

## Detection of Changes in Taste of *japonica* and *indica* Brown and Milled Rice (*Oryza sativa* L.) during Storage Using Physicochemical Analyses and a Taste Sensing System

THI UYEN TRAN,<sup>†,‡</sup> KEITARO SUZUKI,<sup>‡</sup> HIROSHI OKADOME,<sup>‡</sup> HIDEKAZU IKEZAKI,<sup>§</sup>  
SEIICHI HOMMA,<sup>†</sup> AND KEN'ICHI OHTSUBO<sup>\*,†,‡</sup>

Graduate School of Humanities and Sciences, Ochanomizu University,  
2-1-1 Otsuka, Bunkyo, Tokyo 112-8610, Japan; National Food Research Institute,  
2-1-12 Kannondai, Tsukuba, Ibaraki 305-8642, Japan; and Intelligent Sensor Technology, Inc.,  
1800 Onna, Atsugi, Kanagawa 243-8555, Japan

Changes in the taste of *japonica*, hybrid, and *indica* brown and milled rice, stored for 10 months at low (5 °C, 65–70% relative humidity) and room temperatures were observed by physicochemical analyses and a novel method using a taste sensing system. During storage, some properties increased or decreased while others were fairly constant. The main taste components of cooked rice such as sweetness (sucrose) and umami tastes (glutamic acid and aspartic acid) were reduced during storage, whereas glucose and fructose increased. The increase of fat acidity and consequent decrease of the pH value of the cooking solution may contribute to the off-taste of cooked stored rice. A taste sensing system with 10 lipid membrane sensors was also used to classify new and old rice samples using principal component analysis. Fresh and room temperature stored *japonica* and *indica* rice could be clearly distinguished; however, it was not possible to differentiate the samples stored at low temperature.

**KEYWORDS:** Storage; *japonica*; *indica*; brown rice; milled rice; physicochemical properties; taste components; taste sensor

### INTRODUCTION

Rice (*Oryza sativa* L.) is a staple food for more than half of the world's population. A small amount of milled and brown rice is used as material for processing of foods, whereas a large amount of rice grain is used in cooked form. Storage is a usual process in the normal pathway of rice "from farm to table". During storage, a number of changes occur, which is termed aging. Aging is a natural and spontaneous phenomenon involving changes in the physical, chemical, and biological properties of rice that modify the cooking, processing, eating, and nutritional qualities and affect the commercial value of the grain on the market (1). These changes include cooking properties, texture properties, pasting properties, composition, flavor, sensory attributes, and enzyme activity (2).

Storage of milled rice results in increased water absorption during cooking in boiling water and larger volume of the cooked kernels (1). Rice stored for a long time is harder and less sticky than freshly harvested rice in cooked form, as measured by both sensory method (3, 4) and texturometer (5). The viscosity of

rice flour increases dramatically after storage for several months of milled rice as this change depends on storage temperature and duration (5–7). The components of cooked rice flavor change rapidly during storage. Carbonyl compounds, particularly hexanal, are suggested to be the major contributors to the off-flavor because they increase during storage (8). During storage, the appearance of stale flavor has been shown to correspond to higher levels of propanal, pentanal, and hexanal with accompanying decreases in the content of linoleic and linolenic acids. Lipids are hydrolyzed and oxidized to free fatty acids or peroxides, increasing the level of acidity and significantly deteriorating the taste and flavor (2). There is minimal change in gross chemical composition of the rice grain during storage; however, the amount of soluble amylose, which is soluble in boiling water, decreases (9). Some hydrolyses probably occur during the storage process of rice, which lead to a proportional increase in monosaccharides and a decrease in disaccharide (10). The protein content remains unchanged, although its general solubility is reduced with the decrease of albumin amount (1). In addition, the free amino nitrogen content of the outer layer of the rice grain was lost during storage, and this loss was related to Maillard-type nonenzymatic browning as suggested by the parallel losses in free amino nitrogen and whiteness of the rice (1). Furthermore, storage decreased the  $\alpha$ -amylase,  $\beta$ -amylase, and proteolytic activities of milled rice, but increased protease, lipase, and lipoxygenase activities in stored samples (1, 11).

\* Address correspondence to this author at the Cereal Science Laboratory, National Food Research Institute, Kannondai 2-1-12, Tsukuba Science City, Ibaraki 305-8642, Japan (telephone 81-29-838-8045; fax 81-29-838-7996; e-mail kenohtsu@nfri.affrc.go.jp).

<sup>†</sup> Ochanomizu University.

<sup>‡</sup> National Food Research Institute.

<sup>§</sup> Intelligent Sensor Technology, Inc.

**Table 1.** Samples Used for the Experiment and Their Characteristics<sup>a</sup>

no.	variety	abbreviation	type	location	brown rice			milled rice		
					mc (%)	ac (%)	protein (%)	mc (%)	ac (%)	protein (%)
1	Nipponbare	Nip	<i>japonica</i>	Ibaraki prefecture	16.07	15.72	8.15	13.49	17.52	7.43
2	Koshihikari	Koshi	<i>japonica</i>	Ibaraki prefecture	15.70	14.65	7.88	13.28	14.37	6.90
3	Hoshiyutaka	Hoshi	hybrid	Kinki Chugoku ARC	14.96	24.16	7.15	13.77	25.92	6.61
4	Yumetoiro	Yume	<i>indica</i>	Hokuriku ARC	15.70	27.90	6.80	13.91	30.81	6.25
5	Hamasari	Hama	<i>indica</i>	Saitama prefecture	15.30	24.32	8.52	14.06	27.71	7.92

<sup>a</sup> mc, moisture content; ac, amylose content; ARC, Agricultural Research Center; data show initial content of samples when received.

It is well-known that changes in rice qualities during storage can be evaluated by either instrumental or sensory methods. As to the changes of rice tastes, they can be investigated by chemical analyses or sensory evaluation by human beings. During long-term storage or because of high temperature and high humidity, the appearance of mold, fungi, or toxins may occur, which makes the stored rice unsafe for human beings. Thus, the taste sensor (electronic tongue) has been developed to analyze many kinds of foodstuffs and drinks, including changes in the quality of rice during storage (12, 13). Recently, we have shown that the tastes of brown and milled rice with different milling yields could be distinguished using the taste sensor (14).

There have been many attempts to explain the changes in functionalities that are associated with aging on the properties of rice components, such as starch, protein, and lipids and the interactions among them during storage (2). However, the changes of those components, which mainly affect the taste of cooked rice, remain poorly understood. The aims of this study are to determine and compare how the physicochemical properties, the main tastes, and the sweetness and umami taste components of *japonica* and *indica* brown and milled rice change during storage. In addition, we also showed that the aging process of these rices could be detected by a novel method using the taste sensing system.

## MATERIALS AND METHODS

**Rice Samples.** Five cultivated rice samples [*Oryza sativa* L. *japonica* cv. Nipponbare (Nip) and Koshihikari (Koshi), harvested in Ibaraki prefecture, Japan; *O. sativa* L. hybrid cv. Hoshiyutaka (Hoshi), harvested at the Kinki Chugoku Agricultural Research Center; *O. sativa* L. *indica* cv. Yumetoiro (Yume) and Hamasari (Hama), harvested at the Hokuriku Agricultural Research Center and in Saitama prefecture, Japan, respectively, in 2002] were used (Table 1).

**Storage Conditions.** Samples were received in the form of brown rice with initial moisture contents (mc) of 14.96–16.07%. Before being subjected to storage places, the samples were adjusted to mc of 14 ± 1% for 2 or 3 days. This process was carried out by placing the samples in the physical measurement laboratory room where the environmental conditions were controlled at 15–18 °C and 35–50% relative humidity (RH) by a Sanyo Controller System MBCR-2220F (Sanyo Electronic Ltd., Osaka, Japan). The mc of samples was determined every 6 h until the mc reached 14 ± 1%. Both fresh brown and milled rice (milled to 90% milling yield by a Yamamoto polisher, Rice Pal 31, Tendo, Japan) were stored, in paper bags sealed by tape, for up to 10 months from March 2003 to January 2004, at low temperature in an environmental chamber (LPH-350S BioTron LK System, Nihonika Ltd., Osaka, Japan), which was set at 5 °C and 65–70% RH, or at room temperature in a laboratory room without air-conditioning. After 2 months, the samples were collected, divided, and kept in a freezer (–25 °C) between two collection intervals for a maximum of 2 months and used for experiments.

**Preparation of Cooked Rice.** Twenty grams of whole grain (brown or milled and fresh or stored samples) was put into an aluminum cup, 32 mL of distilled water was added, and the rice was soaked for 1 h at

room temperature. The samples were cooked and kept warm in an electric rice cooker (RC183, Toshiba Co. Ltd.) and then left at room temperature for 2 h. After this procedure, the cooked rice samples were frozen (–20 °C) for one night and then lyophilized for 10 h by a freeze-dryer FD-1 (Tokyo Rikakikai Co., Ltd.). The lyophilized cooked rice grains were pulverized with a Cyclone sample mill (UDY Corp., Fort Collins, CO), and the flours were used for experiments as cooked stored rice samples.

