Genotypic Variation in Biomass Production at the Early Vegetative Stage among Rice Cultivars Subjected to Deficient Soil Moisture Regimes and Its Association with Water Uptake Capacity

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Abstract: Genetic improvement in water uptake ability and/or water use efficiency (WUE) of rice cultivars is one option to enhance productivity under water-limited conditions. We examined the genotypic variation in biomass production among 70 rice cultivars (69 cultivars of NIAS global rice core collection and Azucena) under different soil moisture conditions, and to identify whether water uptake ability or WUE is responsible for the variation, if any. Two-week-old seedlings were transplanted into pots and grown for three weeks in an environmentally-regulated growth chamber under three soil moisture regimes: flooded (−0.02 MPa soil water potential) and two unflooded (−0.10 and −0.52 MPa) conditions. Substantial genotypic variations in total dry weight (TDW) were observed under all three regimes. Among all the cultivars tested, TDW was significantly correlated with water uptake ability, but not with WUE. However, several cultivars exhibited comparably higher WUE while showing superior biomass production under the −0.52 MPa regime. The amount of water uptake was significantly correlated with root dry weight among cultivars regardless of moisture regimes, while substantial genotypic difference in the amount of water uptake per unit root dry weight was observed. These results indicate that a marked genotypic difference exists in biomass production at the early vegetative growth under water-deficient conditions, and that this difference appears to be ascribed primarily to greater water uptake capacity, and additionally to higher WUE in drought-tolerant cultivars.

Key words: Rice Diversity Research Set of germplasm, Root formation, Soil moisture, Transpiration, Water uptake ability, Water use efficiency.

Currently, rice production is highly dependent on irrigated ecosystems, which provide 75% of global rice production (Maclean et al., 2002). It is anticipated that the continuous utilization of fresh water for rice production will be threatened by the erratic precipitation caused by global warming (IPCC, 2007) and the increasing competition for fresh water among industrial/urban users (Bouman et al., 2007). Thus, there is an imminent need for the development of novel production systems that enable more water-efficient rice production.

Productivity of rice under rain-fed cultivation is significantly lower than that under irrigated cultivation, largely due to the high sensitivity of rice to water shortage. The degree of yield and biomass reduction caused by water stress is influenced by environmental factors such as the strength and duration of drought, and genotypic differences in drought resistance (Kamoshita, 2011; Serraj et al., 2011).

Drought resistance of rice is considered to depend on the following capacities; drought escape, dehydration avoidance, dehydration tolerance, water use efficiency and drought recovery (Fukai and Cooper, 1995; Hall, 2000). Of these capacities, dehydration avoidance, which refers to the extent to which water content is maintained under drought, is recognized to be an important mechanism under drought conditions (Blum, 2009; Kamoshita, 2011). WUE, the ratio of biomass production to transpiration (Nguyen et al., 1997; Turner, 1997), can also provide an opportunity for improving drought resistance of rice.
cultivars (Tuong et al., 2005), although water uptake capacity was found to be more significant than WUE in determining biomass production under drought (Kobata et al., 1996; Nguyen et al., 1997; Blum, 2005).

Although previous studies found the cultivar difference in responsiveness to water deficit (Lilley and Ludlow, 1996; Courtois and Lafitte, 1999; Kamoshita et al., 2008), information on genotypic diversity among rice cultivars in growth and yield performance under water-limited conditions and the associated morphological and physiological traits is limited because the cultivars employed in previous studies do not cover a wide range of genetically diverse cultivars. Since there are a large number of rice cultivars worldwide, it is necessary to select appropriate cultivars that represent the diverse genetic background in the studies where genetic variation is the issue. For this reason, the Rice Diversity Research Set (RDRS) developed by the National Institute of Agrobiological Science (NIAS), which consists of 69 cultivars and holds 91% of the alleles identified in the representative 332 accessions selected from a global collection of rice cultivars (Kojima et al., 2005), has made possible wide and comprehensive evaluation of the genetic diversity of rice genetic resources.

The objectives of the present study, therefore, were to examine the genotypic variation in biomass production among 70 rice cultivars under different soil moisture conditions, and to identify whether water uptake ability or WUE is responsible for the variation, if any.

