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# HIGH-VALUE PRODUCTS FROM RICE AND ITS ENVIRONMENT

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# High-Value Products from Rice and its Environment

Program Leader: RT Cruz

#### **Executive Summary**

Some of the main causes of poverty in the Philippines are (a) low to moderate economic growth for the past 40 years, (b) high inflation during crisis periods, (c) high level of population growth, (d) weakness in employment generation and quality of jobs generated and (e) failure to fully develop the agriculture sector (Asian Development Bank 2009). Less than a quarter of the Philippine population derived most of its income from agriculture (World Bank Philippine Development Report 2013). Yet poverty was highest among the agricultural households. The gap and severity of poverty among agricultural households were at least twice as high as the national average and at least four times as high compared to those who were mainly dependent on non-agricultural incomes.

High incidence of poverty contributes to prevalence of malnutrition in the country. Protein-energy malnutrition and micronutrient deficiencies remain to be the leading nutritional problems in the Philippines (FAO Report 2010). Therefore, it is imperative to improve the level of income and nutritional status of rice farming households and communities. This can be achieved through improved crop management practices, i.e., pre-harvest and post-harvest management affecting grain yield and quality, reduced farming cost, increased value of their produce, development of sustainable products and generation of additional sources of income from rice and its environment. Adding value to products from rice and its environment can enhance product quality, nutritional and health properties, shelf-life, availability and market value that can benefit the rice farming households, communities, consumers and industry stakeholders.

The High-Value Products from Rice and Its Environment (HVPRE) Program has four Projects: (1) HVP-001 on Program Management to assess accomplishments of the project studies, streamline project studies and allocate funds, (2) HVP-002 on High-Value Rice Grain, (3) HVP-003 on High-Value Products from the Rice Grain and Other Parts of the Rice Plant and (4) HVP-004 on Beneficial Organisms in the Rice Environment.

The HVP-002 Project highlighted the improvement and development of technologies for high value-added products from different rice variants such as brown rice, GABA (gamma-Amino butyric acid) rice, high-iron rice, high-zinc rice, aromatic rice and organic rice. The goals of the Project were to increase production and utilization of high-value added products to increase income of farmers and manufacturers, and help eradicate malnutrition and reduce chronic diseases in the country. Specifically, the Project aimed to (a) develop appropriate technologies to improve nutritional property and keeping quality of brown rice, (b) establish processing technology for the production of high quality GABA rice and GABA rice-based food products and supplements, (c) assess eating quality and consumer acceptability of commercially-fortified and bio-fortified iron rice and zinc rice, and recommend optimum agronomic practices for the production of bio-fortified iron rice and zinc rice, (d) evaluate the suitability of rice blends such rice-adlai and rice-corn as alternative food staples as part of an effort to address food security and (e) establish the optimum preharvest and post-harvest crop management practices for the production of aromatic rice and maintenance of its aroma, and production of organic rice to assess grain yield and quality, marketability and resource use. Significant accomplishments of Project HVP-002 were (a) brown rice intake to lower fasting blood glucose, (b) Nutri Rice Milk with GABA as a marketable beverage, (c) profiling and bioavailability determination of iron and zinc in biofortified rice lines, (d) acceptability of rice blended with brown adlai or ginampay for additional energy source, and (e) improvement of the PalayCheck System with the evaluation of the new key check on postharvest management.

The HVP-003 Project focused on the utilization of rice plant parts such as grain, leaves, panicle, straw, and root as potential sources of proteins, bioactive peptides, carbohydrate-based prebiotics, and secondary metabolites having biomedical and industrial applications. The developed products and technologies can be directly transferred to various stakeholders engaged in the production, marketing and commercialization of functional foods and antioxidant supplements. The Project aimed to (a) establish procedures for the isolation of lignans, terpenoids and flavonoids from the rice plant and assess their health-promoting properties, (b) assess the potential of rice as source of bioactive peptides with anti-hypertensive properties, (c) establish nanotechnology for the encapsulation of antioxidants and proteins from rice bran for functional food and biomedical applications and (d) evaluate the potential of rice grain and its co-products as sources of prebiotics (fiber compounds that stimulate growth of probiotics or beneficial microorganisms) to produce nutraceuticals for nutritional and health improvement or functional foods for health-promotion and disease prevention. Significant accomplishments of Project HVP-003 were (a) utilization of rice cell and organ cultures as sources of secondary metabolites, i.e., lignans, flavonoids and terpenoids, for pharmaceutical and healthrelated applications, (b) bioactive peptides from rice with potential antihypertensive activity, (c) nutritious rice bran as source of natural antioxidants and proteins, and (d) development of a rice cooking method that can yield high amount of prebiotics.

The HVP-004 Project generally aimed to develop products and technologies that will benefit the farmers, industry and rice environment.

Specifically it aimed to formulate (a) symbiotic fungal endophytes that can increase rice yield from an average of 1.5 to 3.5t/ha in drought environment, (b) bio-control agents (BCAs) for the control of white stem borer, rice black bug and rice bug, (c) feed supplement from blue green algae strains with high nutrient content, anti-oxidant and anti-bacterial properties required by the industry and (d) high-quality, living and diversified inoculants from rhizotrophic and endophytic bacteria, and (e) to assess efficiency of Anabaena-azolla symbiosis for nitrogen fixation and carbon sequestration. Significant accomplishments of Project HVP-004 were (a) isolation of rice fungal endophytes that has the potential to enhance rice drought tolerance, (b) utilization of blue-algae as a feed supplement, and (c) isolation of bacteria from the rice root zone and their potential use as biological control agents against pests and for rice plant growth promotion.

Due to realignment of researches in the Institute, Project HVP-004 was transferred to the Applied Biology Center for the Rice Environment in the second half of 2014.

# I. High-Value Rice Grain

Project Leader: MV Romero

In the Philippines, rice is the most important part of the diet because it is the staple food of the population. Thus, rice serves as the primary source of calories needed for the daily energy requirement of Filipinos. It also provides a significant amount of protein to those who cannot afford to buy protein-rich foods such as meat and fish. Majority of Filipinos prefer to eat cooked milled or polished rice because of its excellent eating quality. However, milled or polished rice contains mostly carbohydrates and protein since most of the micronutrients are removed during the milling process. Brown rice or the unpolished form of rice has more health benefits than milled rice because of higher amounts of dietary fiber, fat, vitamins, minerals, and antioxidants. There is also the germinated or sprouted brown rice that contains  $\gamma$ -amino butyric acid (GABA). This form of rice is more nutritious, softer, sweeter, and tastier compared to brown rice. The persistent problem of micronutrient deficiency in the country has prompted various interventions such as commercial fortification and biofortification to enhance the amounts of iron and zinc in rice. Since the Philippines is rich in nutrient-dense crops, they can also serve as alternative staple foods. Efforts are being made to incorporate other staples in the regular diet by substituting them to rice as the main item of the meal. As most consumers are not yet accustomed to eating them as total replacement to table rice, alternative staples may be mixed with rice in certain proportions that would satisfy consumer requirements on eating quality, nutritional improvement, and hunger mitigation. Aromatic and organic rice are becoming popular nowadays and command a higher price in the market. But these two

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properties have to be evaluated following improvements in pre-harvest, harvest and postharvest management components of the integrated crop management, the PalayCheck System, to improve grain yield and quality.

This project evaluated brown rice, germinated brown rice, micronutrient-dense rice, nutrient-rich rice blends, aromatic rice, and organic rice that have added value and health benefits compared to the regularly milled rice. The general objectives were: (1) to improve the nutritional and keeping quality of brown rice and determine its potential health benefits through clinical studies and intervention feeding trial; (2) to develop healthy and nutritious GABA rice from Philippine rice cultivars and identify their potential applications to food and pharmaceutical industries; (3) to determine the grain quality and acceptability of commercially-fortified and biofortified raw and cooked iron- and zinc- rice; (4) to explore the potential of corn and adlai in the preparation of nutrient-rich rice blends with high consumer acceptability; and (5) to develop pre-harvest and postharvest management for aromatic and organic rice

#### **Brown Rice Quality Improvement**

HM Corpuz, MB Dacumos, MJC Ablaza, and MV Romero

Consumption of brown rice (BR) is being encouraged because of its potential contribution to the prevention of chronic diseases and availability of more rice for food. Despite its nutritional and economic advantages, BR remains unappealing to some people because of its short shelf-life due to high amount oil present in the bran. BR easily turns rancid under ambient condition and could only last for 3 to 6 months compared to 12 months for white or milled rice. Heat stabilization techniques have been found effective in prolonging the shelf-life of cereal grains like brown rice. Heat treatments can silence the activity of lipase enzyme which is responsible for the degradation of oil into free fatty acid (FFA). FFAs are then oxidized to secondary products that give rancid odor and flavor to the grains. Parboiling technique can be one of the suitable options to prolong the storability of BR as it involves steaming process needed to deactivate lipases. Thus, this study was conducted to determine the effects of parboiling method on the shelflife, eating, and sensory characteristics of BR.

Studies have shown that regular intake of whole grain cereals instead of refined grains may help lower the risks of many chronic diseases. Since BR belongs to this food group and contains significant amounts health-promoting compounds, there is also a need to evaluate the therapeutic potentials of brown rice against chronic diseases such as diabetes and cancers.

#### **Highlights:**

Parboiled rice grains of NSIC Rc160 and NSIC Rc218 were dehulled and stored in different packaging materials at ambient conditions for 9 months. The physicochemical properties, cooked rice Instron hardness, and sensory attributes of the samples were measured periodically. Analysis of variance revealed that parboiling treatment, type of storage containers, and storage period had significant effect on the physicochemical properties of the samples (Table 1). For both varieties, FFA content and lipase activity of parboiled brown rice were significantly lower compared to that of the untreated samples. However, the opposite was observed on the antioxidant activity of the samples. This could be due to the solubilization and degradation of antioxidants in the grains during soaking and steaming process, respectively. In terms of packaging materials, samples stored in nylon-coated pouch had lower FFA content and lipase activity compared to those samples stored in plastic containers. Nylon-coated plastic pouch could prevent the exposure of the grains to oxygen in the air thereby reducing fat oxidation. Generally, FFA content, enzyme and antioxidant activities increased with storage time and higher values were observed after 6 to 9 months of storage.

- Instron analysis showed that parboiled brown rice samples had significantly softer cooked grains compared to the control. This was due to the higher optimum cooking water used for parboiled brown rice and the grains were partially cooked during the steaming process. However, the cooked grains of parboiled brown rice were fluffier and separated. Glossiness, color, and acceptability of raw samples decreased with storage time. Off-odor was perceived in the untreated samples after 3 months storage period and the unpleasant smell became more distinct in the succeeding months. The rancid off-odor was perceived in parboiled brown rice samples during the 5th month of storage. Rancid off-odor score was higher in the untreated brown rice relative to parboiled brown rice. Similarly, aroma, gloss, and color of cooked brown and parboiled brown rice slightly decreased whereas the rancid offodor, off-taste, taste, and general acceptability increased with storage time. The samples stored in nylon-coated pouch had more acceptable sensory scores than the untreated samples.
- To determine the antidiabetic and anticancer properties of brown rice, pre-clinical studies using animal models were conducted in collaboration with the Pampanga State

Agricultural University. For the antidiabetic screening, the Sprague Dawley rats were injected with alloxan to induce diabetes (Figure 1). Diabetic rats were fed with brown and milled rice grains of NSIC Rc13, NSIC Rc160, and NSIC Rc152. The blood glucose level of the test animal was measured periodically. Preliminary results revealed that diabetic rats fed with brown rice samples had lower fasting blood glucose level compared to those fed with milled rice samples. A follow-up study was conducted using diabetic mutant mice to validate the initial findings. The evaluation of the anticancer property of brown rice through anticlastogenic screening was still in process.

**Table 1.** Summary of means (by factor) for physicochemical properties of brown rice and parboiled brown rice.

Treatments	Free Fa	atty Acid	Lipase Activity		Antio> Acti	
	(mg linol	eic acid /g	(as mg lin	oleic acid	DPPH Sca	avenging
	E	BR)	/g	BR)	Activi	ty (%)
	NSIC	NSIC	NSIC	NSIC	NSIC	NSIC
	Rc160	Rc218	Rc160	Rc218	Rc160	Rc218
Rice Type						
Brown rice	2.40ª	1.79 <sup>a</sup>	0.94ª	0.80 <sup>a</sup>	42.92ª	42.62ª
Parboiled	0.70 <sup>b</sup>	0.47 <sup>b</sup>	0.41 <sup>b</sup>	0.36 <sup>b</sup>	29.53 <sup>b</sup>	29.57 <sup>b</sup>
brown rice						
Packaging						
Materials						
Plastic	1.67ª	1.22ª	0.73ª	0.62ª	36.20ª	36.07ª
container						
Nylon-	1.44 <sup>b</sup>	1.04 <sup>b</sup>	0.62 <sup>b</sup>	0.54 <sup>b</sup>	36.24ª	36.13ª
coated						
plastic						
pouch						
Storage						
Time						
(Months)						
0	0.60 <sup>f</sup>	0.38 <sup>e</sup>	0.04 <sup>g</sup>	0.04 <sup>g</sup>	30.71 <sup>d</sup>	31.19 <sup>e</sup>
1	1.17°	0.89 <sup>d</sup>	0.54 <sup>f</sup>	0.44 <sup>f</sup>	34.43¢	34.84 <sup>d</sup>
2	1.43 <sup>d</sup>	0.93 <sup>cd</sup>	0.65°	0.51 <sup>ef</sup>	36.40 <sup>b</sup>	36.04c
3	1.53 <sup>cd</sup>	1.09 <sup>bcd</sup>	0.70 <sup>de</sup>	0.58 <sup>de</sup>	37.26ª	36.99 <sup>ab</sup>
4	1.54 <sup>cd</sup>	1.18 <sup>abcd</sup>	0.73 <sup>cde</sup>	0.61 <sup>cd</sup>	37.57ª	37.33ª
5	1.73 <sup>bc</sup>	1.21 <sup>abc</sup>	0.76 <sup>bcd</sup>	0.64 <sup>cd</sup>	37.35ª	37.13 <sup>ab</sup>
6	1.79 <sup>ab</sup>	1.35 <sup>ab</sup>	0.78 <sup>abcd</sup>	0.68 <sup>bc</sup>	36.90 <sup>ab</sup>	36.63 <sup>bc</sup>
7	1.84 <sup>ab</sup>	1.36 <sup>ab</sup>	0.82 <sup>abc</sup>	0.73 <sup>ab</sup>	37.04 <sup>ab</sup>	36.99 <sup>ab</sup>
8	1.93 <sup>ab</sup>	1.39 <sup>ab</sup>	0.84 <sup>ab</sup>	0.77ª	37.26ª	36.81 <sup>ab</sup>
9	1.96ª	1.48ª	0.86ª	0.81ª	37.29ª	37.03 <sup>ab</sup>

Mean values with different letters within the same column are significantly different (p<0.05).