**Textural Properties Measurement.** The textural properties of cooked rice were measured with a Tensipresser (My Boy System, Taketomo Electric Co., Tokyo, Japan) according to the method of Okadome et al. (15). The textural properties of hardness and stickiness in a surface layer and an overall layer of a single cooked grain were measured using the low-compression test (LCT) and high-compression test (HCT), respectively. The compression ratio of LCT was 25%, which caused a small deformation in the thickness of the grain, whereas HCT was 90%, at which the grain was compressed completely with great deformation.

**Cooking Properties.** Cooking properties of samples were measured using the method described in Batcher et al. (16). Eight grams of head grains was cooked with 160 mL of distilled water. After cooking, the expanded volume (EV), water uptake ratio (WUR), content of solid substances in cooking solution (SS), iodine blue value (IBV), and pH of the cooking solution were determined.

**Pasting Properties.** Pasting properties of raw flours were measured using an RVA (Rapid-Visco-Analyzer, Newport Science Ltd.), following the procedure of Toyoshima et al. (17). The sample (3.5 g, dry basis) was made equivalent to 14% moisture and mixed with distilled water in an RVA aluminum canister (supplied by Newport Scientific Ltd.). A programmed heating and cooling cycle was as follows: the sample was installed on the rotor of the RVA and heated from 50 to 93 °C (at 50 °C for 1 min and 4 min to reach 93 °C). It was held at 93 °C for 7 min, cooled from 93 to 50 °C over 4 min, and allowed to stand at 50 °C for 3 min. Parameters recorded were maximum (peak), minimum, and final viscosities, setback, breakdown, pasting temperature, and peak time.

**Chemical Component Analyses.** The mc of the flour was measured after 1 h of treatment at 135 °C by an oven-drying method. The amylose content of the sample was measured as described by Juliano (18). The nitrogen content of the sample was measured using a LECO System (LECO FP-528, LECO Corp., St. Joseph, MI). Supplied EDTA from LECO Corp. was used as standard. The protein content of sample was obtained from nitrogen by multiplying it with a nitrogen–protein conversion factor of 5.95. The fat acidity of raw powder was determined according to the spectrophotometric method of Ohtsubo (19), a modification of the Duncombe method (20). The reducing sugar content of cooked stored rice powder was measured according to the Somogyi–Nelson method (21–23). The total carbohydrate content of cooked stored rice powder was measured by using the anthrone method (24). Measurement of free sugar content was performed as follows: 4.0 g of cooked stored rice powder was extracted with 15 mL of 50% ethanol and then centrifuged at 5000 rpm using rotor no. 3 (R10A2) for 20 min at 20 °C with the Hitachi high-speed refrigerated centrifuge, Himac CR 21F (Hitachi Koki Ltd., Tokyo, Japan). The process was repeated twice, and the obtained supernatant was reduced to the volume of 1 mL with an EYELA Centrifugal Evaporator CVE-2000 (Tokyo Rikakikai Co., Ltd., Tokyo, Japan) and then filtered through a 0.45

$\mu\text{m}$  filter (GL chromato-disk 25A, GL Science Co., Ltd., Tokyo, Japan). Twenty microliters of filtrate was injected, and free sugars were analyzed by high-performance liquid chromatography (HPLC). The conditions of HPLC were as follows: controller system (CLS-10AP), refractive index detector (RID-10A), Shimadzu Ltd., Kyoto, Japan; column, Shodex Asahipak NH2P-50 4E (Showa Denko Co., Ltd., Tokyo, Japan); column temperature, 30 °C; solvent, acetonitrile/water (75%:25%); flow rate, 1 mL/min. Measurement of free amino acids content was performed as follows: 2.0 g of cooked stored rice powder was extracted with 5 mL of sulfosalicylic acid (2%) and then centrifuged at 5000 rpm using rotor no. 3 (R10A2) for 15 min at 20 °C with the Hitachi high-speed refrigerated centrifuge, Himac CR 21F (Hitachi Koki Ltd.). The supernatant was filtered through a 0.45  $\mu\text{m}$  filter (GL chromato-disk 25A, GL Science Co., Ltd.). Prepared sample solution (10  $\mu\text{L}$ ) was analyzed by a Hitachi 8500 Amino Acid Analyzer.

**Measurements of Enzyme Activities.**  $\alpha$ -Amylase and  $\beta$ -amylase activities were determined using the kits of Megazyme International Ireland, Ltd.  $\alpha$ -Glucosidase activity was measured according to the method of Imai et al. (25) and Iwata et al. (26). Protease activity was measured by using the method of Palmiano and Juliano (27) with modification. The rice flour (0.5 g) was extracted in sodium phosphate buffer (pH 7.5) containing 0.005 M cysteine for 10 min at room temperature and then centrifuged at 5000 rpm using rotor no. 3 (R10A2) for 15 min at 0 °C with the Hitachi high-speed refrigerated centrifuge, Himac CR 21F. The supernatant was used for assay of protease activity. One milliliter of enzyme solution was incubated with 1.0 mL of 1% bovine albumin (Wako Pure Chemical Co., Kyoto, Japan), which had been dissolved in water, and 1.0 mL of 0.2 M sodium phosphate buffer (pH 6.5) at 40 °C for 90 min. The reaction was stopped with 1.0 mL of 20% TCA, and the precipitate was aged for 1 h on ice. The mixture was centrifuged at 8000 rpm using rotor no. 3 (R10A2) for 15 min at 4 °C. The supernatant was kept at room temperature for 10 min, and the absorbance was read at 280 nm. One unit of protease activity was expressed as the amount of enzyme that produced a 0.1 increase in light absorbance of the solution under the conditions of assay.

**Response of the Taste Sensor to Sample.** Cooked stored rice flours (6 g, dry basis) were mixed and homogenized with 150 mL of deionized water and then centrifuged at 5000 rpm using rotor no. 3 (R10A2) for 20 min at 20 °C with the Hitachi high-speed refrigerated centrifuge, Himac CR 21F. The supernatant was used for measurement as a rice sample solution. The measurements were conducted with a commercial taste sensing system, SA 402 (Anritsu Co., Atsugi, Japan) as described previously (14). The detecting sensor part consisted of electrodes made of lipid membranes, which were purchased from Intelligent Sensor Technology, Inc., Atsugi, Japan. A total of 10 sensors, which were made of lipid membranes, were used. On the basis of the lipid content, the membranes were grouped into hybrid (S1 and S2), negative (S3–S6), and positive membranes (S7–S10). Each lipid membrane was fitted on the part of a plastic tube and called a probe, which had a hole so that the inner part of the cylinder was isolated from the outsider. The end of the cylinder was sealed with a stopper that held an Ag/AgCl electrode. The tubes were filled with saturated AgCl 3.3 M KCl solution. The detecting and reference electrodes were immersed in 35 mL of rice sample solution by a robot arm. Eight detecting electrodes and two reference electrodes were separated into two groups and connected to a computer recording data for differences in potentials between the detecting and the reference electrodes. The principle of the taste sensor was the measurement that was made at equilibrium using a voltmeter with currents that approached zero. When the sample and reference solutions were not the same, a difference in potential was observed. As the electric potential of the reference electrode was constant, changes in the cell potential were due to the detecting electrode. For the membrane potentiometric method, the observed potential was dominated by the Donnan potential that developed across the membrane (28). Measurement for each sample was carried out after all electric potentials of membranes were stabilized in a standard solution, made of 3 mM potassium chloride and 3 mM potassium tartrate (29), for at least 1 week. The response electric potentials were measured relative to zero response potential to fresh Nipponbare cooked brown and milled flours, which were stored in a freezer (–25 °C), and used as a standard samples for brown and milled rice measurements, respectively. Because the

stopper could hold four probes and one reference electrode, the measurement was conducted in two steps: first, the negative and positive membrane probes and, second, the hybrid membrane probes were used. The average values of three measurements were adopted as the response electric potential patterns, and the coefficient of variances was <20%.

**Statistical Analysis.** Means were separated by the least significant difference (LSD) test at  $p \leq 0.05$ . Fresh and stored samples were characterized according to their physicochemical properties and their responses to taste sensor using principal component analysis (PCA). LSD test and PCA were conducted using the data analysis ToolPak of Microsoft Excel, 2000.

## RESULTS AND DISCUSSION

### Changes in Physicochemical Properties during Storage.

Changes in physicochemical properties of samples during storage are summarized in **Table 2**. Textural property is one of the most important factors, which affects the palatability of cooked rice. In Japan, Korea, and North China, sticky rice is preferred, whereas hard rice is consumed mostly in India, South Asia, and part of America. Room temperature storage changed markedly the textural property of cooked *japonica* milled rice grain. Both LCT and HCT showed that the hardness of the surface and overall grain was increased, whereas the stickiness decreased significantly ( $p \leq 0.05$ , Figures 1 and 2 in the Supporting Information). In addition, the balance degree between stickiness and hardness was also decreased (Figure 3 in the Supporting Information). However, the changes in the textural property of cooked stored *indica* rice grains were not clearly observed, as their stickiness was lower but their hardness higher than the respective ones of *japonica*.

Storage of milled rice at room temperature increased water uptake ratio (WUR), iodine blue value (IBV), and expanded volume (EV) but decreased the pH of the cooking solution significantly ( $p \leq 0.05$ , Table 1 in the Supporting Information). The ratio of IBV to SS of most samples significantly increased ( $p \leq 0.05$ ). Thus, after storage at room temperature changes in cooking properties should be taken into account and processing conditions correspondingly adjusted. Low-temperature storage did not cause remarkable changes in cooking property, except that the pH value was slightly reduced. Among the samples, *japonica* rice, such as Koshi and Nip, tended to have lower WUR, EV, and IBV than *indica* rice such as Yume and Hama, as the amylose content of *japonica* rice is lower than that of *indica* (30).

