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RICE CHEMISTRY and FOOD SCIENCE DIVISION

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Rice Science and Food Chemistry Division

Division Head: Riza G. Abilgos - Ramos

Executive Summary

The team is tasked to conduct grain quality (GQ) evaluation of rice lines and varieties to assist breeders in identifying promising rice lines for entry into the National Cooperative Test. Advancement of the GQ assessment methods is also envisioned to upgrade capacity and fast-track varietal screening. Current development in analytical assessment of quality characteristics of rice and the demand for reliable data by heightened breeding activities further necessitate an immediate updating of grain quality techniques. In addition, optimization of new analytical methods for measuring elemental composition and secondary metabolites important in Philippine rice was made. This is to quantify micronutrients and nonnutrients important in health, nutrition, and food safety.

Study 1 of project 1 covers GQ evaluation of rice lines before they are elevated in the advance stages of the National Cooperative Test (NCT). Its purpose is to select promising entries to expedite the screening, thereby reducing cost and maximizing resources. GQ analyses of pre-NCT rice lines for irrigated (transplanted and direct seeded), hybrid, special purpose, and adverse ecosystems are centralized, with a few personnel strictly focused on the task to ensure repeatability and reliability of results. Study 2 evaluated causes of cooked rice spoilage in the Philippine setting and determine the best storage and reheating conditions that will extend the shelf life while keeping the quality of cooked rice.

For the study on grain quality screening, one thousand forty one (1,041) entries from 2013 dry to 2014 wet season under the Hybrid Rice Breeding and Genetics Development of Irrigated Lowland Rice Varieties projects of PBBD and Assessment of Soil Fertility, Plant, Water and Nutrient Management of ASPPD were screened for grain quality. These were distributed to UEM (92), AZU (22), AON (63), M6 (126), DSR-ON (150), LTOF (162), FAV-SUB (48), DAT (24), MYT (10), SV (17), SPSI (31), AON-UEM (109), SUB (196), and 37 (AON-Phytonutrient). The milling and physical qualities and apparent eating quality as predicted by the starch properties of the milled rice were evaluated.

Overall, grain quality evaluation revealed that 28 UEM, 2 AZU, 5 AON, 17 M6, 4 DSR-ON WS, 3 LTOF WS, 4 DAT, and 19 Fav-Sub WS entries; 2 MYT, 1 LTOF WS, 8 DSR-ON DS, 6 SV, 2 SPSI, and 8 LTOF DS entries have satisfied all the requirements for milling, physical, and apparent amylose as eating quality indicator as set by the Rice Varietal Improvement Group.

Results from study 2 revealed the rate of spoilage of popular rice varieties (Japonica, NSIC Rc160, Rc222, SL8, IR64, NFA). SL8 and NFA had perceptible odor after 21 h. SL8, Rc222, and NFA with higher optimum water requirement (1:1.9,1:1.6,1:1.6, respectively) had higher moisture content when freshly cooked and became more cohesive and more moist after 24 h. IR64 had lower microbial growth compared with PSB Rc10. Both IR64 and PSB Rc10 cooked with optimum cooking water. For the second project, three studies were implemented in 2014. For the study on optimization and application of microwave plasma-atomic emission spectroscopy (MP-AES), result showed that forty (40) polished rice samples collected from regions near mining and volcanic activities contain cadmium (Cd), copper (Cu), manganese (Mn), zinc (Zn), and lead (Pb). For the second study under Project 2, 136 rice samples were collected from four regions in Luzon and processed for isotopic fingerprints.

I. Grain Quality Assessment of Rice Lines and Varieties

Project Leader: EH Bandonill

Grain quality (GQ) evaluation is the final trial that rice lines undergo after meeting standards for yield, disease and insect resistance, agronomic and other traits. Study 1 of this project covers GQ evaluation of rice lines before they are elevated in the advance stages of the National Cooperative Test (NCT). Its purpose is to select promising entries to expedite the screening, thereby reducing cost and maximizing resources. GQ analyses of pre-NCT rice lines for irrigated (transplanted and direct seeded), hybrid, special purpose, and adverse ecosystems are centralized, with a few personnel strictly focused on the task to ensure repeatability and reliability of results.

The ultimate objective of the two studies of the project is to enhance breeding efficiency for grain quality and ensure availability of cooked rice with longer shelf-life for consumers. Thus, Study 2 tries to determine the causes of cooked rice spoilage in the Philippine setting and determine the best storage and reheating conditions that will extend the shelf life while keeping the quality of cooked rice.

