialist. PhilRice



VOLUME 12, ISSUE 2, AUGUST 2024

CONTENTS

FULL PAPER

PERFORMANCE OF OYSTER MUSHROOM (PLEUROTUS SPP.) GROWN
IDENTIFICATION AND <i>In-vitro</i> EVALUATION OF PLANT GROWTH-PROMOTING RHIZOBACTERIA
ERGONOMICALLY DESIGNED BROWN RICE MACHINE: ADVANCING GENDER-INCLUSIVE
SILICON DEPOSITION IN RICE PLANT AS AFFECTED BY SILICATE-SOLUBILIZING BACTERIA
ASSESSMENT OF PESTICIDE USAGE AND ORGANOPHOSPHATE PESTICIDE RESIDUE
EFFECTS OF GENOTYPE - ENVIRONMENT INTERACTION ON GRAINVIELD IN HYBRID RICE LINES
POTENTIAL INFLUENCE OF NITROGEN - SILICON FERTILIZATION ON IMPROVING
RESEARCH NOTE
COMPARATIVE STUDY ON PRODUCTIVITY AND PROFITABILITY OF INBRED
BLOCKCHAIN: A DIGITAL PLATFORM DATABASE FOR RICE SCIENCE 79 Eduardo Jimmy P. Quilang, Hazel B. Alfon, Ritesh Amin, Raymond Josef Edward A. Lara, Arturo C. Arocena, and Luis Alejandre I. Tamani

SPECIAL REVIEW ARTICLE

APPLICATIONS OF PARTICIPATORY DEVELOPMENT COMMUNICATION IN THE AGRICULTURE
SECTOR FROM 2013 TO 2023
VENUS B. TAJON, JAIME A. MANALO IV, JAY C. SANTOS, CHRSYL AVEGEIL N. VALLEJO, MARIFE DE TORRES,

VENUS B. TAJON, JAIME A. MIANALO IV, JAY C. SANTOS, CHRSyl AVEGEIL N. REGIDOR G. GABOY, PARSONS N. HAIL

PERFORMANCE OF OYSTER MUSHROOM (PLEUROTUS SPP.) GROWN IN DIFFERENT SUBSTRATE ADDED WITH NITROGEN-RICH PLANTS

RICHARD R. GEGANZO, FRANCIS G. CAPINO, CHESTER IAN TALIDANO, AND ALJEN P. VASQUEZ*

Department of Agriculture Region VI- Research Division, Buntatala, Jaro, Iloilo City *Corresponding Author: aljsvasquez@gmail.com

Abstract

The growing popularity of mushroom production in the Philippines and globally is driven by its social, economic, and environmental benefits. While leguminous crop residues are typically used as organic fertilizers, their potential as substrate enhancers for mushroom cultivation, especially when integrated with rice straw, remains underexplored. Rice straw, an abundant agricultural by-product of rice farming in the country, offers a sustainable base material for mushroom production. However, it is relatively low in nitrogen, a critical element for optimal mushroom growth. This study was conducted to address the increasing demand for mushrooms by enhancing productivity and nutritional quality through the integration of nitrogen-rich leguminous crop residues with rice straw-based growing media. Conducted at the Department of Agriculture - Western Visayas Integrated Agricultural Research Center, the study evaluated the performance of white (Pleurotus ostreatus) and gray (Pleurotus sajor-caju) oyster mushrooms cultivated in rices straw combined with nitrogenous plant residues. Five substrate combinations were tested: rice straw + sawdust (RS+SD), rice straw + soybean straw (RS+SBS), rice straw + peanut straw (RS+PS), rice straw + mungbean straw (RS+MS), and rice straw + ipil-ipil leaves (RS+IpL), all in a 70:30 ratio. Spawns were inoculated under aseptic conditions, and mushroom growth parameters were monitored. All rice straw-based combinations supported mycelial development, but RS+SBS and RS+PS resulted in the fastest colonization and highest yields. Nutrient analysis revealed that enriching rice straw with leguminous residues increased nitrogen content in the substrates and produced fruiting bodies with higher crude protein. The study strongly recommends using rice straw integrated with nitrogen-rich materials to optimize oyster mushroom yield and nutrition while promoting circular use of rice-based farm waste.

Keywords: rice straw, nitrogen-rich plants, oyster mushroom, mycelium, substrate performance

Introduction

Mushroom production has long been an industry in the Philippines and abroad. This technology gained popularity among farmers and food enthusiasts due to its social, economic, and environmental impact (Andam and Dulay, 2014). In the Philippines, most mushrooms consumed are imported from countries in Southeast Asia such as China, Taiwan, Thailand, Malaysia, Korea, and Japan. Despite this, mushroom cultivation remains economically viable in the country due to low production costs, the availability of inexpensive substrates derived from agricultural waste, particularly rice straw, and the high market demand. These factors make mushroom cultivation both profitable and feasible, presenting a valuable opportunity for local farmers.

Among the available agricultural residues, rice straw stands out as one of the most commonly used substrates for oyster mushroom (*Pleurotus spp.*) cultivation in rice-producing regions. However, rice straw is relatively low in nitrogen content, which can limit mushroom growth and yield. To address this constraint, researchers have begun incorporating nitrogen-rich materials, particularly leguminous crop residues, into rice straw-based substrates. This practice has the potential to enhance the nitrogen profile of the growing media and support higher yields and nutritional quality of mushroom fruiting bodies.

Mushrooms present a balanced nutritional composition (Rashidi and Yang, 2016) and are considered an attractive alternative source of highquality protein with essential and non-essential amino acids (AA). All amino acids needed by the human body are present in edible mushrooms (Bach et al., 2017). The nutrient content of food is primarily determined by its protein and carbohydrate (or fat) content. Edible mushrooms are highly nutritious and can be compared to eggs, milk, and meat (Oei, 2003). Pleurotus ostreatus in particular contains numerous essential amino acids such as methionine, isoleucine, lysine, and glutamic acid. It is also rich in physiologically meaningful primary and secondary metabolites and chemical elements. One hundred grams of fresh fruiting bodies contain 15% of the recommended daily intake of vitamin C, 40% of niacin, riboflavin, and thiamine, and 0.5 mg of vitamin B12. This species also contains a high level of oleic acid (40%) and linolenic acid (55%) (Piska et al., 2017).

Cultural practices play a crucial role in achieving efficient mushroom production, leading to higher yields and increased profitability. A substrate is any substance that can facilitate mycelial growth. In the tropical Philippine setting where rice is a major staple and rice straw is abundantly available, strategic substrate enhancement is key to improving mushroom production outcomes. Therefore, it is essential to consider combinations of rice straw with other locally available materials that can increase substrate nitrogen content while reducing input costs and maximizing yield.

High concentrations of carbohydrate and nitrogen sources are usually needed to achieve high-yielding mushrooms (Belewu and Lawal, 2003). While rice straw provides an excellent carbon source, it is often deficient in nitrogen. On the other hand, residues of leguminous crops such as soybean, mungbean, peanut, and ipil-ipil are known to have higher nitrogen content than cereal straws (Lopez et al., 2005). These residues are typically used as organic fertilizers to amend soils, but their potential as nutrient-rich additives to rice straw-based substrates for mushroom cultivation remains underexplored.

Given the growing demand for mushrooms in the Philippines (Chang et al., 2014), there is an urgent need to sustainably increase productivity by utilizing locally available agricultural by-products and rice farming residues. This study was conducted specifically to evaluate the yield and nutritional profile of different *Pleurotus spp.* grown on rice straw-based substrates incorporated with nitrogenrich leguminous plant materials. It further aimed to assess the performance of these substrates in terms of mushroom growth parameters, substrate and fruiting body nutrient content, and economic viability.

Several studies support the viability of nitrogenrich crop residues in mushroom substrate enhancement. Salami et al. (2017) evaluated Pleurotus florida grown on corncob (CC) mixed with soybean shell (SYB), rice bran (RB), and groundnut shell (GS). The combination CC+SYB supported the highest yield and crude protein content of mushrooms, demonstrating of the benefit nitrogen supplementation. Musara et al. (2017) showed that mixing cereal straw (maize or sorghum) with legume straw (bean) improved the biological efficiency of Pleurotus spp., with maize straw + bean straw yielding up to 83% efficiency.

In another related study, Mamiro and Mamiro (2011) assessed two crops of *Pleurotus ostreatus* grown on rice straw as the base substrate. In crop I, rice straw was mixed with various proportions of

banana leaves, *Leucaena leucocephala*, maize bran, or maize cobs. In crop II, rice straw was supplemented with sunflower or cotton seedcake. A 50/50 mixture of rice straw and banana leaves yielded 1,040 g with a biological efficiency (BE) of 98.5%. Supplementation with 2% sunflower or cotton seed hulls achieved yields over 1,070 g and BE exceeding 101%. Notably, non-supplemented rice straw produced the largest mushrooms (21.0 g), although with lower yield efficiency. These findings reinforce the potential of rice straw as a base substrate, and the need for strategic nitrogen enrichment.

In this context, the study aimed to explore how the integration of nitrogen-rich leguminous plant residues into rice straw substrates affects the growth, yield, and protein content of white and gray oyster mushrooms (*Pleurotus ostreatus* and *Pleurotus sajor-caju*). By focusing on rice straw as a foundational component, this study aligns with the goal of maximizing the value of rice by-products for enhanced mushroom production in the Philippines.

Materials and Methods

Materials

The study was conducted at the on-station mushroom production facility of the Department of Agriculture - Western Visayas Integrated Agricultural Research Center (DA-WESVIARC) in Brgy. Buntatala, Jaro, Iloilo City, Philippines, from January 2021 to December 2022. All materials were sourced locally. Substrate materials for the fruiting bags included rice straw, sawdust, ipil-ipil (Leucaena leucocephala) foliage, soybean straw, peanut straw, and mungbean straw. Materials for the preparation and pasteurization of fruiting bags included polypropylene (PP) plastic bags (6 x 12", 0.02 mm thickness), cotton, rubber bands, PVC pipes, manila paper, plastic drums (for substrate soaking), and a metal drum (for fruiting bag sterilization). Additional materials used to facilitate the research included plastic containers/crates, a record book, a digital weighing scale, a digital camera, plastic twine, ballpoint pens, markers, and either a shredder machine or a bolo for material preparation.

Substrate Preparation and Proximate Analysis: Rice straw, peanut straw, soybean straw, mungbean straw, and ipil-ipil leaves were collected from nearby municipalities. These materials were air-dried and allowed to partially decompose before being shredded into smaller pieces using a shredding machine. The shredded substrates were then soaked overnight in clean water within plastic drums. Following soaking, the substrates were air-dried to achieve a target moisture content of 65-75%, which was verified using a moisture meter. The prepared substrates were then bagged into polypropylene plastic bags, with each bag containing 750 g of substrate. The following substrate combinations were prepared: rice straw + sawdust (70:30 ratio), rice straw + soybean straw (70:30 ratio), rice straw + peanut straw (70:30 ratio), rice straw + mungbean straw (70:30 ratio), and rice straw + ipilipil leaves (70:30 ratio). Fruiting bags were sterilized once in a metal drum for 8h and allowed to cool prior to inoculation.

For proximate analysis, 1 kg of dried samples from each substrate combination was submitted to the Department of Agriculture – Integrated Laboratory Division, Feed Chemical Analysis Laboratory. The nitrogen content and crude protein content of the substrates were determined using the Kjeldahl method (Kjeldahl, 1883). The specific formula used for calculation was:

 $N = \frac{14.01 \ x \ (Titre \ value \ mls-blank \ value \ mls)}{Sample \ weight \ used \ x \ 10}$

Spawn inoculation and Incubation: Grain spawn of various oyster mushroom species was acquired from WESVIARC's on-station mushroom production facility. Sterilized fruiting bags were aseptically inoculated, each receiving approximately one teaspoon of high-quality white and gray oyster mushroom spawn. The inoculation chamber was disinfected with 70% alcohol, and all inoculating tools were flame-sterilized to prevent contamination. Fruiting bags were labeled according to their treatment combination and then transferred to an incubation room, ensuring no direct sunlight exposure. The optimal temperature for mycelial colonization (spawn running) ranges 22-26°C (Ladli, 2020). The number of days required for complete mycelial ramification was recorded for each fruiting bag during incubation.

Fruiting Bag Stacking and Management: Upon complete colonization by the mycelium, the fruiting bags were moved to the growing area. These were suspended vertically using nylon twine, with 10 fruiting bags per line representing each treatment. Slits were made at the upper portion of each bag using a sterile knife to facilitate mushroom growth. In this study, 300 fruiting bags were used. A thermometer and hygrometer were used to continuously monitor and maintain the temperature and relative humidity within the fruiting house. Ideal fruiting conditions require a temperature range of 20-30°C and a relative humidity of 75-85%. Regular misting with potable water was performed to help lower the temperature and increase relative humidity.

Harvesting of Mushroom. Oyster mushrooms were harvested when the margins of their caps were

nearly flattened. Harvesting typically occurred 2-3 days after the emergence of pinheads. Mushrooms were weighed according to their respective treatments across three flushes. After the third flush, the total yield and biological efficiency were determined for each treatment.

Chemical Analysis of Mushroom Fruiting bodies. Freshly harvested mushroom fruiting bodies were dried and processed into a powdered form. These powdered samples were then submitted to the Department of Agriculture – Integrated Laboratory Division, Feed Chemical Analysis Laboratory, for nutrient analysis. Specifically, treatment samples underwent proximate analysis to ascertain their crude protein, crude fiber, crude fat, moisture, and ash content when grown on different nitrogen-rich substrates.

Statistical Tool. The study employed a two-way factorial Randomized Complete Block Design (RCBD). Factor A consisted of two different species of oyster mushrooms, while Factor B comprised five distinct substrate combinations. Each treatment combination was replicated three times, with each experimental unit consisting of 10 fruiting bags, totaling 300 fruiting bags for the entire study.

Results and Discussion

The study used a factorial ANOVA to evaluate the effects of incorporating different nitrogen-rich leguminous plants (70% rice straw + 30% nitrogen-rich plants) into the substrate of white and gray oyster mushrooms. The analysis focused on the number of days for mycelial ramification, pinhead formation, survival rate, yield, and biological efficiency.

Number of days of mycelia ramification. Good development of white, cottony mycelium was observed in all substrate combinations for both oyster mushroom species. The survival rate (%) recorded during the incubation period showed no significant difference across substrate combinations (p = 0.43). Similarly, there was no significant difference between white and gray oyster mushrooms, with white oyster recording M = 100% (SD = 0) and gray oyster M = 98.9% (SD = 0.4) (p = 0.33).

However, the number of days for mycelial ramification was significantly different between the white (M = 27.03 days) and gray (M = 30.9 days) oyster mushroom species (p < 0.01). Substrate combinations RS+SBS (M = 27.5), RS+PS (M = 27.9), and RS+MS (M = 27.3) resulted in significantly shorter mycelial colonization periods compared to RS+SD (M = 30.4) and RS+IpL (M = 31.8) (p < 0.001). No significant interaction effect was found between substrate and species (p = 0.06), indicating that the influence of substrate and mushroom species on mycelial development is independent.

All substrate treatments supported mycelial growth, but substrates with leguminous plants especially soybean straw, peanut straw, and mungbean straw, significantly reduced colonization time, with the exception of ipil-ipil foliage (Table 1). This supports the findings of Salami et al. (2017) who highlighted the benefits of organic nitrogen supplementation. According to Nunes et al. (2012), fungi absorb organic nitrogen more efficiently than synthesizing it, conserving energy for growth and fruiting.

Although ipil-ipil foliage is nitrogen-rich (Figure 2), excessive inclusion (beyond 20%) can hinder mycelial growth, as supported by Andrew (2023). In this study, the 30% inclusion of ipil-ipil resulted in the longest colonization period, likely due to excessive nitrogen. As reported by Hassan et al. (2010), mycelial growth largely depends on substrate composition and growing conditions. Since environmental conditions were uniform across treatments, substrate composition is likely the main factor in observed differences.

Number of days of pinhead formation. Pinhead formation refers to the initial emergence of mushroom fruiting bodies. Results (Table 2) show that white

oyster mushrooms formed pinheads significantly faster (M = 34.5 days) than gray oysters (M = 42.8 days) (p < 0.001). Substrate combinations RS+SBS (M = 35.7), RS+PS (M = 35.9), and RS+MS (M = 34.4) led to significantly earlier pinhead formation compared to RS+SD (M = 43.2) and RS+IpL (M = 43.3) (p < 0.001).

The interaction between substrate and species was not significant (p = 0.205), indicating no combined effect. Substrates enriched with soybean straw, peanut straw, and mungbean straw promoted earlier pinhead formation than the control and ipil-ipil foliage. On average, pinhead formation occurred 8–12 days after full mycelial colonization.

This contrasts with Bhattacharjya et al. (2014), who reported 6-8 days for pinhead initiation. Such variation could be attributed to differences in species, substrates, and environmental conditions. Other studies (Imtiaj & Rahman, 2008; Raman et al., 2021) have shown that mushroom strain, spawn rate, and growing conditions affect the interval between spawning and fruiting. Rapid mycelial growth is often associated with quicker pinhead formation and higher yield (Baysal et al., 2003; Naraian et al.,



Figure 1. Survival rate (%) of white and grey oyster mushroom.

Species		Sub	strate			Species	
Species	RS + SD (Control)	RS+SBS	BS RS+ PS RS+		RS+lpL	Mean	
White Oyster	28.9	25.2	25.7	24.8	30.5	27.03 ^b	
Gray Oyster	31.9	29.8	30.0	29.8	33.1	30.9ª	
Substrate Mean	30.4 ^b	27.5°	27.9°	27.3°	31.8ª		

Table 1. The average number of days of mycelium ramification.

RS rice straw, SD sawdust, SBS soybean straw, PS peanut straw, MS mungbean straw, IpL ipil-ipil leaves CV-2.72%

Note: the lower the number of days the better

 Table 2.
 Average number of days of pinhead formation.

Species	Substrate Combination					
	RS + SD (Control)	RS+SBS	RS+ PS	RS+ MS	RS+lpL	Mean
White Oyster	37.8	31.5	30.6	30.3	42.5	34.5 ⁵
Gray Oyster	48.7	39.8	41.3	38.5	44.1	42.8 ª
Substrate Mean	43.2ª	35.7⁵	35.9 ⁵	34.4 ^b	43.3ª	

RS rice straw, SD sawdust, SBS soybean straw, PS peanut straw, MS mungbean straw, IpL ipil-ipil leaves

CV- 9.33%

Note: The lower the number of days the better

2009), confirming the importance of substrate suitability.

Yield. Yield was measured by weighing the freshly harvested mushroom fruiting bodies. No significant difference was found between the yields of white (M = 202.5 g) and gray (M = 193.8 g) oyster mushrooms (p = 0.13). However, substrate type significantly affected yield (p < 0.001). As shown in Table 3, RS+SBS (M = 217.12) and RS+PS (M = 213.97) achieved significantly higher yields compared to RS+MS (M = 198.27), RS+SD (M = 191.42), and RS+IpL (M = 170.08). No interaction effect was found between substrate and species (p = 0.1914).

These findings confirm that incorporating nitrogen-rich leguminous plants into the substrate can significantly improve yield, consistent with Tikdari and Bolandnazar (2012). Musara et al. (2017) emphasized the value of combining cereal straw (for lignocellulose) and nitrogenous substrates (for protein) to enhance mushroom growth.

Despite its nitrogen content, ipil-ipil foliage contains mimosine, a toxic compound that interferes with enzymatic activities, inhibits cell division, and may lead to plant growth suppression and mortality (Kato-Noguchi & Kurniadie, 2022). This may explain the significantly lower yield from RS+IpL. Andrew (2023) similarly found that substrate mixes with 20–100% ipil-ipil foliage resulted in reduced or no yield. Thus, a 30% inclusion, as in this study, corresponded to the lowest yield.

Table 3. Average yield (g) of oyster mushroom.

These results align with Ashraf et al. (2013), who also found no significant yield difference between white and gray oyster species.

Biological efficiency (BE). Biological efficiency (BE) was used to assess the effectiveness of each substrate combination in mushroom production. It was calculated by dividing the total fresh yield by the dry weight of the substrate. No significant difference in BE was observed between white and gray oyster mushrooms (p = 0.18). However, significant differences were found among substrate combinations (p < 0.001).

Post hoc analysis showed that RS+SBS (M = 109.23) and RS+PS (M = 108.58) had significantly higher BE values than RS+MS (M = 98.52), RS+SD (M = 96.67), and RS+IpL (M = 85.15). There was no significant interaction effect between substrate and species (p = 0.07).

These findings mirror yield trends (Tables 3 and 4), as BE is directly correlated with yield. The incorporation of soybean and peanut straw notably increased BE. This study's BE range (85-109%) is higher than that reported by Muswati et al. (2021), which ranged from 66-86%. Thus, using nitrogenrich substrates can significantly improve the biological efficiency of both oyster mushroom species.

Proximate analysis. Proximate analysis of both the mushroom fruiting bodies and the substrates was

Species	Substrate Combination					
Species	RS + SD (Control)	RS+SBS	RS+ PS	RS+ MS	RS+lpL	Mean
White Oyster	188.3	218.3	213.7	204.6	187.9	202.5 ^{ns}
Gray Oyster	194.5	216.0	214.2	192.0	152.3	193.8 ^{ns}
Substrate Mean	191.42°	217.12ª	213.97ªb	198.27 ^{bc}	170.08 ^d	

RS rice straw, SD sawdust, SBS soybean straw, PS peanut straw, MS mungbean straw, IpL ipil-ipil leaves CV-7.78

Table 4. Biological efficiency (%) of Pleurotus spp.

Species	Substrate Combination					
	RS + SD (Control)	RS+SBS	RS+ PS	RS+ MS	RS+lpL	– Mean
White Oyster	95.8	109.4	105.2	102.9	94.5	101.6 ^{ns}
Gray Oyster	97.5	109.1	112.0	94.1	75.8	97.7 ^{ns}
Substrate Mean	96.67 ^ь	109.23ª	108.58ª	98.52 ^b	85.15°	

RS rice straw, SD sawdust, SBS soybean straw, PS peanut straw, MS mungbean straw, IpL ipil-ipil leaves CV- 7.79 conducted in the feed laboratory. Rice straw-based substrates with nitrogen-rich plants showed higher nitrogen content than the rice straw + sawdust control (Figure 2). The RS+IpL and RS+PS substrates had the highest nitrogen content (1.23% and 1.22%, respectively), followed by RS+SBS (0.97%), RS+MS (0.92%), and RS+SD (0.6%) (SD = 0.26). These findings support Salami et al. (2017), who stated that nitrogen content varies with the raw materials used.

Crude protein levels. Crude protein content of the mushroom fruiting bodies was determined using the Kjeldahl method (Figure 3). Among the white oyster mushrooms, those grown on RS+PS recorded the highest protein level at 23.06%. For gray oysters, RS+IpL and RS+MS yielded the highest crude protein levels at 25.54% and 25.06%, respectively.

In contrast, the lowest crude protein content (15.3% and 16.96%) was recorded from mushrooms grown on substrates without nitrogen-rich plant inclusion. Overall, gray oyster mushrooms had a

slightly higher mean protein content (M = 22.09%, SD = 3.1) than white oysters (M = 20.93%, SD = 2.89).

Protein content varied across substrate combinations, with values ranging from 15.3% to 25.54%. While RS+IpL substrates negatively affected yield, they resulted in higher protein levels in gray oyster mushrooms—supporting Andrew's (2023) findings. This suggests that while ipil-ipil may reduce productivity, it can enhance nutritional content. Overall, nitrogen-rich substrates improved the protein content of both oyster mushroom species, consistent with findings from previous studies.

Proximate analysis of crude fiber and crude fat. Results show that gray oyster mushrooms contain a higher crude fiber content (M = 7.74%) compared to white oyster mushrooms (M = 7.4%, SD = 0.22). As shown in Figure 4, the crude fiber content in both oyster species was highest in the RS+SD substrate— 8.93% in white oyster and 9.59% in gray oyster



Figure 2. Nitrogen content of different substrate combinations.



Figure 3. Crude protein of oyster mushroom spp. in different substrates.



Figure 4. The crude fiber, crude fat and ash content of oyster mushroom spp.

mushrooms. Conversely, mushrooms grown in the RS+IpL substrate exhibited the lowest crude fiber content at 6.63% and 6.77%, respectively.

The study also analyzed crude fat content. For white oyster mushrooms, the highest fat content was observed in the RS+SD substrate (4.21%), while the lowest was in the RS+SBS substrate (3.53%). For gray oyster mushrooms, the highest fat content (4.21%) was found in RS+SBS, while the lowest was in RS+SpL (1.65%).

Fresh oyster mushrooms typically contain low fat-between 1% and 8% of their dry weight (Bankole & Salami, 2017). In this study, the crude fat content ranged from 1.65% to 4.21%. On average, white oyster mushrooms had a higher fat content (M =3.85%) compared to gray oyster mushrooms (M = 3.41%, SD = 0.31). These results also indicate that fat content varied depending on the substrate used. The low-fat content observed supports the findings of Onyeka et al. (2018). Mushrooms are known to be a rich source of dietary fiber (Zhao et al., 2022), surpassing conventional sources such as cereals, legumes, fruits, and vegetables (Cheung, 2013). In this study, the crude fiber content ranged from 6.63% to 9.59%, slightly lower than the values reported by Elkanah et al. (2022), which ranged from 9.89% to 11.18%. These findings align with those of Salami et al. (2017), who noted that the nutritional composition of mushrooms can vary depending on the substrate used.

Return on investment (ROI). As shown in Figure 5, white oyster mushrooms grown in the RS+SBS substrate yielded the highest ROI at M = 117.1%,

followed by RS+PS (112.5%), RS+MS (103.5%), RS+IpL (83.2%), and RS+SD (control) at 51.5% (SD = 15.1%). Similarly, for gray oyster mushrooms, the highest ROI was also observed in RS+SBS (M = 114.8%), followed by RS+PS (113%), RS+MS (90%), RS+SD (58.7%), and RS+IpL (51.5%) (SD = 25.6%).

Computation of economic values. The economic analysis was based on the production of 1,000 fruiting bags per cycle. As shown in Table 5a, the highest net income for both mushroom species was obtained from RS+SBS, amounting to PhP29,140.00, followed by RS+PS at PhP28,352.00. The lowest net return was recorded from RS+IpL at PhP17,380.00.

In terms of species-specific performance (Table 5b), white oyster mushrooms generated slightly higher net returns (PhP25,375.00) compared to gray oyster mushrooms (PhP23,200.00). The cost and return analysis indicates that incorporating nitrogenrich (N-rich) plants into the substrate enhances ROI and net income. While the economic returns of white oyster mushrooms were marginally higher, the variations in yield are likely due to differences in substrate combinations. Notably, eliminating sawdust from the substrate reduced total expenses for N-rich substrate formulations.

The average ROI for white oyster mushrooms was 100.5%, and for gray oyster mushrooms, it was 91.9%. These figures are lower than the ROI reported by Guerrero et al. (2015) at 144% and Retnaningsih (2018) at 110%. The differences may be attributed to variation in mushroom prices, types of substrates used, and resulting yields.



Figure 5. Return on investment.

Items/ Particulars		Sul	ostrate Combinat	trate Combination		
	RS+SD (PhP)	RS+SBY (PhP)	RS+PS (PhP)	RS+MS (PhP)	RS+lpL (PhP)	
Gross Income:						
Mushroom Fruit	47,855.00	54,280.00	53,492.50	49,567.50	42,520.00	
Total Gross Income	47,855.00	54,280.00	53,492.50	49,567.50	42,520.00	
Total Variable Costs	18,590.00	18,040.00	18,040.00	18,040.00	18,040.00	
Total Fixed Costs	7,100.00	7,100.00	7,100.00	7,100.00	7,100.00	
Total Cost	25,690.00	25,140.00	25,140.00	25,140.00	25,140.00	
Net Income	22,165.00	29,140.00	28,352.50	24,427.50	17,380.00	
ROI	86.3%	115.9%	112.8%	97.2%	69.1%	

See Table 1 for abbreviations

 Table 5b. Return on investment of different species of oyster mushroom.

Items/ Particulars	Substrate Combination			
	White Oyster (PhP)	Grey Oyster (PhP)		
Gross Income:				
Mushroom Fruit	50,625.00	48,450.00		
Total Gross Income	50,625.00	48,450.00		
Total Variable Costs	18,150.00	18,150.00		
Total Fixed Costs	7,100.00	7,100.00		
Total Cost	25,250.00	25,250.00		
Net Income	25,375.00	23,200.00		
ROI	100.5%	91.9%		

Conclusion

The study concludes that incorporating nitrogenrich plant residues into rice straw-based substrates enhances the nitrogen content of the growing medium for oyster mushrooms. This incorporation promotes faster mycelial run and higher yields in *Pleurotus spp.* production. Soybean, peanut, and mungbean straw substrates yielded better results compared to traditional farmer-used substrates.

Additionally, the inclusion of leguminous plants shortened the cultivation period to around 28 days, enabling more production cycles within a given timeframe. The widespread availability of leguminous crops across various regions presents an opportunity for farmers to adopt N-rich substrate formulations in mushroom farming.

Recommendations

This study is relevant in areas where nitrogenous materials are readily available or considered agricultural waste, offering a sustainable source of nitrogen. It is also recommended for farmers who practice intercropping or crop rotation with legumes. Using nitrogen-rich substrates from soybean, peanut, and mungbean straw improves both the nutritional profile of the substrate and the mushroom fruiting bodies. Scaling up this substrate formulation and disseminating it to mushroom growers could significantly improve the nutritive value of oyster mushrooms.

Furthermore, it is recommended that oyster mushrooms be incorporated more regularly into diets for their protein content and overall health benefits. Continuous efforts to increase mushroom yield should be prioritized in response to rising population and demand. Future studies should explore other abundant agro-waste materials beyond leguminous straw and tree residues to further improve mushroom production sustainability and profitability.

Literature Cited

- Andam C, Dulay H (2014) Oyster mushroom: A treasure from waste. The Philippine Star. Retrieved from https://www. philstar.com/business/agriculture/2014/12/14/1402316/ oyster-mushroom-treasure-waste (accessed October 13, 2021)
- Andrew SM (2023) Production and nutritional value of *Pleurotus* floridanus grown on rice straw supplemented with Leucaena leucocephala foliage. Environmental and Sustainability Indicators 17: 100223
- Ashraf J, Muhammad AA, Waqas Ahmad, Ayyub CM, Shafi J (2013) Effect of different substrate supplements on oyster mushroom (*Pleurotus spp.*) production. Food Science and Technology 1(3): 44 - 51
- Bach F, Helm CV, Bellettini MB, Maciel GM, Haminiuk CWI (2017) Edible mushrooms: a potential source of essential amino acids, glucans and minerals. International Journal of Food Science & Technology 52(11):2382 - 2392
- Bankole FA, Salami AO (2017) Use of Agro-Wastes for tissue culture process and spawn production of oyster mushroom (*Pleurotus florida*). Journal of Applied Life Sciences International 14 (1): 1 - 9

- Baysal E, Parker H, Yalinkilic MK, Temiz A (2003) Cultivation of oyster mushroom on waste paper with some added supplementary materials. Bioresource Technology 89: 95 - 97
- Belewu MA, Lawal RA (2003) Studies of the cultivation of edible mushroom (*Pleurotus pulmonarius*) on cocoa bean shell and sawdust- *Gliricidia* mixture. Res Comm Microbiol *I*(2): 68 - 71
- Bhattacharjya DK, Paul RK, Miah MN, Ahmed KU (2014) Effect of different saw dust substrates on the growth and yield of oyster mushroom (Pleurotus ostreatus). IOSR Journal of Agriculture and Veterinary Science 7(2) 38 - 46
- Chang HY, Jeon SW, Cosadio A, Icalina C, Panganiban R, Quirino R, Song Y (2014) Status and prospect of mushroom industry in the Philippines. JPAIR Multidisciplinary Research 16:1 - 6
- Cheung PC (2013) Mini-review on edible mushrooms as source of dietary fiber: preparation and health benefits Food Science and Human Wellness 2(3-4):162 166
- Elkanah FA, Oke MA, Adebayo EA (2022) Substrate composition effect on the nutritional quality of *Pleurotus ostreatus* (MK751847) fruiting body, Heliyon *8*(11)
- Guerrero MB, Aragones JA, Balane NM, Rantugan HG, Pelagio VP (2015) Evaluation of Different Formulations of Substrate Materials for the Growth and Yield of Two (2) species of Oyster Mushrooms (*Pleurotus florida* and *Pleurotus sajor caju Jack.et Fr*) https://www.academia. edu/33151388/Evaluation_of_Different_Formulations_of_ Substrate_Materials_for_the_Growth_and_Yield_of_ Two_2_Species_of_Oyster_Mushrooms_Pleurotus_ florida_and_Pleurotus_sajor_caju_Jack_et_Fr_1 (accessed September 24, 2023)
- Hassan FRH, Medany GM, Abou Hussein SD (2010) Cultivation of the king oyster mushroom (*Plerrotus eryngii*) in Egypt. Australian Journal of Basic and Applied Sciences 4(1): 99 - 105
- Imtiaj A, Rahman SA (2008) Economic viability of mushrooms cultivation to poverty reduction in Bangladesh. Tropical and Subtropical Agroecosystems 8:93 - 99
- Kato-Noguchi H, Kurniadie D (2022) Allelopathy and allelochemicals of *Leucaena leucocephala* as an invasive plant species. Plants *11*(13): 167
- Kjeldahl J (1883) New Method for the Determination of Nitrogen. Chem. News 1883, 48 (1240), 101–102; Neue Methode zur Bestimmung des Stickstoffs in organischen K"orpern. Z. Anal. Chem. 1883, 22, 366–382; En ny Methode til Kvaelstofbestemmelsei organiske Stooffer. Medd. Carlsberg Lab. 1883, 2 (1), 1–27; Sur une Nouvelle M'ethode de Dosage de l'Azote dans les Substances Organiques (French summary: R'esum'e du CR Trav. Lab. Carlsberg; separately paged section) 1883, 2 (Juni), 1-12
- Ladli (2020) A review on oyster mushroom [*Pleurotus ostreatus*] Cultivation. International Journal of current microbiology and applied sciences *II*:1653 - 1665
- Lopez S, Davies D, Giraldez F, Dhanoa MS, Dijkstra J, France J (2005) Assessment of nutritive value of cereal and legume straws based on chemical composition *in vitro* digestibility. Journal of the Science of Food and Agriculture **85**(9): 1550 -1557

- Mamiro DP, Mamiro PS (2011) Yield and mushroom size of *Pleurotus ostreatus* grown on rice straw basal substrate mixed and supplemented with various crop residues, Journal of Animal & Plant Sciences 10(1): 1211 - 1218
- Musara A, Gasura E, Ngadze A, Matikiti A, Mashingaidze AB, Zvidzai C (2017) Effect of mixing cereal and legume straws on yield of grey oyster mushroom under controlled conditions. African Crop Science Journal 26(2): 175 - 187
- Muswati C, Simango K, Tapfumaneyi L, Mutetwa M, Wonder Ngezimana W (2021) The effects of different substrate combinations on growth and yield of oyster mushroom (*Pleurotus ostreatus*). International Journal of Agronomy 2021. 1-10. 10.1155/2021/9962285 (accessed August 24, 2022).
- Naraian R, Sahu RK, Kumar S, Garg SK, Singh CS, Kanaujia RS (2009) Influence of different nitrogen rich supplements during cultivation of *Pleurotus florida* on corn cob substrate. The Environmentalist 29:1 - 7
- Nunes MD, Da Luz JM, Paes SA, Ribeiro JJ, Da Silva MD (2012) Nitrogen supplementation on the productivity and the chemical composition of oyster mushroom. Journal of Food Research *I*(2):113 113
- Odunmbaku OK, Adenipekun CO (2018) Cultivation of Pleurotus ostreatus (Jacq Fr.) Kumm on Gossypium hirsutum Roxb. (Cotton waste) and Gmelina arborea L. sawdust. International Food Research Journal 25(3): 1140 - 1145
- Oei P (2003) Mushroom cultivation: appropriate technology for mushroom growers (pp 429). (Ed.3), Leiden, English, Book, 90-5782-137-0, Backhuys Publishers: Netherlands
- Onyeka EU, Udeogu E, Umelo C, Okehie MA (2018) Effect of substrate media on growth, yield and nutritional composition of domestically grown oyster mushroom (*Pleurotus ostreatus*) African Journal of Plant Science 12 (7): 141 - 147
- Piska K, Sułkowska-Ziaja, Muszyńska B (2017) Edible mushroom *Pleurotus ostreatus* (Oyster mushroom): its dietary significance and biological activity. *Acta Scientiarum Polonorum Hortorum Cultus* 16: 151 - 161
- Raman J, Jang KY, Oh YL, Oh M, Ima JH, Lakshmanan H, Sabaratnam V (2021) Cultivation and nutritional value of prominent *Pleurotus spp.:* an overview. Mycobiology 49(1):1 - 14
- Rashidi AM, Yang TA (2016) Nutritional and antioxidant values of oyster mushroom (P. sajor-caju) cultivated on rubber sawdust. International Journal on Advanced Science Engineering Information Technology 6(2):161 - 164
- Retnaningsih N (2018) Analysis the feasibility of oyster mushroom (*pleurotus ostreotus*) business in the village of Gondangmanis, Karangpandan sub-district, Karanganyar regency (pp 26 - 29). Atlantis Press
- Salami AO, Bankole FA, Andrew OC (2017) Effect of organic nitrogen supplements on the yield and nutrient content of oyster mushroom (*Pleurotus florida*) Cultivated on Corncobs. SciFed Journal of Mycology Retrieved from https://www.researchgate.net/profile/Faith-Bankole/ publication/329950225_Effect_of_Organic_Nitrogen_ Supplements_on_the_Yield_and_Nutrient_Content_of_ Oyster_Mushroom_Pleurotus_Florida_Cultivated_on_

Oyster Mushroom Yield in Nitrogen-rich Substrates including Rice Straw

Corncobs/links/5c253482a6fdccfc706c482c/Effect-of-Organic-Nitrogen-Supplements-on-the-Yield-and-Nutrient-Content-of-Oyster-Mushroom-Pleurotus-Florida-Cultivated-on-Corncobs.pdf (accessed August 24, 2022)

- Tikdari MM, Bolandnazar S (2012) Application of organic nitrogen supplementations increases the yield of oyster mushroom (*Pleurotus florida*). Research in Plant Biology 2(3):10 - 15
- Zhao H, Wang L, Brennan M, Brennan C (2022) How does the addition of mushrooms and their dietary fibre affect starchy foods. Journal of Future Foods 2:26 - 32

FULL PAPER

IDENTIFICATION AND *IN-VITRO* EVALUATION OF PLANT GROWTH-PROMOTING RHIZOBACTERIA ISOLATED FROM WEEDY RICE AND CULTIVATED RICE (*ORYZA SATIVA L.*) COLLECTED FROM PADDY FIELDS

Leylani M. Juliano^{1*}, Editha V. Evangelista², And Analiza Henedina M. Ramirez³

¹Agronomy, Soils and Plant Physiology Division, Philippine Rice Research Institute, Science City of Muñoz, Nueva Ecija
 ²Crop Biotechnology Center, Philippine Rice Research Institute, Science City of Muñoz, Nueva Ecija
 ³Institute of Weed Science, Entomology and Plant Pathology, University of the Philippines Los Baños, College, Laguna
 *Corresponding Author: lmj.juliano@gmail.com

Abstract

Rhizosphere microorganisms can either benefit or harm plant development. Research on plant growthpromoting rhizobacteria (PGPR) has provided strong evidence of their beneficial roles in crop production. In this study, weedy rice known for its competitive advantage over cultivated rice - was examined for the presence of potential PGPR. Weedy rice is observed to germinate rapidly and exhibit superior root systems, taller shoots, and longer, drooping leaves and panicles compared to cultivated rice. Forty-one bacterial isolates were obtained from the rhizosphere of weedy rice, and 19 from cultivated rice. These isolates were screened for plant growthpromoting traits including starch hydrolysis, indole-3-acetic acid (IAA) production, phosphate solubilization, and 1-aminocyclopropane-1-carboxylate (ACC) deaminase activity. Findings suggest that PGPR associated with weedy rice enhance root and shoot development during the early seedling stage, conferring a competitive growth advantage over cultivated rice. The presence of microorganisms with beneficial metabolic pathways in the weedy rice rhizosphere implies a potential role in improving plant health, suppressing pathogens, and inducing systemic defense. These advantages may contribute to nutrient uptake disadvantages in cultivated rice under paddy conditions.

