Genotypic Variation in Morphological and Physiological Characteristics of Rice (*Oryza sativa* L.) under Aerobic Conditions

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Abstract: The aerobic rice system is a new promising water-saving cultivation technique; however, rice sensitivity to aerobic conditions limits its use. We investigated morphological and physiological responses of seedlings of two upland rice genotypes (Beodien and Sensho) and two lowland rice genotypes (KD18 and Koshihikari) to flooded condition (control) and three aerobic conditions (32, 22, and 14% soil moisture content (SMC), w/w) in 2013 and to a flooded condition and aerobic condition (32% SMC) in 2014. Under aerobic conditions, shoot growth was limited because of a reduction in water uptake. Water uptake capacity was highly controlled by root traits such as root dry weight and root length. Under aerobic conditions, leaf water potential, stomatal conductance, and root hydraulic conductance declined. We found that the 32% SMC condition was the most favorable aerobic condition whereas 22% and 14% SMC conditions strongly suppressed rice growth. Genotypic variations in physio-morphological traits were more clearly detected under well-irrigated conditions (32% SMC) than other two aerobic conditions. In both years, cultivar Sensho adapted to the 32% SMC condition, whereas Koshihikari did not adapt to the aerobic rice system, probably because of its limited root growth and lower root plasticity. The positive correlation between root traits and water uptake indicate that root traits are important for improving rice performance under an aerobic rice system.

Key words: Aerobic rice system, Genotypic variation, Lowland rice, Root system, Upland rice, Water regime.

Rice is the major staple food in Asia and provides 45%–70% of total caloric intake in many rice-consuming countries. Approximately 75% of rice supply comes from 93 million ha of irrigated lowland rice (IRRI, 2013). Therefore, irrigated lowland rice plays an important role in food security in Asia. However, with the increasing water scarcity worldwide, the sustainability of the irrigated lowland rice system is being threatened by water deficit. Various water-saving cultivation techniques have been developed to cope with water shortage, including the relatively new aerobic rice cultivation technique. In this technique, rice is grown in non-puddled and non-saturated soils, similar to wheat and maize cultivation (Bouman, 2001; Bouman et al., 2005), which could save water by minimizing seepage and percolation, and greatly reducing evaporation (Bouman et al., 2002). Therefore, this technique holds promise for farmers in the regions where water is too scarce or too expensive to grow flooded lowland rice (Bouman et al., 2005).

In irrigated lowland rice, a standing water layer is maintained in the field for most of the season when water and nutrient supply is favorable for rice growth. In aerobic rice, irrigation is applied to bring the soil water content up to field capacity when a lower threshold has been reached, such as halfway between field capacity and wilting point (Doorenbos and Pruitt, 1984; Bouman, 2001; Bouman et al., 2005); thus, rice plants might suffer a small water deficit between irrigation events (Okami et al., 2014). Rice is vulnerable to even a small loss of available water (Nguyen et al., 2009). Under aerobic conditions, stomata close and...
transpiration declines even when the soil moisture potential at the 20-cm depth is near field capacity (Kato and Okami, 2010, 2011). As a consequence, rice yield is considerably reduced in aerobic culture in comparison with flooded culture (Belder et al., 2005; Bouman et al., 2005; Peng et al., 2006; Matsuo and Mochizuki, 2009). New rice genotypes adapted to aerobic soil and occasional water deficit are required (Atlin et al., 2006). Therefore, studies of physiological and morphological responses of rice under aerobic conditions could provide necessary information for breeding programs.

The root system is an important trait for increasing yield under soil-related stresses (Serraj et al., 2004; Lynch, 2007). Rice is a shallow-root crop susceptible to drought (Yoshida and Hasegawa, 1982). Rice roots undergo morphological and anatomical changes to adapt to aerobic conditions (Kato and Katsura, 2014); in particular, root growth becomes restricted (Kato and Okami, 2010, 2011; Kato et al., 2010). Water absorption by the root system is determined by root hydraulic conductivity and total root length (or root surface area) (Kato and Okami, 2011; Kato and Katsura, 2014). Under aerobic conditions, total root length is considerably reduced due to down-regulation of adventitious root emergence and lateral root branching (Kato and Okami, 2011). Root hydraulic conductance tended to decrease under aerobic conditions (Matsuo et al., 2009). Restriction of water uptake capacity might make rice plants vulnerable to aerobic conditions (Kato and Okami, 2011). Therefore, rice roots need to be improved to enhance plant adaptation to aerobic rice system.