Centralized Grain Quality Screening

NCRamos, CTEstonilo, JDAdriano

Grain quality screening (GQS) plays an important role in the rice breeding program of PhilRice. Aside from yield, resistance to pests and diseases, and agro-morphologic characteristics, grain quality, which includes milling recovery, physical attributes, physicochemical and eating properties completes the traits of a rice entry. Integration of GQS in the early rice

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breeding stages is essential as it trims down the number of lines advanced for further trials, thus reducing cost and maximizing resources. Furthermore, pre-NCT screening may also allow the identification of entries with properties suited for special purposes.

This year, one thousand forty one (1,041) entries from 2013 dry to 2014 wet season under the Hybrid Rice Breeding and Genetics Development of Irrigated Lowland Rice Varieties projects of PBBD and Assessment of Soil Fertility, Plant, Water and Nutrient Management of ASPPD were screened for grain quality. These were distributed to UEM (92), AZU (22), AON (63), M6 (126), DSR-ON (150), LTOF (162), FAV-SUB (48), DAT (24), MYT (10), SV (17), SPSI (31), AON-UEM (109), SUB (196), and 37 (AON-Phytonutrient). The milling and physical qualities and apparent eating quality as predicted by the starch properties of the milled rice were evaluated.

Highlights:

About 228 entries were analyzed for grain quality and phytonutrient such as antioxidant activity and total anthocyanin content.

- Majority of the entries (53.4%, N=751) met the total milled rice recovery standard of grade 1 to premium, and 82.6% (412 entries) passed the head rice recovery requirement (Table 1). From each varietal development stage, 50 entries from UEM, 1 from AON, 33 from DSR-ON WS, 32 from LTOF DS, 21 from DSR-ON DS, 16 from SV, 3 from SPSI, 3 from LTOF WS, 8 from DAT, and 2 entries from MYT passed the standards for milling quality. However, no entry passed from LTOF DS, A2U and M6. Samples from Favorable Submerged Study were not subjected to milling potential evaluation.
 - Majority of the entries were found to have a preferred dimension of long grain (522 entries) and slender shape (454 entries). In addition, there were 470 entries (62.6%) which passed the requirement for chalkiness (≤5%) level (Table 1). Sixty (60) entries from UEM, 3 from AON, 1 each from AZU, LTOF WS and M6, 12 from DSR-ON WS, 17 from LTOF, 35 from Fav-Sub, 22 from LTOF DS, 3 from SPSI, 33 from DSR-ON DS, and 11 entries from SV have satisfied the chalkiness requirement of less than 5%. However, no entry from MYT passed. Entries from DAT WS were not evaluated for chalkiness as this parameter was not required by breeders.
- Majority (98%) from each varietal development stage met the preferred low to intermediate amylose content of 12-25% (Table 1). One (1) entry from MYT WS was identified as waxy or glutinous. Majority of the entries (82%) were of

intermediate to high-intermediate GT type ($70^{\circ}C - 74^{\circ}C$ GT). Only 7 entries (1 entry each for UEM and M6 and 5 from SPSI) were identified with high GT type (74.5 to 80°C GT). About 118 entries of the remaining samples were of low GT type (<70oC GT).

- Overall, grain quality evaluation revealed that 28 UEM, 2 AZU, 5 AON, 17 M6, 4 DSR-ON WS, 3 LTOF WS, 4 DAT, and 19 Fav-Sub WS entries; 2 MYT, 1 LTOF WS, 8 DSR-ON DS, 6 SV, 2 SPSI, and 8 LTOF DS entries have satisfied all the requirements for milling, physical, and apparent amylose as eating quality indicator as set by the Rice Varietal Improvement Group.
- In December 2014, about 62 samples from AON-UEM for 2014 WS were received and grain quality evaluation is still on-going.

Grain Quality Property		Number of Entries				
			A2U	AON	M6	
Parameter/Level/Classification		2013 D S	2013 DS	2013 DS	2013 DS	
			(N=22)	(N=63)	(N⊨126)	
Milling Recover	y and Physical Attributes					
Brown Rice						
>80%	Good	16	1	12	15	
75.1-79.9%	Fair	64	11	19	41	
<75%	Poor	12	10	44	30	
Milled Rice						
>70.1%	Premium	54	1	6	25	
65.1-70.0%	Grade 1	34	14	39	53	
60.1-65.0%	Grade 2	4	5	16	29	
55.5-60.0%	Grade 3	2	2	2	19	
<55.5%	Below Standards					
Head Rice						
>57%	Premium	10	101		<u>.</u>	
48.0-56.9%	Grade 1	43	1	3	1	
39.0-47.9%	Grade 2	34	5	47	20	
30.0-38.9%	Grade 3	4	6	11	36	
<30.0%	Below Standards	1	10	2	69	
Chalkiness						
0.1-2.0%	Premium	19			24	
2.1-5.0%	Grade 1	41	6	14	43	
5.1-10.0%	Grade 2	24	13	12	39	

