Parboiling of pigmented and non-pigmented Philippine rice (*Oryza sativa* L.) cultivars: Textural properties and carbohydrate quality

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<u>Abstract</u>

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Keywords

parboiling, pigmented rice, resistant starch, retrogradation Parboiling improves rice carbohydrate quality, which is beneficial for diabetes management in Asia. However, parboiling drastically alters cooked rice texture, making it unappealing for Filipinos, who generally prefer tender cooked rice. The present work evaluated the carbohydrate quality and cooked rice hardness of nine local pigmented and non-pigmented rice varieties as affected by parboiling and cooking treatments. The amylose content (AC), retrogradation potential (RP), and resistant starch (RS) content were determined, and the samples with the highest RS were further investigated for their thermal properties and starch morphology. Results showed that parboiling increased RS level by as much as five-fold, particularly in the red rice Minaangan, which had intermediate AC and the highest RP (2,822 cP) among the samples. The highest RS, 2.71 \pm 0.02 g/100 g, was recorded in parboiled NSIC Rc 222 cooked at 1:2.5 (rice:water). It also produced soft cooked rice, which was significantly better than its non-parboiled counterpart. High proportion of water needed to gelatinise parboiled non-pigmented NSIC Rc 222 was due to higher starch crystallinity as indicated by its gelatinisation enthalpy and intact starch granule. In conclusion, parboiling and cooking at appropriate rice:water ratio of NSIC Rc222 and Minaangan could deliver healthier cooked rice with Filipino preferred textural quality.

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Introduction

Rice is an important cereal accounting for 60% of total Asian carbohydrate energy intake (Cui and Dibley, 2012; Moongngarm et al., 2012). However, rice has intermediate to high glycaemic index (Miller et al., 1992; Fitzgerald et al., 2011), which is linked to hyperglycaemia and type 2 diabetes (Zuñiga et al., 2014; Seah et al., 2019). In previous studies, higher postprandial blood glucose levels and poorer insulin sensitivity were observed in Asians than Caucasians when they ingest similar foods (Venn et al., 2010; Kataoka et al., 2013; Tan et al., 2017). With diabetes predicted to afflict an estimated 630 million people globally by 2030 (WHO, 2016), and is already the fourth highest cause of mortality in the Philippines (PSA, 2022), dietary interventions could help alleviate this problem. Wee and Henry (2020) argue

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that combining carbohydrate quantity reduction and quality improvement by way of reduction of digestion rate and amount of available carbohydrates for digestion could be an effective diabetes management strategy in Asia. Carbohydrate quality improvement in rice has huge implications as around 90% of milled rice dry matter is starch (Hizukuri *et al.*, 1989).

Resistant starch (RS) is one of the three dietary starch groups based on *in vitro* digestibility, along with rapidly and slowly digestible starches (Rosin *et al.*, 2002). RS is defined as starch, together with its degradation products, that escapes small intestinal digestion, and is fermented by colonic bacteria (Megazyme, 2019). Reports have shown the health benefits of RS. According to Chen *et al.* (2010), the incidence of colorectal cancer was higher in countries with lower RS intake. A randomised controlled trial of Mohan *et al.* (2016) in 30 human participants revealed significant decrease in mean glycaemic index (GI) in the group eating cooked white rice with 3.9 ± 0.02 g/100 g dry weight (dw) RS than those eating with 0.6 ± 0.03 g/100 g dw RS. An *in vivo* study on mice by Wan *et al.* (2019) showed that daily consumption of cooked white rice with RS as low as 1.07 g/100 g dw reduced fat deposition and adipose weight, while on a moderately high (39%) fat diet. This could be due in part to its lower energy value (8 kJ/g) as compared to fully digestible starch (15 kJ/g) (Fuentes-Zaragoza *et al.*, 2010). Finally, an *in vitro* GI study of Wiruch *et al.* (2019) on parboiled brown rice in a retort pouch containing 2.64 \pm 0.02 g/100 g dw RS showed significant decrease in GI when compared with non-parboiled rice.