Figure 1. (A) Administration of alloxan to induce diabetes; (B) Periodic measurement of fasting blood sugar level of diabetic rats.

# Healthy and Nutritious GABA Rice for Functional Food and Pharmaceutical Industries

RMBulatao, JMMAsuncion, JBChavez and MVRomero

Gamma-amino butyric acid (GABA) is currently one of the most promising compounds due to its health-promoting and cognitive-enhancing properties. It is a non-protein amino acid that serves as neurotransmitter in the central nervous systems. GABA-enriched rice was found to lower blood pressure, inhibit leukemia cell proliferation, and stimulate cancer cell apoptosis. Regular intake of GABA-rich foods or supplements is effective in stimulating immune system and is helpful in treating insomnia, mental irritation, depression, and other neurological disorders such as Parkinson's disease, stiff-man syndrome, and schizophrenia. Because of these, GABA rice is being consumed as a staple food and widely marketed as a functional food in Japan, China, Korea, Malaysia and Thailand. It is used as ingredient in making bread, cookies, crackers, rice balls, rice burger, and instant GABA rice drink. However, GABA rice has not yet been explored in the country given that we have many rice varieties for the production of this novel food stuff. Therefore, this study evaluated the potential of Philippine rice varieties in the production of healthy and nutritious GABA rice for functional food and pharmaceutical products.

#### **Highlights:**

A total of 20 rice varieties were collected from the Business Development Division, PhilRice, Science City of Muñoz, Nueva Ecija. Rice samples were processed into brown rice and their germ size was measured using a compound microscope attached to a computer. The surface area ( $\mu$ m2) and perimeter ( $\mu$ m) of the germ were calculated using DP2-BSW Application Software (Olympus). Among the samples, NSIC Rc160 had the largest surface area and perimeter with the values of  $83,249.60\mu$ m2 and  $1,346.8\mu$ m, respectively. Thus, this variety was selected as sample for the optimization of germination process for the production of dried GABA rice.

- GABA rice was produced by soaking brown rice samples with water at different time intervals (12, 24, 36, 48, and 72 h). The soaked GABA rice was then subjected to different drying methods such as sun-drying (27 to 30°C for 24 h), oven-drying (60°C for 12h), and freeze drying (-20°C for 48h).
- Potential of GABA rice samples in the prevention of diabetes and cancer was evaluated at the Institute of Veterinary Medicines and Zootechnics, Pampanga Agricultural State University. ICR and db/db mice were used to determine the antidiabetic and anticlastogenic potentials of GABA rice samples, respectively.
- GABA analysis was optimized using a chromatographic method through Ultra Performance Liquid Chromatography (UPLC). During the optimization, brown rice, germinated brown rice, and the bran fraction of NSIC Rc160 were extracted, derivatized, and injected into the UPLC system to obtain a chromatogram. Among the samples, the germinated NSIC Rc160 had the highest GABA content of 17.1mg/100g while its brown rice form had the lowest value of 2.7mg/100 (Table 2).
- A prototype of GABA rice-based beverage was developed through a collaborative work with the Philippine Carabao Center at Central Luzon State University. The product was branded as Nutri Rice Milk, a healthy and nutritious drink with GABA rice and fresh buffalo's milk (Figure 2). Nutritional analysis indicated that Nutri Rice Milk was a good source of protein, fiber, vitamin A, calcium, and iron.
- To assess the market potential of Nutri Rice Milk, its shelf-life was evaluated for 14 days under refrigerated temperature (4°C). Physicochemical analysis showed that the water activity (0.92-0.97) and titrable acidity (18.1-19.5%) of the product significantly increased while pH (6.82-6.21) decreased considerably during storage. Microbial analysis indicated that the mold and yeast counts were low (1x102cfu/mL) until 14 days of storage. However, high total plate (1.4x105cfu/mL) and coliform counts (3.3x103 cfu/mL) were observed during the10th day of storage.

Sensory panelists (n=12) perceived the Nutri Rice Milk as sweet and flavorful. The rice bits in the product provided a chewy texture that was pleasing to the palate. Although spoiled odor and flavor were not detected by the panelists during the 10th day of storage, the sensory evaluation of the product was immediately stopped due to the abrupt increase in the total coliform count.

Sample	GABA Content (mg/100g)
Brown Rice	2.7
Germinated Brown Rice	17.1
Rice Bran	3.9

#### Table 2. GABA content of samples.



# Quality Assessment of Iron- and Zinc-Dense Rice

RG Abilgos-Ramos, CT Estonillo

Iron (Fe) and zinc (Zn) are important micronutrients in relation to human health. However, Fe and Zn deficiency remained prevalent among Filipinos. Crop biofortification is one important and sustainable approach to address micronutrient deficiency. Rice with enhanced Fe and Zn content were developed through breeding. There were reports on the uptake of micronutrients in brown rice grains in response to indigenous soil nutrients and N fertilizer application but the influence of nitrogen (N) fertilizer management in relation to the uptake of Fe and Zn in polished rice grains requires further study.

This study aimed to determine the quality of biofortified rice with minerals (iron and zinc) and to assess the effectiveness in terms of bioavailability and acceptability to consumers. This work also evaluated the influence of N fertilization on Fe- and Zn-dense rice genotypes. Results of this study will serve as basis of the recommendation on how to market or improve the iron and zinc-enhanced rice for maximum utilization or how to proceed with the breeding of materials for dissemination and distribution.

## Highlights:

Activity 1. Grain Quality and Nutritional Profile of Fe and Zn Rice

Grain quality evaluation plays a vital role in rice breeding. Grain quality includes milling recovery, physical attributes, and eating properties. In addition to breeding, identification of rice lines with increased micronutrient content like iron and zinc in response to N fertilizer level will enable to assess the extent to which agronomic management can increase the micronutrient content of a certain rice genotype. In 2014 wet season (WS), varieties PSB Rc82, IR68144-2B, PR38142-B, PR35015-GA, and PR38963 were evaluated following application of different N fertilizer levels in the field. The harvested samples from the 5 rice varieties were evaluated for iron and zinc contents on the brown and milled rice forms.

## Grain Quality

- All of the entries met the brown rice and total milled rice recovery standard of grade 1 to premium, but only 44% (11 entries) passed the head rice recovery requirement of at least 48% with grade 1 to premium.
- In terms of physical dimension, samples were long and slender except for IR68144 that had medium and intermediate grains.
- Most of the samples had acceptable % chalky grains except for PSB Rc82 with grade 2 class and did not pass the recommended chalky grain classification.
- Based on oven drying method, all entries passed the recommended grain moisture content that ranged from 10.6 to 11.8%, indicating that samples were stable for long-term storage.
- All entries had intermediate amylose content and predictably with low to high intermediate texture in cooked form. This indicated good eating quality and consumer acceptability of most entries evaluated.
- The crude protein content of samples was evaluated using Kjeldahl method. Crude protein contents ranged from 6.5 to 9.5%. PR38963 with no N fertilizer had the lowest percentage crude protein of 6.5. This could be due to smaller amount of

nutrient absorbed by the plant. PSB Rc82 with high level of N fertilizer had the highest crude protein of 9.5%.

# Seed Production of Biofortified Rice (Fe and Zn)

• Thirty advanced lines derived from different crosses of IR68144 (NSIC Rc172) were seed-increased. Yield performance of top 10 entries ranged from 2047 kg/ha to 2410 kg/ha in 2014 WS (Figure 3). Severe crop lodging at maturity and bacterial leaf blight (BLB) infection were observed on most of the test entries that resulted to lower yields. Seed samples were processed for grain iron and zinc content analysis.

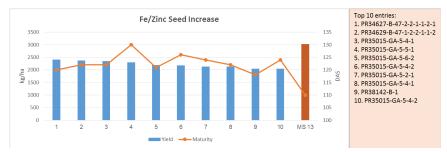


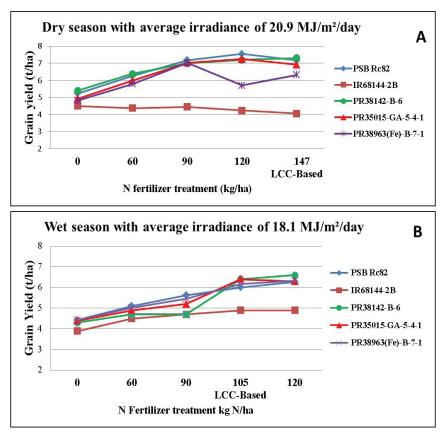
Figure 3. Yield performance of top 10 entries of Fe/Zinc Seed Increase, 2014 wet season.

Activity 2. Influence of Nitrogen Fertilization on Fe- And Zn- Rice Genotypes

Field experiment was conducted at PhilRice Nueva Ecija in 2014 dry and wet seasons (DS and WS) using PSB Rc82, IR 68144-2B-2-2-3-1-166, PR 38142-B-6, PR 35015-GA-5-4-1, and PR 38963 (Fe)-B-7-1. Fertilizer treatments were: (1) Control or 0kg N/ha with 60kg PK/ha applied at 14 days after transplanting (DAT), (2) 60kg N/ha where N fertilizer was applied equally in three splits at 14 DAT, early panicle initiation and heading stages, (3) 90kg N/ha where N fertilizer was applied equally in three splits at 14 DAT, early panicle initiation, and heading stages, (4) 120kg N/ha where N fertilizer was applied equally in three splits at 14 DAT, early panicle initiation and heading stages, and (5) LCC-based N fertilizer application where 30kg NPK/ha using "complete fertilizer', i.e. 14-14-14-12S, was applied at 14 DAT and 35kg N/ha was applied when LCC reading was below the critical value of 4. LCC readings were done weekly starting at 21 DAT until early flowering stage. All P and K fertilizers at 60 kg/ha each were applied in all treatments at 14 DAT.

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Grain yields of rice samples ranged from 4.5 to 5.4t/ha in DS (Figure 4A) and 3.9 to 4.4 t/ha in WS (Figure 4B). Grain yields of PSB Rc82, PR38142-B-6, and PR35015-GA-4-1 increased with increased amount of N fertilizer from 60kg N/ha to 120kg N/ha in DS and WS. However, PR38963 (Fe)-b-7-1 yield increased only in WS. With LCC-based N fertilizer application, optimum yields were achieved at 120kg N/ha in DS and 105kg N/ha in WS. Grain yields of PR38963 (Fe)-B-7-1 were optimum at 90kg N/ha to 120kg N/ha in DS and WS. Grain yields of IR68144-2B were lower than yields of PSB Rc82, PR38142-B-6, PR 38963 (Fe)-B-7-1, and PR35015-GA-4-1 in DS and WS and were similar across N fertilizer treatments. Across treatments, grain yields in 60, 90, and 120kg N/ha and LCC-based N fertilizer application were higher than the control with no N fertilizer. Grain yields in 90kg N/ha, 120kg N/ha and LCC-based N fertilizer application were similar but higher than grain yields in 60kg N/ha. Generally, yields in DS were higher than yields in WS due to higher irradiance in DS. Fe and Zn content of grains as influenced by N fertilization still have to be measured.



**Figure 4.** Grain yields of inbred check varieties PSB Rc82 and IR68144-2B, and genotypes that have high Fe and Zn grain contents PR38142-B-6, PR35015-GA-5-4-1, and PR38963 (Fe)-B-7-1 at different N fertilizer levels in (4A) 2014 dry season and (4B) wet season. N fertilizer treatments of 60, 90 and 120kg N/ha were applied "fixed time" at 14 DAT, early panicle initiation, and heading stage. The N treatment with a total of 147kg N/ha in dry season (4A) and 105kg N/ha in wet season (4B) used the PhilRice leaf color chart (LCC) for "real time" N fertilizer application.

Activity 3. Bioavailability of Fe and Zn rice lines

Profiling and Bioavailability Determination of Iron and Zinc in Biofortified Rice Using Normal and Anemic Rats

 The first part of the experimentation was the iron and zinc profiling of the rice lines (IR68144 and PR35015). Hundred grams of each sample were dehulled and floured. The samples were kept in airtight containers and stored in a refrigerator before processing. An optimized dry-ashing technique was employed. The steps and specific dates of completion are outlined in figure 5.

- Five replicates of each sample, including three blank solutions were prepared as described in Figure 6. Samples were kept in 50mL Eppendorf tubes (Figure 7) and stored in the refrigerator prior to analysis.
- The second part of the experimentation was the determination of the amount of bioavailable iron and zinc and the monitoring of changes in the blood iron and zinc of rats before and after the feeding trials. Body weight, complete blood count (CBC), blood iron and zinc levels were measured to quantitate the effects of the rice diets.
- The rats were procured from the Food and Drug Administration, Filinvest City, Alabang in December 2014. All 36 Sprague-Dawley rats are currently housed in the National Institutes of Health (NIH) animal house facility. All rats are being acclimatized, fed daily, and provided with drinking water ad libitum. Beddings and cages are replaced and cleaned every three days to ensure a favorable living environment for the test subjects in accordance with the guidelines set by the UP Manila Institutional Animal Care and Use Committee (IACUC) as per approval of study implementation in August 2014.

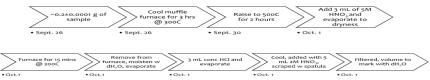
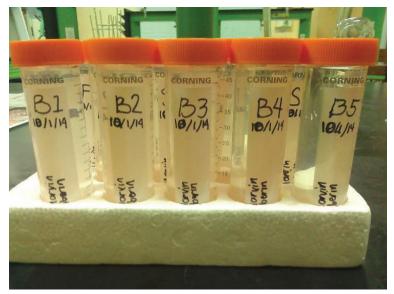


Figure 5. Schematic diagram of the optimized method for dry-ashing of rice samples.



**Figure 6.** (A.) Rice samples were dehulled using a Satake dehuller (B.) Samples were kept in air-tight containers for long-term storage.



**Figure 7.** Dry-ashed samples for AAS analysis. All samples are properly labeled and all pertinent information (e.g. code, date of preparation) are also included in the label.



Figure 8. Rats are currently housed in individual cages and are fed and provided with drinking water, ad libitum.