Materials and Methods

1. Plant materials

We used the RDRS, a global rice core collection developed by NIAS. This collection was developed based on a genome-wide RFLP polymorphism survey of 332 accessions, which were selected on the basis of the passport data of all collections stored at the Genebank of the NIAS, and holds 91% of the alleles identified in these accessions (Kojima et al., 2005). The RDRS consists of 69 cultivars, including 22 indica-I type cultivars, 33 indica-II type cultivars and 14 japonica type cultivars (personal communication with Dr. Ebana) collected from 19 countries. A japonica cultivar Azucena, which had been previously found to be highly drought-tolerant (Sangam et al., 2010), was added as a material.

Plants were grown in an environmentally regulated growth chamber: 12 h light with 450 μmol m−2 s−1 PPFD, day/night temperature of 25/20°C, and 70% relative humidity. Seeds were sown on a seedling tray then two-week-old seedlings were transplanted to 1/10,000-a Wagner pot with a diameter of 127 mm and a height of 198 mm (ICM, Tsukuba), one plant per pot. Each pot contained 1.4 kg of Andosol soil (pH (H2O) 4.7, CEC 28.4 cmolc kg−1, total carbon 50.8 g kg−1, total nitrogen 2.9 g kg−1 and phosphate absorption coefficient 2051 P2O5 g kg−1).

Mixed fertilizer was applied at a rate of 0.26, 0.26 and 0.26 g (N, P2O5 and K2O, respectively) per pot.

2. Soil moisture treatment

After transplanting, plants were subjected to one of three soil moisture regimes: a flooded condition (43%) and two unflooded conditions (33% and 25% (w/w)). For measurement of soil water content, soil samples form each moisture regime were dried at 130°C for 24 hours and soil water potential was estimated. The soil water potentials of each regime were −0.02, −0.10 and −0.52 MPa under the 43%, 33% and 25% soil moisture regimes, respectively. For determination of soil water potential, the soil of each regime was well mixed with water in plastic vat, and the water potential of the soil sample was measured using a dew point microvoltmeter (HR-33T, WESCOR, UT). For adjustment of soil moisture levels, pots were weighed each morning and water was added to maintain the respective soil moisture levels described above. The amount of water uptake was estimated from the daily reduction in pot weight.

3. Measurements

After three weeks of water treatment, the plants were sampled and separated into leaf, leaf sheath + stem and root. Leaf area was measured using an automatic area meter (AAM-9, Hayashi Denko, Tokyo). The crown root number was also counted. Then the samples were dried at 80°C for at least three days before weighing. WUE was calculated as follows:

\[ \text{WUE} = \frac{\text{TDW}}{\text{Water uptake}} \]

where TDW indicates total (shoot and root) dry matter weight and water uptake indicates cumulative amount of water uptake during treatment (for three weeks). For determination of the amount of evaporation from soil surface, we measured the weight change of the pots without plant in each moisture regime. Three replications in each cultivar in each moisture regime were employed.

ANOVA was applied to determine the significance of differences in traits between indica I, indica II and japonica types using JMP Statistical Discovery software (SAS Institute, Cary, NC). Spearman's rank correlation coefficients were also applied to determine the cultivar ranking for individual traits.

Results

1. Genotypic variation in biomass production

Under the three soil moisture regimes, a marked genotypic variation in total dry weight (TDW) was observed among the 70 cultivars. TDW ranged from 0.9 to 3.2 g under the −0.02 MPa regime, 1.2 to 3.4 g under the −0.10 MPa regime and 0.6 to 2.1 g under the −0.52 MPa regime, respectively (Fig. 1). The frequency distribution patterns were similar under the flooded and −0.10 MPa regimes, whereas that under the −0.52 MPa regime was shifted to
lighter weight with a substantially lower mean. The indica II type cultivars exhibited significantly higher TDW than the indica I type cultivars, and japonica type cultivars were intermediate of the two indica groups; the average TDW of the indica I type cultivars was 1.8, 1.9 and 1.1 g plant\(^{-1}\), that of indica II type cultivars was 2.2, 2.2, 1.3 g plant\(^{-1}\), and that of japonica type cultivars was 2.0, 2.0 and 1.3 g plant\(^{-1}\) under the −0.02 MPa (flooded) regime, −0.10 MPa and −0.52 MPa regimes, respectively.

