Changes in NMR Relaxation of Rice Grains, Kernel Quality and Physicochemical Properties in Response to a High Temperature after Flowering in Heat-Tolerant and Heat-Sensitive Rice Cultivars

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Abstract: Rice productivity is related to the ability of plants to adapt to heat stress. The heat-tolerant cultivars ‘Nikomaru’ and ‘Chikushi 64’ and heat-sensitive cultivar ‘Hinohikari’ were grown at 30ºC and 25ºC for 49 days after flowering. At 30ºC, only a few white immature kernels were produced in ‘Nikomaru’ and ‘Chikushi 64’, but about 22% of grains had immature kernels in ‘Hinohikari’. The high temperature(30ºC) caused no significant changes in grain dry weight, water content, and the NMR $T_1$ value during the early ripening stage in ‘Nikomaru’ and ‘Chikushi 64’. It also did not affect grain development, especially with respect to the nucellar epidermis, in ‘Nikomaru’ and ‘Chikushi 64’, but caused clear cessation of development of the nucellar epidermis at 14 days after flowering in ‘Hinohikari’. In addition, high temperature decreased the amylose content and increased hardness vs. adhesion ratio of cooked rice in both ‘Nikomaru’ and ‘Chikushi 64’ resulting a softer, less sticky texture, but not in ‘Hinohikari’. The maximum viscosity and breakdown values were increased, and final viscosity decreased at 30ºC in all three cultivars. These results suggested that starch in the endosperm of grains changed from a fluid state to a doughy state more slowly in ‘Nikomaru’ and ‘Chikushi 64’ than in ‘Hinohikari’, in which the water content and NMR relaxation time decreased, and transported assimilates accumulated slowly during grain development.

Key words: ‘Chikushi 64’, High temperature, Kernel quality, ‘Nikomaru’, NMR relaxation time ($T_1$ and $T_2$), Nucellar epidermis, Rice, Water status.

Changes in crop yield and grain quality, especially in northern Asia, are considered to be serious consequences of global warming. At temperatures above a certain optimum, yield and kernel quality of rice (Oryza sativa L.) decrease (Peng et al., 2004). The effects of high temperature on the kernel quality of rice cultivars can be used to assess their tolerance to heat stress (Nagato et al., 1961; Nagato and Ebata, 1965). High temperatures at the milky stage of grain filling had the greatest influence on the chalkiness of rice grains (Tashiro and Wardlaw, 1991), and the panicule was most sensitive to high temperature (Sato and Inaba, 1973; Morita et al., 2004). Tashiro and Wardlaw (1991) reported that white-backed and milky white kernels occurred at temperatures above 27ºC and that the proportion of these grains increased markedly at 30 and 33ºC, respectively. Moreover, rice grown at 38/21ºC (day/night) contained more chalky grains (Lisle et al., 2000). The chalkiness characteristics were influenced by the shape, size, and packing of the amyloplasts in the rice kernels, and these properties all differed from those in translucent grains. Microscopic observation of the chalky part of grains that ripened at high temperature revealed loosely packed starch granules with air spaces between them that reflected light randomly, leading to the milky appearance (Tashiro and Wardlaw, 1991; Zakaria et al., 2002).

Differences have been observed in grain chalkiness among rice cultivars whose grains ripened at high temperature. For example, the japonica cultivars ‘Hatsuboshi’ and ‘Hinohikari’ produce chalky grains at temperatures above 27ºC, showing sensitivity to high temperature (Wakamatsu et al., 2007). The grains of ‘Hinohikari’ grown at 30ºC had white-backed kernels in 85% of the ripened grains and no perfect kernels (Funaba et al., 2006). Recently, the cultivar ‘Nikomaru’, with tolerance to high temperatures has been bred (Sakai et al., 2007), as well as a similarly tolerant line, ‘Chikushi 64’.

Previously, we showed that the NMR relaxation time of ‘Hinohikari’ grains offer a good non-destructive test that reveals physical changes in rice grains exposed to temperature stress (Funaba et al., 2006). Changes in the NMR spin–lattice relaxation time ($T_1$) in rice...
grains were closely related to the quantity of water in the grains until the mid-mature stage, whereas the spin–spin relaxation time (\( T_2 \)) was a more sensitive diagnostic indicator of the accumulation of dry matter and kernel quality, both of which were influenced by temperature stress. However, varietal differences in heat tolerance are not yet fully understood in relation to grain water status and morphological and physicochemical features. Therefore, here we evaluated the tolerance to high temperature in relation to the water-related grain properties during the grain-filling stage in heat-tolerant ‘Nikomaru’ and ‘Chikushi 64’, and the heat-sensitive ‘Hinohikari’. Additionally, the extremely high temperature before flowering in 2007 seems to have induced heat acclimatization against the 30°C treatment. Therefore, we briefly discuss the influence of the unusually high air temperature in 2007 on the performance of these cultivars.