Grain Quality Property		Number of Entries			
Parameter/Level/Classification		UEM	A2U	AON	M6
		2013 D S	2013 DS	2013 DS	2013 DS
		(N=92)	(N⊨22)	(N=63)	(N ⊨ 126)
101-15.0%	Grade 3	7	1	2	14
>15.0%	Below Standards	1	2	2	3
Grain Length					
>7.5mm	Extra Long	2	1		17
6.4- 7.4mm	Long	64	5	24	70
5.5- 6.3mm	Medium	26	8	39	39
<5.4mm	Short	2	8	2	2
Grain Shape					
>3	Slender	60	8	3	82
2.0- 3.0	Intermediate	32	4	60	44
<2.0	Bold	2	10	2	2
Physicochemical P	Properties				
Amylose Content					
0.0- 2.0%	Waxy	2	2	2	2
2.1-10.0%	Very Low	-	<i>4</i> .	-	-
101-180%	Low	13	-C	16	39
181-25.0%	Intermediate	72	22	47	87
>25.0%	High	7	2	2	2
Gelatinization Ten	nperature by Alkali-Spread	ling Value			
74.5-80.0°C	High	1	1	,	1
70.0-74.0°C	High Intermediate	50	9	30	82
70.0-74.0°C	Intermediate	15	-	29	20
<70.0°C	Low	23	13	4	23

Table 1. Grain quality properties of pre-NCT rice lines (Con't...)

Grain Quality Property		Number of Ent	Number of Entries				
		DSR-ON	LT OF	FAV-SUB	DAT		
Parameter/Level/Classification		2013 WS	2013 WS	2013 WS	2013 WS		
		(N=79)	(N=72)	(N=46)	(N=24)		
Milling Recover	y and Physical Attributes						
Brown Rice							
>80%	Good			n∕a			
75.1 - 79.9%	Fair	71	72	n/a	10		
<75%	Poor	7	-	n∕a	14		
Milled Rice							
≻70.1%	Premium	26	27	n∕a	1		
65.1 - 70.0%	Grade 1	49	45	n/a	12		
60.1-65.0%	Grade 2	4		n∕a	9		
55.5-60.0%	Grade 3	,		n/a	2		
<55.5%	Below Standards		· •	n/a			
Head Rice							
>57%	Premium	12	1	n/a	9		
48.0-56.9%	Grade 1	24	2	n/a	7		
39.0 - 47.9%	Grade 2	19	14	n/a	7		
30.0 - 38.9%	Grade 3	20	17	n∕a	ĩ		
<30.0%	Below Standards	4	38	n∕a			
Chalkiness							
0.1-2.0%	Premium			19	n/a		
2.1-5.0%	Grade 1	12	17	16	n/a		
5.1-10.0%	Grade 2	25	40	6	n/a		

Grain Quality Property		Number of Entries				
		DSR-ON	LT OF	FAV-SUB	DAT	
Parameter/Level	/ Classification	2013 WS	2013 WS	2013 WS	2013 WS	
		(N=79)	(N=72)	(N⊨46)	(N=24)	
10.1-15.0%	Grade 3	21	15		n/a	
>15.0%	Below Standards	21		3	n/a	
Grain Length						
>7.5mm	Extra Long	9	1	6	n/a	
6.4-7.4mm	Long	65	70	34	n/a	
5.5-6.3mm	Medium	5	1	4	n/a	
<5.4mm	Short	÷	÷		n/a	
Grah Shape						
>3	Slender	51	71	34	n/a	
2.0- 3.0	Intermediate	28	1	10	n/a	
<2.0	Bold	2	2	4	n/a	
Physicochemical F	Properties					
Amylose Content						
0.0- 2.0%	Waxy					
2.1- 10.0%	Very Low	2	2		1	
10.1-18.0%	Low	7		32	13	
18.1-25.0%	Intermediate	71	72	14	10	
>25.0%	High	1				
Gelatin Eation Temperature by Alkali-Spreading Value						
74.5-80.0°C	High					
70.0-74.0°C	High Intermediate	44	2	2	4	
70.0-74.0°C	Intermediate	20	70	39	12	
<70.0°C	Low	15	2	3	12	