Response patterns of the TDW of individual cultivars to each water regime are shown in Fig. 2. Of the 70 cultivars examined, 35 exhibited the highest value under the −0.02 MPa regime (flooded), while 35 did so under the −0.10 MPa regime. In most cultivars, TDW was the lightest under the −0.52 MPa regime, but there were some cultivars that exhibited similar or slightly heavier TDW values than under the flooded or −0.10 MPa regime. Spearman’s rank correlation coefficients among cultivars for TDW were significant between the two moisture regimes (P<0.01); they were 0.699, 0.538 and 0.554 between flooded and −0.10 MPa regimes, flooded and −0.52 MPa regimes, and −0.10 MPa and −0.52 MPa regimes, respectively.

A marked genotypic variation in WUE was observed among the 70 cultivars (Fig. 3B). WUE ranged from 4.7 to 8.1, 5.2 to 9.1 and 5.2 to 9.5 g kg\(^{-1}\) under the flooded, −0.10 and −0.52 MPa regimes, respectively. The distribution of WUE under the flooded regime was similar to that

Fig. 1. Genotypic variation in total dry weight among 70 rice cultivars subjected to three soil moisture regimes (flooded (−0.02) and two unflooded conditions (−0.10 and −0.52 MPa of soil water potential)). Number of cultivars vs the value of total dry weight with a range of 0.5 g plant\(^{-1}\) is shown. Triangles indicate the mean value of each moisture regime for indica I, indica II and japonica types. Means with different letters indicate significant differences at P<0.05 according to Tukey’s test.

Fig. 2. Response patterns of total dry weight of 70 individual rice cultivars subjected to three soil moisture regimes (flooded, −0.10 and −0.52 MPa). Bars indicate total dry weight of individual cultivars.

2. Water uptake ability and water use efficiency and their relationship with biomass production

Frequency distributions of the amount of water uptake per plant also varied with soil moisture regimes (Fig. 3A). The amount of the water ranged from 132 to 571 g, 152 to 518 g and 71 to 331 g under the flooded, −0.10 MPa and −0.52 MPa regimes, respectively. As in TDW, the distribution patterns were similar under the flooded and −0.10 MPa regimes, while the range was markedly narrower under the −0.52 MPa regime, which had the lowest mean. A significant difference in the average amount of water uptake per plant was observed between the two indica types regardless of soil moisture regime; the average of the water ranged from 132 to 571 g, 152 to 518 g and 71 to 331 g under the flooded, −0.10 MPa and −0.52 MPa regimes, respectively. In most cultivars, TDW was the lightest under the −0.52 MPa regime, but there were some cultivars that exhibited similar or slightly heavier TDW values than under the flooded or −0.10 MPa regime. Spearman’s rank correlation coefficients among cultivars for TDW were significant between the two moisture regimes (P<0.01); they were 0.699, 0.538 and 0.554 between flooded and −0.10 MPa regimes, flooded and −0.52 MPa regimes, and −0.10 MPa and −0.52 MPa regimes, respectively.

A marked genotypic variation in WUE was observed among the 70 cultivars (Fig. 3B). WUE ranged from 4.7 to 8.1, 5.2 to 9.1 and 5.2 to 9.5 g kg\(^{-1}\) under the flooded, −0.10 MPa and −0.52 MPa regimes, respectively. The distribution of WUE under the flooded regime was similar to that
under the −0.10 MPa regime, and no clear difference between the three groups was observed; the average WUE in indica I group, indica II group and japonica group was 6.5, 6.3 and 6.3 g kg\(^{-1}\) under the flooded regime, 6.8, 6.6 and 6.7 g kg\(^{-1}\) under the −0.10 MPa regime, respectively. In contrast, the distribution of indica I and indica II types under the −0.52 MPa regime shifted to a slightly higher range with a higher mean value than that of japonica type; the average was 7.2, 7.1 and 6.4 g kg\(^{-1}\) in indica I, indica II and japonica group, respectively. Top 20 cultivars with higher WUE under −0.52 MPa regime were all indica type cultivars.
TDW bore a positive correlation with water uptake regardless of soil moisture regime, and no clear differences were observed among moisture regimes in the slopes of regression lines (Fig. 4A). No significant correlation was observed between TDW and WUE (Fig. 4B).