Materials and Methods

1. Plant materials

Seeds of japonica rice (Oryza sativa L. cvs. ‘Hinohikari’ and ‘Nikomaru’, and line ‘Chikushi 64’ bred by Fukuoka Agricultural Research Center) were sown in a seed bed on 25 May 2007. On 21 June, three seedlings were transplanted into each plastic pot (1/5000 a), that contained 3.0 kg soils. Plants were allowed to grow until the heading stage at the experimental field of Kyushu University. Irrigation and pesticide were applied to ensure optimal plant growth. Compound fertilizer (N-P2O5-K2O:4-4-4%) at 0.2 g N was supplied to each pot, as basal dressing. Additionally, 0.1 g N ammonium sulfate (N:21%) was topdressed at the panicle formation and booting stages. At the flowering stage on 19 August, 29 August and 1 September in ‘Chikushi 64’, ‘Hinohikari’ and ‘Nikomaru’, respectively, when the mean temperature was 27.4 and 27.6°C, and the pots were transferred to phytotron a natural lighting growth cabinet, and plants were grown in two temperature treatments 25 (±1)°C and 30 (±1)°C until maturity. After flowering, 3 pots were randomly selected at 7-day intervals from each treatment for analyzing the necessary parameters. Grains (rough rice) on the primary branches and the first (from top) secondary rachis branches were used for the analysis. Each experiment was repeated five times. All parameters were examined at seven stages, 7, 14, 21, 28, 35, 42 and 49 days after flowering (DAF). The pots were arranged in a complete randomized design in a phytotron.

2. \(^1\)H-NMR relaxation time and its analysis and water content

Twenty grains were used for NMR analysis at each sampling time. Spin-lattice relaxation time (\( T_1 \)) of the samples was measured based on the procedure described by Funaba et al. (2006). A \(^1\)H-NMR spectrometer with a magnet operating at 25 MHz for \(^1\)H (Mg25A, JEOL Ltd., Tokyo, Japan) was used for the measurement of \( T_1 \). Twenty grains (rough rice) were prepared for the measurement of NMR relaxation time. Sample was put into an NMR tube (10 mm in diameter) followed by setting in the NMR spectrometer. The probe temperature was controlled by a thermostat connected to the sample chamber of the spectrometer at 30°C.

For \( T_1 \) measurements, the saturation recovery method (90°-\( \tau \)-90° pulse sequence) was used. In this method, \( T_1 \) is determined from \( M_i/M_o \) [1-exp(-\( \tau/T_1 \))] where \( M_i \) is the magnetization amplitude of proton at interval time \( \tau \), and \( M_o \) is the magnetization amplitude of proton in the equilibrium state. In this experiment, free induction decay (FID) signal at every \( \tau \) was obtained by the accumulation of 4 scans. Forty-five \( \tau \) recovery times were logarithmically spaced between 3 and 600 ms. The repetition time of the sequence was always kept more than five times for \( T_1 \), of long fraction. The decay curve of echo signal was analyzed by using a non-linear least-square method on semi-log plots of signal intensity. Two-component analysis by using curve fit method is described (Iwaya-Inoue et al., 2004a). \( T_2 \) was measured by the Carr-Purcell-Meiboom-Gill (CPMG) method or solid echo method. \( T_2 \) is determined from \( M_{2n}=M_o \exp(-2n\tau/T_2) \), where \( n \) is the number of repeated cycles of 180° pulse, \( M_{2n} \) is the magnetization amplitude of the proton signal occurring at time 2\( \tau \) after the initial 90° pulse in CPMG (90° \(-\tau\)-180° \(-2\tau\)-180° \(-2\tau\)-...) pulse sequence. The \( T_2 \) were calculated based on \( n \) of 500 echo signals acquired by accumulation of 16 scans. \( \tau \) was between 0.7 and 1.6 ms. The solid-echo (90°-\( \tau \)-90°) method was also applied for \( T_2 \) measurements when \( T_2 \) values were below 1ms. \( \tau \) was 8 μs. The solid echo signal of the seeds was obtained by accumulation of 128 scans. \( M(t)=\Sigma a_i \times \exp[-(t/T_2i)^{-\beta}] \), where \( m_i \) is Weibull coefficient, and \( a_i \) is signal intensity in each fraction. The relative value of fraction ratio, \( \beta \), is calculated by a formula, \( \beta = a_i/\Sigma a_i \). The decay curve of echo signal was analyzed by using a non-linear least-square method on semi-log plots of signal intensity. For each treatment, four replications were used. Rice grains (rough rice) used for the NMR test were oven-dried at 90°C for 20 hr to determine the water content and dry weight.