Grain Quality P	roperty	Number of Er	Number of Entries				
Parameter/Level/Classification		LT OF	MYT	DSR-ON	SV		
		2014 DS	2014DS	2014 DS	2014 DS		
		(N=18)	(N=10)	(N=71)	(N=17)		
Milling Recov Attributes	ery and Physical						
Brown Rice							
>80%	Good	·		*			
75.1-79.9%	Fair	18	6	68	17		
<75%	Poor	2	4	3	1		
Milled Rice							
>70.1%	Premium	16		24	4		
65.1-70.0%	Grade 1	2	8	47	13		
60.1-65.0%	Grade 2		2				
55.5-60.0%	Grade 3		-				
<5.5%	Below Standards						
Head Rice							
>57%	Premium			1	6		
48.0-56.9%	Grade 1	1	5	20	10		
39.0-47.9%	Grade 2		2	25	1		
30.0-38.9%	Grade 3	9	2	21			
<30.0%	Below Standards	8	1	4			
Chalkiness							
0.1-2.0%	Premium			9	6		
2.1- 5.0%	Grade 1	1		24	5		

Grain Quality P	roperty	Number of Er	ntries		,
		LT OF	MYT	DSR-ON	sv
Parameter/Level/Classification		2014 DS	2014 DS	2014 DS	2014 DS
		(N=18)	(N=10)	(N=71)	(N=17)
5.1-10.0%	Grade 2	10	4	11	4
10.1-15.0%	Grade 3	7	5	9	1
>15.0%	Below Standards	-	1	15	1
	Mixed/opaque				з
Grain Length					
>7.5mm	Extra Long	2	2	3	1
6.4 - 7.4mm	Long	18	9	60	13
5.5-6.3mm	Medium		1	8	з
<5.4mm	Short			-	
Grain Shape				0	
>3	Slender	18	б	25	7
2.0-3.0	Intermediate	2	4	46	10
<2.0	Bold				
Physicochemic	al Properties				
Amylose Conte	nt				
0.0-2.0%	Waxy			-	
2.1-10.0%	Very Low	2	2	3	2
10.1- 18.0%	Low		1	8	5
18.1-25.0%	Intermediate	18	9	57	10
>25.0%	High	-	-	9	2
Gelatinization Value	Temperature by All	kali-Spreading			
74.5-80.0°C	High	-		-	
70.0-74.0°C	High Intermediate	17	4	20	9
70.0-74.0°C	Intermediate	1	4	36	5
<70.0°C	Low	2	2	15	З

		SPSI	LTOF	
Parameter/Lev	vel/Classification	2014 DS	2014 DS	
		(N=31)	(N=72)	
Milling Recov Attributes	ery and Physical			
Brown Rice				
>80%	Good	÷.		
75.1-79.9%	Fair	21	69	
<75%	Poor	10	3	
Milled Rice				
>70.1%	Premium		5	
65.1-70.0%	Grade 1	18	67	
60.1-65.0%	Grade 2	12	-	
55.5-60.0%	Grade 3	1	-	
<55.5%	Below Standards	ч		
Head Rice				
>57%	Premium	1	-	
48.0-56.9%	Grade 1	6	34	
39.0-47.9%	Grade 2	10	38	
30.0-38.9%	Grade 3	7	-	
<30.0%	Below Standards	7	4	
Chalkiness				
0.1- 2.0%	Premium	1		
2.1- 5.0%	Grade 1	2	22	
5.1- 10.0%	Grade 2	v.	45	
10.1-15.0%	Grade 3	1	5	
>15.0%	Below Standards	1		
	Opaque/pigmented	26		

Grain Quality Property		Number of Entries	
		SPSI	LTOF
Parameter/Level/Classification		2014 DS	2014 DS
		(N=31)	(N=72)
Grain Length			
>7.5mm	Extra Long	1.0	-
6.4 - 7.4mm	Long	18	72
5.5-6.3mm	Medium	13	
<5.4mm	Short		ā.
Grain Shape			
>3	Slender	17	72
2.0-3.0	Intermediate	14	-
<2.0	Bold	1.5	÷
Physicochemical	Properties		
Amylose Content			
0.0-2.0%	Waxy		5 .
2.1-10.0%	Very Low	13	
10.1-18.0%	Low	4	ň.
18.1-25.0%	Intermediate	13	72
>25.0%	High	1	
74.5-80.0°C	High	7	70
70.0-74.0°C	High Intermediate		
70.0-74.0°C	Intermediate	19	2
<70.0°C	Low	5	2 ⁻

Factors affecting rice spoilage and optimum cold storage treatment of cooked rice