Aerobic rice could save water but there is a risk of poor performance (Kato et al. 2009) and yield penalty (Matsuo and Mochizuki, 2009) due to reduced water use in aerobic conditions (Belder et al., 2005; Kato and Okami, 2010). Therefore, the most favorable aerobic condition should be applied to mitigate these disadvantages. Some studies have been conducted on the morphological and physiological responses under mild water stress aerobic conditions (Matsuo et al., 2010), and under severe aerobic conditions (Belder et al., 2005; Bouman et al., 2005). However, the effect of soil moisture condition on growth of rice under an aerobic rice system has not been studied in detail. In this study, we conducted two experiments in two years, 2013 and 2014. In 2013, we grew rice under flooded and three aerobic conditions (including severe water stress, mild water stress and well-irrigated conditions) and detected the genotypic variation in physio-morphological traits under well-irrigated conditions. In the second year, we compared the morphological and physiological responses under two conditions: flooded and well-irrigated conditions. The results will help improve water management techniques as well as breeding of aerobic-adapted varieties.

Materials and Methods

1. Experimental design

All experiments were conducted in a greenhouse at the Experimental Farm of Kyushu University, Fukuoka, Japan (33°37' N, 130°27' E) in summer 2013 and summer 2014. The temperature, humidity, and solar radiation during the study period are shown in Fig. 1. In 2013, we used two *japonica* genotypes [Sensho (traditional upland) and Koshihikari (improved lowland)] and two Vietnamese *indica* rice genotypes [Beodien (traditional upland), which were used in a previous study (Matsuo and Mochizuki, 2009; Matsuo et al., 2010) and KD18 (improved lowland)], a popular paddy rice cultivar in Vietnam. Only Sensho and Koshihikari were used in 2014 because they showed the strongest differential responses in 2013.

In 2013, we used water regimes with three different soil moisture contents (w/w) (SMC), well-irrigated (32% SMC), mild water stress (22% SMC) and severe water stress (14% SMC), and flooded condition. In 2014, we used flooded and 32% SMC condition. In each regime, the soil was well mixed with water in a plastic vat, and the water potential of the soil sample was measured by using a Dew...
Point Microvoltmeter (HR-33T; Wescor, UT, USA). Soil water potentials were as follows: 0 kPa (control), > −1 kPa (32% SMC), −25 kPa (22% SMC), and −70 kPa (14% SMC).

The sowing dates were 11 June 2013 and 25 June 2014. Wagner pots 1/10000a (127 mm in diameter and 198 mm in height; ICM, Tsukuba, Japan) with 1 kg dried paddy soil [20.7% clay, 23.2% silt, and 56.1% sand (sandy clay–loam)] was used. We used small pots in order to create uniform soil moisture content in the whole pot to minimize effects of different root systems of different rice genotypes on water uptake ability. Soil had been sieved through a 10-mm mesh screen and pre-mixed with 2 g of chemical fertilizer (16% N:16% P2O5:16% K2O) 2 days before sowing for each pot. Seeds were pre-soaked for 3 days and sown (three seeds per pot); seedlings were thinned to one plant per pot and water treatment was started 5 days after sowing (DAS) in 2013 and 7 DAS in 2014. To avoid water evaporation from the soil surface, each pot was covered with a polystyrene board with a small hole to allow the seedling to grow. Hence, water uptake was approximately the same as plant transpiration. To maintain the target SMC, each pot was weighed and the amount of water lost was replenished every 2 days. The pots were arranged in a randomized complete design with 5 replications for each genotype per water regime.

2. Measurements and calculations

Rice seedlings were sampled at 1 month after sowing. Stomatal conductance ($g_s$) was recorded between 1100 and 1300 using an SC-1 Leaf Porometer (Decagon Devices Inc., Pullman, WA, USA). The chlorophyll index (SPAD) was determined by using a SPAD-502 chlorophyll meter (Spectrum Technology, Inc., Aurora, IL, USA). Root morphological traits (root length, root surface area, root volume, and root diameter) were analyzed by using Win
RHIZO system (Regent Instruments, Inc., Quebec City, Canada) which is interactive with image scanner (Epson Perfection V700/V750 2.80A, Epson, Long Beach, CA, USA) set to a resolution of 400 dpi. The lengths of fine roots (diameter < 0.2 mm) and thick roots (diameter $\geq 0.2$ mm) determined by Win RHIZO were used to calculate the fine to thick root ratio as a measure of the lateral to adventitious root length ratio (Kato and Okami, 2011).