The pasting properties of stored rice were clearly changed during storage for the first 10 months. The maximum, minimum, and final viscosities and breakdown values of all samples increased noticeably with the increase of storage time. Storage at low temperature did not lead to as significant changes as at room temperature (Figures 4 and 5 in the Supporting Information). These results are consistent with those reported by Shibuya et al. (5), Yasumatsu et al. (7), and Perez and Juliano (31). However, Sowbhagya and Bhattacharya (6) and Indudhara et al. (9) reported that after an initial increase in maximum viscosity and a setback during the first 6 months of storage, a steady decrease was noted during the subsequent 3 years of storage of rough and milled rice.

The amylose content of *indica* and hybrid samples was higher than that of *japonica* samples, whereas the protein content varied depending on cultivar. Milled rice had a higher amylose content than brown rice, whereas brown rice had a higher protein content than milled rice (**Table 1**). During storage, the amylose and protein contents were fairly constant (Table 2 in the Supporting Information).

**Table 2.** Summary of the Changes in Physicochemical Properties of Rice during Storage<sup>a</sup>

parameter	brown rice				milled rice			
	room temperature		low temperature		room temperature		low temperature	
	<i>japonica</i>	<i>indica</i>	<i>japonica</i>	<i>indica</i>	<i>japonica</i>	<i>indica</i>	<i>japonica</i>	<i>indica</i>
textural properties								
hardness ( $H_2$ )	+	±	±	±	+	±	±	±
stickiness ( $-H_2$ )	-	-	±	±	-	±	±	+
balance degree ( $-H_2/H_2$ )	-	-	±	±	-	±	±	+
cooking properties								
WUR	+	++	±	±	+	++	±	±
EV	+	++	±	±	+	++	±	±
pH of cooking solution	--	--	-	-	--	--	-	-
IBV/SS	+	+	±	±	+	+	±	±
pasting properties								
max viscosity	++	++	+	+	++	++	+	+
min viscosity	++	++	+	+	++	++	+	+
breakdown	++	++	+	+	++	++	+	+
final viscosity	++	++	+	+	++	++	+	+
setback	++	++	+	+	++	++	+	+
pasting temperature	±	±	±	±	±	±	±	±
time to peak	-	-	-	-	-	-	±	±
chemical components								
amylose	±	±	±	±	±	±	±	±
protein	±	±	±	±	±	±	±	±
total carbohydrate	-	-	±	±	-	-	±	±
reducing sugar	++	++	±	+	++	++	+	+
fat acidity	--	--	-	--	--	--	-	--
enzyme activities								
$\alpha$ -amylase	-	-	±	±	-	-	±	±
$\beta$ -amylase	--	--	±	-	--	--	±	-
$\alpha$ -glucosidase	±	-	-	-	±	-	±	-
protease	±	-	±	-	-	-	±	±

<sup>a</sup> WUR, water uptake ratio; EV, expanded volume; IBV, iodine blue value; SS, solid substances; ++, increased significantly; +, increased; ±, change not clear or fluctuated; --, decreased significantly; -, decreased.

When stored, the lipolysis of lipids occurred and free fatty acids were released, which led to deterioration and off-flavor of rice (2, 7). Fat acidity is one of the best indicators, which is usually used to determine the aging of rice (19). Fat acidity of both *japonica* and *indica* increased significantly ( $p \leq 0.05$ , Table 2 in the Supporting Information) during storage at room temperature. Fat acidity of fresh brown rice was higher than that of fresh milled rice (Table 2 in the Supporting Information). However, after 10 months of storage at room temperature, the fat acidity of milled rice was nearly equal to or higher than that of stored brown rice (Table 2A,C in the Supporting Information). This phenomenon was also observed by Shibuya et al. (5). It is well-known that brown rice has a higher content of lipids than milled rice (32), and the changes of lipid content are greater in the outer 10% layer (by weight) where nonstarch constituents are concentrated (1). The changes observed during storage suggest that at least two processes affect the lipid content; one involves hydrolysis of lipids to produce free fatty acids, and the other is the oxidation of lipids to produce hydroperoxides. It has been shown that in these processes nonstarch lipids (free lipids) are primarily involved (2). Storage conditions of temperature and air are important to the reaction rate for both hydrolysis and oxidation (33, 34). Lipids of milled rice were lipolyzed by hydrolysis and oxidation more quickly and strongly than those of brown rice after 10 months of room temperature storage, probably because brown rice grain is covered by a pericarp called a caryopsis coat (fruit coat), which may protect the brown rice from environmental and physical effects and subsequently inhibit the quick lipolysis caused by lipases, which are a major cause of deterioration of oil in bran, and by lipoxygenase, which is localized in the bran (35). In addition, the ease of disruption of aleurone cells of surface layer of milled

grain, and the accompanying fusion of lipid bodies, may be responsible for faster rancidity in milled grains than brown ones. The increase of fat acidity of *japonica* rice stored at low temperature was at lower level than when stored at room temperature. All *indica* rice samples had significant increase in fat acidity content during storage at low temperature as well as at room temperature. The reason for this phenomenon is probably that *indica* rice samples have a less stable seed coat, stronger lipase and lipoxygenase activities, and higher initial fatty acid and nonstarch lipid contents than *japonica* rice. Brown rice quality changed to a lesser degree than that of milled rice during storage, which may explain why the brown form is often stored in Japan. The rice hull represents ~20% of the rough rice grain. Thus, it is reasonable to store rice in brown form rather than in either milled or paddy forms, because storage in paddy form would occupy a larger space, which is a luxury for a small country such as Japan that has a high population density. In accordance with the increases of fat acidity, there was a consistent decrease in the pH of cooking solutions in all samples with increase of storage time as described above (Table 1, Supporting Information). These changes probably caused off-taste of cooked rice during storage as the increase of sourness, indicated by lower pH value, was observed.

The starch hydrolysis, degradation, or decomposition of stored rice probably occurred, which caused an increase in reducing sugars. Glucose and fructose are the major components of the reducing sugars, whereas sucrose is one of the nonreducing sugars, the concentration of which is several times higher in the outer layer than in the inner layer of the rice grain (1). Fresh milled rice had a higher content of reducing sugar than fresh brown rice. The reducing sugar content of cooked stored rice increased significantly ( $p \leq 0.05$ ) during storage at room

