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分辨米穀粉品質特性之方法

The methods to differentiate rice flour characteristics

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本論文係龔仁玲君（學號 R01641043）在國立臺灣大學食品科技研究所完成之碩士學位論文，於民國 103 年 7 月 22 日承下列考試委員審查通過及口試及格，特此證明

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中文摘要

米穀粉於食品加工領域的應用與日俱增，故須發展針對米穀粉品質檢測之參數指標。本研究之目的為建立分辨米穀粉特性之方法，藉此了解米穀粉之性質並作為米穀粉加工應用之參考。於本研究中，精白米預先被磨成粉後，測定基本組成、粒徑分佈、糊化性質、膨潤力、溶解度、滯留溶劑能力(solvent retention capacity)、破損澱粉、直鏈澱粉含量、α及β澱粉酶之活性及攪拌性質。最終以米穀粉做烘焙產品米麵包。研究結果顯示，蛋白質品質與破損澱粉為影響米穀粉攪拌性質之主要因子。添加1.5%之羥丙基甲基纖維素(hydroxypropylmethylcellulose, HPMC)至米穀粉中使能其在farinograph檢測中達到500 FU之標準。Farinograph之指標受到滯留乳酸及碳酸鈉之能力影響，而兩者又分別與蛋白質品質與破損澱粉有關。吸水率(water absorption)和攪拌耐受指標(mixing tolerance index)與滯留乳酸和碳酸鈉($r \geq 0.9$, $p < 0.001$)之能力呈正相關，然而擴展時間(development time)和攪拌彈性時間(stability time)與滯留乳酸($r \leq -0.9$, $p < 0.001$)之能力呈負相關。

米穀粉之烘焙產品結果顯示滯留溶劑能力(solvent retention capacity), 破損澱粉, 直鏈澱粉含量皆影響米麵包之性質. 其中影像米麵包體積的重要因素藉為破損澱粉及滯留溶劑能力(solvent retention capacity)的五碳聚醣(pentosan)。米麵包之高度、比體積及體積指標皆與滯留蔗糖之能力成正相關($r \geq 0.8$, $p < 0.05$)，然而體積指標與滯留碳酸鈉之能力成負相關($r \leq -0.8$, $p < 0.05$).。本研究所使用之方法能分析米穀粉之特性並作為品工業中米穀粉品質管制之指標。

關鍵詞: 米穀粉, farinograph, 滯留溶劑能力, 五碳聚醣, 米麵包

Abstract

Increased use of rice flour in the food industry has necessitated it to have a set of testing methods that will reveal rice flour characteristics to indicate rice flour quality. Therefore, the objective of this study is to develop differentiating testing methods in rice flour that will help in understanding rice flour properties and that will serve as guidelines in rice flour processing properties. In this study, milled rice were ground to flour, and flour specifications such as proximate composition, particle size distribution, pasting properties, swelling power and solubility, solvent retention capacities, starch damage, amylose content, alpha-amylase and beta-amylase activity, mixing characteristics were determined. Lastly, a bake test was also conducted. Seven commercial including fresh and aged rice samples were selected for testing. Protein quality and starch damage are observed to be the main determining factors in rice flour mixing characteristics. The addition of 1.5% hydroxypropylmethylcellulose to rice flour made it possible for rice flour mixing characteristics to reach a consistency of 500 FU and be studied in a farinograph. Farinograph parameters were greatly influenced by solvent retention capacities of lactic acid and sodium carbonate which are associated with protein quality and starch damage respectively. Water absorption and mixing tolerance index were statistical ($r \geq 0.9$, $p < 0.001$) positively correlated with lactic acid solvent retention capacity and sodium carbonate solvent retention capacity, while development time and stability time were statistical ($r \leq -0.9$, $p < 0.001$) negatively correlated with lactic acid solvent retention capacity and sodium carbonate solvent retention capacity.

Lastly, results for bake test indicated that solvent retention capacity, starch damage and amylose contentaffect rice bread properties. Pentosans and starch damage are key factors that influenced rice bread volume. Height, specific volume and volume index were statistical ($r \geq 0.8$, $p<0.05$) positively correlated with sucrose solvent retention capacity while volume index were negatively correlated with sodium carbonate solvent retention capacity($r \leq -0.8$, $p<0.05$). Therefore, the methods used in this study reveal characteristic rice flour properties and could serve as guidelines in rice flour quality control in the food industry.

Keywords: Rice flour, farinograph, solvent retention capacity, pentosan, gluten free bread, rice bread

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1. Introduction

Rice is a staple food for more than half of world population. It is a world-wide commodity and shows great diversity in properties due to its locality of production and variety. These variations significantly affect its utilization.

Rice is generally grown for its eating qualities as cooked rice. Therefore, literally hundreds of varieties of rice are grown all over the world for their unique characteristics. In accordance, several tests have been developed in measuring physicochemical properties of milled rice.

Rice flours, on the otherhand, is commercially used in babyfoods, meat products, separating powders for refrigerated pre-formed unbaked biscuits, dusting powders, breading, and formulations for pancakes and waffles.

In recent years, rice flour is being used in increasing numbers of novel foods such as tortillas, beverages, processed meats, puddings, salad dressing, and gluten-free breads because of its unique functional properties. (Kadan and Ziegler 1989; McCue 1997; Kadan and others 2001).

However, there is a no set of quality testing methods of rice flour available.Most of the available flour quality testing methods are developed specifically for wheat flour. Rapid visco analyzer and amylograph are used to measure wheat starch properties while farinograph, mixograph and alveograph are used to measure wheat gluten characteristics. Because of differences in composition, rice flour cannot directly adapt tests from that of wheat flour.

The objective of this study is to develop instrumental testing methods in rice flour that will help in understanding rice flour properties and in the same time serve as guidelines in rice flour characterization.

2. Literature Review

2.1. Rice

2.1.1. Origin

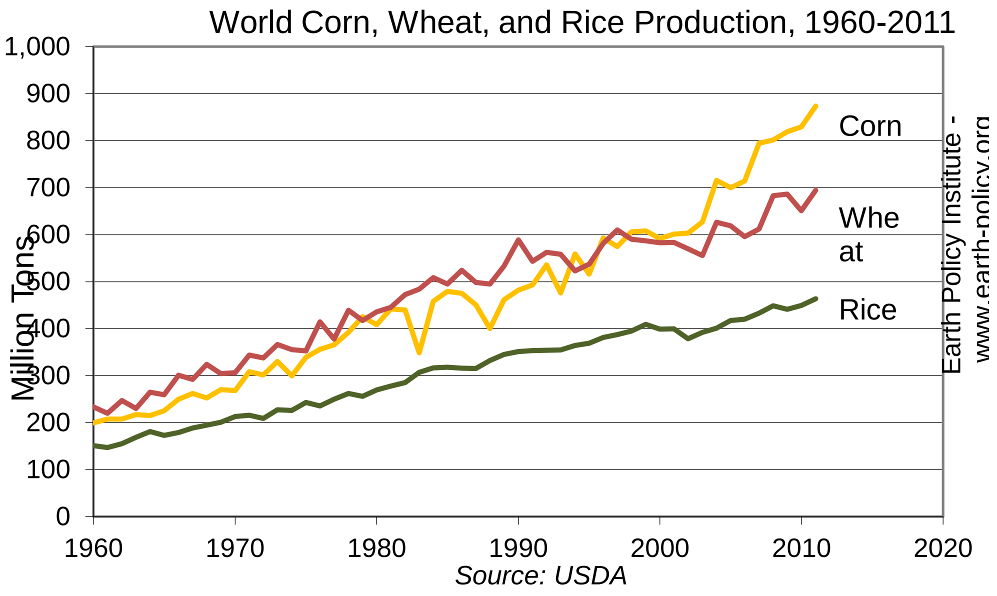
Rice (Oryza sativa) is one of world's oldest and leading food crop species. There are two major subspecies of Oryza sativa: the sticky, short grained japonica, and the nonsticky, long-grained indica variety.It is believed that the original ancestral species no longer exist and that present varieties have evolved from known species(Houston, 1972). Although there are many debates on its origins,rice is believed to have been domesticated approximately 9,000 years ago in China. Molecular evidence showed that indica and japonica were domesticated from the wild rice O.rufipogon(Molina et al., 2011). The domestication of rice is considered as one of the most important developments in human history(IRRI, 2013a).

2.1.2. Production

Rice varieties presently cultivated are mostly *O. sativa* species. It is generally considered a semiaquatic, annual, grass plant. World annual production of the top three cereals, corn, wheat and rice is seen in Figure 1. Rice is grown in more than a hundred countries, with a total harvested area in 2009 of approximately 158 million hectares, producing more than 700 million tons annually (470 million tons of milled rice). About 90% of the rice in the world is grown in Asia (nearly 640 million tons). However, a comparatively small amount of rice moves in the world trade because a high percentage of the worlds rice crop is consumed in the countries where it is produced (Houston, 1972).

Rice yields range from less than 1 ton per hectare under very poor rainfed conditions to more than 10 t/ha in intensive temperate irrigated systems. Small, and in many areas shrinking, farm sizes account for the low incomes of rice farm families.

Rice grows in a wide range of environments and is productive in many situations where other crops would fail. Rice-growing environments are based on their hydrological characteristics and include irrigated, rainfed lowland, and rainfed upland(IRRI, 2013b).


World Corn, Wheat, and Rice Production, 1960-2011

Source: USDA

Figure 1. World Corn, Wheat and Rice Production from 1960 – 2011

2.1.3. Composition

2.1.3.1. Starch

Milled rice, after removal of the bran layers and germ, consists entirely of endosperm with starch (90% dry basis) as the predominant component(Cauvain and Young, 2009).

2.1.3.2. Protein

Protein is the second most abundant constituent in rice. Osborne protein fractions in rice is seen in Table 1, with the glutelin fraction being the highest. The glutelin fraction in rice is oryzenin, the main storage protein in rice. It is composed of subunits that are linked by both intra- and inter- molecular disulphide bridges. During storage of rice, the molecular weight of oryzenin increases significantly, which correlates with an increase in disulphide bonding. The decrease in solubility is thought to explain the decrease in stickiness observed in rice(Martin and Fitzgerald, 2002).

Table 1. Osborne Proteins in Rice and Wheat

	Osborne Protein Fractions (% total)				
	Albumin	Globulin	Prolamin	Glutelin	Lysine (g/16g of N)
Wheat	5-15	5-10	40-50	30-45	2.3
Rice	2-5	2-10	1-5	75-90	3.8

2.1.4. Nutrient comparison of rice and other cereals

Compared with other cereals, rice is comparatively high in caloric value, nitrogen free extract, niacin and is comparatively low in protein, although rice protein has a fairly good balance of the essential amino acids(1985).

2.1.5. Rice flour Specifications

2.1.5.1. Starch

Structures of cakes are mainly based on the properties of starch in flour(Cauvain and Young, 2009).

2.1.5.2. Amylose content

Amylose content of rice starch is an important quality factor. It is directly related to volume expansion and water absorption during cooking, and hardness, whiteness and dullness of cooked rice. High amylose grains also cook dry, are less tender and become harder after cooling(1985).

2.1.5.3. Gelatinization and Pasting Properties

Gelatinization properties reflect starch properties and its influence on the quality of cooked rice.Gelatinization temperature is the temperature at which the rice absorbs water and starch granules swell irreversibly. It also determines the time required for cooking rice. Rice with high gelatinization temperature tends to require more water and time to cook than those with low or intermediate gelatinization temperature (IRRI, 2013c).

The gelatinization of starch is a key contributor to the structural properties of cakes and the evaluation of the pasting properties of cake flours is therefore an important test(Cauvain and Young, 2009).

Pasting properties refer to the viscosity changes that occur during heating, holding and cooling of slurry rice flour with water. These are recorded in the form of a pasting curve. Pasting curves of 20% rice flour slurry provide an approximate indication of the gelatinization temperature while 10% rice flour slurry help predict rice starch behavior during processing of rice-based products. .

2.1.5.4. Batter Viscosity

Batter viscosity is a very important property since it affects the flow of batter to achieve the final product during baking (Cauvain and Young, 2009). However, different factors such as level of added water and batter temperature affect viscosity (Cauvain and Young, 2008) In cakes, the higher level of aeration comes in part from the greater quantity of air incorporated during mixing of cake batters and in part from the action of the leavening agents which are present.

2.1.5.5. Gel consistency

Gel consistency measures the tendency of cooked rice to harden after cooling and it is used to differentiate between high-amylose rice types with differing processing qualities. Gel consistency indicate textural properties of cooked rice. Soft gel consistency indicates a tender cooked rice while hard gel consistency indicates a hard(1985) .

2.1.5.6. Alpha-amylase

Alpha-amylase attacks alpha-glucan molecules (starch and glycogen) hydrolytically resulting to the liquefaction and dextrinization of starch which produces glucose, maltose and other oligosaccharides of varied chain-lengths.

Freshly harvested rice grain has a relatively high alpha-amylase which decreases during storage. It is mainly concentrated in the bran and residual alpha-amylase activity in milled rice is low and readily inactivated during cooking (Desikachar and Subrahmanyan, 1960)(Houston, 1972; 1985).

2.1.5.7. Particle size

Particle size of rice flour greatly affects pasting properties, gel consistency after cooking and palatability of rice flour butter cakes (Nishita and Bean, 1982; Kaletunç and Breslauer, 2003). Similarly, particle size of wheat flour is related to its protein content, utilization and starch damage after milling. Flour particle size is important in evaluation of flour quality, processing behavior and end-product appearance (Kaletunç and Breslauer, 2003).

2.1.5.8. Water absorption

The water absorption capacity of rice flour decreases as it ages(Williams, 2005).

It was reported that freshly harvested rice has a higher water absorption compared to a year-old rice (Desikachar, 1956).

2.1.6. Utilization

2.1.6.1. Milled rice

Milled rice grain is used mainly as direct food consumption, unlike most grains, because of its high cost(BeMiller and Whistler, 2009).Milled raw is consumed mainly as boiled rice. However it is also marketed as precooked, canned, dried, and puffed for breakfast cereals(Maclean *et al.*, 2002).

2.1.6.2. Rice bran

Rice bran which consists about 5% to 8% of the grain weight, is mainly used as an animal feed due to its bitter taste that develops from the lipolytic action of enzymes present in rice oil(BeMiller and Whistler, 2009). It is also used as a pickling medium, a growing medium for mushrooms and for some enzymes, as well as for flours, concentrates, oils, and dietary fiber(Maclean *et al.*, 2002).

2.1.6.3. Hulls and husks

Rice hulls are tough, fibrous, abrasive, have high ash content and low nutritive value (Hoseney, 1986). It has limited applications, such as in animal feed, in chicken litter, as ajuice pressing aid and as fuel(Maclean *et al.*, 2002).

2.1.6.4. Rice polish

Rice polish has a high nutritive value. It is commonly used as plant fertilizer and animal feed. Only a few however utilize rice polish in baby and health foods(Houston, 1972).

2.1.6.5. Rice oil

Rice oil is odorless and tasteless and is considered a high-grade vegetable cooking oil (Houston, 1972). It can be extracted from stabilized and unstabilized bran. Rice oil from stabilized bran is suitable for human food use. Currently, stabilized rice bran is being used in baked goods, energy bars and protein fortification of powdered drink formulations(BeMiller and Whistler, 2009).

2.1.6.6. Rice flour

Rice flours are made from broken milled rice. They have diverse chemical compositions and properties have diverse properties associated with variety and processing history (Houston, 1972). It is used in extrusion-cooked foods, puddings, breads, cakes and crackers noodles and rice paper; fermented foods and vinegars; rice starch; and syrups. Nowadays, a lot of processed foods such as cereals, packaged mixes, pet foods, rice cakes and baby foods incorporate rice flour.In addition, an increasing number of novel food products made from rice flour is attracting interest(Maclean *et al.*, 2002).

2.1.6.7. Rice starch

Rice starch production is quite limited because of the high cost of brewer’s rice. Milled raw and parboiled rice is consumed mainly as boiled rice. Various rices with specific amylose-amylopectin ratios are used in specific rice products and in various regions.Rice starch from white or brown rice exhibits distinct characteristicand acts a functional ingredient in many food products (BeMiller and Whistler, 2009).

2.2. Wheat

2.2.1. Wheat flour specification testing methods

2.2.1.1. Protein

2.2.1.1.1. Farinograph

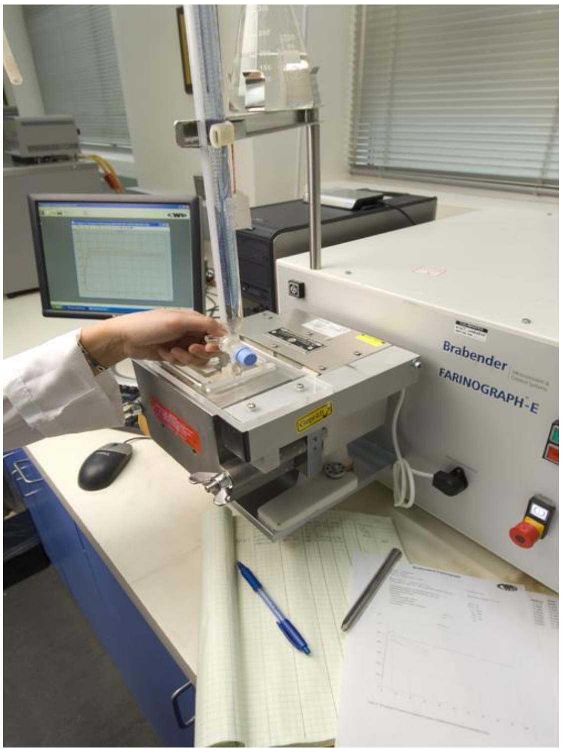
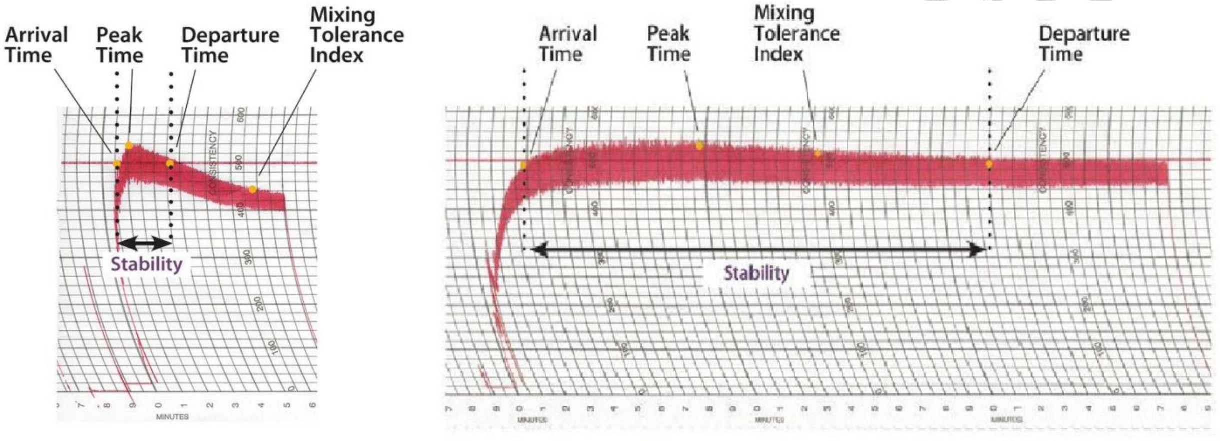


Figure 2. Farinograph

(Kansas State University, 2008)



Weak Gluten Flour **Strong Gluten Flour**

Figure 3. Farinograph curves for weak and strong gluten flours

(Kansas State University, 2008)

The Farinograph determines the mechanical resistance of dough to the mixing action of the paddles. It is the most popular and commonly used instrument in measuring the quality of flour. It evaluates water absorption of flours and measure stability and other characteristics of doughs during mixing.

