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Comparison of Grain Quality Traits between *Japonica* Rice Cultivars from Korea and Yunnan Province of China

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Abstract

Improving eating quality is one of the most important objectives in *japonica* rice breeding programs in Yunnan Province of China. Eating quality and its relevant traits of nine Korean and 11 Yunnan rice cultivars were comparatively analyzed in this study. The grain shape of most Yunnan *japonica* rice cultivars have a relatively slender shape and are slightly larger than Korean rice cultivars. Palatability value of cooked rice of Yunnan rice cultivars was significantly lower, while the protein content of Yunnan rice cultivars was significantly higher than that of Korean cultivars. Peak viscosity and breakdown viscosity of the Yunnan rice cultivars were significantly lower, while setback viscosity of the Yunnan rice cultivars was significantly higher than in Korean rice cultivars. Palatability value of cooked rice was negatively correlated with protein content and setback viscosity but positively correlated with peak viscosity, breakdown viscosity, and cool paste viscosity. Through multiple linear regression analysis, an equation for estimating palatability value (PV) of cooked rice based on quality traits was generated as dependent only upon protein content (PC), $PV=139.024-(10.865\times PC)$ with an R^2 value of 0.822. The results suggest that reducing protein contents should be the major target in improving eating quality of Yunnan japonica rice cultivars through integrated approaches of both cultivar development and appropriate cultural practices. Genetic similarities among cultivars based on DNA markers which had been identified as associated with grain quality seemed not to be directly related to PV.

Key words: *Japonica* rice, grain quality, palatability, physicochemical property, viscosity, DNA marker

Introduction

Rice is the staple food for about half of the world's population. Improving rice grain quality has been a major concern in rice breeding programs to meet the market demand. The traits of rice grain quality include milling quality, appearance quality, cooking and eating quality, and nutritional quality, of which cooking and eating quality is the most important component for the Asian customers who mainly consume rice (Mo 1993). Many studies have been conducted to increase the fundamental understanding on physicochemical properties and the genetic basis of the cooking and eating qualities of rice. Cooking and eating qualities are known to be directly related with some physicochemical properties of endosperm such as amylose content, protein content, and pasting viscosity (Matsue 1993). Protein content (PC) is known to be negatively correlated with cooking and eating quality and amylogram properties (Shen et al. 2003). Rapid Visco-Analyzer (RVA) which can analyze starch pasting viscosity has been widely used in evaluating rice cooking and eating quality because of its simplicity and rapidity with a small amount of samples (Bao and Xia 1999). It was regarded that RVA profiles are appropriate for distinguishing eating quality among rice varieties having similar amylose content (Shu et al. 1998) and the profiles could be used as indices to assist in the

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selection of high quality rice (Sui et al. 2005; Wu et al. 2001;).

With the advantage of DNA markers which produce polymorphic alleles and are suitable for evaluating genetic diversity among closely related rice cultivars, the application of DNA markers has broadened the understanding of the genetic diversity and complex quantitative traits (Ji et al. 1998). Suh et al. (2004) classified *japonica* rice varieties into six major groups by clustering analysis based on 81 SSR markers and has detected 18 SSR markers which are significantly associated with the four grain-quality traits including taste value, alkali digestion value, amylose content, and protein content. Ohtsubo et al. (2003) developed DNA markers for estimating palatability value (PV) of cooked rice. Bao et al. (2006a, 2006b) reported that polymorphisms in starch-synthesizing genes were associated with starch properties and are useful for marker-assisted breeding for improving grain quality.