**Table 3.** Changes in Free Sugar Contents of Cooked Stored Rice during Storage<sup>a</sup>

cultivar	storage period (months)	fructose	glucose	sucrose	maltose	raffinose	maltotriose	sum
<b>A. Brown Rice at Room Temperature</b>								
Nip	0	96.7a	46.3a	823.8a	20.1a	64.0a	11.0a	1061.8a
	2	135.8b	40.0a	745.2b	17.5a	54.4b	—	992.8a
	4	164.9b	45.5a	721.0b	26.7a	67.6a	1.8b	1027.4a
	6	148.9b	83.3b	487.8c	19.9a	38.3c	1.1b	779.3b
	8	225.3c	168.4c	531.5d	25.9ab	45.8b	4.2c	1001.1a
	10	245.1c	212.9d	591.2e	32.1b	51.3b	5.6c	1138.2c
Koshi	0	98.1 a	50.7a	1089.2a	8.8a	70.9a	2.1a	1319.7a
	2	114.3a	58.8a	1008.5a	14.3a	65.0a	—	1260.9a
	4	138.8b	50.8a	963.4ab	17.9a	60.7a	—	1231.6a
	6	166.0c	79.7b	685.3c	26.2b	52.2b	—	1009.4b
	8	127.2ab	96.7b	767.1b	21.5b	58.7a	3.2a	1074.3b
	10	225.6d	81.1b	829.9b	25.2b	58.1a	5.2b	1225.0a
Hoshi	0	110.7a	47.0a	774.8a	15.6a	99.5a	7.0a	1054.5a
	2	127.5a	74.3b	731.5b	13.5a	86.4b	—	1033.2a
	4	142.0a	62.1c	628.0c	21.1ab	74.6c	—	927.8b
	6	111.1a	62.8c	371.3d	26.2b	35.1d	—	606.5c
	8	171.1ab	93.5d	436.2e	15.4a	35.4d	3.2b	754.7d
	10	181.6b	94.9d	448.2e	23.2b	25.6d	3.2b	776.7d
Yume	0	101.7a	48.1a	602.4a	—	106.6a	—	858.9a
	2	130.1a	47.8a	552.8b	—	101.0a	—	831.7a
	4	129.7a	59.4a	393.8c	21.3a	81.0b	4.9	690.0b
	6	210.6b	66.7b	348.5d	8.2b	69.7b	—	703.6b
	8	202.9b	64.7b	354.2d	19.6a	68.8b	—	710.2b
	10	194.5b	67.5b	426.5e	8.4b	77.7b	—	774.6c
Hama	0	116.2a	74.6a	830.0a	19.4a	91.4a	1.4a	1132.9a
	2	148.2a	80.8a	830.6a	10.0a	76.5b	1.5b	1147.7a
	4	276.5b	82.2a	756.5b	26.5b	77.1b	—	1218.8b
	6	316.7b	99.3a	427.1c	23.8ab	51.3c	—	918.1c
	8	209.1c	109.8b	450.5c	24.7ab	51.0c	—	845.0d
	10	170.53	107.5b	473.1c	30.7b	45.9c	—	827.7d
<b>B. Brown Rice at Low Temperature</b>								
Nip	0	96.7a	46.3a	823.8a	20.1a	64.0a	11.0a	1061.8a
	2	49.6ab	35.4a	764.7b	7.9 a	38.0b	—	895.6a
	4	156.5b	32.3a	746.1b	12.1ab	60.6a	3.5b	1011.1a
	6	141.1b	38.9ab	769.7b	10.5a	53.9b	2.2c	1016.3a
	8	130.2bc	84.1b	795.8ab	16.7a	58.5ab	2.9c	1088.3a
	10	156.9b	165.4c	870.0a	22.5ab	59.2ab	2.3c	1276.2b
Koshi	0	98.1a	50.7a	1089.2a	8.8a	70.9a	2.1a	1319.7a
	2	41.3a	38.7a	1023.1a	—	48.4b	—	1151.5a
	4	141.0ab	47.8a	932.0ab	12.0a	65.1a	0.6b	1198.4a
	6	202.9b	45.7a	927.1ab	21.7b	63.8ab	—	1261.2a
	8	200.3b	61.4b	1044.1a	8.5a	64.1a	2.3a	1380.6ab
	10	160.8ab	52.9a	1060.3a	2.0c	56.2b	1.7a	1333.7a
Hoshi	0	110.7a	47.0a	774.8a	15.6a	99.5a	7.0a	1054.5a
	2	94.1a	25.9b	716.9b	9.1ab	71.1b	—	917.0a
	4	184.0b	42.3ab	597.8c	22.8a	73.4b	—	920.3a
	6	163.9b	38.7bc	695.6bc	12.8a	79.7ab	—	990.7a
	8	230.0b	52.8ab	763.7a	2.2b	76.9b	2.4b	1127.9b
	10	207.8b	55.9c	792.2a	3.7b	79.5ab	2.6b	1142.4b
Yume	0	101.7a	48.1a	602.0a	—	106.6a	—	858.9a
	2	41.4a	23.4b	523.2b	—	92.3b	—	680.9b
	4	212.5b	32.3b	440.3c	21.4a	94.8b	—	801.6a
	6	204.3b	36.5c	494.8bc	21.6a	100.9ab	—	858.1a
	8	170.5ab	36.8c	566.2b	—	99.5ab	—	872.9a
	10	288.8bc	34.9c	605.6a	6.7b	110.2a	—	1046.2c
Hama	0	116.2a	74.6a	830.0a	19.4a	91.4a	1.4a	1132.9a
	2	103.4a	50.4b	777.0b	—	56.1b	—	986.9b
	4	261.8b	65.2a	774.5b	28.2b	84.9a	—	1214.6b
	6	236.2b	56.7ab	734.1c	39.0c	72.9c	—	1138.9a
	8	289.2c	70.9a	837.2a	9.0a	69.9c	3.2a	1279.5b
	10	353.6d	86.6c	915.2d	2.8d	72.7c	3.3a	1434.2d
<b>C. Milled Rice at Room Temperature</b>								
Nip	0	78.5a	113.9a	174.0a	32.2a	24.8a	16.4a	439.8a
	2	44.8b	149.5b	148.5b	16.2ab	7.8b	6.1b	372.9b
	4	32.9b	173.5bc	150.1b	22.2a	10.9b	11.8ab	401.4ab
	6	34.8b	135.0b	57.1c	11.9b	4.6c	6.4b	249.8bc
	8	101.7c	228.0d	60.5c	20.0ab	13.6a	7.1b	431.0a
	10	128.7d	285.9e	6.0bc	44.1bc	7.2b	6.3b	532.1cd

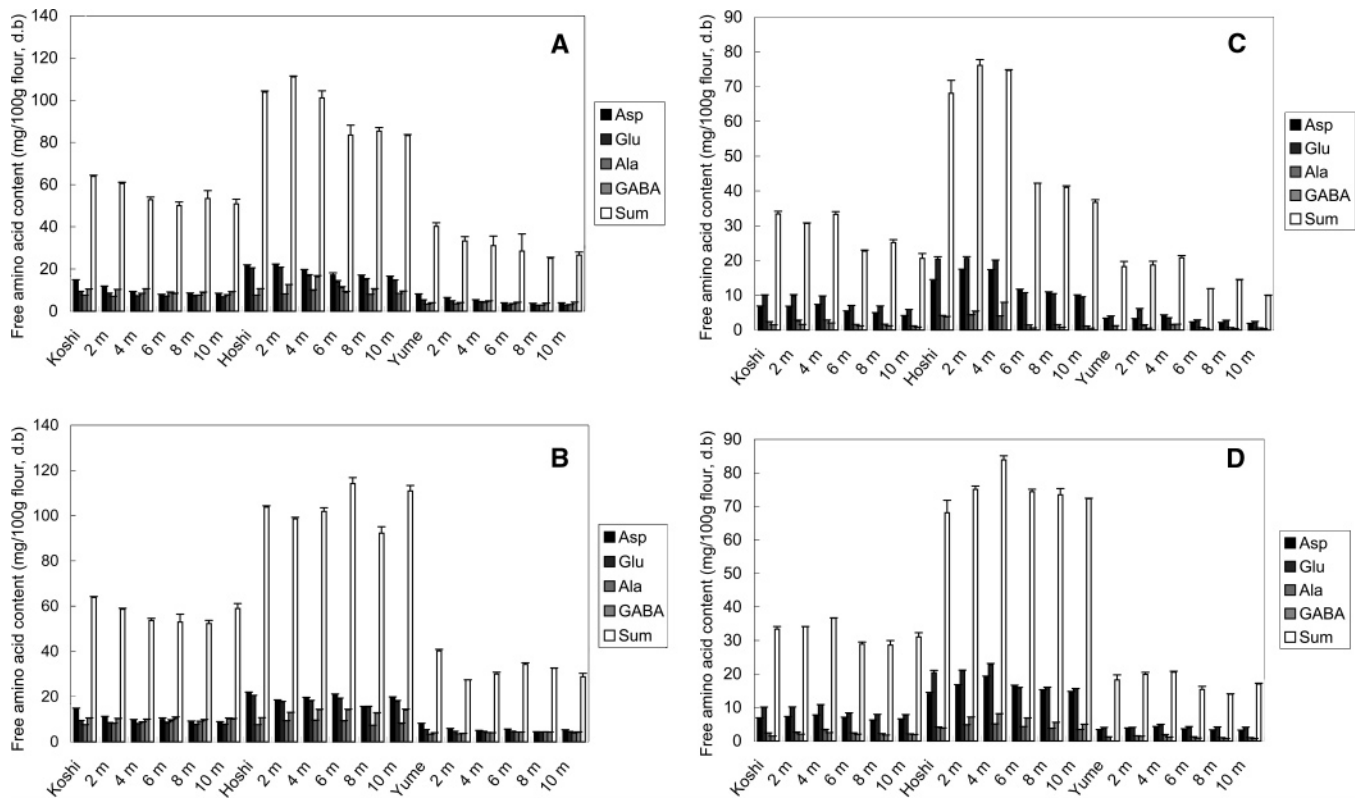
Table 3 (Continued)

