# Effect of Storage Temperature on Biochemical and Mixolab Pasting Properties of Chinese *Japonica* Paddy

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# Abstract

This study investigated the changes in germination rate, contents of oleic acids, protein, water soluble sugars, enthalpy of flour gelatinization, and Mixolab dough pasting properties of three varieties of *japonica* rough rice after 18-month storage at four temperatures of 4,15,25 and 35 °C. After an 18-month storage, the paddy stored at or below 25 °C had more than 70% germination rate, their flour extracts by acid dve mixture of methyl red and bromothymol blue showed greenish color, but the paddy stored at 35 °C was only 30% germination rates, their flour extracts by acid dye mixture seem to be yellowish. With an increase in storage temperature, mositure content in paddy decreased, but total protein changed unsignificantly. In comparison to the paddy stored at 15 °C, the higher storage temperature (25 and 35 °C) tends to decrease the contents of damaged starch, water-soluble reducing sugars, total sugars, and uronic acids. The content of oleic acid in paddy stored at 4 °C was markedly higher than that at the temperatures of 15, 25 and 35 °C. Compared to lower temperate (4 and 15 °C), the higher storage temperature (35 °C) increased the gelatinization enthalpy of paddy flour and its starch determined by a differential scanning calorimetry (DSC), also increased Mixolab characteristic torque parameters such as starch gelatinization peak (C3), starch gelatinization minimum (C4), starch retrogradation minimum (C5), degrees of starch decay (C3-C4) and retrogradation (C5-C4) with a decrease in protein weakening (C2). The results of this study indicate storage temperature is an important factor affecting the physiological and biochemical properties of paddy, and lower temperature below room temperature are recommended to maintain paddy quality.

Keywords: paddy, storage, nutrition, mixolab dough pasting properties, enthalpy of flour gelatinization, ageing

## 1. Introduction

Rice is among the oldest of cultivated crops. History makes first mention of its being grown in China as early as 2800 B.C. Today nearly one half of the world's arable land is used for producing cereal grains, and about one-fifth of this is for the production of rice. As one of primary dietary sources of carbohydrates, rice plays important roles in meeting human energy requirements and nutrient intakes (Yang et al., 2006). China is the world's largest rice producer with annual production over 180 million tonne, and due to its large population, about 40 percent of its production is assigned to store longer periods (two years) in form of paddy under quasi-low temperature (around 20 °C) condition. A great deal of studies deal with the effect of short time storage on physicochemical characteristics and rheological properties of rice (Dhaliwal et al., 1991; Likitwattanasade & Hongsprabhas, 2010; Park et al., 2012 ), but only a few studies (Zhou et al., 2003; 2007; 2010; Zhang et al., 2012) reports the influence of longer time storage on rice deterioration and thermal properties. It is imperative to screen feasible biochemical and thermodynamic parameters for evaluation of paddy quality during storage.

Ageing during storage leads to numerous changes in the chemical and physical properties of rice (Noomhorm et al., 1997; Perdon et al., 1999; Pearce et al., 1999; Suzuki et al., 1999; Sowbhagya & Bhattacharya, 2001; Zhou et al., 2002; Sodhi et al., 2003; Patindol et al., 2005; Singh et al., 2006; Cao et al., 2009). These changes in rice pasting properties, color, flavor, and composition result in rice cooking and eating quality (Meullenet al., 2000; Teo et al., 2000; Jang et al., 2009) and depend on the rice variety, storage conditions, and amylose content. Such changes have been attributed to changes in cell walls and proteins, interaction between proteins and the degradative products of lipid oxidation, and starch-protein interaction (Sodhi et al., 2003; Park et al., 2012). The rapid viscosity analyzer (RVA) is commonly used to analyze the pasting properties of rice flour suspension

(Perdon et al., 1997; Sowbhagya & Bhattacharya, 2001; Zhou et al., 2002), but the mixolab curves could supply with information about water absorption rate, dough protein network and starch gelatinization (Rosell et al., 2007). This study investigated the changes in germination rate, contents of oleic acids and water soluble sugars, damged starch, enthalpy of flour gelatinization, and Mixolab dough pasting properties of three varieties of japonica paddy after 18-month storage at four temperatures of 4,15,25 and 35 °C, with an aim to explain the changes in functionality associated with storage.