Traditional parboiling involves rice paddy soaking until saturation, steaming, and slow drying before husk removal, which changes the overall grain microstructure and nutrient distribution, and inactivates enzymes (Larsen et al., 2000; Kale et al., 2017). Rice has naturally low levels of RS (0 - 1.0%)(Juliano, 2007). However, it has been reported that parboiling had an enhancing effect on RS levels (Walter et al., 2005; Kim et al., 2006). Hydrothermal treatments like parboiling gelatinise starch, which renders it fully digestible. Retrogradation, which occurs during cooling or aging of gelatinised starch, re-assembles the gelatinised starch into a partially crystalline structure leading to the formation of digestion-resistant retrograded RS or RS3 (Garcia-Rosas et al., 2009; Bharath Kumar and Prabhasankar, 2014). Parboiled rice was previously shown in vitro to have a lower glycaemic index (Kale et al., 2017) than non-parboiled rice, which could be due to the presence of amylose crystallites and amylose-lipid complexes (RS5) (Lamberts et al., 2009).

Despite the nutritional benefits of parboiled rice, in addition to its implications on food sufficiency due to improved milling yields, a major challenge in its acceptability as a digestibility-reducing strategy in the Philippines is its harder and flakier cooked rice as opposed to tender texture preferred by most Filipinos (Oli et al., 2014; Bandonill et al., 2017). Any modifying processing method to improve carbohydrate quality, such as parboiling, should have minimal unfavourable effect on the textural and sensory qualities of the food (Wee and Henry, 2020). Therefore, the present work evaluated traditional parboiling to increase the RS content of several Philippine rice varieties, and improve their cooked texture to suit Filipinos' preferences.

Materials and methods

Rice samples and chemicals

Nine Philippine rice varieties (five modern non-pigmented and four traditional pigmented) were collected during 2015 wet - 2016 dry cropping seasons, and used as samples (Table 1). A resistant starch assay kit (K-RSTAR) was purchased from Megazyme International, Ireland. All chemicals used in the present work were of analytical grade.

Table 1. Rice varieties anal	ysed in the	present work.
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Rice	Classification	Cultivation	
variety	Classification	season	
Ingopon	Traditional, pigmented	2015 wet	
Imbuucan	Traditional, pigmented	2015 wet	
Kamanga	Traditional, pigmented	2015 wet	
Minaangan	Traditional, pigmented	2015 wet	
NSIC Rc 242	Modern, non-pigmented	2015 wet	
PSB Rc 10	Modern, non-pigmented	2015 wet	
PSB Rc 14	Modern, non-pigmented	2015 wet	
NSIC Rc 160	Modern, non-pigmented	2016 dry	
NSIC Rc 222	Modern, non-pigmented	2016 dry	

Traditional parboiling

Traditional parboiling was done according to Corpuz *et al.* (2013), with few modifications. Paddy rice (1 kg) was soaked in 3 L of distilled water for 24 h with changing of water halfway to avoid fermentation. Paddy rice was steamed for 1 h, and then sun-dried and/or oven dried to 12 - 14% moisture content as measured by a grain moisture meter (TG 27195, Draminski, Olsztyn, Poland).

Processing of parboiled and non-parboiled rice

Parboiled and non-parboiled paddy rice samples were dehulled using a Satake THU-35A (Tokyo, Japan) machine followed by polishing in a McGill No. 3 milling machine (Dayton Industrial Motor). Head rice for cooking was obtained using a Satake TRG-05A (Tokyo, Japan) rice grader. A portion of the grains were turned into flour using Cyclotec 1039 Sample Mill (Tecator, Hoganas, Sweden) for the analysis of viscosity, thermal properties, flour microscopy, amylose, and RS content.

Amylose content

Raw non-parboiled samples were evaluated for their amylose content (AC) following the modified NH₄Cl method of Juliano et al. (2012). Rice flour (100 mg) was incubated in 95% ethanol (1 mL) and 1 N NaOH (9 mL) at room temperature for 18 - 24 h followed by dilution to 100 mL. Next, 5 mL aliquot was then added with 0.9 N NH₄Cl (1 mL) and 0.5% I₂ in 1.5% KI solution (2 mL), diluted to 100 mL by distilled water, then incubated for 20 min to allow colour development. Absorbance was measured using a UV-Vis Spectrophotometer (DU 730, Beckman Coulter, Fullerton, CA) at 620 nm. Blank, IR2071, IR64, IR8, and IR29 checks, as well as 1, 3, 5, 10, 15, 25, and 35% amylose standards, were run with the samples. Rice varieties were classified based on amylose content as waxy (0 - 2%), very low (2.1 -10%), low (10.1 - 17%), intermediate (17.1 - 22%), and high (> 22%) (NCT, 2022).