cultivar	storage period (months)	fructose	glucose	sucrose	maltose	raffinose	maltotriose	sum
<b>C. Milled Rice at Room Temperature (Continued)</b>								
Koshi	0	91.0a	111.4a	281.6a	18.4a	23.6a	8.8a	534.9a
	2	50.9a	144.0b	264.5b	16.4a	19.0a	7.6a	502.3a
	4	83.9a	176.6b	257.4b	16.8a	18.0a	7.0a	559.7ab
	6	172.0b	143.1c	107.2c	10.4b	8.5b	2.6bc	443.9b
	8	134.7ab	285.1d	137.7d	21.8c	9.3b	1.6c	590.2c
	10	180.4b	314.7e	117.5cb	25.2c	2.3c	3.8ab	643.9d
Hoshi	0	106.8a	100.9a	408.7a	37.4a	79.1a	16.4a	749.4a
	2	75.5a	109.8a	340.0b	25.9b	55.0b	8.7b	614.9b
	4	213.8b	107.1b	315.9c	27.4b	56.3b	9.6b	730.1a
	6	214.1b	138.6b	53.4d	29.9b	7.8c	3.7c	447.4c
	8	328.1c	187.7c	71.7e	20.5b	6.9c	1.6c	616.5b
	10	314.4c	209.8d	52.6d	32.9ab	5.6c	3.5c	618.7b
Yume	0	94.8a	105.2a	194.2a	15.3a	32.6a	—	442.1a
	2	71.7b	134.0b	169.7b	13.4a	32.7a	4.6a	426.1a
	4	177.1c	128.1ab	139.3c	20.3b	32.7a	2.6b	500.0b
	6	229.9d	127.5ab	47.4d	13.0a	12.9b	0.7c	431.5a
	8	146.4e	134.2b	57.7e	13.1a	12.9b	1.8bc	366.2c
	10	174.8c	164.0c	56.7e	16.5a	12.2b	1.3bc	425.5a
Hama	0	118.6a	117.2a	406.7a	29.0a	51.6a	12.8a	735.8a
	2	66.3b	115.8a	327.9b	28.6a	45.2a	10.1a	593.9b
	4	190.7c	120.7a	272.0c	21.6b	27.2b	6.2b	638.3b
	6	262.0d	126.2a	111.0d	17.8b	5.9c	—	522.9c
	8	176.7c	134.1a	142.1e	14.6c	9.9c	—	477.4c
	10	315.3e	156.4b	149.1e	20.1b	8.2c	1.3c	650.4b
<b>D. Milled Rice at Low Temperature</b>								
Nip	0	78.5a	113.9a	174.0a	32.2a	24.8a	16.4a	439.8a
	2	68.1a	98.8b	144.1b	14.8b	8.3b	7.9b	342.1b
	4	57.1bc	121.8bc	101.8bc	17.9ab	10.5b	10.7a	319.8b
	6	38.3b	149.5bc	134.0b	17.3b	13.5b	8.6b	361.1b
	8	57.6bc	152.5cd	160.4ab	17.0b	9.8b	6.1b	403.4ab
	10	67.3ab	149.8bc	173.1a	33.0a	10.2b	7.5b	441.1a
Koshi	0	91.0a	111.4a	281.6a	18.4a	23.6a	8.9a	534.9a
	2	65.9b	95.8a	260.4b	16.8a	15.6b	5.7ab	460.1a
	4	51.3bc	121.8a	264.6b	18.9a	18.8bc	3.0b	478.4a
	6	72.1ab	154.5b	241.6c	14.7a	15.4b	4.5b	502.9a
	8	56.1b	152.5ab	290.0a	15.7a	16.7b	2.6b	608.6ab
	10	65.8b	149.8b	300.2a	27.2b	18.1b	6.0ab	567.2ab
Hoshi	0	106.8a	100.9a	408.7a	37.4a	79.1a	16.4a	749.4a
	2	94.0a	94.5a	358.0b	29.4b	59.8b	8.1b	643.8b
	4	137.4ab	106.4a	347.24b	22.9bc	43.4c	—	657.4ab
	6	147.5b	78.3b	335.6c	24.8b	53.3bc	2.7c	672.2ab
	8	107.9a	92.3a	383.4ab	27.0b	59.7b	5.3c	675.5ab
	10	102.9a	110.8a	407.8a	32.3ab	63.9b	8.4b	726.1ab
Yume	0	94.8a	105.2a	194.2a	15.3a	32.6a	—	442.1a
	2	93.7a	93.8b	175.6b	16.4a	30.4a	1.3a	411.1a
	4	132.1b	106.9a	201.0a	11.0b	23.4c	1.0b	475.3ab
	6	97.2a	75.7c	160.2b	16.0a	33.8a	1.9a	384.7bc
	8	111.5ab	71.5d	182.1b	14.7ab	35.9a	2.5a	418.2a
	10	128.9b	110.8e	206.1a	24.6c	42.6ab	3.6a	516.6b
Hama	0	118.6a	117.2a	406.7a	29.0a	51.6a	12.8a	735.8a
	2	82.4b	103.5b	309.9b	22.5a	35.3b	4.7b	558.2b
	4	74.0b	76.7c	300.5b	27.8a	34.3b	4.2b	517.6b
	6	66.8b	75.5c	303.9b	23.7a	34.4b	3.9b	508.2c
	8	113.1ab	77.5c	361.1c	21.0a	31.7b	3.8b	608.3ab
	10	102.8ab	82.3c	358.7c	24.4a	33.5b	2.3b	604.1ab

<sup>a</sup> Mean value (mg/100 g of rice flour, dry basis). Sections A, B, C, and D show the changes in free sugar contents of cooked stored brown and milled rice during storage at room and low temperature, respectively. Letters (a–e) indicate which value of each variety in each column differed significantly ( $p \leq 0.05$ ) with variation of storage duration. (–) Not detected.

temperature (Table 2, Supporting Information). This result is consistent with the increase of free glucose and fructose contents (Table 3A,C). However, at cold storage, the degree of change was small and there was nearly no significant difference ( $p \leq 0.05$ ), except for cv. Hama, for which the reducing sugar content of both brown and milled forms was increased significantly. Reducing sugar is also an important parameter that may be used

to determine the aging of rice. In addition, changes in total carbohydrate content of cooked stored samples were investigated. The total carbohydrate content of both *japonica* and *indica* brown rice was higher than that of milled ones. This content varied depending on variety. Among the samples, Koshi brown had the highest total carbohydrate content, whereas Yume has the lowest. During storage at room temperature, the



**Figure 1.** Changes in free amino acid contents of cooked stored rice during storage: (A) brown rice at room temperature; (B) brown rice at low temperature; (C) milled rice at room temperature; (D) milled rice at low temperature.

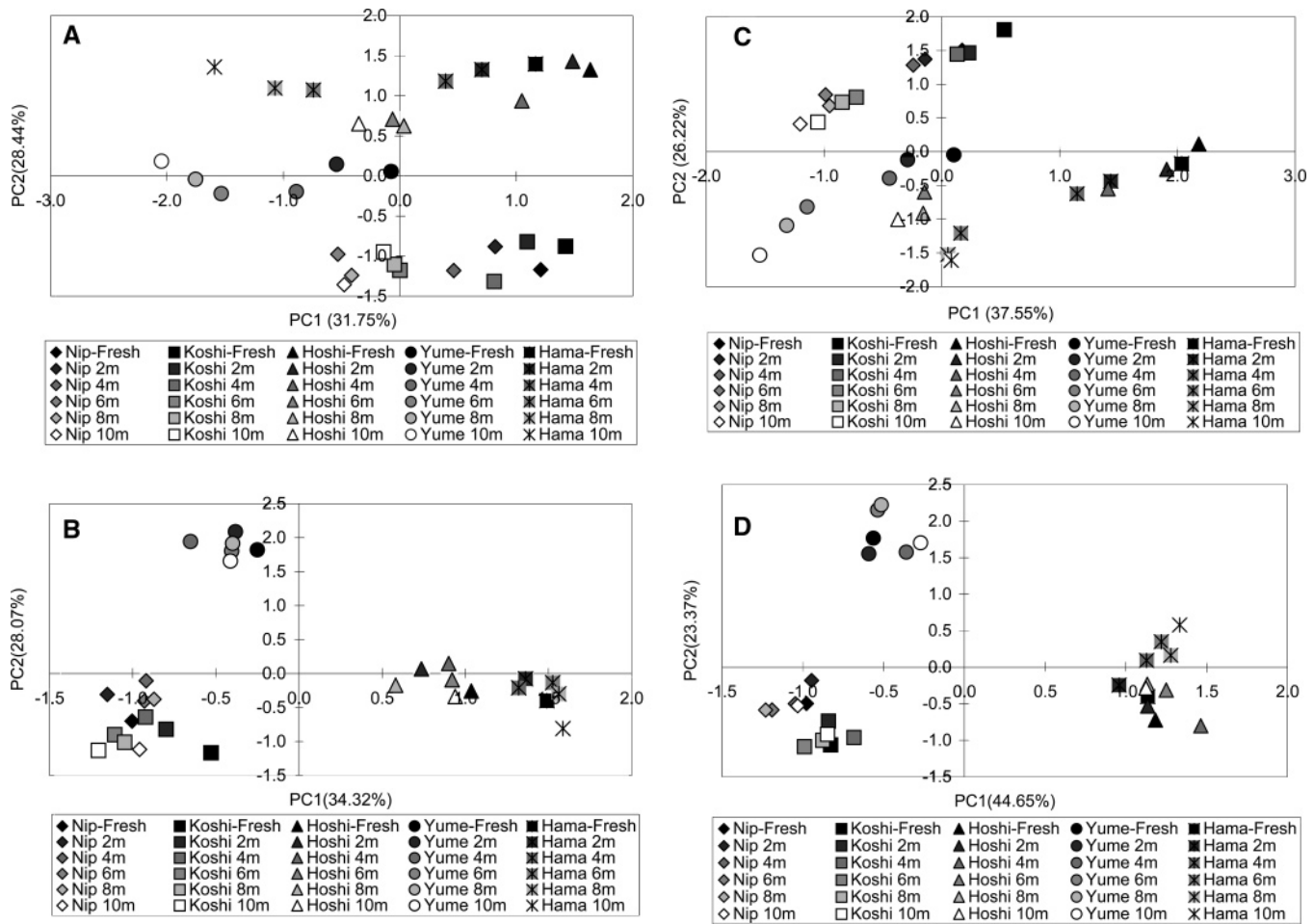
total carbohydrate of either brown or milled rice significantly reduced ( $p \leq 0.05$ ), whereas at low temperature it slightly increased (Table 2, Supporting Information).