3. Relationships between water uptake and root biomass

As shown in Fig. 5, root dry weight (RDW) was significantly correlated with shoot dry weight (SDW), and the regression line (SDW/RDW) became steeper in the following order: flooded > −0.10 MPa > −0.52 MPa regime, indicating that rice plants allocate more biomass to their roots when subjected to reduced water availability. The amount of water uptake was significantly correlated with RDW among the 70 cultivars (Fig. 6). The slope of regression lines varied with soil moisture regimes, being milder under the −0.52 MPa regime than the flooded and −0.10 MPa regimes.

The amount of water uptake per unit RDW varied with the rice type under the unflooded regimes; the average value was higher in the indica II and japonica types than in the indica I type (Fig. 7). Under the flooded regime, however, there was no marked difference in the distribution of RDW between the three types; the average amount of water uptake per unit RDW was 0.79, 0.86 and 0.86 kg H$_2$O g$^{-1}$ in the indica I, indica II and japonica types, respectively. Azucena exhibited the superior water uptake ability per unit RDW regardless of moisture regime, but especially under the −0.10 MPa regime.

4. Water uptake capacity and WUE of the cultivars that exhibited superior biomass production under unflooded regimes

Table 1 lists the top 20 cultivars that exhibited superior biomass production under the unflooded conditions...
### Table 1. Selected traits of top 20 cultivars that exhibited superior biomass production under the −0.10 MPa or −0.52 MPa regime.