## **Development of Nutrient-Rice Blends Using Local Food Crops**

RV Manaois, HF Mamucod, AV Morales, MV Romero

The increasing per capita intake of rice has been identified as one of the factors that contribute to rice shortage. Hence, efforts are being made to incorporate other staples in the diet by substituting them to rice as the main item of the meal. These alternative staples include adlai and corn. Through its Food Staples Self-Sufficiency Program, the Department of Agriculture (DA) regards the consumption of these alternatives as one measure to reduce rice intake and consequently address food security.

Adlai or "katigbi" (Coix lacryma-jobi L. var. ma-yuen) has actually been grown and eaten mostly in Southern Philippines, particularly in Zamboanga del Sur (Dela Cruz, 2011), although it is more popular in neighboring Asian countries. In the Philippines, four varieties are grown: gulian (white), tapol (purple), ginampay (brown), and pulot (white-glutinous). Nutritional value of adlai was reported to be comparable, or even better, than rice. Analysis at the Food and Nutrition Research Institute (Adlai, n.d.) revealed that adlai provides around the same amount of energy as milled rice (Juliano, 2003) at 356 kcal per 100 g, but its protein and total fat content were higher (Adlai, n.d., Dechkunchon and Thongngam, 2007).

Corn (Zea mays), specifically the white variety of Quality Protein Maize (QPM) is another alternative crops being campaigned by the

government. When processed into grits, QPM has acceptable taste similar to rice alone. QPM is nutritionally superior to ordinary white corn in terms dietary fiber and minerals. It also contains two essential amino acids, lysine and tryptophan. Considered as low GI food, QPM has higher amylose content that makes it harder to gelatinize and slower to digest compared with rice.

Despite the nutritional advantages of adlai and QPM, most consumers are not yet accustomed to eating these staples as total replacement to table rice. Consumption of or shifting to pure adlai or corn meal would still be a huge challenge. One way to address this is through substitution or mixing alternative staples with rice in certain proportions that would satisfy consumer requirements on eating quality, nutritional improvement, and satiety. This study aimed to establish the rice:adlai and rice:corn (QPM) blends with good eating quality and high consumer acceptability.

## **Highlights:**

Rice:Adlai Blends

- Ginampay, gulian, and tapol (Figure 9) were procured from the Cagayan Valley Integrated Agricultural Research Center (CVIARC) in Ilagan City, Isabela. The three adlai varieties were harvested in February 2014. While, rice varieties were obtained from the Business Development Division of PhilRice.
- Soaking did not affect the cooking qualities of ginampay, gulian, and tapol, but significantly improved the texture of the samples (Figure 10). Soaking of ginampay for at least 30min gave a comparable tenderness and overall liking with rice. The process had no significant effect on the aroma, taste, and overall acceptability of rice and three adlai varieties.
- Six rice varieties representing three different amylose classifications (AC) were evaluated: PSB Rc10 and NSIC Rc282 (high AC), NSIC Rc216 and NSIC Rc222 (intermediate AC), and NSIC Rc160 and NSIC Rc218 (low AC).
- Blending rice (particularly intermediate to high AC varieties) with ginampay resulted in softer texture in terms of Instron hardness (Figure 11) and higher scores for smoothness, tenderness, and overall acceptability in terms of laboratory sensory evaluation.
- NSIC Rc160 and NSIC Rc218 blended with ginampay had the lowest Instron hardness values making them the softest among

the blends.

- The blend forms of NSIC Rc160 and NSIC Rc218 also received the highest score for gloss, cohesiveness, tenderness, smoothness, and taste.
- Both NSIC Rc160 and NSIC Rc218 blends were acceptable to the consumer panelists, although they preferred the pure rice forms (Table 3).



Ginampay

Gulian

Figure 9. Adlai varieties used in the preparation of rice-adlai blends.

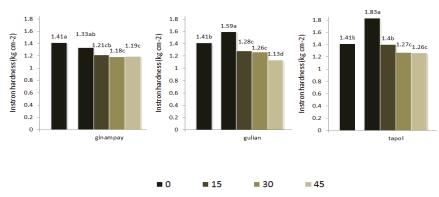


Figure 10. Effect of soaking time on the texture of cooked adlai samples.

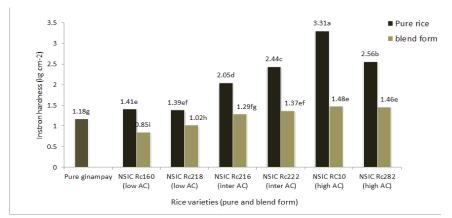


Figure 11. Texture of pure and blend forms of rice with different AC types.

Table 3. Consumer	acceptability	of pure ar	nd blend	forms of	of NSIC Rc16	0 and
NSIC Rc218.						

A 11 11 1	Pure rice		Blend	l form
Attributes –	NSIC Rc160	NSIC Rc218	NSIC Rc160: ginampay	NSIC Rc218: ginampay
% Acceptability <sup>a</sup>	95.83ª	91.67 <sup>ab</sup>	89.58 <sup>ab</sup>	83.33 <sup>b</sup>
Mean ranking <sup>b</sup>	2.17 <sup>b</sup>	2.21 <sup>b</sup>	2.98ª	2.65ª

aBased percent of positive responses (n=48)

b1=highest, 4=lowest

Rice:Corn Blend

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- Corn grits (Quality Protein Maize var. 6 or white flint corn) (Figure 12) was obtained from the Institute of Plant Breeding, University of the Philippines Los Baños, College, Laguna. Rice samples were purchased from the Philippine Rice Research Institute.
- To determine the optimum amount of cooking water, the grits were treated with increasing amount of water (1:1, 1:1.25, 1:1.5, 1:1.75, 1:2, 1:2.5, and 1:3). Results showed that regardless of amount of water, the texture of the cooked corn grits remained hard. The 1:2 and 1:3 corn grits:water ratios were less hard among the samples.

Soaking the corn grits for 15 to 30min had no significant effect on the texture of its cooked form.

#### 20 Rice R&D Highlights 2014

- With different rice:corn ratios, results showed that increasing the ratio of corn on the blend resulted in increased weight, longer cooking time, and higher Instron values compared to pure rice. Meanwhile, height of cooked samples was not affected by the different rice:corn ratios. In terms of laboratory sensory evaluation (n=11), aroma, color, gloss, smoothness, and taste of rice-corn blends, regardless of ratio, were generally comparable to pure rice. No off-odor and off-taste were perceived by the sensory panelists in all rice blend samples. The acceptability of rice blended with corn up to 40% was comparable to pure rice.
  - Using the 60: 40 rice to corn ratio, result of the cooking quality evaluation showed that weight and height of all cooked rice:corn blends were significantly higher than their pure cooked rice forms, except for NSIC Rc10. Rice:corn blends took longer time to cook (additional 2 to 4 min) compared to their pure forms, except for NSIC Rc10. In terms of the laboratory sensory evaluation, pure NSIC Rc218, an aromatic rice variety, had significantly higher score for aroma compared with other cooked rice samples. Off-odor and off-taste were not perceived by the panelists in all samples. Color, glossiness, cohesiveness, tenderness, smoothness, taste, and overall liking of rice-corn blends, regardless of rice variety used, were generally comparable to their pure cooked rice forms. The blend forms of NSIC Rc160 and NSIC Rc218 received overall likings comparable with their pure rice forms.



Figure 12. Corn grits (Quality Protein Maize var. 6 or white flint corn).

**Pre-harvest and Post-harvest Management for Aromatic and Organic Rice** *MUBaradi, MC Quimbo, MFA Magno, RG Ancheta, MV Romero, AJPAnies, MJC Ablaza, RTCruz, MJC Regalado* 

Aromatic and organic rice have become popular and continue to command higher price in local and international markets. Many people are willing to pay more for such types of rice, especially the upper class consumers. Aromatic rice is preferred by consumers because of its distinctive pleasant scent that makes it more special than the ordinary rice. Organic rice or organically-grown rice has also become popular among consumers because of the belief that it has more nutritional benefits (Tafere et al. 2011). Organic rice is grown only with organic amendments/materials/fertilizers and bio-pesticide.

The proposed studies on pre-harvest and post-harvest management for aromatic and organic rice will help the stakeholders, particularly the farmers and processors for the production of better quality aromatic and organic rice demanded by the market. Eventually, results of the studies will enable the farmers to be more competitive in the market, thus, giving them the opportunity to earn higher income from the sale of aromatic and organic rice.

#### A. Dry Season 2013-2014

The effects of nitrogen levels on the grain quality parameters, healthpromoting phytochemicals, and yields of three different rice genotypes grown under the PalayCheck System of integrated crop management for transplanted irrigated lowland rice were determined during the dry season (DS) 2013–2014. Aromatic variety Burdagol-Laguna Type, a pigmented rice variety (Gal-ong) and a non-aromatic PSB Rc82 were tested in response to inorganic and organic fertilizer treatments.

The experiment was conducted in San Nicolas, Ilocos Norte during the dry and wet seasons of 2014. The climate in the area was classified as type I having two pronounced seasons, dry from November to April and wet during the rest of the year. Maximum rain period was from June to September. The soil in the area was classified as San Fernando series clay or clay loam, neutral pH of 7.1, medium organic matter (OM) at 2.5%, low level of total nitrogen (N) at 0.12%, high available phosphorus at 47ppm, and high exchangeable potassium (K) at 0.62 me 100 g soil-1. The nutrient content of the rice straw used in the experiment was 0.76% total N, 0.09% total P, and 1.08% total K. Thus, the 4 t ha-1 rice straw applied was equivalent to 30kg N, 4kg P, and 43kg K. On the other hand, the chicken manure contained 1.82% total N, 4.29% total P, and 3.86% total K. Applying 3 t ha-1 chicken manure provided 55kg N, 129kg P, and 116kg K.

Result of the MOET set-up prior to crop establishment during the 2014 DS showed that the soil was deficient in N and K for Block 3 (Rep 3). T1-T4, T7 and T8 of Blocks 1 (Rep 1) and 2 (Rep 2) were N and S deficient; whereas T5 and T6 were N deficient.

The experiment was replicated three times and the treatments were laid out in the field following a Split Plot Design. Nutrient treatments (zero fertilizer, N-omission plot, P-omission plot, K-omission plot, LCC-based N fertilizer level 1, LCC-based N fertilizer level 2, Organic fertilizer 1, Organic fertilizer 2, and Organic fertilizer 3) were the main plot. Variety (PSB Rc82, Burdagol-Laguna Type, and Gal-ong) was the sub-plot. Each plot size was 4m x 5m (20m2).

# Highlights:

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- The yield was determined by the interaction of the fertilizer treatment and variety used. Highest yields were obtained from those applied with inorganic fertilizers. Yield in the N Omission Plot (-N) was similar to the yields of the Control (no fertilizer applied) and in treatments with organic fertilizers (i.e., chicken manure and rice straw). Gal-ong hadthe lowest yield among the three varieties (Table 4).
- The yield of the three varieties were not significantly different at T1 (Control) and T7 (Rice Straw). However, in the six remaining fertilizer treatments (T2–T6, T8), Gal-ong gave the lowest yield while Burdagol and PSB Rc82 were not significantly different except at T6 where Burdagol outyielded PSB Rc82.

FERTILIZER	Fertilizer Rate		Grain Yield (t ha	a <sup>-1</sup> )
MANAGEMENT	(kg ha <sup>-1</sup> )	Burdagol	PSB Rc 82	Gal-ong*
1. Control	0	1.91 b	1.64 b	1.56 a
2. Omission Plot (-N)	0-42-42	2.11 b	2.07 b	1.31 a
3. Omission Plot (-P)	112-0-42	4.85 a	4.44 a	2.21 a
4. Omission Plot (-K)	112-42-0	4.89 a	4.38 a	2.81 a
5. LCC-based 1	108-42-42	4.95 a	4.80 a	2.47 a
6. LCC-based 2	118-42-42	4.86 a	4.03 a	1.84 a
7. Rice straw	30-4-43 (4 t ha <sup>-1</sup> )	1.92 b	1.94 b	1.72 a
8. Chicken Manure	55-129-116 (3 t ha			
	1)	2.62 b	2.37 b	1.68 a
Significance				
Fertilizer	***			
Variety	***			
Fertilizer x Variety	***			

**Table 4.** Grain yield of the different varieties as influenced by fertilizer treatments, (2014 DS), San Nicolas, Ilocos Norte.

\*\*\* significant at 0.1% level of significance. Means followed by different letter were not significantly different from each other at 5% level of significance using LSD. Yield was low due to bird damage.

#### Effect on Grain Quality Attributes

- The % brown rice of Burdagol and PSB Rc82 was not affected by the fertilizer treatments. Both varieties had fair brown rice recoveries (Table 5). However, for Gal-ong, the brown rice recoveries of the samples treated with T1 to T6 were all poor; only those applied with rice straw and chicken manure produced fair brown rice recoveries.
- Only PSB Rc82 had premium milled rice recoveries, regardless of the fertilizer treatment (Table 6). For Burdagol, T2, T5, and T8 produced premium milled rice; the rest of the fertilizer treatments were all under Grade 1 class. For the case of Galong, T3 and T7 were the only treatments that had premium milled rice recoveries.
- Of the three varieties, the head rice yield of PSB Rc82 was not affected by the fertilizer treatments (Table 7). All the samples belonged to Grade 1 class regardless of fertilizer treatment. However, for Burdagol, T3, T4 and T6 (inorganic treatments or higher N levels) had higher head rice yields (Grade 2) than T1, T2, T7 and T8 (Grade 3). For Gal-ong, T6 gave the premium head rice yield while T7 had Grade 3.

- Gal-ong had medium size grains while Burdagol and PSB Rc82 were long grains (Table 8).
- Burdagol had intermediate shape while Gal-ong was bold and PSB Rc82 was slender (Table 9).
- The amylose content of the different samples was not affected by the fertilizer treatments (Table 10). Burdagol was classified as low amylose while Gal-ong and PSB Rc82 were both intermediate.

FERTILIZER MANAGEMENT	Fertilizer Rate		Brown rice (%, Class)					
	(kg ha-1)	Bur	dagol	Gal-ong	PSE	Rc82		
1. Control	0	75.9	F	73.9 P	76.1	F		
2. Omission Plot (-N)	0-42-42	76.5	F		76.7	F		
3. Omission Plot (-P)	112-0-42	77.2	F	73.1 P	77.0	F		
4. Omission Plot (-K)	112-42-0	76.9	F	74.9 P	78.0	F		
5. LCC-based 1	108-42-42	77.2	F	73.5 P	78.3	F		
6. LCC-based 2	118-42-42	76.0	F	74.4 P	77.9	F		
7. Rice straw	30-4-43 (4 t ha-1)	75.5	F	76.6 F	77.2	F		
8. Chicken Manure	55-129-116 (3 t ha <sup>-1</sup>	<sup>)</sup> 76.4	F	75.2 F	77.7	F		

**Table 5.** Brown rice recovery of the different varieties as influenced by fertilizer treatments, (2014 DS), San Nicolas, Ilocos Norte.