Absorption,expressed in percentage, is the amount of water added until the dough reaches 500BU. This is an important parameter because it directly relates to the quality of the end product.

Farinograph curve results are shown in Figure 3. Arrival time indicates the rate of flour hydration; peak time indicates the dough development time; departure time indicates dough consistency during processing and mixing tolerance index indicates degreee of softenung during mixing. Dough development time is directly proportional to the degree of starch damage and inversely proportional to starch granule size. Mixing tolerance is proportional to the strength of gluten(AACC, 2000; Kansas State University, 2008;

Dapčević Hadnađev et al., 2011) .

Extensigraph

Figure 4. Extensigraph

(Kansas State University, 2008)

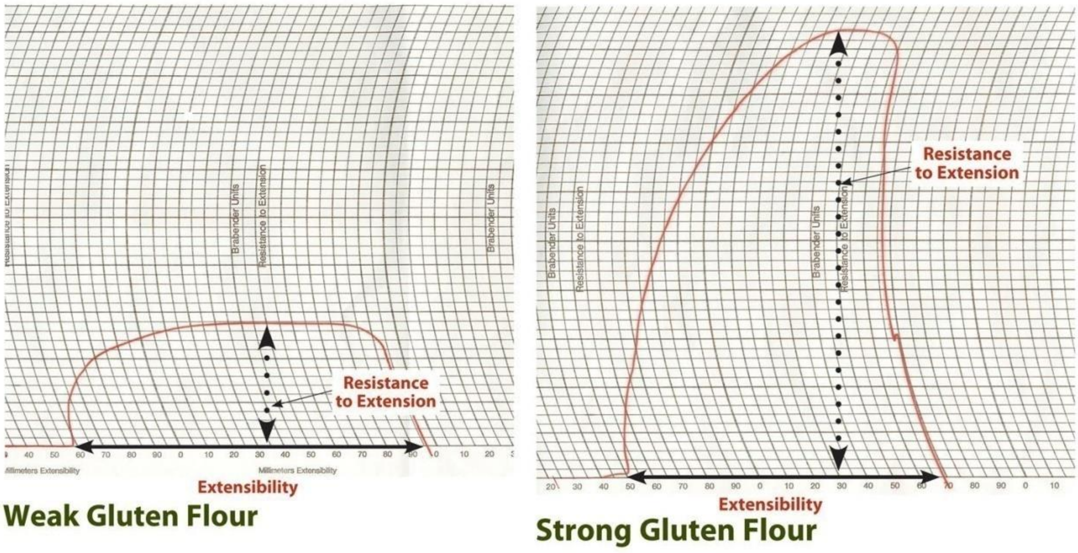


Figure 5.Extensigraph curves of weak and strong gluten flour

(Kansas State University, 2008)

The extensigraph measures the dough resistance and extensibility by recording the force required to pull a hook through a cylindrically shaped piece of dough on a curve. Characteristics of force-time curves are used to assess the general quality of flour and to monitor the effects of added ingredients.

Curve result includes resistance to extension, extensibility, R/E ratio and area under the curve. As seen in Figure 5, weak gluten flour has a lower resistance to extension than strong gluten flour(AACC, 2000; Kansas State University, 2008; Dapčević Hadnađev *et al.*, 2011).

Alveograph

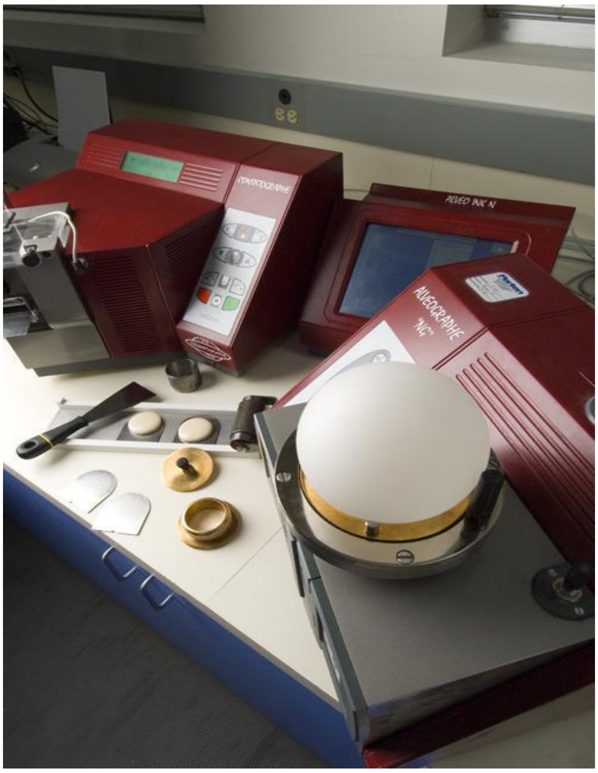


Figure 6. Alveograph

(Kansas State University, 2008)

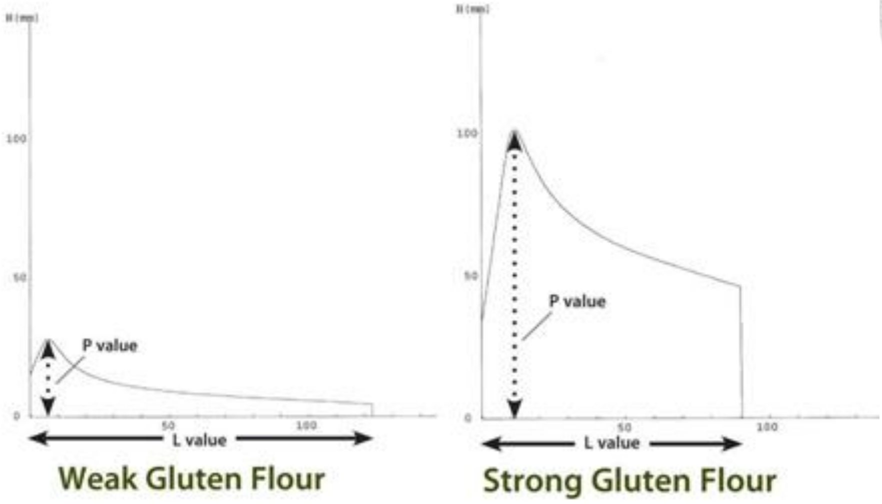


Figure 7. Alveograph curves of weak and strong gluten flour

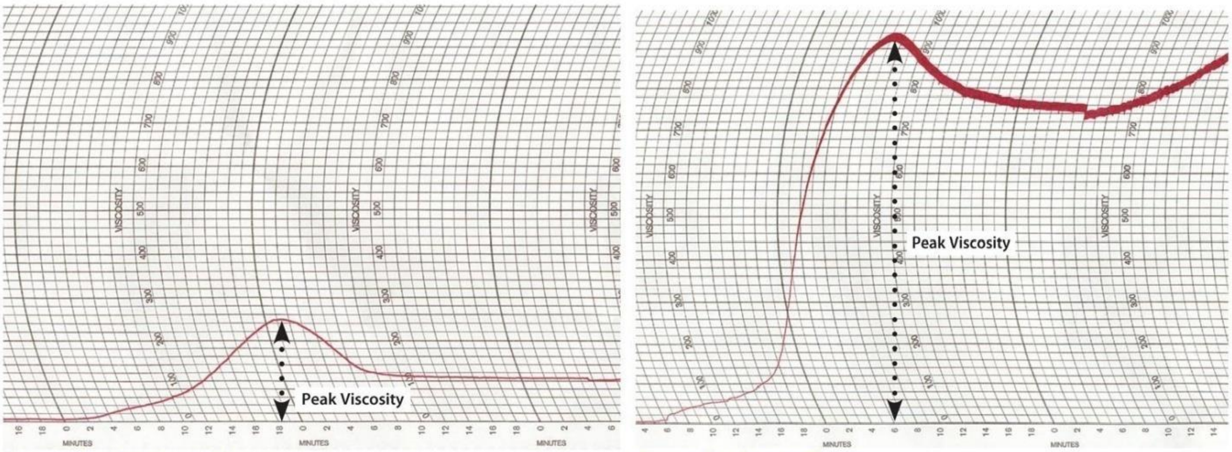
The alveograph determines gluten strength by measuring resistance of a sheet of dough with definite thickness to extension and extent to which it can be stretched after blowing of air to it by air pressure. The internal pressure of the bubble is graphically recorded as a curve on a paper(AACC, 2000; Kansas State University, 2008).

The phases of the Alveograph simulate sheeting, rounding and molding of the dough pieces during the baking process. The five dough pieces are prepared by mixing and extruding, followed by shaping into small disks which are then left for resting during 20 minutes. After that, the air is blown under the disk at a constant rate creating a bubble. The curve will have a P, L value as shown in Figure 7, and W value which is the area under the curve. Since dough made from strong gluten flour requires more force to break and blow the bubble, the resulting curve has a high P and W and low or medium L values. Weak gluten shows the opposite (Kansas State University, 2008; Dapčević Hadnađev et al., 2011)

Amylograph

Figure 8. Amylograph

(Kansas State University, 2008)



Sprouted Wheat Flour **Sound Wheat Flour**

Figure 9. Amylograph curves of sprouted and sound wheat flour

(Kansas State University, 2008)

Amylograph measures viscosity and enzyme activity by measuring the resistance of flour and water slurry to the stirring action of pins or paddles during heating.

Curve result includes gelatinization temperature, peak viscosity and temperature at peak viscosity and this is influenced by starch properties, milling conditions, alpha-amylase enzyme activity.

A high alpha-amylase enzyme activity indicates sprouting in wheat. The amylograph value, or peak viscosity, also called malt index, is inversely proportional to alpha-amylase activity as seen in amylograph curves for sprouted and sound wheat flour shown in Figure 9.

Alpha-amylase largely causes the liquefying of viscous starch paste during heating resulting to a low peak viscosity. High alpha-amylase enzyme activity produces a sticky dough that might cause problems during processing and result to products with poor color and weak texture(AACC, 2000; Kansas State University, 2008; Dapčević Hadnađev et al., 2011).

Glutomatic

Figure 10. Glutomatic

(Kansas State University, 2008)

Glutomatic is an automatic gluten washing apparatus and measures wet gluten content of flour by washing the flour with salt solution to remove the starch and other solubles followed by centrifuging on an especially constructed sieve to force the wet gluten through the sieve.The residue remaining after washing is the wet gluten and the percentage of gluten remaining on the sieve is defined as the Gluten Index, which is directly proportional to its strength. These results provide quantity and quality of gluten present in flour (AACC, 2000; Kansas State University, 2008).

Mixograph

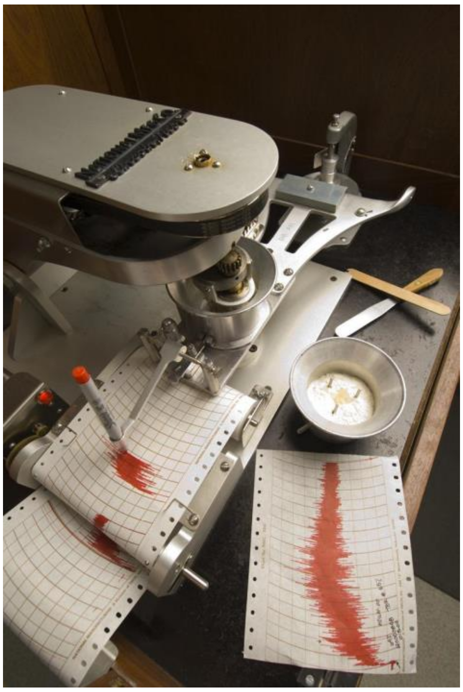
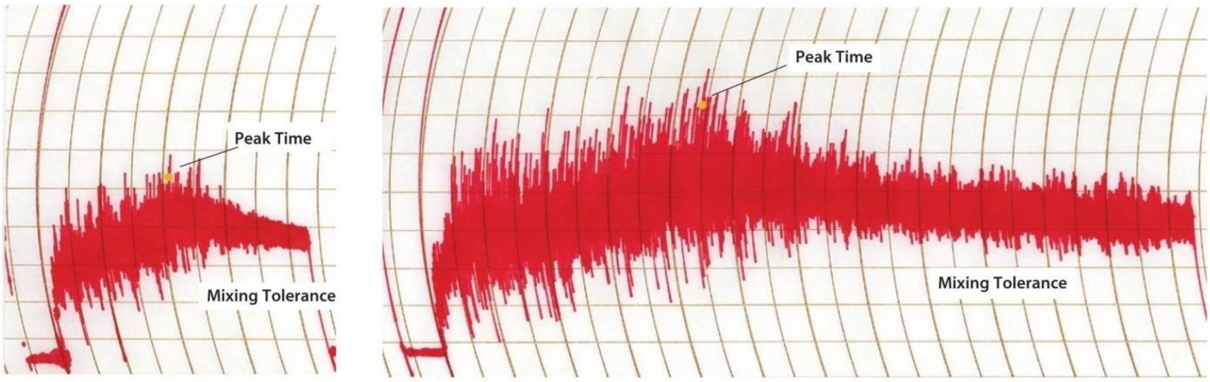


Figure 11. Mixograph

(Kansas State University, 2008)



Weak Gluten Flour

Strong Gluten Flour

Figure 12. Mixograph result of weak and strong gluten flour

(Kansas State University, 2008)

Mixograph measures dough and gluten properties of a flour by measuring the resistance of a dough against the mixing action of pins. Results include peak time and mixing tolerance indicating gluten strength, dough development time, mixing tolerance and bake absorption as seen in Figure 12. Results obtained are quite similar to farinograph except that it operates on a constant amount of water and requires less sample and time (AACC, 2000; Kansas State University, 2008; Dapčević Hadnađev et al., 2011).

Solvent Retention Capacity

Figure 13. Solvent Retention Capacity

(Kansas State University, 2008)

Solvent retention capacity (SRC) is the weight of solvent held by flour after centrifugation. Four solvents which associate with different flour characteristics are used. Sucrose SRC associate with pentosan components, sodium carbonate with damaged starch, lactic acid with gluten properties and water with all adsorbing components. The combined pattern of the four SRC values establishes a practical flour quality/functionality profile useful for predicting baking performance and specification conformance(AACC, 2000; Dapčević Hadnađev et al., 2011).

2.2.1.2. Starch

Rapid Visco Analyzer

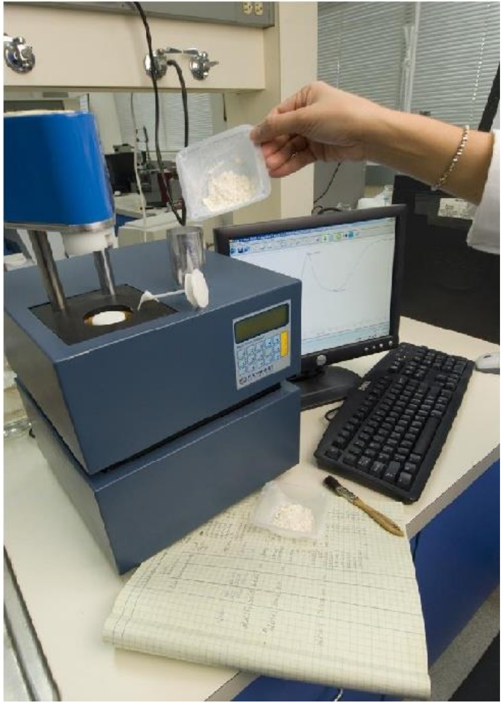


Figure 14. Rapid Visco Analyzer

(Kansas State University, 2008)

Rapid visco analyzer measures viscosity and enzyme activity by measuring the resistance of flour and water slurry to the stirring action of paddles during heating. When the flour slurry is heated, starch granules swell and will make the slurry viscous. This will result to a higher resistance to the paddle and high peak viscosity.

A high alpha-amylase activity indicates sprouting in wheat and willcause the liquefying of viscous starch paste during heating resulting to a low peak viscosity(AACC, 2000; Kansas State University, 2008; Dapčević Hadnađev et al., 2011).

2.2.1.3. Enzyme Activity

Falling number

Figure 15. Falling Number Apparatus

(Kansas State University, 2008)

Falling number (FN) expressed in seconds measures the effect of the present level of alpha-amylase activity in flour rather than the amount. This is based on the ability of alpha-amylase to liquefy a starch gel. Too much enzyme activity indicates that increased level of sugar is present rather than of starch. Starch contributes to the structure of the bread after gluten. Too much sugar will lead to a sticky dough that is hard to process. A high falling number such as above 300 seconds indicates minimal enzyme activity and sound quality flour, while a low falling number below 300 seconds indicates enough enzyme activity and sprout-damage(AACC, 2000; Kansas State University, 2008; Dapčević Hadnađev et al., 2011)

3. Materials and Methods

3.1. Samples

Details of the rice samples used in this study are seen in Table 2. All samples were obtained from commercial sources in milled rice grain form, except for R7 which was in the form of milled rice flour.