Yunnan Province is considered as the center of genetic diversity for Chinese cultivated rice and the central region and natural treasure house of good genetic resources in China (Zeng et al. 2001). In Yunnan Province, rice is cultivated from an altitude of 76 m to 2,650 m above sea level (ASL). In general, *indica* rice is cultivated in the area under 1,500 m ASL, and *japonica* rice is mostly above 1,500 m ASL. Even though the total cultivated area of *japonica* rice is smaller than that of *indica* rice in Yunnan province, the total production of *japonica* rice is higher than that of *indica* rice. Therefore, *japonica* rice cultivars play an important role in rice production in Yunnan Province due to their higher yield potential and greater adaptability. However, cooking and eating quality of these *japonica* rice cultivars are not good as other *japonica* rice cultivars grown in other areas such as Korea and Japan. Moreover, research on the grain quality of Yunnan *japonica* rice cultivars is rare. One recent study, the Sino-Japanese cooperative research program from 1982 to1996 on the evaluation and utilization of rice resources, showed that eating quality of Yunnan *japonica* rice cultivars was very poor and amylose content of the most cultivars ranged from 17 to 24% and grade of appearance quality was very low. Since 1992, the eating quality of many Yunnan rice cultivars has been greatly improved by the breeding program using the Japanese elite cultivars, Koshihikari and Todorokiwase, as crossing parents, but is still relatively lower than Korean and *Japanese* rice cultivars (Liu et al. 2000). In Korea, many studies and breeding programs have been conducted since the end of 1970s to improve eating quality, developing lots of high quality cultivars such as Odae, Jinmi, Ilpum, Dongjin, and Samgwang which show even higher PV than Chucheong and Koshihikari (Han and Sheng 1996).

The objective of this study is to compare the eating quality traits between *japonica* rice cultivars from Yunnan Province, China and Korea and thus to provide the information for effective approaches to improve eating quality of *japonica* rice in Yunnan Province.

Materials and Methods

Rice materials and cultivation

A total of 20 elite commercial *japonica* rice cultivars, nine cultivars harvested in Suwon, Korea (i.e. Ilpum, Gopum, Samgwang, Koshihikari, Dongjin, Chucheong, Nampyeong, Shindongjin, and Odae) and eleven cultivars harvested in Kunming, Yunnan, China (i.e. Dianjingyou 1; Yunjing 17, 19, 21, and 22; Yunjingyou 10, 14, and 15; Chujing 27, Hexi 41, and Yunzikang 21) were used in this study. Korean cultivars were cultivated in Suwon, Korea and Yunnan cultivars were cultivated in Kunming, Yunnan Province, according to conventional cultural practices in each area.

Experimental environments and cultural management

Korean rice cultivars were sown in plastic tunnel-seed beds on 27th April and then 34-day-old seedlings were transplanted in one plant per hill with 30 × 15 cm planting density on the experimental farm of Seoul National University (Suwon, Korea, altitude: 74 m, longitude: 127°36′ E, latitude: 37°51′ N). The fertilizer was applied at the rate of 110-80-80 kg N-P-K ha^{-1}. In Kunming, seeds were sown in the seedling nursery on 20th March then transplanted with one seedling per hill on 9th May on the experimental farm of Yunnan Academy of Agricultural Sciences (Kunming, China, the high-altitude area, altitude: 1,916 m, longitude: 102°41′ E, latitude: 25°1′ N). The planting density was 17.5 × 10 cm and a mixed chemical fertilizer was applied at the rate of 140-14-20 kg N-P-K ha^{-1}. In Suwon, all of the Korean cultivars reached grain-filling stage and maturing stage in the second ten days of August and October, respectively. In Kunming, the cultivars reached grain-filling stage and maturing stage in the last ten days of July and September, respectively. The average monthly temperatures of Suwon during the rice season were higher than those of Kunming from 1.4 to 4.5 ℃ except that of May in which the average monthly temperature of Suwon was 2 ℃ lower than that of Kunming (Table 1).

Table 1. The average monthly temperature, day length, and rainfall during rice-growing period in Suwon, Korea and Kunming, China.

Month	Mean temperature(℃)		Sun duration (hours)		Rainfall (mm)	
	Suwon	Kunming	Suwon	Kunming	Suwon	Kunming
May	17.9	19.9	189.0	198.7	124	111
June	22.0	20.6	158.8	136.8	135	227
July	24.3	20.6	94.5	132.3	382	251
August	25.2	20.7	165.1	145.8	157	115
September	21.2	18.4	172.1	127.5	183	56.9