| Soil moisture regime | Cultivar               | Indica/Japonica | TDW (g plant$^{-1}$) | The amount of water uptake$^a$ (g plant$^{-1}$) | WUE (g kg$^{-1}$) | SDW (g plant$^{-1}$) | Leaf area (cm$^2$ plant$^{-1}$) | RDW (g plant$^{-1}$) | Root number (plant$^{-1}$) |
|---------------------|-----------------------|-----------------|-----------------------|-----------------------------------------------|-----------------|------------------|-----------------------------|-----------------|--------------------------|
| −0.10 MPa           | Jaguary               | Japonica        | 3.4 ± 0.2             | 518 ± 40                                      | 6.6 ± 0.4       | 2.8 ± 0.2        | 498 ± 21                    | 0.7 ± 0.0        | 77 ± 3                    |
|                     | Puluik Arang          | Indica II       | 3.4 ± 0.1             | 464 ± 38                                      | 7.3 ± 0.6       | 2.6 ± 0.0        | 384 ± 31                    | 0.8 ± 0.1        | 87 ± 2                    |
|                     | Chin Galay            | Indica II       | 3.1 ± 0.6             | 481 ± 80                                      | 6.4 ± 0.3       | 2.5 ± 0.4        | 366 ± 58                    | 0.6 ± 0.2        | 85 ± 10                   |
|                     | Bei Khe               | Indica II       | 2.9 ± 0.1             | 463 ± 34                                      | 6.4 ± 0.4       | 2.4 ± 0.1        | 337 ± 17                    | 0.5 ± 0.0        | 93 ± 6                    |
|                     | Badari Dhan           | Indica I        | 2.8 ± 0.4             | 435 ± 83                                      | 7.0 ± 1.6       | 2.3 ± 0.4        | 322 ± 33                    | 0.6 ± 0.1        | 78 ± 9                    |
|                     | Blevio                | Indica II       | 2.8 ± 0.5             | 436 ± 77                                      | 6.4 ± 0.6       | 2.0 ± 0.3        | 325 ± 43                    | 0.7 ± 0.2        | 59 ± 2                    |
|                     | Bingala               | Indica II       | 2.8 ± 0.2             | 497 ± 54                                      | 5.6 ± 0.3       | 2.2 ± 0.2        | 423 ± 52                    | 0.6 ± 0.1        | 78 ± 4                    |
|                     | Deejaohualuo          | Indica II       | 3.2 ± 0.6             | 499 ± 61                                      | 6.1 ± 0.4       | 2.3 ± 0.4        | 310 ± 49                    | 0.5 ± 0.1        | 83 ± 6                    |
| −0.52 MPa           | Puluik Arang          | Indica II       | 2.1 ± 0.3             | 263 ± 39                                      | 8.2 ± 0.5       | 1.6 ± 0.2        | 236 ± 24                    | 0.6 ± 0.1        | 56 ± 8                    |
|                     | Jaguary               | Japonica        | 2.1 ± 0.2             | 330 ± 22                                      | 6.3 ± 0.2       | 1.5 ± 0.2        | 256 ± 33                    | 0.6 ± 0.0        | 50 ± 0                    |
|                     | Davao I               | Indica II       | 2.0 ± 0.1             | 273 ± 28                                      | 7.7 ± 1.1       | 1.5 ± 0.1        | 250 ± 10                    | 0.5 ± 0.1        | 66 ± 4                    |
|                     | Blevio                | Indica II       | 1.9 ± 0.4             | 284 ± 49                                      | 6.8 ± 0.8       | 1.2 ± 0.2        | 198 ± 29                    | 0.7 ± 0.1        | 39 ± 3                    |
|                     | Khao Nok              | Japonica        | 1.9 ± 0.2             | 272 ± 35                                      | 7.0 ± 0.4       | 1.3 ± 0.2        | 228 ± 28                    | 0.6 ± 0.1        | 26 ± 1                    |
|                     | Naba                  | Indica II       | 1.8 ± 0.2             | 302 ± 57                                      | 6.4 ± 0.9       | 1.3 ± 0.1        | 232 ± 14                    | 0.5 ± 0.0        | 64 ± 9                    |
|                     | Bingala               | Indica II       | 1.8 ± 0.1             | 277 ± 23                                      | 6.6 ± 0.3       | 1.3 ± 0.0        | 217 ± 14                    | 0.5 ± 0.1        | 55 ± 4                    |
|                     | Chin Galay            | Indica II       | 1.8 ± 0.5             | 216 ± 50                                      | 8.2 ± 0.5       | 1.3 ± 0.3        | 174 ± 41                    | 0.5 ± 0.1        | 54 ± 4                    |
|                     | Kemasin               | Indica II       | 1.7 ± 0.4             | 250 ± 34                                      | 6.8 ± 0.9       | 1.3 ± 0.3        | 187 ± 30                    | 0.4 ± 0.1        | 46 ± 5                    |
|                     | Tima                  | Japonica        | 1.7 ± 0.3             | 253 ± 23                                      | 6.7 ± 0.4       | 1.3 ± 0.2        | 167 ± 17                    | 0.4 ± 0.1        | 65 ± 4                    |
|                     | Hakhphayhav           | Indica II       | 1.7 ± 0.2             | 225 ± 10                                      | 7.5 ± 0.7       | 1.2 ± 0.1        | 173 ± 16                    | 0.5 ± 0.0        | 57 ± 1                    |
|                     | Ryou Susan Koumai     | Indica II       | 1.7 ± 0.1             | 276 ± 37                                      | 6.2 ± 0.7       | 1.2 ± 0.1        | 176 ± 17                    | 0.5 ± 0.0        | 52 ± 3                    |
|                     | Padi Perak            | Japonica        | 1.7 ± 0.1             | 263 ± 40                                      | 6.5 ± 0.5       | 1.2 ± 0.1        | 185 ± 17                    | 0.5 ± 0.0        | 37 ± 2                    |
|                     | Vandaran              | Indica II       | 1.6 ± 0.5             | 237 ± 56                                      | 6.6 ± 0.5       | 1.1 ± 0.4        | 163 ± 35                    | 0.5 ± 0.1        | 43 ± 2                    |
|                     | Nepal 555             | Indica I        | 1.6 ± 0.3             | 210 ± 42                                      | 7.5 ± 0.3       | 1.2 ± 0.2        | 218 ± 35                    | 0.4 ± 0.1        | 42 ± 5                    |
|                     | Calotoc               | Indica I        | 1.5 ± 0.1             | 248 ± 25                                      | 6.2 ± 0.3       | 1.1 ± 0.1        | 183 ± 20                    | 0.4 ± 0.0        | 49 ± 4                    |
|                     | Radin Goi Sesat       | Indica II       | 1.5 ± 0.1             | 217 ± 24                                      | 6.9 ± 0.4       | 1.0 ± 0.0        | 139 ± 13                    | 0.5 ± 0.1        | 47 ± 4                    |
|                     | Deng Pao Zhai         | Indica II       | 1.5 ± 0.2             | 255 ± 30                                      | 5.7 ± 0.2       | 1.1 ± 0.2        | 184 ± 12                    | 0.3 ± 0.1        | 76 ± 8                    |
|                     | Azucena               | Japonica        | 1.5 ± 0.1             | 331 ± 21                                      | 4.5 ± 0.2       | 1.1 ± 0.0        | 183 ± 9                     | 0.4 ± 0.0        | 24 ± 1                    |
|                     | Surjamukhi            | Indica I        | 1.4 ± 0.1             | 239 ± 17                                      | 6.1 ± 0.7       | 1.0 ± 0.1        | 174 ± 17                    | 0.5 ± 0.0        | 27 ± 1                    |
| Mean                |                      |                 | 2.7 ± 0.3             | 424 ± 63                                      | 6.3 ± 0.3       | 2.1 ± 0.1        | 334 ± 51                    | 0.5 ± 0.1        | 75 ± 5                    |
| LSD (P=0.05)        |                      |                 | 0.9 ± 0.0             | 154 ± 14                                      | 1.5 ± 0.7       | 0.7 ± 0.1        | 107 ± 22                    | 0.2 ± 0.0        | 21 ± 2                    |