Shoot dry weight and root dry weight were determined after drying shoots and roots in an oven at 80ºC for 3 days. Then total dry weight and root to shoot ratio were calculated. The water uptake rate was determined by weighing pots every 2 days; the amount of water uptake during the study period and water use efficiency (g total dry weight per kg water uptake) were determined.

At the end of the experiment, leaf water potential was measured (1300 – 1600) by using a pressure chamber (PMS Instruments, Albany, OR, USA). Briefly, the fully expanded youngest leaf from each plant was cut, covered with a plastic bag, and quickly placed inside the chamber with the cut end exposed outside the chamber. A piece of wet paper was placed at the bottom of the chamber to moisten the chamber air. Pressure was applied until sap appeared at the cut surface and the chamber pressure was recorded.

Root hydraulic conductance ($L_0$) was measured in 2014 as described by Matsuo et al. (2009) with some modifications. Briefly, one day before measurement the rice seedlings of similar height and tiller number were selected, pots were transferred to the laboratory and irrigated with tap water to saturation to enable the plants to regain turgor. Root hydraulic conductance was measured by using pressure chamber as described by Matsuo et al. (2009). The shoot was cut off with a razor blade and exuded sap was collected into pre-weighted cotton wools for 1 min at a pressure of 50, 75 and 100 kPa. The xylem sap flow rate $J_v$ (g cm$^{-2}$ s$^{-1}$) was calculated as $J_v = W/(RSA \times \Delta t)$; $L_0$ (g cm$^{-2}$ kPa$^{-1}$ s$^{-1}$) was then calculated as $L_0 = J_v/\Delta P$, where $\Delta P$ is the pressure difference between 50 or 75 and 100 kPa.

3. Data analysis
All statistical analyses were conducted with SYSTAT 13 (SYSTAT software, Inc., Washington, USA). The effects of genotype and water regime on the level of each trait were assessed by two-way ANOVA. Statistically significant differences between means were identified by a Tukey's test ($p < 0.05$).

Results

1. Climatic data
The climatic data was collected from 10 DAS until the end of the experiment. During these periods, the average temperature was lower in 2013 (28.3ºC) than in 2014 (29.2ºC); however solar radiation was higher in 2013 than in 2014, with values of 20.6 and 16.4 MJ m$^{-2}$ d$^{-1}$, respectively, due to more rainfall and cloudy days in 2014. It also caused higher average air humidity in 2014 (46.3%) than in 2013 (45.4%) (Fig. 1). Because of the lower solar radiation and higher air humidity, plants were less affected by water deficit in 2014 than in 2013.

2. Soil water content
In 2013 (Fig. 2A–D), we found no genotypic variation in the amount of water depleted from soil under the 22% and 14% SMC conditions. Under the 32% SMC condition, Sensho depleted the highest amount of soil water, followed by KD18. The upland genotype Beodien depleted less soil water than lowland KD18. Koshihikari depleted the least amount of soil water. At the end of the experiment (2 days after irrigation), Sensho depleted the largest amount of soil water (137.1 g), followed by KD18 (77.2 g), Beodien (53.7 g), and Koshihikari (36.9 g) under the 32% SMC condition (data not shown); soil moisture content
3. Effects of genotype and water regime on measured traits

Table 1 summarizes the ANOVA for the effects of genotype water regime and their interactions on all measured traits in 2013 and 2014. In 2013, significant effects of genotype were found in all traits except water use efficiency were observed while it was found that water decreased from 32% to 18% (Sensho), 23.9% (KD18), 26.3% (Beodien), and 28% (Koshihikari). In 2014 (Fig. 2E, F), 2 days after irrigation at the end of the experiment, soil water depletion by Sensho (162.5 g) was slightly greater than that by Koshihikari (149.7 g) under the 32% SMC condition.