Keywords: competitive ability, plant growth-promoting traits, rhizobacteria, weedy rice

Introduction

Microorganisms with beneficial effects on plants are promising tools for biofertilizer and biocontrol applications, supporting more sustainable agriculture and environmental protection. The growth-promoting characteristics of rhizosphere microorganisms contribute to improved nutrient absorption, enhanced yield, and reduced dependency on chemical fertilizers and pesticides.

Various physiological traits of plant growthpromoting rhizobacteria (PGPR) contribute to crop development. These traits include the production of: (1) ammonia, (2) siderophores, (3) gibberellic acid, (4) indole-3-acetic acid (IAA), (5) phosphorus solubilization, (6) potassium solubilization, (7) zinc solubilization, and (8) nitrogen fixation via semi-solid nitrogen-free media (Nfm). Nitrogen fixation plays a vital role in the nitrogen cycle and plant nutrition. Crop management practices such as the use of crop residues, fertilizers, and pest control affect the contribution of biological nitrogen fixation (BNF) to the overall nitrogen supply (Cassman et al., 1998). Additionally, nitrogen-fixing bacteria (NFB) and arbuscular mycorrhizal fungi (AMF) have been shown to improve nutrient and water uptake, and to enhance resistance to biotic and abiotic stresses (Sanchez-Navarro et al., 2020).

Microorganisms respond to plant root exudates by enhancing mineral solubilization and organic matter mineralization through the release of phytohormones, organic compounds, and enzymes, ultimately promoting plant growth (Zahir et al., 2005; Nadeem et al., 2014). Thus, identifying and characterizing plant growth-promoting microorganisms from the rice rhizosphere can help understand their role in the competitive growth of weedy rice when associated with cultivated rice.

This study aimed to isolate and characterize rhizosphere microorganisms from weedy and cultivated rice, assess their plant growth-promoting properties, and identify possible mechanisms by which they influence the growth and development of rice plants.

Materials and Methods

Isolation and Purification of Plant Growthpromoting Rhizobacteria

Plant and Rhizosphere Sample Collection. Whole plant samples of weedy and cultivated rice, including the root systems and surrounding soil (root balling), were collected from selected field sites (Table 1). Bacterial isolation followed the method of Cruz and Paterno (2014). The entire root system was carefully removed from the soil and gently tapped to dislodge loosely attached soil particles.

Table 1. Sampling sites and plant samples from each site for
identification of plant growth promoting rhizobacteria.

Site Collection	Weedy Rice	Cultivated Rice
San Roque, San Manuel, Pangasinan (April 5, 2019)	WR SR 1 - straw- colored grain, purple tip, rep pericarp, no awn	NSIC Rc 222
Calzada, Mabini, Pangasinan (April 5, 2019)	brown to reddish grain, brown pericarp, no awn	NSIC Rc 222
La Purisima, Aliaga, Nueva Ecija (April 9, 2019)	straw-colored grain, red pericarp, straw colored medium to long awn	NSIC Rc 222

*Cultivated rice varieties were identified by farmers.

Serial Dilution and Spread Plate Method. The root-soil mixture was suspended in water and serially diluted up to four ten-fold dilutions. From the 10⁻³ and 10⁻⁴ dilutions, 0.1 mL was plated in duplicates on Burk's agar medium (Cruz and Paterno, 2014). Plates were incubated at room temperature. Morphologically distinct colonies were isolated and sub-cultured for further characterization.

Growth Medium. Burk's medium, a nitrogen-free medium containing inorganic salts and carbohydrates but lacking nitrogen, was used to isolate nitrogen-fixing bacteria. The medium was sterilized at 121°C for 20 minutes prior to use and prepared fresh as needed.

Purification of Isolated Bacteria. Colonies were initially distinguished based on color and morphology. A loopful of each unique colony was streaked on Burk's agar plates in duplicate to obtain pure cultures. Each purified isolate was labeled with a code for identification in subsequent experiments.

In-vitro Screening of Bacterial Isolates for Plant Growth-promoting Properties

Bacterial isolates were screened for the following plant growth-promoting (PGP) traits: (1) starch hydrolysis, (2) IAA production, (3) phosphate solubilization, and (4) ACC deaminase activity. All media used were sterilized at 121°C for 20 min prior to use. Only isolates that were successfully maintained as pure cultures were subjected to PGP screening, due to contamination challenges with certain samples in the soil microbiology laboratory.

Starch Hydrolysis. Purified isolates were streaked on starch agar (composition: 3 g beef extract, 10 g soluble starch, 15 g agar, and 1 L distilled water). After 24h of incubation, iodine solution was added to detect starch degradation. A clear halo surrounding the colony indicated starch hydrolysis, while undigested starch turned blue (Collins et al., 2004).

Indole-3-Acetic Acid (IAA) Production Assay. Each bacterial isolate was inoculated in nitrogen-free broth which consists of 31.95 g Burk's medium and 1500 mL distilled H2O supplemented with 0.10 g L-1 Tryptophan (Cruz and Paterno, 2014; Shahab et al., 2009). After seven days of incubation, 1.5 mL of bacterial broth was centrifuged for 10 min at 13,000 rpm in 4oC. Then, 1 mL of the supernatant was mixed with 2 mL of Salkowski's reagent in a test tube. This was kept at room temperature in the dark for 30 min. An aqueous solution (1 mL) containing either 0.25 mM indole-3-acetic acid (IAA), 0.25 mM tolbutamide (TOL), or a combination of 0.25 mM IAA and 0.25 mM TOL was prepared. Each solution was then mixed with 2 mL of Salkowski's reagent. The ultraviolet (UV) spectrum of the resulting mixtures was subsequently measured directly using a Hitachi U3310 Spectrophotometer (Tokyo, Japan) within a wavelength range of 220 to 650 nm. A reddish to pink coloration indicated positive IAA production.

Phosphate Solubilization Assay. Isolates were grown in modified Pikovskaya's medium containing 5.0 g Ca₃(PO₄0), 0.2 g NaCl, 0.2 g KCl, 0.1 g MgSO₄ 7H₂O, 0.00025 g MnSO₄ 7H₂O, 0.00025 g $FeSO_{4}$, $7H_{2}$), 0.5 g (NH₄)2SO₄, 0.5 g yeast extract, 10.0 g glucose, 20.0 agar, and 1L distilled H₂O. Bacterial isolates were spot-inoculated on the agar's surface and incubated for seven days. Clearing zone around the bacterial growth or colony indicates phosphate solubilization (Cruz and Paterno, 2014; Shahab et al., 2009). Then, phosphate-solubilizing capacity of isolates were semi-quantitatively determined in terms of its phosphorus solubilization index (PSI) using the formula by Islam and Hossain (2012). Using a desk ruler, the total diameter of halo zone was measured by getting the length of the halo zone from one edge to the other. The same procedure was followed also in measuring the length of colony diameter.

$$PSI = \frac{Total \, diameter \, of \, the \, halo \, zone \, (mm)}{Colony \, diameter \, (mm)}$$

1-aminocyclopropane-1-carboxylate (ACC) Deaminase Activity Assay. Each isolate was grown in nitrogen-free Dworkin and Foster's minimal salts agar media. Two batches of media were prepared: one supplemented with 0.3 g L⁻¹ ACC and other one supplemented with 0.3 g L⁻¹ (NH₄)₂SO₄. Plates were incubated in the dark for seven days and growth of isolate on the media were taken as an indicator of efficiency of selected isolates to utilize ACC or (NH₄)2SO₄ and to produce ACC deaminase (Cruz and Paterno, 2014). The growth of the colony was observed and scored positive (+) and in case no growth was observed, a negative (-) symbol was used.

Identification of Selected Isolates

Selected isolates were identified using the BiOLOG GEN III Microbial Identification System, following the manufacturer's protocol. The system comprises 94 phenotypic tests, including 71 carbon utilization and 23 chemical sensitivity assays. Isolates were grown on Burk's agar, suspended in inoculating fluid to the recommended cell density, and inoculated into GEN III MicroPlates. Plates were incubated for 24 hours and analyzed using the BiOLOG software at the Natural Sciences Research Institute (NSRI), University of the Philippines Diliman.

In-vitro Evaluation of the Effectiveness of a Highly Potential PGP Isolate on Seedling Growth of Weedy Rice and Cultivated Rice under Laboratory Conditions

The plant growth-promoting (PGP) bacterial isolate identified from weedy rice in the previous study was used for the *in-vitro* evaluation. The isolate found to have multiple plant growth-promoting properties was used as the test material under laboratory conditions. A loopful of the isolate was diluted in 100 mL distilled water. Ten seeds of each weedy rice (WR SR 1) and cultivated rice (NSIC Rc 222) were allowed to germinate in petri dishes lined with filter paper in three replicates. The treatments and test materials were as follows: all uninoculated were labeled with (U); monoculture rice A1R(U), monoculture weedy rice A1WR(U), mixed culture rice A3R(U) and weedy rice A3WR(U); inoculated materials followed the same format but instead letter B was added last with the (U) removed. The diluted PGP isolate was used to soak the test seeds. The petri dishes were covered and remained in the laboratory.

Data gathered. Ten days after inoculation, the shoot and root growth of each seed were measured (cm). The data are presented as the mean of measurements from 10 seeds.

Test materials. Uninoculated control and inoculated weedy and cultivated rice samples were

used for comparison on the qualitative evaluation of the root hairs under electron scanner at 400x magnification.

Results and Discussion

Isolation, Screening and Identification of Plant Growth-promoting Rhizobacteria

There were 60 bacterial isolates taken from the six rice materials; 19 isolates were from cultivated rice (6 from Aliaga, 6 from San Roque, and 7 from Calzada); and 41 isolates from weedy rice biotypes (14 from Aliaga, 18 from San Roque, and 9 from Calzada). During the purification process, the isolates were characterized based on their morphological features, including colony color (light brown, yellow, cream, white, brown, off-white, clear), margin or edge (filiform, entire, undulate, lobate, curled), colony form (circular, irregular, punctiform, filamentous, rhizoid), transparency (opaque, translucent), and colony nature (spread, continuous). These isolates were purified and maintained in culture media for subsequent in vitro screening of plant growthpromoting (PGP) traits.

In-vitro Screening of Bacterial Isolates for Plant Growth-Promoting Properties

Bacterial isolates were evaluated for the following PGP traits: starch hydrolysis, indole-3-acetic acid (IAA) production, phosphate solubilization, and 1-aminocyclopropane-1-carboxylate (ACC) deaminase activity.

Starch Hydrolysis Assay. There were 51 bacterial isolates (30 WR and 21 cultivated rice) subjected to the starch hydrolysis assay. Of these materials, 19 isolates from weedy rice and 6 from cultivated rice showed positive results for starch hydrolysis (Table 2). Amylolytic activity was indicated by the presence of a yellow or clear zone around bacterial colonies grown on a blue medium. This zone signifies the hydrolysis of starch into monosaccharides, which do not bind with iodine, resulting in the absence of blueblack coloration around the colony.

The diameter of the clear zone reflects the extent of enzymatic activity; some isolates exhibited strong amylase activity (+++), characterized by prominent clearing zones, while others showed weak activity (+), indicated by faint or barely visible zones (Figure 1). This finding supports the principle that as iodine binds to starch producing a deep purple color, its conversion to sugars reduces the available starch, creating a clear zone around colonies (Collins et al., 2004). Bacteria that produce the extracellular enzyme amylase hydrolyze starch into simpler sugars such as di- and monosaccharides, which plants can absorb (Bird and



Figure 1. Samples of bacterial isolates from rice and weedy rice showing the presence of starch hydrolysis activity as revealed by the clearing zone surrounding the bacterial growth.

 Table 2. Results of starch hydrolysis assay in 51 bacterial isolates.

No	Sample	Reaction	No	Sample	Reaction
A1	Aliaga Weedy Rice	-	A46	Calzada Weedy Rice	-
A2	Aliaga Weedy Rice	+	A48	Calzada Weedy Rice	+
A3	Aliaga Weedy Rice	-	A50	Calzada Weedy Rice	-
A4	Aliaga Weedy Rice	-	A51	Calzada Weedy Rice	+
A5	Aliaga Weedy Rice	+	A53	Calzada Weedy Rice	+
A6	Aliaga Weedy Rice	-	A54	Calzada Weedy Rice	-
A7	Aliaga Weedy Rice	-	A55	Calzada Weedy Rice	++
48	Aliaga Weedy Rice	+	A58	San Roque Weedy Rice	-
49	Aliaga Weedy Rice	+++	A60	Calzada Weedy Rice	-
A10	Aliaga Weedy Rice	+	B2	Aliaga Rice	-
A11	Aliaga Weedy Rice	-	B3	Aliaga Rice	-
A12	Aliaga Weedy Rice	-	B4	San Roque Rice	-
A13	Aliaga Weedy Rice	-	B5	San Roque Rice	+
A14	Aliaga Weedy Rice	++	B6	San Roque Rice	-
A15	Calzada Weedy Rice	-	B7	Calzada Rice	-
A16	San Roque Weedy Rice	+	B8	Calzada Rice	-
A17	San Roque Weedy Rice	-	B9	Calzada Rice	+
A18	San Roque Weedy Rice	+	B10	Calzada Rice	-
A19	San Roque Weedy Rice	-	B11	Aliaga Rice	+
A20	San Roque Weedy Rice	-	B12	Aliaga Rice	+
A21	San Roque Weedy Rice	-	B13	Aliaga Rice	-
422	San Roque Weedy Rice	+	B17	Aliaga Rice	+
A23	San Roque Weedy Rice	+	B18	Calzada Rice	-
A30	San Roque Weedy Rice	-	B21	Calzada Rice	-
A31	San Roque Weedy Rice	+	B22	Calzada Rice	-
A32	San Roque Weedy Rice	-	B23	San Roque Rice	-
A33	San Roque Weedy Rice	-	B24	San Roque Rice	+
434	San Roque Weedy Rice	+	B25	San Roque Rice	-
435	San Roque Weedy Rice	+++	B25	San Roque Rice	-
436	San Roque Weedy Rice	-		+ weak amylase producers	
A37	San Roque Weedy Rice	++		+++ strong amylase produce	ers
A40	San Roque Weedy Rice	+			

Hopkins, 1954). Variation in clearing zones among isolates likely reflects differences in their ability to break down complex molecules.

Indole-3-acetic acid (IAA) Production Assay. In this study, 26 isolates were assessed for IAA production: 18 from weedy rice (Sample A) and 8 from cultivated rice (Sample B) (Table 3). Among these, only two isolates from weedy rice and four from cultivated rice did not produce IAA.

Previous research has shown that many plant growth-promoting rhizobacteria (PGPR) in the rhizosphere synthesize IAA as a secondary metabolite of L-tryptophan, enhancing root development and nutrient absorption (Patten and Glick, 1996; Keswani et al., 2020). IAA is a key phytohormone involved in various plant growth processes such as seed germination, root elongation, flowering, and fruit clear zone on Pikovskaya's medium, likely due to the production of organic acids by the bacteria. This suggests that rice soils harbor bacteria capable of increasing P availability and supporting plant growth.

In addition to solubilizing P, many PSBs produce IAA, further stimulating plant growth by enhancing metabolic processes such as RNA and protein synthesis and improving nutrient uptake (Shahab et al., 2009).

Phosphorus is the second most critical macronutrient after nitrogen, playing essential roles in photosynthesis, signal transduction, and respiration (Anand et al., 2016; Sharma et al., 2013). Since the 1950s, PSBs have been used as biofertilizers, facilitating the conversion of phosphorus into forms accessible to plants (Amri et al., 2023). Mechanisms involved include secretion of organic acids,

Table 3. Indole-3-acetic acid (IAA) production of the different isolates collected from weedy rice and samples.

Samples	Final Absorbance	Final Concentration (ppm)	Samples	Final Absorbance	Final Concentration (ppm)
A2	0.1959	5.9958	A40	0.1499	4.0792
A4	0.0569	0.2042	A48	0.0839	1.3292
A8	0.2664	8.933	A53	0.0724	0.85
A16	0.0494	-0.1083	A54	0.0689	0.7047
A18	0.0564	0.1833	A58	0.2614	8.725
A19	0.0639	0.4958	B2	0.0724	0.85
A21	0.0569	0.2042	B5	0.0454	-0.275
A22	0.0774	1.5083	B11	0.0979	1.9125
A31	0.0499	-0.0875	B12	0.0429	-0.3792
A32	0.0609	0.3708	B17	0.0789	1.1208
A35	0.0564	0.1833	B21	0.0509	-0.0458
A36	0.0639	0.4958	B23	0.0449	-0.2958
A37	0.0734	0.8917	B24	0.1564	4.35

development. Furthermore, bacterial IAA contributes to increased plant tolerance to abiotic stresses like drought, salinity, and heavy metal toxicity, making it a promising agent for sustainable agriculture (Etesami and Glick, 2024). IAA promotes longer roots with more root hairs and lateral roots, improving nutrient uptake and leading to enhanced plant vigor (Datta and Basu, 2000).

Phosphate Solubilization Assay. Phosphatesolubilizing bacteria (PSB) convert insoluble forms of phosphorus (P) into plant-available forms. In this study, 24 isolates were tested for phosphate solubilization: 16 from weedy rice (Sample A) and 8 from cultivated rice (Sample B). Results showed that 10 isolates from weedy rice and five from cultivated rice exhibited phosphate solubilization activity (Table 4). This activity was evidenced by the formation of a
 Table 4. Phosphorus solubilization assay of the different isolates collected from weedy rice and rice samples.

Isolate	P Sorption Index (mm)	Isolate	P Sorption Index (mm)
A2	1.16	A48	1.1
A4	-	A53	-
A8	1.5	A54	1.14
A16	1.92	A58	-
A18	1.72	B2	1.19
A22	1.11	B5	1.15
A31	-	B11	-
A32	-	B12	1.61
A35	-	B17	-
A36	1.33	B21	1.25
A37	1.12	B23	1.6
A40	1.13	B24	-

production of enzymes, and release of siderophores, along with the synthesis of plant hormones like auxins, cytokinins, and gibberellic acid (Puri et al., 2020; Rawat et al., 2020).

Furthermore, PSBs contribute to nitrogen fixation and pathogen resistance, both of which support plant growth. Their ability to release phosphorus from insoluble or adsorbed forms is crucial for increasing P bioavailability (Habte, 2000). The capacity of microbial biomass to assimilate soluble P and prevent its fixation is well documented (Khan and Joergensen, 2009). Additionally, PSBs mineralize organic P and solubilize precipitated phosphates, thereby enhancing phosphorus uptake by plants (Chen et al., 2002; Kang et al., 2002; Pradhan and Sukla, 2006).

1-aminocyclopropane-1-carboxylate (ACC) Deaminase Activity Assay. Twenty-five bacterial isolates were tested for ACC deaminase activity. Among these, five isolates from weedy rice (Sample A) and three from cultivated rice (Sample B) tested positive (Table 5).

Bacteria that produce ACC deaminase play a significant role in mitigating plant stress responses by degrading ACC, the immediate precursor of ethylene - a stress-induced phytohormone. By lowering ethylene levels, these bacteria help reduce stress-induced growth inhibition (Glick et al., 1998). ACC deaminase catalyzes the conversion of ACC to α -ketobutyrate and ammonia, which can be used by the bacteria as carbon and nitrogen sources (Penrose and Glick, 1997). Studies have shown that these bacteria can enhance plant growth, water use efficiency, membrane stability, and accumulation of compatible solutes, proteins, phenolics, and photosynthetic pigments under stress conditions (Tiwari and Lata, 2018).

 Table 5. Results of the 1-aminocyclopropane-1-carboxylate

 (ACC) deaminase activity of the different isolates collected

 from weedy rice and rice samples.

Isolate	Reading	Isolate	Reading
A2	-	A48	-
A4	-	A53	-
A8	-	A54	+
A16	-	A58	+
A18	-	B2	-
A21	+	B5	+
A22	-	B11	+
A31	-	B12	-
A32	-	B17	-
A35	+	B21	-
A36	+	B23	+
A37	-	B24	-
A40	-		

Summary of Plant Growth Promoting Properties of the Different Isolates

Twenty-one bacterial isolates were evaluated for their plant growth-promoting (PGP) properties, including 14 strains from weedy rice and 7 from cultivated rice (Table 6). The identified PGP traits such as indole-3-acetic acid (IAA) production, ACC deaminase activity, phosphate solubilization, and starch hydrolysis suggest the presence of microbial mechanisms that enhance plant access to soil nutrients. These traits support the notion that weedy rice more efficiently utilizes rhizosphere resources. Microorganisms with these capabilities can activate several metabolic pathways, including those associated with nitrogen and phosphorus acquisition and stress resilience, contributing to improved plant growth and resistance to disease (Shi et al., 2018; Vrancken et al., 2013).

Wu et al. (2009) reported that the rhizosphere bacterial community is significantly more abundant in weedy rice compared to cultivated rice surrounded by weedy rice. This microbial abundance may provide a nutritional advantage to weedy rice in paddy soils. Additionally, rhizosphere communities like Xanthobacteraceae, found in cultivated rice, are associated with disease susceptibility, while Burkholderia may negatively influence the conversion of organic to inorganic nitrogen, potentially hindering rice growth (Li et al., 2021). Microbial symbiosis in the rhizosphere plays a vital role in promoting plant growth by influencing nutrient availability, yield, and crop quality (Olanrewaju et al., 2017; Zhalnina et al., 2018).

Weedy rice, being more phenotypically plastic and invasive than cultivated rice, demonstrates greater adaptability and nutrient uptake efficiency. These traits are linked to the composition of its rhizosphere bacterial community, which appears more stable and resilient to environmental changes (Yang et al., 2021). Comparative studies on co-occurrence networks between weedy and cultivated rice rhizospheres suggest that the former harbors more mutualistic microbes with positive interactions (Wang et al., 2006). For instance, Microbacteriaceae and Micrococcaceae in the weedy rice rhizosphere positively correlate with soil soluble organic nitrogen conversion, whereas Oxalobacteraceae in cultivated rice show a negative correlation (De la Cruz-Barrón et al., 2017; Obermeier et al., 2020). This may further explain the competitive advantage of weedy rice (Wu et al., 2009).

negative ACC deaminase activity, + positive ACC deaminase activity

Table 6. Summary of the plant growth promotion properties of the different isolates from weedy rice and cultivated rice, NSIC	i
Rc 222.	

Isolate	Source*	Starch Hydrolysis	IAA** Production	Phosphorus Solubilization	ACC*** deaminase activity
A2	AL	+	5.9958	1.16	-
A4	AL	-	0.2042	-	-
A8	AL	+	8.9333	1.5	-
A16	CA	+	-	1.92	-
A18	CA	+	0.1833	-	+
A22	CA	+	1.0583	1.11	-
A35	SR	+ + +	0.1833	1.72	+
A36	SR	-	0.4958	1.33	+
A37	SR	++	0.8917	1.12	-
A40	SR	+	4.0792	1.13	-
A48	SR	+	1.3292	1.1	-
A53	CA	+	0.85	-	-
A54	CA	-	0.7042	1.14	+
A58	SR	-	8.725	-	+
B2	AL	-	0.85	1.19	-
B5	SR	+	-0.275	1.15	+
B11	AL	+	1.9125	-	+
B12	AL	+	-0.3792	1.61	-
B17	AL	+	1.1208	-	-
B23	SR	-	-0.2958	1.6	+
B24	SR	+	4.35	-	-

*Source: AL – Aliaga, SR – San Roque, CA – Calzada

**IAA - Indole-3-acetic acid

***ACC -1-aminocyclopropane-1-carboxylate

Rhizosphere microbial composition is strongly influenced by host plant type and its environmental context (Yin et al., 2021). Plants release photosynthetically-fixed carbon via root exudates, which serve as energy sources for soil microbes, reinforcing plant-microbe associations (Li et al., 2019). Another mechanism enhancing microbial diversity in weedy rice is pathogen-mediated recruitment, where plants secrete specific root exudates to attract protective microbes, thus suppressing pathogens and enhancing productivity (Yuan et al., 2018; Dudenhoffer et al., 2016; Li et al., 2021). These protective microbial networks may also explain the relatively low incidence of plant diseases in natural ecosystems (Hacquard et al., 2017).

Identification of Bacterial Isolates through DNA Sequencing and Basic Local Alignment Search Tool Analysis (BLAST)

DNA sequencing was performed on the bacterial isolates, with the first 20 and last 200 bases manually trimmed using Chromas. Forward and reverse sequences were aligned using BioEdit. Consensus sequences were then analyzed using the Basic Local Alignment Search Tool (BLAST) through the National Center for Biotechnology Information (NCBI) platform.

As shown in Table 7, most of the identified isolates belong to the *Bacillus* genus, which is well-known for its beneficial role in plant growth, soil health, and pest management. These bacteria reduce dependence on synthetic inputs, promoting sustainable agricultural practices (El Hadrami, 2023). *Bacillus spp.* are Grampositive, low G+C content bacteria (*Phylum Firmicutes*; Class Bacilli; Order Bacillales; Family Bacillaceae) and are closely related to *Listeria*, *Streptococcus*, and *Staphylococcus* (Ciccarelli et al., 2006). These species are common in the rhizosphere and exhibit high tolerance to environmental stress (Sass and Parkes, 2009).

Members of the *Phylum Firmicutes* have been extensively studied for their roles in sustainable agriculture. They enhance plant nutrient acquisition (e.g., nitrogen, phosphorus, iron), modulate plant hormone levels, and contribute to pathogen suppression (Aguilar-Paredes et al., 2023; Hashmi et al., 2020). For example, *Bacillus megaterium* is

Table 7. Top 5 BLAST hits for the consensus sequences that were aligned with the nucleotide collection (nr/nt) database

Sample Number	Description	% Identity
	Bacillus aryabhattai strain RPW7 16S ribosomal RNA gene, partial sequence	99.86
	Bacillus aryabhattai strain N7 16S ribosomal RNA gene, partial sequence	99.86
A2	Bacillus megaterium strain FK4 16S ribosomal RNA gene, partial sequence	99.86
	Bacillus megaterium strain DK2 16S ribosomal RNA gene, partial sequence	99.86
	Bacillus megaterium strain DW13 16S ribosomal RNA gene, partial sequence	99.86
	Uncultured Bacillus sp. clone T328d75 16S ribosomal RNA gene, partial sequence	99.86
	Bacillus megaterium strain JD-7 16S ribosomal RNA gene, partial sequence	99.86
A18	Bacillus aryabhattai strain MER_75 16S ribosomal RNA gene, partial sequence	99.86
	Bacillus aryabhattai strain JPR41 16S ribosomal RNA gene, partial sequence	99.86
	Bacillus aryabhattai strain 1-Sj-5-1-6-M 16S ribosomal RNA gene, partial sequence	99.86
	Bacillus subtilis strain MDA2 16S ribosomal RNA gene, partial sequence	99.64
	Bacillus subtilis strain MDA1 16S ribosomal RNA gene, partial sequence	99.64
\22	Bacillus siamensis strain 64X-5 16S ribosomal RNA gene, partial sequence	99.64
	Bacillus siamensis strain 56X-1 16S ribosomal RNA gene, partial sequence	99.64
	Bacillus siamensis strain TW2-22 16S ribosomal RNA gene, partial sequence	99.64
	Uncultured Phenylobacterium sp. clone b2-194 16S ribosomal RNA gene, partial sequence	99.10
	Phenylobacterium composti strain 5-39-3-1 16S ribosomal RNA gene, partial sequence	98.80
\35	Phenylobacterium composti gene for 16S rRNA, partial sequence, strain: NBRC 106419	98.13
	Phenylobacterium composti DSM 19425 strain 4T-6 16S ribosomal RNA, partial sequence	98.06
	Phenylobacterium sp. 4T06 16S ribosomal RNA gene, partial sequence	98.06
	Bacillus megaterium strain A plasmid p2, complete sequence	99.86
	Bacillus megaterium strain A chromosome, complete genome	99.86
35	Bacillus aryabhattai strain LW-04 16S ribosomal RNA gene, partial sequence	99.86
	Bacillus sp. (in: Bacteria) strain SNH_K84 16S ribosomal RNA gene, partial sequence	99.86
	Bacillus sp. (in: Bacteria) strain NMS1 16S ribosomal RNA gene, partial sequence	99.86
	Bacillus megaterium strain A plasmid p2, complete sequence	99.78
	Bacillus megaterium strain A chromosome, complete genome	99.78
B12	Bacillus megaterium strain NRCB001 16S ribosomal RNA gene, partial sequence	99.78
	Bacillus aryabhattai strain LW-04 16S ribosomal RNA gene, partial sequence	99.78
	Bacillus sp. (in: Bacteria) strain SNH_K84 16S ribosomal RNA gene, partial sequence	99.78
	Bacillus subtilis strain CL2 16S ribosomal RNA gene, partial sequence	98.99
	Bacillus subtilis strain CL2 16S ribosomal RNA gene, partial sequence	98.99
317	Bacillus sp. (in: Bacteria) strain BSWF 16S ribosomal RNA gene, partial sequence	98.99
	Bacillus subtilis strain P9_B1 chromosome, complete genome	98.99
	Bacillus subtilis strain P8_B3 chromosome, complete genome	98.99
	Bacillus tequilensis strain Z36S 16S ribosomal RNA gene, partial sequence	88.22
	Bacillus tequilensis strain 111-4 16S ribosomal RNA gene, partial sequence	88.16
324	Bacillus amyloliquefaciens subsp. plantarum strain Kk5-11 16S ribosomal RNA gene, partial sequence	88.09
	Bacillus sp. KHR-38 16S ribosomal RNA gene, partial sequence	88.13
	Bacillus sp. LX-110 16S ribosomal RNA gene, partial sequence	88.15

known for nitrogen fixation and phosphate solubilization, facilitating phosphorus availability for plants. Some *Bacillus* strains also improve soil health and serve as bioremediation agents. B. megaterium has been used in combination with earthworms to remediate cadmium-contaminated soil (Wrobel et al., 2023), while B. subtilis SJ-101 has shown potential in the phytoaccumulation of heavy metals (Zaidi et al., 2006).

These bacteria can reduce environmental metal loads via biosorption, EPS-mediated mechanisms, bioaccumulation, or bioprecipitation, targeting heavy metals such as lead, cadmium, mercury, chromium, arsenic, and nickel (Pham et al., 2022; Shao et al., 2019). Bacillus aryabhattai has also been reported to enhance crop yields in saline-sodic soils (Tamilselvi et al., 2022), and B. subtilis contributes to calcite dissolution (Tamilselvi et al., 2018). Additionally, B. tequilensis functions as a biocontrol agent against various fungal pathogens in commercial crops (Kwon et al., 2022).

Other identified isolates include *Phenylobacterium spp.*, which are common soil bacteria with plant growth-promoting and defensive properties, including antibiotic synthesis (Gámez-Arcas et al., 2022). Their volatile compounds influence plant growth, microbial community structure, and hormonal regulation (Garbeva and Weisskopf, 2020). These microbes possess genetic pathways that enable them to produce antimicrobial compounds, allowing them to survive and function in complex soil ecosystems.

In-vitro evaluation of the effectiveness of a highly potential PGP isolate on seedling growth of weedy rice and cultivated rice under laboratory conditions

Isolates identified through DNA sequencing and BLAST analysis (Table 7) were evaluated, with particular focus on isolate A35, identified as *Phenylobacterium sp.*, which exhibited the most promising PGP activity among isolates derived from weedy rice (Table 6). This isolate was selected for further in-vitro testing.

Ten days after inoculation with *Phenylobacterium sp.*, scanning of shoot and root systems revealed a significant promotion of both shoot and root length in weedy rice compared with cultivated rice (Figure 2). Further measurements indicated variability in shoot and root lengths across all treatments and test materials. Among the uninoculated treatments, weedy rice A3WR(U) in mixed culture and A2WR(U) in monoculture had the longest shoot lengths at 8.33 cm and 8.25 cm, respectively (Figure 3). Inoculated weedy rice in monoculture (A2WRB) had the longest shoot length among treated materials at 6.98 cm,

followed by weedy rice in mixed culture (A3WRB) at 5.77 cm, which is comparable to the uninoculated rice in mixed culture (A3R(U), 5.67 cm). In contrast, inoculated monoculture rice (A1RB) and rice in mixed culture (A3RB) exhibited the shortest shoot lengths.



Figure 2. Root and shoot growth of cultivated rice NSIC Rc 222 and weedy rice WR SR 1 inoculated with *Phenylobacterium sp.*

Further measurements indicated variability in shoot and root lengths across all treatments and test materials. Among the uninoculated treatments, weedy rice A3WR(U) in mixed culture and A2WR(U) in monoculture had the longest shoot lengths at 8.33 cm and 8.25 cm, respectively (Figure 3). Inoculated weedy rice in monoculture (A2WRB) had the longest shoot length among treated materials at 6.98 cm, followed by weedy rice in mixed culture (A3WRB) at 5.77 cm, which is comparable to uninoculated rice in mixed culture (A3R(U), 5.67 cm). In contrast, inoculated monoculture rice (A1RB) and rice in mixed culture (A3RB) exhibited the shortest shoot lengths.

Root length data followed a similar trend. The longest roots were observed in uninoculated weedy rice grown in monoculture (A2WR(U), 6.21 cm) and mixed culture (A3WR(U), 6.62 cm). Among inoculated treatments, the weedy rice monoculture (AWRB) showed the longest roots. Generally, the weedy rice seedlings displayed faster and more vigorous growth than cultivated rice seedlings. This suggests that the superior growth performance of weedy rice may be partially explained by differences in microbial composition, especially in the rhizosphere, with root microbial genetics contributing to its rapid early growth (Juliyanti et al., 2024).





In addition to shoot and root lengths, root hair development was assessed using electron scanning at 400x magnification for both inoculated and uninoculated cultivated and weedy rice (Figure 4). Root hair density in weedy rice was visibly enhanced compared to that in cultivated rice, regardless of inoculation with isolate A35 (Phenylobacterium sp.), which was originally isolated from weedy rice (WR SR 1). This enhanced root system in weedy rice is attributed to increased lateral root length in both mono- and mixed-culture setups, facilitating improved water and nutrient uptake from the soil (Suralta et al., 2022). As lateral roots constitute the most active portion of the root system for water uptake and represent the majority of its total surface area (Yamauchi et al., 1996), greater lateral root development naturally supports denser root hair growth.

The increased root hair density observed in weedy rice may indicate a superior capacity for absorbing growth-promoting substances from the soil. Root hairs, which are simple extensions formed at the root apical meristem, enhance the root surface area and establish direct contact with the soil-playing a vital role in water and nutrient absorption from the rhizosphere (Chouhan et al., 2023). In some plant species, root hairs constitute up to 77% of the root surface area, dominating the rhizosphere and facilitating nutrient uptake (Bertin et al., 2003). Their slender structure aids in penetrating soil edges (Mandel and Yanofsky, 1995) and in mediating interactions with soil microbes. Root hairs are also involved in root nodule formation in legumes (Dazzo et al., 1984), extend the functional radius of the root,

of phosphorus (Cai and Ahmed, 2022). During waterlimited conditions, root hairs contribute to the plant's ability to extract water from drier soils. The efficient nutrient uptake and competitive traits

and significantly enhance nutrient uptake, particularly

of weedy rice have been well-documented. Studies by Burgos et al. (2006) and Sales et al. (2011) have shown that weedy rice can access and utilize rhizosphere nutrients more efficiently than cultivated rice. This advantage has been partly attributed to its finer roots, which are approximately six times longer and possess six times more root tips than those of cultivated rice. With a larger root surface area and greater absorptive capacity, weedy rice can outcompete cultivated rice for essential growth resources. The presence of microorganisms such as PGP rhizobacteria may further support these traits. The current study's findings are consistent with these earlier conclusions.

Conclusion

A greater number of plant growth-promoting rhizobacteria (PGPR) were isolated from weedy rice compared to cultivated rice. This finding supports the hypothesis that PGPR associated with weedy rice enhance its competitive abilities in cultivated rice ecosystems. Studies on rhizosphere microbiomes of cultivated, wild, and weedy rice have revealed diverse microbial communities, suggesting that host plants selectively recruit specific microbial populations. The presence of microorganisms with beneficial metabolic pathways in the rhizosphere of weedy rice could explain the nutrient uptake disadvantage experienced by cultivated rice.



Figure 4. Results of root scanning (400x) of cultivated rice and weedy rice showing roots hairs of uninoculated (a) and inoculated samples (b).

PGPR strains not only support plant growth but may also protect weedy rice from pathogens and environmental stressors such as heavy metals or light toxicity in paddy fields. Investigating rhizosphere microbial communities offers valuable insights into the competitive mechanisms employed by weedy rice and other weedy species.

In this study, the application of a PGP bacterium (*Phenylobacterium sp.*) isolated from weedy rice promoted early seedling vigor and enhanced shoot and root growth in weedy rice. Early-stage root analysis confirmed that weedy rice exhibited generally longer root systems with more robust root hairs than cultivated rice, increasing its potential for nutrient and water absorption - resources meant for the cultivated crop.

Weedy rice significantly impacts rice production by reducing yield due to its strong competitive traits. Its similarity in agro-morphological traits to cultivated rice makes it particularly difficult to manage. The results suggest that early seedling vigor and competition should be key targets in agronomic strategies for weedy rice control. Managing weedy rice before flowering can help prevent seed production and reduce its long-term presence in the field.

Cultural management techniques such as the use of a stale seedbed to deplete the soil seed bank and ensuring the use of clean seeds are critical. Given the natural herbicide tolerance of weedy rice, ongoing efforts to identify and develop selective herbicides should continue. Additionally, breeding for cultivated rice varieties with enhanced seedling vigor and robust root systems is recommended.

Lastly, increasing farmer awareness regarding the impact of weedy rice on rice production is essential. Education campaigns and farmer training should emphasize the importance of early intervention and integrated weed management strategies to reduce the competitive threat of weedy rice.