3. Morphological features

Grains of primary branches of ‘Nikomaru’, ‘Chikushi 64’ and ‘Hinohikari’ at 14DAF were fixed in FAA (Formalin acetic alcohol: 80% ethanol:100% acetic acid: formalin=90:5:5). Tissues were cut 30-50 μm in thickness using a cryostat (HM-505E, MICROM Co. Ltd., Japan). These sections were stained with 0.1% Toluidine blue. The morphological features in rice grains of the cultivars and line was observed under
a microscope (ECLISE 80i, Nikon Co.Ltd., Japan). About 10 grains from each treatment were used.

4. Rice quality and physicochemical properties
All rice grains (rough rice) were evaluated for percent ripened grain and 100 kernels of the husked grains for their quality. Total four replications were allocated in the present experiment. The ratio of ripened grain and 1000-grain weight were assessed based on grain width of over 1.8 mm, and kernel quality was visually evaluated and scored according to Hoshikawa (1993).

To analyze the physicochemical properties, we used milled rice polished to 90% with a rice polisher machine Pearlest (Kett Electric Laboratory Co.Ltd., Japan). Contents of protein and amylose were measured with Auto Analyzer type II (Bran+Luebbe Co.Ltd., Germany), as described by Matsue et al. (1991). Amylographic characteristics were measured according to Toyoshima et al. (1997) using a Rapid Visco Analyzer (Newport Scientific Pty. Ltd., Australia). Hardness (H) and adhesion (-H) of cooked rice were measured according to Endo et al. (1980) by using a Texturometer GTX-2 (Zenken Co.Ltd., Japan).

Results

1. Influence of high temperature on dry-matter accumulation, water content, and NMR relaxation time in ‘Chikushi 64’ grains during ripening

The dry weight, water content and NMR relaxation time of ‘Hinohikari’, ‘Chikushi 64’ and ‘Nikomaru’ ripened at 25 and 30°C were determined. The changes in these values of ‘Chikushi 64’ and ‘Nikomaru’ showed similar tendency while those of ‘Hinohikari’ showed a different response to each temperature as reported by Funaba et al. (2006). Therefore, only the results of ‘Chikushi 64’ grains are shown in Fig. 1 as typical profiles of these changes. The dry weight of ‘Chikushi 64’ grains that ripened at 25 and 30ºC increased linearly until 21 DAF and did not change significantly thereafter (Fig. 1A). The water contents of grains at both 25 and 30ºC declined markedly until 14 DAF, and declined gradually thereafter until 49 DAF (Fig. 1B). The changes in dry weight and water content were greater at 30ºC during the early ripening stage.

The NMR relaxation time (\(T_1\), \(T_2\)), which can be used as proxies for the grain’s water status, was measured in the rice grains during ripening. The \(T_1\) values at both temperatures were around 250 ms at 7 DAF, and subsequently declined to about 50 ms at 21 DAF (Fig. 1C). On the other hand, \(T_2\) values at both temperatures were about 100 ms at 7 DAF and decreased abruptly to less than 1 ms by 28 DAF, the yellow-ripe stage (Fig. 1D). In general, \(T_1\) and \(T_2\) values in rice grains grown at 30°C were lower than those at 25°C during the early stages of maturation. Especially during the early stages, \(T_1\) was more sensitive than \(T_2\) to temperature.
Fig. 2. Influence of heat stress on physiological characteristics (grain dry weight, water content, and $T_1$ relaxation time) of ‘Hinohikari’, ‘Nikomaru’, and ‘Chikushi 64’ grains at 7 and 14 days after flowering.