F - Fair; P - Poor

**Table 6.** Milled rice recovery of the different varieties as influenced by fertilizer treatments, (2014 DS), San Nicolas, Ilocos Norte.

FERTILIZER	Fertilizer Rate	Milled rice (%, Class)					
MANAGEMENT	(kg ha-1)	Burg	dagol	Gal-ong	PSE	Rc82	
1. Control	0	69.8	G1	66.8 G1	70.8	Pr	
2. Omission Plot (-N)	0-42-42	70.5	Pr		71.4	Pr	
3. Omission Plot (-P)	112-0-42	69.9	G1	71.1 Pr	71.3	Pr	
4. Omission Plot (-K)	112-42-0	69.9	Gl	69.9 G1	72.3	Pr	
5. LCC-based 1	108-42-42	70.3	Pr	65.7 G1	71.5	Pr	
6. LCC-based 2	118-42-42	69.1	G1	68.0 G1	71.4	Pr	
7. Rice straw	30-4-43 (4 t ha-1)	69.4	G1	70.4 Pr	70.1	Pr	
8. Chicken Manure	55–129–116 (3 t ha						
	1)	70.6	Pr	67.9 G1	71.9	Pr	

Pr - Premium; G1- Grade 1

FERTILIZER MANAGEMENT	Fertilizer Rate	Head rice (%, Class)					
	(kg ha⁻1)	Bur	dagol	Gal-ong	PSE	8 Rc82	
1. Control	0	30.9	G3	46.0 G2	49.9	G1	
2. Omission Plot (-N)	0-42-42	35.0	G3		48.5	G1	
3. Omission Plot (-P)	112-0-42	39.9	G2	48.5 G1	55.2	G1	
4. Omission Plot (–K)	112-42-0	40.0	G2	48.2 G1	54.2	G1	
5. LCC-based 1	108-42-42	31.5	G3	50.8 G1	56.4	G1	
6. LCC-based 2	118-42-42	40.0	G2	57.9 Pr	54.1	G1	
7. Rice straw	30-4-43 (4 t ha <sup>-1</sup> )	36.6	G3	38.7 G3	50.6	G1	
8. Chicken Manure	55-129-116 (3 t ha-	<sup>1)</sup> 33.0	G3	45.8 G2	53.5	G1	

**Table 7.** Head rice recovery of the different varieties as influenced by fertilizer treatments, (2014 DS), San Nicolas, Ilocos Norte.

Pr - Premium; G1 - Grade 1; G2 - Grade 2; G3 - Grade 3

**Table 8.** Grain length of the different varieties as influenced by fertilizer treatments, (2014 DS), San Nicolas, Ilocos Norte.

FERTILIZER	Fertilizer Rate	Grain Length (mm)					
MANAGEMENT	(kg ha-1)	Bur	dagol	Ga	l-ong	PS	B Rc82
1. Control	0	6.9	L	5.6	М	6.7	L
2. Omission Plot (-N)	0-42-42	6.7	L	-	-	6.7	L
3. Omission Plot (-P)	112-0-42	7.0	L	5.5	М	6.7	L
4. Omission Plot (-K)	112-42-0	6.8	L	5.6	М	6.9	L
5. LCC-based 1	108-42-42	6.7	L	5.7	М	6.9	L
6. LCC-based 2	118-42-42	6.8	L	5.7	М	6.8	L
7. Rice straw	30-4-43 (4 t ha-1)	6.7	L	5.6	М	6.8	L
8. Chicken Manure	55-129-116 (3 t ha <sup>-1</sup>	<sup>)</sup> 7.0	L	5.7	М	6.7	L

L - long; M - medium

**Table 9.** Grain shape of the different varieties as influenced by fertilizer treatments, (2014 DS), San Nicolas, Ilocos Norte.

FERTILIZER	Fertilizer Rate	Grain Shape (L/W)					
MANAGEMENT	(kg ha-1)	Bur	dagol	Ga	l-ong	PS	B Rc82
1. Control	0	2.8	I	1.8	В	3.3	S
2. Omission Plot (–N)	0-42-42	2.7	I	-	-	3.2	S
3. Omission Plot (-P)	112-0-42	2.8	I	1.7	В	3.3	S
4. Omission Plot (–K)	112-42-0	2.7	I	1.7	В	3.3	S
5. LCC-based 1	108-42-42	2.6	I	1.8	В	3.3	S
6. LCC-based 2	118-42-42	2.7	I	1.7	В	3.3	S
7. Rice straw	30-4-43 (4tha-1)	2.7	I	1.8	В	3.4	S
8. Chicken Manure	55-129-116 (3tha-1)	2.8	I	1.8	В	3.2	S

S - Slender; I - Intermediate; B - Bold

FERTILIZER	Fertilizer Rate	Amylose Content (%, Class)				
MANAGEMENT	(kg ha-1 )	Bure	dagol	Gal-ong	PSB Rc82	
1. Control	0	15.7	L	18.2 I	22.0 I	
2. Omission Plot (-N)	0-42-42	16.8	L		22.2 I	
3. Omission Plot (-P)	112-0-42	15.0	L	18.3 I	22.3 I	
4. Omission Plot (-K)	112-42-0	16.0	L	18.9 I	23.1 I	
5. LCC-based 1	108-42-42	16.2	L	18.2 I	21.2 I	
6. LCC-based 2	118-42-42	17.2	L	18.7 I	22.8 I	
7. Rice straw	30-4-43 (4 t ha-1)	16.5	L	19.4 I	22.2 I	
8. Chicken Manure	55–129–116 (3 t ha	-				
	1)	15.9	L	18.3 I	22.2 I	

Table 10. Amylose content of the different varieties as influenced by fertilizer treatments, (2014 DS), San Nicolas, Ilocos Norte.

Health-Promoting Phytochemicals

There was a highly significant interaction effect of fertilizer treatment and variety on the DPPH scavenging activity of rice (Table 11). T5 (LCC-based 1) had the highest DPPH (2,2-diphenyl-1-picrylhydrazyl) scavenging activities in Burdagol and PSB Rc82. For Gal-ong, T3 (-P) had the highest DPPH scavenging activity. More basic understanding will be required to explain treatment differences across rice varieties for the radical scavenging activities.

FERTILIZER Fertilizer Rate DPPH Scavenging Activity (%) MANAGEMENT (kg ha-1) Burdagol PSB Rc82 Gal-ong 1. Control 0 17.79 c 63.17 b 18.94 ab 2. Omission Plot (-N) 0-42-42 58.67 c 16.02 d 19.23 ab 3. Omission Plot (-P) 112-0-42 20.09 ab 65.30 a 18.42 bc 4. Omission Plot (-K) 112-42-0 17.86 c 55.62 d 19.86 ab 5. LCC-based 1 108-42-42 21.12 a 62.60 b 20.50 a 6. LCC-based 2 118-42-42 20.78 a 50.72 e 19.58 ab 7. Rice straw 30-4-43 (4 t ha-1) 16.02 d 57.64 c 18.77 b 8. Chicken Manure 55-129-116 (3 t ha-1) 18.54 bc 55.68 d 16.76 c Significance \*\*\* Fertilizer Variety \*\*\* \*\*\* Fertilizer x Variety

Table 11. DPPH (2,2-diphenyl<sup>-1</sup>-picrylhydrazyl) radical scavenging activity, an antioxidant assay for the different rice varieties grown under different fertilizer treatments in 2014 dry season, San Nicolas, Ilocos Norte.

\*\*\*significant at 0.1% level. Means followed the same letter are not significantly different at 0.5% level of significance

#### Validation of Harvest and Postharvest Key Checks

- The improved Key Check 8 (harvest management) and the proposed Key Check 9 (postharvest management) were validated by comparing the recommended technologies to that of the farmers' practice (Table 12). Some of the recommended technologies under the two Key Checks were followed by the farmers but not on drying. The farmers dried their grains up to 9% moisture content (MC), that was lower than the recommended 12 to 14% MC by gravimetric method.
- The overdrying of paddy had significant effect on the final weight of the dried paddy and milling potentials of milled rice. PhilRice samples had higher brown rice, milling and headrice recoveries (Figures 13 to 15). The headrice of the rice samples of the farmers were even below the standards.

**Table 12.** Harvest and postharvest management following recommendedtechnologies (PhilRice Batac) vs. farmers' practice.

PhilRice BatacFarmers' PracticeKeycheck 8: Cut, piled, and threshed the crop at the right time; used laboratory mechanical thresherCut, piled, and threshed the crop at the right time; used motorized pedal-type thresherProposed and qualitySPostharvest sanagementCut, piled, and threshed the crop at the right time; used motorized pedal-type thresher9a. Paddy sorted according to variety type and qualityPaddy sorted according to variety type and quality9b. Dried paddy with good quality (Dried paddy immediately after threshing; grains were spread 2-4 cm thick and stirred every 30 min)Dried the paddy after 1-3 days after threshing. Sundried for 2-3 days; 3-6 times stirring per day. Overdried grains: 9% MCwb.9c. Paddy with premium purity: cleaned paddy using blowerCleaned paddy using electric fan9d. Market quality preserved and losses to pests prevented during storage: used pallets and sacks that are free from residual infestationUsed sacks after drying inside the house9e. Milling and eating quality preserved and losses to pests prevented: ensured that the area is secure; exposed or aerated newly- dried grains for about 4-5 hr before covering them.Single-pass milling machines owned by private rice millers9f. Maximized milling and headrice recovery: used machine that can produce at least 65% milling recovery and 80% head rice on milled rice basisSingle-pass milling machines owned by private rice millers	0 .	1
crop at the right time; used laboratory mechanical thresherright time; used motorized pedal-type thresherProposed managementKeycheck 9:Postharvest managementPaddy sorted according to variety type and quality9a. Paddy sorted according to variety type and qualityPaddy sorted according to variety type and qualityPaddy sorted according to variety type and quality9b. Dried paddy with good quality (Dried paddy immediately after threshing; grains were spread 2-4 cm thick and stirred every 30 min)Dried the paddy after 1-3 days after threshing. Sundried for 2-3 days; 3-6 times stirring per day. Overdried grains: 9% MCwb.9c. Paddy with premium purity: cleaned paddy using blowerCleaned paddy using electric fan9d. Market quality preserved and losses to pests prevented during storage: used pallets and sacks that are free from residual infestationUsed sacks during storage9e. Milling and eating quality preserved and losses to pests prevented: ensured that the area is secure; exposed or aerated newly- dried grains for about 4-5 hr before covering them.Single-pass milling machines owned by private rice millers9f. Maximized milling and headrice recovery: used machine that can produce at least 65% milling recovery and 80% headSingle-pass milling machines owned by private rice millers	PhilRice Batac	Farmers' Practice
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Proposed managementKeycheck 9:Postharvest management9a. Paddy sorted according to variety type and qualityPaddy sorted according to variety type and quality9b. Dried paddy with good quality (Dried paddy immediately after threshing; grains were spread 2-4 cm thick and stirred every 30 min)Pried the paddy after 1-3 days after threshing. Sundried for 2-3 days; 3-6 times stirring per day. Overdried grains: 9% MCwb.9c. Paddy with premium purity: cleaned paddy using blowerCleaned paddy using electric fan9d. Market quality preserved and losses to pests prevented during storage: used pallets and sacks that are free from residual infestationUsed sacks during storage9e. Milling and eating quality preserved and losses to pests prevented: ensured that the area is secure; exposed or aerated newly- dried grains for about 4-5 hr before covering them.Piled sacks after drying inside the house9f. Maximized milling and headrice recovery: used machine that can produce at least 65% milling recovery and 80% headSingle-pass milling machines owned by private rice millers	crop at the right time; used laboratory	right time; used motorized pedal-type
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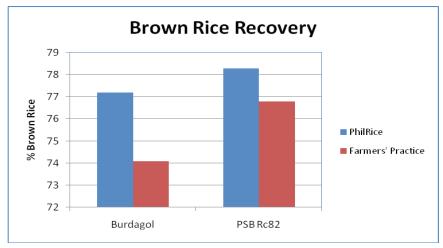


Figure 13. Brown rice recovery of Burdagol and PSB Rc82 comparing PhilRice and farmers' practice.

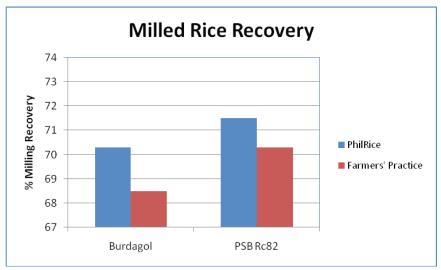


Figure 14. Milled rice recovery of Burdagol and PSB Rc82 comparing PhilRice and farmers' practice.

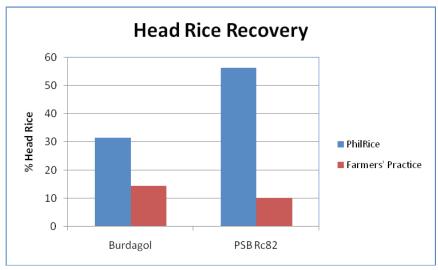


Figure 15. Head rice recovery of Burdagol and PSB Rc82 comparing PhilRice and farmers' practice.

B. Wet Season 2014

Yields were obtained from the field experiments conducted in San Nicolas, Ilocos Norte and Los Baños, Laguna. The highest yields were obtained from potassium omission plot (-K, +N, +P) indicating that soil K was not limiting. Lowest yields were obtained from treatments in the Control or zero fertilizer, -N, and organic fertilizers. Gal-ong consistently had the lowest yield among the three varieties tested, indicating that the low soil N status or the low level of N in the organic fertilizers reduced rice yields.

# **II.** High-value products from the rice grain and other parts of the rice plant

Project Leader: APP Tuaño

PhilRice's Natural Products and Value-adding Systems Program (NVP), with rice and rice-based food products as focus, has recently been expanded to include investigations on the rice grain, other parts of the rice plant, rice cell and organ cultures and other components of the rice environment as potential sources for the generation of high-value products. Technologies, methods, processes and product prototypes for functional foods, nutraceuticals, industrially-useful materials, agricultural and veterinary products are being explored to help address the national concern of minimizing the incidences of poverty and malnutrition. Hence, the High-value Products from Rice and its Environment Program (HVPRE) has been developed – one of the five new R&D programs of PhilRice for its 5-year R&D strategic plan for 2013 to 2017.