Literature Cited

- Aguilar-Paredes A, Valdés G, Araneda N, Valdebenito E, Hansen F, Nuti M (2023) Microbial community in the composting process and its positive impact on the soil biota in sustainable agriculture. Agronomy 13(2): 542
- Amri M, Rjeibi MR, Gatrouni M, Mateus DMR, Asses N, Pinho HJ, Abbes C (2023) Isolation, identification, and characterization of phosphate-solubilizing bacteria from Tunisian soils. Microorganisms 11(3): 783
- Anand K, Kumari B, Mallick MA (2016) Phosphate solubilizing microbes: An effective and alternative approach as biofertilizers. Int. J. Pharm. J. Pharm. Sci. 8(2): 37 - 40
- Bertin C, Yang X, Weston LA (2003) The role of root exudates and allelochemicals in the rhizosphere. Plant and Soil 256: 67 - 83
- Bird R, Hopkins RH (1954) The mechanism of beta-amylase action. 2. Multichain action on amylose fission products. Biochemical Journal 56(1): 140 - 146
- Burgos NR, Norman RJ, Gealy DR, Black H (2006) Competitive N uptake between rice and weedy rice. Field Crops Research 99(2-3): 96 - 105
- Cai G, Ahmed MA (2022) The role of root hairs in water uptake: recent advances and future perspectives. Journal of Experimental Botany 73(11): 3330 - 3338
- Cassman K, Peng S, Olk DC, Ladha J, Reichardt W, Doberman A, Singh U (1998) Opportunities for increased nitrogenuse efficiency from improved resource management in irrigated rice systems. Field Crops Research 56(1-2): 7 - 39
- Chen CR, Condron LM, Davis MR, Sherlock RR (2002) Phosphorus dynamics in the rhizosphere of perennial ryegrass (*Lolium perenne* L.) and radiata pine (*Pinus radiata* D. Don.). Soil Biol Biochem 34(4): 487 - 499
- Chouhan R, Nautiyal AK, Sharma N, Gandhi SG (2023) Plant transcription factors and root development. In Eds: Srivastava V, Mishra S, Mehrotra S, Upadhyay SK, Plant Transcription Factors, Academic Press (pp 63 - 76). Retrieved from https://doi.org/10.1016/B978-0-323-90613-5.00007-8 (accessed September 25, 2024)
- Ciccarelli FD, Doerks T, von Mering C, Creevey CJ, Snel B, Bork P (2006) Toward automatic reconstruction of a highly resolved tree of life. Science 311(5765): 1283 -1287
- Collins C, Lyne P, Grange J, Falkinham III JO (2004) Collins and Lyne's Microbiological Methods. 8th Ed. London: Arnold, a member of the Hodder Headline Group (pp 456). Retrieved from https://mmstcchemistry.weebly.com/ uploads/2/4/1/2/24121933/microbiological_methods.pdf (accessed June 17, 2024)
- Cruz JA, Paterno ES (2014) Isolation and screening of actinomycetes for its growth promoting activities. Asia Life Sci. 23: 413 - 428
- Datta C, Basu PS (2000) Indole acetic acid production by a Rhizobium species from root nodules of a leguminous shrub, *Cajanus cajan*. Microbiol Res. *155*(2): 123 - 127

- Dazzo FB, Truchet GL, Sherwood JE, Hrabak EM, Abe M, Pankratz SH (1984) Specific phases of root hair attachment in the *Rhizobium trifolii*-clover symbiosis. Appl Environ Microbiol 48(6): 1140 - 1150
- De la Cruz-Barrón M, Cruz-Mendoza A, Navarro-Noya YE, Ruiz-Valdiviezo VM, Ortíz-Gutiérrez D, Ramírez-Villanueva DA, Luna-Guido M, Thierfelder C, Wall PC, Verhulst N, Govaerts B, Dendooven L (2017) The bacterial community structure and dynamics of carbon and nitrogen when maize (*Zea mays* L.) and its neutral detergent fibre were added to soil from Zimbabwe with contrasting management practices. Microb Ecol. 73(1): 135 - 152
- Dudenhöffer JH, Scheu S, Jousset A (2016). Systemic enrichment of antifungal traits in the rhizosphere microbiome after pathogen attack. J. Ecol. 104: 1566 - 1575
- El Hadrami A (2023) The use of *bacillus* microorganisms in agriculture. Retrieved from https://omexcanada.com/blog/ the-use-of-bacillus-microorganisms-in-agriculture (accessed October 11, 2024)
- Etesami H, Glick B (2024) Bacterial indole-3-acetic acid: A key regulator for plant growth, plant-microbe interactions, and agricultural adaptive resilience. Microbiological Research, 281: 127602
- Gámez-Arcas S, Baroja-Fernández E, García-Gómez P, Muñoz
 FJ, Almagro G, Bahaji A, Sánchez-López ÁM, Pozueta Romero J (2022) Action mechanisms of small microbial volatile compounds in plants. J. Exp. Bot. 73(2): 498 510
- Garbeva P, Weisskopf L (2020) Airborne medicine: bacterial volatiles and their influence on plant health. New Phytol. 226(1): 32 - 43
- Glick BR, Penrose DM, Li J (1998) a model for the lowering of plant ethylene concentrations by plant growth-promoting bacteria. J. Theor. Biol. 190: 63 - 68
- Guzmán-Moreno J, García-Ortega LF, Torres-Saucedo L, Rivas-Noriega P, Ramírez-Santoyo RM, Sánchez-Calderón L, Quiroz-Serrano IN, Vidales-Rodríguez LE (2022) Bacillus megaterium HgT21: A promising metal multiresistant plant growth-promoting bacteria for soil biorestoration. Microbiology Spectrum 10(5): e0065622
- Habte M (2000) Mycorrhizal fungi and plant nutrition. In: Plant Nutrient Management in Hawaii's Soils, Approaches for Tropical and Subtropical Agriculture (pp 127 - 131). Hawaii: University of Hawaii at Manoa. Retrieved from https://www.ctahr.hawaii.edu/oc/ freepubs/pdf/pnm0.pdf (accessed October 25, 2024)
- Hacquard S, Spaepen S, Garrido-Oter R, Schulze-Lefert P (2017) Interplay between innate immunity and the plant microbiota. Annu. Rev. Phytopathol. 55: 565 - 589
- Hashmi I, Bindschedler S, Junier P (2020) Firmicutes. In Beneficial Microbes in Agro-Ecology (pp 363 - 396). Academic Press: Cambridge, MA, USA
- Islam MT, Hossain MM (2012) Plant probiotics in phosphorus nutrition in crops, with special reference to rice (pp 325 -363). *In:* Maheshwari, DK (ed.), Bacteria in Agrobiology: Plant Probiotics. Berlin: Springer

- Juliyanti V, Itakura R, Kotani K, Lim SY, Suzuki G, Chong CW, Song BK, Rahman S (2024) Comparative analysis of root associated microbes in tropical cultivated and weedy rice (*Oryza* spp.) and temperate cultivated rice. Scientific Reports 14: 9656
- Kang SC, Hat CG, Lee TG, Maheshwari DK (2002) Solubilization of insoluble inorganic phosphates by a soilinhabiting fungus *Fomitopsis* sp. PS 102. Current Science 82(4): 439 - 442
- Keswani C, Singh SP, Cueto L, Garcia-Estrada C, Mezaache-Aichour S, Glare TR, Boriss R, Singh SP, Blazquez MA, Sansinea E (2020) Auxins of microbial origin and their use in agriculture. Applied Microbiology and Biotechnology 104: 8549 - 8565
- Khan KS, Joergensen RG (2009) Changes in microbial biomass and P fractions in biogenic household waste compost amended with inorganic P fertilizers. Bioresource Technology 100(1): 303 - 309
- Kwon HT, Lee Y, Kim J, Balaraju K, Kim HT, Jeon Y (2022) Identification and characterization of bacillus tequilensis GYUN-300: an antagonistic bacterium against red pepper anthracnose caused by *Colletotrichum acutatum* in Korea. Front. Microbiol. *13*: 1 - 18
- Li BB, Roley SS, Duncan DS, Guo J, Quensen JF, Yu HQ, Tiedje JM (2021) Long-term excess nitrogen fertilizer increases sensitivity of soil microbial community to seasonal change revealed by ecological network and metagenome analyses. Soil Biology and Biochemistry, *160*: 108349
- Li X, Jousset A, de Boer W, Carrión VJ, Zhang T, Wang X, Kuramae EE (2019) Legacy of land use history determines reprogramming of plant physiology by soil microbiome. ISME J. 13(3): 738 - 751
- Mandel MA, Yanofsky MF (1995) The *Arabidopsis AGL8 MADS* box gene is expressed in inflorescence meristems and is negatively regulated by APETALA1. The Plant Cell 7 (11): 1763 - 1771
- Nadeem SM, Ahmad M Zahir ZA, Javaid A, Ashraf M (2014) The role of mycorrhizae and plant growth promoting rhizobacteria (PGPR) in improving crop productivity under stressful environments. Biotechnology Advances 32(2): 429 - 448
- Obermeier MM, Gnädinger F, Durai Raj AC, Obermeier WA, Schmid CAO, Balàzs H, Schröder P (2020) Under temperate climate, the conversion of grassland to arable land affects soil nutrient stocks and bacteria in a short term. Science of the Total Environment 703: 135494
- **Olanrewaju OS, Glick BR, Babalola OO** (2017) Mechanisms of action of plant growth promoting bacteria. World J Microbiol Biotechnol. *33:* 1 6
- Patten CL, Glick BR (1996) Bacterial biosynthesis of indole-3acetic acid. Can J Microbiol. 42(3): 207 - 220
- Penrose DM, Glick BR (1997) Enzymes that regulate ethylene levels - 1-aminocyclopropane-1-carboxylic acid (ACC) deaminase, ACC synthase and ACC oxidase. Ind. J. Exptl. Biol. 35: 1 - 17
- Pham VHT, Kim J, Chang S, Chung W (2022) Bacterial biosorbents, an efficient heavy metals green clean-up strategy: Prospects, challenges, and opportunities. Microorganisms 10(3): 610

- Pradhan N, Sukla LB (2006) Solubilization of inorganic phosphate by fungi isolated from agriculture soil. African J. Biotechnol. 5(10): 850 - 854
- Puri A, Padda KP, Chanway CP (2020) In vitro and in vivo analyses of plant-growth-promoting potential of bacteria naturally associated with spruce trees growing on nutrientpoor soils. Appl. Soil Ecol. 149: 103538
- Rawat P, Shankhdhar D, Shankhdhar SC (2020) Plant growthpromoting rhizobacteria: A booster for ameliorating soil health and agriculture production. *In* Soil Health. Soil Biology Vol 59: 47 - 68
- Sales MA, Burgos NR, Shivrain VK, Murphy B, Gbur EE Jr. (2011) Morphological and physiological responses of weedy red rice (*Oryza sativa* L.) and cultivated rice (*O. sativa*) to N supply. American Journal of Plant Sciences 2: 569 - 577
- Sánchez-Navarro V, Zornoza R, Faz Á, Egea-Gilabert C, Ros M, Pascual JA, Fernández JA (2020) Inoculation with different nitrogen-fixing bacteria and arbuscular *mycorrhiza* affects grain protein content and nodule bacterial communities of a fava bean crop. Agronomy 10(6): 768
- Sass H, Parkes RJ (2009) Deep Sub-Surface (pp 64-79). Encyclopedia of Microbiology (Third Edition). Academic Press. Retrieved from https://doi.org/10.1016/B978-012373944-5.00275-3 (accessed June 10, 2024)
- Shahab S, Ahmed N, Khan NS (2009) Indole acetic acid production and enhanced plant growth promotion by indigenous PSBs. African Journal of Agricultural Research 4(11): 1312 - 1316
- Shao W, Li M, Teng Z, Qiu B, Huo Y, Zhang K (2019) Effects of Pb (II) and Cr (VI) stress on phosphate-solubilizing bacteria (*Bacillus* sp. strain MRP-3): Oxidative stress and bioaccumulation potential. Int. J. Environ. Res. Public Health. 16(12): 2172
- Sharma SB, Sayyed RZ, Trivedi MH, Gobi TA (2013) Phosphate solubilizing microbes: Sustainable approach for managing phosphorus deficiency in agricultural soils. Springer Plus 2: 1 - 4
- Shi S, Tian L, Nasir F, Li X, Li W, Tran LS, Tian C (2018) Impact of domestication on the evolution of rhizomicrobiome of rice in response to the presence of *Magnaporthe oryzae*. Plant Physiology and Biochemistry 132: 156 - 165
- Suralta RR, Montero BI, Paner JM, Santiago TY, Urbano MG, Cabral MC, Donayre DK, Asinas FT, Mojica JC, Martin EC, Niones JM (2022) Competition between rice and weedy rice under different water regimes. Philippine Journal of Science 151(5): 1749 - 1759
- Tamilselvi SM, Thiyagarajan C, Uthandi S (2018) Calcite dissolution by *Bacillus subtilis* SSRCI02: An invitro study for the reclamation of calcareous saline-sodic soils. Indian Journal of Geo Marine Sciences 47(06): 1267 - 1273
- Tamilselvi SM, Thiyagarajan C, Elumalai V, Uthandi S (2022) Microbial behavior, responses toward salinity stress, mechanism of microbe-mediated remediation for sustainable crop production (pp 103 - 127).Mitigation of Plant Abiotic Stress by Microorganisms Applicability and Future Directions. Retrieved from https://doi.org/10.1016/ B978-0-323-90568-8.00006-7 (accessed October 12, 2024)

- Tiwari S, Lata C (2018) Heavy metal stress, signaling, and tolerance due to plant-associated microbes: An overview. Front. Plant Sci. 9: 452
- Vrancken K, Holtappels M, Schoofs H, Deckers T, Valcke R (2013) Pathogenicity and infection strategies of the fire blight pathogen *Erwinia amylovora* in *Rosaceae*: state of the art. Microbiology 159(5): 823 - 832
- Wang G, Jin J, Xu M, Liu X (2006) Effects of plant, soil and soil management on soil microbial community diversity. Chinese Journal of Ecology 25(5): 550 - 556
- Wróbel M, Śliwakowski W, Kowalczyk P, Kramkowski K, Dobrzyński J (2023) Bioremediation of heavy metals by the genus *Bacillus*. Int J Environ Res Public Health 20(6): 4964
- Wu D, Hugenholtz P, Mavromatis K, Pukall R, Dalin E, Ivanova NN, Kunin V, Goodwin L, Wu M, Tindall BJ, Hooper SD, Pati A, Lykidis A, Spring S, Anderson IJ, D'haeseleer P, Zemla A, Singer M, Lapidus A, Nolan M, Copeland A, Han C, Chen F, Cheng JF, Lucas S, Kerfeld C, Lang E, Gronow S, Chain P, Bruce D, Rubin EM, Kyrpides NC, Klenk HP, Eisen JA (2009) A phylogeny-driven genomic encyclopedia of bacteria and archaea. Nature 462(7276): 1056 - 1060
- Yamauchi A, Pardales JR, Kono Y (1996) Root system structure and its relation to stress tolerance. *In*: Dynamics of roots and nitrogen in cropping systems of the semi-arid tropics (pp 211 - 233). Japan International Research Center for Agricultural Sciences, Tokyo, Japan
- Yang Y, Shi Y, Kerfahi D, Ogwu MC, Wang J, Dong K, Takahashi K, Moroenyane I, Adams JM (2021) Elevation-related climate trends dominate fungal cooccurrence network structure and the abundance of keystone taxa on Mt. Norikura, Japan. Sci Total Environ. 10(799): 149368

- Yin C, Casa Vargas JM, Schlatter DC, Hagerty CH, Hulbert SH, Paulitz TC (2021) Rhizosphere community selection reveals bacteria associated with reduced root disease. Microbiome 9: 1 - 8
- Yuan J, Zhao J, Wen T, Zhao M, Li R, Goossens P, Huang Q, Bai Y, Vivanco JM, Kowalchuk GA, Berendsen RL, Shen Q (2018) Root exudates drive the soil-borne legacy of aboveground pathogen infection. Microbiome 6(1): 1 - 2
- Zahir ZA, Arshad M, Frankenberger WT Jr. (2005) Plant growth promoting rhizobacteria application and perspectives in agriculture. Advances in Agronomy 81: 97 - 168
- Zaidi S, Usmani S, Singh BR, Musarrat J (2006) Significance of Bacillus subtilis strain SJ-101 as a bioinoculant for concurrent plant growth promotion and nickel accumulation in Brassica juncea. Chemosphere 64(6): 991 - 997
- Zhalnina K, Louie KB, Hao Z, Mansoori N, da Rocha UN, Shi S, Cho H, Karaoz U, Loqué D, Bowen BP, Firestone MK, Northen TR, Brodie EL (2018) Dynamic root exudate chemistry and microbial substrate preferences drive patterns in rhizosphere microbial community assembly. Nat Microbiol. 3(4): 470 - 480

ERGONOMICALLY DESIGNED BROWN RICE MACHINE: ADVANCING GENDER-INCLUSIVE AND HEALTH-CONSCIOUS RICE PROCESSING

ARNOLD S. JULIANO^{*}, JOEL A. RAMOS, PHOEBE R. CASTILLO, AND JOEY P. MIANO

Rice Engineering and Mechanization Division Philippine Rice Research Institute Maligaya, Science City of Muñoz, Nueva Ecija *Corresponding Author: asjuliano@philrice.gov.ph

Abstract

Brown rice contains health-promoting properties such as dietary fiber, micronutrients, and antioxidants that may help prevent diabetes, cancer, and cardiovascular diseases. It also promotes weight loss by supporting healthy bowel movement and increasing metabolic function. While brown rice is accessible to high-income consumers, it remains relatively expensive and is typically only available in supermarkets, making it less accessible to low-income households. To address this, PhilRice developed a gender-friendly, affordable, and easy-to-operate brown rice machine. The device is designed to meet the daily rice requirements (1 kg of uncooked brown rice) of a typical five-member household. The prototype utilizes a pair of rubber roll hullers mounted on a frame with a seat and pedals and can be operated manually or powered by a ¹/₂ hp electric motor, providing users with an option to exercise while milling rice.

The improved prototype demonstrated an average dehulling capacity of 1.98 kg/h with 75.40% recovery using manual power. When powered by a ½ hp electric motor, it yielded a capacity of 1.93 kg/h with a 79.64% recovery rate. At a competitive brown rice price of PhP50/kg, the manually-operated machine used for 150 hours annually generated a net income of PhP13,928, with a payback period of 1.45 years. Meanwhile, the electrically-powered version operating for 260 h per year yielded a net income of Php 8,055 and a payback period of 3.25 years.

Keywords: brown rice, brown rice machine, gender-friendly machine, motor-drive, pedal-type

Introduction

Brown rice, or unpolished rice, is produced through dehulling or dehusking, which removes only the outer husk while preserving the nutrient-rich bran and germ layers. It is not a specific rice variety; rather, all rice varieties can be classified as brown rice once the hull is removed. Traditionally produced using wooden mortar and pestle, brown rice is now also processed by commercial milling machines. Its nutritional profile helps address malnutrition as it contains dietary fiber, magnesium, iron, lysine, and protein. Brown rice yields about 10% higher milling recovery compared to polished rice, which could translate to an additional 1.76 million metric tons (MMT) of edible rice in the Philippines based on 2016 production data (PSA, 2017).

Economically, brown rice production requires less equipment and fuel and reduces labor inputs, as the polishing stage is eliminated (Cuyno, 2003). This further makes it an efficient and sustainable option for rice consumption.

Health Benefits of Brown Rice

Brown rice has gained popularity among healthconscious consumers, particularly high-income earners, due to its numerous health benefits. Rich in dietary fiber, it aids in weight loss, promotes healthy digestion, and supports metabolic function. It also contains B vitamins, phytic acid, and essential minerals (Ohtsubo et al., 2005). Dietary fiber acts as a laxative that helps prevent gastrointestinal diseases and diabetes, while B vitamins prevent beriberi. Brown rice is also an excellent source of selenium, a trace mineral known to reduce the risk of colon cancer through DNA repair and inhibition of cancer cell proliferation.

Moreover, brown rice is abundant in bioactive compounds such as γ -oryzanol, tocopherols, and tocotrienols, which synergistically reduce cancer and cardiovascular disease risks, lower cholesterol, and exhibit anti-inflammatory properties (Lin et al., 2015; Akihisa et al., 2000). It also contains gammaaminobutyric acid (GABA), phytic acid, vitamin E, and phenolic compounds like ferulic and p-coumaric acids. GABA, synthesized from L-glutamic acid, plays a critical role in neurological function and is associated with diseases such as Parkinson's and schizophrenia (Roohinejad et al., 2009). Phenolic compounds in brown rice, particularly those of high molecular weight, effectively scavenge free radicals, depending on structural characteristics like aromatic rings and hydroxyl group substitutions.

In recognition of its health benefits, the U.S. Food and Drug Administration (FDA) has categorized brown rice as a whole grain eligible for health claims related to the prevention of heart disease and cancer.

Brown Rice Commercialization in the Philippines

The Department of Agriculture-PhilRice, in partnership with the National Food Authority (NFA), initiated the "brown4good" project to promote responsible consumption through brown rice. The project also aimed to increase brown rice availability via NFA retail stores, which now sell brown rice at PhP37/kg.

Market scanning revealed that brown rice is primarily available in upscale supermarkets, targeting upper-class and health-conscious consumers. For example, Doña Maria's Jasponica Brown is sold at Walter Mart for PhP309 (2 kg), PhP 450 (5 kg), and PhP 1,205 (10 kg). Robinsons Supermarket offers Sunny Wood's Harvester's Healthy Brown Rice (PhP 137 for 2 kg) and Healthy You's Brown Rice (PhP 170.50 for 2 kg) while SM Supermarket offers Vita Rice at PhP302 per 5 kg.

Brown Rice Machine Development

Traditionally, brown rice is processed using a mortar and pestle (IRRI Rice Knowledge Bank). PhilRice has developed a commercial prototype brown rice milling machine with a capacity of 300 kg/ hour, which is integrated with a double-pass, 1-ton/ hour capacity rice mill. However, this capacity and cost are unsuitable for household use. Additionally, the need for proper storage facilities becomes a concern since brown rice turns rancid quickly under standard conditions. Producing just enough for 5-7 days' consumption (around 15 kg for a 5-member household) is a more practical approach. Though brown rice has a reported shelf life of 3-6 months, spoilage often begins within 1-2 weeks under typical storage conditions.

Generally, the study aimed to develop a portable, gender-friendly brown rice machine suitable for household use. Specifically, it aimed to (1) design and fabricate prototype of a portable brown rice machine; (2) determine the specific operational requirements for safe and efficient use of the technology; (3) identified necessary modifications to improve the prototype based on performance test, durability, and user preference; and (4) conducted a cost analysis of using and adopting the technology.

Materials and Methods

Development of concept design and prototype

Concept design

The design was based on weekly brown rice consumption for a five-member household. Assumptions included:

- One cup of cooked rice per person per meal
- 3 meals/day x 5 members = 15 cups/day
- 15 cups cooked = ~6.6 cups uncooked brown rice
- ~7 kg of brown rice/week

The machine was designed to process a minimum of 7 kg of paddy per batch and be safe for use by all household members, including younger individuals. The rubber roll mechanism uses two rollers rotating at different speeds, with one running 25% faster to produce a shearing action for husk removal. One roller is fixed; the other is adjustable for clearance control. The design also incorporates a pedaling mechanism to promote physical activity during use.

Fabrication of prototype

Fabrication was conducted at the REMD workshop using standard tools and locally available materials. Components such as belts, bearings, and rubber rollers were sourced from nearby suppliers, emphasizing simplicity, ease of procurement, and repairability.

Testing and evaluation

Performance tests used short- and long-grain rice samples. Measurements included input/output weight, milling time, weight of brown rice, unhulled grains, hulls, foreign matter, head rice, broken grains, and brewers. These data were used to compute capacity, purity, hulling efficiency, and other performance indicators.

Fine tuning

User feedback was collected during field demonstrations at farmer field days and similar events. Suggested modifications were incorporated into the final design to improve performance, durability, and user acceptance.

Cost analysis

Cost analysis was based on field test data. Fixed and variable costs were computed assuming a 10% salvage value, five-year equipment lifespan, and

Results and Discussion

Concept design development

The concept design was developed through benchmarking of similar existing equipment, followed by design enhancements using CAD software. The criteria guiding the design were:

- Simplicity and portability Easily fabricated with basic tools and transportable by three persons
- 2. Affordability Comparable in cost to a stationary exercise bike
- 3. Gender-friendliness Usable by both men and women
- Local manufacturability Built using materials and tools commonly found in local workshops
- 5. Household application Usable by any family member while engaging in light exercise

 Sufficient capacity - Produces at least 1 kg of brown rice per hour, enough for one day's family consumption

Description and Features of the Brown Rice Machine

The prototype brown rice machine is manually or electrically powered and features a simple, userfriendly design suitable for household use (Figure 1). It consists of a rubber roll huller assembly mounted on a metal frame with a seat and pedals. Users may either pedal manually or switch to electric motor power. The machine can produce the daily brown rice requirement for a five-member household (approximately 1 kg of uncooked rice per hour), making it both a foodprocessing and fitness device. The machine is lightweight, affordable, made from locally sourced components, and easy to operate and maintain.

An electric motor was also provided to drive the machine, especially to accommodate users who prefer not to operate it manually, such as senior individuals (Figure 2). A clutch mechanism was installed between the momentum weights and the 12-inch pulley, enabling the connection and disconnection of two rotating shafts (Figure 3). This clutch mechanism



Figure 1. Manual/motor driven brown rice machine.



Figure 2. Improved prototype with electric motor.



Figure 3. Clutch lever for shifting of drive.



Figure 4. Hand towel, cellphone, and bottled water holders.

typically consisted of two round plates - one fixed to the drive shaft and the other to the driven shaft.

Following a series of tests, refinements were introduced to improve operator comfort. These included the addition of holders for a hand towel, cellphone, and bottled water (Figure 4).

Modifications were also made to the hopper and collecting buckets (Figure 5). The original hopper had a capacity of 2.5 kg of paddy, which was found excessive for household brown rice requirements. Thus, the hopper was reduced to a 1 kg capacity to

match the daily consumption needs of an average household. The discharge buckets for brown rice and rice husk were also adjusted to accommodate 1 kg.

The input roller was improved by adding a slat to its middle section (Figure 6), which increased the feed rate of the paddy. The original roller had only one slat, which resulted in slower feeding.

The partition guide, which helps separate brown rice from rice husk, was also revised (Figure 7). Initial tests showed that excessive rice husk was ending up in the brown rice collection bucket. To resolve this, the



Figure 5. Modified hopper (left) and collecting buckets (right).



Figure 6. Modified input roller (right).



Figure 7. Originally designed partition arrangement (left) and modified version (right).

Table 1. Problems encountered during testing of the prototype.

Problems Encountered	Cause	Action Taken
High amount of unhulled paddy	Incorrect clearance between the 2 rubber roll hullers (too wide or too narrow).	Adjusted the clearance using the rubber roll clearance adjustment knob.
Brown rice deposited in the rice husk bucket and cyclone	High airflow rate.	Reduced fan speed; Make another pass to recover brown rice.
High quantity of rice husk in the brown rice discharge bucket	Low fan airflow; Wide partition guides/ outlet chute that aid from other impurities when dropping into the collecting bucket.	Increased fan speed; Reduced the length of the outlet chutes to provide enough access for rice husk to reach the rice husk discharge bucket.
Slow feeding of paddy	Lack of slat of input roller.	Added slat in the middle section of input roller to increase the amount of paddy being taken by the machine.
Big hopper size	Too big to hold the needed amount of paddy which is enough for a household.	Reduced hopper size to accommodate the required amount (1kg).
Low capacity of collecting buckets	Low amount of output is being hold and other impurities that dropped to the collecting buckets.	Increased size of collecting boxes which is enough to hold 1kg of brown rice.

length of the partition guide was shortened, accelerating the movement of husk toward the cyclone and reducing husk contamination in the brown rice output.

Additional issues encountered during the improvement phase, along with the corresponding corrective actions, are detailed in Table 1. These included, among other issues, the presence of unhulled paddy in the rice husk discharge bucket and a high volume of brown rice ending up in the husk discharge bucket and cyclone.

Endurance Testing

Endurance or extended-duration testing of the manual/motor-driven brown rice machine was conducted at PhilRice CES after refinements were completed. The prototype was tested by both male and female operators and was also evaluated using a ½ hp electric motor as the power source. Performance indicators included brown rice recovery and output capacity.

When operated manually, the machine produced 1.91 to 2.04 kg of brown rice per hour with a recovery rate of 73.84% to 76.96% after two passes, yielding a grain quality acceptable to consumers. When powered by an electric motor, the machine achieved a capacity of 1.93 kg/h with 79.64% recovery after two passes (Table 2). Slightly lower output was noted with the electric motor due to overheating during prolonged operation. Hence, it is recommended to use a higher-power motor for extended usage. Furthermore, higher throughput is expected when processing in bulk rather than in small, limited samples.

 Table 2. Performance of portable brown rice machine during endurance test.

Devenueter	Manua	Motor	
Parameter	Male	Female	Driven
Input paddy, kg	6.35	2.82	9.27
Brown Rice, kg	16.80	7.30	22.50
Operating time, h	12.93	5.39	17.92
BR Recovery, %	76.96	73.84	79.64
BR Output Capacity, kg/h	2.04	1.91	1.93

In addition, eight individuals operated the machine - six males (75%) and two females (25%). The mean age of the operators was 34 years (Table 3).

Table 3. Profile of operators running the
machine during testing period.

Operator	Mean	Percentage (%)
Sex		
Male		75
Female		25
Age	34	
21-25		25
26-30		25
31-35		25
36-40		12.5
41-45		0
46-50		12.5

Cost Analysis

The initial investment for a manually-operated brown rice machine is PhP 20,200 (Table 4). Assuming a brown rice selling price of PhP 50 kg⁻¹ and usage of 150 hours per year, the machine would yield an average net income of PhP 13,928 with a payback period of 1.45 years.

For the motor-driven version, the initial investment rises to PhP 26,200. At the same price point of PhP 50 kg⁻¹ and assuming 260 hours of use per year, the net income is PhP 8,055 with a payback period of 3.25 years.

 Table 4. Cost analysis of the manual/motor driven brown rice machine.

Parameter	Manual driven	Motor driven
Investment cost, P yr ⁻¹	20,200	26,200
Fixed cost, P yr-1	8,686	10,447
Operating hours, h yr-1	150	260
Net income, P yr-1	13,928	8,055
Payback period, yr	1.45	3.25

Assumptions: PhP 50 kg-1 cost of brown rice; P19/kg dried paddy; 26% interest on Investment, 10% depreciation cost, 10% R&M cost, 3% Insurance cost; 1h operation per day; 5 days a week.

Conclusion and Recommendations

The developed brown rice machine prototype is a gender-friendly technology that promotes both food production and physical activity. The machine delivers a processing capacity of 1.91 to 2.04 kg h⁻¹ after two passes, with a recovery rate of 73.84% to 76.96% using manual operation. When powered by a $\frac{1}{2}$ hp electric motor, it achieves 1.93 kg/h with 79.64% recovery. These outputs are sufficient to meet the daily brown rice needs of a five-member household.

At a brown rice selling price of PhP 50 kg⁻¹, the manually-operated version provides a net income of PhP13,928 with a 1.45-year payback when used for 150 hours annually. The motor-driven version yields a net income of Php 8,055 with a 3.25-year payback at 260 hours per year. These promising results support pilot deployment in rural and urban households to further evaluate the technology's economic viability and social acceptability.

Acknowledgment

The authors extend their sincere appreciation to all individuals who contributed to the success of this project, including the researchers, shop technicians, and technical advisors. Gratitude is also given to the management of PhilRice for their financial support, channeled through the PhilRice's Rice Engineering and Mechanization Division.

Literature Cited

- Akihisa T, Yasukawa H, Yamaura M, Ukiya M, Kimura Y, Shimizu N, Arai K (2000) Triterpene alcohol and sterol ferulates from rice bran and their anti-inflammatory effects. Journal of Agricultural and Food Chemistry 48(6): 2313 -2319
- **Cuyno RV** (2003) The national campaign to combat hidden hunger through brown rice (pp 1 - 10). *In* Consultative meeting nutritional aspect of brown rice, Food & Nutrition Research Institute, Manila Philippines
- Lin YT, Pao CC, Wu ST, Chang CY (2015) Effect of different germination conditions on antioxidative properties and bioactive compounds of germinated brown rice. BioMed Research International 2015(1): 608761
- Nur Yuhasliza AR, Dang Lelamurni AR, Anisah Jamaluddin AJ, Shaiful Adzni Sharifuddin SA, Long K (2015) Bioactive compounds and antioxidant activity of rice bran fermented with lactic acid bacteria (pp 156 – 162). Retrieved from https://www.researchgate.net/publication/ 282269216_Bioactive_compounds_and_antioxidant_ activity_of_rice_bran_fermented_with_lactic_acid_ bacteria (accessed April 1, 2025)
- Ohtsubo KI, Suzuki K, Yasui Y, Kasumi T (2005) Bio-functional components in the processed pre-germinated brown rice by a twin-screw extruder. Journal of food composition and analysis 18(4): 303 - 316
- PSA [Philippine Statistics Authority] (2017) Palay production survey 2016. Retrieved from https://psada.psa.gov.ph/ catalog/113 (accessed April 1, 2025)
- Roohinejad S, Omidizadeh A, Mirhosseini H, Rasti B, Saari N, Mustafa S, Yusof RM, Hussin AS, Hamid A, Manap MY (2009) Effect of hypocholesterolemic properties of brown rice varieties containing different gamma aminobutyric acid (GABA) levels on Sprague-Dawley male rats. Journal of Food, Agriculture and Environment 7(3&4): 197 - 203
SILICON DEPOSITION IN RICE PLANT AS AFFECTED BY SILICATE-SOLUBILIZING BACTERIA

JAYVEE A. CRUZ-KITMA^{1,4*}, BRENDA S. TUBANA¹, HECTOR FAJARDO², AND DIEGO MAYORGA³

¹School of Plant, Environmental, and Soil Sciences, Louisiana State University AgCenter, Baton Rouge, LA, USA ²North Carolina State University, Raleigh, NC, USA ³Virginia State University, VA, USA ⁴Philippine Rice Research Institute, Science City of Muñoz, Nueva Ecija, Philippines *Corresponding Author: jayveeacruz@gmail.com

Abstract

The silicate-solubilizing bacterial (SSB) community plays a significant role in enhancing the availability of silicon (Si) in soils, yet its contribution to Si absorption by plants remains poorly understood. This study aimed to investigate the effect of SSB on Si uptake and deposition in rice leaves. Silica deposition was evaluated using Scanning Electron Microscopy coupled with Energy Dispersive X-ray analysis (SEM-EDX) and silicon mapping. Results revealed clear differences in Si distribution between Si-treated and untreated rice plants. Plants treated with Si were more erect and showed better growth than those without Si application. SEM-EDX proved useful in visualizing silica bodies on rice leaf surfaces. In Commerce silt loam soil, Si mapping showed a higher distribution of silica bodies on the adaxial leaf surface of rice treated with Si (via wollastonite) and SSB inoculated carriers, compared to uninoculated control plants. SEM-EDX analysis revealed that plants treated with SSB-inoculated rice hull had the highest number of silica bodies, with an equivalent Si content of 55% relative to C, O, and K. The lowest number of silica bodies was observed in uninoculated plants. Si content in plants treated with wollastonite, SSB in bagasse, and SSB in slag was 48%, 35%, and 40%, respectively. In Gigger silt loam, Si mapping showed higher silica deposition in plants treated with wollastonite (48%) and uninoculated control (46%), followed by SSB in rice hull (40%), bagasse (26%), and slag (37%). Leaf surfaces treated with wollastonite and SSB exhibited trichomes, wart-like protuberances, Si papillae, and dumbbell-shaped silica bodies. To further elucidate SSB's role under field conditions, future studies should utilize soils with low Si content and subject test plants to biotic and abiotic stresses. Field assessments are necessary to understand the interaction between soil types, silicon application, SSB inoculation, and rice productivity.

Keywords: rice, scanning electron microscope-energy dispersive x-ray (SEM-EDX), silicate-solubilizing bacteria (SSB), silicon, silica bodies

Introduction

Soil microorganisms play crucial roles in mineral dissolution and nutrient cycling, significantly influencing soil fertility (Uroz et al., 2007; Calvaruso et al., 2006; Ehrlich, 1996). Beneficial soil microbes, such as nitrogen-fixing and phosphate-solubilizing bacteria, are well-known plant growth-promoting agents. A lesser-known but equally important group, silicate-solubilizing bacteria (SSB), facilitates the conversion of insoluble silicates into bioavailable mono-silicic acid.

Silicon is a critical micronutrient for the optimal growth and stress resilience of many cereal crops, particularly rice (Brunings et al., 2009). Though not considered essential for higher plants, its role in enhancing plant defense and growth, especially in Si-accumulating species such as rice, is welldocumented (Ma and Takahashi, 2002; Liang et al., 2007; Lee et al., 2019). Despite its abundance in the earth's crust, Si predominantly exists in insoluble forms and becomes available to plants only through weathering or microbial solubilization (Naureen et al., 2015).

Current research on Si largely focuses on its contribution to soil fertility, with limited studies exploring its biological interactions via microorganisms. With increasing concerns about the environmental and financial sustainability of chemical fertilizers, biological alternatives like SSB offer promising, eco-friendly solutions. This study investigated the effect of SSB application using various organic carriers on Si uptake in rice plants grown in two different Louisiana soil types.

Materials and Methods

The soils were collected from fields grown to soybean, wheat, corn, sugarcane, and rice at various locations in Louisiana, U.S.A. Two soils were selected on the basis of Si content: Gigger silt loam (low Si, <40 mg kg⁻¹) and Commerce silt loam (medium Si, 40-100 mg kg⁻¹). Two rates of wollastonite fertilization (no wollastonite and with wollastonite (500 kg Si ha⁻¹) and SSB inoculation (uninoculated and inoculated) were used in this study.

Collection and Processing of Bulk Soil Samples

Soil collection and processing soil samples were collected from agricultural fields in Louisiana cultivated with soybean, wheat, corn, sugarcane, and rice. Two soils were selected based on Si content: Gigger silt loam (low Si, <40 mg kg⁻¹) and Commerce silt loam (medium Si, 40-100 mg kg⁻¹). Treatments included two wollastonite rates (0 and 500 kg Si ha⁻¹) and two SSB inoculation levels (uninoculated and inoculated).

Soils were collected from St. Gabriel (Commerce silt loam; coordinates: 30.2625, -91.09722) and Winnsboro (Gigger silt loam; coordinates: 32.1418, -91.6862). To prevent cross-contamination, soils were sieved (5 mm), homogenized, and air-dried. Composite samples were taken for chemical characterization.

Seed Surface Sterilization

Mermentau rice seeds were washed five times with tap water, soaked in 95% ethanol for 2.5 minutes, and rinsed five times with sterile distilled water. They were then treated with 30% bleach for 30 seconds, followed by five additional rinses with sterile distilled water.

Preparation of Inoculant Carriers and SSB Inoculants, and Inoculant Application

Sterilized carriers (slag, bagasse, rice hull; 100 g each) were inoculated with SSB cultured in tryptic soy broth (TSB) for 24 h at 28-30°C. Moisture content was adjusted to field capacity, and the inoculated carriers were incubated at room temperature for one week.

Surface-sterilized seeds were coated with the SSB-inoculated carriers for 30 min. Five seeds were planted 2.5 cm deep in each pot.

Fertilizer Application

Urea was applied at 267 kg ha⁻¹ based on LSU AgCenter recommendations. At the 2-3 leaf stage, urea was incorporated into the top 15 cm of soil followed by flooding.