Determination of retrogradation potential

Total setback or retrogradation potential (RP) of raw non-parboiled rice was determined as the difference between duplicate final and trough viscosity values obtained using Rapid Visco Analyser (RVA 4D, Newport Scientific, Australia) following the AACC (1999) method. TSB is an indirect measure of starch retrogradation or the recrystallisation of gelatinised starch as it cools (Akintayo and Ashogbon, 2012).

Cooking of rice

Rice grains were cooked using the method of the National Cooperative Project for Rice (NSIC, 1996). Briefly, head rice of parboiled samples was placed into the inner pan of an electric automatic rice cooker, added with tap water in a 1:1.5 (T1) and 1:2.5 (T2) rice:water ratio, and swirled 20 times to wash the rice. Wash water was decanted, measured, and replaced with same amount of tap water. The rice was cooked until the audible switch off sound of the cooker. Cooked rice was left undisturbed for 15 min before subsequent analyses. Non-parboiled rice cooked in 1:1.5 rice:water ratio served as control.

Texture evaluation

Cooked rice hardness was evaluated using an Instron hardness tester (Norwood, MA, USA). Approximately 17 g of cooked rice was transferred into a cell measuring 10 cm² with 24.5-mm holes, and extruded through the Ottawa Texture Measuring System. Based on the Instron hardness value (kgf/cm²) obtained using Instron BluehillTM texture

software version 2.17 (Norwood, MA, USA), cooked rice was classified as very soft (VS), 0.5 - 1.0; soft (S), 1.1 - 1.8; medium (M), 1.9 - 2.5; and hard (H), > 2.5.

Resistant starch content

Duplicate determination of RS content for both raw and cooked rice flours were performed using the Megazyme kit based on the AOAC Official Method 2002.02/AACC Official Method 32-40.01 (Megazyme, 2019). A kidney bean powder (4.9% w/w RS) (Megazyme International Ireland Ltd., Bray, Co. Wicklow, Ireland) served as process control across all RS determinations.

Physical and thermal analyses

To determine signs of gelatinisation and retrogradation following parboiling, raw rice flours were evaluated for their physical and thermal properties. These analyses were done on one pigmented and one non-pigmented rice with the highest RS among the samples before and after parboiling. Granule size and morphology were assessed using a scanning electron microscope (SEM) (SU3500, Hitachi, Japan). Briefly, a small amount of raw flour particles was sprayed onto a carbon tape fixed on the specimen stub. Excess powder was blown off, and the samples were viewed with an acceleration voltage of 1.50 kV and magnification of 6,000×.

Flour thermal property was determined using a differential scanning calorimeter (Q2000, TA Instruments, Delaware, USA) following the ASTM No. D3418-12 method (ASTM, 2013). Gelatinisation enthalpy was recorded, and percentage of gelatinised starch was calculated based on the enthalpy change after parboiling (ΔH_{par}) as compared to non-parboiled (ΔH_{raw}) samples (Patindol *et al.*, 2008), using Eq. 1:

% Gelatined starch (%) =
$$\left[1 - \left(\frac{\Delta H_{par}}{\Delta H_{raw}}\right)\right] x \ 100$$
(Eq. 1)

Statistical analysis

ANOVA with subsequent pairwise comparison using Tukey's HSD test on the amylose content, RP, cooked rice hardness, and RS values were performed using IBM SPSS Statistics version 20 (Cary, NC). The level of significance used was p < 0.05. All measurements were performed in triplicates, unless otherwise stated.

Results and discussion

Amylose content

Table 2 shows the AC of the rice samples in increasing order.

Table	2.	Amylose	content	and	retrogradation
potential of raw non-parboiled rice varieties.					