![Figure 3](image)

Fig. 3. Genotypic variation in (A) shoot dry weight (SDW), (B) root dry weight (RDW), (C) total dry weight (TDW), (D) root to shoot ratio (RSR), (E) water uptake (WU), (F) water use efficiency (WUE), (G) leaf water potential (LWP) and (H) stomatal conductance ($g_s$) under flooded and aerobic conditions in 2013. Vertical bars indicate standard error. Bars with the same letter in each genotype are not significantly different at $p < 0.05$ by Tukey’s test ($n = 5$).
regime significantly affected all the traits. There were significant interactions between genotype and water regime in all traits except for water use efficiency. In 2014, the analysis showed significant effects of genotype on shoot dry weight, root dry weight, total dry weight, root to shoot ratio, root surface area, root volume and root diameter whereas water regime showed significant effects on all traits except root surface and total root length. The effects of genotype and water regime showed significant interactions in root dry weight, total dry weight, root to shoot ratio, root surface area and root volume.

4. Data obtained in 2013

1) Shoot and root growth
Shoot dry weight, root dry weight, total dry weight and root to shoot ratio in all genotypes are shown in Fig. 3 (A–D). In all genotypes, shoot dry weight was reduced gradually in the following order: flooded > 32% SMC > 22% SMC > 14% SMC. In 32% SMC, shoot dry weight of Sensho was reduced by 50.3% in comparison with flooded condition, whereas in other genotypes shoot dry weight decreased by 70.4% – 77.8%. Especially, under the same condition, Koshihikari was strongly affected with a decline of 77.8% in comparison with the flooded condition. Root dry weight was also reduced under aerobic conditions. Sensho was the least affected, whereas Koshihikari was the most affected by aerobic conditions. The lowland genotype KD18 had the same root dry weight as the upland genotype Beodien, except that it was lower in KD18 than in Beodien under the 14% SMC condition. The decline of root growth (32.1% – 88.1%) was less than that of shoot growth (50.3% – 90.3%). Total dry weight decreased with the same tendency as shoot dry weight and root dry weight; total dry weight was highest under the flooded condition and lowest under the 14% SMC condition. Water treatment had the smallest effect in Sensho and the greatest effect in Koshihikari. Aerobic conditions increased the root to shoot ratio in all genotypes except Koshihikari in comparison with the flooded condition.

2) Water uptake and water use efficiency
Water uptake was highest under the flooded condition and was reduced significantly under aerobic conditions in the following order: flooded > 32% SMC > 22% SMC > 14% SMC (Fig. 3E). Under aerobic conditions, water uptake by Koshihikari was the smallest; under the 14% SMC condition, water extraction was only 8.3% compared to flooded condition. Among all genotypes under aerobic conditions, Sensho had the highest ability to take up water under the 32% SMC condition and it was more than twice that of Koshihikari in the same condition. Water use efficiency increased with decreasing water uptake and total dry weight (Fig. 3F); in all genotypes, water uptake was lower (by 51.6% – 90.7%) under aerobic conditions compared to the flooded condition whereas water use efficiency increased up to 137.6%. Water use efficiency was highest under the 22% SMC and 14% SMC conditions. No significant differences in water use efficiency were detected among different genotypes (Table 1).

3) Leaf water potential, stomatal conductance, and SPAD index
Leaf water potential was highest under the flooded condition and declined with the reduction of soil moisture content (Fig. 3G). Under aerobic conditions, leaf water potential of KD18 was similar to that of Sensho and higher than that of Beodien and Koshihikari; in Koshihikari, leaf water potential strongly decreased. Stomatal conductance (g.) was highest under the flooded condition and was significantly reduced under aerobic conditions (Fig. 3H); stomatal conductance in Sensho and KD18 under the 32% SMC condition was higher than those under the 22% SMC and 14% SMC conditions, but there were no differences in the stomatal conductance in Beodien and Koshihikari under the aerobic conditions. The SPAD indices in all genotypes except Koshihikari under the flooded condition were higher than those under the aerobic conditions, but the differences were small (data not shown).

4) Root morphological traits
Root morphological traits are shown in Fig. 4. Root length was reduced under the 32% SMC and 22% SMC conditions in comparison with the flooded condition but had a tendency to increase under the 14% SMC condition in comparison with the 22% SMC condition. Root length in Beodien was nearly the same under the 14% SMC and flooded conditions. Under the 32% SMC condition, root length, root surface area and root volume were increased in Sensho, but decreased in Koshihikari, and the proportion of each trait under the 32% SMC condition to that under flooded condition was 72, 74 and 76%, respectively, in Sensho and 20, 18 and 16% respectively, in Koshihikari. Under aerobic conditions, rice root tended to be smaller and thinner and consequently root dry weight was lighter as compared with the flooded condition (Fig. 3). Under the 32% SMC condition, lateral root length and adventitious root length in Sensho were highest, followed by KD18, Beodien, and Koshihikari. Fine to thick root length ratio in Sensho Beodien and KD18 tended to decrease under the 32% and 22% SMC conditions, but was unchanged in Koshihikari. These data showed these genotypes had more thick roots than Koshihikari under these aerobic conditions.