Phenotypic analysis

After harvest, the rice grains were air-dried and stored at room temperature for three months and hulled to produce brown rice. Then the rice samples were polished to 90% of brown rice by a polishing machine (MC-90A, Toyoseiki), and then milled through a 100-mesh screen by Cyclotec Sample Mill (Foss North America (Tecator) No.1093-003). Grain size and shape of brown rice were measured by profile-magnifying projector analyzer. Amylose content(AC)was measured according to the method of Williams et al. (1969). Protein content (PC) was

measured by Kjeldahl method (Vapodest Gerhardt, Germany). Palatability value (PV), which is a good measure of eating quality (Hwang 2004), was evaluated with 33 g of milled rice per sample using TOYO Mito meter (MB-90A and MA-90B, Japan). Alkali digestion value (ADV) was evaluated after incubation in 1.4% KOH solution at 30 °C for 23 h. Viscogram parameters were analyzed with a Rapid Visco-Analyzer (RVA-4, Newport Scientific, Australia) according to the American Association of Cereal Chemists (AACC) Standard Method (1995 61-02). Rice paste viscosity properties were described by eight parameters of the pasting curve: peak time (PET), pasting initial temperature (PAT), peak viscosity (PKV), hot paste viscosity (HPV), cool paste viscosity (CPV), breakdown viscosity (BDV = PKV − HPV), setback viscosity (SBV = CPV − PKV), and consistency viscosity (CSV= CPV − HPV). All viscosity parameters were measured in rapid visco units (RVU) with three replications for each sample.

DNA extraction and analysis

Total genomic DNA of each cultivar was extracted from fresh young leaves using the CTAB method (Causse et al. 1994). The primers used in detecting polymorphism of 20 *japonica* rice cultivars are shown in Table 7. STS-series were designed according to Ohtsubo et al. (2002, 2003), SNP-series were designed according to Bao et al. (2006a, 2006b), and Indel-series were designed by Crop Molecular Breeding Lab of Seoul National University in Korea (Chin et al. 2007). Each 20 ul amplification reaction mixture contained 50 ng DNA, 5 pM of each primer, 2 ul PCR buffer [100 mM Tris-HCl (pH 8.3), 500 mM KCl, 20 mM $MgCl_2$, and Enhancer solution], 250 uM of each dNTPs, and 0.5 unit Taq polymerase. All amplifications were performed on the MJ Research PCR system (Applied Biosystem) under the following conditions: 5 min at 94 °C, followed by 35 cycles of 45 s at 94 °C, 45 s at 55 °C or 58 °C, and 45 s at 72 °C, then 5 min at 72 °C for a final extension. The amplified PCR products were resolved by electrophoresis in 3% agarose gel. The DNA band patterns were recorded as 1 for present and 0 for absent for each allele and were subjected to the multiple linear regression analysis for estimating PV and cluster analysis for the total 20 cultivars used in this study.

Data analysis

T-test analysis, correlation analysis, and multiple regression analysis were performed using SAS 6.12 software (SAS, Institute Inc., Cary, NC, USA, 1990). Genetic similarities between cultivars were obtained by the UPGMA method using the NTSYS program to construct a dendrogram (Rohlf 2002).

Results

Variations of appearance quality of rice

Grain size, grain shape, 1000-grain weight, and percentage of chalky grains were not significantly different between Korean and Yunnan rice cultivars (Table 2). Percentage of chalky grains was relatively lower in Korean cultivars but without a significant difference. This indicates that all rice cultivars tested had ellipse grains and favorable appearance quality in both Korea and Yunnan except Dianjingyou 1, a Yunnan cultivar which exhibited a long slender grain shape.

Table 2. Grain size and shape of brown rice of 20 cultivars from Korea and Yunnan, China.

Month	Varietal origin	No. of cultivars	Grain length(mm)	Grain width(mm)	Length-width ratio	1000-grain weight (g)	Ratio of chalky grains (%)
Mean	Korea	9	5.05	2.84	1.78	21.86	4.95
	Yunnan	11	5.18	2.72	1.92	20.62	11.97
Range	Korea	9	4.85-5.58	2.77-2.93	1.70-1.91	19.80-26.10	0.17-21.67
	Yunnan	11	4.76-6.18	2.40-2.99	1.60-2.57	17.70-25.00	0.83-79.87
C.V. (%)	Korea	9	4.79	1.50	4.16	9.52	151.48
	Yunnan	11	7.13	6.32	12.53	10.79	191.52
T-test			ns	ns	ns	ns	ns

ns: Not significant at $\alpha = 0.05$ by t-test.