Rice has been cultivated in the country mainly as source of food. However, its potential as source of industrially important compounds (macromolecules and secondary metabolites) has not yet been fully investigated in the country. This is due to the main interest of the public on the rice grain, particularly as polished/milled rice, that consists of around 80 to 90% starch serving as primary source of dietary caloric energy needed by Filipinos (Juliano 2010). This project aims to explore the potential of the rice grain and its processing by-products, rice plant parts and rice cell and organ cultures as sources of biomolecules such as proteins, peptides, carbohydrates and secondary metabolites like lignans, terpenoids, flavonoids, antioxidants, anthocyanins, phytic acid and other bioactive components having nutritional, biomedical, health-related and industrial applications. It includes survey, screening and characterization of rice cultivars (modern, traditional and wild rices), their plant parts and cell and organ cultures derived from them, in order to determine best sources of the compounds of interest geared towards relevant characterization of their biological activities.

# Rice Plant Lignans, Terpenoids and Flavonoids with Health-Related and Industrial Applications

APP Tuaño, HP Hernandez, VC Lapitan, RB Valdez, EV San Gabriel

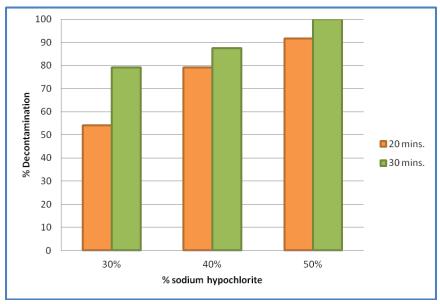
The science of drug discovery has been dependent on natural products researches that mainly involve extraction, isolation, purification and characterization of secondary metabolites from various organisms, particularly from plants. Secondary metabolites like lignans, terpenoids, flavonoids, other phenolic compounds, saponins, steroids, their derivatives and their conjugates have been studied in relation to their bioactivities and applications to biomedicine and pharmacology. In recent years, the advances in plant tissue culture made possible the production of a wide variety of beneficial secondary metabolites at significantly huge amounts.

Plant tissue culture is the process of in vitro production of plant using cells, tissues and other organs under aseptic conditions with specific nutritional and environment requirements. This system will allow us to explore the potential of the rice cell and organ cultures as sources of secondary metabolites with pharmaceutical and health-related applications such as lignans, flavonoids and terpenoids. This undertaking will aid drug discovery research in the country with the potential of helping the rice farming communities widen their economic opportunities from the rice plant parts/rice cultures as sources of industrially important metabolites.

# Highlights:

A. Decontamination of explants

Seeds of NSIC Rc240 were decontaminated. The highest decontamination percentage of 100% was obtained from seeds treated with 50% bleach soaked for 30 minutes (Figure 16). All the seeds survived and germinated when cultured on Murashige and Skoog (MS) medium with or without growth hormones.





#### 32 Rice R&D Highlights 2014

• The above protocol was utilized for aseptic germination of five selected pigmented traditional rice varieties with medicinal properties based on PhilRice gene bank database.

#### B. Callus Induction

• Only the culms and roots of the four week-old aseptic seedlings of the five pigmented varieties were utilized as sources of explants for callus induction (Figure 17). The leaves were no longer used since previous results showed that most of the leaf explants turned brown and eventually died within two weeks after culture.

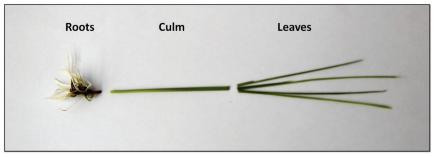


Figure 17. A four week old seedlings cut into three parts: roots, culm and leaves.

Callus induction of both explants was enhanced by the addition of dichlorophenoxyacetic acid (2,4-D) and combination of other growth hormones such as kinetin, 6-benzylaminopurine (BAP), and 1-naphthaleneacetic acid (NAA) in the MS basal media. However, between the two explants, the roots induced more calli than culms, thus in the succeeding experiment, roots were used for callus induction of the five pigmented varieties on different media (Figure 18). On the average, the medium supplemented with NAA (Medium 3) produced the highest percent callus induction followed by kinetin-supplemented medium (Medium 2) while the medium with BAP (Medium 4) had the lowest percent callus induction.

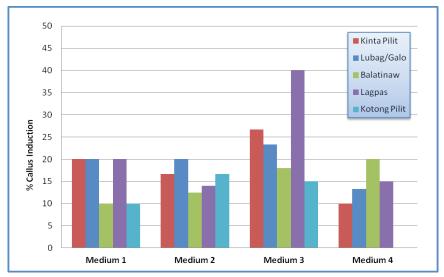
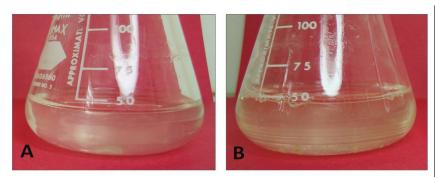


Figure 18. Percent callus induction of 5 pigmented/medicinal varieties using root explants on different induction media. Medium 1 - MS basal salt + 1mg/L 2,4-D; Medium 2 - MS basal salt + 1mg/L 2,4-D + 0.5 mg/L Kinetin; Medium 3 - MS basal salt + 1mg/L 2,4-D + 0.5 mg/L NAA; Medium 4 - MS basal salt + 1mg/L 2,4-D + 0.5 mg/L BAP.

- Callus induction was observed to be genotype-dependent. The response varied among genotypes across media. The sample Lagpas had the highest callus induction response in Medium 3 but not in other media. Kinta Pilit and Lubag/Galo had consistently good responses in Medium 1, 2, and 3 while Balatinaw, the lowest performing genotype in these three media, had the highest response in Medium 4.
- The type of calli obtained from this experiment using root explants was mostly embryogenic and were used for the establishment of cell suspension culture.
- C. Cell Suspension Culture.
  - Cell suspension cultures were established by transferring 5.0g of embryogenic calli to 50mL N6 liquid media. These were supplemented with 2,4-D and kinetin and treatments with and without proline were compared. Media were incubated on rotary shaker (180rpm) in the growth room. Addition of proline in liquid media was observed to produce more cells. The suspension culture became turbid two weeks after culture and the calli disintegrated into small sizes (Figure 19). Cell

culture samples (50mL) were submitted to the Rice Chemistry and Quality Laboratory for extraction of bioactive metabolites.



**Figure 19.** Cell suspension culture: (A) 0 week after subculture (B) 2 weeks after subculture.

- Subculture was done every two weeks where 10ml of suspension culture was taken from the old culture and transferred to a new 50ml medium with the same composition.
- D. Flavonoids from Mestiso 20 Plant Parts
  - Roots, stems and leaves of Mestiso 20 were air-dried, ground and extracted with hexane and methanol, separately. Yields of hexane extracts from various plant parts ranged from 0.46% to 1.89% while methanol extraction yields ranged from 8.06% to 15.16%. Highest extraction yields were noted using rice leaves in both hexane and methanol. Total phenolic content of leaf methanolic extract was 25.7mg gallic acid equivalent (GAE)/g extract while total flavonoid content was 89.7%mg quercetin equivalent/g extract.
  - Column chromatography followed by thin-layer chromatography showed promise of separation and purification of the components of the leaf methanolic extract using ethyl acetate-hexane solvent system (Figure 20).

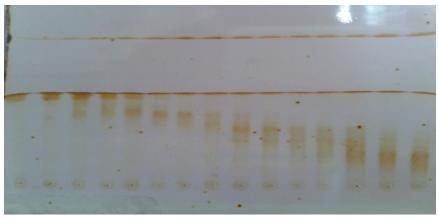
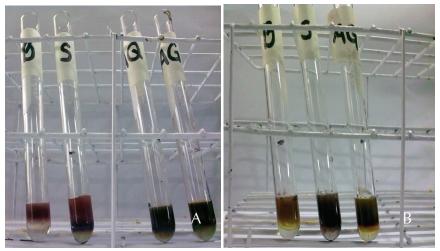


Figure 20. Chromatographic profile of fractions 1 to 15 using 2:3 ethyl acetate-hexane as mobile phase.

• Previous methanol-eluted fractions that might contain putative flavonoid glycosides were hydrolyzed using dilute HCl and tested for the presence of cleaved carbohydrate portion. After acid hydrolysis, the flavonoid aglycone (AG) showed an unclear interface (Figure 21A) but thoroughly washed AG showed no purple interface (Figure 21B) indicating the absence of carbohydrate moiety in the sample. The aqueous layer after hydrolysis showed the presence of carbohydrate (purple interface) through the Molisch test.



**Figure 21.** Molisch test on flavonoid aglycone hydrolysate (AG), aqueous layer after acid hydrolysis (AQ), blank (B) and sugar standard (S). 6A. AQ and AG after acid hydrolysis; 6B. AG after thorough washings with distilled water.

Washed aglycone was confirmed to contain flavonoids with a positive result on Shinoda test (Figure 22). Similar strategy as presented above is ongoing using seedlings and mature plants of purple rice and Minaangan, a medicinal rice variety from the PhilRice gene bank.



**Figure 22.** Shinoda test result for AG. AG0 – without test reagent; AG1 – with test reagent. Solution turning into red means a positive test for flavonoids.

- E. Terpenoids Extraction from Mestiso 19 Seedlings.
  - Shoots of Mestiso19 seedlings were air-dried, ground and extracted with dichloromethane in 1:100 (g/mL) powder to solvent ratio. Crude extract yield was 2.12%. Preliminary column chromatography separation trials followed by thin-layer chromatography in various solvent systems is in progress.

# Survey, Isolation, Purification and Characterization of Bioactive Peptides with Anti-Hypertensive Activity from Rice.

MAO Torio, RC de Leon, APP Tuaño

Bioactive peptides are potential health-enhancing components for food and pharmaceutical applications. They can be helpful in preventing and treating lifestyle-related diseases such as cardiovascular disease, cancers, osteoporosis, stress and obesity. Due to their many applications, there is a need to find biologically active peptides of medicinal value from cheap and readily available sources. Their purification and sequence determination are required to investigate their potential uses and, thus, increase the commercial value of the plant sources.

This study focused on utilizing different variants of rice, namely non-waxy rice, glutinous rice, pigmented rice, and brown rice, rice plant parts and rice cell cultures as potential sources of bioactive peptides, particularly with anti-hypertensive activity.

#### **Highlights:**

- Crude proteins were extracted from milled rice, bran, leaves, seedlings, and cell suspension culture of different rice varieties. Hydrolysates were prepared from the crude and partially purified proteins.
- Rice bran proteins and hydrolysates from 10 varieties of rice were evaluated for antihypertensive activity. These were PSB Rc18, Rc10, NSIC Rc13, Rc160, Rc240, Mestiso 1, Mestiso 19, Mestiso 20, Improved Malagkit Sungsong 2, and Inipot Ibon. The percentage angiotensin I converting enzyme (ACE) inhibition of rice bran protein hydrolysates ranged from 61.4 to 82.9%, with the M19 peptic digest having the highest (82.9%).
- Peptic digest of crude proteins from NSIC Rc240 cell suspension culture showed high ACE inhibition activity (98.5%) at 41.0 µg/ml protein content. Peptic and tryptic digests of the soluble proteins from rice leaves inhibited the activity of ACE by 49.1% and 38.8%, respectively. Protein fractions from Mestiso 19 and Mestiso 20 milled rice digested using pepsin showed 33.0 to 55.0% ACE inhibitory activity.

## Extraction and encapsulation of antioxidants and proteins from rice bran for functional food and biomedical applications

RM Bulatao, JMM Asuncion, HM Corpuz, RV Manaois, HFMamucod, AVMorales

Last year, our country significantly increased its rice production to about 18.03 million metric tons, or 8.09% higher than the previous year (BAS, 2012). The significant increase in rice production translates to an increased production of agricultural by-product like bran. Rice bran is a lowvalued by-product of rice processing but a good source of useful ingredients and products. It contains substantial amount of hypoallergenic protein, dietary fiber, vitamins, minerals, and natural phytochemicals including lignans, phytosterols, higher alcohols,  $\gamma$ -oryzanol, tocopherols, tocotrienols, and phenolic compounds (Schimitz et al., 2008; Rao et al., 2010). These bioactive compounds are known to reduce serum cholesterol, decrease incidence of atherosclerosis, and possess antitumor properties (Jariwalla, 2001).

The bran, particularly from pigmented rice, is nutritionally superior as it contains higher amount of nutrients and phytochemicals compared to ordinary white rice. Pigmented rice bran contains special phenolic compounds like anthocyanins, that have anticancer, hypoglycaemic, and anti-inflammatory health benefits that inhibit cell proliferation, alter cycle progression, and initiate apoptosis in malignant cells (Jariwalla, 2001; Min et al., 2009). In this regard, utilization of nutritious rice bran as source of natural antioxidants and proteins could be one of the sustainable strategies to maximize the use of essential nutrients and phytochemicals that are usually removed during milling process. Furthermore, this cheap agricultural by-product when properly utilized as food ingredients might be a good response to the escalating malnutrition problems facing our country today. Thus, this study was intended to extract and characterize antioxidants and proteins from rice bran for functional food, pharmaceutical and biomedical applications.

#### Highlights:

A. Extraction and nanoencapsulation of anthocyanins

A total of twenty one pigmented rice varieties were collected from the local rice farmers in the provinces of Tarlac, Kalinga, Mt. Province, Cavite, Quezon, Northern Samar and North Cotabato. The control rice variety NSIC Rc160 was obtained from the local seed grower at the Science City of Muñoz, Nueva Ecija. After collection, rice samples were cleaned, dehulled, and polished to obtain the bran. The bran samples were characterized in terms of pericarp color, L\*a\*b\* values, proximate composition, and antioxidant properties.

> The bran recovery varied significantly from 5.2% to13.8%. Chor-choros produced the highest recovered bran while Dikdik yielded the lowest. NSIC Rc160 and Mismis had the lightest bran color among the samples as indicated by their very high L\* values (Table 13). The bran of Saluyaw and Chorchor-os had the reddest bran color while Mismis, Dikdik, and NSIC Rc160 had the yellowest bran color as denoted by their significantly higher a\* and b\* values, respectively. The bran of Ominio obtained the lowest L\*a\*b\* confirming that this variety had the darkest bran color among the samples.