Rice variety	Amylose content	Retrogradation potential (cP)
	(%)	(n = 2)
Ingopon	0.6 (Wx)	$396 \pm 1^{\text{g}}$
NSIC Rc 242	11.5 (L)	$1146\pm11^{\rm f}$
NSIC Rc 160	13.7 (L)	$1285\pm16^{\rm f}$
Minaangan	19.4 (I)	2822 ± 30^{a}
PSB Rc 14	20.0 (I)	$1588 \pm 13^{\text{e}}$
Imbuucan	22.7 (H)	$1754 \pm 10^{\rm d}$
Kamanga	23.6 (H)	$1788\pm 6^{\rm d}$
PSB Rc 10	24.2 (H)	$2261 \pm 137^{\circ}$
NSIC Rc 222	25.2 (H)	1762 ± 1^{d}

Wx: waxy, 0.0 - 2.0%; L: low, 10.1 - 17.0%; I: intermediate, 17.1 - 22.0%; and H: high, > 22.0%. Values are mean \pm SD. Means followed by different superscripts within the same column are significantly different at *p* < 0.05.

The values ranged from 0.6 - 25.2%, with all amylose classifications represented. Most of the samples had intermediate to high AC, which generally had higher RP. The highest RP value was recorded in Minaangan, which is classified as an intermediate-AC rice. Retrogradation of starch is influenced by several factors, which include amylose content (Englberger et al., 2010). Hydrothermal treatment and subsequent drying during parboiling induce starch gelatinisation followed by amylose recrystallisation (Ranawana et al., 2009). This creates RS3 structures which are double helical in nature stabilised with hydrogen bonds, has a characteristically high thermal stability (117 125°C), and do not fit amylase binding site (Chen et al., 2010; Birt et al., 2013; Raigond et al., 2014; Sonia et al., 2015). Higher amylose translates to higher retrogradation tendency (Sobolewska-Zielinska and Fortuna, 2010; Gu et al., 2011), and that starch retrogradation enthalpy (ΔH_r) of cooked rice was shown to be positively correlated with AC values (r =0.650, p = 0.01) (Yu *et al.*, 2009). Moreover, single helical-complex structures from amylose and fatty acids/fatty alcohols (RS5) could form during

parboiling which prevents starch binding and cleavage by amylase (Birt *et al.*, 2013; Raigond *et al.*, 2014). RS5 also restricts starch granule swelling and enzyme hydrolysis through entanglement of amylopectin molecules (Seneviratne and Biliaderis, 1991; Hasjim *et al.*, 2010). While RS is usually high for high AC foods, high AC alone does not necessarily correlate with GI (Morita *et al.*, 2007; Choi *et al.*, 2010; Kumar *et al.*, 2018). However, a combination of high AC and RS in rice substantially reduces GI (De Guzman *et al.*, 2017).

Retrogradation potential

Pasting parameters are determined by the swollen granules and materials leached from the granules, and highly correlated with amylose content (Akintayo and Ashogbon, 2012; Chaiwanischiri et al., 2012). The intermediate amylose variety Minaangan had the highest total setback among the rice samples (2821 cP), while the waxy variety Ingopon had the lowest (396 cP) (Table 2). Total setback indicates RP, which is the quantitative measure of the recrystallisation of the gelatinised starch as it cools, and an indirect measure of starch retrogradation (Akintayo and Ashogbon, 2012). Generally, RP positively correlated with AC values such that high AC varieties had higher RP. Minaangan, however, despite not having the highest AC, had the highest RP. This may be partially attributed to its low crude protein content (5.80%). Protein retards retrogradation by preventing the recrystallisation of amylose during retrogradation (Chaiwanischiri et al., 2012).

Cooked rice hardness

Cooked rice hardness of non-parboiled samples at 1:1.5 rice:water ratio ranged from 0.97 - 2.07 kgf/cm², with most samples having soft to medium texture (Figure 1). Parboiling resulted in harder cooked rice, with increases of up to 203% in the high amylose variety PSB Rc10 in the same rice:water ratio. The texture of cooked parboiled rice improved drastically upon increasing the rice:water ratio to 1:2.5, even reducing the hardness value of the high amylose variety NSIC Rc 222 from 4.41 (Hard) to 1.79 kgf/cm² (Soft). The higher amount of cooking water needed to achieve comparable or better texture in parboiled rice could be due to formed RS3 which has a higher water holding capacity than granular starch (Sanz *et al.*, 2008).



Figure 1. Instron hardness of parboiled and non-parboiled rice varieties. VS: very soft; S: soft; M: medium, and H: hard.