Physicochemical characteristics and palatability

PV, PC, ADV, and AC are the basic physicochemical traits for evaluating rice grain quality. PVs of Korean rice cultivars were significantly higher than those of Yunnan rice cultivars, while PC was significantly lower than that of Yunnan rice cultivars (Table 3). The lowest PV was 70.7 and the highest PC was 6.76% in Korean rice cultivars, while the highest PV was 66.4 and the lowest PC was 6.75% in Yunnan rice cultivars. Even the lowest PV of Korean rice cultivars was higher than the highest PV of Yunnan rice cultivars, indicating that eating quality of most of the representative cultivars in Yunnan Province was lower than Korean cultivars. On the contrary, the highest PC of Korean rice cultivar was similar to the lowest PC of Yunnan rice cultivars. However, the average values of ADV and AC were similar between Korean and Yunnan rice cultivars.

Table 3. Physicochemical properties of rice grain and palatability of cooked rice from Korea and Yunnan, China.

	Origin	PV[1)]	PC (%)	ADV (1~7)	AC (%)
Mean	Korea	76.4	5.95	6.64	18.71
	Yunnan	57.1	7.39	6.64	19.15
Range	Korea	70.7-80.8	5.11-6.76	6.47-6.77	16.69-20.19
	Yunnan	50.5-66.4	6.75-8.26	5.97-6.87	16.97-21.89
C.V. (%)	Korea	4.4	9.37	1.33	6.08
	Yunnan	8.5	6.64	4.00	6.46
T-test		**	**	ns	ns

[1)] PV: Palatability value of cooked rice, measured by TOYO Mito meter (MA-90); PC: Protein content; ADV: Alkali digestion value; AC: Amylose content. **: Significant at $\alpha = 0.01$ by t-test. ns: Not Significant at $\alpha = 0.05$ by t-test

Viscogram characters

Among viscogram parameters, PKV and BDV of Korean rice cultivars were significantly higher than those of Yunnan rice cultivars but SBV of Korean rice cultivars were significantly lower than that of Yunnan rice cultivars. HPV, CPV, and CSV of Korean rice cultivars and Yunnan rice cultivars were not significantly different but the averages of Korean rice cultivars were slightly higher than those of Yunnan rice cultivars (Table 4).

Table 4. Comparison of viscogram components between *japonica* rice cultivars from Korea and Yunnan, China.

	Origin	PT	PAT	PKV	HPV	BDV	CPV	CSV	SBV
Mean	Korea	6.36	68.1	234.9	177.3	57.6	253.4	76.0	18.5
	Yunnan	6.45	68.1	201.8	168.4	33.4	241.6	73.2	39.8
Range	Korea	6.07-6.58	68.0-68.2	210.6-276.9	143.8-226.1	44.5-76.6	232.2-293.9	63.3-94.1	9.3-29.6
	Yunnan	6.11-7.00	68.0-68.6	155.6-255.3	125.5-232.3	23.0-45.3	194.3-296.0	55.4-87.3	29.5-48.0
C.V. (%)	Korea	2.36	0.1	9.1	13.1	20.9	7.9	12.8	40.0
	Yunnan	4.67	0.2	13.6	17.6	22.3	10.8	12.8	12.1
T-test		ns	ns	**	ns	**	ns	ns	**

PT: Peak time (min); PAT: Pasting initial temperature (°C); PKV: Peak viscosity; HPV: Hot paste viscosity; BDV: Breakdown viscosity (= PKV – HPV); CPV: Cool paste viscosity, SBV: Setback viscosity (= CPV – PKV); CSV: Consistency viscosity (= CPV – HPV).
**: Significant at $\alpha = 0.01$ by t-test. ns: Not significant at $\alpha = 0.05$ by t-test.

Correlation analysis among quality traits

Correlation analysis showed that PV was negatively correlated with PC and SBV but positively correlated with PKV, BDV, and CPV (Table 5). PC was negatively correlated with PKV and BDV but positively correlated with SBV. These results are in accordance with the previous report (Shen et al. 2003) in which the relationship between PV and other quality traits was analyzed. The analysis results also showed that ADV and AC were negatively correlated with PKV, HPV, and CPV, respectively.

Table 5. Correlation coefficients among quality traits of rice.