## 2. Method and Materials

## 2.1 Experimental Sample and Preparation

Three varieties of japonica paddy, "Changyou No. 3", "Wuyuanjng", and "Yanfeng" " were used in this study (Table 1). Yanfeng was harvested in October, 2011 from Panjing city, Liaoning province. Changyou No.3 was obtained in late September, 2011 in Changyou city, Jiangsu province, and Wuyunjing was collected in November 2011 from Nanjing, Jiangsu province. Some samples were cleaned, repacked in 2-kg units in a polyethylene film bags and stored for 18 months in different temperature controller at 4 °C, 15 °C, 25 °C, or 35 °C before use. The other samples were treated at 40 °C and 80% RH for three days, and then stored at 35 °C, i.e. 40-35 °C treatment. After storage, the paddy samples were dehulled and milled, with an 800 µm screen used.

#### Table 1. Paddy samples

Paddy variety (abbr)	Producing region	Harvest time
Changyou No. 3 (CY)	Jiangsu Province	Sep. 2011
Wuyunjing (WYJ)	Jiangsu Province	Oct. 2011
Yanfeng (YF)	Jinlin Province	Oct. 2011

### 2.2 Moisture Content, Germination Rate and Rice Freshness

The moisture content of brown rice flour was determined at 105 °C in an air-oven for 3 h. The germination potential and rate of paddy was determined respectively at the fifth and the tenth day after being seminated at 30 °C. Freshness was determined by measuring light transmittance at 620 nm on rice flour extract with acid dye mixture of 0.05% methyl red and 0.15% bromothymol blue.

### 2.3 Contents of Free Fatty Acids and Oleic Acid

The content of free fatty acids (FFA) in rice flour was extracted by anhydrous ethyl alcohol, and then determined by hand titration with 0.01 M sodium hydroxide–ethanol solution using phenolphthalein as indicator. The FFA content was also determined by automatic potentiometric titration method with a potentiometer (ET38, electrode DG 111-SG, Mettler-Toledo).

Oleic acid content in brown rice flour was analyzed by the method of Goffman and Bergman (2003) with some revision. The lipid was extracted by vortex 6 g of milled rice for 15 min with 10 ml of isooctane, and then centrifuged at 4000 rpm for 5 min to collect the supernatant. An assay solution containing 4 ml of isooctane extract and 2 ml of 3% pyridine /5% cupric acetate reagent was shaken 5 min. After aqueous phase was fully fractionated, take 1mL isooctane fraction with a pipette to place in a 1-cm cuvette and measure absorbance at 715 nm. The oleic acid content of each extract was obtained from a calibration curve. Oleic acid (Sigma) was dissolved in isooctane to produce oleic acid solutions of 0-14.5 µmol/ml.

### 2.4 Content of-Soluble Total Sugars, Reducing Sugars and Pectic Acids of Paddy

Brown rice flour (1.0 g) was weighed into 50 ml plastic centrifuge tube, mixed with 15 ml of hexane, and then shaken at room temperature for 3 h. After centrifuged at 4000 rpm and 4 °C for 10 min, the precipitate was kept and fully volatilized hexane at a fume hood. The defatted meal was ground using a pestle and mortal with 15 ml of distilled water, then extracted for 5 min at room temperature. After centrifuged at 4000 rpm for 10 min, the supernatant was sample extracts. The content of total sugars was determined using phenol- sulfuric acid (Dubois et al., 1956). The extract (0.5 ml) was added 0.5 ml of 5% phenol aqueous solution, then 3 ml sulfuric acid, and carefully shaken. After cooled, the absorbency of 490 nm was measured.

The content of reducing sugars was determined by 3,5-dinitro salicylic acid (DNS) colorimetric method. The extract (800  $\mu$ l) was mixed with 600 ul of 4.4 mM DNS reagent, and boiled in water bath for 5 min. After cooled,

the solution was diluted to a volume of 10 ml with distilled water and the absorbency of 540 nm was measured. Glucose was used to make standard curve.

Uronic acid content was determined as described by Blumenkrantz et al. (1973). The extract (0.5 ml) was added 3 ml of 12.5 mM sodium tetraborate/ sulfuric acid, mixed, and boiled in water bath for 5 min. After cooled on ice to room temperature, the reactant was added 50  $\mu$ l of 0.15% m-hydroxydipheny/ 0.5% NaOH reagent, mixed and the absorbency of 520 nm was measured. Galacturonic acid (sigma) was used to make standard curve.

