Chemical and sensory evaluation of aromatic rice cultivars.

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Abstract

It is known that there is a distinct difference in aroma strength among aromatic rice cultivars. In the present study, we determined 2-acetyl-1-pyrroline concentration (2AP) of some aromatic rice cultivars and evaluated the intensity of the characteristic aroma of them using a sensory test to elucidate the relationship between 2AP concentration and the sensory intensity. About a 3-fold difference of 2AP levels was observed among the 4 aromatic rice cultivars. The linear relationship was shown between the aroma rating and logarithmic value of the concentration of 2AP in samples,

Key words

Aromatic rice, Cultivar difference, *Oryza sativa* L., Sensory evaluation, 2-Acetyl-1-pyrroline.

Introduction

Aromatic or scented rice, a rice type that emits a special aroma when cooked, has been cultivated in many regions throughout Japan and in South and South-East Asian countries from ancient days. In the course of our study on the aromatic rice cultivars from all over Japan and other countries, we found that there is a distinct difference in aroma strength among cultivars. Namely some are highly aromatic, some moderate while others have just a trace of aroma. Some researchers examined the aroma of aromatic rice by a sensory test (Tsuzuki et al., 1977; Tripathi and Rao, 1979; Sanchez and Raymundo, 1991). Buttery et al. (1983) identified the characteristic aroma compound in steam volatile oils of cooked aromatic rice as 2-acetyl-1-pyrroline (2AP).

Fushimi (1992) and Fushimi et al. (1996) developed a simple method for analysis of the 2AP concentration from a small amount of rice flour directly extracted with hot ethanol instead of collecting the headspace gas over cooking rice. However they have not conducted a sensory test for their analyzing method. In the present study, we adopted a method similar to Fushimi's analyzing method of 2AP to some aromatic rice cultivars cultivated and harvested under the same conditions and evaluated the intensity of the characteristic aroma of them using a sensory test to elucidate the relationship between 2AP concentration and the sensory intensity. We adopted two-steps of sensory tests using experienced and non-experienced panelists.

Materials and Methods

Plant materials: We selected four aromatic rice cultivars; ‘Tohwanishiki’ and ‘Hieri’ from Japan, ‘Sari Queen’ bred from a cross between ‘Nipponbare’ and ‘Basmati 370’, and ‘Surjamukhi’ from India. We cultivated the four aromatic rice cultivars and two non-aromatic rice cultivars (‘Koshihikari’ and ‘Nakateshinsenbon’) under the same conditions in the rice fields of Hiroshima Prefectural University in 1998. All the harvested materials were kept in a refrigerator and hulled just before the experiments during January and February in 1999. We used brown rice for the sensory test and analysis of 2AP because 2AP decreased logarithmically from the surface to the center of rice grains and milling degree may influence the concentration of 2AP (Harada et al., 1990).

Experiment 1: The five kinds of rice samples, blended with 100, 75, 50, 25 and 0 % of aromatic rice ‘Hieri’ in non-aromatic rice ‘Koshihikari’, were analyzed for 2AP concentration and their aroma was scored by sensory test. Two grams of brown rice samples and 3 mL of water in a sealed test tube were treated in boiling water for 30 minutes. After cooling to room temperature, three panelists (staff) ranked the aroma strength of the samples and gave a score of 0 (non-aromatic) to 3 (most aromatic).

Experiment 2: The concentrations of 2AP in the ten rice samples composed of 4 aromatic rice cultivars, 4 blended samples (10% aromatic rice with 90% non-aromatic ‘Nakateshinsenbon’) and 2 non-aromatic rice cultivars were analyzed and subjected to the sensory test. In this experiment, the aroma intensities of the 10 samples were rated on the following 5-point category scale by 24 panelists (four groups of 6 members, 10 male and 14 female students aged between 20 and 22 years): 0 (non-aromatic), 1(subtle aromatic), 2(clearly aromatic but not so strong), 3(aromatic strong as Hieri), and 4 (more aromatic than Hieri). The reference samples, Nakateshinsenbon for the score 0 and Hieri for the score 3, were presented to panelists before test. The sensory test was conducted freely by a six-panel group in turn in a quiet laboratory during the lesson.

Analysis of 2AP

Extraction of 2AP: The brown rice sample was ground into a powder (less than 0.5 mm in diameter). A half of one gram of sample was placed into pressure-resistant vials, and suspended in 2.0 mL of ethanol (99.5%). Extraction was carried out in an 80?C water bath with shaking (160 cycles/min) for 60 minutes. After cooling to room temperature, 100μL of 0.03% 3-hexanol solution in ethanol was added as an internal standard (IS) to the suspension. After filtration using a membrane filter (pore size 0.2 μm), the filtrate was used for quantitative analysis of 2AP. Each sample was extracted three times.