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

Leaf samples were collected at the reproductive stage. Six small leaf sections per sample were analyzed using SEM-EDX to evaluate Si deposition on the adaxial leaf surface. Two leaf samples per replicate, with three subsamples each, were examined. SEM imaging was conducted at 1,500x and 2,000x magnification at 5.0 kV. EDX analysis followed at 20.0 kV. Elemental composition (%Si, %C, %O, %K) was quantified, and Si content was visually represented using green coloration. Si mapping was performed by calculating the total pixel area associated with Si deposition.

Results and Discussion

Figure 1 illustrates the morphological effects of Si treatment in rice plants. Si-treated plants were more erect and vigorous compared to untreated controls. Silicon accumulates in rice primarily as amorphous silica gel (SiO2 nH2O), with deposition observed in epidermal layers, vascular bundles, and sclerenchyma tissues (Kim et al., 2002; Agarie et al., 1996).



Figure 1. Rice plants without (-Si) and with silicon (+Si).

Scanning electron microscopy followed by energy dispersive X-ray analysis is useful in locating silica bodies on the leaf surface of rice plants. Figure 2 shows the cross sections of leaves (red arrow highlighting the presence of silica bodies or phytolith) of rice grown on soil treated with SSB using SEM-EDX. The SEM image shows silica bodies were mostly concentrated on the epidermal cell walls of rice leaf. Kim et al. (2002) reported that electron microscopy and in situ X-ray microanalysis provided evidence that Si was deposited within sub-epidermal tissues of Si-treated rice leaves in epidermal cell walls, middle lamellas, and intercellular spaces. In the epidermal cell wall structure, the most significant variations were observed between Si-treated rice plants and control plants. These results were also consistent with the findings of Yoshida et al. (1962). In the analysis by Agarie et al. (1996), as seen in the cross-section of Si-treated rice plants, leaf silica cells occurred in the epidermal layer above and below the vascular bundles. However, Silicon deposition was not limited to electron-dense epidermal cell walls and was also observed as polymorphic aggregates in middle lamella and intercellular spaces (Kim et al., 2002). Silicon is believed to be associated with components of the cell wall, such as polysaccharides and proteins (Carpita, 1996). Silicon is likely to be



Figure 2. Cross sections of rice leaves (red arrow highlighting the presence of silica bodies or phytolith) treated with SSB (a) and silica deposition in the adaxial leaf surface of rice using SEM-EDX at 5,000x (b) and 2,000x (c) magnification.

integrated into cell walls as Si-aromatic ring contacts in rice leaves between lignin and carbohydrate (Inanaga et al., 1995).

The deposition of silica in rice leaves was assessed using scanning electron microscopy coupled with energy-dispersive X-ray spectroscopy (SEM-EDX) and silicon (Si) mapping. Figures 3 and 4 present the SEM-EDX Si maps of the adaxial leaf surfaces of rice plants grown in Commerce silt loam and Gigger silt loam soils, respectively. The treatments included a control (no Si), wollastonite, and three different solidstate bacterial (SSB) inoculations using rice hull, bagasse, and slag as carriers.

SEM-EDX analysis revealed distinct differences in Si distribution between Si-treated and untreated plants. In Commerce silt loam, Si mapping visually showed a greater abundance of silica bodies on the adaxial leaf surfaces of plants treated with wollastonite and SSB-inoculated carriers, compared to the uninoculated control. Among the treatments, plants treated with SSB-inoculated rice hull exhibited the highest density of silica bodies, corresponding to a Si content of 55% relative to carbon (C), oxygen (O), and potassium (K) levels. In contrast, the control group had the lowest silica body distribution (30% Si). Other treatments yielded the following Si contents: wollastonite (48%), SSB in bagasse (35%), and SSB in slag (40%). In Gigger silt loam, a similar trend was observed. Wollastonite-treated plants exhibited the highest Si content (48%), followed closely by the control group (46%). Si contents in the SSB treatments were as follows: SSB in rice hull (40%), SSB in slag (37%), and SSB in bagasse (26%).

Several structural features such as trichomes, wart-like protuberances, Si papillae, and dumbbellshaped silica bodies were evident on the leaf surfaces of plants treated with either wollastonite or SSBinoculated carriers (Figures 3 and 4). Similar silica body structures on rice leaves were previously reported by Kim et al. (2002). Point analysis of the Si X-ray spectra confirmed the presence of Si within trichomes and wart-like protuberances. Conversely, epidermal regions lacking these structures, as well as stomatal guard cells, accumulated significantly less Si. Notably, high Si X-ray counts consistently aligned with protruded areas on the leaf surface.

Dumbbell-shaped silica bodies were particularly prominent in leaves of plants treated with SSBinoculated rice hull and bagasse. As leaves matured, these silica cells exhibited a gradual morphological transformation, indicating early flexibility in the silica cell wall that transitioned to a more rigid, sandwichlike structure over time, consistent with the findings of Zhang et al. (2013). The development of silica cells in the rice leaf epidermis involves two key processes: (1)



Figure 3. Scanning electron microscope-energy dispersive X-ray (SEM-EDX) silicon map of adaxial leaf surface of uninoculated (**or control – a**), with wollastonite (**b**), SSB-inoculated rice hull (**c**), SSB-inoculated bagasse (**d**), and SSB-inoculated slag (**e**) rice plants planted in Commerce silt loam soil. Values are %Si relative to %C, %O, and %K. SC, silica cell in dumbbell-shaped; SP, silicon papilla; SG, stomatal guard cell.



Figure 4. Scanning electron microscope-energy dispersive X-ray (SEM-EDX) silicon map of adaxial leaf surface of uninoculated (or control – a), with Wollastonite (b), SSB-inoculated rice hull (c), SSB-inoculated bagasse (d), and SSB-inoculated slag (e) rice plants planted in Gigger silt loam soil. Values are %Si relative to %C, %O, and %K. SC, silica cell in dumbbell-shaped; SP, silicon papilla; SG, stomatal guard cell.

lignification and silicification of the cell wall, resulting in a dumbbell shape, and (2) the formation of needle-like biosilica, initiated from the inner wall and expanding in a defined direction until the lumens are fully filled.

Despite these insights, the precise mechanisms regulating Si biomineralization in higher plants remain unclear and warrant further investigation. Silicon deposition in the cell walls and silica cells, particularly those formed within dumbbell structures, may play a crucial role in plant defense against biotic stress (Zhang et al., 2013). In untreated plants, Si was more diffusely distributed across the leaf surface, as previously noted by Ranganathan et al. (2006). In contrast, Si-treated plants exhibited a marked increase in Si content and larger silica cell sizes (Cai et al., 2008).

Conclusion

This greenhouse experiment evaluated the effectiveness of previously identified multi-SSB formulations, applied via various carriers, in enhancing Si content in rice. Semi-quantitative assessment of silica body distribution via SEM-EDX revealed distinct differences across soil types, with a notably higher concentration observed in rice grown in Commerce silt loam. The analysis confirmed increased Si deposition in treated versus untreated plants. Silicon-treated rice plants appeared more erect and less stunted compared to controls.

SEM-EDX proved to be an effective tool for visualizing silica body distribution on rice leaf surfaces. The findings suggest that SSB inoculation presents a promising approach for enhancing Si availability to crops. However, its full potential has yet to be realized. Future studies should optimize factors such as SSB concentration, application method, and timing. Additionally, trials using soils with inherently low Si content under stressful field conditions are recommended to better demonstrate the benefits of this technology.

Literature Cited

- Agarie S, Agata W, Uchida H, Kubota F, Kaufman PB (1996) Function of silica bodies in the epidermal system of rice (*Oryza sativa* L.): testing the window hypothesis. Journal of Experimental Botany 47(5): 655 - 660
- Brunings AM, Datnoff LE, Ma JF, Mitani N, Nagamura Y, Rathinosabapathi B, Kirst M (2009) Differential gene expression of rice in response to silicon and rice blast fungus *Magnaporthe oryzae*. Annals of Applied Biology 155(2): 161 - 170
- Cai K, Gao D, Luo S, Zeng R, Yang J, Zhu X (2008) Physiological and cytological mechanisms of siliconinduced resistance in rice against blast disease. Physiologia Plantarum 134(2): 324 - 333

- Calvaruso C, Turpault MP, Frey-Klett P (2006) Root associated bacteria contribute to mineral weathering and to mineral nutrition in trees: a budgeting analysis. Applied and Environmental Microbiology 72(2): 1258 - 1266
- Carpita NC (1996) Structure and biogenesis of the cell walls of grasses. Annual Review of Plant Biology 47: 445 - 476
- Ehrlich H (1996) How microbes influence mineral growth and dissolution. Chemical Geology *132* (1-4): 5 9
- Tanaka S (1995) Does silicon exist in association with organic compounds in rice plant? Soil Science and Plant Nutrition 41: 111 - 117
- Jinab H, Solond M, Varietel M (2008) Functional food product development (pp 354). Book. Pub. Smith & Charter
- Kim SG, Kim KW, Park EW, Choi D (2002) Silicon-induced cell wall fortification of rice leaves: a possible cellular mechanism of enhanced host resistance to blast. Phytopathology 92(10): 1095 - 1103
- Lee KE, Adhikari A, Kang SM, You YH, Joo GJ, Kim JH, Kim SJ, Lee IJ (2019) Isolation and characterization of the high silicate and phosphate solubilizing novel strain *Enterobacter ludwigii* GAK2 that promotes growth in rice plants. Agronomy 9 (3): 144
- Liang Y, Sun W, Zhu YG, Christie P (2007) Mechanisms of silicon-mediated alleviation of abiotic stresses in higher plants: a review. Environmental Pollution 147(2): 422 -428
- Ma JF, Takahashi E (2002) Soil fertilizer and plant silicon research in Japan. Elsevier Science, Amsterdam. Retrieved from https://api.semanticscholar.org/CorpusID:85579928
- Naureen Z, Aqeel M, Hassan MN, Gilani SA, Bouqellah N, Mabood F, Hussain J, Hafeez FY (2015) Isolation and screening of silicate bacteria from various habitats for biological control of phytopathogenic fungi. American Journal of Plant Sciences 6(18): 2850
- Ranganathan SV, Suvarchala V, Rajesh YB, Prasad M, Padmakumari AP, Voleti SR (2006) Effects of silicon sources on its deposition, chlorophyll content, and disease and pest resistance in rice. Biologia Plantarum 50: 713 -716
- Uroz S, Calvaruso C, Turpault MP, Pierrat JC, Mustin C, Frey-Klett P (2007) Effect of the mycorrhizosphere on the genotypic and metabolic diversity of the bacterial communities involved in mineral weathering in a forest soil. Applied and Environmental Microbiology 73(9): 3019 - 3027
- Yoshida S, Ohnishi Y, Kitagishi K (1962) Histochemistry of silicon in rice plant. III. The presence of cuticle-silica double layer in the epidermal tissue. Soil Science and Plant Nutrition $\delta(2)$: 1 5
- Zhang C, Wang L, Zhang W, Zhang F (2013) Do lignification and silicification of the cell wall precede silicon deposition in the silica cell of the rice (*Oryza sativa* L.) leaf epidermis? Plant and soil 372: 137 - 149

ASSESSMENT OF PESTICIDE USAGE AND ORGANOPHOSPHATE PESTICIDE RESIDUE SCREENING OF RICE, CORN, AND MUNGBEAN IN SELECTED AREAS IN NUEVA VIZCAYA AND ISABELA, CAGAYAN VALLEY REGION, PHILIPPINES

LAWRENCE C. GASPAR*, SARAH B. AQUINO, MARIELLA B. SINGSON, YVONNE KREESHNA A. BALDERAMA, MINDA FLOR M. AQUINO, GERLY T. ZULUETA, CHONALYN A. PASCUA, KAY S. OLIVAS, ROSE MARY G. AQUINO

Department of Agriculture Regional Field Office No. 2 – Integrated Laboratory Division- Regional Crop Protection Center- Plant Health Clinic, Carig Sur, Tuguegarao City, Cagayan, Philippines *Corresponding Author: ildphc.darfo2@yahoo.com

Abstract

Food safety remains a critical public concern globally. In Region 2, the Department of Agriculture-Regional Field Office 2 (DA-RFO2) has been actively monitoring pesticide residues in fruits and vegetables. Elevated levels of pesticide residues have been found in trading centers and public markets, prompting concerns about consumer health. These findings highlight the necessity to also evaluate pesticide residues in other priority commodities such as rice, corn, and mungbean - crops with strong export potential in the region. This initiative aims to strengthen producers' adherence to Good Agricultural Practices (GAP) and assess export compliance.

A coordination meeting was held with the Municipal Local Government Unit (MLGU) to identify respondents for baseline data collection. Fifty-eight farmers were interviewed, reporting frequent pesticide use to control pests and diseases in their crops. They reported the use of insecticides, fungicides, herbicides, and molluscicides. Insecticide use was widespread across commodities: rice (89%), corn (89%), and mungbean (75%). For rice, 47% of the farmers used pyrethroids, 19% used carbamates, and 16% used organophosphates. In corn production, 39% used pyrethroids, 24% organophosphates, and 20% neonicotinoids. For mungbean, 80% used diamides, 10% carbamates, and 10% neonicotinoids. These pesticides are categorized from 'moderately toxic and moderately irritant' to 'least toxic and not an irritant,' potentially posing health risks to farmers and consumers.

To determine whether farmers complied with Pre-Harvest Interval (PHI) and recommended dosages, a pesticide residue screening was conducted using Gas Chromatography - Mass Spectrometry (GC-MS). Samples from rice, corn, and mungbean were tested specifically for organophosphates. Results showed residue levels were below detection limits, indicating compliance with GAP. These findings suggest that Region 2's agricultural products are competitive and export-ready.

Keywords: organophosphate residues, gas chromatography-mass spectrometry, good agricultural practices, pesticide compliance, food safety, export standards

Introduction

Pesticides are commonly applied during crop production and postharvest storage to manage pests, as well as to preserve product quality (Naylor, 2003). Organophosphates and carbamates are frequently used in storage facilities to deter insect infestation in grains. Previous research has reported the presence of pesticide residues in stored grains. For instance, Jagadish et al. (2015) detected chlorpyrifos as the most prevalent pesticide in rice, wheat, and pulses. Similarly, a Nigerian study found organophosphate and carbamate residues in maize from local markets, with some levels exceeding the Maximum Residue Limit (MRL) (Ogah and Coker, 2012).

Food safety is a priority of DA-RFO2, particularly in promoting pesticide-safe vegetables through residue analysis. Gas Chromatography - Mass Spectrometry (GC-MS) is a powerful tool for residue analysis due to its sensitivity, specificity, and reliability. Beyond regulatory compliance and food safety, this technology facilitates market readiness, particularly for plant-based export commodities such as mungbean. Customs protocols in many countries require initial screening followed by laboratory tests to verify pesticide residue levels and other key parameters.

Under this conditions, the region needs a reliable pesticide residue testing method aligned with international MRL and GAP standards to support farmers in accessing export markets. Establishing a local testing facility would aid in organic certification and MRL compliance, benefitting both producers and consumers. This study focused on the determination of pesticide residues in low-water, high-starch crops namely rice, corn, and mungbean - as representatives of grains and pulses. It also serves as a baseline assessment of pesticide usage among producers of these crops. The objectives were to gather baseline data on pesticide application in rice, corn, and mungbean production; to screen samples for organophosphate residues using GC-MS; and to evaluate the suitability of these commodities for export in terms of food safety compliance.

Materials and Methods

Survey on Pesticide Usage

A survey was conducted in 2022 in selected agricultural areas of region 2. The focus was on major production zones and active Farmer Cooperative Associations (FCAs) identified by the respective MLGUs. Key areas included Nueva Vizcaya for rice known for its triple-cropping system and high pesticide usage due to pest vulnerability - and Isabela for corn and mungbean, the province's dominant crops.

Survey Participants and Methodology

Fifty-eight farmers participated in the pesticide usage survey: 28 rice farmers from Nueva Vizcaya, 18 corn farmers from Ilagan, Isabela, and 12 mungbean farmers from Cabatuan, Isabela. These farmers were members of associations previously identified by the Municipal Local Government Units (MLGUs). To complement the producer data, a preliminary survey of 15 agricultural traders was conducted in 2023. This group included five mungbean traders from Cabatuan and ten rice and corn traders from Ilagan. All traderrespondents were selected based on their active involvement in agricultural commerce within Region 2.

Data were gathered through focus group discussions (FGDs) and the administration of a structured questionnaire. Farmers were asked about their personal profiles, including age, sex, and years of farming experience. Farm-specific information was also collected, such as the total land area under cultivation, the type of cropping system employed, and the common pest problems they encountered in the field.

To assess pesticide practices, respondents were asked to identify the types of pesticides they used including insecticides, fungicides, herbicides, and molluscicides as well as the specific active ingredients. They provided information on dosage rates, frequency of application, and adherence to the pre-harvest interval (PHI). Farmers also indicated the date of their most recent pesticide application and the sources of irrigation water used on their farms. Additionally, data were collected on whether they used personal protective equipment (PPE) when handling or applying pesticides.

For traders, the survey focused on pesticide use during postharvest storage. Questions covered the types of pesticides applied to stored grains and the frequency of these applications.

Due to constraints in time and resources, the survey was limited to select municipalities and does not fully represent pesticide usage across all five provinces of Region 2. Nevertheless, the results provide a valuable initial dataset that can guide more extensive, region-wide investigations in the future.

A detailed breakdown of survey participants is presented in Table 1.

Collection of Samples

Grain samples were collected in 2024 from market stalls and storage facilities in Ilagan (for rice and corn) and Cabatuan (for mungbean), Isabela. Ten 1-kg samples were randomly selected for each commodity, properly labeled, and stored at 4°C until further analysis.

Chemicals and Reagents

All chemicals used in the analysis were of analytical grade with 99% purity. Acetonitrile served as the extraction solvent. Triphenyl phosphate (TPP) was used as the internal standard, and formic acid was used to stabilize extracts.

Sample Preparation, Extraction, and Clean-up

The pesticide residue analysis was conducted at the DA-RFO2 Integrated Laboratory Division -Regional Crop Protection Center, specifically at the Plant Health Clinic Chemical Laboratory. The analytical procedure followed the British Standard Institution (BSI) EN 15662 (2008) QuEChERS protocol, which is widely recognized for its efficiency in multi-residue pesticide testing.

To begin the process, 5 g of homogenized crop sample were accurately weighed and transferred into a conical tube. This was hydrated with 10g of ultrapure water to ensure proper sample preparation. For recovery monitoring, 100 μ L of a 20 ppm solution of triphenyl phosphate (TPP) were added as an internal standard.

Extraction was then performed by adding 10 ml of acetonitrile to the sample. The mixture was vortexed for one minute to ensure thorough mixing before being subjected to phase separation using a QuEChERS extraction kit. This step was followed by

Location	Name of FCA	Crem	Sex		Age Bracket		
Location		Crop	Male	Female	18-35	36-55	>56
Brgy. Casat, Bayombong,	Magus Farmers Irrigators Association	Rice	17	6	2	10	11
Nueva Vizcaya	Casat-Ipil Farmers Association Inc.	Rice	3	2	0	1	4
Brgy. Marana, Ilagan City, Isabela	San Andres Marana Corn Cluster Association	Corn	9	9	0	4	14
Brgy. Sampaloc, Cabatuan, Isabela	Flow of Pariir Agricultural Cooperative	Mungbean	9	3	1	5	6

Table 1. Summary of respondents.

centrifugation at 3,500 rpm for 5 min to allow clear separation of the aqueous and organic layers.

The supernatant obtained from the centrifugation step underwent further purification using dispersive solid-phase extraction (dSPE) tubes containing 150 mg of magnesium sulfate (MgSO₄), 25 mg of octadecylsilane (C18), and 25 mg of primary secondary amine (PSA). The purified extract was then vortexed and centrifuged once more. Following this, it was acidified with 5% formic acid to stabilize the analytes and then transferred into gas chromatography–mass spectrometry (GC-MS) vials.

All prepared samples were directly analyzed using GC-MS to screen pesticide residues.

Measurement of Organophosphate Pesticide Residue Using GC-MS

The Shimadzu QP2020 GC-MS system was used, featuring a split/splitless AOC 20i auto-injector, AOC 20s autosampler, and MS-QP2020 mass selective detector. The gas chromatograph was fitted with a SH-Rxi-5Sil MS fused silica capillary column (30 m × 0.25 mm i.d., 0.25 μ m film thickness; RESTEK). Helium (99.999% purity) served as the carrier gas at a flow rate of 5.0 mL/min. The oven was initially set to 100°C for 1 min, increased at 25°C/min to 150°C and finally at 10°C/min to 280°C/min and held for 9 min.

A 1 μ L volume was injected in splitless mode at 250°C. The mass spectrometer operated in Electron Impact (EI) mode with a detector voltage of 70V, ion source temperature of 230°C, and GC interface temperature of 250°C. Acquisition was done using Selected Ion Monitoring (SIM) mode.

Screening Detection Limit (SDL) and Compound Identification.

SDLs were established based on a signal-to-noise (S/N) ratio of \geq 3. Compound identification was based on retention time (RT) comparison and spectral matching using the NIST and Shimadzu EI libraries. Specific pesticides screened included: Malathion (10.615 min), Fenthion (10.855 min), Chlorpyrifos (10.780 min), Triazophos (13.735 min), Profenofos (12.615 min), and Diazinon (8.945 min).

Results and Discussion

Rice Production

Survey results showed that farmers commonly practice a triple-cropping system for rice cultivation. This intensive farming approach has been associated with a higher incidence of pest outbreaks, particularly of stem borers, rice black bugs, rice leaffolders, and leafhoppers. Farmers reported these issues during interviews, and the findings were validated through field monitoring by Regional Crop Protection Center (RCPC) personnel. In response to these pest pressures, improper pesticide application practices such as cocktailing or the combined use of multiple pesticides in a single application - were noted.

Nueva Vizcaya, identified as a hotspot for insect pests, exhibited high availability and use of synthetic pesticides. This accessibility, combined with ongoing capacity-building activities spearheaded by various agencies and bureaus, has contributed to a relatively high level of farmer awareness regarding pest identification and corresponding management practices.

As illustrated in Figure 1, farmers use several types of pesticides, with insecticides being the most dominant at 89%. Fungicides are used by 7% of respondents, of which 3.57% apply Ionic Copper Concentrate (ICC), which also functions as a bactericide. Herbicides and molluscicides are each used by 7% of farmers, suggesting that weed and mollusk pressures are comparatively less severe in the area. Notably, 7% of surveyed farmers reported not using any form of pesticide on their crops.

Table 2 presents the variety of pesticides applied during both wet and dry seasons for rice cultivation. Commonly used products include Magnum (Cypermethrin), Dupont Lannate (Methomyl), Chix (Beta-Cypermethrin), and Ranger (a combination of BPMC and Chlorpyrifos). The most frequently applied pesticide classes are pyrethroids, organophosphates, and carbamates, with some farmers also using diamides and neonicotinoids. These substances range from moderately toxic to least toxic classifications. Among organophosphates,

Chlorpyrifos, Malathion, and Profenofos were prioritized as target analytes for screening in milled rice samples using GC-MS.

Corn Production

Corn is planted from May to September during the dry season and from December to April in the wet season. Farmers reported infestations of corn borer, corn plant hopper, and fall armyworm, which necessitate the use of insecticides. Observations also indicated a reduced susceptibility of these pests to pesticides, suggesting the possible emergence of insecticide resistance. As shown in Figure 2, insecticides are the most widely used pesticide for corn (89%), followed by herbicides (22%) and fungicides (6%). Another 6% of corn farmers reported not using any pesticides.

Table 3 details the pesticides applied in corn production, including their active ingredients, pesticide classes, toxicity levels, and usage frequency. The most commonly used insecticides include pyrethroids (Cypermethrin, Beta-Cypermethrin, Lambda-cyhalothrin, Beta-Cyfluthrin), neonicotinoids (Thiamethoxam, Imidacloprid), and organophosphates (Chlorpyrifos). For fungicides, farmers used the triazole class, while herbicides fell under the organophosphate, triketone, phenoxy, and



Figure 1. Percentage of pesticide usage during the production of rice 2022, wet season (WS).



Figure 2. Percentage of pesticide usage in corn production, 2022 WS.

Pesticide Used	Active Ingredient	Chemical Class	FPA Color Band*	FPA Toxicity Category*	the p	ers Using esticide <u>N=28)</u>
Insecticide					Ν	%
Magnum 5 EC	Cypermethrin	Pyrethroid	Green	4	5	17.86%
Cymbush 5 EC	Cypermethrin	Pyrethroid	Green	4	2	7.14%
Surekill	Pyrethrin	Pyrethroid	Green	4	2	7.14%
Lancer 75 SP	Acephate	Organophosphate	Green	4	2	7.14%
Dupont Prevathon 5 SC	Chlorantraniliprole	Diamide	Green	4	1	3.57%
Gold 20 SC	Clothianidin	Neonicotinoids	Green	4	1	3.57%
Malathion 57 EC	Malathion	Organophosphate	Green	4	1	3.57%
Chix	Beta-Cypermethrin	Pyrethroid	Blue	3	8	28.57%
Dupont Lannate 40 SP	Methomyl	Carbamate	Yellow	2	6	21.43%
Ranger 31.5 EC	BPMC + Chlorpyrifos	Carbamate + Organophosphate	Yellow	2	3	10.71%
Karate 2.5 EC	Lambda-Cyhalothrin	Pyrethroid	Yellow	2	2	7.14%
Solomon 300 OD	Beta-Cyfluthrin and Imidacloprid	Pyrethroid + Neonicotinoid	Yellow	2	2	7.14%
Wild Kid 40 SP	Methomyl	Carbamate	Yellow	2	1	3.57%
Brodan 31.5 EC	Chlorpyrifos	Organophosphate	Yellow	2	1	3.57%
Bida 2.5 EC	Lambda-Cyhalothrin	Pyrethroid	Yellow	2	1	3.57%
Kriss	Lambda-Cyhalothrin	Pyrethroid	Yellow	2	1	3.57%
Pulsar 2.5 EC	Lambda-Cyhalothrin	Pyrethroid	Yellow	2	1	3.57%
Topkill 500 EC	Profenofos	Organophosphate	Yellow	2	1	3.57%
Molluscicide						
Kuholkil	Metaldehyde	Tetroxocane	Yellow	2	2	7.14%
Fungicide						
Index	Myclobutanil	Triazole	Yellow	2	1	3.57%
Ionic Copper Concentrate	Copper sulfate pentahydrate	Inorganic Copper	Green	4	1	3.57%
Herbicide						
Topshot	Penoxsulam+-cyhalofop- butyl	Triazolopyrimidine + Aryloxyphenoxy propionate	Green	4	1	3.57%
2,4-D	Amine	Phenoxy	Yellow	2	1	3.57%
No Pesticide Used	N/A			N/A	2	7.14%

Table 2. Pesticide usage in rice production, 2022 WS.

*Toxicity category 1 is highly toxic and severely irritating, 2 is moderately toxic and moderately irritating, 3 is slightly toxic and slightly irritating, 4 is least toxic and not an irritant.

*Reference: Fertilizer and Pesticide Authority (2024)

chloroacetamide categories. Commonly used brands include Alika, Lorsban, Spitfire, Chix, and Karate. Among the organophosphates, Chlorpyrifos was selected for residue analysis via GC-MS. Many of the insecticides used fall into the moderate toxicity category, which underscores the potential health and environmental risks when not properly applied.

Mungbean Production

Mungbean planting typically occurs from March to June. Farmers pay close attention to pod development to time their harvests precisely, ensuring pods are collected before they over-mature or shatter. Prior to harvesting, they also conduct soil preparation for the succeeding cropping season, which includes soil testing and nutrient application.

Figure 3 shows that 75% of mungbean farmers rely on synthetic insecticides, primarily due to issues with pod borers. The remaining 25% of farmers practice organic farming methods.

Table 4 presents the specific insecticides used in mungbean production. Prevathon (Chlorantraniliprole) is the most commonly used, applied by 66.67% of farmers. Other insecticides include Lannate (Methomyl) and Gold (Clothianidin), used by 8.33% each. These products fall under the diamide, carbamate, and neonicotinoid classes and range from

least toxic to moderately toxic based on toxicity classification.

Practices of Traders in Storage Facilities

During the second year of implementation, the study expanded to assess pesticide usage in postharvest stages. This component was carried out in collaboration with local government units (LGUs) of Cabatuan and Ilagan, focusing on pesticide application in the storage of milled rice, corn, and mungbean.

Traders from various warehouses in Ilagan and Cabatuan, Isabela were interviewed, and samples were collected from their facilities for laboratory testing. Table 5 presents the data collected regarding their postharvest practices.

Survey findings indicated that some traders apply pesticides such as Scorpio, Cymbush, and Lannate daily to control common storage pests like weevils. Lannate (Methomyl) is widely used for rice and corn due to its affordability and availability. While the storage duration for rice and corn ranges from one to two months, mungbean is not treated with pesticides as it is typically sold quickly.

Pesticide residue analysis using GC-MS

Thirty samples of milled rice, corn, and mungbean obtained from trading warehouses were subjected to

Table 3. Pesticide usage in corn production, 2022 WS.

Pesticide Used	Active Ingredient	Chemical Class	FPA Color Band*	FPA Toxicity Category*	ре	r Using the sticide <u>N=18</u>
Insecticide					Ν	%
Cymbush 5 EC	Cypermethrin	Pyrethroid	Green	4	1	5.56%
Chix	Beta-Cypermethrin	Pyrethroid	Blue	3	3	16.67%
Alika 247 ZC	Lambda-cyhalothrin +thiamethoxam	Pyrethroid +Neonicotinoid	Yellow	2	6	33.33%
Lorsban	Chlorpyrifos	Organophosphate	Yellow	2	5	27.78%
Karate 2.5 EC	Lambda-Cyhalothrin	Pyrethroid	Yellow	2	3	16.67%
Solomon 300 OD	Beta-Cyfluthrin +Imidacloprid	Pyrethroid+ Neonicotinoid	Yellow	2	2	11.11%
Ariba 2.5 EC	Lambda-Cyhalothrin	Pyrethroid	Yellow	2	1	5.56%
Fungicide						
Armure 300 EC	Difenoconazole +propiconazole	Triazole	Blue	3	1	5.56%
Herbicide						
Spitfire 480 SL	Glyphosate ipa	Organophosphate	Green	4	4	22.22%
Halex GT 525 ZC	Mesotrione, glyphosate, s-metochlor	Triketone, Organophosphate, Chloroacetamide	Green	4	1	5.56%
2,4-D	Amine	Phenoxy	Yellow	2	1	5.56%
No Pesticide Used	N/A			N/A	1	5.56%

*Toxicity category 1 is highly toxic and severely irritating, 2 is moderately toxic and moderately irritating, 3 is slightly toxic and slightly irritating, 4 is least toxic and not an irritant.

*Reference: Fertilizer and Pesticide Authority (2024)



Figure 3. Percentage of pesticide usage in mungbean, 2022 dry season (DS).

Table 4. Pesticide usage in mungbean production, 2022 DS.

Pesticide Used	Active Ingredient	Chemical Class	FPA Color Band*	FPA Toxicity Category*	pe	r Using the sticide <u>N=12</u>
Insecticide					Ν	%
Dupont Prevathon 5 SC	Chlorantraniliprole	Diamide	Green	4	8	66.67%
Dupont Lannate 40 SP	Methomyl	Carbamate	Yellow	2	1	8.33%
Gold 20 SC	Clothianidin	Neonicotinoid	Green	4	1	8.33%
No Pesticide Used	N/A			N/A	3	25.00%

*Toxicity category 1 is highly toxic and severely irritating, 2 is moderately toxic and moderately irritating, 3 is slightly toxic and slightly irritating, 4 is least toxic and not an irritant.

*Reference: Fertilizer and Pesticide Authority (2024)

Table 5. Pesticide usage during postharvest in 2023.

Pesticide Used	Active Ingredient	Chemical Class	FPA Color Band*	FPA Toxicity Category*
Cymbush	Cypermethrin	Pyrethroid	Green	4
Lannate	Methomyl	Carbamate	Yellow	2
Scorpio	Methomyl	Carbamate	Yellow	2

*Toxicity category 1 is highly toxic and severely irritating, 2 is moderately toxic and moderately irritating, 3 is slightly toxic and slightly irritating, 4 is least toxic and not an irritant.

*Reference: Fertilizer and Pesticide Authority (2024)



Figure 4. Chromatogram of 6 Organophosphate Standards: (a) Diazinon, Chlorpyrifos; (b) Malathion, Fenthion, Profenofos; and (c) Triazophos.

qualitative screening for pesticide residues using Gas Chromatography-Mass Spectrometry (GC-MS). The analysis did not detect any target compounds, indicating that samples were either free from organophosphate pesticide residues or contained amounts below the method's screening detection limits (Figure 4).

Conclusion and Recommendations

This study was conducted to establish baseline data on pesticide usage among rice, corn, and mungbean producers; to perform screening for organophosphate pesticide residues through Gas Chromatography-Mass Spectrometry (GC-MS); and to assess the export potential of these commodities with respect to food safety compliance.

Findings revealed the predominant use of insecticides in the production of rice, corn, and mungbean, largely due to widespread pest infestations. Among the commonly applied insecticide classes organophosphates, were carbamates, and pyrethroids. In rice and corn cultivation, triazole fungicides were frequently used, while herbicides such as amine and glyphosate were applied during the production period. Despite efforts to adopt Good Agricultural Practices (GAP), Integrated Pest Management (IPM), and organic farming methods, most farmers continued to rely on synthetic pesticides - primarily those classified as moderately toxic (yellow label pesticides).

Traders, particularly those handling rice and corn, reported daily applications of synthetic insecticides such as Scorpion, Cymbush, and Lannate to manage storage pests like weevils. These insecticides belong to the carbamate and pyrethroid classes and range from least toxic (green label) to moderately toxic (yellow label) categories.

Initial GC-MS screening of samples collected from trading warehouses and market stalls indicated no detectable levels of the target organophosphate pesticide residues. This result suggests that either the commodities were free from organophosphate contamination or residues were present below detection limits. Specifically, yellow corn sourced from Ilagan, Isabela was found safe for livestock feed production, thereby supporting food safety in beef, dairy, poultry, and other animal sectors. Similarly, milled rice and yellow corn from Ilagan City, along with mungbean from Cabatuan, Isabela, were deemed relatively safe for export or further processing, presenting opportunities for value chain enhancement and increased income for producers.

In light of these findings, there is a need to expand baseline data collection to other locations across the region. This would enable a more comprehensive analysis of farming practices and facilitate the development of appropriate interventions. Further, pesticide residue screening should be broadened to include other active ingredients and classes beyond organophosphates, to ensure greater protection of consumer health and to strengthen the export readiness of milled rice, corn, and mungbean from region 2. It is also recommended that the government introduce incentive programs to promote the adoption of GAP and to recognize and reward farmers and traders who demonstrate safe pesticide practices and compliance with local and international standards.

Literature Cited

- BSI [British Standard Institution] (2008) Foods of plant origin Determination of pesticide residues using GC-MS and/or LC-MS/MS following acetonitrile extraction/ partitioning and cleanup by dispersive SPE — QuEChERS-method. European Standard. Retrieved from http://www.chromnet. net/Taiwan/QuEChERS_Dispersive_SPE/QuEChERS_ %E6%AD%90%E7%9B%9F%E6%96%B9%E6%B3 %95_EN156622008_E.pdf. (accessed January 9, 2023)
- Fertilizer And Pesticide Authority (2024) List of registered agricultural pesticides as of May 2, 2024. Department of Agriculture. Retrieved from https://fpa.da.gov.ph/wpcontent/uploads/2024/05/Updated-List-of-Registeredpesticide-final-as-of-May-2-2024.pdf (accessed May 28, 2024)
- Jagadish GK, Jaylakshmi SK, Sreeramulu, K (2015) Evaluation of pesticide residue in rice, wheat and pulses of Bidar district Karnataka, India. Issues in Biological Sciences and Pharmaceutical Research 3(9): 100 - 106
- Naylor REL (2003) Crop health. Responding to weed, diseases and pests (pp 213 - 230). The Agricultural Notebook, Oxford, Blackwell Science
- Ogah CO, Coker HB (2012) Quantification of organophosphate and carbamate pesticide residues in maize. Journal of Applied Pharmaceutical Science 2(9): 093 - 097

FULL PAPER

EFFECTS OF GENOTYPE-ENVIRONMENT INTERACTION ON GRAIN YIELD IN HYBRID RICE LINES

OLIVER E. MANANGKIL^{*} AND MAYBEL DM. BANTING

Plant Breeding and Biotechnology Division, Philippine Rice Research Institute, Science City of Muñoz, Nueva Ecija *Corresponding Author: revilo0514@gmail.com

Abstract

The development and promotion of hybrid rice remain among the government's key strategies, implemented through the agriculture sector to achieve rice self-sufficiency and improve the well-being of both consumers and farmers. With the National Cooperative Test (NCT) now adopting a major rice-producing area (MRPA) classification instead of the former regional approach, it has become essential to analyze data by incorporating genotype-by-environment interaction (GEI) to assess the differential responses of promising rice lines to varying environmental risk factors across MRPAs.

This study aimed to identify high-yielding and stable hybrid rice lines by evaluating ten genotypes across 18 sites over three cropping seasons - 2017 wet season (2017 WS), 2018 dry season (2018 DS), and 2018 wet season (2018 WS). The additive main effects and multiplicative interaction (AMMI) and genotype \times genotype \times environment (GGE) models were employed. In addition, the study assessed agro-morphological traits, grain quality, and resistance to major insect pests and diseases. Results revealed that genotype performance in grain yield varied across locations due to the effects of genotype, environment, and GEI. Most genotypes exhibited early maturity, semi-dwarf plant height, high milling recovery, and intermediate to resistant reactions against major pests and diseases. However, some genotypes showed susceptibility to tungro virus, indicating the need for further resistance breeding.

Keywords: additive main effects and multiplicative interaction (AMMI) model, genotype x environment interaction (GEI), hybrid yield potential, genotype x genotype x environment (GGE) model

Introduction

Rice serves as the staple food in the Philippines and plays a vital role in the nation's economy, particularly for low-income populations. Enhancing rice productivity through modern technological interventions is therefore a critical strategy for poverty alleviation and increasing the economic return of the agricultural sector. Since the early 1960s, rice self-sufficiency has been the target of various government programs in agriculture; however, this goal remains elusive (Balisacan & Ravago, 2003).

One promising approach to increasing rice production is hybrid rice technology, which provides a yield advantage of 15–30% over modern inbred rice varieties (Abebrese et al., 2019). Hybrid rice has achieved significant breakthroughs in modern agricultural science (He et al., 2020), especially in Asia, where it has substantially improved productivity (Revathi et al., 2025). Expanding the availability of hybrid rice varieties is thus essential in ensuring food security, especially in developing countries (Nguyen & Scrimgeour, 2022). However, realizing the full yield potential of hybrid rice is complex, often affected by genotype × environment interaction.

In the Philippines, hybrid rice development and promotion are integral to the government's strategy for achieving rice self-sufficiency and improving farmers' income. Gramaje et al. (2020) reported superior hybrid performance in terms of grain yield and morpho-agronomic traits compared to check varieties. Nevertheless, hybrids have limitations, and their successful deployment depends heavily on adaptability (Atal, 2024). Therefore, further research is needed to improve yield advantage, grain quality, and overall productivity of hybrid rice.

Yield advantage is often assessed by comparing the performance of farmers growing hybrids versus inbred varieties. However, this advantage cannot be attributed solely to varietal differences. Farmers usually cultivate hybrids in environments favorable in terms of soil condition, climate, topography, and water availability. A study by Peng et al. (2003) involving trials across various tropical Asian locations estimated the yield advantage of hybrids while minimizing variation from non-genetic factors such as management practices and input levels.