## 2.5 Contents of Protein and Damaged Starch of Paddy

The protein content in brown rice flour was determined by automatic nitrogen element rapid analyzer (Carbon Hydrogen Nitrogen Element Analyzer M366774, Elementar, Germany), the measured value was multiplied a coefficient of 5.95.

The damaged starch content was determined by SD matic analyzer (Chopin Technologies, France). One gram rice flour was weighed and put into small dipper of the analyzer, one drop of 95% ethanol, 3 g boric acid, 3 g potassium iodide and 120 ml distilled water were put into the reaction cup. The analyzer automatically raises temperature of the reaction cup to 35 °C, and flour sample automatically falls into reaction cup. This analyzer automatically calculates the content of damaged starch based on the residual iodide concentration. The result was present as the percentage of iodine adsorption (Ai%), since the content of damaged starch in brown rice flour expressed by either the UCD unit (the instrument manufacturer supply the calculation method) or UCDC unit (consider the effect of moisture and protein based on the UCD calculation) is negative.

### 2.6 Gelatinization Properties

The thermal properties of rice flour and purified starch were determined with a different scanning calorimeter (DSC) 200F3 (Netzsch, Germany). The sample (3.0-3.1 mg) was weighed at aluminium crucible and added distilled water to the ratio of water/sample as 2:1. The aluminium crucible was sealed and equilibrated at 4 °C overnight. DSC temperature was raised from 30 to 110 °C, with heating rate 10 °C /min.

Rice starch was isolated by combining 3 g brown rice flour with 30 ml of petroleum ether/anhydrous ethanol (v/v, 4:1) and shaking for 2 h at room temperature to defat. After centrifuged at 4000 rpm for 10 min, the residue was again added 30 ml of petroleum ether/anhydrous ethanol (v/v, 4:1) and shaken for 2 h. The precipitate was dipped in 12 ml of 0.1% Na<sub>2</sub>SO<sub>3</sub> solution for 1 h, and then centrifuged at 4000 rpm for 10 min. The precipitate was dissolved by 30 ml of 1% SDS solution and shaken 4 h. After centrifuged, the residue was washed four times with distilled water. Finally, the starch solid was dried at 45 °C over 5 h.

### 2.7 Mixolab Measurement

The mixing and pasting behavior of rice flour dough was measured using a Mixolab (Chopin Technologies, France) according to Rosell et al. (2007). Dough weighed at 90 g with 60% of water (14% w.b.) was evaluated. Both initial mixing temperature and water tank temperature were 30 °C. The hydration was 55%-65%. Target torque is  $0.8\pm0.5$  Nm. The mixing speed during the entire assay was 80 rpm/min. Firstly, the temperature was held at 30 °C for 8 min for the initial mixing. Secondly, the temperature increased at 4 °C /min to 90 °C within 15 min and then held for 7 min. Thirdly, the temperature decreased at 4 °C /min to 50 °C within 10 min and held for 5 min. The total test time is 45 min. The parameters of interest, given in Nm and including C1-C2 as well as C3-C4 and C5-C4, were recorded and calculated.

### 2.8 Statistical Analysis

Experimental data were subjected to analysis of variance using SPSS 11.5 for windows. Treatment means were tested separately for least significant difference at a 5% level of probability.

#### 3. Results

## 3.1 Changes in Moisture Content, Germination Rate and Freshness of Paddy

Table 2 shows the changes in moisture content, germination rate and freshness of paddy. The moisture content (m.c.) of paddy decreased with an increase in storage temperature. The germination potential and rate of three japonica paddy tended to decrease with increaseing storage temperature. Stored at 35 °C for 15 months, Changyou and Wuyunjing did not germinate, Yanfeng germinated 31%. At 25 °C storage condition, Changyou, Wuyunjing, and Yanfeng germinated 72%, 85%, 92%, respectively. At lower temperature (15 °C) storage, their germination rates all were over 95%.

The brown rice flour extracts by acid dye mixture of methyl red and bromothymol blue show that the paddy stored at 4 °C to 25 °C gave grass green or greenish color, but paddies stored at 35 °C was yellowish, even brown.