Rice Bran Samples	Pericarp Color	L*	a*	b*
NSIC Rc160	Light brown	$64.5 \pm 0.1^{a}$	1.0±0.1 <sup>i</sup>	$15.6 \pm 0.1^{b}$
Binerhen	Light brown	$61.5 \pm 0.2^{b}$	$1.3{\pm}0.1^{hi}$	14.9±0.1c
Mimis	Light brown	$63.5\!\pm\!0.0^{ab}$	0.3±0.1 <sup>i</sup>	$16.9{\pm}0.0^{a}$
Dikdik	Light brown	57.2±0.9 <sup>c</sup>	$1.8{\pm}0.1$ <sup>h</sup>	$16.4{\pm}0.1^a$
Kamuros	Red	$50.4 \pm 0.1^d$	3.8±0.1 <sup>e</sup>	$13.5\!\pm\!0.0^d$
Malido red	Red	$51.0 \pm 1.3^{d}$	6.7±0.1 <sup>b</sup>	$13.1\!\pm\!0.1^{de}$
Kalinayan	Red	$42.7{\pm}0.4^{h}$	6.0±0.1c	$13.1\!\pm\!0.0^{de}$
Kasagpi	Red	46.8±0.1e	$6.5{\pm}0.2^{b}$	$12.5{\pm}0.2^{\text{ef}}$
Dinorado	Red	$46.0{\pm}1.4^{\rm f}$	$6.4{\pm}0.3^{bc}$	10.8±0.3g
Baysilanon	Red	$44.2{\pm}0.4g^h$	6.7±0.1 <sup>b</sup>	$12.1 \pm 0.1^{f}$
Besaya	Red	$52.0{\pm}0.0^{\text{d}}$	$6.3{\pm}0.1^{\text{bc}}$	$13.2 {\pm} 0.1^{d}$
Saluyaw	Red	$43.5\!\pm\!0.0^{gh}$	$7.6{\pm}0.0^{a}$	11.3±0.1g
Cho-chor-os	Red	$44.9{\pm}0.6^{\text{gh}}$	$7.4{\pm}0.0^{a}$	$12.0{\pm}0.1^{f}$
Ominio	Black	31.9±0.1m	$0.8 \pm 0.1^{i}$	$1.0 \pm 0.1^{1}$
Pirurutong	Black	37.4±0.1 <sup>kl</sup>	$2.9{\pm}0.2^{\text{fg}}$	$2.5{\pm}0.1^k$
Inipot-ibon	Grayish black	$45.2{\pm}5.2^{\text{fg}}$	$3.6{\pm}0.3^{e}$	$5.6{\pm}0.4^{i}$
Inatipan	Black	$38.0{\pm}0.6^{kl}$	$3.5{\pm}0.1^{e}$	$5.8{\pm}0.3^{i}$
Kotinaw	Black	$36.0\pm0.0^{I}$	$2.6{\pm}0.0^{g}$	4.4±0.1 <sup>i</sup>
Sinomay	Black	$45.3{\pm}0.0^{\text{fg}}$	$4.5{\pm}0.1^d$	$6.6{\pm}0.1^{h}$
lttum	Black	$40.9{\pm}0.6^{ij}$	$3.5 \pm 0.1^{e}$	$4.2{\pm}0.4^j$
Inugsa	Black	$39.4{\pm}0.1^{jk}$	3.8±0.1 <sup>e</sup>	5.7±0.1 <sup>i</sup>
Labitaris	Black	$37.1\!\pm\!0.0^{kl}$	$3.3{\pm}0.2^{\text{ef}}$	$4.5\!\pm\!0.3^i$

 Table 13. Pericarp color and L\*a\*b values of rice samples.

\*Means followed by the same letter within a column are not significantly different (p = 0.05) using Tukey's test.

Proximate analysis showed that Chor-chor-os, Kamuros, and Pirurutong had the highest protein content while Saluyaw had the lowest (Figure 23). The bran of Ominio had the highest ash and fat content while Kalinayan, Ominio, Dikdik, Ittum, and NSIC Rc160 had the highest crude fiber content. The highest carbohydrate content was noted in the bran of Dikdik while the lowest was recorded in Ominio.

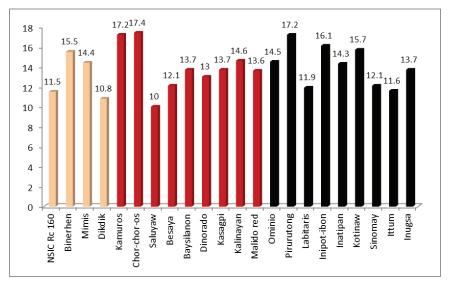


Figure 23. Crude protein content (%) of 22 rice bran samples.

The bran of black rice Ominio had the highest total anthocyanin and carotenoid contents among the samples (Table 14 and Figure 24, respectively). Anthocyanins in rice exist mainly in the form of cyanidin-3-glucoside and peonidin-3-glucoside (Abdel-Aal et al. 2006; Yawadio et al. 2007). The bran of Besaya had the highest total phenolic content while NSIC Rc160 had the lowest (Figure 25). The total antioxidant activity of pigmented rice bran samples ranged from 80.1 to 87.8%. The bran of Besaya and Malido red exhibited the strongest antioxidant activity among the samples (Figure 26). The bran of these varieties demonstrated greater capability of donating electrons to free radicals resulting in a more stable product, thus terminating series of radical chain reactions that may cause oxidative stress (Phetpornpaisan et al. 2014). In comparison with NSIC Rc160, all pigmented rice bran samples had considerably higher total antioxidant activity.

Rice Bran Samples	Anthocyanin Content (mg/Kg)
NSIC Rc160	554.5±23.7 <sup>h</sup>
Galo 1	4402.4±38.5 <sup>f</sup>
GaloMalagkit	10,722.7±291.8°
Inatipan	5,261.6±244.9e
Ittum	7,077.1±475.39 <sup>d</sup>
Kotinaw	7,094.8±185.9 <sup>d</sup>
Labitaris	3,455.7±182.9 <sup>g</sup>
Masbate	5,271.2±229.2°
Ominio	36,110.8±726.7ª
Palanqui	3,732.3±112.1 <sup>fg</sup>
Pirurutong	19,581.2±250.2 <sup>b</sup>

Table 14. Anthocyanin content of selected rice bran samples.

\*Means followed by the same letter within a column are not significantly different (p  $\leq$  0.05) using Tukey's test.

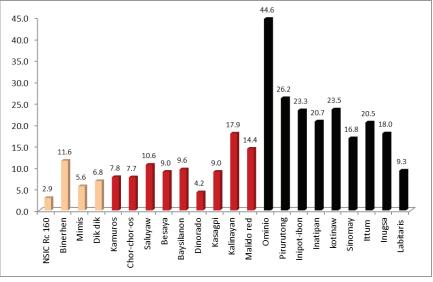


Figure 24. Carotenoid content ( $\mu$ g/g) of 22 rice bran samples.

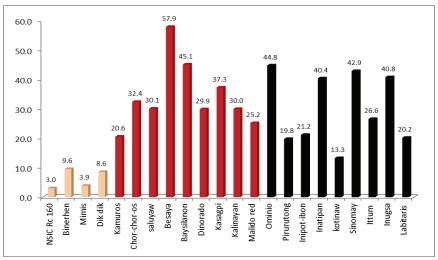


Figure 25. Total phenolic content (mg GAE/g) of 22 rice bran samples.

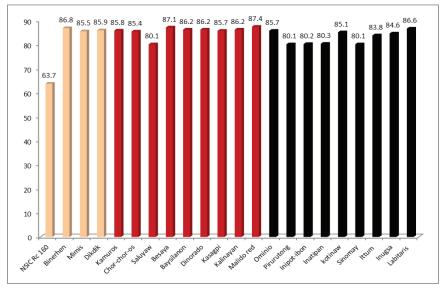


Figure 26. Total antioxidant activity (%) of 22 rice bran samples.

• Varying concentrations of acidified methanol and ethanol were used to extract anthocyanin fractions from the bran of Ominio (Figure 27). Results showed that absolute (100%) methanol was the most efficient solvent system in extracting anthocyanins. However, methanol is not recommended particularly for food use due to its toxicity to human and environment (Hue et al. 2013). Therefore, 85% acidified ethanol, which had comparable extracting efficiency with that of concentrated methanol, will be used in preparing rice bran extracts for food and biomedical applications.

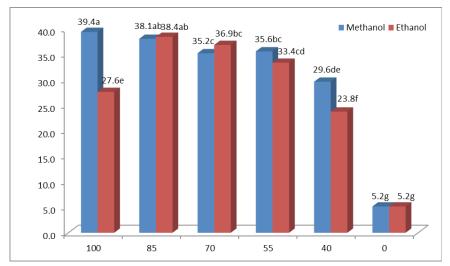


Figure 27. Anthocyanin content (g/kg) of methanolic and ethanolic rice bran extracts.

- The developed stabilized NSIC Rc160 bran contained 2.2% free fatty acid (FFA), which was below the acceptable range of FFA level (3%). Stabilized rice bran contained 11.4% ash, 22.0% fat, 14.2% protein, 12.5% fiber and 45.1% carbohydrates. Product development of rice bran supplemented- pandesal showed that different supplementations of bran (0%, 1%, 3%, 5%, 10%, and 15%) had no significant effects on the odor, aroma, off-odor, denseness, moistness, and tenderness of pandesal. However, increasing proportions of rice bran had minimal effects on the color intensity, texture, after-taste, mouthfeel and overall acceptability of pandesal. Sensory panelists preferred the control bread followed by 1%, 3%, and then 5% bran supplementation. Incorporation of bran up to 5% level was comparable with the control pandesal.
- Anthocyanin was then extracted using acidified ethanol from defatted Ominio bran that had the highest anthocyanin content. The extract was oven-dried, pulverized into powder and encapsulated using chitosan-alginate nanoparticles.

The anthocyanin nanocapsules were evaluated in terms of encapsulation efficiency, particle size, and surface morphology. Encapsulation efficiency of the prepared anthocyanin capsules was determined by obtaining the anthocyanin content of the unencapsulated (supernatant) and encapsulated anthocyanins (residue). Figure 28 shows the encapsulation efficiency of the prepared capsules. Among the different dosages, 30mg of anthocyanin extract had the highest encapsulation efficiency (68.9%) while 10 and 20mg of anthocyanin extract had the lowest with the efficiency of 56.9% and 56.3%, respectively. The low efficiency of the prepared capsules might be due to the leaching of anthocyanin during encapsulation process since it is water soluble. Water-in-oil emulsion is currently being explored to improve the efficiency of the prepared capsules. The surface morphology of the prepared anthocyanin capsules is shown in Figure 29.

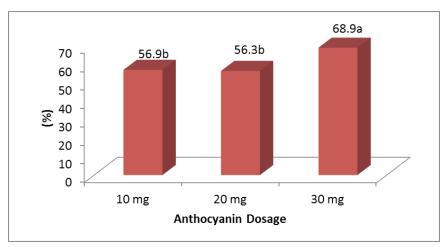
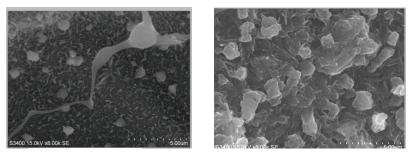
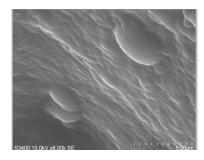


Figure 28. Encapsulation efficiency of the prepared anthocyanin nanocapsules.



10 mg Anthocyanin

20 mg Anthocyanin



30 mg Anthocyanin

Figure 29. Scanning electron micropgraphs of the prepared anthocyanin nanocapsules.

Ethanolic bran extracts of 5 black rices (Ominio, Pirurutong, GaloMalagkit,Ittum, and Kotinaw) and 3 red rice varieties (NSIC Rc19, Chor-chor-os, and Red blonde) were evaluated for the presence of secondary metabolites and antimicrobial activities. Results showed that all pigmented rice samples contained saponins, tannins, alkaloids, flavonoids, steroids, phenols, and sterols. Microbial analysis indicated that both black and red rice varieties exhibited bacteriostatic effect against Staphylococcus aureus, Pseudomonas aeruginosa, and Escherichia coli (Figure 30). Moreover, only Chor-chor-os and Ominio brans showed minimal bacteriostatic effect against Bacillus subtilis. All pigmented rice bran samples had no fungistatic activity against Aspergillusniger, Penicilliumcitrinum, Fusariumoxysposrum, and Trichodermaharzianum.

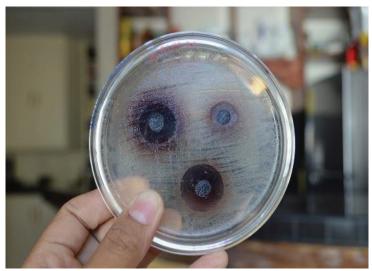


Figure 30. Zone of inhibition (mm) of pigmented rice bran samples.

#### B. Extraction and nanoencapsulation of gamma-oryzanol.

- Bran samples were collected from four modern varieties differing in amylose type. Bran of NSIC Rc160 produced the highest fat content, but did not differ significantly with that of NSIC Rc13 while NSIC Rc218 had the lowest (Figure 31). Hence, bran of NSIC Rc160 was used to extract rice bran oil for the nanoencapsulation of gamma-oryzanol.
- Purification of rice bran oil was carried-out based on the modified procedure of PhilRice (2007). Crude rice bran oil was subjected to degumming, alkali refining, bleaching, and winterization. The purified oil was encapsulated by cationic pre-gelation and polyelectrolyte complex formation using chitosan-alginate nanoparticles. The nanoencapsulated rice bran oils were submitted for the analysis of surface morphology using Scanning Electron Microscopy (Institute of Chemistry, UP Diliman) and particle size using zetasizer (Institute of Chemistry, UP Los Baños).

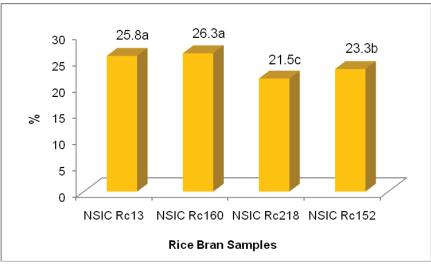


Figure 31. Crude fat content of rice bran samples from Philippine modern rice varieties.