Means ± SE are shown. Underlined cultivars appear in both regimes.

$^a$: Cumulative amount of water uptake during soil moisture treatment (for three weeks).
that WUE was an additional trait for achieving superior than the average value of the top 20 cultivars, suggesting the top 10 cultivars that exhibited superior TDW under the −0.52 MPa regime, seven cultivars exhibited higher WUE regime. For example, Puluik Arang exhibited the greatest associated with the larger amount of water uptake, whereas showed higher RDW, leaf area, and water uptake per RDW. Under the −0.10 MPa regime, TDW of these cultivars was the largest amount of water uptake among the cultivars. Among TDW under the −0.52 MPa regime did not exhibit the effects of soil water deficit were examined under field conditions, the water potential of −0.10 MPa appeared to be a moisture level to give a considerable water stress, and the soil moisture treatment with ca. −30 kPa significantly reduced grain yield of rice (Zhang et al., 2009). In the present study, we grew the plants in growth chamber in which environmental conditions (light, temperature, humidity etc.) were regulated within an optimum range, whereas these environmental attributes can substantially fluctuate under the field conditions. Therefore, the −0.10 MPa regime did not appear to be a condition that adversely affected biomass production for many of the cultivars examined in the present study.

Matsuo and Mochizuki (2009) examined the genotypic differences in growth and yield under several water managements and found that the upland cultivars maintained or increased grain yield under aerobic conditions (the soil water potentials were −15 and −30 kPa). Similarly, previous studies conducted in the field observed superior growth and yield in upland conditions compared to flooded conditions (Kato et al., 2006b; Matsunami et al., 2009, 2010; Nakaide and Katsura, 2010). In these previous studies, upland rice cultivars showed reduction of grain yield under flooded conditions, suggesting the involvement of physiological differences between lowland and upland rice cultivars in response to different soil moisture conditions. Even though the cultivars used in the present study were mostly lowland cultivars (according to the passport data provided by NIAS, except for Urasan1 (WRC51; japonica type) (NIAS, 2010) and Azucena, about half exhibited superior growth under the upland condition (−0.10 MPa regime) compared to the flooded condition. Thus, it is suggested that many lowland cultivars have high adaptability to upland conditions, as long as adequate soil moisture is maintained, comparable to that under flooded conditions. In this regard, Ichwantoari et al. (1989) found that there were no differences in the response to soil moisture levels between lowland and upland cultivars.