The acid dye extracts from paddy stored at higher temperature had higher transparency ( $T_{620}$ ) than those of paddy stored at 4 °C.

Rice	Storage	Moisture content	Germination	Germination	Freshness	
variety	Temp(°C)	(%)	Potential (%)	Rate(%)	T 620	Color
СҮ	4	14.71	95.7±4.9a	96.0±4.4a	18.45±0.04e	greenish
	15	14.33	93.0±1.0a	95.0±1.7a	31.88±0.01b	greenish
	25	13.63	65.7±2.1b	72.0±3.6b	26.57±0.03d	greenish
	35	13.28	0±0c	0±0c	71.76±0.06a	yellowish
	40-35	12.07	0±0c	1.0±1.7c	31.02±0.02c	yellowish
WYJ	4	14.75	95.7±2.1a	97.0±2.6a	16.31±0.06e	greenish
	15	13.35	97.7±1.5a	98.3±0.6a	34.26±0.04a	greenish
	25	12.90	91.3±1.5ab	92.3±2.5ab	27.35±0.05d	greenish
	35	11.49	0±0d	0±0d	33.46±0.03b	yellowish
	40-35	10.85	1.0±0c	1.3±0.6c	29.70±0.02c	yellowish
YF	4	13.92	86.3±0.6a	91.3±3.1a	19.46±0.03e	greenish
	15	13.17	91.7±2.9a	95.0±2.6a	24.57±0.17d	greenish
	25	12.41	81.0±1.7b	84.7±2.5b	25.65±0.07c	greenish
	35	10.76	29.0±2.6c	31.3±3.1c	28.95±0.01a	yellowish
	40-35	9.91	1.7±0.6d	6.3±3.5d	26.79±0.08b	yellowish

Table 2. Germination rate and freshness of paddy stored for eighteen months

Data are given as the means±SD (standard deviation) for triplicate.

3.2 Changes in Free Fatty Acid (FAA) and Oleic Acid of Paddy

Table 3. Fatty acid content of paddy stored for eighteen months

	Storage temp.( °C)	СҮ	WYJ	YF
Free fatty acids	4	23.01±7.53b	32.97±2.85b	8.15±2.82c
(mgKOH/100g)	15	65.35±2.26a	52.51±5.69a	44.39±17.09b
Hand titration	25	59.99±2.34a	60.31±7.94a	39.16±12.44b
	35	64.58±10.43a	60.90±5.61a	90.3±7.74a
	40-35	47.76±15.76a	$0\pm0c$	31.03±0b
Free fatty acids	4	44.44±0.37b	52.47±1.02a	62.80±4.42a
(mgKOH/100g)	15	45.92±2.44b	43.58±0.71c	46.72±0.96e
Potentiometric	25	45.54±2.43b	48.96±2.02b	48.71±0.32c
titration	35	66.18±3.68a	49.08±2.12b	48.08±0.19d
	40-35	45.21±1.17b	48.19±1.80b	53.38±2.09b
Oleic acid	4	938.36±55.91a	2202.76±27.28a	1517.34±85.94a
(µg/gdw)	15	403.81±9.65e	608.42±10.53c	651.75±5.95c
	25	495.22±26.21c	522.49±6.18d	698.28±11.6b
	35	567.51±43.79b	869.90±37.78b	696.12±3.22b
	40-35	459.23±6.07d	432.9±20.52d	657.07±16.46c

Data are given as the means  $\pm$  SD for triplicate.

Table 3 shows changes in the content of paddy FAA determined by different methods. With phenolphthalein as an indicator, the KOH- ethanol solution was used to manually titrate the paddy FFA extracted by anhydrous ethyl ethanol solution. The FFA in paddy stored at 15 °C to 35 °C was clearly higher than that stored at 4 °C. The paddy FFA measured by this method did not show linear relationship with storage temperature in the range of 15 °C to 35 °C.

The paddy FFA extracted by anhydrous ethyl ethanol was titrated with KOH- ethanol solution using potentiotitration method and pH glass electrode as indicating electrode. With addition of titration reagent, the potential (E) changed with the volume (V) of titration reagent. When E occur a transition, the end of titration reached. The FFA contents in Wuyun jing and Yanfeng stored at 15 to 35 °C were clearly lower than that stored at 4 °C, but Changyou stored at 15 to 35 °C had higher FFA content than at 4 °C.