Table 2. Rice samples

Samples*	Abbreviation	Source
Chang Hsien	R1	Yeedon Enterprise Co., Ltd.
Chang Hsiang	R2	Hua-dong Co., Ltd.
Tainan 11	R3	China Grain Products Research and Development Institute
Chung Hsing	R4	The Union Rice Co., Ltd.
Kung Liang	R5	China Grain Products Research and Development Institute
Tainung Sen 14	R6	China Grain Products Research and Development Institute
Lian Hwa	R7	A commercial rice flour from Lian Hwa Enterprise Co. Ltd.,

*All samples, except R7, were received as milled rice and ground in laboratory scale Cyclotec mill.

3.2. Grinding of rice grains

Commercial rice samples R1 to R6in Table 2 were ground into flour using Cyclotec mill with 1.0 mm sieve (Cyclotec 1093, Tecator, Sweden).Sample R7 was not ground because it is already in rice flour form.

3.3. Rice flour specification methods

3.3.1. Proximate analysis

Ash content was determined following Ash content for soy flours (AACC Approved Method 08-16). Fat was determined following Crude fat for flours (AACC Approved Method 61-01) with modification of using 95% Ethanol (Echo Chemical, Taipei, Taiwan) as extraction solvent. Protein was determined by multiplying 5.25 by the nitrogen obtained after sample combustion in LECO FP-628 Nitrogen Determinator (LECO Corporation, St. Joseph, Michigan, USA)

3.3.2. Particle Size Distribution

Particle size dristribution was determined by using light scattering and by using different mesh size sieves. Samples were suspended in isopropyl alcohol (JT Baker, Sweden), and analyzed using a diffraction particle analyzer Coulter LS 100 (Micro Volume Module Cell, Coulter Electronics, USA). 25 grams of samples were pass through 30, 60, 80 and 100 mesh sizes. Flour retained in each sieve was weighed and recorded.

3.3.3. Pasting properties

Pasting propertieswere determined using Brabender Viscograph E (Brabender GmbH & Co. KG, Germany) followingthe Amylograph Method for milled rice (AACC Approved Method 61-01). 10% rice flour slurry was subjected to the following temperature cycle: held at 35^{o}C for 5 minutes, heated with a rate of 1.5^{o}C per minute to 95^{o}C, held for 20 minutes, and cooled with a rate of 1.5^{o}C per minute to 50^{o}C.

3.3.4. Swelling Power and Solubility Index

0.10 g (dry basis) of rice flour was placed in 50 mL centrifuge tubes (Falcon tube). Flours were hydrated with 10 mL of distilled water and placed in water bath held at different temperatures (60, 70, 80 and 90^{o}C) for 30 minutes. Samples were vortexed after 2, 4, 6, 8, 10, 15, and 25 minutes in water bath to maintain suspension of flour in water. Centrifuge tubes were placed in a room temperature water bath and wereallowed to cool for 30 minutes. Samples were then centrifuged (3000×g , 15 min) and the supernatant wasdried at 105 °C until constant weight. The Water solubility index and swelling power were calculated as described in (Li and Yeh, 2001; Cozzolino et al., 2013).

Water Solubility Index (%) = (W1 / flour dry weight) X100
Swelling Power (g/g) = W2 / [flour dry weight x (100% - W1)]
Where, W1 is the supernatant dried to a constant weight
(105 °C); and W2 is the weight of the sediment.

3.3.5. Solvent Retention Capacity Profile

Solvent retention capacity profile of rice flour was determined using four solvents: deionized water, 50 % sucrose solution, 5% lactic acid and 5% sodium carbonate (AACC Approved Method 56-11). 5.0 grams of rice flour (with known moisture content) was placed in 50 mL centrifuge tubes (Falcon tube). Flours were hydrated with 25 grams of the solvent and placed in a water bath at 30^{o}C) for 20 minutes. Samples were vortexed after 5, 10, 15, and 20 minutes in water bath to maintain suspension of flour in water. Samples were then centrifuged (1000xg, 15 minutes). Tubes were inverted and drained on a paper towel for 10 minutes. Solvent retention capacity was calculated as follows:

% SRC = [(gel weight / flour weight) X (86/(100-%flour moisture content)) – 1] X 100

3.3.6. Starch damage

Damaged starch wasdetermined using the Megazymekit (K-SDAMMegazyme International Ireland, Wicklow, Ireland). Damaged starch granules were hydrated; followed by hydrolysis to maltosaccharidesand limit dextrins by fungal alpha-amylase. Amyloglucosidase is then used to convert dextrins to glucose, which is specifically determined spectrophotometrically after glucose oxidase/peroxidase treatment (AACC Approved Method 76-31).

3.3.7. Amylose content

Amylose content was determined using colorimetric determination at 620 nm of the greenish-blue starch-iodine complex developed in ammonium buffer following modifications (Juliano *et al.*, 2012)based on amylose content determination (AACC Approved Method 61-03). Rice amylose was isolated from rice starch by complexing amylose with butanol (Takeda *et al.*, 1986) and used as standard.

3.3.8. Alpha-amylase and Beta-amylaseactivities

The enzyme activities were measured by using Megazyme Kit (K-MALTA, Megazyme International Ireland, Wicklow, Ireland)with colored substrates. One unit of enzyme activity is defined asthe amount of enzyme that causes the hydrolysis of one μmole ofthe specific substrate per minute (Haros *et al.*, 2002).The alpha-amylase and beta-amylase activity were measured by using a blocked p-nitrophenyl maltoheptaoside(BPNPG7, Megazyme International Ireland, Wicklow, Ireland) and p-nitrophenyl beta-maltotrioside (PNP β -3G, Megazyme International Ireland, Wicklow, Ireland)) respectively as substrate(Santos and Riis, 1996; McCleary *et al.*, 2002).

Falling numbers were determined(AACC Approved Method 56-81) using a falling number apparatus (Model 1500, Perten Instruments, Huddinge, Sweden).

3.3.9. Mixing characteristics

Mixing characteristics were measured following a farinograph (Brabender OHG, Kulture, 51-55, d- 47055, Duisburg, Germany)following the method farinograph test (AACC Approved Method 54-22) with some modifications. Rice flours were first blended with 1.5% Hydroxypropylmethylcellulose (SFE-4000, Shin-Etsu chemicals

Ltd., Japan) by sifting together five times.

3.3.10. Bake test

Rice bread was chosen for this test. Rice bread was made using Panasonic bread machine’s setting for rice bread containing no wheat flour (SD-BM 103T, Panasonic Co., Ltd., Taiwan). Recipe provided in the manual was followed with 300 g rice flour, 5 g salt, 9 g sugar, 10 g oil, 50 g fructose, 220 g water and 3 g yeast. Zai-lai-mi rice flour was used as control rice flour (Sunright Co., Ltd, Taiwan). As for the wheat flour control, wheat flour bread machine’s setting for wheat bread was used with high gluten flour (Milk international Co., Ltd., Taiwan).

Bread volume was determined following the Rapeseed displacement method(AACC Approved Method 10-05).

Rice breadwere cut vertically. 20 mm thick slice from the center was used to measure bread parameters. Parameters were measured following the layer cake measuring chart (AACC Approved Method 10-91) with some modifications. Height of bread at vertical lines B, C and D to the nearest 1.0 mm were read. These lines were designated as illustrated in Figure 16 for calculations of volume index, symmetry index and uniformity index

Volume Index = B + C + D

Symmetry Index = 2C – B – D

Uniformity Index = B - D

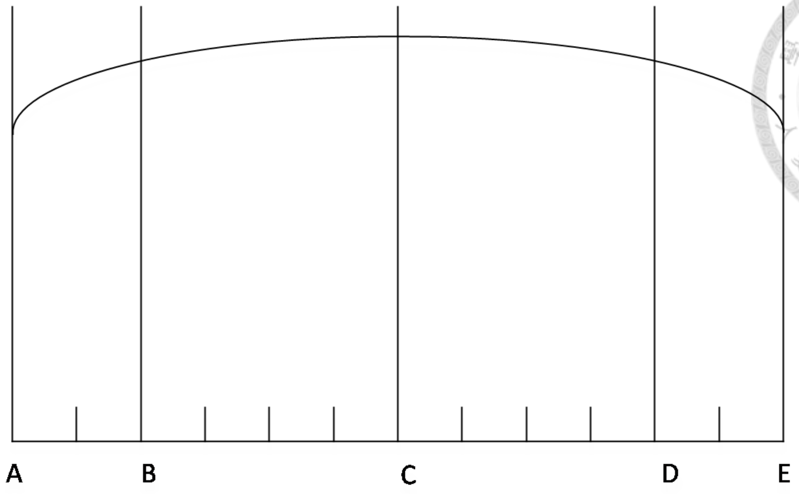
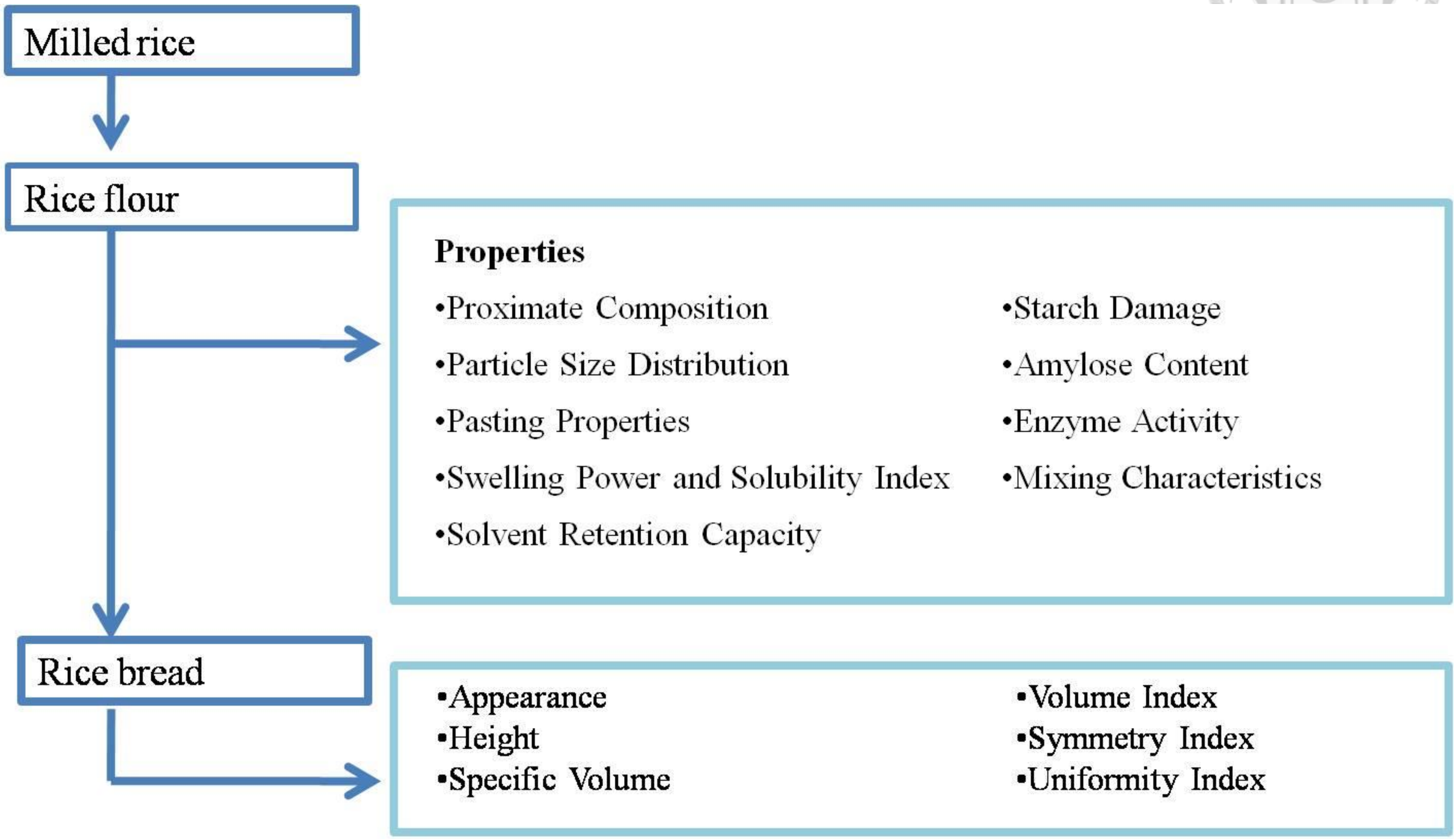


Figure 16. Template for measuring bread

4. Experimental Design



Milled rice

Rice flour

Properties

- Proximate Composition
- Particle Size Distribution
- Pasting Properties
- Swelling Power and Solubility Index
- Solvent Retention Capacity
- Starch Damage
- Amylose Content
- Enzyme Activity
- Mixing Characteristics

Rice bread

- Appearance
- Height
- Specific Volume
- Volume Index
- Symmetry Index
- Uniformity Index

5. Results and Discussion

5.1. Proximate Composition

Results for the proximate composition of rice flour samples are shown below. The protein, ash and fat content of selected rice samples were in the range of 7.0 – 11.6, 0.4 -0.5, and 0.7 – 2.1% respectively. Generally, the protein content of indica rice is higher than that of japonica rice (Sun *et al.*, 2008). However, this trend was not observed in this set of indica (R1, R2, R5, R6) and japonica (R3, R4, R7) rice samples. The reason for this could be due to variety differences such as the so called soft indica R1 and R2. Soft indica is an indica rice but with a low amylose content.

Table 3.Proximate composition of rice flours (on dry basis)

	Protein (%)	Ash (%)	Fat (%)
R1	8.75 ± 9.10*	0.49 ± 0.02	2.10 ± 0.40
R2	7.91 ± 0.50	0.48 ± 0.01	0.80 ± 0.10
R3	7.05 ± 0.00	0.50 ± 0.00	0.90 ± 1.50
R4	8.32 ± 8.32	0.47 ± 0.02	0.70 ± 0.20
R5	11.57 ± 0.52	0.52 ± 0.03	1.00 ± 0.20
R6	7.70 ± 7.91	0.38 ± 0.01	1.30 ± 0.20
R7	9.10 ± 0.49	0.52 ± 0.01	1.60 ± 0.10

*Means ± standard deviations, n=3

5.2. Particle Size Distribution

R7 had the finest particle size distribution in this set of samples. As seen inTable 4, 66.2% of R7 was in the range of 0-50μm and 99.8% in 0-150μm inTable 5. Particle size distribution of samples R1 to R6 was relatively the same, but differed much from that of R7. Same laboratory cyclone mill was used to grind samples R1 to R6 into rice flour (page 25) which lead to their similarity. R7, on the other hand was received in rice flour form so no further grinding was done.

Table 4.Particle Size Distribution of rice flours by light scattering

	Particle size distribution (%)			
	0-50 μm	51-100 μm	101-200 μm	201-2000 μm
R1	54.3*	21.3	15	9.4
R2	43	22.4	22.9	11.7
R3	58.3	24.3	13.8	3.7
R4	47.6	20.9	20.8	10.6
R5	45.2	22.7	27.6	4.6
R6	43.7	19.4	25.5	11.4
R7	66.2	20.9	10.9	2

*Means, n=3

Table 5.Particle Size distribution of rice flours by mesh size

	particle size distribution (%)				
mesh size	< 100	> 100	> 80	> 60	> 30
	0-150 µm	151-176 µm	177 µm-249	251-594 µm	> 595 µm
R1	55.1*	13.4	11.6	19.6	0.3
R2	55.1	14.9	11.7	18.2	0.2
R3	56.6	12.7	11.1	19.3	0.3
R4	62.4	11.1	11.5	14.8	0.2
R5	60.0	12.4	10.7	16.5	0.4
R6	67.2	10.5	9.0	13.3	0.1
R7	99.8	0.2	0.0	0.0	0.0

*Means, n=3

5.3. Pasting Properties

By recording viscosity changes that occur during heating, holding, and cooling of 10% rice flour slurry, pasting properties help predict rice starch behavior during processing of rice-based product.

As seen, each rice flour sample achieved a distinct pasting profile. There is an observed distinction for aged rice flour K3 and S7. They had the lowest peak viscosity and breakdown as seen in Table 6. This low peak viscosity and breakdown indicated that the starch granules in aged rice flour was more resistant to swelling and more resistant to rupture after cooking, respectively (Noomhorm *et al.*, 1997; Teo *et al.*, 2000). There is a reported significant increase in the number of disulfide bonds and the average molecular weight of the major protein in rice - oryzenin(Chrastil, 1990). These disulfide bonds possibly restrict starch granuleswelling during gelatinization and make the swollen granule less susceptible to disruption by shear (Hamaker and Griffin, 1993).

In addition, pasting properties: hot paste viscosity ($r=0.889$, $P<0.01$), cold paste viscosity($r=0.938$, $p<0.01$) and setback ($r=0.951$, $p<0.01$) were found to be positively correlated with amylose content while breakdown ($r=0.869$, $p<0.01$) is negatively correlated with amylose content.