EH Bandonill, GGCorpuz, MJCAblaza

Filipinos regularly eat boiled rice three times a day where most of the consuming population belongs to below poverty line and depends highly on white rice for their cheap source of carohydrates and protein. To save on fuel cost, cooking rice once in the morning enough for the whole day's consumption is a common practice. In the absence of a refrigerator, cooked rice is usually kept at ambient temperature. This practice prevents rice retrogradation caused by refrigeration. However, in several instances, food poisoning caused by the consumption of unrefrigerated cooked rice has been reported. Homemakers have also noted that cooked rice spoils easily during hot weather and when using specific cooking pans, or when cooking a particular rice variety. The issue of rice spoilage has been raised but the specific causes have not been identified. The presence of Bacillus cereus, a Gram-positive bacteria is often associated with rice spoilage which can cause foodborne illnesses where its toxin can be fatal in some cases.

Meanwhile, some private companies are also getting interested in knowing the optimum cold storage and reheating conditions of cooked rice since most of their ready-to-eat rice meals are stored under refrigerated and/or chiller temperatures. To improve the stability of cooked rice and disseminate information to interested groups/clients, it is therefore important to investigate the best processing techniques that will not only extend the shelf life but also maintain the quality of cooked rice. In the advent of rice conservation and to help in the attainment of rice sufficiency in the country, it is important to keep both raw and cooked rice, safe for consumption even for an extended period. The results of the study will provide not only valuable information on cooked rice spoilage and its prevention but may also save lives of the rice consuming public.

Highlights:

About 100 respondents from Nueva Ecija (57%), Pangasinan and Tarlac (15% each), Aurora, Zambales, Bulacan, Tarlac, Pangasinan and Pampanga were surveyed on their perception of rice varieties that spoil easily and the possible causes of spoilage (Figure 1). Among the respondents, seldom have spoiled rice (69%) while those with rice left-overs often reserve the rice (65%) by reheating and re-cooking in the form of fried rice (Figure 2). NFA rice (41%), grains with short and bold length and shape, brokens (72%), rice cooked in excess water (92%), kept with tight cover (72%), and at room temperature (88%) particularly during hot, humid days, were the type of rice and the factors identified to cause faster spoilage of rice (Figure 3).

- Determined the rate of spoilage of popular rice varieties (Japonica, NSIC Rc160, Rc222, SL8, IR64, NFA). SL8 and NFA had perceptible odor after 21h. SL8, Rc222, and NFA with higher optimum water requirement (1:1.9,1:1.6,1:1.6, respectively) had higher moisture content when freshly cooked and became more cohesive and more moist after 24h.
- Total plate count (TPC) was within the tolerable limit (105) for all the popular varieties tested (raw and freshly cooked) and did not produce any off- odor.
- After 18 h of storage at ambient temperature (29.9 to 30.5oC) and 55 to 61% Relative Humidity (RH), SL8 and NFA rice had perceptible off-odor while microbial load was beyond 105. In contrast, NSIC Rc160 and Rc222 remained to have no offodor and count was within 105. IR64 and Japonica remained to have no off-odor but microbial load was already beyond 105. Interestingly, IR64 remained to have no off-odor even after 21h.
- Determined the rate of spoilage of IR64 and PSB Rc10 using different head rice percentage, namely: 0, 25, 50, 75, and 100%. Freshly cooked and 24-hr staled IR64 had lower moisture content (56.9 to 65.5%) than PSB Rc10 (64.2 to 69.3%) due to lower optimum cooking water of IR64 (1.5) than PSB Rc10 (2.1).
- Highest total plate count was observed in 0% head rice of the 2 varieties although IR64 had lower count than PSB Rc10 (Table 2).
- Similar trend was observed in the sensory evaluation of IR64 where 0% head rice had the highest off-odor score (2.43) at 24 h storage. Gloss, cohesiveness, and moistness of IR64 decreased with increasing ratio of head rice.
- In contrast, 0% head rice of PSB Rc10 had the lowest off-odor score at 30 h storage while gloss, cohesiveness, and moistness of 25% headrice was the lowest.
- Determined the rate of spoilage of rice using different amounts of cooking water, namely: 1:10 (LW), 1:1.5 (OW), and 1:2.0 (EW) for IR64 and 1:1.4 (LW), 1:2.1 (OW), and 1:2.8 (EW) for PSB Rc10. Increasing moisture content was obvious in the 2 varieties as the amount of cooking water was increased.