The paddy FFA content was also determined by colorimetric method with cupric acetate-pyridine as a color developing reagent. The oleic acid was used to make standard curve. The content of oleic acid in paddy stored at 4 °C was markedly higher than that at the temperatures of 15 °C, 25 °C and 35 °C.

### 3.3 Changes in Water-Soluble Sugars, Crude Protein and Damaged Starch of Paddy

Table 4.	Changes in	water-soluble sugars,	protein and	damaged s	starch of <i>i</i>	paddy	v stored for	r eighteen	months
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Rice	Storage	Reducing	Total sugars	Pectic acids	Protein	Iodine value
variety	temp.	sugars	(mg/g dw)	(mg/g dw)	(%d.b.)	(Ai%)
	(°C)	(mg/g dw)				
CY	4	7.56±0.64d	27.83±1.41a	0.68±0.01cd	8.81	80.47±0.06b
	15	17.96±0.09a	25.07±0.24b	1.03±0.01a	8.98	81.52±0.58a
	25	13.68±0.64b	22.59±0.40c	0.79±0.06c	8.81	78.23±0.23c
	35	13.44±0.14b	17.90±1.86d	$0.72 \pm 0.07c$	8.79	77.36±0.38d
	40-35	8.88±0.67c	20.17±1.38d	0.96±0.05b	8.88	75.85±0.36e
WYJ	4	8.23±0.07c	21.30±0.94b	0.30±0.01d	10.03	82.47±0.47a
	15	15.04±0.42a	27.62±1.97a	1.20±0.05a	10.29	78.49±0.22b
	25	11.38±0.25b	21.29±0.75b	0.77±0.06c	10.21	78.45±0.30b
	35	11.80±0.24b	20.96±0.40b	$0.87 \pm 0.02b$	10.11	77.50±0.34c
	40-35	8.59±0.51c	21.60±1.02b	0.92±0.03b	9.93	75.96±0.05d
YF	4	12.88±0.36b	19.45±0.64ab	0.71±0.01c	10.07	80.03±0.13a
	15	15.66±0.12a	21.66±1.57a	1.01±0.02a	9.21	76.56±0.59b
	25	14.27±0.48a	21.83±1.17a	0.87±0.07ab	10.72	73.60±0.12c
	35	11.85±0.57c	19.81±0.87a	0.95±0.05a	11.36	73.02±0.42d
	40-35	8.09±0.47d	20.61±1.59a	0.65±0.06c	10.43	72.36±0.53d

Data are given as the mean±SD for triplicate. The protein content is the mean of two deterimations.

Table 4 shows the changes in water-soluble reducing sugar, total sugar, pectic acid, and crude protein, as well as damaged starch of paddy. In comparison to 15 °C storage, the contents of water-soluble reducing sugar, total sugar, and pectic acid tended to decrease in paddy stored at 25 °C to 35 °C. The content of paddy crude protein changed insignificantly with an increase in storage temperature. Compared to 4 °C and 15 °C storage, the damaged starch in paddy stored at 25 °C was reduced.