Starch is degraded by the combined actions of enzymes such as  $\alpha$ -amylase,  $\beta$ -amylase, and  $\alpha$ -glucosidase, during cooking and processing of rice (36). The  $\alpha$ -amylase and  $\beta$ -amylase distribution in rice endosperm is mainly in the outer 20% of the endosperm. These enzymes are thought to be responsible for the tastes of cooked rice, and their activities were reduced during storage (1). In this study, we observed that  $\alpha$ -amylase,  $\beta$ -amylase, and protease activities of both *japonica* and *indica*, either brown or milled grains, reduced significantly during storage at room temperature (Table 3, Supporting Information). The degree of reduction was stronger in *indica* than in *japonica*, as well as in milled form than in brown form. The activity of  $\alpha$ -glucosidase increased after 6 months and then slightly decreased from the 8th to the 10th month. *japonica* rice had a higher  $\alpha$ -glucosidase activity than *indica* and hybrid samples. On the other hand, its  $\alpha$ -amylase,  $\beta$ -amylase, and protease activities were lower than those of *indica* and hybrid rice. However, after 6 months of storage at room temperature, these enzyme activities of *indica* and hybrid samples reduced quickly and reached the level of *japonica* types. Among *indica* samples, the enzyme activities of cv. Yume, both brown and milled forms, were lowest compared to those of cv. Hama. The same as for chemical components, enzyme activities did not change so much when samples were stored at low temperature. The decrease of enzyme activities during storage at room temperature probably led to reductions of free sugar and free amino acid contents and then, in part, affected the taste of cooked rice. The physicochemical properties diversified depending on cultivar, and their changes in those samples depended on storage conditions such as room or cold temperature.

**Changes in Taste Components.** Sweetness and umami taste are the main attributes that affect the palatability of cooked rice.

Free sugars, such as glucose and sucrose, and free amino acids such as glutamic acid and aspartic acid, are the main components (37, 38) that directly influence the sweetness and the umami taste of cooked rice. To investigate the changes of those tastes of cooked stored rice during storage, free sugar and free amino acid contents were determined. Fresh *japonica* Koshi brown rice had the highest cooked free sugar content, whereas *indica* cv. Yume had the lowest (Table 3A). It is well-known that Koshi belongs to the group of rice which has the best quality and commands a high price on the Japanese rice market, because it has good palatability such as stickiness, less hardness, sweet, and good appearance. Among the free sugar contents, the sucrose content is the highest one in both brown and milled forms. At a 90% milling degree, among milled samples, fresh Hoshi has the highest content of free sugar (Table 3C). These results indicated that the milling process affected the chemical composition of milled rice as previously reported (14, 37–39). Moreover, chemical compositions may depend on the grain structure and shape of the cultivars such as the composition of the outer and inner layers and short or long grain. After 6 months of storage at room temperature, the contents of sucrose and raffinose of all samples were reduced, whereas the contents of glucose and fructose increased significantly ( $p \leq 0.05$ ; Table 3A,C). These results suggested that starch and high molecular weight sugars were hydrolyzed at room temperature and catalyzed in part by some active debranching enzymes; thus, the content of disaccharides and/or polysaccharides, such as sucrose and raffinose, was reduced, whereas that of monosaccharides, such as fructose and glucose, which are the main components of reducing sugar, increased during storage at room temperature. The change in maltose content was small and tended to increase. During storage at low temperature, changes in free sugar contents were low (Table 3B,D).

The changes in free amino acid contents are presented in Figure 1. Among the samples, hybrid cv. Hoshi, both brown



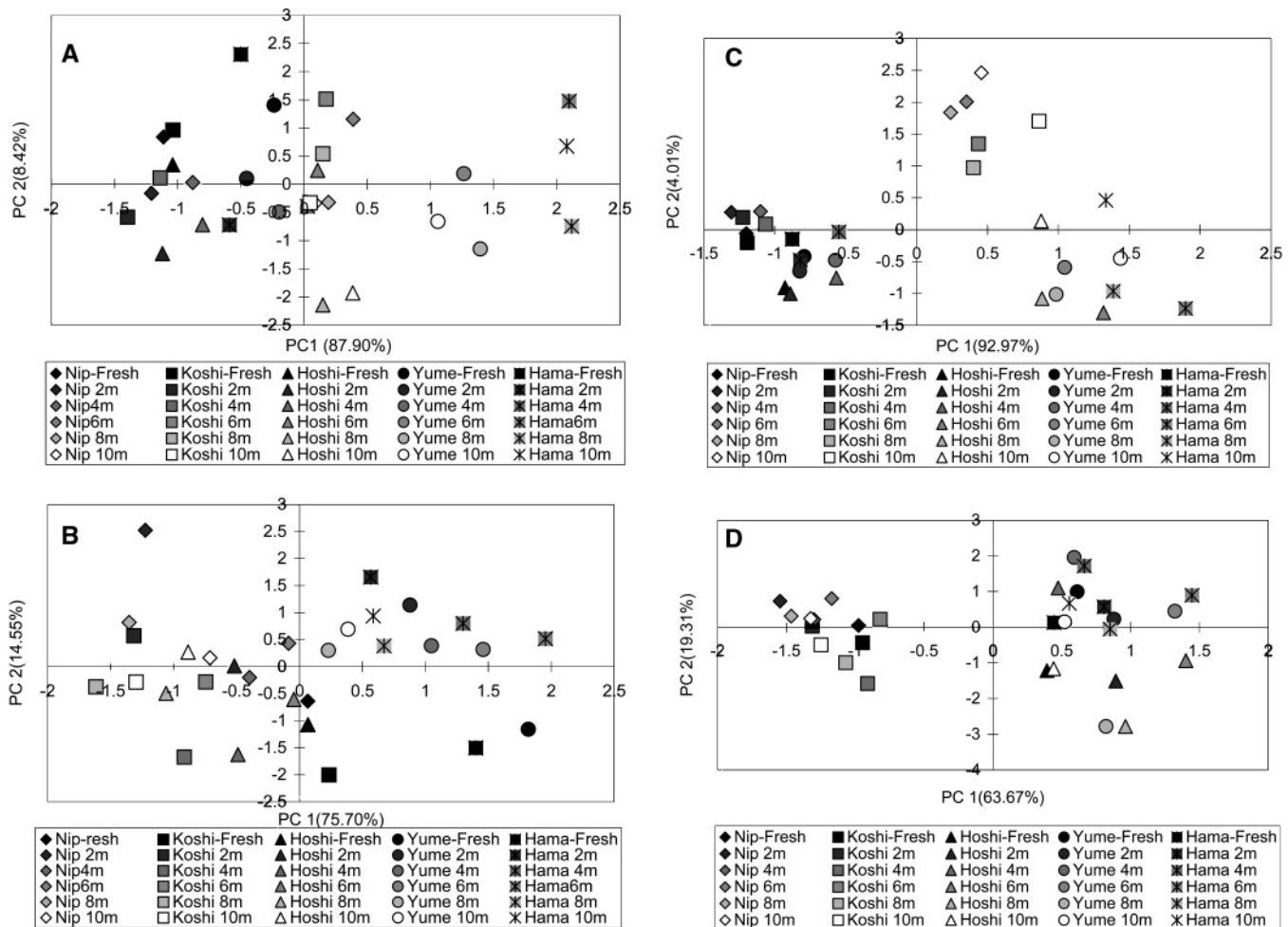
**Figure 2.** Applied PCA to the physicochemical characteristics for samples: (A) brown rice at room temperature; (B) brown rice at low temperature; (C) milled rice at room temperature; (D) milled rice at low temperature.

and milled types, had higher free amino acid contents than others, whereas brown and milled *indica* cv. Yume had lower. The free amino acid contents of all *japonica*, hybrid, and *indica* rice, either brown (Figure 1A,B) or milled (Figure 1C,D), was significantly reduced ( $p \leq 0.05$ ) during storage at room temperature, whereas the degree of reduction was small at low temperature. These results are in agreement with those reported previously (40). Among amino acids, Asp, Glu, Ala, and GABA are the main amino acids of cooked rice, and the change of their contents during storage was clearly observed, whereas the change of contents of other amino acids was not as their contents are low. Taken together, our data demonstrated that both free sugars and free amino acids changed noticeably during storage at room temperature. These changes may have an effect on the sweet and umami tastes of cooked stored rice.