- IR64 had lower microbial growth compared with PSB Rc10. Both IR64 and PSB Rc10 cooked with optimum cooking water (OW) was observed to have the highest microbial growth than LW and EW (Table 3). Slight off-odor was perceived only after 24h standing with OW having the highest off-odor score (2.15). Glossiness and moistness of sample was observed to be highest in IR64_OW, followed by IR64_EW, and IR64_LW (Figure 4A).
- Off-odor was perceived at 12h in PSB Rc10_EW. Glossiness and moistness was highest in PSB Rc10_EW and lowest in PSB Rc10_LW. (Figure 4B).



Figure 1. Demographic profile of 100 survey respondents.



Figure 2. Presence of left-overs among households, what they usually do with leftovers and the frequency of having spoiled rice.



Figure 3. Consumers' perception on factors affecting rice spoilage.

nee ratio.						
Line d Direc Develop to ge	Total Plate Count (Cfu/G X 10 ⁵)					
Head Kice Percentage	0h	6h	9h	12h	24h	
IR64_0%	1.41	0.16	0.09	0.20	39.00	
IR64_25%	0.52	0.17	0.06	0.10	9.00	
IR64_50%	0.51	0.07	0.25	0.20	1.00	
IR64_75%	0.28	0.09	0.63	0.10	1.00	
IR64_100%	0.15	1.44	0.05	0.60	8.00	
PSB Rc10_0%	0.15	0.06	0.30	2.69	114.10	
PSB Rc10_25%	0.03	0.01	0.07	1.19	60.10	
PSB Rc10_50%	0.04	0.03	0.33	1.76	168.40	
PSB Rc10_75%	0.02	0.09	0.01	0.30	62.80	
PSB Rc10_100%	0.02	0.11	0.07	1.84	16.50	

Table 2. Microbial load of cooked IR64 and PSB Rc10 with different head

 rice ratio.

 Table 3. Microbial load of IR64 and PSB Rc10 cooked with different cooking water ratio.

HEAD	RICE	TOTAL PLATE COUNT (cfu/g x10 ⁴)					
PERCENTAGE		0h	6h	9h	12h	2 4h	
IR64_LW		0.05	0.15	0.30	0.45	58.00	
IR64_OW		0.75	0.10	10.65	7.90	271.00	
IR64_EW		0.40	0.00	1.30	0.10	2.00	
PSB Rc10_LW		0.07	0.23	0.18	0.34	2.50	
PSB Rc10_OW		0.13	0.47	6.21	51.00	955.00	
PSB Rc10_EW		0.05	0.37	3.04	18.90	520.00	





12.00

10.00

8.00







Figure 4A. Sensory characteristics of cooked IR64 with different cooking water and staled up to 24h.









Figure 4B. Sensory characteristics of cooked PSB Rc10 with different cooking water and staled up to 24h.

II. Nutritional Quality Assessment and Elemental Composition of Philippine Rice

RG Abilgos-Ramos, NC Ramos, CT Estonilo, JD Adriano, CA Arcilla, LL Quirit

Beside grain quality, nutritional properties, source, and true identity of rice are also important considerations for consumers. This is due to an increasing awareness and demand for more nutritious foods, reliable sources of supply, and concern on health and safety of food for consumption. The macro and micronutrients in rice varies depending on variety, location, and other factors. This project looked into the important elements and isotopic fingerprints of imported and popular rice planted in different parts of the country.

Simultaneous Multi-Element Determination in Rice Using Microwave Plasma Atomic Emission Spectroscopy (MP-AES): Method Optimization and Application

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The elemental composition of rice is of great importance for its nutritional value. It is a major consideration due to emerging issues concerning contamination of rice farms near mining areas leading to decreased confidence in the quality and safety of rice produced outside local region. Many studies have reported essential and trace element concentrations of copper (Cu), manganese (Mn), sodium (Na), phosphorus (P), potassium (K), and zinc (Zn) in polished rice (Al-Saleh et al. 2001, Antoine et al. 2012, Bennet et al, 2000). However, other elements that are detrimental to health such arsenic (As), cadmium (Cd), lead (Pb), and mercury (Hg) are also found in low concentrations (Al-Saleh et al. 2001, Bennet et al. 2000, Jung et al. 2005, Shimbo et al. 2001, Sommella et al. 2013). In Japan, the concentrations of Cd and Pb in rice were reported at 0.05 and 0.002mg/kg, respectively. In Wisconsin, USA, rice contain about 0.136, 0.016, and 0.250mg/kg of As, Cd, and Pb, respectively. In South Korea, As, Cd, and Pb in rice were reported at 0.122, 0.021, and 0.206mg/ kg, respectively.