The performance of rice varieties, whether inbred or hybrid, is the result of genotype (G), environment (E), and their interaction (GEI). As noted by Blanche et al. (2009), evaluating GEI is critical for assessing yield stability and identifying genotypes with consistent performance across diverse environments and production years. GEI suggests that genotypes respond differently to environmental variation, thus reinforcing the need for multilocation testing (Tariku et al., 2013). According to Hill (1975), a highperforming genotype maintains acceptable performance across seasons and environmental stress levels.

Genotype \times environment interactions arise when environmental factors influence genotype performance in differing ways (Lindheimer et al., 2009). Beyond rice, studies such as that by Mutari et al. (2022) on navy beans demonstrate how GEI analysis identifies genotypes with high adaptability and links grain yield with other agronomic traits.

Understanding GEI and crop stability aids decisions during varietal development. It helps identify adaptation patterns, guide screening for release, and delineate recommendation domains (Yan et al., 2007). Quantifying GEI involves evaluating genotypes across environments, using either univariate or multivariate statistical methods. Common univariate approaches include regression of genotype means on environmental indices (Yates & Cochran, 1938; Finlay & Wilkinson, 1963; Eberhart & Russell, 1966). Multivariate methods, such as the AMMI model, complement these analyses by separating main effects from interaction patterns (Crossa, 1990; Gauch, 1992).

The AMMI model, combining ANOVA and principal component analysis, dissects the GEI into interpretable interaction principal components (IPCs), thereby providing a multiplicative model (Romagosa & Fox, 1993; Zobel et al., 1988; Gauch & Zobel, 1996). Several parameters derived from AMMI, including the AMMI stability value (ASV) and its modified form (MASV), help quantify and rank genotype stability (Purchase et al., 2000; Karimizadeh et al., 2012). These metrics are comparable with traditional methods such as those proposed by Shukla (1972) and Eberhart & Russell (1966). ASV has been validated as an effective tool in identifying stable genotypes (Sabaghnia et al., 2008).

In the study of Bose et al. (2014a), AMMI analysis of 17 early-maturing rice genotypes across four seasons revealed that the first two IPCs explained 93.76% of total GEI effects. Similarly, Tarang et al. (2013) characterized two stable rice lines using AMMI across three years. Bose et al. (2014b) evaluated 12 genotypes over three years, identifying stable lines using significant AMMI components (AMMI-1 to AMMI-3). In another study, Tariku et al. (2013) analyzed 16 rainfed lowland rice genotypes across eight environments and found that the first four IPCs accounted for 91.13% of the total GEI variance. These examples show that multivariate models like AMMI provide powerful tools for GEI interpretation and guide variety recommendation decisions. This study aimed to identify high-yielding, stable rice genotypes and determine the environments where they are best adapted using the AMMI model. Promising genotypes exhibiting favorable grain quality, high yield, and stability may be recommended for varietal release.

Field performance trials were conducted by the NCT across diverse locations to evaluate elite hybrid lines for potential release. Stability in grain yield is a highly desirable trait in rice cultivars, facilitating broad adaptability. Genotype adaptability is evaluated on yield responses across different based environments within a given rice ecosystem. Additional data were also collected on grain quality and resistance to major insect pests and diseases. The performance and stability of the genotypes were validated using the AMMI and GGE models. Addressing these challenges requires a robust breeding pipeline capable of delivering varieties that specific regional and meet provincial recommendation requirements.

Materials and Methods

At the time this study was conducted, the National Cooperative Testing (NCT) followed its previous protocol, which required evaluation over three cropping seasons - one dry season (DS) and two wet seasons (WS). Accordingly, hybrid trials were conducted across multiple NCT locations under this framework. This multi-location, multi-season testing ensures rigorous evaluation of genotypic performance under varied environmental conditions. Only hybrid lines that consistently meet NCT's stringent criteria are recommended for varietal release by the National Seed Industry Council (NSIC) and approved for commercial cultivation. This system is intended to ensure yield stability of new hybrids under favorable environments (Shakhatreh et al., 2001).

Ten hybrid genotypes were evaluated under this protocol across the 2017 WS, 2018 DS, and 2018 WS seasons. These trials were conducted at 18 sites, generating 35 environment-specific data points (Table 1). Comprehensive data wrangling was undertaken to ensure accurate dataset organization, allowing for reliable, efficient, and unbiased GEI analysis.

The hybrid rice genotypes were assigned codes G1 through G10 to maintain confidentiality. This coding was necessary because the evaluated lines were precommercial and lacked official varietal names, accession numbers, or registered identities. Most of these lines are under development by private breeding companies, and confidentiality is required to protect intellectual property rights.

Location	2017 WS	2018 DS	2018 WS
PhilRice site in Agusan del Norte	E1	-	E1
PhilRice Bicol	E2	E1	E3
Bohol APC	E3	-	E2
PhilRice CES	E4	E2	E4
CMU Musuan, Bukidnon	E5	-	-
Hagonoy, Davao del Sur	-	E3	E3
INREC, Dingras, Ilocos Norte	E6	E4	E7
Dingras, Ilocos Norte	-	-	E6
PhilRice Isabela	E7	-	E8
Southern Leyte	-	E5	E9
PhilRice Midsayap	-	-	E10
PhilRice Negros	-	E6	-
PhilRice site in Agusan del Sur	-	E7	E11
PREC Sta. Barbara, Pangasinan	-	E8	-
SCRC Cagayan	E8	E9	E12
VSU Leyte	E9	E10	E13
DA-WESCIARC	E10	E11	E14

Table 1. Test sites of hybrid Group 1 during three seasons (2017 WS, 2018 DS, and 2018 WS).

*E- environment *L- location

*LxS- location x season code

*Hybrid group 1 tested in 10 sites during 2017 WS, 11 in 2018 DS, and 14 in 2018 WS. Statistical design – refer to hybrid protocol

Only data from test sites with a coefficient of variation (CV) less than 20% and a coefficient of determination (r^2) greater than 0.5 were included in the analysis. Per-season analysis was conducted to determine the adaptability of specific genotypes to particular environments, while across-season (AS) analysis was used to assess stability over all three cropping seasons. The AMMI and GGE models were employed to generate graphical representations and to interpret genotype responses across environments.

Results and Discussion

Yield prediction and estimation in this study were based on multi-location trials conducted with limited experimental data. These trials help determine yield stability across environments and provide reliable guidance for selecting the best genotypes or agronomic treatments for future planting seasons and expansion into new areas (Crossa, 1990). Building on Crossa's work, key agronomic strategies for future plantings emphasize the importance of multi-location testing, analysis of genotype × environment (G×E) interactions, and stability assessments. These approaches support the identification of robust and high-performing genotypes under diverse conditions.

Ssemakula and Dixon (2007) demonstrated the importance of $G \times E$ interaction (GEI) in genotype evaluation and breeding programs. Hybrid genotypes often exhibit distinct performance across different

locations, with variations attributed to the interaction between genotype and environment (Martin, 2000). The existence of GEI complicates the evaluation, selection, and recommendation of hybrids in rice ecosystems (Ebdon and Gauch, 2002).

The yield performance of genotypes varied significantly across environments, as reflected by a statistically significant interaction (p < 0.0001). Analysis of variance (ANOVA) showed that the main effects including genotype, environment, and GEI were all highly significant ($p \le 0.0001$) across all three seasons and across seasons (AS), except for GEI during the 2018 wet season (2018 WS) (Table 2). A significant GEI suggests that the yield performance of genotypes is inconsistent across environments (Manigbas et al., 2019). Despite the insignificant GEI in 2018 WS, data from that season were still included in the AMMI and GGE biplot analyses to capture and compare trends across all three seasons.

The AMMI model and GGE biplot captured GEI effectively, with combined principal component scores (PC1 and PC2) explaining more than 50% of the G+GE variation across yield trials. This level of variation was sufficient for the models to explain GEI and generate mega-environments (Manigbas et al., 2016). Partitioning the sum of squares further showed that environmental effects accounted for 88.2% of total variation, while genotype and GEI contributed 1.4% and 10.3%, respectively.

Interaction effects on hybrid rice grain yield

Table 2. Analysis of variance of the	10 genotypes across environments in 20	017 WS, 2018 DS, and 2018 WS.

Source of Variation	DF	Sum of Squares	Mean Square	F value	PR > F	Sum of Squares (%)
2017 WS						
G	9	19506826.5	2167425.2	6.8	<.0001	1.9
E	9	870416493.8	96712943.8	302.3	<.0001	85.4
REP	2	392508.9	196254.5	0.6	0.5425	0.0
G*E	81	128694638.9	1588822.7	5.0	<.0001	12.6
AMMI Analysis						
PC1	17	61662696	3627217	6412.68	0	47.9
PC2	15	22425992	1495066	2643.18	0	17.4
GGE Biplot Analysis						
PC1	17	61672449	3627791	6413.7	0	41.6
PC2	15	39024317	2601621	4599.5	0	26.3
Residuals/Error	198	63347995	319939			
2018 DS						
G	9	52888722	5876525	9.0	<.0001	3.7
E	10	1233692376	123369238	189.0	<.0001	85.3
REP	2	2617565	1308783	2.0	0.1372	0.2
G*E	90	156481964	1738688	2.7	<.0001	10.8
AMMI Analysis						
PC1	18	77194169	4288565	5308.6	0	48.3
PC2	16	26810699	1675669	2074.2	0	16.8
GGE Biplot Analysis					-	
PC1	18	112924617	6273590	7765.7	0	53.5
PC2	16	31266265	1954142	2418.9	0	14.8
Residuals/Error	208	135746749	652629			
2018 WS						
G	9	31612224	3512469	4.7	<.0001	2.0
E	13	1449290860	111483912	149.4	<.0001	89.6
REP	2	390880	195440	0.3	0.7698	0.0
G*E	117	137113543	1171911	1.6	0.0014	8.5
AMMI Analysis					0.0011	0.0
PC1	21	47981778	2284847	2644.7	0	35.0
PC2	19	30532486	1606973	1860.1	0	22.3
GGE Biplot Analysis	10		1000070	1000.1	5	22.0
PC1	21	51298810	2442800	2827.6	0	30.4
PC2	19	37772125	1988007	2301.1	0	22.4
Residuals/Error	278	207492011	746374	2001.1	U U	<i></i> .7
Across Season	2.0					
G	9	57894385	6432709	7.96	<.0001	1.4
E	31	3593566054	115921486	143.39	<.0001	89.5
REP	2	2076183	1038092	1.28	0.2775	0.1
G*E	279	418763471	1500944	1.86	<.0001	10.4
AMMI Analysis				1.00		10.7
PC1	42	153951330	3665508	4076.8	0	32.7
PC2	40	97429377	2435734	2709.0	0	20.7
GGE Biplot Analysis			2100704	_,	Ŭ	20.7
PC1	42	168704746	4016780	4467.5	0	32.0
PC2	40	97774143	2444354	2718.6	0	18.5
	-10	0,,,,,,,,,,		2,10.0	5	10.0

*G- genotype, E-environment

Environmental effects accounted for a substantial portion of total yield variation: 85.4% in 2017 WS, 85.3% in 2018 dry season (2018 DS), 89.6% in 2018 WS, and 89.5% across seasons (AS). These results indicate a high level of environmental diversity among test locations. A significant GEI further suggests differences in genotypic responses to varying environments (Asfaw et al., 2009; Manigbas et al., 2019). In contrast, genotype contributed only a small percentage to the variation—1.9% in 2017 WS, 3.7% in 2018 DS, 2.0% in 2018 WS, and 1.4% across seasons. A small genotypic contribution implies low efficiency of indirect selection for improving yield potential (Manigbas et al., 2016).

2017 WS Yield Performance and Stability of Hybrid Genotypes AS (AMMI analysis)

Table 3 presents the ranking of genotypes based on mean yield, AMMI Stability Value (ASV), and Yield Stability Index (YSI). AMMI analysis showed that G7 had the highest yield at 6.1 t ha⁻¹ while G2 was the most stable genotype based on ASV. Notably, G3 ranked first as the most stable and high-yielding genotype across 10 locations during 2017 WS based on YSI.

Table 3. Ranking of genotypes based on YSI, ASV, and meanyield in 2017 WS.

Construct	Y	SI	Α	sv	Меа	n Yield
Genotype	Rank	Value	Rank	Value	Rank	(t ha-1)
G3	1	4	2	23.81	2	6080.13
G7	2	5	4	33.67	1	6183.63
G2	3	7	1	8.56	6	5550.00
G4	4	8	5	57.35	3	5821.03
G1	5	10	3	33.32	7	5521.20
G5	6	12	7	69.94	5	5594.03
G10	7	13	9	87.38	4	5730.20
G8	8	15	6	67.78	9	5465.70
G6	9	18	8	73.99	10	5409.63
G9	9	18	10	90.78	8	5466.50

*YSI- yield stability index *ASV- AMMI stability value

Figure 1 shows the AMMI1 biplot illustrating the yield and PC1 scores of genotypes and environments. The vertical axis indicates interaction differences while the horizontal axis reflects the main effects of genotypes and environments (Asfaw et al., 2009; Manigbas et al., 2016; Manigbas et al., 2019). Genotypes and environments located on the right side of the ordinate tend to exhibit higher yields (Tariku et al., 2013).

In general, the high-yielding genotypes in 2017 WS were G3, G4, G7, and G10. These genotypes performed well in environments such as PhilRice Bicol, Albay (E2); Bohol-APC, Bohol (E3); PhilRice CES, Nueva Ecija (E4); WESVIARC, Iloilo (E10); and PhilRice Isabela (E7). Conversely, genotypes G1, G2, G5, G6, G8, and G9 had lower yields, as did environments including PhilRice site in Agusan del Sur, Agusan del Norte (E1); CMU, Bukidnon (E5); INREC, Ilocos Norte (E6); SCRC, Cagayan (E8); and VSU, Leyte (E9) (Figure 1).

Genotypes and environments with large positive or negative PC1 scores demonstrated strong interactions while those with scores near zero were considered stable across environments. Genotypes with high yield but located far from zero along the interaction axis were deemed suitable only under favorable conditions (Tariku et al., 2013; Manigbas et al., 2016; Manigbas et al., 2019). Atakora et al. (2023) also noted that PC scores near zero indicate stability, whereas larger deviations indicate stronger interactions.

G10 and G4 were high-yielding genotypes but were only suitable under favorable conditions. Environments such as PhilRice Bicol (E2) and Bohol APC (E3) were identified as stable and high-yielding while PhilRice Agusan (E1) and INREC (E6) were stable low-yielding environments. G3, as supported by the YSI ranking (Table 3), was high-yielding and stable across environments. Genotype G2 demonstrated wide adaptation with PC scores near zero (Figure 2). Specific genotype-environment adaptations were also observed: G10 to CMU (E5), G8 to SCRC (E8), G3 to PhilRice CES (E4), and G7 to PhilRice Isabela (E9).

Identification of Most Discriminating and Most Representative Environment (GGE Biplot Analysis)

Figure 3 presents the GGE biplot, showing relationships among environments based on yield performance. The length of the environment vectors indicates their discriminating ability (Yan and Tinker, 2006; Manigbas et al., 2016). PhilRice Isabela (E7) was the most discriminating environment, followed by CMU (E5), PhilRice CES (E4), and VSU, Leyte (E9). Less discriminating environments included Bohol-APC (E3), INREC (E6), PhilRice Agusan (E1), PhilRice Bicol (E2), WESVIARC (E10), and SCRC (E8).





Figure 1. The AMMI1 Biplot showing the main and interaction (PC1) effects of genotypes and environment on mean yield in 2017 WS.



Figure 2. AMMI Biplot model shows PC1 and PC2 contributed to 65.3% of the GEI total variation of 10 genotypes and 10 environments in 2017WS.

The "what-won-where" biplot of the GGE model is considered one of the most effective tools for analyzing GEI patterns (Gauch and Zobel, 1997; Manigbas et al., 2016). Demelash (2024) emphasized that this type of analysis effectively identifies the topperforming genotypes within each megaenvironment, allowing for efficient genotype evaluation across fewer but more relevant testing zones.

Based on the biplot (Figure 4), genotypes were spread across six sections, while environments were grouped into five. The first section, comprising PhilRice Agusan (E1), PhilRice Bicol (E2), PhilRice CES (E4), VSU (E9), and WESVIARC (E10), had G3 and G7 as the top performers, consistent with the yield response plot (Figure 5). G4 was specifically adapted to PhilRice Isabela (E7), forming the second section. The third section included Bohol APC (E3) and INREC (E6), with G6 and G5 as the highest yielding genotypes. G9 and G10 were specifically adapted to SCRC (E8) and CMU (E5), respectively. The observed biplot pattern supports the recommendation of genotypes tailored for each environmental section.



¹E1- PhilRice Agusan, Agusan del Norte; E2- PhilRice Bicol, Albay; E3- Bohol APC, Bohol; E4- PhilRice CES, Nueva Ecija; E5- CMU, Bukidnon; E6- INREC, Ilocos Norte; E7- PhilRice Isabela; E8- SCRC, Cagayan; E9- Visayas State University (VSU), Leyte; E10- WESVIARC, Iloilo ²Genotypes 1-10 (G1-G10): Environment 1-10 (E1-E10).





Figure 4. The GGE what-won-where biplot showing GEI of 10 genotypes over 10 environments. Genotypes 1-10 (G1-G10); Environment 1-10 (E1-E10) in 2017 WS.



Figure 5. Yield response plot of 10 genotypes over 10 environments in 2017 WS. Genotypes 1-10 (G1-G10); Environment 1-10 (E1-E10).

2018 DS Yield Performance and Stability of Hybrids Genotypes Across Environments (AMMI analysis)

The Additive Main Effects and Multiplicative Interaction (AMMI) analysis showed that genotype G10 attained the highest mean yield at 7.56 t ha⁻¹, whereas G3 was identified as the most stable genotype based on the AMMI Stability Value (ASV). Moreover, when both yield and stability were considered using the Yield Stability Index (YSI), G3 ranked first among all tested genotypes across 11 environments during the 2018 dry season (Table 4).

 Table 4. Ranking of genotypes based on mean yield, ASV, and YSI in 2018 DS.

Constune	Y	SI	ASV		ASV Mean Yie		
Genotype	Rank	Value	Rank	Value	Rank	(t ha-1)	
2018DS							
G3	1	5	1	15.32	4	6687.12	
G8	2	7	4	37.35	3	6730.36	
G2	3	9	7	85.85	2	6877.79	
G10	3	9	8	93.38	1	7560.52	
G9	4	10	3	29.32	7	6358.79	
G6	5	11	5	47.44	6	6531.33	
G7	5	11	6	69.75	5	6593.05	
G1	6	12	2	25.26	10	6028.15	
G5	7	18	9	95.09	9	6316.17	
G4	7	18	10	98.31	8	6357.50	

*YSI- yield stability index *ASV- AMMI stability value

High-yielding environments included PhilRice Bicol (E1), PhilRice CES, Nueva Ecija (E2), Hagonoy, Davao del Sur (E3), Southern Leyte (E5), and SCRC, Cagayan (E9). In contrast, low-yielding environments were observed in INREC, Ilocos Norte (E4), PhilRice Negros (E6), Agusan del Sur (E7), PREC, Pangasinan (E8), VSU, Leyte (E10), and WESVIARC, Iloilo (E11). Among these, PhilRice Bicol (E1) was a consistently high-yielding and stable environment, while WESVIARC, Iloilo (E11) represented a stable but low-yielding site (Figure 6).

Genotype G3 was consistently high-yielding and stable, as evidenced by its top YSI ranking (Table 4). While genotypes G2, G8, and G10 also demonstrated high yields, their performance was notable only under favorable conditions. In contrast, genotypes G1, G4, G5, G6, and G9 showed relatively low yields across the test locations.



Figure 6. The interaction (PC1) effects of genotypes and environment on mean yield based on the AMMI Biplot in 2018DS. Genotypes 1-10 (G1-G10); Environment 1-11(E1-E11).

Identification of Most Discriminating and Most Representative Environment (GGE Biplot Analysis)

Based on the GGE biplot, Hagonoy, Davao del Sur (E3) was identified as the most discriminating environment, followed by VSU, Leyte (E10), PhilRice Negros (E6), WESVIARC, Iloilo (E11), and PhilRice CES, Nueva Ecija (E2). In contrast, Agusan del Sur (E7), PhilRice Bicol (E1), SCRC, Cagayan (E9), Southern Leyte (E5), INREC, Ilocos Norte (E4), and PREC, Pangasinan (E8) were identified as less discriminating (Figure 7).



Figure 7. Demonstration of GGE Biplot on the relationship among test environments based on yield in 2018 DS. Genotypes 1-10 (G1-G10); Environment 1-11 (E1-E11).

The "which-won-where" GGE biplot (Figure 8) divided the genotypes and environments into distinct sectors. PhilRice Negros (E6) and PREC, Pangasinan (E8) formed one section where G7 emerged as the highest-yielding genotype. Another sector included PhilRice Bicol (E1), Hagonoy (E3), INREC (E4), Southern Leyte (E5), SCRC (E9), VSU (E10), and WESVIARC (E11), with G10 as the top-performing genotype. The remaining section grouped PhilRice CES (E2) and Agusan del Sur (E7), where G4 and G5 performed best. These patterns of genotype adaptability were consistent with the observed yield responses (Figure 9).



Figure 8. The GGE what-won-where biplot showing GEI of 10 genotypes over 11 environments in 2018 DS. Genotypes 1-10 (G1-G10); Environment 1-11 (E1-E11).



Figure 9. The response plot of 10 genotypes over 11 environments based on yield in 2018 DS. Genotypes 1-10 (G1-G10); Environment 1-11 (E1-E11).

2018 WS Yield Performance and Stability of Hybrid Genotypes Across Environments (AMMI analysis)

Genotype G2 achieved the highest mean yield at 6.55 t ha⁻¹ during the 2018 wet season based on yield ranking. However, G6 emerged as the most stable and high-yielding genotype when both ASV and YSI results were considered across 14 test environments (Table 5).

Environments with consistently high yields included PhilRice Agusan (E1), Bohol APC (E2), Hagonoy, Davao del Sur (E5), PhilRice Isabela (E8), Agusan del Sur (E11), and WESVIARC, Iloilo (E14). Conversely, low-yielding environments included PhilRice Bicol (E3), PhilRice CES (E4), Dingras, Ilocos Norte (E6), INREC, Ilocos Norte (E7), Southern Leyte (E9), PhilRice Midsayap (E10), SCRC, Cagayan (E12), and VSU, Leyte (E13). Among these, PhilRice Agusan (E1) was recognized as a stable high-yielding site, while PhilRice Midsayap (E10) and VSU, Leyte (E13) were stable low-yielding environments (Figure 10).



Figure 10. AMMI1 Biplot showing the main and interaction (PC1) effects of genotypes and environment on mean yield in 2018 WS. Genotypes 1-10 (G1-G10); Environment 1-14(E1-E14).

Genotypes G2, G4, G6, and G10 were classified as high-yielding, while G1, G7, G8, and G9 were lowyielding. Based on the YSI ranking, G6 was confirmed as the most stable and high-yielding genotype (Table 5).

Identification of Most Discriminating and Most Representative Environment (GGE Biplot Analysis)"

In the GGE biplot analysis for 2018 WS, Bohol-APC (E2) emerged as the most discriminating environment, followed by INREC, Ilocos Norte (E7),

Table 5. Ranking of	genotypes	based	on	mean	yield,	ASV,
and YSI in 2018 WS.						

Constra	Y	SI	ASV		Mean Yield		
Genotype	Rank	Value	Rank	Value	Rank	(t ha⁻¹)	
2018 WS							
G6	1	4	1	7.74	3	6553.24	
G2	2	5	4	23.88	1	6753.43	
G4	3	7	3	13.08	4	6517.60	
G3	4	11	6	25.99	5	6490.98	
G10	4	11	9	43.53	2	6692.67	
G1	5	12	2	11.21	10	5856.57	
G5	6	13	7	35.75	6	6430.00	
G8	7	14	5	24.70	9	5968.76	
G9	8	16	8	38.41	8	6299.50	
G7	9	17	10	78.77	7	6326.00	

*YSI- yield stability index

*ASV- AMMI stability value

PhilRice Isabela (E8), Dingras, Ilocos Norte (E6), and Agusan del Sur (E11). Less discriminating environments included PhilRice Agusan (E1), PhilRice Bicol (E3), PhilRice CES (E4), Hagonoy (E5), Southern Leyte (E9), PhilRice Midsayap (E10), SCRC (E12), VSU (E13), and WESVIARC (E14) (Figure 11).

The "which-won-where" biplot (Figure 12) and the corresponding genotype response plot (Figure 13) divided genotypes into four groups and environments into three sectors. The first group showed specific adaptation of G1 to PhilRice Agusan (E1). The second group included environments such as PhilRice CES (E4), Dingras (E6), INREC (E7), PhilRice Isabela (E8), PhilRice Midsayap (E10), Agusan del Sur (E11), SCRC (E12), and WESVIARC (E14), where G4, G5,



Figure 11. GGE Biplot showing the relationship among test environments based on yield in 2018 WS. Genotypes 1-10 (G1-G10); Environment 1-14 (E1-E14).

and G8 demonstrated superior performance. In the third group, G7 emerged as the highest yielding genotype in PhilRice Bicol (E3), Hagonoy (E5), and Southern Leyte (E9).



Figure 12. GGE what-won-where biplot showing GEI of 10 genotypes over 14 environments in 2018WS. Genotypes 1-10 (G1-G10); Environment 1-14 (E1-E14).

Yield Performance and Stability of Hybrid Group 1 Genotypes Across Environments (AMMI analysis)

Across seasons, the genotype-by-environment interaction (GEI) analysis for Hybrid Group 1 involved data from 35 environments (Table 6), consisting of 10 locations in the 2017 WS; 11, 2018 DS; and 14, WS.

Table 6. Test sites of hybrid Group 1 across season 2017WS, 2018 DS, and 2018 WS.

Results from the NCT-Hybrid trials showed that genotype G3 was the most stable and high-yielding genotype. It ranked first in both the Yield Stability Index (YSI) and the AMMI Stability Value (ASV), and third in terms of mean yield (Table 7).

Table 7. Ranking of genotypes based on YSI, ASV, and mean yield, across seasons (2017 WS, 2018 DS & WS).

0	YSI		Α	sv	Mean Yield					
Genotype	Rank	Value	Rank	Value	Rank	(t ha⁻¹)				
Across Season										
G3	1	4	1	13.23	3	6435.24				
G2	2	5	3	21.96	2	6448.68				
G9	3	10	2	21.00	8	6080.13				
G10	4	11	10	78.87	1	6690.43				
G6	4	11	5	40.37	6	6219.61				
G4	5	13	8	62.56	5	6268.26				
G7	5	13	9	66.20	4	6369.25				
G1	6	14	4	22.11	10	5814.68				
G5	6	14	7	59.96 7		6155.38				
G8	7	15	6	47.50	9	6064.39				

*YSI- yield stability index

*ASV- AMMI stability value

Figure 13 presents the AMMI Model 1 biplot for the 10 hybrid genotypes. Among the genotypes, G2, G3, G7, and G10, along with 13 environments namely, E2 (PhilRice Agusan; 2018 WS), E5 (PhilRice Bicol; 2018 DS), E7 (Bohol-APC; 2018 WS), E9 (PhilRice CES; 2018 DS), E12 and E13 (Hagonoy, Davao del Sur; 2018 DS and 2018 WS), E18 and E19 (PhilRice Isabela; 2017 WS and 2018 WS), E20 (Southern Leyte; 2018 DS), E25 (Agusan del Sur; 2018 WS), E28 (SCRC, Cagayan; 2018 DS), and E33 and E35 (WESVIARC, Iloilo; 2017 WS and 2018 WS) - were situated on the right side of the grand mean, signifying high-yielding genotypes and environments. In contrast, low-yielding genotypes and environments appeared on the left side of the grand mean (Figure 14).

The AMMI1 analysis categorized the hybrid genotypes into three distinct groups based on yield performance and stability: (1) the most stable and high-yielding genotype (G3); (2) less stable but high-yielding genotypes (G2, G7, and G10); and (3) low-yielding genotypes (G1, G4, G5, G6, G8, and G9).

Identification of Most Discriminating and Most Representative Environment (GGE Biplot Analysis)

Among the test environments, E12 (Hagonoy, Davao del Sur; 2018 DS) emerged as the most discriminating site, followed by E31 (VSU, Leyte; 2018 DS), E11 (CMU, Bukidnon; 2017 WS), E23 (PhilRice Negros; 2018 DS), and E18 (PhilRice Isabela; 2017 WS), as shown in Figure 15.



Figure 13. Response plot of yield of 10 genotypes over 14 environments across seasons. Genotypes 1-10 (G1-G10); Environment 1-14 (E1-E14).



Figure 14. AMMI1 Biplot showing the main and interaction (PC1) effects of genotypes and environment on mean yield across seasons. Genotypes 1-10 (G1-G10); Environment 1-35 (E1-E35).

The genotype-by-environment interaction pattern is further illustrated in the "which-won-where" GGE biplot (Figure 16) and corresponding response plot (Figure 17). Genotypes and environments were distributed across four distinct sectors. In the first sector, G10 was the top-performing genotype at multiple locations: E12 (Hagonoy; 2018 DS), E31 and E32 (VSU, Leyte; 2018 DS and 2018 WS), E34 and E35 (WESVIARC, Iloilo; 2018 DS and 2018 WS), E2 (PhilRice Agusan; 2018 WS), E4 and E5 (PhilRice Bicol; 2017 WS and 2018 DS), E22 (PhilRice Midsayap; 2018 WS), E28 (SCRC, Cagayan; 2018 DS), and E25 (Agusan del Sur; 2018 WS).



*G-genotypes, E-environment

Figure 15. GGE Biplot showing the relationship among test environments based on yield. Genotypes 1-10 (G1-G10); Environment 1-35 (E1-E35).

The second sector comprised E7 (Bohol APC; 2018 WS), E8 (PhilRice CES; 2017 WS), E13 (Hagonoy, Davao del Sur; 2018 WS), E30 (VSU; 2017 WS), E23 (PhilRice Negros; 2018DS), E11 (CMU, Bukidnon; 2017 WS), E21 (Southern Leyte; 2018 WS), E26 (PREC, Pangasinan; 2018 DS), E6 (PhilRice Bicol; 2018 WS), E27 (SCRC, Cagayan; 2017 WS), E1 (PhilRice Agusan; 2017 WS), and E16 (INREC, Ilocos Norte; 2018 DS), where G7 was identified as the top-yielding genotype.

Genotype G1 showed specific adaptation to E18 (PhilRice Isabela; 2017 WS), which occupied the



Figure 16. The GGE what-won-where biplot showing GEI of 10 genotypes over 35 environments. Genotypes 1-10 (G1-G10); Environment 1-35 (E1-E35).



Figure 17. The yield response plot of 10 genotypes over 14 environments. Genotypes 1-10 (G1-G10); Environment 1-35 (E1-E35).

third sector of the biplot. The fourth sector included E19 (PhilRice Isabela; 2018 WS), E17 (INREC, Ilocos Norte; 2018 WS), E9 and E10 (PhilRice CES; 2018 DS and 2018 WS), E24 (Agusan del Sur; 2018DS), E33 (WESVIARC, Iloilo; 2017 WS), E3 (Bohol APC; 2017 WS), and E14 (Dingras, Ilocos Norte; 2018 WS), where G4 and G5 were the highest yielding genotypes.

The most high-yielding and stable environmental sites were E2 (Agusan del Norte; 2018 WS), E5 (Bicol; 2018 DS), E13 (Hagonoy, Davao del Sur; 2018 WS), E20 (Southern Leyte; 2018 DS), E28 (SCRC, Cagayan; 2018 DS), and E35 (WESVIARC, Iloilo; 2018 WS), as illustrated in Figures 18 and 20. The consistently high-yielding genotypes were G2, G7, and G10 while G3 demonstrated the highest yield stability and a strong mean yield of 6.4 t ha^{-1} with an ASV of 13.23.

Figure 19 shows the vector lengths, which indicate the discriminating ability of each environment. Among these, E12 (Hagonoy, Davao del Sur; 2018 DS) exhibited the strongest discriminating power, followed by E31 (VSU, Leyte; 2018 DS), E11 (CMU, Bukidnon; 2017 WS), E23 (Negros; 2018 DS), and E18 (Isabela; 2017 WS). Environments with vectors close to zero are considered the most representative sites.







Figure 19. The GGE Biplot indicating the relationship among test environments based on yield.



Figure 20. Response plot yield of 10 genotypes over 35 environments.

Furthermore, G10 performed best at the following locations: E31 and E32 (VSU, Leyte; 2018 DS and 2018 WS), E12 (Hagonoy, Davao del Sur; 2018 DS), E34 and E35 (WESVIARC, Iloilo; 2018 DS and 2018 WS), E2 (Agusan del Norte; 2018 WS), E28 (SCRC, Cagayan; 2018 DS), E22 (Midsayap; 2018 WS), and E4 and E5 (Bicol; 2017 WS and 2018 DS). Similarly, G7 was the top-yielding genotype at E7 (Bohol APC; 2018 WS), E8 (PhilRice CES; 2017 WS), E30 (VSU; 2017 WS), E13 (Hagonov; 2018 WS), E20 (Southern Leyte; 2018 DS and 2018 WS), E26 (PREC, Pangasinan; 2018 DS), E6 (Bicol; 2018 WS), E27 (SCRC, Cagayan; 2017 WS), E1 (Agusan del Norte; 2017 WS), E16 (INREC, Ilocos Norte; 2018 DS), E11 (CMU; 2017 WS), and E23 (Negros; 2018 DS). Genotype G1 was specifically adapted to E18 (Isabela; 2017 WS) while genotypes G4 and G5 yielded highest in E9 and E10 (PhilRice CES; 2018 DS and 2018 WS), E24 (Agusan del Sur; 2018 DS), E17 (INREC; 2018 WS), E19 (Isabela; 2018 WS), E33 (WESVIARC; 2017 WS), and E3 (Bohol; 2017 WS).

Conclusion

The test environments in this study exhibited significant diversity, and the presence of notable genotype-by-environment interaction (GEI) underscores the varying responses of genotypes to different environmental conditions, as shown in the ANOVA results. Genotype G3 emerged as the most stable and high-performing entry across multiple locations and seasons. Meanwhile, G1 demonstrated specific adaptation to the E19 site in Isabela, emphasizing the importance of localized selection in rice breeding programs.

Understanding GEI provides deeper insights into the range of environmental stresses faced across NCT sites and helps pinpoint genotype-specific responses to these challenges.

In the 2017 wet season, G7 posted the highest average yield at 6.10 t ha⁻¹, while G2 was the most stable genotype based on the AMMI Stability Value (ASV). However, G3 topped the Yield Stability Index (YSI), indicating its strong balance of high yield and stability across 10 sites.

During the 2018 dry season, G10 had the highest yield (7.56 t ha⁻¹), but G3 again proved the most stable genotype. G3 also ranked first in YSI, confirming its adaptability across the 11 test locations.

In the 2018 WS, G2 achieved the highest mean yield at 6.55 t ha⁻¹. However, G6 emerged as the most stable and high-yielding genotype when both ASV and YSI were considered, across 14 sites.

Thirty-five environments - 10 from 2017 WS, 11 from 2018 DS, and 14 from 2018 WS - were evaluated in this multi-season analysis. Across these varied conditions, G3 consistently ranked highest in both ASV and YSI. While it placed third in mean yield, its overall performance underscores its potential for varietal release due to its adaptability and consistent stability.

These findings highlight the importance of understanding genotype-specific responses to environmental conditions in breeding programs. They also point to the need for deeper, statistically rigorous analyses in multi-location trials. For more effective varietal recommendations under the major riceproducing area (MRPA) framework, selection decisions must be supported by broad, evidence-based datasets.

Acknowledgment

This work was funded by the Department of Agriculture - Bureau of Agricultural Research (DA-BAR), in collaboration with PhilRice and in partnership with various government and nongovernment agencies.