## 3.4 Changes in Thermal Properties of Paddy

Rice	Storage temp.	To	T <sub>p</sub>	T <sub>c</sub>	Peak width	Peak enthalpy	ΔH
Variety	(°C)	(°C)	(°C)	(°C)	(°C)	(W/mg)	(J/g flour)
CY	4	61.8±0.4a	67.2±0.4a	72.9±0.5a	11.1±0.9a	0.1132±0.0094a	5.5193±0.775b
	15	61.5±0.2a	66.9±0.3a	72.3±0.6a	10.8±0.4a	0.1131±0.0145a	5.724±0.503b
	25	61.7±0.6a	67.0±0.3a	72.5±0.9a	10.8±0.6a	0.1121±0.0129a	5.254±0.894b
	35	61.7±0.4a	67.4±0.2a	74.0±0.6a	12.3±1.7a	0.1122±0.0122a	7.145±0.549a
	40-35	61.8±0.2a	67.3±0.2a	73.1±0.5a	11.3±0.5a	0.1206±0.0116a	7.0170±0.672a
WYJ	4	64.7±0.8a	70.7±0.5ab	77.1±0.7a	12.0±1.2a	0.1339±0.0174a	6.046±0.591c
	15	66.2±0.5a	71.9±0.5a	77.4±0.6a	11.2±0.3a	0.1204±0.0134a	6.551±0.461c
	25	65.7±0.9a	71.5±0.5a	77.4±0.5a	11.7±0.3a	0.1367±0.0166a	6.498±0.327c
	35	66.2±0.4a	71.7±0.2a	77.5±0.2a	11.3±0.6a	0.1286±0.0118a	7.498±0.261b
	40-35	66.5±0.6a	71.7±0.8a	77.8±0.4a	11.3±0.5a	0.1208±0.0166a	8.216±0.432a
YF	4	64.2±0.7a	70.6±0.2a	76.4±0.3a	12.3±0.6a	0.1050±0.0072ab	6.125±0.541c
	15	64.3±0.4a	70.6±0.2a	76.5±0.1a	12.3±0.3a	$0.1069 {\pm} 0.0081 ab$	6.402±0.919c
	25	64.1±0.4a	70.8±0.1a	76.2±0.9a	12.0±0.5a	0.1286±0.0136a	7.868±0.422ab
	35	64.6±0.2a	70.9±0.3a	76.4±0.2a	11.8±0.4a	0.1267±0.0158a	8.289±0.741a
	40-35	64.7±0.8a	70.7±0.5a	77.1±0.7a	12.1±0.2a	0.1339±0.0174a	9.014±0.846a

Table 5. DSC data for rice flour ground from paddy storage for eighteen months

 $T_{o}$ , onset of gelatinaiztion;  $T_{p}$ , peak temp.;  $T_{c}$ , conclusion temp.;  $\Delta H$ , enthalpy of gelatinization; Data are given as the mean  $\pm$  SD for triplicate.

Table 5 shows the effect of storage temperature on the thermal properties of rice flour ground from three japonica paddy. With increase in storage temperature, the onset temp.  $(T_o)$ , peak temp.  $(T_p)$ , and conclusion temp.  $(T_c)$  of three paddies, as well as peak width and peak enthalpy, all changed insignificantly, but their flour gelatinization enthalpy clearly increased. The values of  $T_o$ ,  $T_p$  and  $T_c$  for Changyou were markedly lower than those of Wuyunjing and Yanfeng. The gelatinization enthalpy of Changyou was slightly lower than those of Wuyunjing and Yanfeng. The values of  $T_o$ ,  $T_p$ ,  $T_c$  and gelatinization enthalpy of Wuyunjing was similar to those of Yanfeng.

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Rice Variety	Storage temp.	T <sub>o</sub> (°C)	T <sub>p</sub> (°C)	T <sub>c</sub> (°C)	Peak width	Peak enthalpy	∆H (J/g starch)
	(°C)				(°C)	(W/mg)	
СҮ	4	60.1	64.9	71.4	11.3	0.1531	7.055
	35	60.2	65.4	70.6	10.4	0.1665	7.210
WYJ	4	68.3	72.3	77.5	9.2	0.2129	8.738
	35	66.2	71.4	76.9	10.7	0.1847	8.748
YF	4	66.4	71.3	76.7	10.3	0.1757	8.072
	35	67.1	71.9	77.6	10.5	0.1841	8.342

Table 6. DSC data for purified starch from rice flour after paddy storage for eighteen months

 $T_o$ , onset of gelatinaiztion;  $T_p$ , peak temp.;  $T_c$ , conclusion temp.;  $\Delta H$ , enthalpy of gelatinization; Data are given as the mean for two determinations.

In order to preclude the influence of lipid, protein, cellulose, and hemicellulose, as well as soluble sugars and minerals on starch thermal property, we used purified rice starch to analyze DSC data. It was confirmed that the gelatinization enthalpy of rice starch from paddy stored at 4 °C tended to be lower than that stored at 35 °C (Table 5). The values of  $T_0$ ,  $T_p$ ,  $T_c$ , peak width, and enthalpy of gelatinization in purified rice starch were similar to those of brown rice flour, but its peak enthalpy markedly increased. The value of  $T_0$ ,  $T_p$  and  $T_c$  in purified starch of Changyou were markedly lower than those of Wuyunjing and Yanfeng. The enthalpy of gelatinization in purified starch of Changyou was slightly lower than those of Wuyunjing and Yanfeng. The values of  $T_0$ ,  $T_p$ ,  $T_c$  and enthalpy of gelatinization in purified starch of Yanfeng.