Currently, a range of elemental analytical techniques, including flame and graphite furnace atomic absorption spectroscopy (F-AAS and GF-AAS), X-ray fluorescence spectrometry (XFR), inductively coupled plasma atomic emission spectroscopy (ICP-AES), and inductively coupled plasma mass spectroscopy (ICP-MS) are available for use [Balaram et al., 2013]. A relatively new and promising analytical technique, microwave plasma atomic emission spectroscopy (MP-AES), was introduced by Hammer in 2011. The development of method for this instrument should be explored to screen rice varieties for mineral and heavy metals content. Hence, study utilized the newest commercially available instrument (4100 MP-AES) based on microwave induced plasma. Instead of using the inductively coupled argon plasma, the MP-AES uses nitrogen plasma which is sustained by a lower power microwave source operating at atmospheric pressure. However, nitrogen plasma is cooler and features different performance characteristics. Since the temperature is lower, most of the elements are in the atomic state resulting in a simpler spectrum than the argon plasma. Thus, optimization of operating procedure is important. In addition, the limit of detection (LOD) and limit of quantification (LOQ) for MP-AES method in rice will be measured using a certified reference material (CRM). A CRM will validate the analytical method and will calibrate the instrument. The LOD is approximately equal to method detection limit (MDL) if a CRM is used. The LOD and LOQ are the lowest concentration level that can be determined in an analyte in a specified matrix.

Highlights:

Optimization: Method validation is a tool used to investigate if a specific analytical method actually measures what it intends to measure, and thus is suitable for its intended purpose (Taverniers et al. 2004). The CRM identified for rice is Sigma IRMM804-15G. The Institute for Reference Materials and Measurement (IRMM) is one of the institutes of Joint Research Centre (JRC) that produces reference materials with reliable European measurement. The analytes present in the CRM are copper, zinc, selenium, cadmium, arsenic, and lead. The accuracy of the dry-ashing method in this study was evaluated by calculated recovery using CRM 804. Calculation of acceptable percent recoveries and precision (Table 5) were based on the criteria according to Horwitz and AOAC Peer Verified Methods (PVM) Program where the levels of acceptability of the %RSD is closely linked to the concentration of the analyte. Following the procedure for cereal and plants of AOAC (1990), rice flour was ash and digested with nitric acid and hydrochloric acid. The concentrations obtained for Cu, Mn, Zn, Cd, and Pb were 2.21, 29.5, 18.7, 1.58, and 0.41 mg/kg, respectively. The recoveries computed for each element were 80.8% for Cu, 86.2% for Mn, 80.9% for Zn, 98.0% for Cd, and 101.2% for Pb. The relative standard deviations for the five detected elements ranged from 0.13 to 15.23%. All recoveries and precision values are acceptable for the target elements.

• The LOD and LOQ in rice were determined using standard solutions. A set of concentration levels fortified in method blank in 6 replicates to optimize the method for rice. Table 5 shows the detection limit for 5 elements. It can be observed that LOD has close values to the LOQ for each element suggesting that the solutions has a clear analyte line

emission intensities during quantification. It could be that the nebulization process removes the water in the aerosol efficiently that limits emission interferences.

- Application: Farm areas near mining and volcanic activities were identified through coordination with the Regional Field Officer (RFO) and Provincial Agriculturist Officer (PAO) in the country. There are two (2) regions namely, Central Luzon and Bicol, identified to be affected by volcanic activities. While there are four (4) regions, Ilocos, Bicol, Davao, and Caraga, identified near mining doings. From each farm site, 1kg of sample was randomly collected. A total of forty (40) rough rice samples were collected and analyzed. A control sample was also used to evaluate the trace elements present if the area was not affected by the possible contamination from mining and volcanic activities. Two (2) rice samples were obtained from the Business Development Division of PhilRice during the wet season of 2014.
- All samples were digested and analyzed similar to CRM 804. Table 6 shows the mineral concentrations in polished rice collected from identified regions in the Philippines that were potentially contaminated from mining and volcanic activities. The control sample, where the location was neither affected by both activities, revealed that Cd and Pb were both undetected. The Cu content was 1.4mg/kg, Mn was 17.9mg/kg and Zn was 17.5mg/kg. The rice samples collected from Central Luzon came from one province only, namely Pampanga, which was impacted by the explosion of Mount Pinatubo in 1991. From the results shown in Table 3, all elements were detected except for Cd. Pb was found to be the highest with 3.1mg/kg, followed by Cu with 2.9mg/ kg, Zn with 13.0mg/kg, and Mn with 11.1mg/kg. The llocos region identified La Union as potentially contaminated from a mining activity. From the results, all elements were present where Zn concentration was 14.2mg/kg, followed by Mn at 7.9mg/kg, Cu at 3.7mg/kg, Pb at 1.5mg/kg, and Cd at 0.8mg/ kg. The Bicol region has 4 provinces where both mining and volcanic activities are present. Camarines Sur and Masbate are potentially affected by mining, while Albay, and Sorsogon by volcanic activities (Mount Mayon and Mount Bulusan). Samples in Bicol affected by mining had Zn concentration of 17.0mg/kg, Mn at 7.9mg/kg, Cu at 2.4mg/kg and Pb at 1.6mg/ kg. Zn content was found at 8.9mg/kg, Mn at 7.0mg/kg, Cu at 1.3mg/kg and Pb at 1.14mg/kg for samples from Bicol affected by volcanic activities. Similar to Central Luzon, Bicol