Characters	PV	ADV	AC	PC	PKV	HPV	BDV	CPV	CSV
ADV	-0.240								
AC	-0.286	0.298							
PC	-0.907**	0.219	0.152						
PKV	0.719**	-0.496*	-0.623**	-0.672**					
HPV	0.395	-0.504*	-0.607**	-0.349	0.852**				
BDV	0.689**	-0.080	-0.143	-0.678**	0.440	-0.095			
CPV	0.454*	-0.524*	-0.649**	-0.429	0.915**	0.938**	0.131		
CSV	0.022	0.108	0.086	-0.091	-0.110	-0.473*	0.603**	-0.137	
SBV	-0.847**	0.182	0.245	0.780**	-0.635**	-0.239	-0.798**	-0.269	0.000

*, **: Significance at $P < 0.05$ and $P < 0.01$, respectively. Refer to Table 3 and Table 4 for abbreviations.

Cluster analysis using DNA markers

Cluster analysis of 20 cultivars was conducted based on their genotypes using 19 discriminative DNA markers (Figs. 1, 2) which were selected due to their association with quality characters in previous reports (Bao et al. 2006a, 2006b; Ohtsubo et al. 2002; Ohtsubo et al. 2003) and developed by the Crop Molecular Breeding Lab of Seoul National University in Korea. The genetic similarity coefficient (GS) among all cultivars varied from 0.57 to 1. All cultivars were divided into five groups with a GS value criterion (< 0.68). Group I included four Korean cultivars (Ilpum, Gopum, Samgwang, and Nampyeong) and ten Yunnan cultivars (Yunjing 17, 19, 21, and 22; Yunjingyou 10, 14, and 15; Chujing 27, Hexi 41, and Yunzikang 21), group II was composed of Odae and Koshihikari, group III was of Dianjingyou 1, group IV was of Dongjin, and group V was of Chucheong and Shindongjin. Korean rice cultivars were distributed over five groups, while all of Yunnan rice cultivars belonged to two groups, I and IV.

Fig. 1. PCR profile of tested cultivars amplified by STS primer A6 and M2CG.
M. 100 bp Ladder Molecular Weight DNA Marker, 1.Ilpum, 2.Gopum, 3.Samgwang, 4.Koshihikari, 5.Dongjin, 6.Chucheong, 7.Nampyeong, 8.Shindongjin, 9.Odae, 10.Dianjingyou 1, 11. Yunjing 17, 12.Yunjing 19, 13.Yunjing 21, 14.Yunjing 22, 15.Yunjingyou 10, 16.Yunjingyou 14, 17.Yunjingyou 15, 18.Chujing 27, 19.Hexi 41, 20.Yunzikang 21

Regression analysis with palatability value (PV) and other quality traits

Multiple linear regression analysis was performed to know the relationship between PV with nine quality traits (i.e. ADV, AC, PC, PKV, HPV, BDV, CPV, CSV, and SBV) and then the best model for estimating PV was selected. When all nine traits were included, the coefficient of determination (R^2) of the regression equation was 0.900 (Table 6). If five traits (PC, PKV, BDV, CPV, and SBV) which showed a significant correlation with PV were used, R^2 for estimating PV was 0.888. The R^2 value from the regression model in which PC was used as a single independent variable was 0.822, and the regression equation was $PV=139.024-(10.865 \times PC)$. This suggests that eating quality, measured by PV, can be estimated efficiently using only PC in this batch of Korean and Yunnan cultivars.

Table 6. Multiple linear regression analysis for estimating the palatability of cooked rice using physicochemical and viscogram parameters of rice.

Traits	All traits			Traits correlated with PV			Significant trait		
	Estimate	*t* value	R^2	Estimate	*t* value	R^2	Estimate	*t* value	R^2
PC	-7.907	-2.81**	0.441	-7.314	-3.75**	0.501	-10.865	-9.11**	0.822
SBV	-169.825	-0.79	0.058	-41.959	-0.90	0.055			
CPV	189.719	0.64	0.039	41.624	0.89	0.054			
PKV	-152.385	-0.54	0.028	-41.592	-0.89	0.054			
BDV	-17.142	-0.07	0.000	-0.047	-0.42	0.013			
AC	-1.371	-0.94	0.082						
HPV	-37.362	-0.41	0.016						
CSV	-20.283	-0.11	0.001						
ADV	-0.094	-0.01	0.000						
Intercept	166.623	2.02*		120.437	5.04**		139.024	17.16**	
Total R^2			0.900			0.888			0.822

* : Significance at P < 0.05. ** : Significance at P < 0.01. Refer to Table 3 and Table 4 for abbreviations.