**Use of Physicochemical Measurements to Characterize Stored Samples.** PCA was used to summarize the physicochemical properties measured to characterize fresh and stored rice samples. Figure 2 shows the PCA results from a model containing all samples based on physicochemical properties (5 samples  $\times$  6 storage duration  $\times$  17 variables). Parts A and B of Figure 2 are presented for brown rice stored at room and low temperature, respectively. For brown rice stored at room temperature, PC1 and PC2 accounted for 31.75 and 28.44% of the total variance, respectively (Figure 2A). *japonica* cv. Koshi and Nip are shown in the lower part of the graph, whereas *indica* is in the upper part of the graph. *japonica* and *indica* samples were therefore distinguished by PC2, which mainly depended on positive loading variables (Glu,  $\beta$ -amylase, Asp, sum AA,

AC) and negative loading variables ( $H_1$ ,  $-H_1$ , max visc). The fresh samples loaded positively, whereas stored samples loaded negatively on PC1, which loaded negatively for variables such as max visc, FA, EV, WUR, reducing sugar, AC, and glucose and positively for variables such as protein,  $H_1$ ,  $-H_1$ ,  $\beta$ -amylase, sum AA, Glu, Asp, sum sugar, sucrose, and pH. Therefore, the fresh brown samples tended to appear on the right part, whereas the stored ones appeared on the left part of the graph. At low temperature, the fresh and stored brown rice samples were not separated. However, the differences between *japonica* and *indica* samples were divided with PC1 and PC2, which represented 34.32 and 28.07%, respectively (Figure 2B). Cv. Yume appeared on the upper part of the graph, which mainly depended on PC2 with positive loading for AC, EV, and WUR, whereas Koshi and Nip appeared on the left and Hoshi and Hama appeared on the right part of the graph, which mainly depended on PC1 with positive loading for sum AA, Glu, Asp, and  $\beta$ -amylase and negative loading for  $H_1$ ,  $-H_1$ , and max visc. These results are consistent with the above findings, which showed that brown *indica* rice samples had higher values of AC, EV, WUR, amino acids, and  $\beta$ -amylase, whereas brown *japonica* rice was stickier. The effect of storage on milled rice, which was studied on the basis of physicochemical properties, was similar to that for brown rice, and the results are shown in parts C and D of Figure, respectively. Fresh and room-temperature-stored milled rice samples were distinguished with PC1 and PC2, which accounted for 37.55 and 26.22% of the total variance, respectively. When PC1 was 44.65% and PC2 23.37%, fresh and low-temperature-stored samples were not





**Figure 3.** Applied PCA to the response patterns of sensors for samples: (A) brown rice at room temperature; (B) brown rice at low temperature; (C) milled rice at room temperature; (D) milled rice at low temperature.

separated; however, differences could be detected between *japonica* and *indica*. We concluded that on the basis of physicochemical analyses, when stored at room temperature, fresh and room-temperature-stored rice could be distinguished. However, in the case of storage at low temperature, fresh and stored samples could not be separated. In addition, *japonica* and *indica* rice could be identified using physicochemical measurements.

**Changes Observed by the Taste Sensing System and Correlation Coefficient between Sensor Responses and Taste Components.** The potential response patterns (mV) from a total of 10 sensors collected from three types of sensors (negative, positive, and hybrid sensors) to fresh and stored brown and milled samples were subjected to PCA (5 samples  $\times$  6 storage duration  $\times$  responses of 10 sensors). *japonica* and *indica* rice could be distinguished using the taste sensing system. The results of brown rice stored at room and low temperatures are shown in parts A and B of Figure 3, and those of milled rice are presented in parts C and D of Figure 3, respectively. Although the differences among brown rice were not easily recognized, *japonica* and *indica* milled rice were differentiated, and the changes of milled rice during storage were clearly distinguished. As presented in Figure 3A, fresh and room-temperature-stored brown rice samples were characterized by the potential responses of sensors, PC1 and PC2, which accounted for 87.90 and 8.41% of the total variance, respectively. The fresh harvested samples appeared on the upper left, which exhibited high positive values in the response of the S10 and high negative values in the response of the S7 that mainly depended on PC2. Samples stored

for 2 and 4 months at room temperature appeared on the lower left and samples stored for 6 months on the right part of the graph, where PC1 had a strong effect. The fresh brown rice appeared in the lower right part of the figure. However, other low-temperature-stored samples were scattered (Figure 3B). Therefore, there was no discrimination with respect to storage at low temperature. As shown in Figure 3C, with 92.97 and 4.01% contribution rates of PC1 and PC2, respectively, when stored at room temperature, fresh and stored samples of *japonica* and *indica* milled rice were clearly identified. Fresh Koshi and Nip appeared on the lower left, whereas 2- and 4-month-stored samples appeared on the upper left and 6-month-stored samples on the upper right of the graph, respectively. In contrast, fresh and 2- and 4-month-stored *indica* and hybrid samples are shown on the lower left, whereas samples stored for  $>6$  months are shown on the lower right of the graph. The loading values of PC1 and PC2 indicated that all of the sensors positively correlated with PC1, which was mainly a component for the discrimination of the samples following the storage duration, with the exception of S10. With respect to PC2, the sensors could be mainly split into three groups: S10, which had high positive value; S2, S7, and S4, which exhibited lower positive value; and S3, S5, and S6, which negatively correlated with PC2. At low temperature of storage, as illustrated in Figure 3D, milled rice was separated into two groups, *japonica* in the left part and hybrid and *indica* on the right part of the graph, with the contribution rates of PC1 and PC2 of 63.67 and 19.31%, respectively.

**Table 4.** Correlation Coefficients (*r*) between Taste Components and Responses of Sensors<sup>a</sup>

sensor	room temperature				low temperature			
	FA	pH	sum sugar	sum AA	FA	pH	sum sugar	sum AA
<b>A. Brown Rice</b>								
S1	0.77a	-0.87a	-0.54a	-0.04	0.54a	0.07	-0.26	0.14
S2	0.84a	-0.95a	-0.52a	-0.06	0.68a	-0.17	-0.44b	0.03
S3	0.80a	-0.84a	-0.56a	0.07	0.51a	0.01	-0.25	0.30
S4	0.78a	-0.90a	-0.54a	-0.10	0.39b	0.12	-0.30	0.08
S5	0.81a	-0.94a	-0.48a	-0.03	0.78a	-0.11	-0.27	0.22
S6	0.77a	-0.76a	-0.61a	0.11	0.40b	0.03	-0.24	0.30
S7	0.82a	-0.93a	-0.47a	-0.03	0.79a	-0.17	-0.22	0.25
S8	0.83a	-0.90a	-0.50a	0.01	0.64a	0.07	-0.27	0.26
S9	0.83a	-0.92a	-0.51a	-0.02	0.63a	0.00	-0.22	0.25
S10	-0.36	0.39b	0.44b	-0.06	-0.12	0.60a	-0.12	0.02
<b>B. Milled Rice</b>								
S1	0.81a	-0.95a	-0.04	0.00	0.31	-0.23	0.34	0.28
S2	0.60a	-0.94a	-0.19	-0.13	-0.14	0.05	-0.50a	-0.35
S3	0.82a	-0.94a	-0.07	0.00	0.63a	-0.24	0.46b	0.58a
S4	0.82a	-0.93a	-0.05	0.00	0.60a	-0.23	0.49a	0.54a
S5	0.75a	-0.98a	-0.18	-0.13	0.88a	-0.45b	0.11	0.45b
S6	0.80a	-0.94a	-0.01	0.03	0.72a	-0.38b	0.49a	0.62a
S7	0.74a	-0.98a	-0.13	-0.12	0.74a	-0.31	0.01	0.41b
S8	0.79a	-0.97a	-0.12	-0.07	0.76a	-0.33	0.43b	0.59a
S9	0.81a	-0.97a	-0.10	-0.05	0.68a	-0.24	0.37b	0.56a
S10	-0.71a	0.63a	0.05	-0.17	-0.20	0.25	-0.26	0.12

<sup>a</sup> FA, fat acidity; sum sugar, sum of free sugars; sum AA, sum of free amino acids; S1–S10, sensors 1–10. Significant correlation a and b at 1 and 5%, respectively.

Using PCA with the responses of 10 sensors, new and old rice could be differentiated. To investigate the effect of main taste components to response of sensors, the correlation coefficient between them was analyzed (Table 4). There were high significant correlations between FA, pH values, and responses of sensors. However, the correlation between responses of sensors and sum sugar and sum AA was smaller and only fairly significant. These results suggested that the taste sensor seemed to be able to taste the aging of rice with respect to off-taste, which was caused mainly by the increase of fat acidity and sourness. Similar to physicochemical measurements, the changes of both *japonica* and *indica*, brown and milled, rice could be detected by the taste sensing system during storage at room temperature. However, it was impossible to distinguish samples stored at low temperature. In comparison with physicochemical measurements, the taste sensing system is a powerful tool, which can be used to detect the aging process of rice grain.

The present investigation has implications for rice consumers, processors, and inspectors. Quality and taste properties of rice stored in different conditions vary greatly. Room temperature reduces quality quickly, whereas low temperature does not. Storage of rice is better in the form of brown rice than milled rice. Thus, rice consumers are advised to store rice in cold places. Stored rice has an increase in WUR, EV, and pasting properties. In addition, *indica* has higher amylose content, WUR, and EV than *japonica*. Therefore, changes in cooking properties should be taken into account and processing conditions correspondingly adjusted. For rice processors, the good method to keep the desired quality of cooked rice is to store rice under cold condition and in brown form to yield the minimum loss in rice quality. For rice inspectors, this study showed that the taste sensing system could be used to show differences between fresh and aged rice samples and also between *japonica* and *indica* rice samples.

## ABBREVIATIONS USED

cv., cultivar; RH, relative humidity; mc, moisture content; RVA, Rapid-Visco-Analyzer; EDTA, ethylenediaminetetraacetate; TCA, trichloroacetic acid; HPLC, high-performance liquid chromatography; LSD, least significant difference; PCA, principal component analysis; PC1, first principal component; PC2, second principal component; LCT, low-compression test, 25%; HCT, high-compression test, 90%; WUR, water uptake ratio; IBV, iodine blue value; EV, expanded volume; SS, solid substances.

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**Supporting Information Available:** Cooking properties of milled rice, chemical components, enzyme activities, textural properties, and pasting properties raw data. This material is available free of charge via the Internet at <http://pubs.acs.org>.