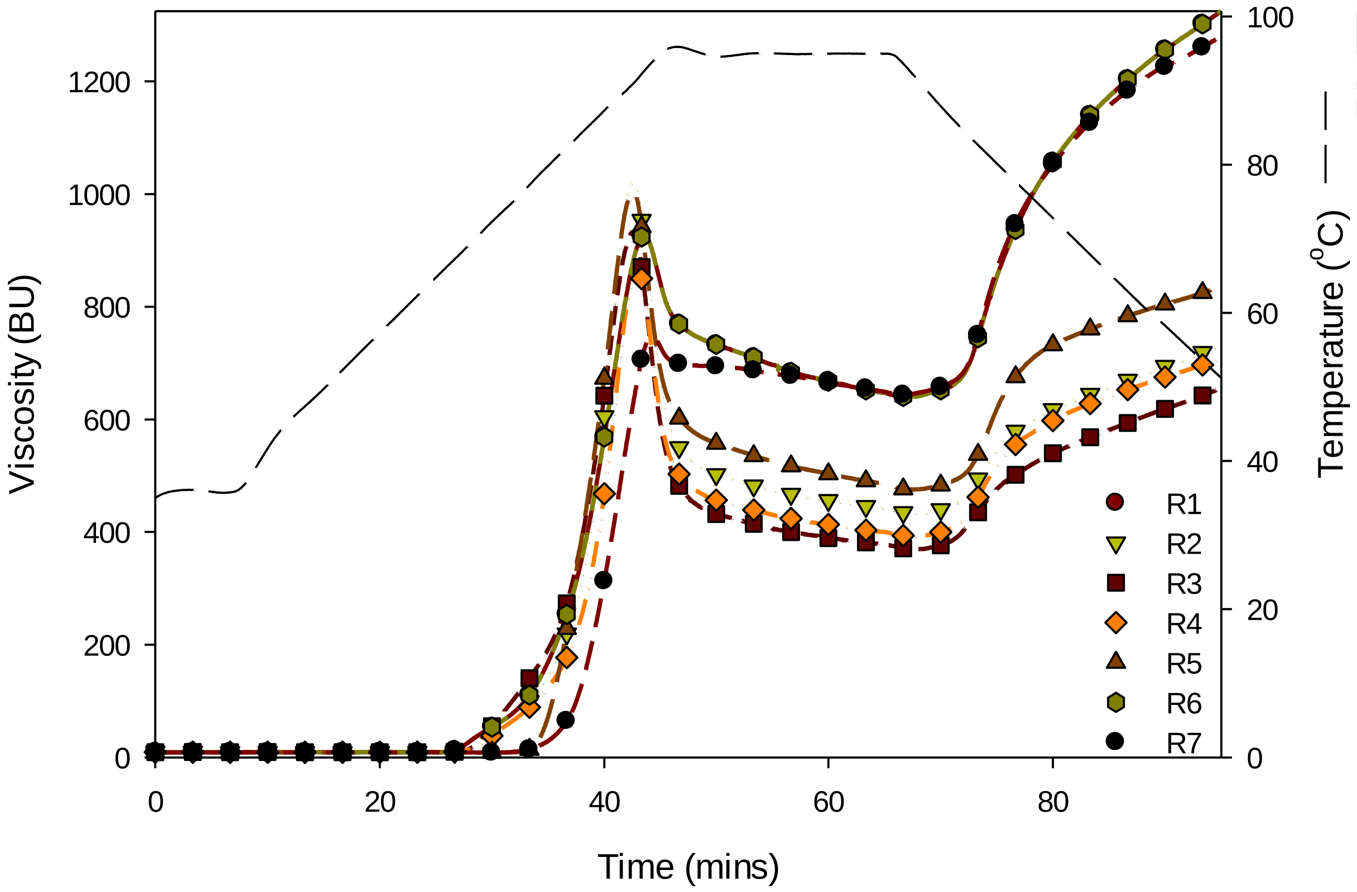


Figure 17. Pasting profile of rice flours

Table 6.Pasting properties of rice flours

	Peak Viscosity** (BU)	Hot paste viscosity (BU)	Cold paste viscosity (BU)	Breakdown (BU)	Setback (BU)
R1	*1021.3 ± 6.8 ^{a}	441.7 ± 3.9 ^{c}	732.7 ± 5.2 ^{e}	579.7 ± 3.3 ^{a}	291 ± 1.4 ^{f}
R2	1006.3 ± 4.8 ^{b}	485.7 ± 0.9 ^{b}	836.3 ± 0.9 ^{c}	520.7 ± 5.19 ^{c}	350.7 ± 0.9 ^{d}
R3	933 ± 11.3 ^{c*}	378 ± 0.8 ^{f}	655.3 ± 1.7 ^{g}	555 ± 10.6 ^{b}	277.3 ± 1.2 ^{g}
R4	865.7 ± 3.7 ^{e}	400 ± 6.5 ^{d}	708.7 ± 11.4 ^{f}	465.7 ± 3.09 ^{e}	308.7 ± 5 ^{e}
R5	748.3 ± 5.6 ^{f}	649 ± 2.4 ^{a}	1280.7 ± 5.9 ^{b}	99.3 ± 3.3 ^{g}	631.7 ± 3.9 ^{b}
R6	927 ± 7.1 ^{c}	647.3 ± 1.9 ^{a}	1324.3 ± 4.6 ^{a}	279.7 ± 6.34 ^{f}	677 ± 4.5 ^{a}
R7	880.3 ± 6.2 ^{d}	389 ± 1.4 ^{e}	780.7 ± 4.2 ^{d}	491.3 ± 4.92 ^{d}	391.7 ± 2.9 ^{c}

*Means ± standard deviations, n=3; values followed by different letters in the same column are significantly different (Duncan’s test $p<0.05$)

**Peak viscosity: the maximum viscosity reached during initial heating

Hot paste viscosity: viscosity reached at the end of heating at 95^{o}C

Cold paste viscosity: viscosity when cooled to 50^{o}C

Breakdown= peak viscosity – Hot paste viscosity

Setback= cold paste viscosity – hold paste viscosity.

5.4. Swelling Power and Solubility Index

Swelling power and solubility increased with increase in temperature and starch damage and decreased with storage time.

Swelling powers and solubility index of all rice samples as shown in Figure 18 and Figure 19increased with increase in temperature from 60 to 90^{o}C.

As seen, the swelling power for R7 is high even at relatively low temperature of 60^{o}C. R7 has the smallest particle size distribution in this set of samples Table 4and thus has the highest starch damage Figure 20. These highly damaged starch granules present in flour lead the granules to absorb more water and therefore increase the swelling power (Gujska *et al.*, 1994; Hasjim *et al.*, 2012).

As observed, flour from aged rice R5 had a low swelling powerat 60 and 70^{o}C. This indicated that aged rice starch granule is more resistant to swelling than that of fresh rice. Similar results was also seen in granular morphology of swollen fresh and aged rice starch (Wattinee and Sanguansri, 2012). Protein might be responsible in the inhibition of swelling in aged rice starch or flour. During storage of rice, the number of disulfide bonds and the average molecular weight of oryzenin, which is a major protein in rice, increased (Chrastil, 1990). These disulfide bonds possibly restrict starch granuleswelling during gelatinization (Hamaker and Griffin, 1993).

Starch granular swelling is known to be a primary property of amylopectin. High proportions of long chain molecules in amylopectin contribute to the high swelling power of waxy starch (Tester and Morrison, 1990; Cozzolino *et al.*, 2013). However, in this study no evident trend was observed for amylopectin content and swelling power. This might have occurred due to the complexity of flour components that may have masked or overpowered the swelling property of amylopectin in pure starch.

It is interesting to note that the solubility indices of flour from aged rice R5 maintained a relatively low solubility index compared to other samples. This agreed with low water SRC in Table 7 for R5. Also, similar results was observed for hot water solubility of amylose in aged rice starch and flour (Bhattacharya, 2011). Starch and protein in aged flour might be the components responsible for low solubility index in the same way, aged wheat starch is known to have increased hydrophobicity, more starch granule surface protein and thus decreased hydrophilicity (Seguchi, 1993).

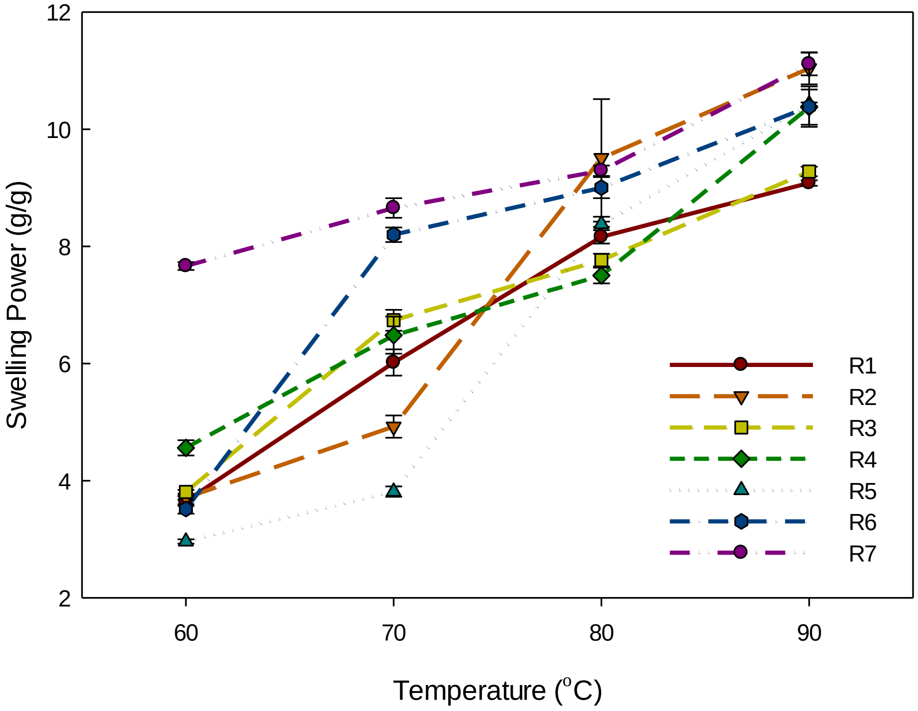


Figure 18. Swelling Power of rice flours at 60, 70, 80 and 90°C

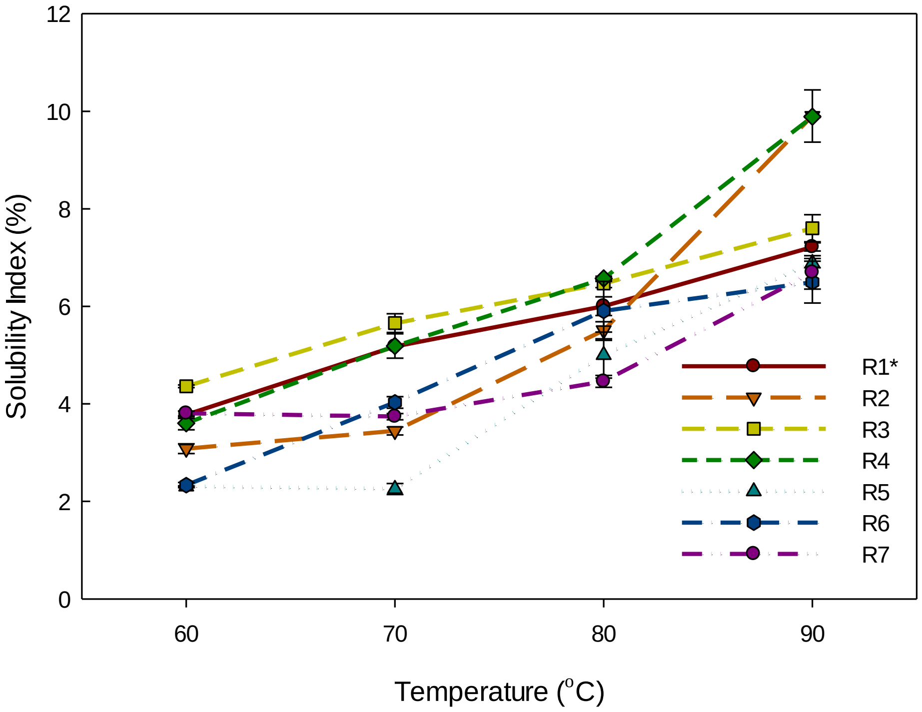


Figure 19. Solubility Index of rice flours at 60, 70, 80 and 90°C

5.5. Solvent Retention Capacity Profile

Each rice flour sample had a distinct SRC profile which implies that SRC test can be a good tool to provide a characteristic description of the functionality of not only wheat flour but also rice flour.

The SRC test is a solvation assay for wheat flours that is based on the enhanced swelling behavior of functional flour components or individual flour polymer networks in selected single diagnostic solvents-- lactic acid in water (for glutenin), sodium carbonate in water (for damaged starch), and sucrose in water (for pentosans)—which are used to predict the functional contribution of each individual flour component(Kweon *et al.*, 2011).

R7 had the highest solvent retention capacity profile for all solvents. The biggest difference between R7 and all the other samples is the particle size. This indicates that in addition to flour components, particle size also plays a great role in SRC test. The small particle size of R7 may have caused high adsorption of all solvents resulting to high SRC values.

Lactic acid as previously described, enhances the swelling of the major wheat protein glutenin. Although rice do not contain glutenin, this study remains to use lactic acid in its SRC profile. Major proteins in wheat (glutenin) and rice (oryzenin) are both glutelin Osborne proteins. These glutelin Osborne proteins can be extracted by dilute acids such as lactic acid (Chrastil and Zarins, 1994). However, in this test, the protein was not dissolved and extracted but instead, it swelled and adsorb the solvent. Presence of disulphide bonds and high molecular weight of oryzenin might be responsible for its decreased solubility. Also, it is important to note that lactic acid SRC indicates flour protein functionality and not flour protein quality (Kweon *et al.*, 2011) , which explains why protein content presented in Table 3 does not show the same trend with lactic acid SRC.

Sodium carbonate as mentioned above enhances the swelling of damaged starch. In accordance, results for the sodium carbonate SRC and for damaged starch as indicated in Figure 20 showed the same trend.

Table 7.Solvent Retention Capacity of rice flours

	Deionized water	50% Sucrose	5% Lactic acid	5% Sodium carbonate
R1	*124.2 ± 0.0 ^{b}	136.0 ± 0.0 ^{e}	109.5 ± 0.3 ^{d}	119.7 ± 0.0 ^{b}
R2	102.0 ± 0.0 ^{e}	137.1 ± 0.0 ^{d}	106.6 ± 0.2 ^{e}	118.8 ± 0.0 ^{b}
R3	104.1 ± 1.3 ^{d}	138.4 ± 0.4 ^{c}	114.2 ± 0.6 ^{b}	120.2 ± 2.2 ^{b}
R4	114.9 ± 0.0 ^{c}	131.0 ± 0.0 ^{g}	101.2 ± 0.6 ^{g}	112.5 ± 0.0 ^{c}
R5	102.6 ± 0.7 ^{e}	133.4 ± 0.2 ^{f}	111.4 ± 1.1 ^{c}	112.2 ± 0.7 ^{c}
R6	99.9 ± 0.2 ^{f}	210.5 ± 0.0 ^{a}	103.0 ± 2.1 ^{f}	105.8 ± 0.0 ^{d}
R7	175.1 ± 0.0 ^{a}	203.8 ± 0.0 ^{b}	186.0 ± 1.0 ^{a}	201.0 ± 0.0 ^{a}

*Means ± standard deviations, n=3; values followed by different letters in the same column are significantly different (Duncan’s test $p<0.05$).

5.6. Starch damage

Damaged starch is the breakage of starch structure such as fragmented starch granules with exposed interior observed in flour after grinding or milling. Although not comparable, it is interesting to note that the typical starch damaged for bread flours are around 5-10 %. Certain degree of damaged starch granules is desirable in flour to maintain the quality of food products as damaged starch granules increase water-absorption capacity of flour and amount of substrates for yeast fermentation.

Results obtained for starch damage showed a similar trend with sodium carbonate SRC as shown in Table 7. Starch damage is found to be positively correlated with sodium carbonate SRC (r=0.9493, p<0.001). Compared to the other samples, K8 had the highest starch damage and the smallest particle size distribution Table 4. This is a likely indication that K8 was subjected to mechanical damage by dry milling to fine flour. All samples, except K8, were milled to flour using the same cyclone mill. However, starch damage was still significantly different from each flour. This suggests that there are other factors that may affect starch damage in addition to the type of mill. Possible factors are grain hardness and rate of milling.

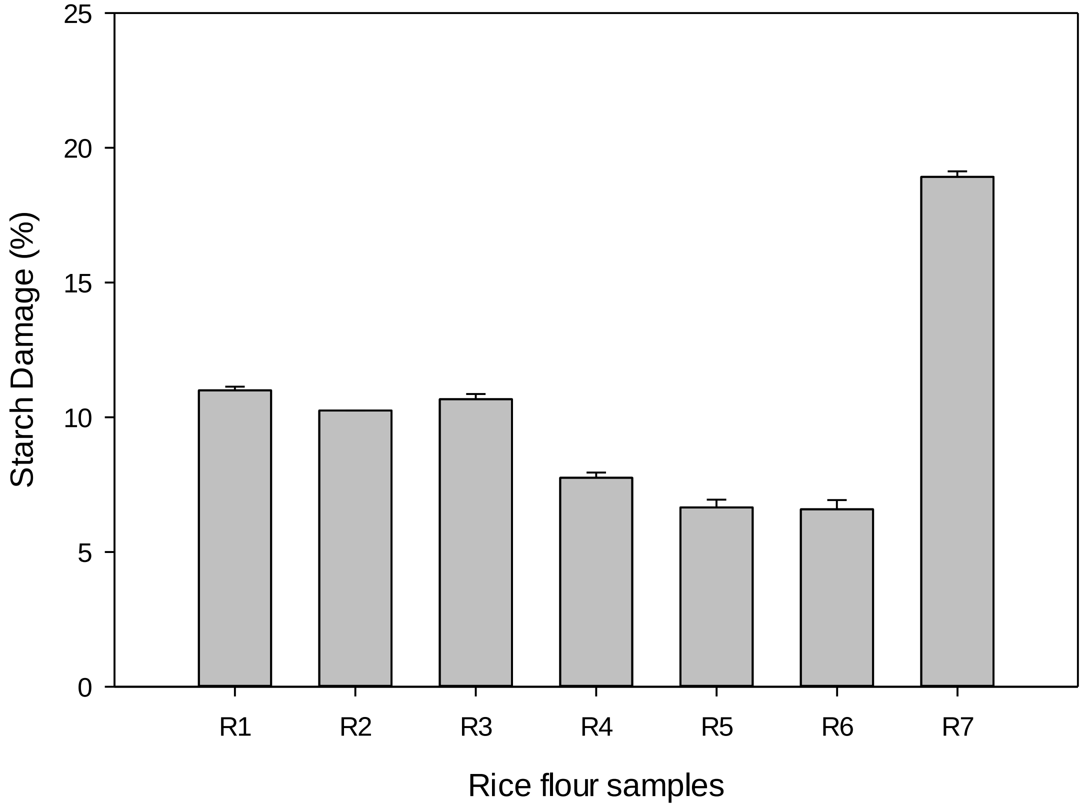


Figure 20.Starch damage of rice flour

5.7. Amylose content

Generally, japonica rice should have lower amylose content compared to that of indica rice. However, this trend was not shown in the results found in Table 8for indica (R1, R2, R5, R6) and japonica (R3, R4, R7) rice samples. Samples R1 and R2 had a relatively low amylose content compared to japonica rice.. This might be because they were store bought rice and were the so called soft indica rice.

Correlation analysis revealed that pasting properties: hot paste viscosity, cold paste viscosityand setback were found to be positively correlated with amylose content while breakdown is negatively correlated with amylose content as seen in Table 9.

Table 8.Amylose content of rice flours

	Amylose Content (%)**		
R1	11.42	±	0.33*
R2	14.47	±	0.76
R3	14.47	±	0.28
R4	15.67	±	0.23
R5	21.46	±	0.43
R6	25.03	±	0.79
R7	14.22	±	0.91

*Means ± standard deviations, n=3
** flour basis.