Discussion

In general, it was known that Yunnan *japonica* rice cultivars had poor eating quality compared to Korean varieties. In order to compare the eating quality and quality-associated traits between two cultivar groups, we comparatively analyzed the eating quality and its relevant traits of Korean and Yunnan rep-

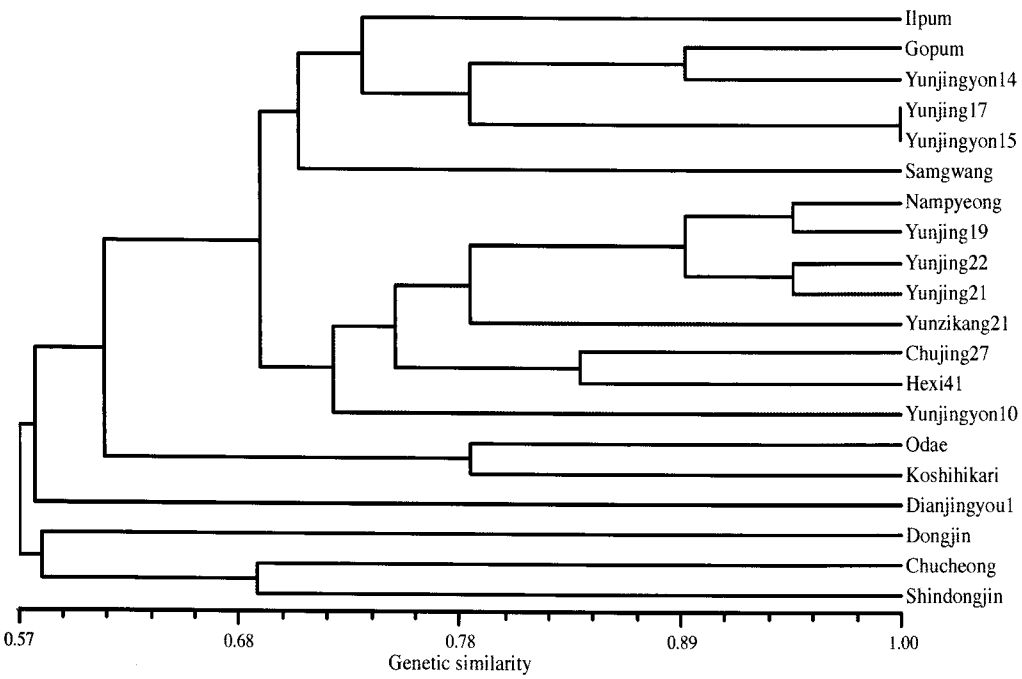

Fig. 2. Dendrogram for 20 cultivars according to UPGMA clustering based on genetic similarity determined by molecular markers.

resentative rice cultivars. Palatability value (PV), peak viscosity (PKV), and breakdown viscosity (BDV) of Korean rice cultivars were significantly higher than those of Yunnan rice cultivars, while protein content (PC) and setback viscosity (SBV) of Korean rice cultivars were significantly lower than those of Yunnan cultivars. Correlation analysis showed that PV was negatively correlated with PC and SBV but positively correlated with PKV and BDV. The best linear regression model for estimating PV using physicochemical and viscogram properties of rice grain revealed that PV was mainly influenced by PC but not directly influenced by viscogram characters such as PKV, BDV, and SBV although there were significant correlations between them (Table 5). This is probably due to the fact that viscogram parameters should be largely affected by protein content (Table 5) (Fitzgerald and Reinke 2006; Kim et al.1994). Thus, even though other factors might be involved in eating quality of rice, developing low PC variety should be the major target in improving the eating quality of Yunnan *japonica* rice cultivars.