## LITERATURE CITED

- Barber, S. Milled rice and changes during aging. In *Rice: Chemistry and Technology*, 1st ed.; Houston, D. F., Ed.; American Association of Cereal Chemists: St. Paul, MN, 1972; pp 215–263.
- Zhou, Z.; Robards, K.; Helliwell, S.; Blanchard, C. Aging of stored rice: changes in chemical and physical attributes. *J. Cereal Sci.* **2002**, *35*, 65–78.
- Meullenet, J.-F. C.; Marks, B. P.; Griffin, K.; Daniels, M. J. Effects of rough rice drying and storage conditions on sensory profiles of cooked rice. *Cereal Chem.* **1999**, *76* (4), 483–486.
- Meullenet, J.-F. C.; Marks, B. P.; Hankins, J.-A.; Griffin, V. K.; Daniels, M. J. Sensory quality of cooked long-grain as affected by rough rice moisture content, storage temperature, and storage duration. *Cereal Chem.* **2000**, *77* (2), 259–263.
- Shibuya, N.; Iwasaki, T.; Yanase, H.; Chikubu, S. Studies on deterioration of rice during storage. Part I. Changes of brown rice and milled rice during storage (in Japanese). *Nippon Shokuhin Kougyougaku Kaishi* **1974**, *21* (12), 597–603.
- Sowbhagya, C. M.; Bhattacharya, K. R. Changes in Pasting Behavior of Rice During Aging. *J. Cereal Sci.* **2001**, *34*, 115–124.
- Yasumatsu, K.; Morikata, S.; Karinuma, T. Effect of changes during storage in lipid composition of rice and its amylograph. *Agric. Biol. Chem.* **1964**, *28*, 265–272.
- Villareal, R. M.; Resurreccion, A. P.; Suzuki, L. B.; Juliano, B. O. Changes in physicochemical properties of rice during storage. *Starch* **1976**, *28*, 88–94.
- Indudhara, S. Y. M.; Sowbhagya, C. M.; Bhattacharya, K. R. Changes in physicochemical properties of rice with aging. *J. Sci. Food Agric.* **1978**, *29*, 627–639.
- Cao, Y.; Wang, Y.; Chen, X.; Ye, J. Study on sugar profile of rice during aging by capillary electrophoresis with electrochemical detection. *Food Chem.* **2004**, *86* (1), 131–136.
- Dhaliwal, Y. S.; Sekhon, K. S.; Nagi, H. P. S. Enzymatic activities and rheological properties of stored rice. *Cereal Chem.* **1991**, *68*, 18–21.
- Toko, K. *Biomimetic Sensor Technology. Taste Sensor (6)*; Cambridge University Press: Cambridge, U.K., 2000; pp 164–165.
- Ohtsubo, K.; Ikezaki, H.; Taniguchi, A.; Okadome, H.; Toyoshima, K.; Inbe, T. Investigation on palatability assay of rice grains by the taste sensor system (in Japanese). *Tech. Rep. IEICE* **2000**, 25–29.

- (14) Tran, T. U.; Suzuki, K.; Okadome, H.; Homma, S.; Ohtsubo, K. Analyses of the tastes of brown and milled rice with different milling yields using a taste sensing system. *Food Chem.* **2004**, *88* (4), 557–566.
- (15) Okadome, H.; Toyoshima, H.; Ohtsubo, K. Multiple measurements of physical properties of individual cooked rice grains with a single apparatus. *Cereal Chem.* **1999**, *76* (6), 855–860.
- (16) Batcher, O. M.; Helmlintoller, K. F.; Dawson, E. H. Developments and application of methods for evaluating cooking and eating quality of rice. *Rice J.* **1956**, *59*, 4–8.
- (17) Toyoshima, H.; Okadome, H.; Ohtsubo, K.; Suto, M.; Horisue, N.; Inatsu, O.; Narizuka, A.; Inouchi, N.; Fuwa, H. Cooperative test on the small-scale rapid method for the gelatinization properties test of rice flour with a RVA. *Nippon Shokuhin Kagaku Kogakukaishi* **1997**, *44*, 579–584 (in Japanese).
- (18) Juliano, B. O. A simplified assay for milled-rice amylose. *Cereal Sci. Today* **1971**, *12*, 334–360.
- (19) Ohtsubo, K.; Yanase, H.; Ishima, T. Colorimetric determination of fat acidity of rice—Relation between quality change of rice during storage and fat acidity determined by improved Duncombe method. *Rep. Natl. Food Res. Inst.* **1987**, *51*, 59–65 (in Japanese).
- (20) Duncombe, W. G. The Colorimetric Micro-Determination of Long-Chain Fatty Acids. *Biochem. J.* **1963**, *88*, 7–10.
- (21) Somogyi, M. A new reagent for the determination of sugars. *J. Biol. Chem.* **1945**, *160*, 61–68.
- (22) Somogyi, M. Notes on sugar determination. *J. Biol. Chem.* **1952**, *195*, 19–23.
- (23) Nelson, N. A photometric adaption of the Somogyi method for the determination of glucose. *J. Biol. Chem.* **1944**, *153*, 375–380.
- (24) Loewus, F. A. Improvement in anthrone method for determination of carbohydrates. *Anal. Chem.* **1952**, *24*, 219.
- (25) Imai, Y.; Tokutake, S.; Yamaji, N.; Suzuki, M. An improved method for measuring of  $\alpha$ -glucosidase activity in rice *koji*. *J. Brew. Soc. Jpn.* **1997**, *92* (4), 296–302 (in Japanese).
- (26) Iwata, H.; Iwase, S.; Takahama, K.; Matura, H.; Itani, T.; Aramaki, I. Relationship between  $\alpha$ -glucosidase activity and physical and chemical properties of rice. *Nippon Shokuhin Kagaku Kaishi* **2001**, *48* (7), 482–490 (in Japanese).
- (27) Palmiano, E. P.; Juliano, B. O. Biochemical changes in the rice grain during germination. *Plant Physiol.* **1972**, *49*, 751–756.
- (28) Ivarsson, P.; Kikkawa, Y.; Winquist, F.; Krantz-Rulcker, C.; Hojer, N. E.; Hayashi, K.; Toko, K.; Lundstrom, I. Comparison of a voltammetric electronic tongue and a lipid membrane taste sensor. *Anal. Chim. Acta* **2001**, *449*, 59–68.
- (29) Arikawa, Y.; Toko, K.; Ikezaki, H.; Shinha, Y.; Ito, T.; Oguri, I.; Baba, S. Analysis of sake taste using multi-electrode taste sensor. *Sensor Mater.* **1995**, *7* (4), 261–270.
- (30) Tran, T. U.; Okadome, H.; Murata, M.; Homma, S.; Ohtsubo, K. Comparison of Vietnamese and Japanese rice cultivars in terms of physicochemical properties. *Food Sci. Technol. Res.* **2001**, *7* (4), 323–330.
- (31) Perez, C. M.; Juliano, B. O. Texture changes and storage of rice. *J. Text. Stud.* **1981**, *12*, 321–333.
- (32) Park, J. K.; Kim, S. S.; Kim, O. K. Effect of milling ratio on sensory properties of cooked rice and on physicochemical properties of milled and cooked rice. *Cereal Chem.* **2001**, *78* (2), 151–156.
- (33) Sowbhagya, C. M.; Bhattacharya, K. R. Lipid autoxidation in rice. *J. Food Sci.* **1976**, *41*, 1018–1023.
- (34) Pomeranz, Y. Chapter 3: Biochemical, functional, and nutritive changes during storage. In *Storage of Cereal Grains and Their Products*, 4th ed.; Sauer, D. B., Ed.; AACC: St. Paul, MN, 1992; pp 55–141.
- (35) Champagne, E. T.; Wood, D. F.; Juliano, B. O.; Bechtel, D. B. The rice grain and its gross composition. In *Rice Chemistry and Technology*, 3rd ed.; Champagne, E. T., Ed.; AACC: St. Paul, MN, 2004; pp 77–107.
- (36) Awazuhara, M.; Nakagawa, A.; Yamaguchi, J.; Fujiwara, T.; Hayashi, H.; Hatae, K.; Chino, M.; Shimada, A. Distribution and characterization of enzymes causing starch degradation in rice (*Oryza sativa* Cv. Kohihikari). *J. Agric. Food Chem.* **2000**, *48*, 245–252.
- (37) Tajima, M.; Horino, T.; Maeda, M.; Rok Son, J. Maltooligosaccharides extracted from outer-layer of rice grain. *Nippon Shokuhin Kogyo Gakkaishi* **1992**, *39* (10), 857–861 (in Japanese).
- (38) Saikusa, T.; Horino, T.; Mori, Y. Distribution of free amino acids in the rice kernel and kernel fractions and the effect of water soaking on the distribution. *J. Agric. Food Chem.* **1994**, *42*, 1122–1125.
- (39) Champagne, E. T.; Bett, K. L.; Vinyard, B. T.; Wedd, B. D.; McClung, A. M.; Barton, F. E.; Lyon, B. G.; Moldenhauer, K.; Linscombe, S.; Kohlwey, D. Effects of drying conditions, final moisture content and degree of milling on rice flavor. *Cereal Chem.* **1997**, *74*, 566–570.
- (40) Aibara, S.; Ismail, I. A.; Yamashita, H.; Ohts, H.; Sekiyama, F.; Morita, Y. Changes in rice bran lipids and free amino acids during storage. *Agric. Biol. Chem.* **1986**, *50* (3), 665–673.

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