Table 9. Correlation coefficent of pasting properties and amylose content

		Amylose content
Pasting properties	peak viscosity	-0.615 [a]
	hot paste viscosity	0.889**
	cold paste viscosity	0.938***
	breakdown	-0.869**
	setback	0.951***

[a] significantly different (Pearsons' correlation , $*p<0.05$, $**p<0.01$, $***p<0.001$).

5.8. Alpha-amylase and Beta-amylase activities

Alpha-amylase and beta-amylase activities for all samples were generally low. Rice grains have generally conceded to contain alpha amylase and beta amylase (Shinke *et al.*, 1973). Alpha-amylase and beta-amylase are present in the bran and in the endosperm respectively. As seen below in Table 11, alpha-amylase seems to be more abundant than beta-amylase. However, the two are not comparable having different units,Ceralpha unit for one and Betamyl unit for another. In addition, although not comparable, falling number was also done for all samples and all attained more than 300 seconds Table 10.

Table 10. Falling number of rice flours

	Falling number (sec)
R1	430.5 ± 0.5*
R2	472.5 ± 1.5
R3	410.0 ± 1.0
R4	403.0 ± 11.0
R5	>500
R6	>500
R7	479.00 ± 4.00

*Means ± standard deviations, n=2.

Table 11.Enzyme activities of rice flours

	alpha-amylase (CU/g of flour)**	beta-amylase (BU/g of flour)
R1	4.86 ± 0.08*	1.32 ± 0.01
R2	4.19 ± 0.19	1.15 ± 0.00
R3	4.05 ± 0.58	1.25 ± 0.01
R4	3.93 ± 0.13	1.31 ± 0.05
R5	4.30 ± 0.06	1.44 ± 0.02
R6	3.56 ± 0.13	1.30 ± 0.01
R7	5.97 ± 0.04	1.47 ± 0.02

*Means ± standard deviations, n=3

** CU = Ceralpha Unit; Amount of enzyme, in thepresence of excess thermostable α-glucosidase, required to releaseone micromole of *p*-nitrophenol from BPNPG7 in one minute under thedefined assay conditions

BU = Betamyl-3 Unit; Amount of enzyme, in thepresence of excess thermostable β-glucosidase, required to releaseone micromole of *p*-nitrophenol from PNPβ-G3 in one minute underthe defined assay conditions.

5.9. Mixing Characteristic

The addition of 1.5% hydroxypropyl methylcellulose to rice flours enabled the rice flour-water mixture reach a consistency of 500 FU in the farinograph. Farinograph parameters are described in Table 12. Most of the rice flour had a gradual water uptake and gradual increase in consistency. As the consistency reached 500 FU, it remained at that point until the end of the test indicating that the rice dough is quite stable. This is reflected in the samples’ stability time. However R7 has a noticeable breakdown at 4.5 minutes as seen in Figure 24. This seemed similar to the breakdown that is observed for wheat flour farinograms. However, this is not caused by gluten breakdown caused by shear. Rather, the peak observed was caused by abrupt water absorption of K8 flour, forming lumps. The lumps are responsible for the 500 FU consistency observed for a few minutes. Breakdown is observed when water absorbed in the lumps are distributed throughout the flour.

In addition, it is interesting to note that farinogram parameters were found to be correlated with each other and several other flour physicochemical properties such as swelling power, solvent retention capacity, starch damage, enzyme activities as seen in Table 13. This indicates that mixing characteristics of rice flours are affected by many factors and the combination of these factors are evidently seen in the farinogram. This also proves that the use of farinograph and the addition of 1.5% hydroxypropyl methylcellulose to the rice flour is an effective tool in studying rice flour mixing characteristics

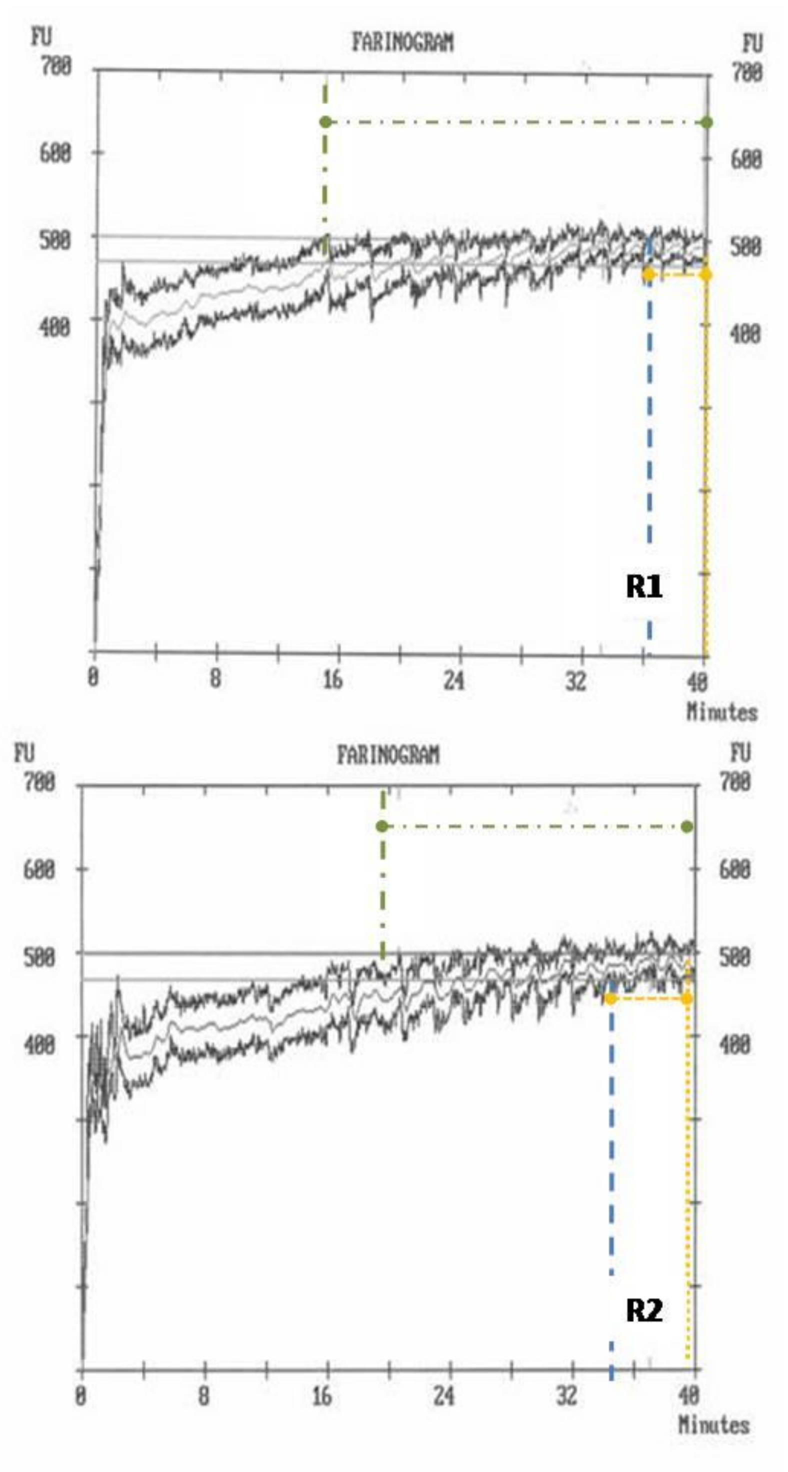


Figure 21.The farinograms of rice flour R1 andR2. (rice flour with 1.5% hydroxypropylmethylcellulose)

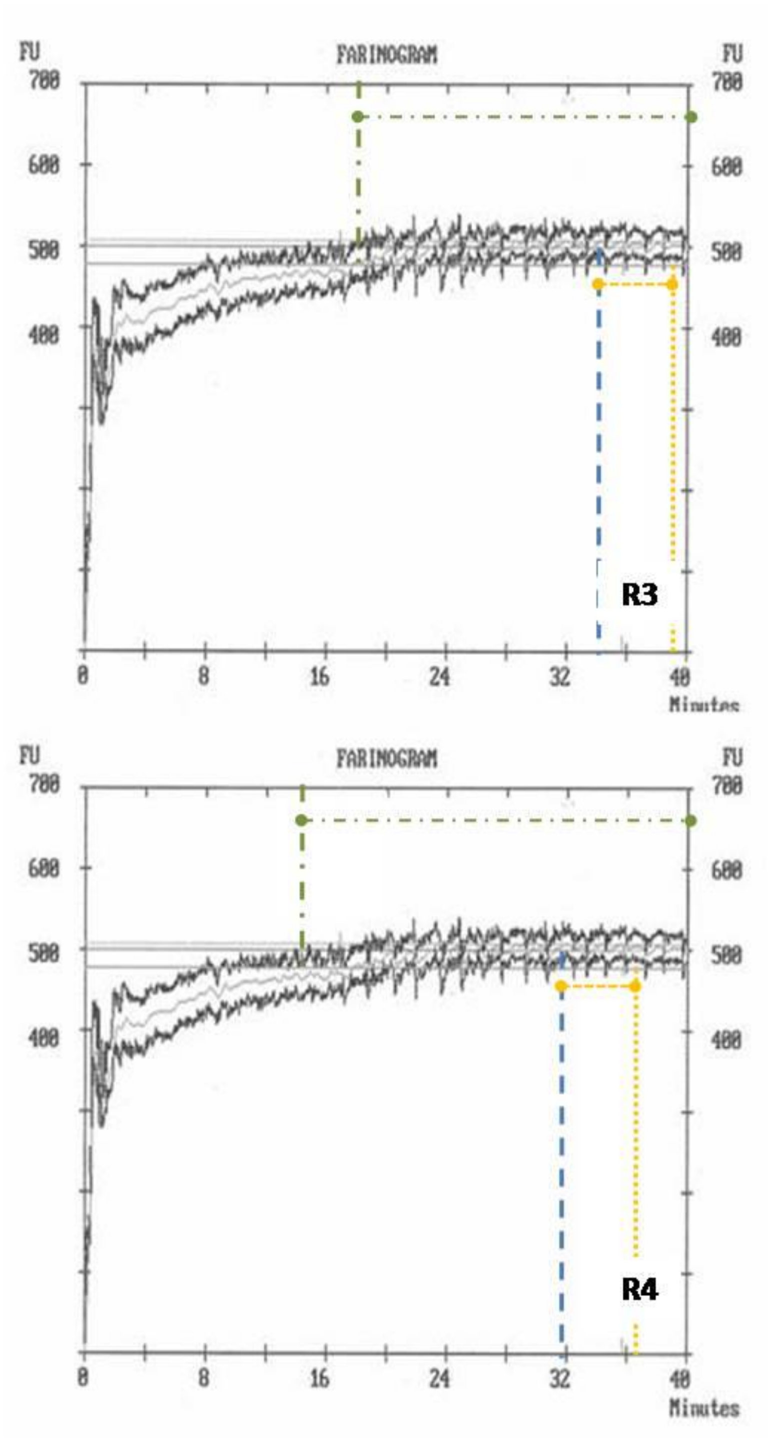


Figure 22. The farinograms of rice flour R3 and R4(rice flour with 1.5% hydroxypropylmethylcellulose)

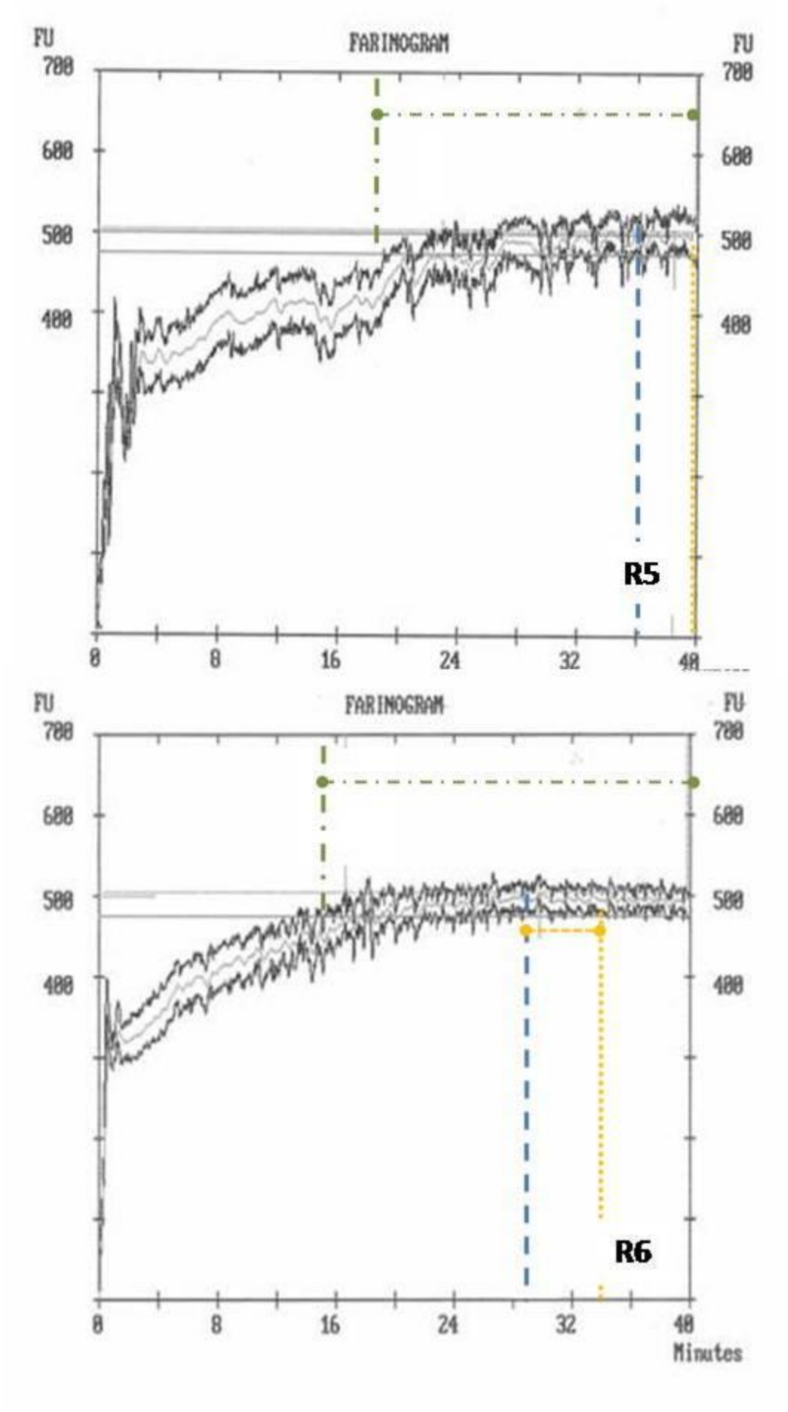


Figure 23. The farinograms of rice flour R5 and R6(rice flour with 1.5% hydroxypropylmethylcellulose)

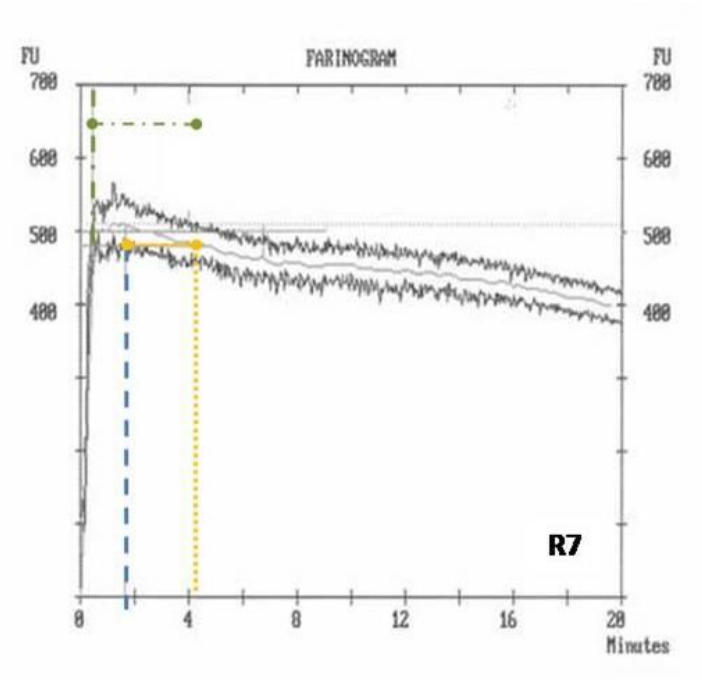


Figure 24. The farinogram of rice flour S7 (rice flour with 1.5% hydroxypropylmethylcellulose)

Table 12. Farinograph Parameters of rice flours

	Consistency (FU)	Water absorption (%)	Development time (mins)	Stability (mins)	Mixing Tolerance Index (FU)
R1	*502 ± 1 ^{bc}	66.6 ± 0 ^{e}	36.2 ± 3 ^{ab}	24.7 ± 0 ^{a}	7.5 ± 2 ^{bc}
R2	494 ± 2 ^{d}	67.2 ± 0 ^{d}	34.8 ± 2 ^{abc}	20 ± 1 ^{b}	8 ± 1 ^{b}
R3	504.3 ± 3 ^{b}	67.5 ± 0 ^{b}	33.7 ± 2 ^{abc}	22.1 ± 1 ^{ab}	7 ± 1 ^{bc}
R4	497.3 ± 3 ^{cd}	66.4 ± 0 ^{f}	31.5 ± 5 ^{bc}	24.6 ± 2 ^{a}	3.3 ± 1 ^{cd}
R5	506.3 ± 8 ^{d}	62.5 ± 0 ^{c}	36.7 ± 1 ^{abc}	18.3 ± 1 ^{b}	10.7 ± 1 ^{b}
R6	505.7 ± 1 ^{b}	65.4 ± 0 ^{h}	29.1 ± 2 ^{c}	24.8 ± 2 ^{a}	9.7 ± 1 ^{b}
R7	514 ± 3 ^{a}	109.7 ± 0 ^{a}	1.7 ± 0 ^{d}	3.5 ± 0 ^{c}	48.7 ± 4 ^{a}

*Means ± standard deviations, n=3; values followed by different letters in the same column are significantly different (Duncan's test $p<0.05$)

** Development time= time at the highest point of the curve
Stability = time of arrival at 500 ± 20FU consistency – time of departure at 500 ± 20 FU consistency
Mixing Tolerance Index = highest consistency – consistency after 5 minutes.