Resistant starch

RS levels of raw rice samples were low (0.04 - 0.27 g/100 g dw) (Table 3). These results agree with those reported by Juliano (2007) that rice RS ranges from 0 - 1.0%. However, recent reports from Kumar *et al.* (2018) observed RS levels ranging 0.35 - 2.57 g/100 g in 24 Indian rice varieties. RS levels depend on several factors including cultivation environment,

variety, and processing (Birt *et al.*, 2013; Nissar *et al.*, 2017). Flour milling destroys structures entrapping starch granules, thus increasing particle size, and in turn, digestibility (Al-Rabadi *et al.*, 2009; Dhital *et al.*, 2016). Conventional cooking of rice with rice:water ratio only slightly increased RS levels (0.05 - 1.82 g/100 g).

	Resistant starch (mg/100 g sample dw) $(n = 2)$				
Rice variety	Non-parboiled		Parboiled		
	Raw Cooked T ₀	Cooked	D	Cooked	
		T ₀	Kaw	T_1	T_2
Ingopon	0.16 ± 0.01^{abc}	$0.06\pm0.01^{\text{g}}$	$0.18\pm0.02^{\text{d}}$	$0.08\pm0.01^{\text{e}}$	$0.12\pm0.02^{\text{g}}$
NSIC Rc 242	$0.11\pm0.06^{\text{bc}}$	$0.05\pm0.01^{\text{g}}$	$0.08\pm0.00^{\rm e}$	$0.12\pm0.02^{\text{e}}$	$0.05\pm0.01^{\text{g}}$
NSIC Rc 160	$0.04\pm0.01^{\circ}$	$0.30\pm0.01^{\rm f}$	$0.21\pm0.05^{\text{d}}$	$0.18\pm0.03^{\rm e}$	$0.51\pm0.02^{\rm f}$
Minaangan	0.18 ± 0.00^{ab}	$1.57\pm0.03^{\rm b}$	$1.07\pm0.00^{\rm a}$	$2.06\pm0.14^{\text{a}}$	$2.07\pm0.04^{\rm b}$
PSB Rc 14	0.13 ± 0.02^{bc}	$0.67\pm0.04^{\rm e}$	$0.42\pm0.02^{\rm c}$	$0.49\pm0.02^{\text{d}}$	$1.21\pm0.00^{\rm e}$
Imbuucan	0.16 ± 0.02^{abc}	$0.90\pm0.01^{\text{d}}$	$0.39\pm0.00^{\rm c}$	$1.09\pm0.01^{\rm c}$	$1.98\pm0.09^{\rm b}$
Kamanga	0.15 ± 0.00^{abc}	$0.96\pm0.01^{\text{d}}$	$0.48\pm0.02^{\rm c}$	$0.99\pm0.01^{\rm c}$	$1.83\pm0.01^{\circ}$
PSB Rc 10	$0.27\pm0.04^{\rm a}$	$1.25\pm0.00^{\rm c}$	$0.68\pm0.04^{\text{b}}$	$0.87\pm0.11^{\rm c}$	$1.65\pm0.01^{\text{d}}$
NSIC Rc 222	$0.27\pm0.07^{\mathrm{a}}$	1.82 ± 0.02^{a}	$0.70\pm0.02^{\rm b}$	$1.57\pm0.14^{\rm b}$	2.71 ± 0.02^{a}

Table 3. Resistant starch levels of non-parboiled and parboiled rice varieties.

T₀: non-parboiled rice cooked at 1:1.5 rice:water ratio; T₁: parboiled rice cooked at 1:1.5 rice:water ratio; T₂: parboiled rice cooked at 1:2.5 rice:water ratio. Values are mean \pm SD. Means followed by different lowercase superscripts within the same column are significantly different at p < 0.05.

RS levels slightly increased when samples were parboiled, especially in intermediate- and high-AC varieties (Table 3). The highest levels were reported in the high amylose NSIC Rc 222 for the non-pigmented sample (0.70 g/100 g), and the intermediate-amylose Minangan among the pigmented rice (1.07 g/100 g). Starch is the most affected rice component during parboiling with major changes occurring as gelatinisation and recrystallisation (Oli et al., 2014; Kale et al., 2017). Generally, higher amylose varieties had more RS after parboiling with positive correlation of AC with RS observed in previous reports (Choi et al., 2010; Englberger et al., 2010; Garcia et al., 2013). Amylose's linear helical chain could form compact structures with high crystallinity that limits enzymatic access (Hu et al., 2004; Syahariza et al., 2013).