- C. Extraction and nanoencapsulation of proteins
  - Freshly milled bran of NSIC Rc160 was collected and analyzed for protein content using standard Kjeldahl method (AOAC 2005). The protein content of the sample was 13.14%, which falls within the known value for rice bran (10 to 16%) as reported by Fabian and Ju (2011).
  - Alkali extraction was used to isolate proteins from the rice bran based on the modified method of Gnanasambandam and Hettiarachchy (1995). About 10g of sieved sample was weighed into a beaker and distilled water added at different dilution ratios (wt/vol), namely: T1=1:5; T2=1:10; and T3=1:1.5. Bran and water were mixed well and the pH of the mixture was adjusted to 9.5 using a 1.0N NaOH. The bran residues were separated by centrifugation at 8,000rpm for 30min. The bran residues were further clarified by vacuum filtration using a filter paper. The pH of the clarified filtrate was readjusted to 4.5 with 1.0N HCl and the filtrates were re-centrifuged using the same setting as described above. The supernatants were further filtered under vacuum. The collected residues (protein isolates) were sequentially washed with pH 4.5 and pH 7.0 distilled water. The filter paper containing the rice bran protein concentrate was then dried in an oven at 40°C for 18hr. The dried filter paper was weighed

and the % protein isolated was determined using the formula below:

Protein Isolated (PI) (%) = weight of protein concentrate (g) x 100 weight of bran sample (g)

• The protein recovered from the bran was calculated as shown below:

Protein Recovered (%) = <u>protein isolated (PI) x 100</u> crude protein content of bran

> Figure 32 shows the different protein concentrates extracted by alkaline treatment. Protein concentrate obtained from the bran dispersed in the lowest amount of water (T1) appeared to be less clumpy than the other treatments. More aggregation was observed in T3, but its amount did not differ from that obtained in T2 (Table 15). Protein isolated was the amount of protein extracted from the bran sample, and was thus determined by dividing the amount of protein concentrate by the amount of rice bran used for the extraction. The amount of protein isolated using T1 was 7.00% indicating that more water binding was required due to the large particle size of the bran sample. This was confirmed by Gnanasambandam and Hettiarachchy (1995) who reported that protein extraction becomes less efficient with increasing particle size. Hence, increasing the amount of water to 10 parts resulted in significantly higher yield (10.21%), but increasing it to 15 parts had no yield increment.

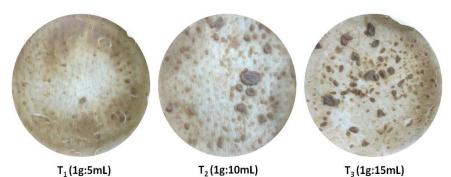


Figure 32. Protein isolates from rice bran dispersed in distilled water at different dilution ratios.

Treatment	Bran:Water (g/mL)	Protein Isolated (%)
TI	1:5	$7.00 \pm 0.37^{b}$
T2	1:10	$10.21 \pm 0.49^{a}$
Т3	1:15	$9.44 \pm 0.57^{a}$

Table 15. Protein isolated from rice bran at different dilution ratios.

Mean values with the same letter are not significantly different at p = 0.05.

Table 16 shows the protein recovered from rice bran at different dilution ratios. Protein recovered was defined as the amount of protein isolated divided by the total protein content of the bran sample. Similar trend was obtained with of the protein isolated, wherein higher amount of protein was recovered when bran was dispersed in at least 10 volumes of distilled water.

Table 16. Protein recovered from rice bran at different dilution ratios.

Treatment	Bran:Water (g/mL)	Protein Recovered (%)
TI	1:5	$55.95 \pm 2.92^{b}$
T2	1:10	$81.64 \pm 3.90^{a}$
Т3	1:15	$75.45 \pm 4.59^{a}$

Mean values with the same letter are not significantly different at p = 0.05.

## Prebiotics from Rice: Dietary Fiber, Resistant Starch and Xylooligosaccharides

HF Mamucod, RV Manaois, JEI Zapater

Prebiotics are among the functional food components that promote human health and wellness and help combat chronic degenerative diseases. Prebiotics are not digested and absorbed in the small intestines, signifying possible hypoglycemic effects. They ferment in the large intestines and are metabolized by resident beneficial microorganisms, resulting in improved colonic health (Birkett, 2000, 2008).

Resistant starch (RS) is a starch fraction that exhibits prebiotic properties. Among its reported beneficial effects in the body are reduction of plasma glucose and insulin levels, prevention of gastrointestinal diseases, and formation of short-chain fatty acids with health-promoting properties. Furthermore, it increases fecal bulk and supports the growth of beneficial microorganisms in the gut. Hence, the present study assessed the potentials of rice and its parts as sources of prebiotics and their utilization as nutraceuticals or functional food products.

### Highlights:

- Six rice varieties (NSIC Rc292, NSIC Rc11, NSIC Rc13, PSB Rc68, NSIC Rc186 and NSIC Rc188) were subjected to different heat-moisture treatments (conventional cooking, cooking + freezing, autoclaving-chilling and parboiling + cooking) for retrograded resistant starch (RS3) formation. RS3 levels of the samples were estimated using the optimized enzyme-gravimetric assay.
- Results showed that all varieties were classified as high gelatinization temperature or GT (79.15 to 80.79°C), except for the waxy NSIC Rc13, which had intermediate GT class (73.40°C) (Table 17).
- Analysis of the pasting properties of the six rice varieties showed that PSB Rc68 had the highest total setback in terms of viscosity profile or cP (i.e., 2083 cP), indicating its retrogradation potential. NSIC Rc13 was lowest with 427 cP.
- Conventional cooking produced RS3 with values ranging from 6.45 to 8.89%. Cooking + freezing gave considerably lower RS3 values ranging from 6.00 to 7.57%, except for NSIC Rc292 with 9.4% RS3. Autoclaving-chilling showed no significant effect among the rice varieties with extreme RS values of 4.28 to 11.67%. The highest RS3 levels were produced from the parboiling + cooking process. Five out of six rice varieties had RS values ranging from 9.65 to 10.58% (w/w).

Inole III Boladineador	temperature (GT) of the nee	samsiesi
Rice Variety	Gelatinization Temperature (°C) <sup>1</sup>	GT Type <sup>2</sup>
NSIC Rc292	79.74 $\pm$ 0.35 <sup>A</sup>	High
NSIC Rc11	$79.81 \pm 0.12$ <sup>A</sup>	High
NSIC Rc13	$73.40 \pm 0.28$ <sup>B</sup>	Intermediate
PSB Rc68	79.19 $\pm$ 0.54 <sup>A</sup>	High
NSIC Rc186	79.15 $\pm$ 0.28 A	High
NSIC Rc188	$80.79\pm0.26$ ^	High

Table 17. Gelatinization temperature (GT) of the rice samples.

<sup>1</sup>Mean values with the same letter are not significantly different at p = 0.05. <sup>2</sup>High (74.5-80°C), Intermediate (70-74.5 °C)

	Pasting Properties <sup>1</sup>							
Rice Varie ty	Peak Viscosit y (cP)	Trough Viscosit y (cP)	Breakd own (cP)	Final Viscosity (cP)	Setback (cP)	Peak Time (min)	Pasting Temperat ure (°C)	Retrograda tion Potential (cP)
NSIC Rc29 2	2278± 12 <sup>D</sup>	1609± 8 <sup>B</sup>	669±1 7 <sup>c</sup>	3461±3 0 <sup>BC</sup>	1184± 41 <sup>B</sup>	5.75±0. 04 <sup>A</sup>	80.33±0. 42 <sup>B</sup>	1853±23 <sup>c</sup>
NSIC Rc11	2506± 67 <sup>в</sup>	1594± 27 <sup>в</sup>	913±4 0 <sup>B</sup>	3530±4 1 <sup>B</sup>	1024± 26 <sup>c</sup>	5.53±0. 00 <sup>в</sup>	78.27±0. 03 <sup>c</sup>	1937±14 <sup>B</sup>
NSIC Rc13	2655± 42 <sup>A</sup>	1697± 13 <sup>A</sup>	958±2 9 <sup>ab</sup>	2125±2 0 <sup>E</sup>	– 530±2 2 <sup>E</sup>	4.53±0. 00 <sup>D</sup>	70.38±0. 08 <sup>E</sup>	427±8 <sup>D</sup>
PSB Rc68	2701± 27 <sup>A</sup>	1688± 12 <sup>A</sup>	1014± 16 <sup>A</sup>	3771±6 9 <sup>A</sup>	1070± 71 <sup>c</sup>	5.35±0. 04 <sup>c</sup>	76.70±0. 00 <sup>D</sup>	2083±65 <sup>A</sup>
NSIC Rc18 6	2387± 12 <sup>c</sup>	1491± 8 <sup>c</sup>	896±7 <sup>B</sup>	3226±1 7 <sup>D</sup>	839±9 <sup>D</sup>	5.51±0. 03 <sup>B</sup>	78.38±0. 03 <sup>c</sup>	1735±9 <sup>B</sup>
NSIC Rc18 8	2028± 57 <sup>E</sup>	1507± 21 <sup>c</sup>	521±1 7 <sup>D</sup>	3392±3 0 <sup>C</sup>	1364± 68 <sup>a</sup>	5.73±0. 07 <sup>A</sup>	81.18±0. 43 <sup>A</sup>	1885±37 <sup>BC</sup>

Table 18. Pasting properties of the rice samples.

<sup>1</sup>Mean values with the same letter are not significantly different at p = 0.05.

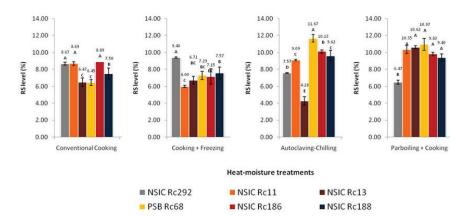


Figure 33. RS3 levels (%) of six rice varieties subjected to different heatmoisture treatments.

### III. Beneficial Organisms in the Rice Environment

Project Leader:Truong Hoai Xuan, PhD

There are many beneficial organisms in the rice environment like fungus and bacteria. Some species of fungus have pesticidal and stress tolerance properties which can be beneficial to its host plant. Other species of bacteria have nutritional properties and antibiotic properties. HVP-004 aims to: (1) assess the potential of endophytic fungi for improvement of stress-tolerance of rice, (2) assess the efficiency of microbial control agents as alternative pest control measure, (3) assess the efficiency of rhizotrophic and cyanobacteria as a substitute for commercial N fertilizer in rice production to reduce the environment problems, (4) assess the efficiency of Azolla as an alternative and renewable source of N and sequester carbon, and (5) assess the potential of of blue-green algae (BGA) as source of bioactive compounds and feed supplement.

#### Fungi with Pesticidal and Stress Tolerance

XH Truong

Drought and mineral toxicity are considered as the primary stresses that affect crop growth and development. One promising approach to improve crop plant productivity is to utilize the endophytic fungi. Such microbe-plant partnership known as "symbiosis", can improve the tolerance of host plants to a wide variety of stresses, including disease, drought, nutrient shortages, and extreme temperature. Remarkable symbiogenesis reported was the ability of Fusarium culmorum isolated from a weed species in Costal Beach Salt and Curvularia protuberata isolated from a weed species in geothermal soil to confer stress tolerance, as well as increase seed yields and root systems in Japonica and Indica rice varieties. These rice varieties and the weeds are genetically unrelated and came from different habitats. The rice plants provide the energy for the microorganisms through photosynthesis while the fungi or bacteria provide water and several nutrient elements for the plants in a natural way- without changing the genetic make-up of the host plants. This suggests that endophytic symbiosis can turn any susceptible varieties to be abiotic stress tolerant and fungal pathogen suppressor. Endophytes only colonize the vegetative tissues, but not in the seed embryo. However, the fungal endophytes are transmitted through the seed. Intensive research worldwide has proven that the endophytic and rhizospheric biota have multi-bioactivities and can be explored to produce agro-industrial products.

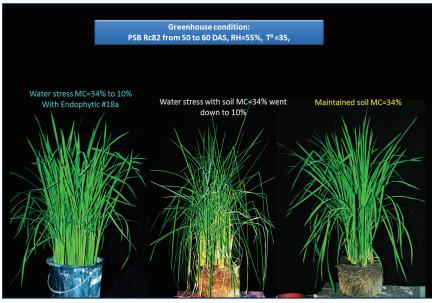
The general objective is to explore the mutualistic fungi in rice habitat that will enhance the tolerance of susceptible rice varieties to abiotic stresses and their secondary metabolites for plant growth promoting. Initially, target environments were characterized and plant samples were collected for isolation of endophytic fungi. Isolates were initially tested for drought tolerance using polyethylene glycol as the stress medium.

#### Highlights:

- Forty endophytic fungal isolates (EFI) were collected from rice plant, sedges, grasses, and broadleaves
- Among the EFI, four isolates such as Au4-1, Au8-6, Z4-2, and Z7-2 showed potential under induced drought tolerance.
- More plant samples were collected at different environments isolate EFI and evaluate rice drought tolerance, biocontrol of brown spot and leaf scald, and for nutrient management (P, Fe)..
  - Endophytic fungi may allow the symbiotic plants to activate stress response systems stronger than non-symbiotic plants and by triggering osmotic response (Redman et al. 1999) to maintain turgor and physiological processes. Endophytic #18a had positive results in terms of plant growth compared to the other isolates shown in Table 19. Total plant water potential = osmotic potential + turgor potential. Osmotic adjustment is the lowering of osmotic potential due to net accumulation of compatible solutes, i.e., amino acids, sugars and ions. With endophytic fungi isolate 18a, rice variety PSB Rc82 appeared to tolerate drought stress more than rice plant without fungi isolate (Figure 34).

Collection site	Endophytic Fungi isolate	Heat treatment (50 °C for 12hr)	Host plant	BCA test
15° 46' 09" LAT. N	Z4-2	+	Monocots	+
119° 55' 21" LON. E	18a	?	Dicots	-
24.12m ASL				
15° 22' 58" LAT. N	Au4-1	+	Monocots	+
121° 22' 46" LON. E	Au3-2	+	Monocots	+
24.69m ASL	Au7-1	?	Monocots	+
?= To be re-tested				

**Table 19.** Endophytic fungal isolates with biocontrol agent (BCA) potential and heat tolerance.



**Figure 34.** PSB Rc82 grown from 50 to 60 days after sowing in (a) soil with 10 to 34% misture content + endophytic fungi #18a, (b) soil with 10 to 34% moisture content and (c) soil with 34% moisture content.