## 3.5 Changes in Mixolab Dough Pasting Properties of Paddy

Table 7 shows the effect of storage temperature on mixolab dough pasting properties of paddy after 18-month storage. The first part of the curve in Figure 1, before the heating-cooling cycle starts, allows water absorption of the flours to be determined. The water addition level of kneading dough was constant 60% or 65% flour basis for three rice varieties. The dough development time (DDT) increased as the storage temperature of paddy increased. Compared to 15 °C storage, C1 consistency peak torque (Nm) reduced as the storage temperature of paddy increased. The amplitude of kneading dough (AKD) at 30 °C was in the range of 0.02 Nm to 0.04 Nm for three rice varieties. Dough stability time (min) shows the dough behavior during mixing, which increased as the storage temperature of paddy increased.

Paddy	<b>C</b>	Water	DDT	C1	AKD	DST	C2	C1-C2	C3	C4	C3-C4	C5	C5-C4
Var.	Storage temp. (° C)	level%	(min)	torque (Nm)	(Nm)	(min)	torque (Nm)	(Nm)	Torque (Nm)	torque (Nm)	(Nm)	torque (Nm)	(Nm)
CV	4	65	2.03	0.29	0.02	3.63	0	2.03	1.59	0.86	0.73	1.37	0.51
CI	15	60	1.52	0.75	0.04	2.93	0.23	1.29	1.77	1.16	0.61	1.81	0.65
	25	60	2.08	0.42	0.03	5.93	0.13	1.95	1.86	1.1	0.76	1.73	0.63
	35	60	6.35	0.58	0.04	9.1	0.37	5.98	2.18	1.32	0.86	1.98	0.66
	40-35	60	5.77	0.33	0.04	8.5	0.17	5.6	1.96	1.17	0.79	1.8	0.63
XX/X/ I	4	65	2.62	0.55	0.02	5.87	0.24	2.38	1.66	1.16	0.5	1.68	0.52
WYJ	15	60	1.8	0.6	0.03	6.03	0.19	1.61	1.68	1.2	0.48	1.78	0.58
	25	60	2.78	0.3	0.03	8.7	0.07	2.71	1.74	1.13	0.61	1.75	0.62
	35	60	6.63	0.31	0.03	9.85	0.13	6.5	1.9	1.23	0.67	1.89	0.76
	40-35	60	4.83	0.31	0.03	8.22	0.09	4.74	1.75	1.17	0.58	1.82	1.65
VE	4	65	2.88	0.32	0.02	6.32	0.12	2.76	1.52	1	0.52	1.44	0.44
ΥΓ	15	60	1.97	0.41	0.03	5.48	0.1	1.87	1.68	1.12	0.56	1.7	0.58
	25	60	4.55	0.33	0.02	7.12	0.07	4.48	1.8	1.16	0.64	1.79	0.63
	35	60	5.13	0.26	0.03	9.1	0.04	5.09	1.78	1.13	0.65	1.7	0.57
	40-35	65	5.17	0.57	0.02	9.85	0.32	0.25	2.32	1.56	0.76	2.29	1.13

Table 7. Mixolab pasting properties of rice flours from paddy

Note: Water level%, constant water level (%flour basis); DDT,dough development time (min); C1, consistency peak torque (Nm); AKD, amplitude of kneading dough (Nm); DST, dough stability time (min); C2, consistency minimum torque (Nm); C1-C2, protein weakness; C3, pasting viscosity peak torque (Nm); C4, pasting viscosity minimum torque (Nm); C3-C4, starch decay (Nm); C5, starch retrogradation final torque (Nm); C5-C4, Starch setback (Nm).



Figure 1. Mixolab analysis of brown rice dough behavior during mixing, heating and cooling

The minimum torque (C2) of dough consistency is related to weakening of the protein network from mechanical and thermal constraints. The high C2 minimum torque shows strong protein network. At five storage temperatures, C2 minimum torque was in the range of 0.01 to 0.37 Nm, 0.07 to 0.24 Nm and 0.04 to 0.12 Nm for Changyou, Wuyunjing and Yanfeng, respectively. Protein weakness (C1-C2) increased with an increase in storage temperature of three varieties of paddy.