Literature Cited

- Abebrese SO, Yeboah A, Dogbe W, Dartey PKA, Akromah R, Gracen VE, Offei SK, Danquah EY (2019) Evaluation of yield, reaction to diseases, and grain physical attributes of some introduced rice hybrids in Ghana. International Journal of Agronomy. Retrieved from https://doi.org/ 10.1155/2019/3926765 (accessed November 11, 2024)
- Asfaw A, Alemayehu F, Gurum F, Atnaf M (2009) AMMI and SREG GGE biplot analysis for matching varieties onto soybean production environments in Ethiopia. Scientific research and essay *4*(11): 1322 - 1330
- Atakora K, Essilfie ME, Agyarko K, Dapaah HK, Santo KG (2023) Evaluation of yield and yield components of some cowpea (Vigna unguiculata (L.) Walp) genotypes in forest and transitional zones of Ghana. Agricultural Sciences 14(7): 878 - 897
- Atal JC (2024) Productivity of hybrid rice (Mestizo 27) under different water and nutrient management systems. Journal of Asian Scientific Research 14(4): 535
- Balisacan AM, Ravago ML (2003) The rice problem in the Philippines: Trends, constraints, and policy imperatives. Transactions of the National Academy of Science and Technology Philipine 25: 221 - 236
- Blanche SB, Utomo HS, Wenefrida I, Myers GO (2009) Genotype x environment interactions of hybrid and varietal rice cultivars for grain yield and milling quality. Crop Sci. 49(6): 2011 - 2018
- **Bose LK, Jambhulkar NN, Pande K, Singh ON** (2014b) Use of AMMI and other stability statistics in the simultaneous selection of rice genotypes for yield and stability under direct-seeded conditions. Chilean journal of agricultural research 74(1): 3 - 9

- Bose LK, Jambhulkar NN, Singh ON (2014a) Additive Main effects and Multiplicative Interaction (AMMI) analysis of grain yield stability in early duration rice. JAPS: Journal of Animal & Plant Sciences 24(6)
- Crossa J (1990) Statistical analysis of multi-location trails. Advances in Agronomy 44: 55 - 85
- Ebdon JS, Gauch Jr. HG (2002) Additive main effect and multiplicative interaction analysis of national turf grass performance trials: I. Interpretation of genotype × environment interaction. Crop Science 42(2): 489 - 496
- Eberhart ST, Russell W (1966) Stability parameters for comparing varieties 1. Crop science, 6(1): 36 - 40
- Finlay KW, Wilkinson GN (1963) The analysis of adaptation in a plant-breeding programme. Australian journal of agricultural research 14(6): 742 - 754
- Gauch HG (1992) Statistical analysis of regional yield trials: AMMI analysis of factorial designs. Amsterdam, Netherlands: Elsevier Science Publishers
- Gauch Jr HG, Zobel RW (1996) Optimal replication in selection experiments. Crop Science 36(4): 838 843
- Gauch Jr, HG, Zobel RW (1997) Identifying mega-environments and targeting genotypes. Crop science 37(2): 311 - 326
- Gramaje LV, Caguiat JD, Enriquez JOS, dela Cruz, QD, Millas RA, Carampatana JE, Tabanao D.A (2020) Heterosis and combining ability analysis in CMS hybrid rice. Euphytica 216: 1 - 22
- He Q, Deng H, Sun P, Zhang W, Shu F, Xing J, Peng Z (2020) Hybrid rice. Engineering 6(9): 967 - 973
- Hill J (1975) Genotype x environment interactions: A challenge for plant breeding. J Agric Sci. 85(3): 477 - 493
- Karimizadeh R, Mohammadi M, Sabaghnia N, Shefazadeh MK (2012) Using Huehn's nonparametric stability statistics to investigate genotype× environment interaction. Notulae Botanicae Horti Agrobotanici Cluj-Napoca 40(1): 293 -301
- Lindheimer MD, Roberts JM, Cunningham FG (2009) Chesley's hypertensive disorders in pregnancy. *In* Ward K, Lindheimer MD (Eds.) (pp 51–71). Genetic Factors in the Etiology of Preeclampsia/Eclampsia. Academic press, Elsevier. Retrieved from https://utsouthwestern. elsevierpure.com/en/publications/chesleys-hypertensivedisorders-in-pregnancy-3 (Accessed March 05, 2024)
- Manigbas NL, Lambio LAF, Magat MB, Bartolome VI (2016) AMMI and GGE biplot analyses of multi-environment test in rice (Oryza sativa L.) for irrigated lowland conditions in the Philippines. Philippine Journal of Crop Science 41(1): 60 - 69
- Manigbas NL, Madrid LB, Badajos AT (2019) Adaptability of Korean cultivars and its potential as genetic donor for rice breeding in the Philippines. Philippine journal of crop science 44(1): 25 - 35
- Martin N (2000) Gene-environment interaction and twin studies. In Spector T, Snieder H, MacGregor A, (Eds.). Advances in Twin and Sib-Pair Analysis (pp 43 - 150). London: Greenwich Medical Media

- Mutari B, Sibiya J, Gasura E, Kondwakwenda A, Matova PM, Chirwa R (2022) Genotype x environment interaction and stability analyses of grain yield and micronutrient (Fe and Zn) concentrations in navy bean (Phaseolus vulgaris L.) genotypes under varied production environments. Field Crops Research 286: 108607
- Nguyen Chau T, Scrimgeour F (2022) Productivity impacts of hybrid rice seeds in Vietnam." Journal of Agricultural Economics 73(2): 414 - 429
- Peng S, Yang J, Laza MRC, Sanico AL, Visperas RM, Son TT (2003) Physiological bases of heterosis and crop management strategies for hybrid rice in the tropics. *In* Virmani SS, Mao CX (Eds.). Hybrid rice for food security, poverty alleviation, and environmental protection (pp 153-173). Retrieved from https://www.researchgate.net/profile/ Mohammad-Zaman-Nouri/publication/269518415_ Hybrid_rice_technology_and_achievements_in_Iran/ links/54b8f9200cf28faced6264b1/Hybrid-ricetechnology-and-achievements-in-Iran.pdf#page=157 (accessed December 11, 2024)
- Purchase JL, Hatting H, Van Deventer CS (2000) Genotype x environment interaction of winter wheat (*Triticum* aestivum L.) in South Africa: II. Stability analysis of yield performance. South African Journal of Plant and Soil 17(3): 101 - 107
- Revathi P, Satheesh Naik SJ, Sundaram RM (2025) Hybrid Seed Production of Rice: Status and Outlook. Hybrid Seed Production for Boosting Crop Yields: Applications, Challenges and Opportunities 1: 41 - 78
- Romagosa I, Fox PN (1993) Genotype× environment interaction and adaptation. In Plant breeding: Principles and prospects (pp 373 - 390). Dordrecht: Springer Netherlands.
- Sabaghnia N, Sabaghpour SH, Dehghani H (2008) The use of an AMMI model and its parameters to analyse yield stability in multi-environment trials. The Journal of Agricultural Science 146(5): 571 - 581
- Shakhatreh Y, Kafawin O, Ceccarelli S, Saoub H (2001) Selection of barley lines for drought tolerance in lowrainfall areas. Journal of Agronomy and Crop Science 186(2): 119 - 127
- Shukla GK (1972) Some statistical aspects of partitioning genotype-environmental components of variability. Heredity 29(2): 237 - 245
- Ssemakula G, Dixon AGO (2007) Genotype x environment interaction, stability and agronomic performance of carotenoid-rich cassava. Scientific Research and Essay 2(9): 390-399
- Tarang A, Chaleshtary MH, Tolghilani A, Esfahani M (2013) Evaluation of grain yield stability of pure lines of rice in Guilan province. Iranian Journal of Crop Sciences 15(1)
- Tariku S, Lakew T, Bitew M, Asfaw M (2013) Genotype by environment interaction and grain yield stability analysis of rice (*Oryza sativa* L.) genotypes evaluated in north western Ethiopia. Net Journal of Agricultural Science *I*(1): 10 - 16
- Yan W, Kang MS, Ma B, Woods S, Cornelius PL (2007) GGE biplot vs. AMMI analysis of genotype-by-environment data. Crop science 47(2): 643 - 653

- Yan Weikai, Tinker NA (2006) Biplot analysis of multienvironment trial data: Principles and applications. Canadian journal of plant science 3(2006): 623 - 645
- Yates F, Cochran WG (1938) The analysis of groups of experiments. The Journal of Agricultural Science 28(4): 556 - 580
- Zobel RW, Wright MJ, Gauch Jr HG (1988) Statistical analysis of a yield trial. Agronomy journal *80*(3): 388 - 393

FULL PAPER

POTENTIAL INFLUENCE OF NITROGEN – SILICON FERTILIZATION ON IMPROVING NUTRIENT AVAILABILITY AND GRAIN YIELD OF RICE (*ORYZA SATIVA* L.)

KIM ANGELO B. GONZALES¹, MARIA LUISA T. MASON¹, JAYVEE C. KITMA^{2*}, AND JAN NICO A. LAZARO²,

¹Central Luzon State University, Science City of Muñoz, Nueva Ecija ²Philippine Rice Research Institute, Science City of Muñoz, Nueva Ecija *Corresponding Author: jayveeacruz@gmail.com

Abstract

Improper fertilizer management limits rice productivity. The combined application of nitrogen (N) and silicon (Si) enhances rice growth, stress tolerance, and yield. This study evaluated the influence of Si (with and without) under varying N levels (0%, 50%, and 100%) on NSIC Rc160 in a screenhouse experiment. Parameters examined included soil chemical properties, silicon uptake, plant NPK levels, and grain yield.

Initial soil analysis revealed high nitrogen (0.15%) and phosphorus (24.9 ppm) levels. After harvest, nitrogen remained high (0.15–0.16%), while phosphorus increased significantly, ranging from 29.7 to 34.6 ppm. Potassium improved from an initial 13.1 ppm to 55.8–66.6 ppm across treatments, suggesting effective N–Si interaction in enhancing potassium uptake. Scanning Electron Microscopy (SEM) of Treatment S2N2 (with Si; 50% N) revealed well-developed silica bodies and protective structures in Si-treated leaves. Although plant tissue N content remained low (0.42–0.50%), likely due to grain filling, phosphorus and potassium levels were maintained within optimal ranges. The highest number of filled grains was observed in Treatment S2N3 (with Si; 100% N), recording 941 grains and outperforming all non-Si treatments at every nitrogen level. Silicon showed minimal influence on 1,000-grain weight, indicating that its role may be synergistic with nitrogen and other factors in supporting grain yield. The results highlight a promising interaction between N and Si in improving nutrient uptake and productivity. However, further field validation is needed to confirm these effects under diverse growing conditions and to understand the mechanisms underlying enhanced soil nutrient availability and yield improvement in rice.

Keywords: nitrogen, silicon, rice, scanning electron microscopy, yield

Introduction

Rice is the staple food of more than 1.3 billion people in Asia, including millions of Filipinos who rely on it as a constant part of their diet (Mamiit et al., 2021; Cuong et al., 2017). The demand for rice continues to increase, yet production faces ongoing challenges due to limited access to technologies, low crop profitability, and poor soil and crop management practices. Additionally, the continued use of lowyielding cultivars hampers productivity growth in the rice sector (Datta et al., 2017).

Although rice output has seen dramatic increases in recent decades, the challenge remains: improving yield per unit area while reducing the environmental impact (Lobell et al., 2009). Addressing this issue requires both improved management and more efficient nutrient use.

Nitrogen (N) is a key nutrient in rice cultivation, essential for chlorophyll synthesis and photosynthesis, which influence the number and size of leaves, internode length, and overall vegetative growth (Zhu et al., 2017; Rütting et al., 2018; Wang et al., 2022). Nitrogen enhances both grain yield and food quality. Silicon (Si), although considered a beneficial rather than essential element, has proven value in rice cultivation. It increases resistance to pests and diseases (Sarma et al., 2019; Kumar and Bhandari, 2022), and improves tolerance to abiotic stresses such as drought and strong winds (Epstein, 2009; Guntzer et al., 2012; Cuong et al., 2017). Application of Si fertilizer results in stronger cell walls, thicker culm tissues, larger vascular bundles, and higher peroxidase activity - factors that enhance stress resilience and help prevent lodging (Ma and Takahashi, 2002; Chaiwong and Prom-U-Thai, 2022).

Despite its abundance in nature, Si is often found in insoluble forms. Common sources include slags from the iron and alloy industries, calcium silicates (e.g., wollastonite), and thermophosphate fertilizers (Gascho, 2001). For high-value crops and research, more soluble forms such as potassium silicate, sodium silicate, and silica gel are used. The synergistic application of nitrogen and silicon (N–Si) in rice has been shown to enhance plant growth, grain yield, and stress resistance through complex interactions. In this context, the present study evaluated the effects of N–Si fertilization on soil chemical properties, silicon uptake, NPK nutrient levels in plant tissues, and grain yield.

Materials and Methods

Place of the Study

The experiment was conducted in a screenhouse located at the Agronomy, Soils, and Plant Physiology Division (ASPPD) of the Philippine Rice Research Institute - Central Experimental Station (PhilRice CES), Science City of Muñoz, Nueva Ecija. Soil analysis was performed at PhilRice CES, while plant tissue analysis was conducted at the DA Regional Field Office III, City of San Fernando, Pampanga. Silicon deposition in plant tissue was evaluated through Scanning Electron Microscopy (SEM) at the Nanotechnology Research and Development Laboratory, Central Luzon State University.

Treatments and Experimental Design

The study was laid out using a split-plot arrangement in a randomized complete block design (RCBD) with four replications, conducted under screenhouse conditions. Each experimental unit consisted of a pot filled with ten kilograms of soil. The main plot factor was silicon application, which had two levels: with silicon and without silicon. The subplot factor was nitrogen, applied at three levels: 0%, 50%, and 100% of the recommended rate. These combinations resulted in six treatment groups, specifically designated as follows: S1N1 (without Si; 0% N), S1N2 (without Si; 50% N), S1N3 (without Si; 100% N), S2N1 (with Si; 0% N), S2N2 (with Si; 50% N), and S2N3 (with Si; 100% N).

Silicon was applied once and incorporated directly into the soil at the time of pot preparation. The nitrogen treatments followed standard recommendations based on site-specific nutrient management (SSNM) protocols. This factorial arrangement allowed the evaluation of individual and interactive effects of silicon and nitrogen on rice growth and productivity parameters.

Soil Sampling and Application of Inorganic Fertilizer

Soil was collected from Brgy. Maligaya and PhilRice CES at a depth of 0.5m. The samples were pulverized, sieved using a two-millimeter mesh, and quartered to obtain a one-kilogram sample for initial analysis. The fertilizer application followed SSNM guidelines and recommendations from Rice Crop Manager tests conducted on the sampled soil.

Seedling Preparation

Rice seeds were placed in a mesh bag and heated in an oven at 65°C for at least 5 h to break dormancy. They were left to cool overnight in the oven. The next day, seeds were transferred to sanitized petri dishes lined with moist tissue paper soaked in distilled water for pre-germination over two days. Five pregerminated seeds were sown at a depth of 3 cm in each pot containing well-puddled soil. Thinning was done 10 days after sowing, retaining only three healthy seedlings per pot.

Scanning Electron Microscopy

Leaf samples were collected during the reproductive stage. Three small sections were cut from the leaves for SEM analysis to examine silicon deposition on the adaxial leaf surface. The analysis was performed using a Hitachi SU3800 SEM at 1,000x magnification and an operating voltage of 5.0 kV. Focus and brightness were optimized to ensure image clarity. Each treatment was replicated three times.

Plant Tissue Analysis

Nitrogen content in plant tissue was determined using the Kjeldahl method, which involves acid digestion, distillation, and titration (Bremner, 1965). Phosphorus content was assessed through the development of a blue complex with ammonium molybdate and measured at 880 nm using a spectrophotometer (Murphy and Riley, 1962). Potassium was measured via flame photometry or atomic absorption spectroscopy, based on its emission or absorption at approximately 766.5 nm (Chapman and Pratt, 1962).

Data Analysis

All data were tabulated and analyzed using RStudio. Analysis of variance (ANOVA) was performed based on the split-plot design to determine significant differences among treatments. The Bonferroni test was used to compare treatment means at the 5% level of significance.

Results and Discussion

Soil Chemical Analysis

Initial and post-harvest analyses of soil nutrients are presented in Table 1. The total nitrogen and available phosphorus levels were initially high, indicating that the soil was already rich in these elements even before treatment application. After treatment, nitrogen remained high (0.15–0.16%), while phosphorus increased to 29.7 - 34.6 ppm across treatments.

In contrast, potassium levels, initially low at 13.1 ppm, increased to moderate levels after treatment, ranging from 55.8 to 66.6 ppm. The improvement in potassium levels is likely attributable to the synergistic effects of N and Si application, which may

have enhanced potassium availability and uptake. This improved nutrient profile contributes to better soil fertility and potentially higher crop performance.s

The initial soil pH was 5.9, indicating moderate acidity. Post-treatment pH levels varied slightly: S1N1 (without Si; 0% N) and S1N3 (without Si; 100% N) remained moderately acidic at 5.93 and 5.99, respectively. Treatments S1N2, S2N1, S2N2, and S2N3 exhibited slightly acidic pH values. These variations may be attributed to the types and amounts of nitrogen fertilizers applied, which can acidify the soil over time.

According to Frings et al. (2014), soil pH between 5 and 6 is generally sufficient for silicon availability, though texture influences solubility. At a pH of 9.8, maximum Si adsorption occurs. Sirisuntornlak et al. (2021) reported that Si's influence on pH is typically non-significant, suggesting that its application is viable across a broad pH range.

Soil organic matter (SOM) was initially very low. After treatment, SOM ranged from 2.98% to 3.44% still categorized as low, though slightly improved by N and Si application. These findings imply that Si might contribute to moderating soil acidity and marginally enhancing organic matter levels.

Assessment of Silicon Deposition in Rice Leaves using SEM-EDX

Silicon deposition in the leaves of NSIC Rc 160 was examined using scanning electron microscopy

(SEM) at 1,000x magnification (Figure 1). The resulting micrographs revealed silica bodies on the adaxial leaf surface, including papillae and stomata. Treatments with silicon fertilizer demonstrated a well-developed, ladder-like arrangement of silica bodies, in contrast to the undeveloped forms observed in treatments without silicon.

Among the silicon-treated plants, Treatment S2N2 (with Si; 50% N) (Figure 1c) displayed more distinctly defined dumbbell-shaped silica bodies. In these Si-treated samples, papillae and guard cells were visible over the stomatal complex adjacent to the dumbbell-shaped silica bodies (Figure 1a–d), potentially contributing to defense against pathogens. This observation aligns with the findings of Takahashi et al. (2006), who reported significant silicon deposition on papillae and trichomes. In the present study, trichomes were noticeably more prominent and elongated in Treatment S2N2 (with Si; 50% N) compared to those in Treatment S2N1 (with Si; 0% N).

Rice plants absorb silicon in the form of monosilicic acid (H_4SiO_4). According to Zhang et al. (2013) and Ma and Takahashi (2002), silicon is absorbed through the roots and gradually develops into dumbbell-shaped or bulliform-shaped silica bodies that align in rows along the leaf veins within the epidermis. Active silicon uptake in rice involves two key types of transporters, as noted by Rao and Susmitha (2017): the influx transporters Lsi1 and Lsi2, which move silicon from the root cells to the

Table 1. Effect of nitrogen and silicon fertilizer application on physicochemical properties of soil samples at initial and after harvest. The readings are based on the soil analysis conducted in PhilRice CES, Maligaya, Science City of Muñoz, Nueva Ecija. Ratings are from Landon (1991).

Soil samples	Total N (%)		Available P (ppm)		Available K (ppm)		рН		OM (%)	
	Reading	Rating	Reading	Rating	Reading	Rating	Reading	Rating	Reading	Rating
Initial	0.15	High	24.91	High	13.1	Low	5.96	Moderately Acidic	2.63	Very low
S1N1 (without Si; 0% N)	0.15	High	33.90	High	66.6	Moderate	5.93	Moderately Acidic	3.44	Low
S1N2 (without Si; 50% N)	0.16	High	30.52	High	56.9	Moderate	6.02	Slightly Acidic	3.44	Low
S1N3 (without Si; 100% N)	0.15	High	30.05	High	57.9	Moderate	5.99	Moderately Acidic	2.98	Low
S2N1(with Si; 0% N)	0.15	High	34.66	High	63.1	Moderate	6.06	Slightly Acidic	2.84	Low
S2N2 (with Si; 50% N)	0.15	High	31.35	High	55.8	Moderate	6.01	Slightly Acidic	2.80	Low
S2N3 (with Si; 100% N)	0.15	High	29.72	High	56.4	Moderate	6.05	Slightly Acidic	2.98	Low



Figure 1. Silicon deposition on the adaxial leaf surface of NSIC Rc160 at 1,000x magnification with different N-Si fertilization treatments; (a) S1N3 (without Si; 100% N), (b) S2N1 (with Si; 0 % N), (c) S2N2 (with Si; 50 % N), and (d) S2N3 (with Si; 100 % N).

apoplast, and the efflux transporter Lsi6, which facilitates silicon transport from large vascular bundles.

In this study, a considerable number of papillae were detected in treatments that received silicon. These findings corroborate the results of Ng et al. (2019), who found that Si-treated tissues exhibited more papillae compared to untreated samples. The formation of papillae - small outgrowths on plant surfaces associated with structural reinforcement and stress response - was enhanced by silicon application. Ning et al. (2014) similarly reported that silicon promoted more extensive silicification in rice leaves, along with larger and more elaborate papillae. The resistance of Si-treated leaves to fungal penetration has been attributed to this elaborated papillae formation, as discussed by Zhang et al. (2006) and Cai et al. (2008).

Overall, SEM-EDX analysis indicates that silicon application, particularly when combined with nitrogen, promotes structural defense features in rice leaves - including enhanced silica bodies, numerous papillae, and extended trichomes. These adaptations may strengthen the plant's resilience to biotic and abiotic stressors, thereby contributing to improved plant health and potentially higher yield performance.

Plant Tissue Analysis

The effect of silicon and nitrogen treatments on nutrient uptake in rice plants is presented in Table 2. Nitrogen levels in plant tissues were found to be deficient across all treatments, ranging from 0.42% to 0.50%. This deficiency may be attributed to the translocation of nitrogen to the developing grains during the ripening phase, a critical stage for yield formation. Huang et al. (2004) noted that nitrogen absorbed during earlier growth stages is largely translocated to the grains as the plant matures, with about 39% of absorbed N at mid-tillering and 46% at panicle initiation moving to the grain.

Regular monitoring of nutrient status across growth stages is thus essential for effective nutrient management and optimizing yield. In contrast to nitrogen, phosphorus (P) and potassium (K) levels were within the optimal range across all treatments, indicating sufficient availability and efficient uptake by rice plants.

Effects of N – Si Fertilizer in the Number of Filled Grains and 1,000 Grain Weight of Rice

The application of 100% nitrogen in combination with silicon resulted in the highest number of filled grains (941), outperforming the corresponding treatment without silicon (S1N3), which produced
891 filled grains. Similarly, Treatment S2N2 (with Si; 50% N) achieved a mean of 758 filled grains, which was higher than Treatment S1N2 (without Si; 50% N) with 739 filled grains. Even at 0% nitrogen, plants treated with silicon (S2N1) recorded a greater number of filled grains (521) than those without silicon (S1N1), which had 486 grains (Table 3).

These results underscore the beneficial role of silicon in improving rice yield, particularly when applied with nitrogen. Enhanced grain filling may be attributed to improved stress tolerance and increased chlorophyll content, as reported by Ligaba-Osena et al. (2020). The synergistic effect of Si and N appears to enhance nitrogen use efficiency, contributing to greater yield. Jawahar et al. (2015) similarly noted that silicon fertilization promotes carbohydrate assimilation in the panicles, increasing the number of filled grains.

In terms of 1,000-grain weight, the treatment with 100% nitrogen and no silicon recorded a slightly higher weight (24.2 g) compared to the treatment with silicon and 100% nitrogen (S2N3), which had a grain weight of 24.1 g. A similar trend was observed at the 50% nitrogen level: Treatment S1N2 (without Si) had a mean grain weight of 24.0 g, while S2N2 (with Si) had 23.9 g. In contrast, at 0% nitrogen, silicon-treated plants (S2N1) recorded a higher 1000-grain weight (23.7 g) than untreated ones (S1N1), which had 23.6 g. These results are supported by Pati et al. (2016) and Cuong et al. (2017), who found that silicon application can increase 1,000-grain weight.

Conclusion

This study demonstrated the significant contributions of silicon and nitrogen fertilization to rice growth and yield, supported by soil and tissue nutrient analyses. The initial soil assessment showed high total nitrogen and available phosphorus levels, with potassium increasing from a low baseline of 13.1 ppm to between 55.8 and 66.6 ppm across treatments after harvest, indicating that the interaction between Si and N may have enhanced K availability. Soil pH remained suitable for rice cultivation (5.9–6.0), although organic matter content was low (2.9–3.4%).

Silicon deposition, observed on the adaxial leaf surface via SEM, was most pronounced in Treatment S2N2 (with Si; 50% N). This treatment exhibited well-developed, ladder-like silica bodies and a greater abundance of protective structures such as papillae and trichomes, providing strong evidence for silicon's role in reinforcing rice plant defenses. Tissue analysis showed deficient nitrogen levels (0.42–0.50%), likely due to redistribution during grain filling, while phosphorus and potassium remained within optimal ranges, suggesting efficient uptake and utilization.

Yield assessments revealed that silicon application significantly increased the number of filled grains, particularly when combined with nitrogen. The highest grain count (941) was recorded in Treatment S2N3 (with Si; 100% N), outperforming its non-Si counterpart. Si-treated plots outperformed those

Table 2. Effect of silicon and nitrogen application on nutrient uptake of rice.

Treatmente	Present N (%)		Present P (%)		Present K (%)	
Treatments	Reading	Rating	Reading	Rating	Reading	Rating
S1N1 (without Si; 0% N)	0.50	Deficient	0.12	Optimum	1.64	Optimum
S1N2 (without Si; 50% N)	0.46	Deficient	0.10	Optimum	1.57	Optimum
S1N3 (without Si; 100% N)	0.47	Deficient	0.10	Optimum	1.41	Optimum
S2N1(with Si; 0% N)	0.36	Deficient	0.11	Optimum	1.67	Optimum
S2N2 (with Si; 50% N)	0.47	Deficient	0.11	Optimum	1.61	Optimum
S2N3 (with Si; 100% N)	0.42	Deficient	0.09	Optimum	1.50	Optimum

Ratings are from Landon (1991).

Table 3. Number of filled grains and weight of 1000 grains of NSIC Rc 160.

Treatments	Number of filled grains	Weight of 1,000 grains (g)
S1N1 (without Si; 0% N)	486 f	23.6
S1N2 (without Si; 50% N)	739 d	24.0
S1N3 (without Si; 100% N)	891 b	24.2
S2N1(with Si; 0% N)	521 e	23.7
S2N2 (with Si; 50% N)	758 c	23.9
S2N3 (with Si; 100% N)	941 a	24.1

Treatments with the same letter are not significantly different.

without Si across all nitrogen levels, indicating improved nitrogen use efficiency and grain filling.

The results support the use of silicon as a supplementary nutrient to enhance rice productivity and stress resilience. Further investigation is recommended to validate the performance of siliconnitrogen fertilization under field conditions and across various rice cultivars. In addition, research into the physiological and biochemical mechanisms underlying the interaction between silicon and nitrogen could inform the development of more effective and sustainable nutrient management practices for rice farming.

Literature Cited

- Bremner JM (1965) Nitrogen availability indexes. Methods of Soil Analysis: Part 2 Chemical and Microbiological Properties (pp 1324 - 1345)
- Cai K, Gao D, Luo S, Zeng R, Yang J, Zhu X (2008) Physiological and cytological mechanisms of siliconinduced resistance in rice against blast disease. Physiologia Plantarum 134(2): 324 - 333
- Chaiwong N, Prom-U-Thai C (2022) Significant roles of silicon for improving crop productivity and factors affecting silicon uptake and accumulation in rice: a review. Journal of Soil Science and Plant Nutrition 22(2): 1970 - 1982
- Chapman HD, Pratt PF (1962) Methods of analysis for soils, plants and waters. Soil Science 93: 68
- Cuong TX, Ullah H, Datta A, Hanh TC (2017) Effects of siliconbased fertilizer on growth, yield and nutrient uptake of rice in the tropical zone of Vietnam. Rice Science 24 (5): 283 -290
- Datta A, Ullah H, Ferdous Z (2017) Water management in rice (pp 255 277). *In* Rice Production Worldwide. Springer
- **Epstein E** (2009) Silicon: its manifold roles in plants. Annals of applied Biology *155*(2): 155 -160
- Frings PJ, Clymans W, Jeppesen E, Lauridsen TL, Struyf E, Conley DJ (2014) Lack of steady-state in the global biogeochemical Si cycle: emerging evidence from lake Si sequestration. Biogeochemistry 117: 255 - 277
- Gascho GJ (2001) Silicon sources for agriculture. In Studies in plant science (Vol. 8, pp. 197 207). Elsevier
- Guntzer F, Keller C, Meunier JD (2012) Benefits of plant silicon for crops: a review. Agronomy for sustainable development 32: 201 - 213
- Huang ZA, Jiang DA, Yang Y, Sun JW, Jin SH (2004) Effects of nitrogen deficiency on gas exchange, chlorophyll fluorescence, and antioxidant enzymes in leaves of rice plants. Photosynthetica 42: 357 - 364
- Jawahar S, Jain N, Suseendran K, Kalaiyarasan C, Kanagarajan R (2015) Effect of silixol granules on silicon uptake, stem borer and leaf folder incidence in rice. Int J Curr Res Acad Rev 3: 168 - 174

- Kumar S, Bhandari D (2022) Silicon: As a potential source tpests management. International Journal of Tropical Insect Science 42(5): 3221 - 3234
- Landon JR (ed) (1991) Booker Tropical Soil Manual; John Wiley and Sons Inc.; New York
- Ligaba-Osena A, Guo W, Choi SC, Limmer MA, Seyfferth AL, Hankoua BB (2020) Silicon enhances biomass and grain yield in an ancient crop tef [*Eragrostis tef* (Zucc.) Trotter]. Frontiers in Plant Science 11: 608503
- Lobell DB, Cassman KG, Field CB (2009) Crop yield gaps: their importance, magnitudes, and causes. Annual review of environment and resources 34: 179 - 204
- Ma JF, Takahashi E (2002) Soil, fertilizer, and plant silicon research in Japan. Elsevier
- Mamiit RJ, Yanagida J, Miura T (2021) Productivity hot spots and cold spots: Setting Geographic priorities for achieving food production targets. Frontiers in Sustainable Food Systems 5: 727484
- Murphy J, Riley JP (1962) A modified single solution method for the determination of phosphate in natural waters. Analytica Chimica Acta 27: 31 - 36
- Ng LC, Hafiz ES, Sariam O, Razi RI (2019) The effect of calcium silicate as foliar application on aerobic rice blast disease development. European journal of plant pathology *153*: 533 - 543
- Ning D, Song A, Fan F, Li Z, Liang Y (2014) Effects of slag-based silicon fertilizer on rice growth and brown-spot resistance. *Plos one* 9(7):e102681
- Pati S, Pal B, Badole S, Hazra GC, Mandal B (2016) Effect of silicon fertilization on growth, yield, and nutrient uptake of rice. Communications in Soil Science and Plant Analysis 47(3): 284 - 290
- Rao GB, Susmitha PJ (2017) Silicon uptake, transportation and accumulation in Rice. J Pharmacogn Phytochem 6(6): 290 - 293
- Rütting T, Aronsson H, Delin S (2018) Efficient use of nitrogen in agriculture. Nutrient Cycling in Agroecosystems 110: 1 - 5
- Sarma RS, Shankhdhar D, Shankhdhar SC (2019) Beneficial effects of silicon fertilizers on disease and insect-pest management in rice genotypes (*Oryza sativa*. L). Journal of Pharmacognosy and Phytochemistry 8(3): 358 - 362
- Sirisuntornlak N, Ullah H, Sonjaroon W, Anusontpornperm S, Arirob W, Datta A (2021) Interactive effects of silicon and soil pH on growth, yield and nutrient uptake of maize. Silicon 13(2): 289 - 299
- Takahashi N, Kato Y, Isogai A, Kurata K (2006) Silica distribution on the husk epidermis at different parts of the panicle in rice (Oryza sativa L.) determined by X-ray microanalysis. Plant Production Science 9(2): 168 - 171
- Wang B, Zhou G, Guo S, Li X, Yuan J, Hu A (2022) Improving nitrogen use efficiency in rice for sustainable agriculture: strategies and future perspectives. Life *12*(10): 1653

- Zhang C, Wang L, Zhang W, Zhang F (2013) Do lignification and silicification of the cell wall precede silicon deposition in the silica cell of the rice (*Oryza sativa* L.) leaf epidermis? Plant and Soil 372: 137 - 149
- Zhang GL, Dai QG, Zhang HC (2006) Silicon Application enhances resistance to sheath blight (*Rhizoctonia solani*) in rice. Journal of plant Physiology and Molecular Biology 32(5): 600 - 606
- Zhu D, Zhang H, Guo B, Xu K, Dai Q, Wei C, Zhou G, Huo Z (2017) Effects of nitrogen level on structure and physicochemical properties of rice starch. Food Hydrocolloids 63: 525 - 532

COMPARATIVE STUDY ON PRODUCTIVITY AND PROFITABILITY OF INBRED LOWLAND RICE (*ORYZA SATIVA* L.) UNDER DIFFERENT CROP ESTABLISHMENT METHOD

Flerida A. Demamay^{*}, William P. Opher, Jr., Lyka M. Hisu-an, Rener Jay B. Duran, Denmarie T. Lodovice, Emheryn Grace S. Calitisin, Weldien C. Gequillo, and Aljen P. Vasquez

Department of Agriculture 6-Research Division and DA-WESVIARC, Hamungaya, Buntatala, Jaro, Iloilo City *Corresponding Author: fleridademamay@gmail.com

Abstract

Rice production practices in the Philippines are evolving with advancements in technology and government initiatives aimed at addressing the country's agricultural challenges. Traditional methods like manual transplanting have been supplemented by technologies such as mechanical transplanters and knapsack-type seeding machines. Concurrently, manual broadcasting or line-seeding of pre-germinated seeds on wet soils is gaining popularity among farmers. This study aimed to determine the most profitable method for achieving optimal rice growth and potential yield. The research compared mechanical and manual transplanting with mechanical and manual direct seeding on 0.25 ha plots replicated thrice across wet and dry cropping seasons in irrigated lowland ecosystems. Results showed that mechanical transplanting produced the highest number of productive tillers (23), tallest plants (109.06 cm), and highest 1,000-grain weight (25.02 g), resulting in the highest yield (5.27 t ha⁻¹). Conversely, manual direct seeding showed lower performance in these parameters, yielding 4.47 t ha⁻¹ with 17 tillers. Cost and return analysis indicated that mechanical direct seeding with a knapsack-type machine provided the highest return on investment (ROI) at 64.05%. Mechanization not only reduced labor costs but also enhanced crop establishment efficiency, contributing to a significant increase in farmer income.

Keywords: crop establishment methods, direct seeding, mechanical transplanting, manual transplanting, return of investment

Introduction

In 2018, the Philippines ranked eighth globally in rice production, with an average annual yield of 3.83 t ha⁻¹ (GYGA, 2020). Rice is predominantly cultivated in Luzon, Western Visayas, Southern Mindanao, and Central Mindanao. Western Visayas, for instance, contributes 11.90% to the national production, led by Iloilo (43%), followed by Negros Occidental (21%) and Capiz (16%) (DA-Western Visayas, 2021). The region's rice sufficiency reached 122% in 2020, up from 115% in 2019, driven by yield increases in Aklan and Iloilo (DA-Western Visayas, 2021).

The landscape of rice production in the Philippines is shaped by evolving technologies and government programs designed to meet the dynamic needs of its population. With rice serving as a staple for the majority of the 100.98 million Filipinos, ensuring its steady supply is crucial (Tallada, 2019). Moreover, rice production is politically significant, reflecting the nation's food security status.

The Department of Agriculture – Philippine Rice Research Institute (DA-PhilRice) advocates for optimal seeding rates to enhance yields and reduce production costs. Recommended rates include 40 kg ha⁻¹ for transplanted rice and 60–80 kg ha⁻¹ for directseeded rice (DA, 2022). Farmers, who previously used 200–250 kg ha⁻¹, now opt for 100–150 kg ha⁻¹, citing these as buffers against pests such as snails and birds (DA, 2022).

Traditional manual transplanting remains widely practiced despite its labor-intensive nature, requiring up to 30 laborers per hectare compared to mechanized methods requiring only 1 - 2 laborers (IRRI, 2021). However, the availability of manual labor has dwindled, posing challenges to traditional practices.

To address these challenges, various rice cultivation technologies have emerged, including mechanical transplanters and seed sowers. Concurrently, manual broadcasting of pre-germinated seeds on wet soils is gaining traction among farmers.

Mechanical transplanting involves planting young rice seedlings from trays or dapog beds using selfpropelled machines capable of planting up to 2 - 3 hectares per day. This method minimizes transplant shock, promotes uniform crop stands, and requires less water compared to manual transplanting (DA-PhilRice, 2021).

Direct seeding methods vary by region and season, with dry-direct seeding common in upland and rainfed areas, and wet-direct seeding practiced in irrigated lowlands. Direct seeding reduces labor and crop management costs by allowing earlier maturation than transplanted rice.

The Department of Agriculture - Western Visayas has distributed 1,543 units of knapsack-type seed sowers to farmer associations to address seed shortages. These machines, powered by gasoline engines, facilitate efficient broadcasting of seeds, fertilizers, and pesticides (DA-Western Visayas, 2021).

This study aimed to fill gaps in research by comparing the yield and profitability of four rice establishment methods: manual transplanting, mechanical transplanting, manual direct seeding, and mechanical direct seeding. The findings can inform future innovations and promote mechanization in rice production.

Objectives

General Objectives:

To compare the performance of four establishment methods in irrigated inbred lowland rice for optimal growth and yield.

Specific Objectives:

Evaluate growth and yield parameters - plant height, number of tillers, panicle length, filled grains, 1000-grain weight, and grain yield - across different establishment methods.

Determine the economic benefits of each establishment method in irrigated inbred lowland rice.

Materials and Methods

Research Design

This study employed a Randomized Complete Block Design (RCBD) with three blocks, each containing four plots for the four treatments. Each plot covered 0.25 hectares, replicated thrice across wet and dry cropping seasons in an irrigated ecosystem. Quantitative data were statistically analyzed using Analysis of Variance (ANOVA).

The study included four establishment methods: mechanical transplanting, manual transplanting, mechanical direct seeding, and manual direct seeding. Figure 1 illustrates the experimental layout.

Research Procedure

The rice cultural practices conducted in the field followed standard guidelines, with modifications specific to each planting method.

These are detailed as follows:

Land preparation followed PalayCheck System Key Check No. 2.

Mechanical Transplanting

For mechanical transplanting: In addition to Key Check No. 2, the field underwent final leveling 2 - 3 days before transplanting. During final leveling:

- The field was filled with water to a depth of at least 10 cm, with no soil mounds visible above the water surface.
- After 24 hours, once the water had cleared and golden apple snails resurfaced, a molluscicide was applied.
- The field was then left undisturbed for another 24 hours.

This timing allowed the newly leveled soil to settle and prevented it from liquefying during mechanical transplanting operations.

For manual transplanting, mechanical direct seeding, and manual direct seeding: Final leveling was done a day before or on the day of crop establishment, also in accordance with Key Check No. 2.

BLOCK 1			BLOCK 2			BLOCK 3						
	ТЗ	T2	T1	T4	T2	ТЗ	T4	T1	ТЗ	T2	T1	T4

Figure 1. Experimental layout arranged in RCBD.

where:

T1 - Mechanical Transplanting

T2 - Manual Transplanting T3 - Mechanical Direct Seeding

T3 - Mechanical Direct Seeding



Figure 2. Satellite image of the research area.

Seedbed and Seedling Preparation

For manual transplanting:

- Land for the seedbed was saturated with irrigation water for 1-2 weeks and harrowed twice to remove weeds and old crop residues.
- During the last harrowing, 100 kg of compost or vermicast was incorporated.
- A 500 m² seedbed was prepared for every 1 ha planting area, raised 3 cm from ground level, and 1.5 m wide.
- Pre-germinated seeds (at 20 kg ha⁻¹ or 50 g m⁻²) were evenly sown and covered with sacks to protect against birds.
- Sacks were removed after emergence of 1 2 leaves.
- T-14 fertilizer was applied when seedlings were 12 15 days old.
- At 20 days, seedlings were pulled and bundled manually for field transplanting.
- For mechanical transplanting:
- Registered NSIC Rc 480 seeds were used at 40 kg ha⁻¹ to produce 230 250 seedling trays per hectare.
- The seedling media consisted of 8 bags of sieved garden soil (50 kg each) and 2 bags of organic fertilizer.
- Seeds were soaked for 24 h and incubated for another 24 h before sowing.
- A seed sowing machine was used to distribute pre-germinated seeds in trays, which were then covered with a thin layer of soil.

- Trays were stacked for three days, then exposed to sunlight.
- Watering was done twice daily using a knapsack sprayer for the first three days.
- Once roots had anchored, a water sprinkler or hose was used.

Seed Pre-Germination for Direct Seeding

Mechanical direct seeding: 60 kg ha⁻¹

Manual direct seeding: 80 kg ha-1

For these methods, registered NSIC Rc 480 seeds were used. Seeds were soaked for 24 h and incubated for another 24 h, three days prior to crop establishment.

Crop Establishment

Crop was established simultaneously for all planting methods.

Direct seeding:

- Pre-germinated seeds were sown directly after final leveling.
- Mechanical direct seeding used a granular spreader/applicator at 60 kg ha⁻¹.
- Manual direct seeding used a rate of 80 kg ha⁻¹.

Manual transplanting:

- 18-day-old seedlings were transplanted immediately after pulling to avoid stress.
- Planting rate: 1 seedling per hill, spaced 20 cm x 20 cm (row x hill).

Mechanical transplanting:

• 14-day-old seedlings were transplanted using a riding-type rice mechanical transplanter.

 Table 1. Crop establishment method with its corresponding applied seeding rate.

Crop Establishment Method	Applied Seeding Rate (kg ha ⁻¹)
Mechanical Transplanting	40
Manual Transplanting	20
Mechanical Direct Seeding	60
Manual Direct Seeding	80

Fertilizer Application

Rice Crop Manager (RCM) fertilizer recommendation was used for nutrient management.