5) Relationship between root-related parameters and plant growth and water uptake
A high significant positive correlation between water uptake and total dry weight was observed regardless of
water regime and genotype (Fig. 5A). The same trend was observed for the relationship between root dry weight and shoot dry weight (Fig. 5B). These data indicate that the development of the root system increased the water uptake ability to support shoot growth regardless of water regime.

Root dry weight, root length, root surface area and root volume were significantly positively correlated with water uptake in all water regimes in four genotypes (Fig. 5E–F). These results show that the root length and thickness may play a major role in water uptake under aerobic conditions.

Fig. 4. Genotypic variation in (A) root length (RL), (B) root surface area (RSA), (C) root volume (RV), (D) root diameter (RD), (E) fine root length (FRL), (F) thick root length (TRL) and (G) fine to thick root ratio (FTRR) under flooded and aerobic conditions in 2013. Vertical bars indicate standard error. Bars with the same letter in each genotype are not significantly different at $p < 0.05$ by Tukey’s test ($n = 5$).
conditions.

5. Data obtained in 2014

(1) Shoot and root growth

As compared with the flooded condition, shoot dry weight was 33% and 19% lower under the 32% SMC in Koshihikari and Sensho, respectively (Fig. 6A). Root dry weight was significantly higher in Sensho but not different in Koshihikari (Fig. 6B). Total dry weight was not different in Sensho, but significantly lower in Koshihikari (Fig. 6C). Root surface area was 183% and 132% higher under the 32% SMC condition in Sensho and Koshihikari, respectively (Fig. 6D). These trends were similar in both years; however, the differences were slightly smaller in 2014 than in 2013, probably due to the difference in weather conditions. More rainy days in 2014 led to higher air humidity (Fig. 1), which might have mitigated the effect of water deficit in soil on the growth of rice plants.

(2) Water uptake and water use efficiency

Under the 32% SMC condition, water uptake and water use efficiency were not significantly different from those under the flooded condition in Sensho, but significantly decreased in Koshihikari (Fig. 6E, F). In 2013, under the

![Fig. 5. Correlation of root related parameters with (A,B) plant growth and (C,D,E,F) water uptake (WU) in four rice genotypes under flooded and aerobic conditions in 2013. ** and *** indicate significance at p < 0.01 and p < 0.001, respectively.](image-url)
32% SMC condition, the amount of water uptake in Sensho was more than twice that in Koshihikari. No significant differences in water use efficiency were detected among different genotypes (Table 1).

32% SMC condition, the amount of water uptake in Sensho was more than twice that in Koshihikari. No significant differences in water use efficiency were detected among different genotypes (Table 1).

(3) Leaf water potential, stomatal conductance and SPAD index

In both genotypes leaf water potential and stomatal conductance were lower under the 32% SMS condition than the flooded condition. Sensho showed a stronger reduction in leaf water potential and stomatal conductance than Koshihikari (Fig. 6G, H). The SPAD index of Sensho
was not affected, whereas that of Koshihikari was slightly but significantly reduced (data not shown).

(4) Root morphological traits and root hydraulic conductance ($L_0$)

In both genotypes root length was shorter, under the 32% SMC condition than the flooded condition with less difference in Sensho (Fig. 7A). Root surface area was unaffected in Sensho, but was significantly reduced in Koshihikari (Fig. 7B). In Sensho, root volume was significantly higher under the 32% SMC condition, but not different in Koshihikari (Fig. 7C). Lateral root length

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**Fig. 7.** Genotypic variation in (A) root length (RL), (B) root surface area (RSA), (C) root volume (RV), (D) root diameter (RD), (E) fine root length (FRL), (F) thick root length (TRL), (G) fine to thick root ratio (FTRR) and (H) root hydraulic conductance ($L_0$) under flooded and aerobic conditions in 2014. Vertical bars indicate standard error. Bars with the same letter in each genotype are not significantly different at $p<0.05$ by Tukey's test ($n=5$).
was shorter under the 32% SMC condition in both genotypes; adventitious root length was slightly but significantly shorter in Koshihikari but not different in Sensho (Fig. 7E, F). Fine to thick root length ratio decreased in both genotypes (Fig. 7G). In both genotypes, root $L_0$ was lower under the 32% SMC condition but the reduction was 55% in Koshihikari and 41% in Sensho (Fig. 7H).