The second part of the curve shows the behavior of rice starch gelatinization. C3 pasting viscosity peak (Nm) shows starch gelatinization rate, which increased as the storage temperature of paddy increased, but C4 pasting viscosity minimum torque (Nm) and C5 starch retrogradation end (Nm) tended to increase. In contrast to lower temperature storage, higher temperature storage increased starch gelatinization peak (C3), starch gelatinization minimum (C4), starch retrogradation minimum (C5), degrees of starch decay (C3-C4) and setback (C5-C4).

## 4. Discussion

Fat acidity is usually used as an index of quality deterioration during rice storage since lipid dissolution is considered to be more rapidly than protein and starch (Genkawa et al., 2008). This study showed that long 35 °C storage seemed to increase FFA content, but significantly decreased oleic acid content compared with 4 °C storage. This demonstrated that the rice triglycerides were degraded during storage. These changes might involve the hydrolysis of lipids to produce FFA and the oxidation of lipids (including FFA) into hydroperoxides and other secondary products (Gregory et al., 2008). Yasumatsu et al. (1964) suggested that FFA quantity increased during storage and bind with amylose in starch, and the increase in fatty acid-amylose complex may affect the maximum viscosity of the amylogram. Hence, the increase of FFA content in paddy during long time storage (over 15 months) should be cautious to adopt, and other sensitive indicator is need to explore.

The duration and temperature of storage were found to markedly affect the enthalpy and temperature of gelatinization and retrogradation of rice flour (Fan & Marks, 1999). Starch and in particular, amylose became more insoluble following the higher temperature storage (Rajendra & Zakiuddin, 1991; Patindol et al., 2005; Zhou et al., 2007). The RVA pasting curves of rice flours have shown that the changes in pasting properties of milled rice during storage contained increased setback (Sowbhagya and Bhattacharya, 2001) and decreased breakdown(Zhou et al., 2003; Park et al., 2012). In this study, compared to lower temperate (4 and 15 °C), the higher storage temperature (35 °C) increased the enthalpy of paddy flour gelatinization determined by a differential scanning calorimetry (DSC) and decreased damaged starch, also increased Mixolab characteristic torque parameters such as starch gelatinization peak (C3), starch gelatinization minimum (C4), starch retrogradation minimum (C5), degrees of starch decay (C3-C4) and retrogradation (C5-C4) with a decrease in protein weakening (C2). This suggested that longtime higher temperature storage lead to more ordered structure of deteriorated rice, reducing the swelling and cracking of starch granula and leaking of starch components. This change in aged rice thermal behaviors maybe increase optimum cooking time. Interestingly, from the DSC thermograms we found the gelatinization enthalpy of rice purified starch from 4 °C storage tended to be higher than that of 35 °C long storage. This indicates the effect of storage on rice thermal properties relates to the interaction between starch and non-starch components after long storage. The effect of non-starch components such as protein and non-starch polysaccharides on the thermal properties of ageing rice need further study.

## 5. Conclusion

After18-month storage, the paddy stored at or below 25 °C had more than 70% germination rate, their flour extracts by acid dye mixture of methyl red and bromothymol blue showed greenish color, but the paddy stored at 35 °C was only 30% germination rates, their flour extracts by acid dye mixture seem to be yellowish. With an increase in storage temperature, moisture content in paddy decreased. The content of oleic acid in paddy stored at 4 °C was markedly higher than that at the temperatures of 15, 25 and 35 °C. Compared to lower temperate (4 and 15 °C), the higher storage temperature (35 °C) increased the enthalpy of paddy flour gelatinization determined by a differential scanning calorimetry (DSC), also increased Mixolab characteristic torque parameters such as starch gelatinization peak (C3), starch gelatinization minimum (C4), starch retrogradation minimum (C5), degrees of starch decay (C3-C4) and retrogradation (C5-C4) with a decrease in protein weakening (C2). The results of this study indicate storage temperature is an important factor affecting the physiological and physicochemical properties of paddy, and lower temperature below room temperature are recommended to maintain paddy quality.

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