Fig. 3. Influence of high temperature on the morphological features of the rice grains at 14 days after flowering. Left, Illustration of central cross section of a rice grain; Right, Samples were stained with 0.1% Toluidine Blue.
2. Influence of high temperature on the grains at the early ripening stage

We examined the influence of high temperature on grains at the early ripening stage, 7 and 14 DAF, to determine whether we could diagnose adverse changes early (Fig. 2). The high temperature increased the dry weight and decreased the water content in ‘Hinohikari’, and both the long and short fractions of $T_1$. $T_1$ values between 100 ms and 3 s are thought to show the existence of free water that is present mainly in vacuoles (Hills and Remigereau, 1997; Iwaya-Inoue and Nonami, 2003; Iwaya-Inoue et al., 2004a, 2004b; Funaba et al., 2006). The changes of $T_1$ in the ‘Hinohikari’ grains were closely related to the water content until the mid-mature stage. In contrast, there was no significant influence of high temperature on grain dry weight or water-related parameters in ‘Nikomaru’. The temperature responses of ‘Chikushi 64’ grains were intermediate between those of ‘Nikomaru’ and ‘Hinohikari’.

At 14 DAF, development of the nucellar epidermis was observed in the kernels of ‘Nikomaru’, ‘Chikushi 64’, and ‘Hinohikari’ grown at 25ºC and 30ºC (Fig. 3). The high temperature had little influence on the morphology of the nucellar epidermis in ‘Nikomaru’ and ‘Chikushi 64’ grown at 30ºC, whereas the nucellar epidermis of ‘Hinohikari’ clearly ceased formation at 30ºC. A similar lack of change in the nucellar epidermis was observed in the kernels of ‘Nikomaru’ and ‘Chikushi 64’ at 21 DAF (data not shown).

3. Influence of high temperature on the quality and physicochemical properties of grains at maturity

A high temperature did not significantly affect the grain yield per plant or the 1000-grain weight in

| Cultivar    | Temp. (ºC) | Grains per pot | Ripened grain (%) | 1000-grain weight (g) | Milky white kernel (%) | White core kernel (%) | White backed kernel (%) | White based kernel (%) | White immature kernel (%) |
|-------------|------------|----------------|-------------------|-----------------------|------------------------|----------------------|------------------------|------------------------|--------------------------|
| Hinohikari  | 25         | 1244           | 88.8              | 21.8 ab               | 0.6 a                  | 2.4 a                | 0.2 a                  | 1.4 ab                 | 5.3 a                    |
|             | 30         | 1337           | 91.5              | 21.6 ab               | 4.6 b                  | 5.6 b                | 8.6 b                  | 3.1 b                  | 22.1 b                   |
| Nikomaru    | 25         | 1513           | 84.1              | 22.2 b                | 0.6 a                  | 1.5 a                | 0.0 a                  | 0.6 ab                 | 2.8 a                    |
|             | 30         | 1389           | 84.8              | 21.7 ab               | 1.1 a                  | 1.6 a                | 1.3 a                  | 0.4 ab                 | 4.4 a                    |
| Chikushi64  | 25         | 1473           | 82.7              | 21.4 ab               | 0.4 a                  | 0.0 a                | 0.0 a                  | 0.0 a                  | 0.4 a                    |
|             | 30         | 1494           | 88.6              | 20.9 a                | 1.1 a                  | 0.8 a                | 1.2 a                  | 0.6 ab                 | 3.9 a                    |

Cultivar (A) ns ns * ns ** ns ** ns ** ns
Temperature (B) ns ns ns ns ** ns ** ns
A×B ns ns ns ns ns ns ns ns ns

Table 1. Effects of high temperature on the proportion of ripened rice grains, the 1000-grain weight, and the proportions of various types of white immature kernel.

| Cultivar    | Protein (%) | Amylose (%) | Amylographic characteristics (R.V.U.) | Textural characteristics |
|-------------|-------------|-------------|---------------------------------------|-------------------------|
|             | Max.visco.  | Min.visco.  | Break down | Final visco. | H –H | –H/–H |
| Hinohikari  | 5.6         | 17.3 d      | 344 a     | 171 ab       | 175 a | 288 bc |
|             | 5.6         | 14.8 b      | 407 bc    | 174 ab       | 253 bc | 280 ab |
| Nikomaru    | 5.4         | 17.5 d      | 343 a     | 175 ab       | 168 a | 301 d  |
|             | 5.4         | 15.2 b      | 391 bc    | 179 b        | 212 b | 293 cd |
| Chikushi64  | 5.3         | 16.2 c      | 379 b     | 170 ab       | 208 b | 286 bc |
|             | 5.4         | 14.1 a      | 419 c     | 165 a        | 254 c | 269 a  |

Cultivar (A) ns ** ns ns * ns ** ns
Temperature (B) ns ns ns ns ns * ns
A×B ns ns ns ns ns ns ns

Abbreviations: R.V.U.; rapid visco units, H; hardness, –H; adhesion, H/–H; hardness/adhesion. ** and * are significant at 1% and 5% levels, respectively; ns: not significant at 5% level. Values followed by the same letter within a column are not significantly different at 5% level, based on Tukey’s multiple range test.