Table 13.Correlation coefficient of rice flour specifications with farinogram parameters

Rice flour specification		farinogram parameters				
		consistency	water absorption	development time	stability time	mixing tolerance index
Proximate composition	protein	*0.142	-0.104	0.191	-0.170	-0.049
	ash	0.477	0.402	-0.382	-0.224	0.457
	fat	0.127	0.287	-0.098	-0.468	0.231
Pasting properties	peak viscosity	-0.259	0.076	-0.076	0.168	0.036
	hot paste viscosity	-0.006	-0.476	0.422	0.261	-0.343
	cold paste viscosity	0.147	-0.337	0.265	0.134	-0.202
	breakdown	-0.132	0.333	-0.299	-0.072	0.230
	setback	0.254	-0.228	0.146	0.040	-0.096
Swelling Power	60^{o}C	0.571	0.968***	-0.964***	-0.82904**	0.909**
	70^{o}C	0.512	0.629	-0.744	-0.331	0.600
	80^{o}C	0.179	0.415	-0.402	-0.509	0.483
	90^{o}C	0.064	0.415	-0.449	-0.557	0.434
Solubility Index	60^{o}C	-0.392	-0.389	-0.549	0.257	0.378
	70^{o}C	0.074	0.036	-0.108	0.461	0.121
	80^{o}C	0.164	0.056	-0.010	0.538	0.139
	90^{o}C	-0.357	-0.583	-0.448	0.426	0.695

Cont.

Rice flour specification		farinogram parameters				
		consistency	water absorption	development time	stability time	mixing tolerance index
Solvent Retention Capacity	deionized water	0.645	0.952***	-0.906**	-0.833**	0.911**
	50% sucrose	0.667	0.612	-0.735	-0.469	0.668
	5% lactic acid	0.759*	0.974***	-0.921***	-0.958***	0.963***
	5% sodium carbonate	0.677	0.991***	-0.937***	-0.940***	0.965***
Starch Damage	starch damage	0.536	0.921***	-0.821*	-0.837**	0.854**
Amylose content		0.136	-0.349	0.226	0.207	-0.244
Enzyme activity	alpha	0.572	0.822*	-0.691	-0.827**	0.794*
	beta	0.819	0.564	-0.595	-0.603	0.665
Farinograph Parameters	consistency					
	water absorption	0.684				
	development time	-0.710*	-0.974***			
	stability time	-0.685	-0.918***	0.863**		
	mixing tolerance index	0.770*	0.973	-0.962***	-0.941***	
Bake test	height	0.307	0.021	-0.682	0.623	0.245
	specific volume	0.514	-0.063	-0.542	0.559	0.381
	volume index	0.410	-0.179	-0.584	0.491	0.301
	symmetry	-0.482	0.615	-0.552	0.498	-0.247
	uniformity	-0.692	0.488	0.054	-0.324	-0.197

[a] significantly different (Pearsons' correlation , *p<0.05, **p<0.01, ***p<0.001).

5.10. Bake test

Bake test revealed that pentosans, starch damage and amylose content are key factors that influenced rice bread volume.

As expected, all rice bread properties obtained from the rice samples were all lower than the control wheat and control rice bread. Gluten in wheat flour was responsible for the structure of the control wheat bread while additional starches were responsible for the structure of the control rice bread. The control rice bread used commercially bought rice flour contained potato, wheat and corn starch which was blended in by the manufacturer.

Pearson's correlation analysis revealed that height, specific volume and volume index were positively correlated with sucrose solvent retention capacity (r=0.928, p<0.05; r=0.892, p<0.01; r=0.886, p<0.05) while volume index were negatively correlated with sodium carbonate solvent retention capacity (r=-0.797, p<0.05).

Most of the symmetry indices obtained were negative values unlike the those of the control. This indicated that rice breads had a slightly collapsed center. This could be due to the low gas retention ability of rice flours. Uniformity indices obtained were approximately 0, revealing that height of cakes are approximately uniform without one side exceedingly higher or lower.

As seen in the Figure 24, bread from aged rice flourR5 had a more cracked top layer.

This is also reflected in its corresponding rough farinogram curve found inFigure 23. This could be caused by the low ability of flours from aged rice to absorb water and swell as explained in page 38.

Regarding the air cell, R3, R4 and R2 had a very compact air cell at the bottom and had few medium sized air cells. Aged rice R5 also had a very compact air cell at the bottom but had a larger sized air cell on the top. R1 however had a more evenly spread air cell. R6 had the largest air cell among all the samples. It was more uniform and circular. This is reflected in volume and height of the bread in Table 14. Rice flour R6 with an intermediate to high amylose content obtained highest parameters in height, specific volume and volume index. Similar findings have found that intermediate amylose rice produce cakes with high volume expansion (Mohamed and Hamid, 1998).

Surprisingly, there is an observed negative correlation between ash content and height ($r=-0.929$, $p<0.01$), specific volume ($r=-.839$, $p<.05$), volume index ($r=-0.857$ $p<0.05$,).In addition, researches have also found a correlation ash content and cookie crumb grain made from wheat flour (Geng *et al.*, 2012). More details in correlation analysis done could be seen inTable 15.

Further studies regarding internal structures of rice flour bake tests however is suggested.

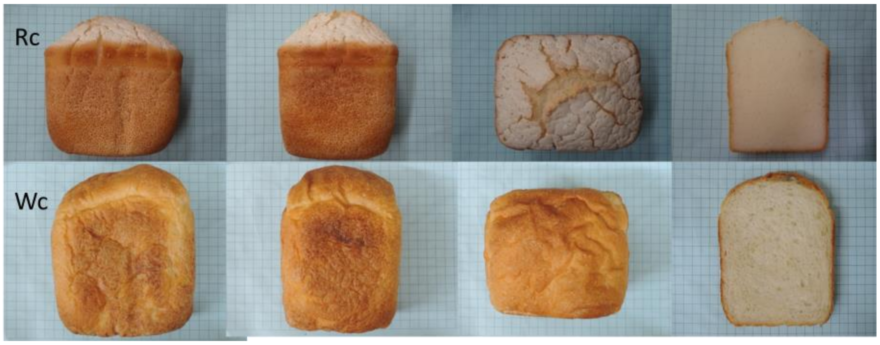


Figure 25. Control bread from rice flour (Rc) and wheat flour (Wc).

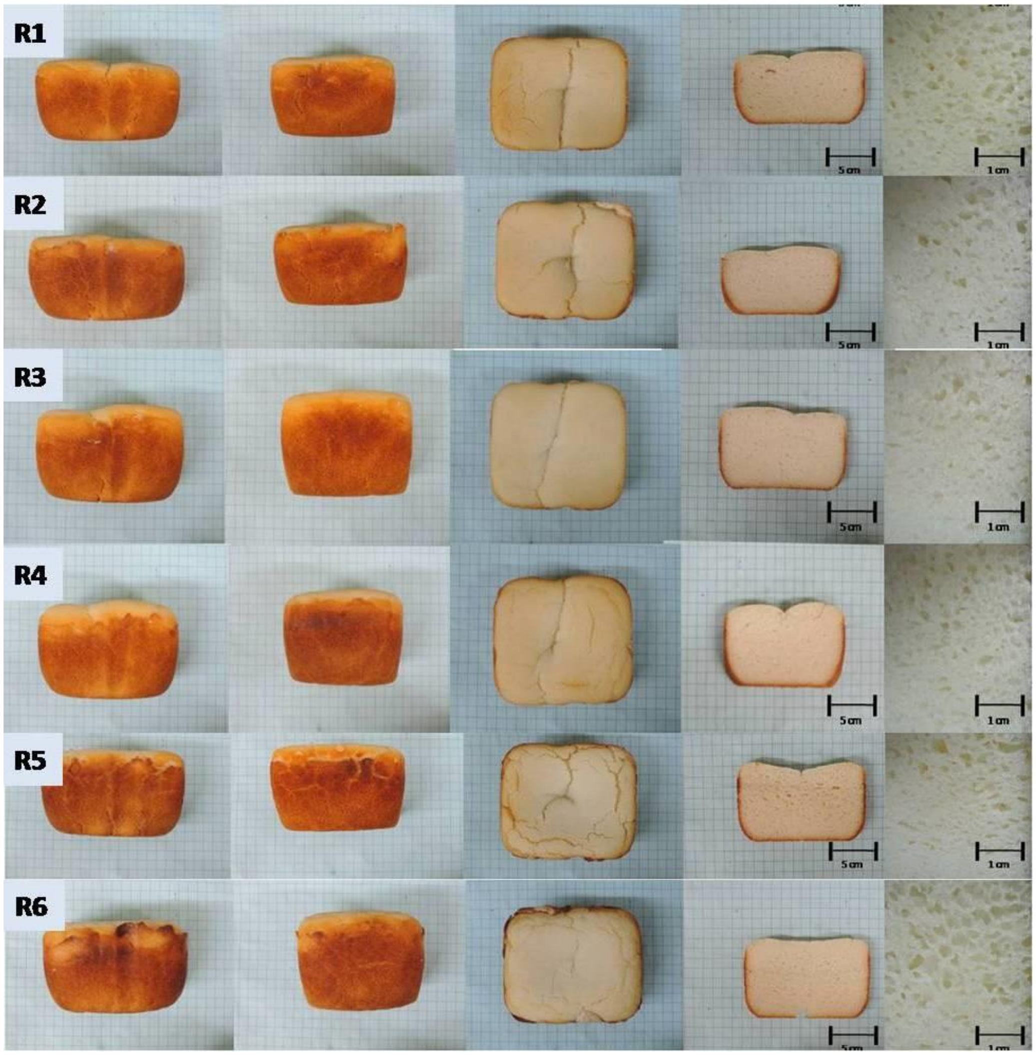


Figure 26. Rice breads

*R7 not shown : following the recipe used R7 cannot form a rice bread.

Table 14. Rice bread properties

	Height (cm)	Specific Volume (cc/g)	Volume Index	Symmetry Index	Uniformity Index
Rc*	11.7	2.65	32.3	2.8	1
Wc	14.9	4.47	42.2	2.5	1.2
R1	6.7 ± 0.4^{b}	1.56 ± 0.06^{b}	20.2 ± 0.9^{b}	-0.3 ± 0.4^{b}	-0.1 ± 0.2^{b}
R2	6.4 ± 0.3^{b}	1.43 ± 0.02^{c}	19.2 ± 0.5^{cd}	0 ± 0.4^{a}	0.2 ± 0.2^{a}
R3	6.3 ± 0.2^{b}	1.44 ± 0.06^{c}	18.8 ± 0.4^{d}	-0.1 ± 0.1^{a}	0.1 ± 0.1^{a}
R4	6.7 ± 0.4^{b}	1.47 ± 0.01^{bc}	20.2 ± 0.3^{b}	-0.1 ± 0.4^{a}	0 ± 0.3^{ab}
R5	6.4 ± 0.3^{b}	1.49 ± 0.05^{bc}	20.1 ± 0.5^{b}	-0.6 ± 0.3^{b}	-0.1 ± 0.1^{b}
R6	7.7 ± 0.2^{a}	1.71 ± 0.07^{a}	23 ± 0.6^{a}	0 ± 0.4^{a}	-0.1 ± 0.2^{ab}

*Means ± standard deviations, n=3; values followed by different letters in the same column are significantly different (Duncan's test p<0.05); Rc = rice flour control, Wc = wheat flour.

Table 15.Correlation coefficients between rice flour specifications and rice bread properties

Rice flour specification		bake test				
		height	specific volume	volume index	symmetry	uniformity
Proximate composition	protein	-0.301 [a]	-0.198	-0.103	-0.720	-0.376
	ash	-0.929**	-0.838*	-0.857**	-0.534	0.235
	fat	0.334	0.546	0.324	-0.209	-0.535
Pasting properties	peak viscosity	0.177	0.169	0.002	0.527	0.317
	hot paste viscosity	0.369	0.427	0.527	-0.330	-0.456
	cold paste viscosity	0.419	0.463	0.572	-0.296	-0.478
	breakdown	-0.120	-0.158	-0.306	0.475	0.435
	setback	0.449	0.483	0.597	-0.270	-0.489
Swelling Power	60^{o}C	0.077	-0.105	-0.070	0.539	0.290
	70^{o}C	0.694	0.621	0.547	0.597	-0.127
	80^{o}C	0.245	0.211	0.244	0.249	0.260
	90^{o}C	0.084	-0.110	0.146	0.174	0.298
Solubility Index	60^{o}C	-0.392	-0.389	-0.549	0.257	0.378
	70^{o}C	0.074	0.036	-0.108	0.461	0.121
	80^{o}C	0.164	0.056	-0.010	0.538	0.139
	90^{o}C	-0.357	-0.583	-0.448	0.426	0.695

Cont.

Rice flour specification		bake test				
		height	specific volume	volume index	symmetry	uniformity
Solvent Retention Capacity	deionized water	-0.081	0.006	-0.115	-0.165	-0.298
	50% sucrose	0.927**	0.892**	0.886**	0.409	-0.338
	5% lactic acid	-0.636	-0.427	-0.592	-0.472	0.106
	5% sodium carbonate	-0.702	-0.619	-0.797	-0.013	0.536
Starch Damage	starch damage	-0.527	-0.471	-0.673	0.229	0.528
Amylose content		0.525	0.496	0.640	-0.093	-0.416
Enzyme activity	alpha	-0.556	-0.368	-0.501	-0.516	-0.098
	beta	0.127	0.303	0.317	-0.803*	-0.827*
Farinograph Parameters	consistency	0.307	0.514	0.410	-0.482	-0.692
	water absorption	0.021	-0.063	-0.179	0.615	0.488
	development time	-0.682	-0.542	-0.584	-0.552	0.054
	stability time	0.623	0.559	0.491	0.498	-0.324
	mixing tolerance index	0.245	0.381	0.301	-0.247	-0.197
Bake test	height					
	specific volume	0.950***				
	volume index	0.970***	0.955***			
	symmetry	0.367	0.134	0.152		
	uniformity	-0.513	-0.681	-0.651	0.546	

[a]significantly different (Pearsons' correlation , *$p<0.05$, **$p<0.01$, ***$p<0.001$).

6. Conclusion

There is a need for set of quality tests to control rice flour processing. Rice flour tests cannot directly adapt the well established set of quality tests for wheat flour because of the differences in composition, physicochemical and rheological properties. The methods used in this study such as proximate composition, particle size distribution, pasting properties, swelling power and solubility, solvent retention capacities, starch damage, amylose content, alpha-amylase and beta-amylase activity, mixing characteristics and bake test prove to reveal distinct characteristics of rice flour samples.

Proximate composition and particle size distribution gave a brief overview of rice flour samples.

Results in pasting properties of rice flours revealed that rice hot paste viscosity, cold paste viscosityand setbackwere statistical ($r \geq 0.8$, $p < 0.01$) positively correlated with amylose content while breakdown ($r = 0.869$, $p < 0.01$) is negatively correlated with amylose content.

In swelling power and solubility index, flour from aged rice R5 had a low swelling power of 2.97 g/g and 2.96 g/g at 60 and 70^{o}C respectively. In addition solubility indices were also low at 60, 70, 80 and 90^{o}C with 2.32, 2.26, 5, 6.89 for R5.

R7 had the highest solvent retention capacity profile for all solvents. Since the biggest difference between R7 and all the other samples is the particle size. This indicates that in addition to flour components, particle size also plays a great role in SRC test.

Starch damage is found to be positively correlated with sodium carbonate SRC (r=0.949, p<0.001). R7 with the highest starch damage of 18.9% also had the highest sodium carbonate solvent retention capacity of 201.

The addition of 1.5% hydroxypropyl methylcellulose to rice flour made it possible for rice flour mixing characteristics be studied in a farinograph. Mixing characteristics of rice flours are affected by many factors and the combination of these factors are evidently seen in the farinogram. Farinograph parameters were correlated with each other and with other rice flour physicochemical properties. Water absorption is positively correlated with swelling power at 60^{o}C (r=0.968, p<0.01), water solvent retention capacity (r=.952, p<0.001), lactic acid solvent retention capacity (r=0.974, p<0.001), sodium carbonate solvent retention capacity (r=.991, p<0.001), alpha-amylase activity (0.822, p<0.05) and starch damage (r=0.921, p<0.001). Development time is negatively correlated with swelling power at 60^{o}C (r=-0.964, p<0.001), water solvent retention capacity (r=-0.906, p<0.01), lactic acid solvent retention capacity (r=-0.921, p<0.001), sodium carbonate solvent retention capacity (r=-0.937, p<0.001) and beta-amylase activity (r=-0.821, p<0.001). Stability time is negatively correlated with swelling power at 60^{o}C (r=--0.833198, p<0.01), water solvent retention capacity (r=-0.833, p<0.01), lactic acid solvent retention capacity (r=-0.958, p<0.001), sodium carbonate solvent retention capacity (r=-0.940, p<0.001), beta-amylase activity (r=-0.827, p<0.01), and starch damage (r=-0.837, p<0.01). Mixing Tolerance Index is positively correlated with swelling power at 60^{o}C (r=0.909, p<0.01), water solvent retention capacity (r=.912, p<0.01), lactic acid solvent retention capacity (r=0.964, p<0.001), sodium carbonate solvent retention capacity (r=.965, p<0.001), alpha-amylase activity (0.794, p<0.05) and starch damage (r=0.854, p<0.01).

In the bake test, differences in rice breads produced by different rice flours were observed in their cross sections through air cells and the top surface. Differences were also observed in specific volume and volume index. Further analysis of the bake test breads are suggested to further observe the effects of rice flours in breads.

The methods used in this study, revealed characteristics of rice flour that could serve as guidelines in rice flour quality control in the food industry.

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Appendix

The methods to differentiate rice flour characteristics

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Abstract

Increased use of rice flour in the food industry has necessitated it to have a set of testing methods that will reveal rice flour characteristics to indicate rice flour quality. Therefore, the objective of this study is to develop differentiating testing methods in rice flour that will help in understanding rice flour properties and that will serve as guidelines in rice flour processing properties. In this study, milled rice were ground to flour, and flour specifications such as proximate composition, particle size distribution, pasting properties, swelling power and solubility, solvent retention capacities, starch damage, amylose content, alpha-amylase and beta-amylase activity, mixing characteristics were determined. Lastly, a bake test was also conducted. Seven commercial including fresh and aged rice samples were selected for testing. Protein quality and starch damage are observed to be the main determining factors in rice flour mixing characteristics. The addition of 1.5% hydroxypropylmethylcellulose to rice flour made it possible for rice flour mixing characteristics to reach a consistency of 500 FU and be studied in a farinograph. Farinograph parameters were greatly influenced by solvent retention capacities of lactic acid and sodium carbonate which are associated with protein quality and starch damage respectively. Water absorption and mixing tolerance index were statistical ($r \geq 0.9$, $p<0.001$) positively correlated with lactic acid solvent retention capacity and sodium carbonate solvent retention capacity, while development time and stability time were statistical ($r \leq -0.9$, $p< 0.001$) negatively correlated with lactic acid solvent retention capacity and sodium carbonate solvent retention capacity. Lastly, results for bake test indicated that solvent retention capacity, starch damage and amylose content affect rice bread properties. Pentosans and starch damage are key factors that influenced rice bread volume. Height, specific volume and volume index were statistical ($r \geq 0.8$, $p<0.05$) positively correlated with sucrose solvent retention capacity while volume index were negatively correlated with sodium carbonate solvent retention capacity($r \leq -0.8$, $p<0.05$). Therefore, the methods used in this study reveal characteristic rice flour properties and could serve as guidelines in rice flour quality control in the food industry.