## Cyanobacteria for nitrogen fixation, nutrient elements, and carbon sequestration

EH Bandonill, GA Nemeño, DLG Escañan, HM Corpuz, MB Dacumos, MR Martinez-Goss

Cyanobacteria is a phylum of bacteria that obtains its energy through photosynthesis. They are the most successful group of microorganisms on earth and most genetically diverse. Rice plantations utilize healthy populations of nitrogen-fixing cyanobacteria (Anabaena, as symbiotes of the aquatic fern Azolla) for use as rice paddy fertilizer. The endosymbiont, which is nitrogen-fixing, provides sufficient N for itself and its host (Peters, 1978). The fern on the other hand, provides a protected environment for the alga and also supplies it with a fixed carbon source (Peters, 1976; Van Hove, 1989). Under suitable conditions Azolla can double in weight every 3 to 5 days and fix N at a rate exceeding that of legume/Rhizobium symbiotic relationship and can accumulate 2 to 4kg of N ha-1 day-1 (Lumpkin and Plueknett, 1980). In addition, Azolla has enormous potential to sequester of atmospheric CO2 due to its rapid growth in freshwater without the need for a soil-based nitrogen source. Thus, this natural resource endemic in the area should be maximized as an alternative and renewable source of N that can supply the requirement of the rice crop, improve soil fertility in a sustainable way, and sequester C that can minimize the climate change.

The Philippines possess about 91 taxa of algae being used as food, namely red algae, green algae, brown algae and the cyanobacteria group or blue-green algae (BCA) (Martinez 1993). They are known to be rich in phycocyanin (PC), a photosynthetic pigment with antioxidant and antiinflammatory properties. They are also recognized for their protective effects against viral infections, cancer, allergy, diabetes, and hyperlipidemia. In other parts of the world, algae are already exploited. However, in the Philippines minimal number of strains is utilized as food or animal feed. With the vast potential of algae, collection and characterization of other BGA strains from different areas of the country is necessary. Moreover, extraction of their bioactive compounds as possible feed supplement will maximize their utilization and exploit this commodity present in the rice environment. Once farmers are able to mass produce the strain that will be collected and characterized from this study, this will help augment their economic status and contribute to the goal of the program that is to improve the well-being of rice-based farming communities and consumers.

#### **Highlights:**

Utilization of azolla for N fixation and carbon sequestration

- Collected Azolla pinnata in the locality and three strains in PhilRice Los Baños and mass-produced as planting material.
- Established Azolla nursery pond (on-station) for the maintenance of different Azolla strains (Figure 35a).
- Established the field experimental plots (on-station) for the evaluation of the potentials of Azolla to fix N and sequester C in the atmosphere (Figure 35b).
- Collected and air dried soil samples for processing prior to complete soil analysis.
- Scouted for farmers to be interviewed regarding the use of Azolla as biofertilizer in order to document farmers' acceptability of Azolla in reference to the previous program on Azolla or the National Azolla Action Program (NAAP).



**Figure 35.** Azolla nursery pond (a) and experimental plots for N fixation and C sequestration (b).

#### Blue-green algae as feed supplement

- Identified the collected BGA samples from PhilRice ponds and rice paddies from Pangasinan and described in terms of shape and the presence of reproductive cells (Table 20). Photos of algae strains are shown on Figure 36.
- The moisture content as dry basis ranged from 2.76% to 16.93%. HMC's culture had the highest crude protein content (47.38%) probably because it was cultured with the addition of urea as the probable source of nitrogen (Table 21). The lowest crude protein content was obtained from the Atrium sample, but had the highest amount of calcium (23,800mg/100g sample) and magnesium (610mg/100g sample). The crude ash of all algae samples ranged from 29.44% and 46.45%. Algae strain collected from Mindanao had the highest total phenolic content (64.28mg GAE/100g sample), sodium (4215mg/100g sample), and zinc (65mg/100g sample) among all algae samples collected. Vitamin E, thiamine, and total sugars were very low at <0.2, <0.3mg/100g sample, and <0.6g/100g sample, respectively for all the algae samples.

Sources	Description	Tentative Identification
Greenhouse	Filamentous, unbranched, cells cylindrical,	Rhizoclonium
	11–13 µ broad, 30–35 µ long; only	
	vegetative cells were observe d.	
Pangasinan	Cells ovate, 7-8 µ long, 6 µ broad,	Aphanothece pallida
Brown	scattered throughout gelatinous mass	(Kuetz.)Rabenh.
Atrium	Filamentous, cells 11 µ long, 4–5 µ broad;	Oedogonium
	no reproductive cells were not observed	
Pangasinan	Cells ovate 3 µ long, 2 µ broad, scattered	Aphanothece
Green	throughout gelatinous mass	<i>microscopica</i> Naeg.
HMC's Culture	Cells with cup-shaped chloroplast and	Chlamydomonas
	eyespot, spherical, 2 $\mu$ in dia; did not	<i>Dinobryonii</i> GM Smith
	observed any remnants of lorica of	
	Dinobryon. Hence, the id is tentative	
PhilRice Marker	Filamentous, unbranched, cells cylindrical,	Oedogonium
	4-5 μ broad, 11-15 μ long; did not	
	observe any reproductive cells	

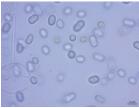
Table 20. Sources, description, and identification of collected algae strains.



Rhizoclonium

Rhizoclonium

Aphanothece pallida (Kuetz.)Rabenh



Aphanothece pallida (Kuetz.) Rabenh



Chlamydomonas



Dinobryonii Aphanothece microscopica Naeg.





Figure 36. Identity of collected algae strains.

microscopica Naeg.

			Reading		
	Atrium	Mindanao	Greenhouse	Pangasinan	HMC's Culture
Parameter					
Moisture, %	2.76	16.93	6.01	6.12	7.1
Crude Protein, %	5.28	18.82	15.51	15.11	47.38
Crude Ash, %	46.45	35.72	29.44	42.29	-
Crude Fiber, g/100g	3.6	3.4	10.9	-	-
Total Fat & Other	1.2	0.2	0.9	-	-
Solvent-Extractable					
Substances, g/100g					
Iron, mg/100g	152	223	1,000	-	-
Calcium, mg/100g	23,800	2,015	5,120	-	-
Magnesium, mg/100g	610	453	595	-	-
Zinc, mg/100g	7	65	23	-	-
Sodium, mg/100g	52	4,215	104	-	-
Vitamin E, mg/100g	< 0.2	< 0.2	< 0.2	-	-
Thiamine, mg/100g	< 0.3	< 0.3	< 0.3	-	-
Total Sugar, g/100g	< 0.6	< 0.6	< 0.6	-	-
Total Phenolic Content,	52.95	64.28	32.59	55.73	_
mg GAE/100 g sample					

**Table 21.** Nutritional composition and total phenolic content of collected algae strains.

- insufficient amount of sample

#### **Rhizotrophic Bacteria with Nutritional and Antibiotic Properties** *HXTruong*

Biofertilizers (BFs) can improve rice crop growth and reduce dependence of rice farmers on chemical fertilizers. However, it is important to assess the efficacy of biofertilizers in terms of nutritional and antibiotic properties with strict experimental control as well as the consistency in the production of the biofertilizers. This study assessed the identity of bacterial inoculants such as N-fixers, P and K-solubilizers, growth hormones and siderophores. Tests were done with the use of simple kits, then materials were characterized based on biochemical database.

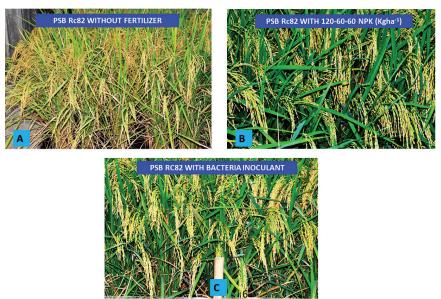
#### Highlights:

Biological Control Agent (BCA). Eighteen bacterial isolates from root zone of rice were characterized and in- vitro assayed against two major rice pathogens, bacterial leaf blight (BLB) and sheath blight (ShB).

Five Bacillus isolates belonging to B. subtilis and B. putilis inhibited the growth of ShB (Rhizoctonia solani). Of these, B. subtilis isolate S6-4 was the most potential BCA (inhibition zone 31mm or inhibition ratio of 3.4).

- Eight BCA isolates inhibited the growth of BLB pathogen. The isolate B. subtilis 184 is the most potential (inhibition zone 35mm).
- The isolate BCA isolate 192 inhibited both BLB and ShB with medium inhibition.
- Three isolates: B. subtilis S4-3, B. megaterium A, and Pseudomonas species S5-3 capable of IS, IAA production, and PS promoted rice plant growth.

Biofertilizer. Without fertilizer, yield of PSB Rc82 was 3.0 t/ha (Figure 37). With N-fixing bacteria, yield of PSB Rc82 was 4.0t/ha. With 120-60-60kg NPK/ha, yield of PSB Rc82 was 7.0t/ha.



**Figure 37.** (A) Grain yield of 3t/ha yield without fertilizer. (B) Grain yield of 7t/ha with the recommended fertilizer rates of 120-60-60kg NPK/ha. (C) Grain yield of 4t/ha with N-fixing bacteria at a rate of 20g powder/4 m<sup>2</sup>.

#### Abbreviations and acronymns

ABA – Abscicic acid Ac – anther culture AC – amylose content AESA – Agro-ecosystems Analysis AEW – agricultural extension workers AG – anaerobic germination AIS – Agricultural Information System ANOVA – analysis of variance AON – advance observation nursery AT – agricultural technologist AYT – advanced yield trial BCA - biological control agent BLB - bacterial leaf blight BLS – bacterial leaf streak BPH – brown planthopper Bo - boron BR - brown rice BSWM - Bureau of Soils and Water Management Ca - Calcium CARP - Comprehensive Agrarian Reform Program cav – cavan, usually 50 kg CBFM - community-based forestry management CLSU - Central Luzon State University cm - centimeter CMS - cystoplasmic male sterile CP - protein content CRH – carbonized rice hull CTRHC - continuous-type rice hull carbonizer CT - conventional tillage Cu – copper DA - Department of Agriculture DA-RFU - Department of Agriculture-**Regional Field Units** DAE - days after emergence DAS – days after seeding DAT - days after transplanting DBMS - database management system DDTK - disease diagnostic tool kit DENR - Department of Environment and Natural Resources DH L- double haploid lines DRR – drought recovery rate DS - dry season DSA - diversity and stress adaptation DSR - direct seeded rice DUST - distinctness, uniformity and stability trial DWSR – direct wet-seeded rice EGS - early generation screening EH – early heading

EMBI – effective microorganism-based inoculant EPI – early panicle initiation ET - early tillering FAO – Food and Agriculture Organization Fe – Iron FFA - free fatty acid FFP - farmer's fertilizer practice FFS - farmers' field school FGD – focus group discussion FI - farmer innovator FSSP – Food Staples Self-sufficiency Plan g – gram GAS - golden apple snail GC - gel consistency GIS - geographic information system GHG – greenhouse gas GLH - green leafhopper GPS - global positioning system GQ - grain quality GUI – graphical user interface GWS - genomwide selection GYT – general yield trial h – hour ha – hectare HIP - high inorganic phosphate HPL - hybrid parental line I - intermediate ICIS - International Crop Information System ICT - information and communication technology IMO - indigenous microorganism IF – inorganic fertilizer INGER - International Network for Genetic Evaluation of Rice IP - insect pest IPDTK – insect pest diagnostic tool kit IPM – Integrated Pest Management IRRI – International Rice Research Institute IVC - in vitro culture IVM - in vitro mutagenesis IWM - integrated weed management JICA – Japan International Cooperation Agency K – potassium kg – kilogram KP - knowledge product KSL - knowledge sharing and learning LCC – leaf color chart LDIS - low-cost drip irrigation system LeD – leaf drying LeR – leaf rolling lpa – low phytic acid LGU - local government unit

LSTD – location specific technology development m – meter MAS - marker-assisted selection MAT - Multi-Adaption Trial MC – moisture content MDDST - modified dry direct seeding technique MET – multi-environment trial MFE - male fertile environment MLM - mixed-effects linear model Mg - magnesium Mn - Manganese MDDST - Modified Dry Direct Seeding Technique MOET - minus one element technique MR - moderately resistant MRT – Mobile Rice TeknoKlinik MSE – male-sterile environment MT – minimum tillage mtha-1 - metric ton per hectare MYT – multi-location yield trials N - nitrogen NAFC - National Agricultural and Fishery Council NBS – narrow brown spot NCT – National Cooperative Testing NFA – National Food Authority NGO - non-government organization NE – natural enemies NIL – near isogenic line NM - Nutrient Manager NOPT - Nutrient Omission Plot Technique NR - new reagent NSIC – National Seed Industry Council NSQCS - National Seed Quality Control Services OF - organic fertilizer OFT - on-farm trial OM – organic matter ON - observational nursery OPAg – Office of Provincial Agriculturist OpAPA – Open Academy for Philippine Agriculture P - phosphorus PA - phytic acid PCR – Polymerase chain reaction PDW – plant dry weight PF - participating farmer PFS - PalayCheck field school PhilRice - Philippine Rice Research Institute PhilSCAT - Philippine-Sino Center for Agricultural Technology PHilMech - Philippine Center for Postharvest Development and Mechanization PCA – principal component analysis

PI - panicle initiation PN – pedigree nursery PRKB – Pinoy Rice Knowledge Bank PTD – participatory technology development PYT – preliminary yield trial QTL - quantitative trait loci R - resistant RBB - rice black bug RCBD – randomized complete block design RDI – regulated deficit irrigation RF – rainfed RP - resource person RPM - revolution per minute RQCS – Rice Quality Classification Software RS4D - Rice Science for Development RSO – rice sufficiency officer RFL - Rainfed lowland RTV - rice tungro virus RTWG – Rice Technical Working Group S – sulfur SACLOB - Sealed Storage Enclosure for Rice Seeds SALT – Sloping Agricultural Land Technology SB – sheath blight SFR - small farm reservoir SME - small-medium enterprise SMS - short message service SN - source nursery SSNM - site-specific nutrient management SSR – simple sequence repeat STK – soil test kit STR – sequence tandem repeat SV – seedling vigor t – ton TCN – testcross nursery TCP – technical cooperation project TGMS – thermo-sensitive genetic male sterile TN – testcross nursery TOT – training of trainers TPR – transplanted rice TRV - traditional variety TSS – total soluble solid UEM – ultra-early maturing UPLB – University of the Philippines Los Baños VSU – Visayas State University WBPH – white-backed planthopper WEPP – water erosion prediction project WHC – water holding capacity WHO - World Health Organization WS – wet season WT – weed tolerance YA – yield advantage Zn – zinc ZT – zero tillage

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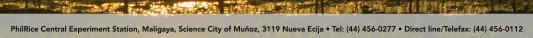
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