Table 2. Effect of high temperature on protein and amylose contents, viscosity of milled rice, and on the texture of cooked rice.
‘Chikushi 64, ‘Nikomaru’ and ‘Hinohikari’. However, ‘Nikomaru’ and ‘Chikushi 64’ grown at 30ºC for 49 days during the grain development and ripening stages produced 85% and 89% ripened kernels and 4.4% and 3.9% white immature kernels, respectively (Table 1). In contrast, the proportions of ripened and white immature kernels in ‘Hinohikari’ grown at 30ºC were 92% and about 22%, respectively. These results show that a high temperature markedly enhanced the occurrence of white immature kernels in ‘Hinohikari’. Among those white immature kernels in ‘Hinohikari’, the proportions of white-based, white-backed, milky white and white-cored kernels were all higher at 30ºC.

Table 2 shows the physicochemical properties of the milled rice. The high temperature decreased the amylose content of milled rice, and the hardness (H) and the adhesion (-H, except for ‘Hinohikari’) of cooked rice, resulting a softer, less sticky texture. In addition, the maximum viscosity and breakdown values were higher, and the final viscosity was low in grains grown at 30ºC. A high temperature was previously reported to enhance the protein content of rice grains (Honjyo, 1971) but decrease the amylose content (Chamura et al., 1979; Yamakawa et al., 2007), and to increase the maximum viscosity and breakdown values (Maeshige, 1984). Our results generally support these previous results, but the protein contents of the milled rice ranged from 5.3% to 5.6% and did not differ significantly between the two temperatures.

Discussion

1. Features of dry weight, water content and water status of rice cultivars grown at a high temperature during the early ripening stage

The characteristics of ‘Nikomaru’ and ‘Chikushi 64’ grains indicated heat tolerance, whereas those of ‘Hinohikari’ clearly indicated heat sensitivity (Table 1). High temperature accelerates rice kernel development during the early ripening period, but interferes with development during the late ripening period, thereby decreasing grain weight somewhat (Matsushima and Manaka, 1957; Nagato and Ebata, 1965). The changes we observed in the dry weight of ‘Chikushi 64’ grown at a high temperature (Fig. 1A) were similar to those in the previous reports.

NMR microimaging clearly showed the area that generated a striped pattern of high signal intensity on the surface of the endosperm of rice grains at 10 and 15 DAF (Horigane et al., 2001). In the present study, ‘Nikomaru’, ‘Chikushi 64’, and ‘Hinohikari’ grains showed a close correlation with water content and dry matter accumulation at 14 DAF (Fig. 2), as was shown by Funaba et al. (2006). The transport of assimilates is associated with water movement in rice grains. For example, in previous research, a decrease in the NMR signal intensity in the endosperm of rice grains during development of the caryopsis corresponded to a change in the starch storage tissue from fluid to doughy, and a change in appearance from milky white to translucent (Horigane et al., 2001). The latter study also showed that water in the caryopsis was mostly distributed along the peripheral layer and in the pericarp vascular bundle of the grains before 15 DAF.

Table 3. Air temperature and duration of sunshine during the 14 days before and after flowering in 2004 and 2007.

| Year | Cultivar | Flowering Stage | 14 days before flowering | 14 days after flowering |
|------|----------|-----------------|--------------------------|------------------------|
|      |          |                 | Max.temp. (ºC) | Min.temp. (ºC) | Ave.temp. (ºC) | Sunshine (hour) | Sunshine (hour) |
| 2004 | Hinohikari | 29.Aug. | 31.9 | 25.3 | 28.0 | 68.9 | 46.5 |
| 2007 | Hinohikari | 29.Aug. | 33.8 | 26.6 | 29.6 | 122.5 | 63.8 |
|      | Nikomaru | 1.Sep. | 33.7 | 26.1 | 29.0 | 86.6 | 80.8 |
|      | Chikushi 64 | 19.Aug. | 34.5 | 26.7 | 30.1 | 128.7 | 78.4 |
|      | 30 years average | — | 31.4 | 24.3 | 27.4 | 85.0 | 80.9 |

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2. Relationship of histological characteristics to kernel quality in grains of heat-tolerant cultivars grown at a high temperature during the early ripening stage