Keywords: Rice flour, farinograph, solvent retention capacity, pentosan, gluten free bread, rice bread

Introduction

Rice is a staple food for more than half of world population. It is a world-wide commodity and shows great diversity in properties due to its locality of production and variety. These variations significantly affect its utilization.

Rice is generally grown for its eating qualities as cooked rice. Therefore, literally hundreds of varieties of rice are grown all over the world for their unique characteristics. In accordance, several tests have been developed in measuring physicochemical properties of milled rice. Rice flours, on the other hand, is commercially used in babyfoods, meat products, separating powders for refrigerated pre-formed unbaked biscuits, dusting powders, breading, and formulations for pancakes and waffles.

In recent years, rice flour is being used in increasing numbers of novel foods such as tortillas, beverages, processed meats, puddings, and gluten-free breads because of its unique functional properties. (Kadan and Ziegler 1989; McCue 1997; Kadan and others 2001).

However, there is a no set of quality testing methods of rice flour available. Most of the available flour quality testing methods are developed specifically for wheat flour. Rapid visco analyzer and amylograph are used to measure wheat starch properties while farinograph, mixograph and alveograph are used to measure wheat gluten characteristics. Because of differences in composition, rice flour cannot directly adapt tests from that of wheat flour.

The objective of this study is to develop instrumental testing methods in rice flour that will help in understanding rice flour properties and in the same time serve as guidelines in rice flour characterization.

Materials and Methods

Samples

All samples were obtained from commercial sources in milled rice grain form, except for R7 which was in the form of milled rice flour.

Table1.Rice samples

Samples*	Abbreviation	Source
Chang Hsien	R1	Yeedon Enterprise Co., Ltd.
Chang Hsiang	R2	Hua-dong Co., Ltd.
Tainan 11	R3	China Grain Products Research and Development Institute
Chung Hsing	R4	The Union Rice Co., Ltd.
Kung Liang	R5	China Grain Products Research and Development Institute
Tainung Sen 14	R6	China Grain Products Research and Development Institute
Lian Hwa	R7	A commercial rice flour from Lian Hwa Enterprise Co. Ltd.,

*All samples, except R7, were received as milled rice and ground in laboratory scale Cyclotec mill.

Grinding of rice grains

Commercial rice samples R1 to R6 in Table 2 were ground into flour using Cyclotec mill with 1.0 mm sieve (Cyclotec 1093, Tecator, Sweden). Sample R7 was not ground because it is already in rice flour form.

Rice flour specification methods

Proximate analysis

Ash content was determined following Ash content for soy flours (AACC Approved Method 08-16). Fat was determined following Crude fat for flours (AACC Approved Method 61-01) with modification of using 95% Ethanol (Echo Chemical, Taipei, Taiwan) as extraction solvent. Protein was determined by multiplying 5.25 by the nitrogen obtained after sample combustion in LECO FP-628 Nitrogen Determinator (LECO Corporation, St. Joseph, Michigan, USA)

Particle Size Distribution

Particle size dristribution was determined by using light scattering and by using different mesh size sieves. Samples were suspended in isopropyl alcohol (JT Baker, Sweden), and analyzed using a diffraction particle analyzer Coulter LS 100 (Micro Volume Module Cell, Coulter Electronics, USA). 25 grams of samples were pass through 30, 60, 80 and 100 mesh sizes. Flour retained in each sieve was weighed and recorded.

Pasting properties

Pasting properties were determined using Brabender Viscograph E (Brabender GmbH & Co. KG, Germany) following the Amylograph Method for milled rice (AACC Approved Method 61-01). 10% rice flour slurry was subjected to the following temperature cycle: held at 35^{o}C for 5 minutes, heated with a rate of 1.5^{o}C per minute to 95^{o}C, held for 20 minutes, and cooled with a rate of 1.5^{o}C per minute to 50^{o}C.

Swelling Power and Solubility Index

0.10 g (dry basis) of rice flour was placed in 50 mL centrifuge tubes (Falcon tube). Flours were hydrated with 10 mL of distilled water and placed in water bath held at different temperatures (60, 70, 80 and 90^{o}C) for 30 minutes. Samples were vortexed after 2, 4, 6, 8, 10, 15, and 25 minutes in water bath to maintain suspension of flour in water. Centrifuge tubes were placed in a room temperature water bath and were allowed to cool for 30 minutes. Samples were then centrifuged (3000×g , 15 min) and the supernatant was dried at 105 °C until constant weight. The Water solubility index and swelling power were calculated as described in (Li and Yeh, 2001; Cozzolino et al., 2013).

Water Solubility Index (%) = (W1 / flour dry weight) X100
Swelling Power (g/g) = W2 / [flour dry weight x (100% - W1)]

Where, W1 is the supernatant dried to a constant weight
(105 °C); and W2 is the weight of the sediment.

Solvent Retention Capacity Profile

Solvent retention capacity profile of rice flour was determined using four solvents: deionized water, 50 % sucrose solution, 5% lactic acid and 5% sodium carbonate (AACC Approved Method 56-11). 5.0 grams of rice flour (with known moisture content) was placed in 50 mL centrifuge tubes (Falcon tube). Flours were hydrated with 25 grams of the solvent and placed in a water bath at 30^{o}C) for 20 minutes. Samples were vortexed after 5, 10, 15, and 20 minutes in water bath to maintain suspension of flour in water. Samples were then centrifuged (1000xg, 15 minutes). Tubes were inverted and drained on a paper towel for 10 minutes. Solvent retention capacity was calculated as follows:
% SRC = [(gel weight / flour weight) X (86/(100-%flour moisture content)) – 1] X 100

Starch damage

Damaged starch was determined using the Megazyme kit (K-SDAM Megazyme International Ireland, Wicklow, Ireland) . Damaged starch granules were hydrated; followed by hydrolysis to maltosaccharidesand limit dextrins by fungal alpha-amylase. Amyloglucosidase is then used to convert dextrins to glucose, which is specifically determined spectrophotometrically after glucose oxidase/peroxidase treatment (AACC Approved Method 76-31).

Amylose content

Amylose content was determined using colorimetric determination at 620 nm of the greenish-blue starch-iodine complex developed in ammonium buffer following modifications (Juliano *et al.*, 2012) based on amylose content determination (AACC Approved Method 61-03). Rice amylose was isolated from rice starch by complexing amylose with butanol (Takeda *et al.*, 1986) and used as standard.

Alpha-amylase and Beta-amylase activities

The enzyme activities were measured by using Megazyme Kit (K-MALTA, Megazyme International Ireland, Wicklow, Ireland) with colored substrates. One unit of enzyme activity is defined as the amount of enzyme that causes the hydrolysis of one µmole of the specific substrate per minute (Haros *et al.*, 2002). The alpha-amylase and beta-amylase activity were measured by using a blocked p-nitrophenyl maltoheptaoside (BPNPG7, Megazyme International Ireland, Wicklow, Ireland) and p-nitrophenyl beta-maltotrioside (PNP β -3G, Megazyme International Ireland, Wicklow, Ireland)) respectively as substrate (Santos and Riis, 1996; McCleary *et al.*, 2002).

Falling numbers were determined (AACC Approved Method 56-81) using a falling number apparatus (Model 1500, Perten Instruments, Huddinge, Sweden).

Mixing characteristics

Mixing characteristics were measured following a farinograph (Brabender OHG, Kulture, 51-55, d-47055, Duisburg, Germany) following the method farinograph test (AACC Approved Method 54-22) with some modifications. Rice flours were first blended with 1.5% Hydroxypropylmethylcellulose (SFE-4000, Shin-Etsu chemicals Ltd., Japan) by sifting together five times.

Bake test

Rice bread was chosen for this test. Rice bread was made using Panasonic bread machine’s setting for rice bread containing no wheat flour (SD-BM 103T, Panasonic Co., Ltd., Taiwan). Recipe provided in the manual was followed with 300 g rice flour, 5 g salt, 9 g sugar, 10 g oil, 50 g fructose, 220 g water and 3 g yeast. Zai-lai-mi rice flour was used as control rice flour (Sunright Co., Ltd, Taiwan). As for the wheat flour control, wheat flour bread machine’s setting for wheat bread was used with high gluten flour (Milk international Co., Ltd., Taiwan).

Bread volume was determined following the Rapeseed displacement method (AACC Approved Method 10-05).

Rice bread were cut vertically. 20 mm thick slice from the center was used to measure bread parameters. Parameters were measured following the layer cake measuring chart (AACC Approved Method 10-91) with some modifications. Height of bread at vertical lines B, C and D to the nearest 1.0 mm were read. These lines were designated as illustrated in Figure 1. for calculations of volume index, symmetry index and uniformity index
Volume Index = B + C + D
Symmetry Index = 2C – B – D
Uniformity Index = B – D

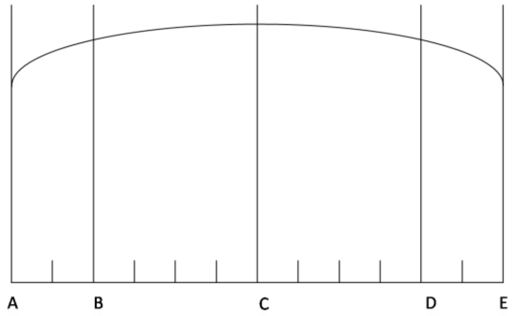

Figure 1.Template for measuring bread

Results and Discussion

Proximate Composition

Results for the proximate composition of rice flour samples are shown below. The protein, ash and fat content of selected rice samples were in the range of 7.0 – 11.6, 0.4 -0.5, and 0.7 – 2.1% respectively. Generally, the protein content of indica rice is higher than that of japonica rice (Sun *et al.*, 2008). However, this trend was not observed in this set of indica (R1, R2, R5, R6) and japonica (R3, R4, R7) rice samples. The reason for this could be due to variety differences such as the so called soft indica R1 and R2. Soft indica is an indica rice but with a low amylose content.

Particle Size Distribution

R7 had the finest particle size distribution in this set of samples. As seen in Table366.2% of R7 was in the range of 0-50μm and 99.8% in 0-150μm inTable 4Table 5. Particle size distribution of samples R1 to R6 was relatively the same, but differed much from that of R7. Same laboratory cyclone mill was used to grind samples R1 to R6 into rice flour (page 25) which lead to their similarity. R7, on the other hand was received in rice flour form so no further grinding was done.

Pasting Properties

heating, holding, and cooling of 10% rice flour slurry, pasting properties help predict rice starch behavior during processing of rice-based product.

As seen, each rice flour sample achieved a distinct pasting profile. There is an observed distinction for aged rice flour K3 and S7. They had the lowest peak viscosity and breakdown as seen in Figure 1. This low peak viscosity and breakdown indicated that the starch granules in aged rice flour was more resistant to swelling and more resistant to rupture after cooking, respectively (Noomhorm et al., 1997; Teo et al., 2000). There is a reported significant increase in the number of disulfide bonds and the average molecular weight of the major protein in rice - oryzenin (Chrastil, 1990). These disulfide bonds possibly restrict starch granule swelling during gelatinization and make the swollen granule less susceptible to disruption by shear (Hamaker and Griffin, 1993).In addition, pasting properties: hot paste viscosity (r=0.889, p<0.01), cold paste viscosity (r=0.938, p<0.01) and setback (r=0.951, p<0.01) were found to be positively correlated with amylose content while breakdown (r=0.869, p<0.01) is negatively correlated with amylose content.

Swelling Power and Solubility Index

Swelling power and solubility increased with increase in temperature and starch damage and decreased with storage time. Swelling powers and solubility index of all rice samples as shown in Figure 4 amd 3Figure 18 increased with increase in temperature from 60 to 90oC.As seen, the swelling power for R7 is high even at relatively low temperature of 60oC. R7 has the smallest particle size distribution in this set of samples and thus has the highest starch damage Figure 5. These highly damaged starch granules present in flour lead the granules to absorb more water and therefore increase the swelling power (Gujska et al., 1994; Hasjim et al., 2012).

Table 2.Proximate composition of rice flours (on dry basis)

	Protein (%)			Ash (%)			Fat (%)		
R1	8.75	±	9.10*	0.49	±	0.02	2.10	±	0.40
R2	7.91	±	0.50	0.48	±	0.01	0.80	±	0.10
R3	7.05	±	0.00	0.50	±	0.00	0.90	±	1.50
R4	8.32	±	8.32	0.47	±	0.02	0.70	±	0.20
R5	11.57	±	0.52	0.52	±	0.03	1.00	±	0.20
R6	7.70	±	7.91	0.38	±	0.01	1.30	±	0.20
R7	9.10	±	0.49	0.52	±	0.01	1.60	±	0.10

Means ± standard deviations, n=3

Table 3.Particle Size Distribution of rice flours by light scattering

	Particle size distribution (%)			
	0-50 μm	51-100 μm	101-200 μm	201-2000 μm
R1	54.3*	21.3	15	9.4
R2	43	22.4	22.9	11.7
R3	58.3	24.3	13.8	3.7
R4	47.6	20.9	20.8	10.6
R5	45.2	22.7	27.6	4.6
R6	43.7	19.4	25.5	11.4
R7	66.2	20.9	10.9	2

*Means, n=3

Table 4.Particle Size distribution of rice flours by mesh size

	particle size distribution (%)				
mesh size	< 100	> 100	> 80	> 60	> 30
	0-150 μm	151-176 μm	177 μm-249	251-594 μm	> 595 μm
R1	55.1*	13.4	11.6	19.6	0.3
R2	55.1	14.9	11.7	18.2	0.2
R3	56.6	12.7	11.1	19.3	0.3
R4	62.4	11.1	11.5	14.8	0.2
R5	60.0	12.4	10.7	16.5	0.4
R6	67.2	10.5	9.0	13.3	0.1
R7	99.8	0.2	0.0	0.0	0.0

*Means, n=3

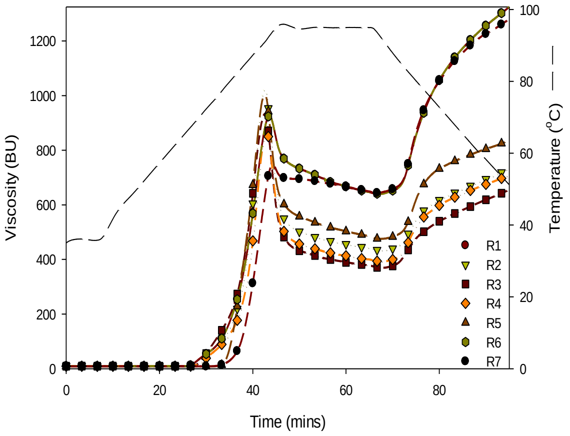

Figure2.Pasting properties of rice flours

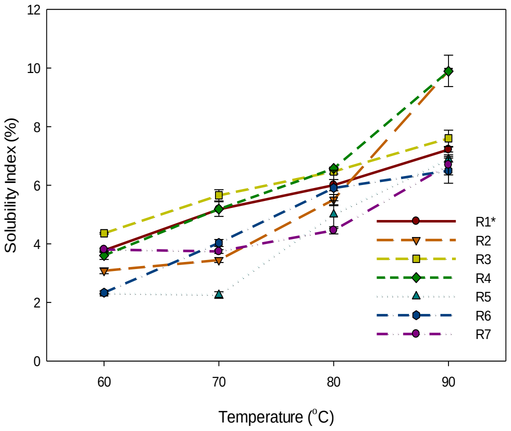

Figure3.Solubility indices of rice flours

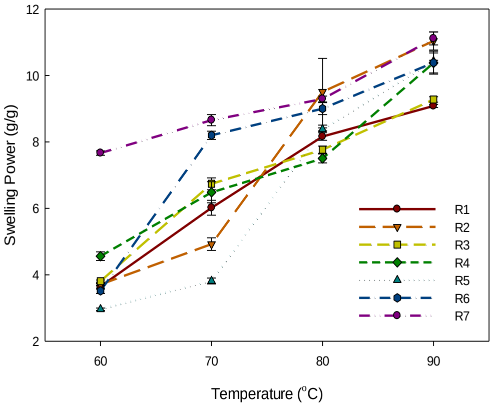

Figure4.Swelling Power of rice flours

As observed, flour from aged rice R5 had a low swelling power at 60 and 70°C. This indicated that aged rice starch granule is more resistant to swelling than that of fresh rice. Similarresults was also seen in granular morphology of swollen fresh and aged rice starch (Wattinee and Sanguansri, 2012). Protein might be responsible in the inhibition of swelling in aged rice starch or flour. During storage of rice, the number of disulfide bonds and the average molecular weight of oryzenin, which is a major protein in rice, increased (Chrastil, 1990). These disulfide bonds possibly restrict starch granule swelling during gelatinization (Hamaker and Griffin, 1993).

Starch granular swelling is known to be a primary property of amylopectin. High proportions of long chain molecules in amylopectin contribute to the high swelling power of waxy starch (Tester and Morrison, 1990; Cozzolino *et al.*, 2013). However, in this study no evident trend was observed for amylopectin content and swelling power. This might have occurred due to the complexity of flour components that may have masked or overpowered the swelling property of amylopectin in pure starch.

It is interesting to note that the solubility indices of flour from aged rice R5 maintained a relatively low solubility index compared to other samples. This agreed with low water SRC in for R5. Also, similar results was observed for hot water solubility of amylose in aged rice starch and flour (Bhattacharya, 2011). Starch and protein in aged flour might be the components responsible for low solubility index in the same way, aged wheat starch is known to have increased hydrophobicity, more starch granule surface protein and thus decreased hydrophilicity (Seguchi, 1993).