region also had negative detection for cadmium. The samples from Davao region were collected from Compostela Valley potentially contaminated from mining. Zn content was also found highest at 20.1mg/kg, followed by Mn at 13.3mg/kg, Cu at 3.0mg/kg, Pb at 3.0mg/kg, and Cd at 0.5mg/kg. Caraga region includes the Agusan Del Sur and Surigao Del Norte provinces for the collection where mining is present. The Zn content was 21.2mg/kg, Mn was 15.8mg/kg, Cu was 3.9mg/ kg, Pb was 3.7mg/kg, and Cd was 0.2mg/kg.

Analyte	LOD (mg/L)	LOQ (mg/L)	
Cu	0.02	0.05	
Mn	0.01	0.04	
Se	0.03	0.10	
Fe	0.29	0.97	
Zn	0.02	0.05	
к	0.09	0.30	
Cd	0.01	0.04	
Pb	0.01	0.04	

Table 4. Concentration levels in MP-AES (mg/kg) (n=6).

Table 5: Acceptable %recoveries in terms of %RSD values for Cu, Mn, Zn, Cd, and Pb in CRM 804 based on the concentrations present in the digestion mixture (n=6).

Element	Certified value, mg kg ^{°1}	Experimental value, mg kg ^{°1}	% RSD	Experimental %Recovery	Accept able %Recovery ^{a,b}	Horwitz %RSD ^b	AOAC P∨M %RSD ^{a,b}
Mn	34.2±2.3	29.5±1.9	6.50	86.2	80-110	11.3	7.3
Zn	23.1±1.9	18.7±1.4	7.37	80.9	80-110	11.3	7.3
Cd	1.61±0.07	1.58±0.09	5.83	98.0	80-110	16	11
Pb	0.42±0.07	0.41	0.13	101.2	80-110	22.6	15

a – Taverniers et al., 2004

b – AOAC, 1990

			Cu .	Mn.	Zn .	Cd .	РЬ.
Region	n		mg kg '	mg kg 1	mg kg 1	mg kg 1	mg kg 1
Control	2	Mean ± SD	1.4±1.0	17.9±3.2	17.5±2.7	nd	nd
		Min-max	0.6-2.1	15.6-20.1	15.6-19.5	nd	nd
Central Luzon	4	Mean ± SD	2.9±0.4	11.1±0.5	13.0±0.3	nd	3.1±0.5
Volcanic)		Min-max	1.5-4.0	6.2-17.2	8.0-19.9	nd	2.2-3.5
locos	5	Mean ± SD	3.7±0.2	7.9±0.3	14.2±0.4	0.8	1.5±0.4
Mining)		Min-max	1.5-5.7	6.0-8.7	10.5-17.4	0.5-3.5	0.5-3.0
Bicol	12	Mean ± SD	1.3±0.1	7.0±0.2	8.9±0.2	nd	1.1±0.3
Volcanic)		Min-max	0.5-3.0	4.0-10.2	7.0-12.2	nd	0.0-2.2
Bicol	12	Mean ± SD	2.4±0.2	7.9±0.2	17.0±0.2	nd	1.6±0.3
Mining)		Min-max	1.0-3.5	1.0-12.9	7.4-30.0	nd	0.5-2.7
Davao	з	Mean ± SD	3.7±0.1	13.3±0.2	20.1±0.1	0.5	3.0±0.2
Mining)		Min-max	3.0-5.2	10.5-14.7	13.5-33.2	0.5-3.5	1.7-3.7
Caraga	4	Mean ± SD	3.9±0.1	15.8±0.2	21.2±0.3	0.2	3.7±0.6
Mining)		Min-max	2.0-5.7	10.2-28.7	10.9-41.2	0-0.5	2.7-5.0

 Table 6. MP-AES determination in rice (mg/kg).

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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