Water Management. The field was irrigated with 5 - 7 cm water depth from 10 to 35 days after transplanting/sowing (DAT/DAS) to facilitate fertilizer application and weed control. Starting at 42 DAT/DAS, the field was irrigated intermittently until seven days before harvest.

Pest Management. Correct pest identification and the application of integrated pest management technologies were observed.

Weed Management. A pre-emergence herbicide was applied at 1–3 DAT/DAS, or an early post-emergence herbicide was applied at 6–8 DAS. If necessary, post-emergence herbicide was applied at 15–30 DAT/DAS, followed by hand weeding as needed

Harvesting and Processing. The rice crop was harvested using a Rice Combine Harvester when 85–90% of the grains at the upper portion of the panicle were golden yellow or straw-colored, and 20% of the grains at the base of the panicle were at the hard dough stage.

Data Gathered:

- 1. Plant Height Plant height was measured from 10 randomly selected hills using a meter stick for each treatment. It refers to the length from the base of the plant to the tip of the highest flag leaf and was recorded before harvest.
- Number of Productive Tillers Data was collected from three randomly selected 1 m² quadrants at 45 days after planting for each treatment.
- 3. Panicle length Panicle length refers to the measurement from the base to the tip of the panicle, excluding the awn. This was measured from 10 randomly selected hills for each treatment.

- Number of filled grains This was determined by counting the filled grains from 10 randomly selected hills per treatment.
- 5. 1,000 grain weight This refers to the weight of 1,000 grains collected from each treatment.

Parameters calculated:

Grain Yield. Grain yield was determined using the crop cut method, a standard technique for estimating yield within a specific unit area. This was done by establishing three randomly selected 5 m^2 quadrants for each replication per treatment. The crop within each quadrant was manually harvested, stripped, threshed, and winnowed. The fresh paddy was then weighed, and the result served as the basis for determining the grain yield of the study.

It was computed using the formula:

$$Grain Yield = \left(\frac{yields \, per \, sub \, plot}{sub \, plot \, area \, (m^2)}\right)\left(\frac{10,000 \, (m^2)}{1000}\right)$$

Return of Investment. This was determined by dividing the net profit to the total investment utilized in this study, which is expressed in percentage.

$$ROI = (\frac{Net \, Profit}{Total \, Investment})(100)$$

Results and Discussion

Wet Season Growth Performance of Rice Using Different Crop Establishment Methods

The data on the number of productive tillers, plant height, panicle length, number of filled grains, weight of 1,000 grains, and yield using different crop establishment methods during the 2020 and 2021 wet seasons are shown in Table 2. Analysis showed comparable results among the four crop establishment methods in terms of productive tillers, panicle length, number of filled grains, 1,000-grain weight, and yield.

However, in terms of plant height, similar values were observed between the two direct seeding methods (manual and mechanical), and between the two transplanting methods (manual and mechanical). A significant difference, however, was found between direct seeding and manual transplanting methods, indicating that method selection may affect plant height.

Dry Season Growth Performance of Rice Using Different Crop Establishment Methods

The data for the 2021 and 2022 dry seasons are presented in Table 3. Results showed that the four crop establishment methods produced comparable values for productive tillers, plant height, panicle length, number of filled grains, 1,000-grain weight, and yield, indicating that each method performed similarly under dry season conditions.

Yield Performance of Rice Using Different Crop Establishment Methods

Table 4 presents the yield performance across four cropping seasons using different crop establishment methods. In the 2020 wet season (WS) and 2022 dry season (DS), results revealed no significant difference in yield among the four methods.

However, in the 2021 dry season, the yield was comparable among manual direct seeding, mechanical direct seeding, and manual transplanting, but the mechanical transplanting method yielded significantly higher than both direct seeding methods and was statistically similar to manual transplanting.

In contrast, the 2021 wet season data showed comparable yields between the two direct seeding methods (manual and mechanical), and also between the two transplanting methods (manual and mechanical). Additionally, manual and mechanical transplanting methods produced significantly higher yields than the manual and mechanical direct seeding methods.

Growth Performance of Rice Using Different Crop Establishment Methods

The growth parameters across all trials are summarized in Table 5. Results showed comparable outcomes for productive tillers, plant height, panicle length, 1000-grain weight, and yield among the four methods.

However, in terms of number of filled grains, manual and mechanical direct seeding methods recorded comparable values (86 and 90 grains, respectively). In contrast, manual and mechanical transplanting produced a notably higher number of filled grains (115 and 116 grains, respectively). A significant difference was also observed between

Table 2. Means for agronomic and yield properties of lowland rice during 2020 and 2021 wet season cropping.

Treatment	Productive Tiller ^{ns}	Plant Height	Panicle Length ^{ns}	Filled Grains ^{ns}	1,000 grains ^{ns}	Yield ^{ns} (t ha ⁻¹)
Manual Direct seeding	20.34	106.25b	21.99	82.34	25.30	3.62
Mechanical Direct seeding	22.17	106.88b	22.04	73.67	25.47	4.51
Mechanical Transplanting	19.50	112.16a	22.87	104.34	26.68	4.74
Manual Transplanting	22.84	114.45a	22.31	106.17	26.68	4.61
Mean	21.21	109.94	22.30	91.63	26.03	4.37
CV (%)	7.46	1.50	3.94	21.42	1.74	15.50

means with the same letter are not significantly different at 5% level by Least Significant Difference (LSD) Test

Table 3. Means for agronomic and yield properties of lowland rice during 2021 and 2022 dry sea
--

Treatment	Productive Tiller ^{ns}	Plant Height ^{ns}	Panicle Length ^{ns}	Filled Grains ^{ns}	1,000 Grains⁵	Yield ^{ns} (t ha ⁻¹)
Manual Direct Seeding	14.34	101.09	21.03	88.5	22.28	5.32
Mechanical Direct Seeding	17.34	97.16	21.18	105.83	23.48	5.43
Mechanical Transplanting	17.00	95.05	23.51	124.83	21.55	5.22
Manual Transplanting	23.00	103.67	23.09	125.50	23.32	5.92
Mean	17.92	99.25	22.20	111.17	22.66	5.47
CV (%)	17.45	2.82	3.47	9.27	2.06	5.59

Table 4. Yield properties of lowland rice for four cropping season.

Treatment	Yield (t ha-1)						
	2020 WS ^{ns}	2021 DS	2021 WS	2022 DS ^{ns}			
Manual Direct Seeding	2.58	5.88b	4.66b	4.76			
Mechanical Direct Seeding	3.86	5.97b	5.17b	4.89			
Mechanical Transplanting	3.03	6.51a	6.18a	5.32			
Manual Transplanting	3.06	6.21ab	6.42a	4.22			
Mean	3.13	6.14	5.61	4.80			
CV (%)	21.98	3.24	7.23	9.22			

means with the same letter are not significantly different at 5% level by Least Significant Difference (LSD) Test

Treatment	Productive Tiller ^{ns}	Plant Height ^{ns}	Panicle Length ^{ns}	Filled Grains	1,000 Grains ^{ns}	Yield ^{ns} (t ha ⁻¹)
Manual Direct Seeding	17	103.67	21.5	86 ^b	23.78	4.47
Mechanical Direct Seeding	20	102.02	21.61	90 ^b	24.48	4.97
Mechanical Transplanting	23	109.05	22.7	116ª	24.98	5.26
Manual Transplanting	18	103.6	23.18	115ª	24.11	4.98
Mean	19.5	104.59	22.25	101.75	24.34	4.92
CV (%)	13.09	3.25	3.95	13.93	3.43	10.68

Table 5. Means for agronomic and yield properties of lowland rice across season.

means with the same letter are not significantly different at 5% level by Least Significant Difference (LSD) Test

Table 6. Time of operation for different crop establishment method per hectare.

Treatment	Crop Establishment Method	Time (per hectare)
1	Mechanical Transplanting	1:2:04
2	Manual Transplanting	4:52:10
3	Mechanical Direct Seeding	33:34
4	Manual Direct Seeding	49:36

mechanical transplanting and both direct seeding methods.

profitable option, with a net income of PhP 12,251.50 and an ROI of 22.06%.

Length of Operating Time for Each Crop Establishment Method

The time required for field operations per hectare using different establishment methods is presented in Table 6. Among the four, the Seed Sower (mechanical direct seeding) completed operations in the shortest time, while manual transplanting required the longest.

Cost and Return Analysis

Tables 7 and 8 present the cost and return analysis of the different crop establishment methods across two consecutive wet and dry cropping seasons. During the wet seasons, mechanical direct seeding emerged as the most profitable method, registering the highest net income at PhP 18,105.50 and a return on investment (ROI) of 44.78%. This was followed closely by mechanical transplanting, which posted a net income of PhP 16,372.50 and an ROI of 37.64%. Manual direct seeding generated a net income of PhP 5,777.50 and an ROI of 14%, while manual transplanting recorded the lowest net income of PhP 5,478.00, with an ROI of 9.76%.

In the dry seasons, the cost and return data revealed a different trend. Mechanical transplanting resulted in the highest net income of PhP 36,251.00 and an ROI of 89.19%. This was followed by mechanical direct seeding, which produced a net income of PhP 32,183.50 and an ROI of 83.80%. Manual direct seeding yielded a net income of PhP 29,543.00 and an ROI of 74.47%. As in the wet season, manual transplanting remained the least

Table 9 summarizes the average labor cost, input cost, total production cost, gross income, net income, and ROI across all establishment methods. Mechanical transplanting achieved the highest average yield at 5,260 kg ha⁻¹, with a production cost of PhP 42,068.25 per hectare. Given a selling price of PhP 13 kg⁻¹, this translated to a gross income of PhP 68,380.00 and a net income of PhP 26,311.75 ha⁻¹.

When ranked based on net profit, mechanical transplanting led all methods, followed by mechanical direct seeding, manual direct seeding, and manual transplanting. However, in terms of ROI, mechanical direct seeding outperformed all other methods, posting the highest ROI at 64.05%, largely due to its lower production costs. Manual transplanting consistently registered the lowest ROI, primarily attributable to higher labor expenses incurred during crop establishment.

Conclusion

Based on the study results, mechanical transplanting showed superior performance in terms of productive tillers, number of filled grains, plant height, and 1000-grain weight. In contrast, manual direct seeding had the lowest performance across these indicators.

Mechanical transplanting minimized transplanting shock caused by root damage, resulting in early seedling vigor and a uniform crop stand. Younger seedlings, proper row spacing (30 x 16 cm), and Table 7. Cost and return analysis during wet season cropping.

	Mechanical Transplanting	Manual Transplanting	Mechanical Direct Seeding	Manual Direct Seeding
Labor Cost (PhP)	24,582.00	33,278.50	21,132.00	21,232.00
Input Cost (PhP)	18,910.50	22,863.50	19,430.50	20,050.50
TOTAL PRODUCTION COST	43,492.50	56,142.00	40,430.50	41,282.50
Production				
Gross Yield (kg)	4605	4740	4513	3620
Price kg ⁻¹ (PhP)	13.00	13.00	13.00	13.00
Gross Income (PhP)	59,865.00	61,620.00	58,669.00	47,060.00
Net Income (PhP)	16,372.50	5,478.00	18,106.50	5,777.50
Return of Investment	37.64%	9.76%	44.78%	14.00%

Table 8. Cost and return analysis during dry season cropping.

	Mechanical Transplanting	Manual Transplanting	Mechanical Direct Seeding	Manual Direct Seeding
Labor Cost (PhP)	20,850.00	36,217.00	18,025.00	18,925.00
Input Cost (PhP)	19,794.00	19,326.50	20,381.50	20,744.00
TOTAL PRODUCTION COST	40,644.00	55,543.50	38,406.50	39,669.00
Production				
Gross Yield (kg)	5915	5215	5430	5324
Price kg ⁻¹ (PhP)	13.00	13.00	13.00	13.00
Gross Income (PhP)	76,895.00	67,795.00	70,590.00	69,212.00
Net Income (PhP)	36,251.00	12,251.50	32,183.50	29,543.00
Return of Investment	89.19%	22.06%	83.80%	74.47%

Table 9. Cost and return analysis across season.

	Mechanical Transplanting	Manual Transplanting	Mechanical Direct Seeding	Manual Direct Seeding
Labor Cost (PhP)	22,716.00	34,747.75	19,491.00	19,991.00
Input Cost (PhP)	19,352.25	19,028.50	19,906.00	20,397.25
TOTAL PRODUCTION COST	42,068.25	53,776.25	39,397.00	40,388.25
Production				
Gross Yield (kg)	5260	4977.5	4971.5	4472
Price kg ⁻¹ (PhP)	13.00	13.00	13.00	13.00
Gross Income (PhP)	68,380.00	64,707.50	64,629.50	58,136.00
Net Income (PhP)	26,311.75	10,931.25	25,232.50	17,747.75
Return of Investment	62.54%	20.38%	64.05%	43.94%

sufficient light penetration encouraged vigorous growth and reduced pest infestation.

Among the four methods tested over four cropping seasons, mechanical transplanting consistently produced the highest yields due to adequate plant spacing and strong root anchorage, which promoted better tillering behavior. Conversely, the lower yield in manual direct seeding was likely due to poor plant spacing, which led to nutrient competition.

Furthermore, the mechanical methods (especially transplanting) required less labor, reduced seed input, and delivered higher ROI, making them more economically viable.

Despite this, the mechanical direct seeding method was found to be the most profitable for both dry and wet seasons, due to its lower costs and higher ROI.

Recommendations

Analysis of the data on productive tillers, plant height, panicle length, number of filled grains, grain yield, time of operation, and return on investment (ROI) suggests differentiated recommendations based farmers' production goals. on Mechanical transplanting is recommended for farmers engaged in seed production, as it enables faster and more efficient field operations with reduced physical labor requirements. This method minimizes health risks and stress while enhancing operational efficiency per hectare - particularly advantageous in areas labor shortages. experiencing Furthermore, mechanical transplanting facilitates timely and synchronized planting, which contributes to improved yield performance and higher income.

Conversely, mechanical direct seeding is best suited for farmers targeting commercial milling and grain consumption markets. This method is characterized by a shorter operating time and lower input costs, and it demonstrated the highest ROI among all crop establishment methods evaluated in the study. Thus, it offers an economically attractive option for maximizing profit while minimizing resource inputs.

Literature Cited

- Bautista E, Javier E (2005) The evolution of rice production practices. Philippine Institute for Development Studies. Discussion paper series no. 2005-14. Retrieved from https:/ /pidswebs.pids.gov.ph/CDN/PUBLICATIONS/ris-oldbackups/dps/pidsdps0514.pdf (accessed February 25, 2021)
- Business World (2021) Palay production costs top 47,000 per hectare in 2020. Retrieved from https://www.bworldonline. com/agribusiness/2021/11/01/407319/palay-productioncosts-top-p47000-per-hectare-in-2020/ 2021) (accessed February 25, 2021)
- DA [Department of Agriculture] (2022) From PNA: Rice farmers urged to follow seeding rate to improve yield. Retrieved from https://www.da.gov.ph/from-pna-rice-farmers-urgedto-follow-seeding-rate-to-improve-yield/. (accessed March 12, 2022)
- **DA-PHILRICE** [Department of Agriculture-Philippines Rice Research Institute] (2021) RICE (pp 1 - 2). Technology Bulletin
- DA-Western Visayas [Department of Agriculture-Western Visayas] (2021) Agricultural performance Retrieved from https://westernvisayas.da.gov.ph/agriculturalperformance/.(accessed February 25, 2021)
- GYGA [Global Yield Gap Atlas] (2020) Rice production in the five southeast Asian countries, Vietnam, Thailand, Myanmar, Philippines, and Cambodia Retrieved from https://www.yieldgap.org/philippines#: ~:text=The%20Philippines%20ranked%20eighth%20in,2 008%20(FAOSTAT%2C%202020) (accessed February 25, 2021)
- IRRI [International Rice Research Institute] (2021) Manual Transplanting. Retrieved from http://www. knowledgebank.irri.org/training/fact-sheets/cropestablishment/manual-transplanting (accessed February 25, 2021)

BLOCKCHAIN: A DIGITAL PLATFORM DATABASE FOR RICE SCIENCE

Eduardo Jimmy P. Quilang^{1*}, Hazel B. Alfon¹, Ritesh Amin², Raymond Josef Edward A. Lara³, Arturo C. Arocena¹, and Luis Alejandre I. Tamani

> ¹Philippine Rice Research Institute, Maligaya, Science City of Muñoz, Nueva Ecija ²IBM ASEANZK ³IBM Philippines, Incorporated *Corresponding Author: ejpquilang@philrice.gov.ph

Abstract

Harnessing timely and location-specific data is essential for designing innovative and cost-effective interventions to achieve the Philippines' rice self-sufficiency and affordability goals. However, ensuring farmers' sustainable engagement in data sharing requires a transparent, secure, and positively reinforced system where the blockchain technology can play a transformative role.

This study aimed to develop a blockchain artifact that is capable of gathering, securing, and managing data across multiple activities, functions, locations, processes, stakeholders, and other components, initially focusing on the higher seed class production function of the Philippine Rice Research Institute (PhilRice). Once proven as a strategic initiative, the artifact may be expanded to cover the entire Philippine rice value chain. Moreover, should this artifact be adopted for national implementation, the resulting system is expected to enhance data transparency, traceability, and security, ultimately contributing to the modernization and sustainability of the Philippine rice industry.

The study outlines the Proof of Experience (PoX) methodology for developing a Blockchain-based Traceability Management artifact, tracking rice from breeder seed production to distribution. It also showcases PoX processes within a public-private partnership (PPP) model.

The PoX for developing the Blockchain-based Traceability Management artifact was executed in collaboration with an international IT industry leader, IBM, through IBM ASEANX and IBM Philippines, Inc. The experiences gained by PhilRice from this initiative can serve as a blueprint for similar applications across government programs in agriculture, forestry, and fisheries.

Keywords: artifact, blockchain technology, provenance, rice science, supply chain management, traceability

Introduction

Atzori (2016) and Tasca and Tessone (2018), as cited in Pelt et al., (2021), described blockchain technology as a system that allows unacquainted parties to reach consensus on a shared data administration without requiring human intervention, centralized control, or regulatory oversight. Hacker (2017) further explains that a blockchain's software framework defines the rules governing transactions, the timing of new block additions, and the size limitations of data blocks (Pelt et al., 2021).

Blockchain is a software-based data management protocol that secures information by timestamping recorded data and distributing it across multiple computers, ensuring it cannot be lost, altered, or manipulated without consensus or approval from the entire network. To simplify, imagine a field researcher's notebook filled with valuable data. If the notebook is lost or damaged and there is no backup, all the time, effort, and research investments are wasted. If the researcher saves the data on a computer, it might still be lost due to system failure or tampered with by hackers. Now, think of blockchain as a network of digital notebooks stored across many computers. When data is recorded on a blockchain, it is replicated and stored on multiple computers worldwide. If someone tries to alter or erase the data, they would have to modify every copy on all computers in the network making fraud or manipulation nearly impossible.

Additionally, while new information can be added or updated, the original record remains intact, ensuring a transparent, secure, and tamper-proof history of all entries.

History of Blockchain

Blockchain technology was first introduced in 2008 by the pseudonymous entity Satoshi Nakamoto (Tripathi et al., 2023). Since then, it has evolved through four distinct generations. The first generation marked the inception of decentralized digital currencies, enabling secure and transparent online financial transactions most notably, the development of Bitcoin (Tripathi et al., 2023). The second generation expanded the technology's capabilities through the introduction of smart contracts and self-

executing scripts that automate processes within blockchain networks (The Investopedia Team, 2024). This innovation extended blockchain's applications to areas such as loans, equity trading, and the management of digital assets. The third generation broadened its use beyond finance, supporting critical sectors including scientific research, healthcare, and supply chain management. Currently, the fourth generation of blockchain is being developed with a focus on integration with emerging technologies such as artificial intelligence (AI), the Internet of Things (IoT), and other forms of digital intelligence (Tripathi et al., 2023).

Provenance Data Management for Rice Seed Traceability Using Blockchain Technology

In 2020, the Philippine Rice Research Institute (PhilRice) initiated a blockchain-based program in rice science aimed at addressing persistent inefficiencies in rice production and marketing in the Philippines. The primary objective was to uncover the root causes of high production costs and limited market returns, which have contributed to elevated consumer prices without translating into improved livelihoods for the majority of rice farmers. This disconnect between market pricing and farmer welfare necessitates a thorough investigation of systemic inefficiencies so that precise, cost-effective interventions can be developed and implemented.

Central to this effort is the collection and analysis of provenance (or historical) data directly from farmers and other stakeholders across the Philippine rice value chain. These include government agencies mandated to ensure national rice self-sufficiency and enhance sector productivity. Identifying the underlying inefficiencies not only enables bettertargeted policy and investment decisions but also ensures these decisions support the long-term welfare of both producers and value chain actors.

This report contextualizes "rice science" by aligning it with the institutional mandate of PhilRice, as defined in Executive Order No. 1061, Series of 1985. The mandate establishes PhilRice as the lead agency responsible for advancing rice productivity in the Philippines through innovations that improve farmer incomes, expand rural employment, and promote national self-sufficiency in rice production (Office of the President of the Philippines, 1985). The agency is tasked with addressing both biological and socio-economic challenges that arise within dynamic agricultural systems. Key focus areas include improving lowland irrigated rice production through the deployment of high-yielding cultivars, optimal use of agro-inputs, and sustainable water management. Additionally, PhilRice works to address yield gaps in ecologically challenging zones such as uplands, saline

soils, drought- and flood-prone areas, and rainfed and cool environments.

Complementing this institutional perspective is the broader academic framework provided by the Crop Science Society of America (CSSA), which defines "crop science" as the discipline committed to ensuring a reliable supply of safe and nutritious food (CSSA, 2024). Key domains under this umbrella include crop breeding, seed technology, genetics, physiology, conservation, sustainability, and nutritional quality. When applied to rice, these domains are reflected in PhilRice's core activities: (1) breeding of modern rice varieties, (2) rice seed technology, (3) rice genetics, (4) agronomy, soils, and physiology, (5) rice-based farming systems, (6) conservation of rice genetic resources, and (7) innovation in novel rice products and nutritional enhancements.

One of PhilRice's most significant contributions to the national rice economy is the development of new rice varieties and the production of high-quality seed classes such as breeder, foundation, and registered seeds. This report will therefore focus on how blockchain technology was applied to enhance traceability in the production and distribution of inbred rice seed varieties. Specifically, the system was designed to securely collect, store, and manage provenance data across the supply chain from breeder seed production through to registered seed production within PhilRice's branch and satellite stations.

Related Studies

The Blockchain-based Traceability Management System for Rice Science aims to enhance the efficiency and transparency of the Philippine rice production system through data-driven, targeted interventions. These interventions are informed by provenance data gathered directly from stakeholders across the rice value chain; farmers, seed producers, processors, and distributors ensuring that solutions are grounded in actual field realities. However, the scope of this report is intentionally narrow: it focuses solely on the development of a blockchain-based artifact tailored for higher seed class production and seed traceability. Specifically, the concept is about documenting the technical development of the system through an outsourced approach, without addressing its broader deployment or integration across the full rice value chain.

This section reviews foundational studies and frameworks that inform the conceptual and technical design of the artifact. It surveys literature on the use of blockchain for traceability and provenance management, with particular attention to agricultural applications. Emphasis is placed on the seed-to-rice continuum, from breeder and foundation seed production to the traceability of certified seeds highlighting the specific domain addressed by the system under development.

Supply Chain Management as the Foundation of the Blockchain Artifact

The immediate application of the blockchain artifact in this study centers on enhancing the efficiency of supply chain management (SCM) across the seed-to-milled rice production and consumption spectrum. Kamble et al. (2019), drawing from the Council of Supply Chain Management Professionals (CSCMP), defined SCM as "the planning and management of all activities involved in sourcing and procurement. conversion, and all logistics management activities. Importantly, it includes coordination and collaboration with channel partners, which can be suppliers, intermediaries, third-party service providers, and customers" (CSCMP, 2019, as cited in Kamble et al., 2019).

Historically, SCM literature and applications have focused predominantly on manufacturing sectors, with limited attention to agriculture, forestry, and fisheries (Routroy and Behera, 2017). However, a shift has been observed, with SCM increasingly being adopted in agricultural development. This transition aligns with Shukla and Jharkharia's (2013) argument that agriculture plays a pivotal role in the global economy as a key provider of raw materials for various industries (as cited in Routroy and Behera, 2017).

The integration of advanced information and communication technology (ICT) into agriculture has significantly improved the efficiency of food production systems, supporting food safety and sustainability amidst global population growth (Arena et al., 2019). Real-time data collection from supply chain stakeholders enables the identification of operational inefficiencies, and blockchain technology is emerging as a promising solution for ensuring secure, transparent, and tamper-proof data recording and sharing. Kamble et al. (2019) highlighted that traceability is among the primary drivers behind the adoption of blockchain technology in agricultural supply chains.

Routroy and Behera (2017) identify the five key actors in the agricultural supply chain (ASC) as the farmer, processor, distributor, retailer, and end-user. Major operational domains include traceability, logistics, and information technology, while significant challenges revolve around post-harvest losses, food safety, and product quality. These issues underscore the need for innovative technologies that enhance transparency and efficiency.

Rice, a staple crop with a notably intricate supply chain, exemplifies the need for such innovations.

Purwandoko et al. (2019) emphasized that the rice supply chain requires tools capable of monitoring processes from production to consumption (land-totable), and recommended the implementation of traceability systems to ensure food transparency. The Codex Alimentarius Commission (FAO, 2012) defines a traceability system as a "management system that is able to track the movement of food at certain stages of production, processing and distribution."

Blockchain technology, as defined by Casino et al. (2019) based on Nakamoto's (2008) seminal work, is "a structured and decentralized ledger initially developed to provide a practical solution to reaching consensus in an untrusted decentralized distributed environment" (p. 2730). Information is organized into chains of blocks, each storing a set of transactions at a point in time. These blocks specific are cryptographically linked to their predecessors, forming a secure, immutable chain. Blockchain combines cryptography, mathematics, economic modeling, and distributed algorithms, enabling a peerto-peer network to achieve consensus without central authority. Its immutability ensures that once data is recorded, it cannot be modified or tampered with, making it highly suitable for supply chain applications.

Building on these insights, this study proposes a definition of Blockchain for Rice Science - aligned with the research and development mandate of PhilRice - as a tamper-proof, distributed digital recording system. This system aims to protect intellectual assets and support various solutions designed to bolster the competitiveness of Philippine rice through PhilRice's initiatives in research, development, and extension.

Materials and Methods

The preliminary phase of this study focused on optimizing a blockchain-based solution designed to cost-effectively trace the seed-to-shelf journey of Philippine rice from breeder seed production to its eventual distribution in milled form through supermarkets and e-commerce platforms. As illustrated in Figure 1, this effort outlines the Blockchain for Rice Science journey spearheaded by PhilRice. The initial stages of value creation for the project, which commenced in 2020, are detailed below to show the progression of development and integration within the rice supply chain.

It can be noted that the bedrock of the project is about institutionalizing blockchain for rice science nationwide. The purpose is to gather as much data as possible to brace data-driven strategy and policy formulation toward rice self-sufficiency and



Figure 1. Blockchain for rice science value creation journey.

affordability while properly considering profitability among the rice value chain actors. Thus, credible data must come from all the players of the rice value chain, especially the farmers.

In developing the system, series of consultations were conducted with the PhilRice experts on plant breeding, agronomy, crop protection, seed production and seed health as well as information communication technology (ICT) practitioners. The constraints and challenges of PhilRice in terms of developing, managing and maintaining an IT-system was also considered on the investment decision. IT industry experts from global business operators were also consulted for the design of the Blockchain-based Traceability Management System.

In phasing the project, scoping was also done with individual farmers, regional offices of the Department of Agriculture and agriculture officers of the provincial, municipal and barangay local government units (LGUs). The observations from the scoping activity were discussed with the experts on roundtable format.

Based on the recommendations distilled from the insights of rice science experts, the traceability system can be tested initially for managing provenance data for the production of higher seed classes: (1) nucleus to breeder seeds; (2) breeder to foundation seeds; (3) foundation to registered seeds; and, (5) registered to certified seeds. The responsibility centers and production processes are illustrated, as follows:

It can be noted that the supply chain aspects are only up to the registered to certified seed production. The next steps or the continuation of the supply chain, which is rice production from certified seeds and rice retail will be included in the medium-term plan.

It can be noted as well from Figure 2 that the responsible centers and locations can vary per seed class, the PhilRice Central Experiment Station, which is located in the Science City of Muñoz, Nueva Ecija, Philippines undertakes the production from nucleus seeds up to registered seeds while the branch and satellite stations of PhilRice can undertake foundation to registered seed production activities. PhilRice operates nationwide through its branch and satellite stations that are located in major rice producing areas nationwide. The details of the actors in the preliminary digital supply chain design are provided in Table 1.

Limitations of the Study

A key limitation of this study is that PhilRice currently lacks the in-house capacity to independently develop a fully customized blockchain system. Building such a technology from the ground up particularly one that integrates mobile phone application access for farmers - requires substantial time, technical expertise, and resource investment. While mobile accessibility is critical for real-time data collection and user engagement at the grassroots level, the current dependency on external platforms or thirdparty developers may delay full-scale implementation and limit immediate responsiveness to evolving user needs.

In this initiative in (Table 1), the term firewall refers to a realm or specific work package, which is the smallest unit comprising a complete process comprising the product being sought. For example,



Figure 2. Illustration of higher seed class production processes and responsibility centers (firewalls).

Realm	Activity Description	Responsible Center(s)/ Process Owners	Number of Sites
Firewall 1	Nucleus to Breeder Seed Production	PhilRice Central Experiment Station	
Firewall 2	Breeder to Foundation Seed Production	PhilRice Central Experiment Station	1
Firewall 3	Distribution of Foundation Seeds to PhilRice Branch and Satellite Stations	Trucking from PhilRice Central, Branch and Satellite Stations; Logistics Services Providers	variable
Firewall 4	Foundation to Registered Seed Production	PhilRice Central, Branch and Satellite Stations	About 10 sites
Firewall 5	Registered to Certified Seed Production	Accredited Seed Growers	At least 14,000 nationwide*

*Based on the data provided by the National Seed Quality Control Services – Bureau of Plant Industries (NSQCS-BPI)

inside Firewall 1 is the entire process of producing inbred breeder rice seeds from nucleus seeds. The divisions, units, personnel and/or service providers who will do the activities within the said work package are responsible for the outputs/outcomes. Thus, all the activities to ensure quality such as the handing of nucleus seeds to the authorized recipient; proper land preparation; proper seeding; monitoring and appropriate management of pests and diseases; and proper harvesting, drying, seed cleaning, packaging and storage must be done with utmost diligence.

The next step was the development of the preliminary traceability system design. The development was outsourced from the International Business Machine (IBM) - a global IT industry leader. Briefly, the provenance data will start when the nucleus seeds of a specific variety is handed to the personnel concerned under the Plant Breeding and Biotechnology Division (PBBD). The day-to-day activities may vary depending on the seed class and rice variety as well as the weather condition. Based on the database of PBBD, there are 395 approved rice varieties - 247 are inbred rice while 149 are hybrid rice seed varieties. All the activities and phenomena will be recorded in the system so that factors responsible for consistent good performance or failure to pass the quality tests by the National Seed Quality Control Services - Bureau of Plant Industries (NSQCS-BPI) can be identified and improved. It is planned that the recording will be handled by the researchers or the process owners. The provenance data can also be the basis of performance incentives and sanctions.

The preliminary contents of the platform that was developed following the illustrated processes are provided in Figure 3.

Results and Discussion

The implementation of the initial Proof of Experience (PoX) phase demonstrated notable cost efficiencies resulting from the methodological approach adopted. Specifically, the design and deployment of the blockchain-based solution leveraged a lean, agile framework, which avoided the need for traditional resource-intensive processes. This included:

1. Elimination of IT hiring requirements, thereby reducing the demand on the Human Resource Management Division and associated hiring costs;



Figure 3. Overview of the components of the preliminary platform developed in partnership with the IBM.

- 2. No procurement of additional hardware or cloud services, as the system was built on existing digital infrastructure and modular technologies;
- **3.** Minimal utility and office overhead expenses, since the development and testing were conducted in virtual or hybrid environments; and
- **4. Pay-as-you-go resource use**, allowing the project to scale only according to actual, validated needs.

This methodological approach enabled rapid iteration, where researchers and PhilRice partners could immediately initiate learning cycles, collect data in real time, and iteratively test and refine interventions. Customizing the platform based on specific, on-the-ground requirements allowed for adaptive fine-tuning, reinforcing a feedback-driven development cycle that is particularly suited for dynamic agricultural ecosystems.

Despite the promising early results, the benefits observed in Phase I are still preliminary. Further validation through broader deployment and optimization phases is required to establish the full value and scalability of the system. The project's multi-phase strategy is outlined as follows:

- **PoX Phase I**: Focused on system optimization with a single account, serving as a controlled testbed to evaluate core functionality and validate initial design assumptions.
- **PoX Phase II**: Expansion to at least 100 user accounts across the country, specifically

targeting key seed production personnel stationed at Central and Branch Stations. Surplus capacity may be extended to supply chain partners such as the National Seed Quality Control Services (NSQCS). Seed Growers will also participate to test usability and interface intuitiveness.

- **PoX Phase III**: Scaling up to 1,000 accounts to include seed growers, farmers, and additional stakeholders across the rice value chain. This phase will evaluate performance under realistic operational conditions and broader user diversity.
- **Nationwide Roll-out**: Subject to successful outcomes in Phase III, the system will be formally adopted for national use in rice seed production.
- System Optimization Across PhilRice Programs: Expansion of blockchain-based tracking and analytics to support PhilRice's full range of research, development, and extension activities.
- **Inclusion of Other Crops**: Evaluation of the system's adaptability for other crops within rice-based farming systems, facilitating broader agricultural transformation.

This phased approach ensures that each level of implementation builds upon validated learning from the previous stage, thereby reinforcing reliability, scalability, and contextual fit within the Philippine rice sector.

Conclusion

This research represents a pioneering initiative by PhilRice to explore the potential of blockchain technology in enhancing transparency, traceability, and efficiency within the rice supply chain. As a preliminary endeavor, the blockchain-based artifact developed in this study serves as a proof of concept laying the foundation for broader digital transformation in Philippine rice science and agriculture.

Currently, PhilRice does not yet possess in-house blockchain development capabilities. The decision to outsource the initial development reflects the current absence of respectable expertise, which refers to the specialized technical proficiency and applied experience required to design, deploy, and manage decentralized systems at scale. Outsourcing allowed the institution to accelerate the learning process, avoid technical debt, and focus on context-specific customization without compromising operational timelines. Should the system undergo successful optimization through subsequent phases, PhilRice could serve as a model case study for other government agencies and development institutions. The lessons learned and methodologies established through this project have the potential to inform digital transformation across the broader domains of agriculture, forestry, and fisheries in the Philippines.

In the long term, PhilRice can contribute to the institutionalization of a blockchain regime defined as a structured, policy-aligned digital governance framework that leverages blockchain technology to ensure integrity, security, and traceability of data across agricultural value chains. Such a regime would not only support data-driven decision-making but also empower stakeholders at all levels through equitable access to trusted information.

Through continued investment in technical capacity and cross-sector collaboration, PhilRice has the opportunity to assert global leadership in blockchain applications for rice science transforming it from an early adopter into a central enabler of national digital agriculture strategies.

Literature Cited

- Alimentarius C (2006) Principles for traceability/product tracing as a tool within a food inspection and certification system. In C. A. Commission, Principles for Traceability (pp 1 - 4). Codex Alimentarius Commission
- Arena A, Bianchini A, Perazzo P, Vallati C, Dini G (2019) BRUSCHETTA: An IoT blockchain-based framework for certifying extra virgin olive oil supply chain. Proceedings of the 5th IEEE International Conference on SMART Computing (pp 173-179). Washington, United States: 5th IEEE International Conference on SMART Computing. Retrieved from https://ieeexplore.ieee.org/document/ 8784020. (accessed October 24, 2023)
- Casino F, Kanakaris V, Dasaklis TK, Moschuris S, Rachaniotis NP (2019) Modeling food supply chain traceability based on blockchain technology. Ifac-Papersonline *52*(13): 2728 - 2733
- CSSA [Crop Science Society of America](2024) About Crops: What is Crop Science?. Retrieved from Crop Science Society of America: www.crops.org (accessed January 30, 2024)
- FAO [Food and Agriculture Organization of the United Nations] (2012) Principles for Traceability/Product tracing as a tool within a food inspection and certification system. Codex Alimentarius Commission (CAC/GL 2006). In Food import and export inspection and certification systems (5th ed.). Retrieved from https://www.fao.org/4/i2483e/i2483e. pdf (accessed December 19, 2023)
- Kamble SS, Gunasekaran A, Sharma R (2019) Modeling the blockchain enabled traceability in agriculture supply chain. International Journal of Information Management 52: 101967

- Nakamoto S (2008) Bitcoin: A peer-to-peer electronic cash system. Retrieved from bitcoin.org: https://bitcoin.org/bitcoin.pdf (accessed January 30, 2024)
- Office of the President of the Philippines (1985) Executive Order No. 1061, s. 1985. Establishing the Philippine Rice Research Institute (PRRI). Official Gazette. Retrieved from https://www.officialgazette.gov.ph/1985/11/05/executiveorder-no-1061-s-1985/ (accessed November 17, 2023)
- Pelt RV, Jansen S, Baars D, Overbeek S (2021) Defining blockchain governance: a framework for analysis and comparison. Information Systems Management 38(1):21 -41
- **PSA** [Philippine Statistics Authority] (2021) Agricultural indicators system. output and productivity. Quezon City, the Philippines: Philippine Statistics Authority. Retrieved from https://psa.gov.ph/system/files/main-publication/1.1_ Output%2520%2526%2520Prod%25202016-2020%2520Publication_AIS_signed.pdf. (accessed October 24, 2023)

- Purwandoko PB, Seminar K, Sutrisno, Sugiyanta (2019) Development of a smart traceability system for the rice agroindustry supply chain in Indonesia. Information 10(10): 288
- **Routroy S, Behera A** (2017) Agriculture supply chain: a systematic review of literature and implications for future research. Journal of Agribusiness in Developing and Emerging Ecnomics 7(3): 275 302
- Shukla M, Jharkharia S (2013) Agri-fresh produce supply chain management: a state-of-the-art literature review. International Journal of Operations & Production Management 33(2): 114 - 158
- The Investopedia Team (2024) Investopedia. Retrieved from Investopedia: www.investopedia.com
- Tripathi G, Ahad MA, Casalino G (2023) A comprehensive review of blockchain technology: Underlying principles and historical background with future challenges. Decision Analytics Journal 9:100344

SPECIAL REVIEW

APPLICATIONS OF PARTICIPATORY DEVELOPMENT COMMUNICATION IN THE AGRICULTURE SECTOR FROM 2013 TO 2023

VENUS B. TAJON¹, JAIME A. MANALO IV^{1,2*}, JAY C. SANTOS¹, CHRYLS AVEGEIL N. VALLEJO¹, MARIFE DT PASCUAL¹, REGIDOR G. GABOY¹, PARSONS N. HAIL¹

> ¹Central Luzon State University, Science City of Muñoz, Nueva Ecija, Philippines ²Philippine Rice Reseazrch Institute, Science City of Muñoz, Nueva Ecija, Philippines *Corresponding Author:jaime.manalo@clsu2.edu.ph

Abstract

This review examines the applications, benefits, and challenges of Participatory Development Communication (PDC) in agricultural development from 2013 to 2023. PDC represents a paradigm shift from conventional topdown communication approaches to collaborative, inclusive, and horizontal dialogues. It empowers farmers, extension workers, and development stakeholders to actively participate in planning, decision-making, and implementation processes. This participatory approach fosters the co-creation of solutions that are contextspecific, culturally appropriate, and sustainable. Using a systematic review methodology, 12 peer-reviewed studies were analyzed based on defined inclusion criteria. The studies span diverse geographical regions and agricultural contexts, highlighting the transformative potential of PDC. Key themes identified include enhanced local ownership of development initiatives, improved adoption of agricultural technologies, strengthened knowledge exchange, and increased resilience to environmental and economic shocks. The review also underscores the role of PDC in integrating indigenous knowledge with scientific practices, leading to more relevant and sustainable agricultural outcomes. However, despite its documented benefits, the review identifies several persistent barriers to the effective implementation of PDC. These include rigid institutional communication structures, weak integration with technical disciplines, limited exposure to participatory tools, and sociopolitical constraints such as power imbalances and stakeholder distrust. To maximize the effectiveness and sustainability of PDC, the review proposes several recommendations: institutionalizing participatory mechanisms in agricultural policies and programs; integrating scientific, technical, and participatory approaches; leveraging digital tools for broader engagement; adopting context-specific and flexible communication frameworks; fostering cross-learning between local and global innovations; and embedding PDC in climate change adaptation strategies. In conclusion, PDC holds significant promise as a catalyst for inclusive, equitable, and sustainable agricultural transformation. Its success depends on its integration within broader development systems and policies, and its ability to adapt to the complexities of contemporary agricultural and environmental challenges.