The heavy application of nitrogen fertilizer causes increased PC in rice grains as well and then eventually decreases the eating quality and viscogram property (Shen et al. 2003). The amount of nitrogen fertilizer application at Kunming (140 kg/ha) was larger than at Suwon, Korea (110 kg/ha). Poor eating quality of Yunnan rice cultivars might be attributable to the heavy dose of nitrogen fertilizer which led increased PC. Another possible explanation for poor eating quality is that the average daily air temperature was relatively low during grain-filling stage at Kunming. That is, the average temperature of August and September at Suwon was 25.2 °C and 21.1 °C, respectively, while at Kunming it was 20.7 °C and 18.4 °C, respectively. In general, the optimum average daily air temperature during grain filling is from 21 to 26 °C (Cheng and Zhong 2001; Lu et al. 1998; Zhang and Wang 2006). As a consequence, low temperature during ripening at Kunming might cause a relatively poor eating quality through increasing the protein content and modifying other quality characters (Fitzgerald and Reinke 2006; Gomez et al. 1975; Jeong et al. 1997).

The genetic diversity of Yunnan *japonica* rice cultivars was obviously less than that of Korean rice cultivars because most of these Yunnan rice cultivars were derived from Hexi or Dianxi varieties widely grown in Yunnan (Fig. 2). It should be considered in breeding programs to broaden the genetic base of Yunnan cultivars through introducing germplasm from various origins. Using four Korean cultivars, belonging to Group I and genetically close to Yunnan cultivars, as crossing parents would be effective to improve the grain quality of Yunnan rice cultivars in a short time. However, it was unlikely that there might be a significant relationship between PV and genetic similarity coefficient as appeared in Fig. 2, although the markers used here had been selected as associated with PV or starch-related characters (Bao et al. 2006a, 2006b; Ohtsubo et al. 2002, 2003).

In conclusion, the eating quality of Yunnan *japonica* rice cultivars can be improved by lowering the protein content by integrated approaches through cultivar development and appropriate cultural technology.

Table 7. Primer sequences used for genotyping.

Primer	type	Forward (5'→3')	Reverse (5'→3')	Tm(°C)
A6	STS	CCAGCTGTACGCCTGTACTAC	CCAGCTGTACGTCTTCCCCAGC	55
A7	STS	TGCCTCGCACCAGAAATAG	TGCCTCGCACCATGAG	55
B1	STS	GTTTCGCTCCTACAGTAATTAAGGG	GTTTCGCTCCCATGCAATCT	55
B43	STS	GGCCGGCATGACTCAC	ACTGGCCGGCATCAAGAC	55
F6	STS	ACCACTCCATATATATCATCCAAAG	ACCACTCCATATCACCACAAGG	55
G4	STS	GAGACCGATATGCGATTC	GTGGTGTTTAGATCCAGAGACTTA	55
G22	STS	CTCACTCAAATTTACAGTGCATTTTCTTG	AGGGCCATGATACAAGACTCTGT	55
G28	STS	GGCGGTCGTTCTGCGAT	GGAGAATCCCACAGTAAGTTTTTCTTTG	55
J6	STS	GTCGGAGTGGTCAGACCG	GTCGGAGTGGATGGAGTAGC	55
M2CG	STS	ACAACGCCTCCGATGA	ACAACGCCTCCGACAACAAGAT	55
M11	STS	GTCCACTGTGACCACAACAT	GTCCACTGTGGGGATTGTTC	55
P5	STS	ACAACGGTCCGTCCTTGCTT	ACAACGGTCCAACAGATACTTTTGA	55
S13	STS	GTCGTTCCTGTGGTTAGGACAGGGT	GTCGTTCCTGCTGGTGTCTCAGAT	55
WK9	STS	CCCGCAGTTAGATGCACCATT	CCGCAGTTAGATCAAGTGGC	55
E30	STS	TACCTGGTTGATGTATACAGATCTGGTT	ATCCCTCGATCCCTCTAGCATTAT	55
SBE3	SNP	492:GTCTTGGACTCAGATGCTGGACTC	493:ATGTATAACTGGCAGTTCGAACGG	55
$F7A_{-F7}$	one	CTGGATCACTTCAAGCTGTACGAC		57
$F7A_{-F22}$	set	CAAGGAGAGCTGGAGGGGGC		68
$F7A_{R1}$	for		GCCGGCCGTGCAGATCTTAAC	57
$F7A_{R21}$	SNP		ACATGCCGCGCACCTGGAAA	64
TreB1	Indel	CACTCCAGTTCCTGCTCAAA	CACCTCCAAAACGAATATGG	55
S3c I	Indel	CCACTCTCATGTCCTTGAAC	GCCATGACATTTGGACAT	55

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