Solvent Retention Capacity Profile

Each rice flour sample had a distinct SRC profile which implies that SRC test can be a good tool to provide a characteristic description of the functionality of not only wheat flour but also rice flour.The SRC test is a solvation assay for wheat flours that is based on the enhanced swelling behavior of functional flour components or individual flour polymer networks in selected single diagnostic solvents--lactic acid in water (for glutenin), sodium carbonate in water (for damaged starch), and sucrose in water (for pentosans)—which are used to predict the functional contribution of each individual flour component (Kweon *et al.*, 2011).

R7 had the highest solvent retention capacity profile for all solvents. The biggest difference between R7 and all the other samples is the particle size. This indicates that in addition to flour components, particle size also plays a great role in SRC test. The small particle size of R7 may have caused high adsorption of all solvents resulting to high SRC values.

Table 5. Solvent Retention Capacity

	Deionized water	50% Sucrose	5% Lactic acid	5% Sodium carbonate
R1	*124.2 ± 0.0^{b}	136.0 ± 0.0^{e}	109.5 ± 0.3^{d}	119.7 ± 0.0^{b}
R2	102.0 ± 0.0^{e}	137.1 ± 0.0^{d}	106.6 ± 0.2^{e}	118.8 ± 0.0^{b}
R3	104.1 ± 1.3^{d}	138.4 ± 0.4^{c}	114.2 ± 0.6^{b}	120.2 ± 2.2^{b}
R4	114.9 ± 0.0^{c}	131.0 ± 0.0^{g}	101.2 ± 0.6^{g}	112.5 ± 0.0^{c}
R5	102.6 ± 0.7^{e}	133.4 ± 0.2^{f}	111.4 ± 1.1^{c}	112.2 ± 0.7^{c}
R6	99.9 ± 0.2^{f}	210.5 ± 0.0^{a}	103.0 ± 2.1^{f}	105.8 ± 0.0^{d}
R7	175.1 ± 0.0^{a}	203.8 ± 0.0^{b}	186.0 ± 1.0^{a}	201.0 ± 0.0^{a}

*Means ± standard deviations, n=3

Lactic acid as previously described, enhances the swelling of the major wheat protein glutenin. Although rice do not contain glutenin, this study remains to use lactic acid in its SRC profile. Major proteins in wheat (glutenin) and rice (oryzenin) are both glutelin Osborne proteins. These glutelin Osborne proteins can be extracted by dilute acids such as lactic acid (Chrastil and Zarins, 1994). However, in this test, the protein was not dissolved and extracted but instead, it swelled and adsorb the solvent. Presence of disulphide bonds and high molecular weight of oryzenin might be responsible for its decreased solubility. Also, itis important to note that lactic acid SRC indicates flour protein functionality and not flour protein quality (Kweon *et al.*, 2011) , which explains why protein content presented in Table 2does not show the same trend with lactic acid SRC.

Sodium carbonate as mentioned above enhances the swelling of damaged starch. In accordance, results for the sodium carbonate SRC and for damaged starch as indicated in Figure 5 showed the same trend.

Starch damage

Damaged starch is the breakage of starch structure such as fragmented starch granules with exposed interior observed in flour after grinding or milling. Although not comparable, it is interesting to note that the typical starch damaged for bread flours are around 5-10 %. Certain degree of damaged starch granules is desirable in flour to maintain the quality of food products as damaged starch granules increase water-absorption capacity of flour and amount of substrates for yeast fermentation. Results obtained for starch damage showed aimilar trend with sodium carbonate SRC as shown in Table 5. Starch damage is found to be positively correlated with sodium carbonate SRC (r=0.9493, p<0.001). Compared to the other samples, K8 had the highest starch damage and the smallest particle size distribution Table 4. This is a likely indication that K8 was subjected to mechanical damage by dry milling to fine flour. All samples, except K8, were milled to flour using the same cyclone mill. However, starch damage was still significantly different from each flour. This suggests that there are other factors that may affect starch damage in addition to the type of mill. Possible factors are grain hardness and rate of milling.

Amylose content

Generally, japonica rice should have lower amylose content compared to that of indica rice. However, this trend was not shown in the results and japonica (R3, R4, R7) rice samples. Samples R1 and R2 had a relatively low amylose content compared to japonica rice.. This might be because they were store bought rice and were the so called soft indica rice.

Correlation analysis revealed that pasting properties: hot paste viscosity, cold paste viscosityand setback were found to be positively correlated with amylose content while breakdown is negatively correlated withamylose content as seen in Table 9Table 6.

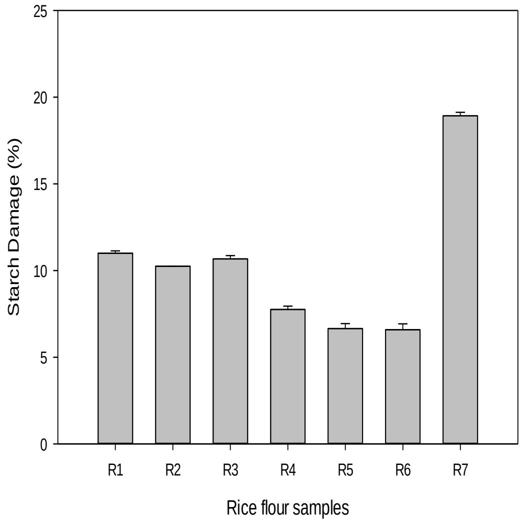

Figure 5. Starch damage

Table 6. Amylose content of rice flours

	Amylose Content (%)**		
R1	11.42	±	0.33*
R2	14.47	±	0.76
R3	14.47	±	0.28
R4	15.67	±	0.23
R5	21.46	±	0.43
R6	25.03	±	0.79
R7	14.22	±	0.91

*Means ± standard deviations, n=3
** flour basis.

Table 7. Correlation coefficent of pasting properties and amylose content

		Amylose content
Pasting properties	peak viscosity	-0.615 [a]
	hot paste viscosity	0.889**
	cold paste viscosity	0.938***
	breakdown	-0.869**
	setback	0.951***

[a] significantly different (Pearsons' correlation , *$p<0.05$, **$p<0.01$, ***$p<0.001$).

Alpha-amylase and Beta-amylase activities

Alpha-amylase and beta-amylase activities for all samples were generally low. Rice grains have generally conceded to contain alpha amylase and beta amylase (Shinke *et al.*, 1973). Alpha-amylase and beta-amylase are present in the bran and in the endosperm respectively. As seen below in Table 9 alpha-amylase seems to be more abundant than beta-amylase. However, the two are not comparable having different units, Ceralpha unit for one and Betamyl unit for another. In addition, although not comparable, falling number was also done for all samples and all attained more than 300 seconds in Table 8.

Table 8. Falling number of rice flours

	Falling number (sec)		
R1	430.5	±	0.5*
R2	472.5	±	1.5
R3	410.0	±	1.0
R4	403.0	±	11.0
R5	>500		
R6	>500		
R7	479.00	±	4.00

*Means ± standard deviations, n=2.

Table 9. Enzyme activities of rice flours

	alpha-amylase (CU/g of flour)**			beta-amylase (BU/g of flour)		
R1	4.86	±	0.08*	1.32	±	0.01
R2	4.19	±	0.19	1.15	±	0.00
R3	4.05	±	0.58	1.25	±	0.01
R4	3.93	±	0.13	1.31	±	0.05
R5	4.30	±	0.06	1.44	±	0.02
R6	3.56	±	0.13	1.30	±	0.01
R7	5.97	±	0.04	1.47	±	0.02

*Means ± standard deviations, n=3

** CU = Ceralpha Unit; Amount of enzyme, in the presence of excess thermostable α-glucosidase, required to release one micromole of *p*-nitrophenol from BPNPG7 in one minute under the defined assay conditions

BU = Betamyl-3 Unit; Amount of enzyme, in the presence of excess thermostable β-glucosidase, required to release one micromole of *p*-nitrophenol from PNPβ-G3 in one minute under the defined assay conditions.

Mixing Characteristic

The addition of 1.5% hydroxypropyl methylcellulose to rice flours enabled the rice flour-water mixture reach a consistency of 500 FU in the farinograph. Farinograph parameters are described in Table 10. Most of the rice flour had a gradual water uptake and gradual increase in consistency. As the consistency reached 500 FU, it remained at that point until the end of the test indicating that the rice dough is quite stable. This is reflected in the samples' stability time. However R7 has a noticeable breakdown at 4.5 minutes as seen in Figure 2. This seemed similar to the breakdown that is observed for wheat flour farinograms. However, this is not caused by gluten breakdown caused by shear. Rather, the peak observed was caused by abrupt water absorption of K8 flour, forming lumps. The lumps are responsible for the 500 FU consistency observed for a few minutes. Breakdown is observed when water absorbed in the lumps are distributed throughout the flour. In addition, it is interesting to note that farinogram parameters were found to be correlated with each other and several other flour physicochemical properties such as swelling power, solvent retention capacity, starch damage, enzyme. This indicates that mixing characteristics of rice flours are affected by many factors and the combination of these factors are evidently seen in the farinogram. This also proves that the use of farinograph and the addition of 1.5% hydroxypropyl methylcellulose to the rice flour is an effective tool in studying rice flour mixing characteristics

Table 10. Farinogram parameters

	Consistency (FU)	Water absorption (%)	Development time (mins)	Stability (mins)	Mixing Tolerance Index (FU)
R1	*502 ± 1 ^{bc}	66.6 ± 0 ^{e}	36.2 ± 3 ^{ab}	24.7 ± 0 ^{a}	7.5 ± 2 ^{bc}
R2	494 ± 2 ^{d}	67.2 ± 0 ^{d}	34.8 ± 2 ^{abc}	20 ± 1 ^{b}	8 ± 1 ^{b}
R3	504.3 ± 3 ^{b}	67.5 ± 0 ^{b}	33.7 ± 2 ^{abc}	22.1 ± 1 ^{ab}	7 ± 1 ^{bc}
R4	497.3 ± 3 ^{cd}	66.4 ± 0 ^{f}	31.5 ± 5 ^{bc}	24.6 ± 2 ^{a}	3.3 ± 1 ^{cd}
R5	506.3 ± 8 ^{d}	62.5 ± 0 ^{c}	36.7 ± 1 ^{abc}	18.3 ± 1 ^{b}	10.7 ± 1 ^{b}
R6	505.7 ± 1 ^{b}	65.4 ± 0 ^{h}	29.1 ± 2 ^{c}	24.8 ± 2 ^{a}	9.7 ± 1 ^{b}
R7	514 ± 3 ^{a}	109.7 ± 0 ^{a}	1.7 ± 0 ^{d}	3.5 ± 0 ^{c}	48.7 ± 4 ^{a}

*Means ± standard deviations, n=3; values followed by different letters in the same column are significantly different (Duncan's test $p<0.05$)

** Development time= time at the highest point of the curve

Stability = time of arrival at 500 ± 20FU consistency – time of departure at 500 ± 20FU consistency

Mixing Tolerance Index = highest consistency – consistency after 5 minutes.

Bake test

Bake test revealed that pentosans, starch damage and amylose content are key factors that influenced rice bread volume. As expected, all rice bread properties obtained from the rice

Table 11.Rice bread properties

	Height (cm)	Specific Volume (cc/g)	Volume Index	Symmetry Index	Uniformity Index
Rc*	11.7	2.65	32.3	2.8	1
Wc	14.9	4.47	42.2	2.5	1.2
R1	6.7 ± 0.4 ^{b}	1.56 ± 0.06 ^{b}	20.2 ± 0.9 ^{b}	-0.3 ± 0.4 ^{b}	-0.1 ± 0.2 ^{b}
R2	6.4 ± 0.3 ^{b}	1.43 ± 0.02 ^{c}	19.2 ± 0.5 ^{cd}	0 ± 0.4 ^{a}	0.2 ± 0.2 ^{a}
R3	6.3 ± 0.2 ^{b}	1.44 ± 0.06 ^{c}	18.8 ± 0.4 ^{d}	-0.1 ± 0.1 ^{a}	0.1 ± 0.1 ^{a}
R4	6.7 ± 0.4 ^{b}	1.47 ± 0.01 ^{bc}	20.2 ± 0.3 ^{b}	-0.1 ± 0.4 ^{a}	0 ± 0.3 ^{ab}
R5	6.4 ± 0.3 ^{b}	1.49 ± 0.05 ^{bc}	20.1 ± 0.5 ^{b}	-0.6 ± 0.3 ^{b}	-0.1 ± 0.1 ^{b}
R6	7.7 ± 0.2 ^{a}	1.71 ± 0.07 ^{a}	23 ± 0.6 ^{a}	0 ± 0.4 ^{a}	-0.1 ± 0.2 ^{ab}

*Means ± standard deviations, n=3; values followed by different letters in the same column are significantly different (Duncan's test *p*<0.05); Rc = rice flour control, Wc = wheat flour.

samples were all lower than the control wheat and control rice bread. Gluten in wheat flour responsible for the structure of the control rice bread. The control rice bread used commercially bought rice flour contained potato, wheat and corn starch which was blended in by the manufacturer.

Pearson's correlation analysis revealed that height, specific volume and volume index were positively correlated with sucrose solvent retention capacity (*r*=0.928, *p*<0.05; *r*=0.892, *p*<0.01; *r*=0.886, *p*<0.05) while volume index were negatively correlated with sodium carbonate solvent retention capacity (*r*=-0.797, *p*<0.05).

Most of the symmetry indices obtained were negative values unlike the those of the control. This indicated that rice breads had a slightly collapsed center. This could be due to the low gas retention ability of rice flours. Uniformity indices obtained were approximately 0, revealing that height of cakes are approximately uniform without one side exceedingly higher or lower. Bread from aged rice flour R5 had a more cracked top layer. This is also reflected in its corresponding rough farinogram curve. This could be caused by the low ability of flours from aged rice to absorb water and swell as explained in page 38.Regarding the air cell, R3, R4 and R2 had a very compact air cell at the bottom and had few medium sized air cells. Aged rice R5 also had a very compact air cell at the bottom but had a larger sized air cell on the top. R1 however had a more evenly spread air was responsible for the structure of the control wheat bread while additional starches were cell. R6 had the largest air cell among all the samples. It was more uniform and circular. This is reflected in volume and height of the bread in Table 11 Rice flour R6 with an intermediate to high amylose content obtained highest parameters in height, specific volume and volume index. Similar findings have found that intermediate amylose rice produce cakes with high volume expansion (Mohamed and Hamid, 1998).

Surprisingly, there is an observed negative correlation between ash content and height (*r*=-0.929, *p*<0.01), specific volume (*r*=-.839, *p*<.05), volume index (*r*=-0.857 *p*<0.05,). In addition, researches have also found a correlation ash content and cookie crumb grain made from wheat flour (Geng *et al.*, 2012). Further studies regarding internal structures of rice flour bake tests however is suggested.

Conclusion

There is a need for set of quality tests to control rice flour processing. Rice flour tests cannot directly adapt the well established set of quality tests for wheat flour because of the differences in composition, physicochemical and rheological properties. The methods used in this study such as proximate composition, particle size distribution, pasting properties, swelling power and solubility, solvent retention capacities, starch damage, amylose content, alpha-amylase and beta-amylase activity, mixing characteristics and bake test prove to reveal distinct characteristics of rice flour samples.

Proximate composition and particle size distribution gave a brief overview of rice flour samples.

Results in pasting properties of rice flours revealed that rice hot paste viscosity, cold paste viscosity and setback were statistical ($r\geq0.8$, $p<0.01$) positively correlated with amylose content while breakdown ($r=0.869$, $p<0.01$) is negatively correlated with amylose content.

In swelling power and solubility index, flour from aged rice R5 had a low swelling power of 2.97 g/g and 2.96 g/g at 60 and 70^{o}C respectively. In addition solubility indices were also low at 60, 70, 80 and 90 ^{o}C with 2.32, 2.26, 5, 6.89 for R5.R7 had the highest solvent retention capacity profile for all solvents. Since the biggest difference between R7 and all the other samples is the particle size. This indicates that in addition to flour components, particle size also plays a great role in SRC test.

Starch damage is found to be positively correlated with sodium carbonate SRC ($r=0.949$, $p<0.001$). R7 with the highest starch damage of 18.9% also had the highest sodium carbonate solvent retention capacity of 201.

The addition of 1.5% hydroxypropyl methylcellulose to rice flour made it possible for rice flour mixing characteristics be studied in a farinograph. Mixing characteristics of rice flours are affected by many factors and the combination of these factors are evidently seen in the farinogram. Farinograph parameters were correlated with each other and with other rice flour physicochemical properties. Water absorption is positively correlated with swelling power at 60^{o}C ($r=0.968$, $p<0.01$), water solvent retention capacity ($r=.952$, $p<0.001$), lactic acid solvent retention capacity ($r=0.974$, $p<0.001$), sodium carbonate solvent retention capacity ($r=.991$, $p<0.001$), alpha-amylase activity (0.822, $p<0.05$) and starch damage ($r=0.921$, $p<0.001$). Development time is negatively correlated with swelling power at 60^{o}C ($r=-0.964$, $p<0.001$), water solvent retention capacity ($r=-0.906$, $p<0.01$), lactic acid solvent retention capacity ($r=-0.921$, $p<0.001$), sodium carbonate solvent retention capacity ($r=-0.937$, $p<0.001$) and beta-amylase activity ($r=-0.821$, $p<0.001$). Stability time is negatively correlated with swelling power at 60^{o}C ($r=--0.833198$, $p<0.01$), water solvent retention capacity ($r=-0.833$, $p<0.01$), lactic acid solvent retention capacity ($r=-0.958$, $p<0.001$), sodium carbonate solvent retention capacity ($r=-0.940$, $p<0.001$), beta-amylase activity ($r=-0.827$, $p<0.01$), and starch damage ($r=-0.837$, $p<0.01$). Mixing Tolerance Index is positively correlated with swelling power at 60^{o}C ($r=0.909$, $p<0.01$), water solvent retention capacity ($r=.912$, $p<0.01$), lactic acid solvent retention capacity ($r=0.964$, $p<0.001$), sodium carbonate solvent retention capacity ($r=.965$, $p<0.001$), alpha-amylase activity (0.794, $p<0.05$) and starch damage ($r=0.854$, $p<0.01$).

In the bake test, differences in rice breads produced by different rice flours were observed in their cross sections through air cells and the top surface. Differences were also observed in specific volume and volume index. Further analysis of the bake test breads are suggested to further observe the effects of rice flours in breads.

The methods used in this study, revealed characteristics of rice flour that could serve as guidelines in rice flour quality control in the food industry.

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