Keywords: participatory development communication, agricultural development, farmer participation, indigenous knowledge, sustainable agriculture, climate change adaptation

Introduction

Participatory Development Communication (PDC) has emerged as a vitally important approach in agricultural development, especially in addressing complex challenges. PDC shifts the paradigm from traditional top-down models to collaborative and horizontal dialogues by engaging farmers, extension workers, and stakeholders in decision-making processes (Waisbord, 2020). This approach empowers farmers to articulate their needs, share indigenous knowledge, and co-create solutions tailored to their contexts, unique ensuring that agricultural interventions are relevant and sustainable.

The importance of participatory communication in agriculture cannot be overstated. Globally, it has proven effective in various contexts by promoting local ownership, enhancing the adoption of new technologies, and building resilience against environmental and economic shocks. For instance, studies have shown that Farmer Field Schools in East Africa significantly improved farmers' knowledge and adoption of sustainable practices through participatory methods (Friis-Hansen and Duveskog, 2012). Similarly, research in Indonesia highlights the critical role of Communication for Development (C4D) training for extension workers in addressing the complexities of modern agricultural development (Agunga and Putra, 2015). In Kenya, involving local communities in decision-making has led to more effective pest management strategies, while in Ethiopia, a shift from top-down to inclusive communication models has been shown to enhance agricultural production and improve rural livelihoods (Gebeyehu and Jira, 2023). In Ethiopia, shifting from top-down to participatory communication models in agricultural interventions has been shown to enhance

agricultural production and improve rural livelihoods (Gebeyehu and Jira, 2023).

This article reviews applications of participatory communication in the agriculture sector from 2013 to 2023. By doing this review, the overarching aim is to highlight best-fit practices, challenges, and opportunities for optimizing participatory communication to advance sustainable agricultural development globally.

Unlike previous studies focusing on localized case studies or specific participatory approaches, this review provides a comprehensive, cross-regional analysis of participatory communication in agriculture over the past decade. By synthesizing diverse applications, identifying emerging trends, and evaluating the scalability of participatory strategies, it addresses gaps in understanding how PDC can be systematically integrated into agricultural policies and programs. This broader perspective offers valuable insights for policymakers, researchers. and practitioners seeking to enhance the effectiveness of participatory communication in fostering sustainable agricultural development.

Methodology

Literature Search and Selection

The review was conducted by systematically searching for literature from Google Scholar, PubMed, and JSTOR. The search process involved the following: First, each database was searched using "Participatory predefined keywords, including Development Communication," "agricultural development," "farmer participation," "communication strategies," and "rural livelihoods," aiming to capture a wide range of studies related to participatory communication in agricultural contexts. Second, the initial search results were screened for relevance by examining the titles and abstracts of each study. Studies that did not meet the inclusion criteria were excluded. From an initial pool of 80 articles, 32 articles that met the inclusion criteria were further screened; hence, the number was reduced to 20. The 20 articles were then downloaded for thorough review. After thoroughly reviewing the 20 articles, the number was further reduced to 12. Studies were excluded if they focused on sectors other than agricultural development, only briefly mentioned participatory communication, or lacked detailed analysis. Weak methodologies such as unclear research designs also led to exclusion. Similar studies were streamlined, retaining only the most comprehensive ones, while some were removed to maintain regional diversity. Studies without full-text access or from non-peer-reviewed sources were excluded, along with those with poor language quality

or unclear presentation. Relevant data from these selected studies were systematically extracted and organized, including information on the study's objectives, methodologies, findings, and implications related to participatory communication in agricultural development.

Studies were included in the review if they met the following criteria: published between 2013 and 2023 to ensure coverage of the most recent and relevant literature, written in English to ensure consistency and accessibility, centered participatory on communication in agricultural development, and examined the role, impact, and effectiveness of participatory communication strategies and approaches. Additionally, the studies had to be published in peer-reviewed journals, academic reports, case studies, or conference proceedings to ensure credibility and reliability.

Analysis

The extracted data were then analyzed to identify common themes, best-fit practices, challenges, and opportunities related to participatory communication in agricultural development. This analysis aimed to synthesize the findings from the selected studies to provide a comprehensive understanding of the impact and effectiveness of participatory communication in promoting sustainable rural agricultural practices. By following this systematic approach, the review aimed to provide a thorough and credible assessment of the current state of participatory communication in agricultural development, highlighting key insights and recommendations for future practice and research. Adhering to these criteria and following a rigorous search process, this review aimed to provide a comprehensive and reliable synthesis of current knowledge on participatory communication in agricultural development.

Results and Discussion

Key Themes

The analysis of the literature on Participatory Development Communication (PDC) in agricultural development has revealed several key themes and findings that are pivotal to understanding its impact and effectiveness. The reviewed studies highlight the transformative potential of PDC in fostering inclusive and responsive agricultural frameworks. Key themes include the role of PDC in promoting local ownership, enhancing technology adoption through participatory building resilience methods. and against environmental and economic shocks. Furthermore, the findings underscore the importance of collaborative and horizontal dialogues in empowering farmers, incorporating indigenous knowledge, and cocreating tailored solutions. By examining these themes, the review provides a comprehensive overview of the current state of PDC in agricultural development. It offers insights into best-fit practices, challenges, and future research and implementation opportunities.

Transforming Agricultural Communication to a more Collaborative, Farmer-centered Approach

Challenges in Ethiopia's Agricultural Sector. The study by Gebeyehu and Jira (2023) highlights the challenges of а predominantly top-down communication approach in Ethiopia's agricultural sector. which restricts interaction between development agents and farmers. This results in policies and interventions that often fail to address farmers' actual needs and experiences. Gebevehu and Jira advocate for a shift towards more inclusive communication strategies, likely involving workshops or focus groups to better engage farmers. The key strength of this approach lies in its focus on integrating farmers' indigenous knowledge into policy-making, which promotes a sense of ownership and ensures that policies are tailored to the farmers' specific needs. However, the existing top-down model limits effective participation, rendering the PDC approach less impactful without systemic changes. Although the study emphasizes the need for PDC, it does not integrate technical approaches from agricultural science, suggesting that PDC would be more effective if combined with these technical strategies. While the study underscores the importance of PDC, it lacks integration with technical agricultural approaches, suggesting that combining PDC with these strategies could enhance its effectiveness.

Indonesian *Opportunities* in Agricultural Extension. Agunga and Putra (2015) underscore the critical need for training Indonesian agricultural Communication extension workers in for Development (C4D). They highlight the importance of equipping agricultural extension workers with a deep understanding of development theory, policy, and practice to enhance their ability to facilitate dialogue and collaboration among stakeholders. The study emphasizes the use of training programs that integrate Participatory Development Communication (PDC) with C4D. This approach involves workshops and training sessions aimed at improving extension workers' skills in participatory methods and communication. The key strength of this approach is its ability to enhance dialogue and collaborative learning, making extension workers more effective in engaging with farmers and improving agricultural initiatives. However, without adequate training, participatory communication efforts can be

ineffective, especially in diverse agricultural contexts. PDC was not used in isolation but was combined with technical training, illustrating a blended approach that merged participatory methods with expert knowledge to enhance the effectiveness of agricultural extension services.

Integrating Socio-cultural Insights and Trustbuilding in Agricultural Practices

Development of Sheep Farming in Malang: The study by Putri Yulinarsari et al. (2021) examines the development of sheep farming in Malang, Indonesia, focusing on the socio-cultural and communication elements influencing the industry. Participatory Development Communication (PDC) in this context involved methods such as interviews, workshops, and peer knowledge sharing. These methods directly engage farmers in the communication process. facilitating the exchange of knowledge and experiences, which leads to improved farming practices and business outcomes. One of the key strengths of this approach is its effectiveness in enhancing business development and farming practices through active farmer involvement. However, a notable weakness is its limited impact on the sociocultural dimensions of the community. This suggests that while PDC can advance business and farming practices, it requires integration with social science approaches to address and incorporate sociocultural aspects more comprehensively. Although PDC was effective in promoting business development, its impact would be significantly enhanced by incorporating socio-cultural or anthropological studies. The statement implies that in the context of the study, the implementation of PDC lacked sufficient integration of sociocultural or anthropological insights. This suggests that the challenge lies not in the theory of PDC but in how it was executed, indicating that the researchers or practitioners may not have fully incorporated sociocultural dimensions during implementation. To clarify, while PDC as a framework is inherently capable of addressing socio-cultural factors, the study indicates that its effectiveness could have been enhanced through a more comprehensive integration of these insights.

Barriers to Participatory Extension in Egypt. McDonough et al. (2015) investigate the barriers to implementing participatory extension services in Egypt, emphasizing the need for trust and support among farmers, extension workers, and other stakeholders. The study primarily uses extension services with participatory methods, including stakeholder dialogue and workshops. One of the key strengths of this approach is its capacity to build trust and foster strong relationships through open communication, thereby enhancing participatory practices. However, the study highlights a significant weakness: the lack of trust and effective communication among stakeholders impedes full engagement and limits the effectiveness of the participatory process. While PDC is used as a standalone approach, the study suggests that integrating additional techniques, such as social network strengthening or leadership training, would greatly benefit the approach. These additional methods could help overcome the barriers of trust and communication, leading to more effective stakeholder collaboration and enhanced participatory extension practices.

Participatory Approaches in Agricultural Modeling and Planning

Shrimp Aquaculture in Vietnam. Joffre et al. (2015) illustrate the integration of participatory approaches with agent-based modeling for planning shrimp aquaculture in Vietnam. The study employs role-playing games (RPGs) and local consultations as its primary Participatory Development Communication (PDC) methods. RPGs offer an innovative approach for stakeholders to simulate various scenarios and assess potential risks in realtime, thereby enhancing the relevance and effectiveness of the model by incorporating local knowledge and perspectives. This participatory method facilitates active involvement and learning among farmers and decision-makers, improving decision-making processes and planning outcomes. However, the approach is resource-intensive, requiring significant facilitation and expertise in both communication and technical modeling. PDC incorporates agent-based modeling as a key component, blending social and technical sciences. This blended approach merges social and technical sciences, optimizing decision-making and ensuring that the management strategies are more accurate and tailored to local conditions.

The study by Joffre et al. (2015) describes a participatory approach that integrates role-playing games (RPGs) and local consultations, both of which established Participatory are Development Communication (PDC) methods. RPGs engage stakeholders by allowing them to simulate different scenarios, assess risks, and explore outcomes in a controlled yet interactive environment. This facilitates mutual learning and collective decision-making, which are core principles of PDC. Local consultations further support this by incorporating local knowledge and ensuring that stakeholders' voices are reflected in the decision-making process. The unique aspect of this study is the integration of PDC methods with agent-based modeling, a technical tool used to simulate complex systems and predict outcomes based on stakeholder behavior and environmental

factors. While agent-based modeling itself is not a PDC method, combining it with RPGs and consultations creates a hybrid participatory approach. This blended method strengthens the relevance and accuracy of management strategies by merging social insights (through PDC) with technical modeling. Therefore, the RPGs and local consultations are clearly PDC methods, but the agent-based modeling serves as a complementary analytical tool rather than a separate PDC method. This underscores how PDC can be effectively integrated with technical approaches to improve planning and decision-making in complex agricultural systems.

Management Strategy Evaluation (MSE) for Fisheries Management. Goethel et al. (2019) examine the role of stakeholder participation in Management Strategy Evaluation (MSE) for fisheries management, utilizing workshops and consultations to involve various stakeholders in defining roles and responsibilities. This approach enhances transparency and fosters stakeholder buy-in, ensuring that management strategies are informed by diverse perspectives and needs. A key strength of the PDC method is its ability to integrate various viewpoints, which leads to more balanced and comprehensive management strategies. However, the process of reconciling diverse stakeholder perspectives can be challenging, and issues such as miscommunication or lack of clarity may impede progress. The PDC approach is not used in isolation but is combined with Management Strategy Evaluation (MSE), blending participatory methods with scientific analysis. This integrated approach creates a more effective framework for fisheries management by aligning participatory insights with technical expertise.

Enhancing Stakeholder Engagement

Stakeholder Workshops in the Philippines. Cleland and San Jose (2018) investigate the use of stakeholder workshops in the Philippines within participatory conservation-for-development projects. Their approach includes traditional face-to-face interactions and modern digital tools. Face-to-face interactions are a key strength as they build trust, facilitate dialogue, and ensure that all voices are heard. However, the integration of digital platforms can create barriers, such as power imbalances that favor expert opinions and tech-savvy participants over those of less experienced or local participants. This can overshadow valuable local knowledge, reduce the workshop's inclusivity, and introduce jargon or concepts that may hinder effective communication. These challenges can lead to the exclusion of some voices and reinforce existing inequities. The PDC method used involves stakeholder workshops that blend traditional communication methods with digital tools. While this combination can enhance

engagement and inclusivity, it requires careful balancing to prevent the exclusion of non-technical participants (may include farmers, community members, or local stakeholders who rely more on practical experience and indigenous knowledge rather than formal technical training) and to ensure that the workshop remains truly inclusive. PDC in this context is not a standalone approach; it is combined with technological tools, indicating that while participatory methods are employed, they need to be carefully integrated with technology to address and mitigate potential barriers to full participation. In this context, PDC incorporates technological tools, meaning that while participatory methods are employed, careful integration of these tools is essential to prevent or reduce potential barriers to full participation.

Co-designing Management Recommendations in the Baltic Sea. Ehrlich et al. (2023) examine the codesign process for developing management recommendations for northern pike in the Baltic Sea. The study utilizes regular "roundtable" workshops as the primary PDC method, engaging a diverse range of stakeholders, including local community members, environmentalists. and policymakers. These workshops provide a structured platform for open dialogue and collective problem-solving, facilitating consensus-building and improving decision-making by integrating varied perspectives. The key strength of this PDC approach lies in its ability to reconcile differing viewpoints and build consensus among stakeholders, ensuring that management recommendations are well-informed and more likely to be accepted and implemented. However, a notable weakness is that continuous engagement required for these workshops can be resource-intensive, and conflicting interests among stakeholders may impede progress. In this case, PDC is not used in isolation but is integrated with environmental science. The combination of participatory dialogue with scientific analysis supports a more comprehensive approach to addressing complex environmental management challenges. In this case, PDC is integrated with environmental science, combining participatory dialogue with scientific analysis to create a more comprehensive approach to addressing complex environmental management challenges.

Incorporating Local Knowledge

Integration of Farmers' Local Soil Knowledge in Madagascar. Ravonjiarison et al. (2023) introduce a conceptual framework to integrate farmers' indigenous soil knowledge with scientific research in Madagascar. Local soil classifications and indicators used by farmers align with scientific findings, demonstrating their effectiveness. Recognizing and incorporating local expertise enhances agricultural practices in ways that are scientifically valid and contextually relevant. The study highlights that farmers in the Alaotra Lake Region have developed detailed local soil classifications, identifying twelve soil types in the south and nine in the north, reflecting their extensive indigenous knowledge. It also shows that farmers use 14 local indicators to assess soil fertility, which aligns with scientifically validated methods, demonstrating that local practices are consistent with broader agricultural standards. These local classifications not only complement existing scientific knowledge but also provide additional insights beyond the French Soil Classification (CPCS), suggesting that integrating indigenous knowledge can enhance scientific soil management. The study confirms that these local soil fertility indicators are effective and comparable to those used globally, highlighting the scientific robustness of indigenous knowledge. Additionally, the article introduces a conceptual framework for integrating local knowledge with scientific research, offering a structured approach to incorporating indigenous expertise into agricultural development. The approach bridges the gap between traditional knowledge and modern science, fostering a more holistic understanding of soil management and improving agricultural practices.

Participatory Approaches in Soil Research and Management. Wadoux and McBratney (2023) synthesize various participatory approaches in soil research and management, focusing on involving nonexperts, such as local farmers and community members. in data collection and analysis. Participatory projects significantly contribute to scientific progress by incorporating diverse perspectives and local expertise, ensuring research outcomes are more relevant and applicable to realworld conditions. These projects often lead to positive social outcomes, such as increased community engagement and empowerment. However, the longterm impacts of these participatory approaches are often underreported, limiting the understanding of their sustained effects on agricultural development and soil management. The research highlights the benefits of integrating local knowledge with scientific inquiry and the need for more comprehensive evaluations of long-term impacts.

Addressing Climate Change and Disaster Risks

Evaluation of climate-induced hazards risk for coastal Bangladesh. A participatory approach-based assessment, Geomatics, Natural Hazards and Risk: Faisal et al. (2021) investigate the impacts of climateinduced hazards in coastal regions of Bangladesh using a participatory approach that combines workshops and field interviews. Workshops are organized to facilitate collaborative discussions among community members, stakeholders, and experts, fostering the sharing of insights and collective assessment of climate-related risks. Field interviews further support this by collecting firsthand information from residents about the effects of climate hazards and their specific needs and experiences. The key strengths of this approach lie in its comprehensive engagement, ensuring that disaster risk reduction strategies are informed by local knowledge and real-world conditions, which leads to more accurate and contextually relevant solutions. By tailoring strategies to the specific needs of the affected areas, the participatory process enhances contextual relevance. However, the approach has weaknesses, such as dependence on external resources for implementing some recommended strategies, like infrastructure improvements, which may extend beyond the capabilities of PDC alone. Additionally, without incorporating additional scientific or technical expertise, the participatory approach may not fully address the technical aspects of disaster risk reduction. Thus, while PDC effectively engages local knowledge and develops relevant strategies, it is used alongside technical disaster risk reduction methods to ensure a comprehensive approach that addresses broader climate adaptation needs. Thus, while PDC effectively engages local knowledge and develops relevant strategies, integrating it with technical disaster risk reduction methods strengthens the approach, ensuring that both local insights and scientific expertise are combined to address broader climate adaptation needs.

Faisal et al. (2021) investigate the impacts of climate-induced hazards in coastal Bangladesh using a participatory approach that combines workshops and field interviews. The workshops facilitate collaborative discussions among community members, stakeholders, and experts, promoting the sharing of insights and collective assessment of climate-related risks. Field interviews complement this by collecting firsthand information from residents about the effects of climate hazards and their specific needs and experiences.

A key strength of this approach is its comprehensive engagement, ensuring that disaster risk reduction strategies are informed by local knowledge and real-world conditions, leading to more accurate and contextually relevant solutions. By tailoring strategies to the specific needs of the affected areas, the participatory process enhances the relevance and effectiveness of interventions.

Conclusion and Recommendations

This review affirms the transformative potential of Participatory Development Communication (PDC) in fostering inclusive, responsive, and sustainable agricultural development. PDC enhances local ownership, integrates indigenous knowledge, promotes collaborative decision-making, and supports technology adoption tailored to farmers' unique contexts. Across various studies, its effectiveness is evident in improving agricultural outcomes and building resilience against environmental and economic shocks.

However, several constraints hinder its full realization. Chief among these are entrenched topdown communication structures, limited exposure to innovative participatory tools, inadequate integration with technical disciplines, and sociopolitical challenges such as stakeholder distrust. These limitations underscore the need to reposition PDC not as an isolated approach, but as part of an integrated development strategy that includes scientific and technological expertise.

To maximize its impact and sustainability, PDC must evolve into a dynamic and adaptive framework one that is responsive to the complexities of contemporary agricultural systems. Its success hinges on its ability to embrace co-innovation, leverage digital technologies, and foster knowledge co-creation through inclusive dialogue.

Based on the findings, the following recommendations are proposed:

- Institutionalize Participatory Communication in agricultural policies and programs Policymakers should embed participatory mechanisms in every stage of program development - from planning to implementation and evaluation. This includes establishing platforms such as farmer consultation councils and participatory monitoring systems to ensure continuous and meaningful stakeholder engagement.
- Integrate scientific, technical, and participatory approaches Development strategies should encourage collaboration among farmers,
- *extension workers*, researchers, and communication specialists. *Jointly designed interventions* that combine expert knowledge with local practices will enhance relevance, adoption, and effectiveness.
- Leverage digital tools for enhanced farmer engagement Stakeholders should invest in user-friendly digital platforms such as mobile apps, SMS services, community radio with feedback channels, and online forums, that facilitate real-time knowledge sharing and farmer participation. These tools are particularly vital in reaching remote areas and

enabling rapid response to emerging challenges.

- Adopt context-specific, flexible frameworks for implementation Rigid, one-size-fits-all strategies must give way to adaptable frameworks that respect local cultures, resources, and ecological conditions. Participatory Action Research (PAR), Communication for Social Change (CFSC), and Agricultural Knowledge and Information Systems (AKIS) should be localized to meet the distinct needs of each farming community.
- Foster cross-learning between local innovations and global best practices Encouraging farmer-to-farmer exchanges, regional learning hubs, and open-access knowledge repositories can facilitate the dissemination of effective practices. This cross-learning can accelerate innovation and help communities adapt to globalization, climate change, and shifting market demands.
- *Embed PDC in climate change adaptation strategies* as climate-related shocks become more frequent, participatory approaches must be central to designing and implementing climate-smart agricultural solutions. This ensures that farmers not only adapt more effectively but also shape the innovations that safeguard their livelihoods.

In conclusion, Participatory Development Communication holds significant promise as a catalyst for equitable and sustainable agricultural transformation. Its strategic integration with other disciplines, technologies, and policy structures will be essential in addressing the complex challenges of food security, environmental change, and inclusive rural development in the years ahead.

Literature Cited

- Agunga R, Putra RARS (2015) Training needs of indonesian agricultural extension workers for the 21st century: A recommendation based on a field study. Asian Journal of Agricultural and Development *12*(2): 45 - 58
- Cleland D, San Jose RO (2018) Rehearsing inclusive participation through fishery stakeholder workshops in the Philippines. Conservation and Society 16(3): 351 - 362

- Ehrlich E, Bath AJ, Niessne D, Koemle D, Cyrus EM, Arlinghaus R (2023) Co-designing management recommendations with stakeholders: a case study about northern pike (*Esox lucius*) in the southern Baltic Sea. Fisheries Research 263:106687
- Faisal M, Saha MK, Sattar MA, Biswas AM, Hossain MA (2021) Evaluation of climate induced hazards risk for coastal Bangladesh: a participatory approach-based assessment. Geomatics, Natural Hazards and Risk 12(1): 2477 - 2499
- Friis-Hansen E, Duveskog D (2012) The empowerment route to sustainable livelihoods: a case study of farmer field schools in East Africa. World Development 40(2):414 - 27
- Gebeyehu HZ, Jira YS (2023) Exploring participatory communication implemented to improve the livelihood of rural Ethiopia. Humanities and Social Sciences Communications. Retrieved from https://www.nature.com/ articles/s41599-023-02286-6 (accessed October 6, 2024)
- Goethel DR, Lucey SM, Berger AM, Gaichas SK, Karp MA, Lynch PD, Walter III JF, Deroba JJ, Miller S, Wilberg MJ (2019) Closing the feedback loop: on stakeholder participation in management strategy evaluation. Canadian Journal of Fisheries and Aquatic Sciences 76(10): 1895 -913
- Joffre OM, Bosma RH, Ligtenberg A, Tri VP, Ha TT, Bregt AK (2015) Combining participatory approaches and an agentbased model for better planning shrimp aquaculture. Agricultural Systems *141*: 149 - 159
- McDonough C, Nuberg IK, Pitchford WS (2015) Barriers to participatory extension in Egypt: agricultural workers' perspectives. The Journal of Agricultural Education and Extension 21(2): 159 - 176
- Putri Yulinarsari A, Azizah S, Suwandi UWN (2021) The development of sheep farming business with participatory development communication in Malang. International Journal of Scientific Advances 2(4): 10
- Ravonjiarison N, Albrecht A, Penot E, Razafimbelo TM (2023) A conceptual framework for assessing farmers' soil knowledge: lessons from the lake alaotra region in Madagascar. Biotechnol. Agron. Soc. Environ. 27(1): 42 -60
- Wadoux AM, McBratney AB (2023) Participatory approaches for soil research and management: A literature-based synthesis. Soil Security 10: 100085
- Waisbord S (2020) Family tree of theories, methodologies, and strategies in development communication (pp 93 -132). Handbook of communication for development and social change. Retrieved from https://doi.org/10.1007/978-981-10-7035-8 56-1 (accessed October 6, 2024)

CALL FOR PUBLICATION Rice-based Biosystems Journal

SCOPE

The Rice-based Biosystems Journal encourages publication of original research and review articles that have impact on applied and integrated rice and rice-based cropping systems in a particular ecosystem. The Journal provides information on rice-based researches on soil and crop management, crop protection, crop improvement, grain quality, farm machinery, resource use efficiency, plant biology, nutraceuticals, food value-adding systems, biofertilizers, biopesticides, biomaterials, and system analysis and simulation. It also covers the economics, social, and communication systems that influence the landscape of rice and rice-based cropping systems.

AUTHOR'S GUIDELINES

1. Submission and Acceptance of Manuscripts

Manuscripts are submitted to Rice-based Biosystems Journal: rbbj.philrice@gmail.com. Manuscripts should be formatted as described in the Rice-based Biosystems Journal Author Guidelines and follow the PhilRice style guide. When preparing your file, please use Times New Roman as font type, and 12 as font size for the text. Please do not use Japanese or other Asian fonts. Do not use automated or manual hyphenation. With your submission, you will have to complete, sign, and send the Copyright Transfer Agreement. Authors may provide names of potential reviewers for their manuscript. Authors must inform the editorial assistant of any possible conflict of interest capable of influencing their judgement, and if necessary, a disclaimer will be included. Revised manuscripts must be submitted two weeks after the authors are notified of conditional acceptance pending satisfactory revision. Authors resubmitting manuscripts should follow the same procedures as for submission of new manuscripts. If accepted, papers become the copyright of the journal. Photos and tables must be high resolution scans (JPEG at 300 dpi).

2. Requirements for Manuscripts

2.1. Language

The language of publication is English.

2.2. Format

The first page should contain the name and address of the institute where the work has been done, the title of the paper, name(s) and initial(s) of the author(s), the e-mail address of the corresponding author, and the number of figures and tables.

The main text shall be preceded by an abstract, which is always in English and contains the background for the research undertaken, reference to the material and methods used, as well as main results and conclusions. It should not exceed 220 words. Up to seven 'keywords' should be added. A short version of the title (running title) should also be given.

The main text should be divided into the following sections: Introduction, Materials and Methods, Results and Discussion, Conclusion, Recommendation, Acknowledgment, and Literature Cited. Facts explained by tables or figures need no lengthy explanation in the text. Numerical material should be submitted only after statistical processing.

The manuscript comprises a printout of the text and a list of all figures and tables with their captions and titles on a separate piece of paper. In anticipation of the online edition, we ask that you convey the essential information within the first 60 characters of the captions. Each figure, table, and bibliographic entry must have a reference in the text. The preferred position for the insertion of figures and tables should be marked on the margin of the text of the manuscript. Any corrections requested by the reviewer should already be integrated into the file. The text should be prepared using standard software (Microsoft Word). Please do not include footnotes.

2.3. Length

The manuscript should be typed double spaced with a 4 cm left margin. Manuscripts, including figures and tables, should not exceed 25 printed pages. The publication of shorter papers may be given priority.

2.4. Units, Abbreviations, and Nomenclature

All units and measures must conform to the international standard-system (SI). Botanical genus and species names should be set in italics.

2.5. Illustrations and Tables

The number of tables and figures should be kept to the minimum necessary, and have a maximum of 13 cm in height and 17 cm in width. All figures should include reproducible copies marked with the author's name, short title, and figure number. Figures submitted as electronic file should be saved in PNG instead of JPEG for better quality. Powerpoint and Word graphics are unsuitable for reproduction.

Submit high-contrast photographic materials suitable for reproduction. Images should be of high quality with respect to detail, contrast, and fineness of grain to withstand the inevitable loss of contrast and detail during the printing process.

Scanned figures (usually in JPEG format) should have a resolution of 300 dpi (halftone) or 600 to 1200 dpi (line drawings) in relation to the reproduction size. You may submit figures in color or black and white. Graphs with an x and y axis should not be enclosed in frames; only 2-dimensional representations. Place labels and units.

Captions for the figures should give a precise description of the content and should not be repeated within the figure. Tables should be created with the table function of a word processing program. Spreadsheets are not acceptable.

2.6. References

The literature cited should be arranged alphabetically and contain: the author's surname, first name and middle initial, year of publication, title of paper, name of journal, volume number, and first and last page number of the publication.

Bibliographic references to books or other established publications should contain: author's surname, first name and middle initial, year of publication, and edition, publishing house and place of publication. The name of the author and the date of publication should be included within the text. If more than one publication of the same author appeared in one year, these should be marked by small letters after the year, e.g., 2015a; 2015b. References to publications by more than two authors should be cited as follows: Luna et al. (2015) or (Luna et al., 2015).

3. Copyright

If your paper is accepted, the author identified as the formal corresponding author for the paper will receive an email.

4. Proof Corrections and Offprints

The corresponding author will receive an e-mail with the laid out publication. A working e-mail address must therefore be provided for the corresponding author. Further instructions will be sent with the proof. We will charge for excessive changes made by the author in the proofs, excluding typesetting errors.

5. Submission and Acceptance of Research Notes

A research note is a short discussion on key research findings and advances on a particular theory, study, or methodology that does not sum up to a full research article. The format and guidelines of a research note resembles that of a full-length manuscript except for the number of words, figures and/or tables. A 3000 to 4000-word paper with an abstract and a maximum of 2 figures and/or 2 tables may be submitted as a research note.

6. Submission of Invited Papers

The Editorial Team can invite a member of the Advisory Board and Editorial Board of the Rice-based Biosystems Journal or an expert to submit a paper in line with the theme of the volume to be published. Invited papers may be in the form of a full paper, research note or a review article. A review article gives information on a particular field of study, recent major advances and discoveries, significant gap in the research, current debates, and ideas or recommendations for future advances.

At least one expert on the subject matter will review the invited paper. Instructions for submitting a full paper and research note are in numbers 1-5 of the author guidelines.

6.1 Format

The Abstract consists of 220 words or less that summarizes the topic of the review. The current challenges and perspective on the topic are addressed, with significant conclusion and recommendations.

The Introduction states the purpose of the review. It presents a short background of the nature of the problem and its aspects of being resolved. The limitations of current solution or studies are included.

The Body presents the current studies and major advances or discoveries and impact on the present situation of the problem. Evaluation of studies such as applicability and availability of the methods used to certain areas and situation or statistical significance are elaborated.

The Conclusion summarizes the overall or major impacts and main points of the current studies. Recommendations for future advances of the research on the subject matter are presented.

The Literature Cited follows the instructions in number 2.6 of the author guidelines.

EDITORIAL POLICY

Authors should:

- designate a corresponding author who will be responsible in coordinating issues related to submission and review, including ensuring that all authorship disagreements are resolved appropriately;
- submit original work that has been honestly carried out according to rigorous experimental standards;
- give credit to the work and ideas of others that led to their work or influenced it in some way;
- declare all sources of research funding and support;
- submit manuscripts that are within the scope of the journal by ensuring that they abide by the journal's policies and follow its presentation and submission requirements;
- explain in a cover letter if there are special circumstances when the manuscript deviates in any way from a journal's requirements or if anything is missing and ensure that the manuscripts do not contain plagiarized material or anything that is libelous, defamatory, indecent, obscene or otherwise unlawful, and that nothing infringes the rights of others;
- ensure they have permission from others to cite personal communications and that the extent, content, and context have been approved;
- provide details of related manuscripts they have submitted or have in press elsewhere; and
- check the references cited to ensure that the details are correct.

Authors should not:

- submit the same or a very similar manuscript to more than one journal at the same time, present their work, or use language, in a way that detracts from the work or ideas of others;
- be influenced by the sponsors of their research regarding the analysis and interpretation of their data or in their decision on what to, or not to publish and when to publish;
- divide up the papers inappropriately into smaller ones in an attempt to increase their list of publications;
- be involved in 'ghost' or 'gift' authorship;
- use information privately obtained without direct permission from the individuals from whom it was obtained;
- make exaggerated claims about the novelty or significance of their findings;
- misrepresent or inappropriately enhance their results by any means;
- make significant changes to their manuscript after acceptance without the approval of the editor or journal editorial office; and
- submit a manuscript that has been rejected by one journal to another journal without considering the reviewers' comments, revising the manuscript, and correcting presentational errors.

Reviewers should:

- Maintain the confidentiality of all manuscript content throughout the review process Provide an unbiased evaluation of the scientific papers; and
- Contribute to improving the quality of submitted manuscripts by reviewing them carefully, thoughtfully, and objectively Submit their evaluations on or before the stated deadline. If a delay is unavoidable, they must inform the managing editor promptly.

Reviewers should not:

- Share or copy information from the manuscripts for any purpose;
- Accept an invitation to review a manuscript if the intention is only to gain insights without submitting a completed evaluation form Use any information from the evaluated manuscript for their own research if the manuscript is still under review; and
- Suggest that authors cite their own research papers unless it is genuinely relevant to the work being reviewed.

Plagiarism Policy

• The journal not only promotes and advocates for the original works of its authors but also supports the protection of intellectual property rights. The use of plagiarized content in papers is a violation of research and originality standards. All authors submitting manuscripts to the journal are required to declare the originality of their work, avoid all forms of plagiarism, and adhere to ethical guidelines. If plagiarism is identified during the peer review process, the manuscript will be rejected.

Post-publication and Malpractice Statement

• In the event that plagiarism or misconduct is discovered after publication, the journal reserves the right to take appropriate action to correct any inaccuracies or retract the paper. Requests for corrections by authors to address errors that impact the interpretation of the manuscript after publication are subject to approval by the editor. If a reader identifies a serious error, they are encouraged to contact the authors of the paper directly.

Ethical Guidelines for Authors

- Ensure that the manuscript submitted for review is original, based on research, written by the listed authors, has not been published elsewhere, and is not being submitted simultaneously to other journals.
- Obtain approval from all authors and the research institutions where the study was conducted before submitting the manuscript.
- Ensure that individuals who contributed to the research receive appropriate acknowledgment. Avoid plagiarism and the use of fabricated data. Declare any conflicts of interest. Do not cite unpublished work.

Peer Review Policy

- The journal employs a double-blind peer review process, ensuring that both reviewers and authors remain anonymous to one another. Under this process, the journal invites two experts to evaluate each manuscript. The manuscript will be returned to the authors for revisions based on the referees' comments and suggestions. Revisions will continue until the referees approve the manuscript.
- The editorial board reserves the right to close the review process if authors fail to respond to necessary revisions or if the referees do not recommend the manuscript for publication. The final recommendation of the referees will be considered by the editor in chief when making the decision to publish.



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

BRANCH STATIONS:

DA-PhilRice Batac, MMSU Campus, City of Batac, 2906 llocos Norte; Mobile: 0919-944-3016; Email: batac_1.station@mail.philrice.gov.ph DA-PhilRice Isabela, Malasin, San Mateo, 3318 Isabela; Mobile: 0999-889-3027; Email: isabela.station@mail.philrice.gov.ph; philriceisabela3318@gmail.com DA-PhilRice Los Baños, UPLB Campus, Los Baños, 4031 Laguna; Tel: (49) 501-1917; Mobile: 0993-631-9175; Email: losbanos.station@mail.philrice.gov.ph DA-PhilRice Bicol, Batang, Ligao City, 4504 Albay; Tel: (52) 431-0122; 742-0690; 742-0684; Email: bicol.station@mail.philrice.gov.ph DA-PhilRice Negros, Cansilayan, Murcia, 6129 Negros Occidental; Mobile: 0909-129-3763; Email: negros.station@mail.philrice.gov.ph DA-PhilRice Agusan, Basilisa, RTRomualdez, 8611 Agusan del Norte; Telefax: (85) 806-0463; Email: agusan.station@mail.philrice.gov.ph DA-PhilRice Midsayap, Bual Norte, Midsayap, 9410 Cotabato; Mobile: 0938-374-1040; Email: midsayap.station@mail.philrice.gov.ph

SATELLITE STATIONS:

Mindoro: Alacaak, Sta. Cruz, 5105 Occidental Mindoro; Mobile: 0919-495-9371 Samar: UEP Campus, Catarman, 6400 Northern Samar; Mobile: 0921-555-5500; 0948-754-5994; Email: philricesamar@gmail.com Zamboanga: WMSU Campus, San Ramon, 7000 Zamboanga City; Mobile: 0975-526-0306

DA-PhilRice Field Office, CMU Campus, Sayre Highway, Musuan, Maramag, 8714 Bukidnon; Mobile: 0909-822-9813; Email: philricefocmu.2019@gmail.com Liaison Office, 3rd Floor, ATI Building, Elliptical Road, Diliman, Quezon City; Mobile: 0928-915-9628



CALL FOR PAPERS

The Rice-Based Biosystems Journal is a peer-reviewed online and an open-accessed journal in The University Library of University of the Philippines Diliman, Cagayan State University Library, and recently in Caloocan City Science High School. It is managed by the Philippine Rice Research Institute (PhilRice) through a competent editorial team, reputable advisory and editorial boards, and invited referees. The journal encourages publication of original research and review articles that have impact on and are interconnected with rice and rice-based crops. It publishes rice and rice-based research studies on soil and crop management, crop protection, crop improvement, grain quality, farm machinery, resource use efficiency, plant biology, nutraceuticals, food value-adding systems, biofertilizers, biopesticides, biomaterials, and systems analysis and stimulation. It also covers the economics, social, and communication systems that may influence the landscape of rice and rice-based production. For your submission, you can email your manuscript to rbbj.philrice@gmail.com with subject and file format as: RBBJ_Title_Surname.doc. For more information about the journal, please visit www.philrice.gov.ph/grants/call-publication/.



Volume 12 • Issue 2 • August 2024

CALL FOR PAPERS

visit www.philrice.gov.ph/grants/call-publications/

Mailing address: PhilRice Central Experiment Station Science City of Muñoz, 3119 Nueva Ecija





Philippine Rice Research Institute Central Experiment Station Maligaya, Science City of Murice, S119 Nueva Edja