**Discussion**

In this 2-year study, we compared shoot and root characteristics of rice genotypes grown under aerobic and flooded conditions. In 2013, we found that shoot and root growth of all four genotypes studied was severely reduced under aerobic conditions in comparison with the flooded conditions, and that the decline of root growth was less pronounced than that of shoot growth (Fig. 3). None of the four genotypes could tolerate severe (14% SMC) or mild (22% SMC) water stress, and no clear genotypic variation was detected under these conditions. Under the well-irrigated aerobic (32% SMC) condition, upland rice Sensho adapted and grew better than the other genotypes, whereas lowland rice Koshihikari was most affected by water deficit. In 2014, we investigated in detail the difference in root growth between Sensho and Koshihikari under two water regimes: flooded condition and 32% SMC condition.

A large genotypic variation was observed in shoot and root growth in Sensho and Koshihikari under aerobic conditions. In 2013 under 32% SMC condition, up to 80% reduction in shoot dry weight and root dry weight was observed in Koshihikari, whereas these parameters in Sensho were less affected (a reduction of ~50% in shoot dry weight and 30% in root dry weight). In 2014 under the 32% SMC condition, shoot dry weight also decreased in both genotypes. However, root dry weight of Koshihikari was not affected, whereas that of Sensho was significantly increased (Fig. 6). Shoot growth was closely correlated with root growth, and with water uptake (Fig. 5); therefore, root growth restriction in Koshihikari may have limited shoot growth. Under water-limited conditions, root to shoot ratio increases (Banba and Ohkubo, 1981; Kondo et al., 2000; Singh et al. 2000; Price et al., 2002; Kato et al., 2006) because root growth is favored over shoot growth (Kato et al., 2006). In 2013, the root to shoot ratio in all genotypes except Koshihikari was higher under the 32% SMC condition than under the flooded condition (Fig. 3D). In Koshihikari, no change was observed, because the shoots and roots showed little growth. In 2014, the root to shoot ratio in both Sensho and Koshihikari was higher under the 32% SMC condition than under the flooded condition; the increase was higher in Sensho than in Koshihikari (Fig. 6D). These results imply that shoot and root growth of Koshihikari was more vulnerable to aerobic conditions than Sensho.

The ability to develop the root system in response to a changing environment, such as changes in soil moisture, reflects phenotypic plasticity (O’Toole and Bland, 1987; Yamauchi et al., 1996; Wang and Yamauchi, 2006; Kano et al., 2011). In 2013 under 32% SMC condition, root length, root surface area, root volume and root diameter were highest in Sensho and lowest in Koshihikari. The length of adventitious and lateral roots under the 32% SMC condition in Sensho was approximately four times that in Koshihikari (Fig. 4). In 2014 under the 32% SMC condition, the values of all root characteristics were higher in Sensho than in Koshihikari, but were nearly the same in both genotypes under the flooded condition. The length of adventitious roots did not change in Sensho but decreased in Koshihikari under 32% SMC condition in comparison with the flooded condition (Fig. 7). Therefore under the aerobic conditions, Koshihikari had fewer thick roots and a smaller root system. These results indicate that the root system of Koshihikari has less plasticity under aerobic conditions than that of Sensho.