Cooked parboiled rice generally had higher measured RS than non-parboiled rice (Table 3). Again, the highest RS levels were those of NSIC Rc 222 (2.72 g/100 g) and Minaangan (2.07 g/100 g). Hydrothermal treatments destroy RS1 (physically inaccessible starch) and RS2 (granular starch), but amylose re-associates into a semi-crystalline form during retrogradation which is totally impervious to amylase action (Faraj et al., 2004; Sajilata et al., 2006; Deckardt et al., 2013). Parboiling of rice induces repeated gelatinisation-recrystallisation of amylose, which initially happens as pre-gelatinisation during parboiling, and complete gelatinisation during cooking (Itoh and Kawamura, 1985; Kim et al., 2006). Generally, increased cooking water also increased RS content of parboiled samples. Additional cooking water allowed for further swelling of starch amorphous regions and disruption

of non-RS3 crystalline regions, thus resulting in higher RS formation (Sagum and Arcot, 2000). Previously formed RS3 before cooking has high thermal stability (117 - 125°C) before reverting to its digestible form (Sonia *et al.*, 2015), while RS5 could easily reform after cooking (Birt *et al.*, 2013). Kim *et al.* (2006) likewise observed similar RS-enhancing effect of increasing the amount of cooking water in relation to that of rice in their rice samples subjected to autoclaving, another heat-moisture processing treatment.

Physical and thermal properties of high RS flours

Figures 2 and 3 show the flour morphologies of Minaangan and NSIC Rc 222, respectively. In Minaangan, parboiling resulted in gelatinisation as observed through the loss of polyhedral structure, and reduction of particle size (Figure 2). Parboiling induces gelatinisation, which involves preferential amylose leaching and melting of crystallites (Lamberts et al., 2009). Thermal data suggested gelatinisation (18.70%)with partial lower gelatinisation enthalpy of parboiled Minaangan flour $(258.75 \pm 37.12 \text{ J/g})$ as compared to its non-parboiled $(317.55 \pm 14.02 \text{ J/g})$ counterpart. Gelatinised starches could undergo further retrogradation especially with the high RP (2,822 cP) of Minaangan relative to those of other rice samples. This could account for Minaangan's almost two-fold increase in RS when parboiled form is cooked (Table 3).

Granules of parboiled NSIC Rc 222 appeared to be intact, and no significant change in structure was observed (Figure 3), thus suggesting high crystallinity. Higher crystallinity provides more structural stability which makes granules more



Figure 2. Scanning Electron Microscope (SEM) micrographs at 6,000× magnification of raw non-parboiled (**A**) and parboiled (**B**) Minaangan rice flour.



Figure 3. Scanning Electron Microscope (SEM) micrographs at 6,000× magnification of raw non-parboiled (**A**) and parboiled (**B**) NSIC Rc 222 rice flour.

resistant to gelatinisation (Abd Elgadir *et al.*, 2009). Moreover, gelatinisation enthalpy of parboiled NSIC Rc 222 flour (271.15 \pm 8.27 J/g) was higher than its non-parboiled (248.1 J/g) counterpart. Wang *et al.* (2016) concluded in their study that the enthalpy of retrograded starch gels was greater than that of native starch due to the melting of recrystallised starch formed during retrogradation, and the residual crystallites remaining in the retrograded starch gels.

Conclusion

Parboiling was able to enhance carbohydrate quality of the local rice cultivars by increasing the amount of RS by as much as five-fold for the intermediate-AC variety Minaangan, and almost sixfold in NSIC Rc 222 in raw form. High starch crystallinity plays a part in starch gelatinisation and retrogradation, and subsequently in RS formation. Cooking of parboiled rice at appropriate rice:water ratios could deliver rice with high RS and textural quality preferred by most Filipinos. Consumption of cooked parboiled Minaangan and NSIC Rc 222 could be explored as lower glycaemic index foods to reduce postprandial glucose response of Filipinos.

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