Quantitative analysis of 2AP: Gas chromatograph mass spectrometry was performed with a Hewlett Packard 6890 gas chromatograph coupled to a Hewlett Packard 6890 MSD. Capillary column Ultra-1 (Hewlett Packard) 50m x 0.2mm was used. The injector temperature was 150?C, and the splitless mode was used. The injection volume was 2μL. The flow rate of the carrier gas (helium) was 1.0 mL/min. The GC column-oven temperature was increased from 50?C to 140?C at a rate of 6?C /min and then increased at a rate of 20?C /min to 240?C and held at that temperature for 20 min. 2AP was identified by its characteristic fragments m/z 68, 69, 83, and 111. Then the concentration of 2AP in the extract was determined using a selected ion monitoring (SIM) mode at the peak area of m/z 83. The IS (3-hexanol) was quantified at m/z 73. Samples were quantified using a calibration curve, which was prepared from the standard solutions of 2AP. The concentration of 2AP in the rice is shown as a weight ratio of 2AP per dry matter of the rice.

Results

Experiment 1: The ethanol extract clearly emitted characteristic aroma. The concentrations of 2AP and sensory intensity of the 5 blended samples are shown in Table 1. The level of 2AP in 75, 50, 25 and 0 % of aromatic rice decreased in proportion to the blending ratio of the aromatic rice, and every value showed within plus minus 10 % of the expected value. The aroma scores nearly corresponded with the blending ratio of aromatic rice.

Experiment 2: The concentration of 2AP in 6 cultivars and aroma intensity of 10 samples are shown in Table 2. About a 3-fold difference of 2AP levels, 135 ppb in Tohwanishiki and 52 ppb in Surjamukhi was observed among the 4 aromatic rice cultivars. In the case of non-aromatic rice, the level of 2AP was under the detection limits of this analytical method (2 or 3 ppb).

Analysis of variance on sample difference in the aroma rating obtained from the sensory test showed highly significant difference among both members and samples. The mean aroma rating increased with the level of 2AP from 0 to 50 ppb of 2AP and did not increase significantly over 50 ppb of 2AP (Table 2). According to Fetchner‘s law (Fetchner, 1860), the relationship between physical intensity and sensation intensity is expressed by linear model obtained on a semi-log plot. Fig. 1 shows the linear relationship between the aroma rating and logarithmic value of the concentration of 2AP in samples, where 2AP concentrations of 2 non-aromatic rice samples were regarded as 1 ppb.

Discussion

2AP is a chemical compound that emits a buttered-popcorn-like aroma at specific concentrations in rice and is reported to be an important component of the toasted or sweet odor in wheat bread, popcorn, boiled beef and so on (Schieberle, 1991). 2AP has a lower odor threshold than the many other volatile compounds in rice and the concentration in rice is very low (Buttery et al. 1983, 1988). In this study we used a simple method for analysis of the 2AP concentration from a small amount of rice flour directly extracted with hot ethanol instead of collecting the headspace gas over cooking rice because the raw rice grains emit aroma when chewed. The 2AP concentration quantified by using this analyzing method were not so different from those reported by Buttery et al. (1983), and the results of the panel test do not contradict with the quantification by this method. Yoshihashi (2002), by a method using isotope-labeled analogues of 2AP, reported that 2AP does not form during cooking or post-harvest processing. We used a 2AP analyzing method similar to Fushimi’s method. The difference of both methods is the ratio of rice powder to ethanol and an internal standard. Fushimi used an identical rice sample alternately with every sample (Fushimi 1992; Fushimi and Ishitani 1994).

About a 3-fold difference of 2AP levels was observed among the 4 aromatic rice cultivars. Many researchers have examined the inheritance of aroma (Pinson, 1994; Kato and Itani, 1996). Most of these studies indicate that this character is controlled by a small number of major genes. In addition, pre- and post-harvest conditions can influence aroma strength in aromatic rice. For excellent aromatic rice production, accumulation of studies and information on aroma intensity is needed because it depends on genetic and environmental conditions.

Table 1 2AP concentrations of five samples blended with various % of aromatic rice to non-aromatic rice.

Blending % of	2AP of brown rice			Aroma rating of cooked rice
aromatic rice[1)]	(ppb)		ratio(%)	(0-3)
100	104?16[2)]	a[3)]	100	2.3?0.6[2)]
75	72?12	b	69	1.7?0.6
50	56?19	b	54	1.0?0.0
25	28?10	c	27	1.0?0.0
0	0?0		0	0.0?0.0

1) Aromatic rice 'Hieri' was blended to non-aromatic rice 'Koshihikari'. 2) Means ? standard deviations (n=3). 3) Means followed by the same letter do not differ at the 5% level by LSD method.

Table 2 Scores for aroma of cooked rice samples with various 2AP concentrations.

Samples	2AP (ppb)		Scores for aroma	
Towanishiki	135?8	a [2)]	2.04?0.86	a [2)]
Hieri	98?5	b	2.04?0.86	a
Sari Queen	64?8	c	1.88?1.03	ab
Surjamkhi	52?4	d	1.92?0.97	ab
Towanihiki (10%)[1)]	14		1.46?0.88	bc
Hieri (10%)	10		1.58?0.93	bc
Sari Queen (10%)	6		0.77?0.83	c
Surjamkhi (10%)	5		0.67?0.82	c
Koshihikari	0	e	0.38?0.58	d
Nakateshinsenbon[4)]	0	e	0.21?0.41	d

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