Rice root characteristics significantly correlated with the water uptake capacity (Fig. 5C–F). In 2013 under the 32% SMC condition, water uptake in Sensho was about twice that of Koshihikari, which may be explained by differences in changes in root characteristics such as root dry weight and root length. Thick roots persist longer and produce more branches, which are also larger, and thereby increase root length density and water uptake capacity (Ingram et al., 1994). Matsuo et al. (2009) found that Sensho had thicker roots than Koshihikari in pot and hydroponic cultures. In the present study, root length, root surface area and root volume in Sensho under a well-irrigated condition were about four times those in Koshihikari under the same condition in 2013 (Fig. 4A–C). Root diameter in Sensho showed no significant difference between flooded condition and well-irrigated and mild stress conditions, but was reduced markedly under aerobic conditions in Koshihikari (Fig. 4D). Therefore, Sensho could have a greater ability to extract water and to maintain a high water potential because of more extensive root systems. As a consequence, Sensho grew vigorously under aerobic conditions and the yield was similar to that under the flooded condition; while Koshihikari could not grow well in aerobic rice systems and grain yield was 80% lower than that under the flooded condition (Matsuo and Mochizuki, 2009). Matsuo et al. (2009) also studied root hydraulic conductance of different rice genotypes under hydroponic and soil conditions, including mild water stress (control) and severe water stress (water-saving) condition, and found that root hydraulic conductance of Koshihikari was markedly reduced under control (32.2 times) and water-saving condition (92.5 times) compared to hydroponic condition while the reduction rate in Sensho
were 11.9 and 17.4 times. The reason may be repeated water stress might damage the root systems of Koshihikari, resulting in a small root hydraulic conductance, which in turn caused the low water uptake (Matsuo et al., 2009). In current study, we compared root hydraulic conductance in Sensho and Koshihikari between flooded and well-irrigated aerobic condition (non-water stress condition) and found that root hydraulic conductance in Sensho was higher than those of Koshihikari under both conditions, however, root hydraulic conductance in Koshihikari more severely decreased under the 32% SMC condition as compared to the flooded condition (Fig. 7H). The similar results between our study and Matsuo et al. (2009) indicate that root activity in Koshihikari was more sensitive to the soil water condition and was affected even under a favorable condition (32% SMC condition). These results suggest that Koshihikari may not be suitable for aerobic cultivation. Our results also demonstrate that a large root system might be a desirable trait under aerobic conditions.

Reducing water use and increasing water use efficiency have been the objectives of water-saving cultivation techniques in general and the aerobic rice system in particular (Borrel et al., 1997; Codon et al., 2004; Tuong et al., 2005). In 2013, water use was lower under aerobic conditions compared to flooded condition (Fig. 3E), whereas water use efficiency increased (Fig. 3F). However, increased water use efficiency might reduce photosynthesis rate (due to decreased transpiration rate and lower g, and thus reduce growth (Ceron et al., 2004), resulting in smaller plant or reducing their lifespan (Deivanai, 2012). In our experiments, higher water use efficiency and lower g, under aerobic than under flooded conditions resulted in a lower biomass production under aerobic conditions.

Blum (2009) argued that water use efficiency cannot be used as a measure of drought resistance or yield improvement under water-limited conditions. Thus, plant breeders should avoid using water use efficiency as a major trait for yield improvement in drought-prone environments (Blum, 2009). Our data that no genotypic variation was detected in water use efficiency agree with the study of Kobata et al. (2000). The similarity of water use efficiency values (calculated from the ratio of dry matter to transpiration) was due to the close relationship between dry biomass and water uptake (transpiration) (Fig. 5A).

In this study, the effects of different soil moisture contents on growth of rice seedling were clearly observed. It was showed by the consistent significant effects of water regime on almost the traits in both years, except for root surface area and total root length in 2014 (Table 1). The well-irrigated aerobic condition (32% SMC) was moistened and considerably favorable for upland rice Sensho, especially in 2014 when the climate condition was more humid, the growth of Sensho was little affected compared to flooded condition. While Koshihikari showed unadapted even under well-irrigated condition. Higher performance of Sensho compared to Koshihikari under this condition could be explained by more extended root system of Sensho in both years. The larger root system of Sensho helped this genotype absorb more water from soil and therefore grew better compared to Koshihikari under this condition. The mild and severe water stresses were harmful for growth of all genotypes since all parameters markedly decreased under these conditions. The results reveal that the 32% SMC condition could be taken into consideration in water management for an aerobic rice system.

Although aerobic rice is widely grown in China, Brazil, India, and the Philippines, the threshold at which irrigation is needed is still unknown. Some authors have reported that the threshold is halfway between field capacity and wilting point (Doorenbos and Pruitt, 1984; Bouman, 2001; Bouman et al., 2005) whereas others pointed out that it is when soil water tension at the 20-cm depth reaches ~30 kPa (Peng et al., 2006; Matsuo and Mochizuki, 2009). Our study suggests that none of the four genotypes tested can tolerate severe or even mild water stress, but under well-irrigated aerobic condition upland rice Sensho adapts and grows better than other genotypes. Further studies are needed to determine the threshold when irrigation is needed so that growth and development of rice plants are not affected and to search for genotypes without yield penalty under well-irrigated aerobic conditions.

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* In Japanese with English abstract.