Nagato et al. (1961) reported that the occurrence of “white-ridge” kernels was closely correlated with the adaptability of the rice cultivar to high temperature. We found few white immature kernels in ‘Nikomaru’
and ‘Chikushi 64’ grains, but these kernels accounted for 22% of the total in the heat-sensitive cultivar ‘Hinohikari’ when the plants were grown at 30°C for 49 DAF (Table 1). A high temperature has also been reported to cause loosely packed starch granules, decreased kernel weight, and thus enhanced occurrence of abnormal and chalky rice kernels (Resurreccion et al., 1977; Lisle et al., 2000). Grains with white, opaque parts in their cores or on the ventral sides of the endosperm (white-cored and white-belly rice kernels, respectively) have an unusually loose cell arrangement (Matsuda et al., 1988).

We found no influence of high temperature on the development of rice grains, especially of the nucellar epidermis, in ‘Nikomaru’ and ‘Chikushi 64’ (Fig. 3). However, ‘Hinohikari’ grown at 30°C for 14 DAF clearly revealed cessation of the development of the nucellar epidermis. Morphological studies of endosperm development in rice grains showed the transport of assimilates and water into the caropsis during this stage (Hoshikawa, 1967, 1972). Starch in the inner portion of grains of ‘Koshihikari’ was affected by the exposure to 32/27°C (day/night) during the early ripening stage (He et al., 1990). Furthermore, in situ localization of mRNA revealed that rice cell wall invertase 1 (OsCIN1) is produced preferentially in the vascular parenchyma of the dorsal vein, integument, and surrounding cells, and weakly in the nucellar projection and the nucellar epidermis (Hirose et al., 2002).

3. Influence of the unusually hot summer in 2007 on kernel quality of heat-sensitive cultivar, ‘Hinohikari’ grains

The high temperature influenced grain growth rate, grain weight, water status, anatomical features and kernel quality of ‘Hinohikari’. (Figs. 1-3). This tendency was observed in other rice cultivars and other cereals during the grain-filling stage (Hong et al., 1995; Lisle et al., 2000; Morita et al., 2005; Tahir et al., 2005). On the other hand, in the present study, the high temperature did not significantly affect the percentage of ripened grains and the 1000-grain weight of ‘Hinohikari’, ‘Chikushi 64’, or ‘Nikomaru’ grown in 2007 (Table 1). There was also no significant influence of high temperature on protein content (Table 2). In the study in 2004, the grains of ‘Hinohikari’ grown at 30°C had no perfect kernels and had more than 85% white-backed kernels (Funaba et al. 2006). However, the grains of the same cultivar grown in 2007 produced only about 22% white immature kernels, including white-backed kernels. Matsushima and Manaka (1957) reported that sunshine enhanced the recovery from kernel damage caused by heat stress. In 2004, the duration of sunshine for the 14 days before and after flowering of ‘Hinohikari’ was about 69 and 47 hr, respectively in 2004, and 123 and 64 hr, respectively, in 2007 (Table 3). The longer duration of sunshine in 2007 may have reduced the proportion of white immature kernels by some recovery mechanism. Furthermore, although the flowering date of ‘Hinohikari’ was 29 August in both 2004 and 2007, the mean air temperature during the 14 days before flowering was 28.0°C in 2004, and 29.6°C in 2007. The large difference in the kernel quality of heat-sensitive ‘Hinohikari’ suggests that the high temperature before flowering in 2007 induced acclimatization of the grains to the 30°C treatment. HSP family genes, HSP82 and HSP22, were markedly induced by ripening under 33/28°C treatment (Yamakawa et al., 2007). Furthermore, rhSP90 has been shown to play additional roles in various cellular signaling, such as heat stress signaling and protein transport/translocation via ER and/or chloroplast envelope in rice plants (Liu et al, 2006). Recently we found that heat shock (at 37°C for 2 hr) significantly increased the accumulation of ER-type ER-PDI and HSP90ER proteins in ‘Hinohikari’ seedlings (Tanaka et al., 2008). These results suggest that signal transduction to regulate expression of the genes for ER-specific heat-shock proteins (HSPs) was more sensitive to a high temperature than that for cytosolic HSPs.

In heat-sensitive rice cultivar, such as ‘Hinohikari’ grains grown under high temperature condition changed rapidly from a fluid state to doughy state, accompanied by marked lowering of water content as well as an accumulation of transported assimilates. Further studies are needed to clarify the function of HSP family induced by heat stress in rice grains to support efforts to improve rice kernel quality.

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** In Japanese with English summary.

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