A comparison of the average TDW among the three types (indica I, indica II and japonica) revealed that indica II type cultivars exhibited substantially heavier TDW than the other two types under the different soil moisture conditions (Fig. 1). In previous studies in which drought tolerance of indica was compared with that of japonica types, the results were not consistent; Lilley and Ludlow (1996) evaluated indica types as possessing better dehydration tolerance, while Courtois and Lafitte (1999) found that within-subspecies, variation tended to be as great as the between-subspecies variation.

Since the distribution pattern of TDW under the flooded regime was similar to that under the −0.10 MPa regime (a moisture condition comparable to wet upland), it appears that in most cultivars examined, plants grown under the −0.10 MPa water regime were not water stressed. In fact, about half of the cultivars exhibited superior growth under the −0.10 MPa regime compared to the flooded regime (Fig. 2). When compared with previous studies in which the effects of soil water deficit were examined under field conditions, the water potential of −0.10 MPa appeared to be a moisture level to give a considerable water stress, and the soil moisture treatment with ca. −30 kPa significantly reduced grain yield of rice (Zhang et al., 2009). In the present study, we grew the plants in growth chamber in which environmental conditions (light, temperature, humidity etc.) were regulated within an optimum range, whereas these environmental attributes can substantially fluctuate under the field conditions. Therefore, the −0.10 MPa regime did not appear to be a condition that adversely affected biomass production for many of the cultivars examined in the present study.

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### Discussion

#### 1. Genotypic diversity of biomass production among rice cultivars grown under different soil moisture regimes

The primary objective of this study was to evaluate the diversity of biomass production among rice cultivars subjected to different soil moisture conditions. The results revealed a marked genotypic variation in TDW among the 70 cultivars (Fig. 1). Moreover, the genotypic variation was substantial in all three soil moisture regimes ranging from −0.02 MPa (flooded) to −0.52 MPa; the cultivar difference in TDW (ratio of the maximum to minimum value) amounted to 3.6, 2.8 and 3.5 times under the flooded, −0.10 MPa and −0.52 MPa regimes, respectively. Particularly, the variation in TDW of cultivars subjected to the most critically deficient moisture condition (−0.52 MPa) was comparable to that of those under the flooded condition, suggesting that there is a great possibility to exploit the genes available in existing cultivars for rice breeding programs in which vigorous early growth under water-deficient conditions is the main breeding target.

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#### 2. Relationship between biomass production, water uptake ability and WUE under different soil moisture conditions

The other objective of this study was to identify whether water uptake ability or WUE is responsible for the variation, if any. Previous studies revealed that the cultivar difference in biomass production under water-limited conditions was associated with water uptake ability, but not with WUE (Kobata et al., 2000; Wade et al., 2000). In
contrast, Peng et al. (1998) examined the transpiration efficiency (the ratio of photosynthesis to transpiration) of seven indica and seven japonica cultivars under irrigated conditions, and found that the efficiencies of tropical japonica type cultivars were 25-30% higher than those of indica type cultivars, due to the reduction in transpiration, suggesting that WUE may be responsible for cultivar difference in biomass production.

Marked genotypic differences in water uptake ability among cultivars were observed under the three soil moisture regimes (Fig. 3A), which was associated with the genotypic variation in TDW (Fig. 4A). Despite the substantial cultivar difference in WUE under unflooded conditions (Fig. 3B), WUE was not correlated with TDW (Fig. 4B). Therefore it is likely that water uptake ability rather than WUE is primarily responsible for the genotypic variation in TDW. Similarly, Kobata et al. (1996) indicated that dry matter production ability of rice under drought conditions was caused by a high ability of transpiration (≒ water uptake), but not by a high WUE.

Generally, deficient soil moisture causes a reduction of transpiration, resulting in an increase in WUE without increasing biomass production (Nguyen et al., 1997; Blum, 2005). However, a comparison among all the 70 cultivars of the relationship between the amount of water uptake and WUE revealed the contrasting variation patterns among the soil moisture regimes (Fig. 8). The most deficient soil moisture regime (−0.52 MPa) resulted in the greatest variation in WUE with the smallest variation in the amount of water uptake, whereas the flooded and the unflooded with relatively wet soil moisture regime (−0.10 MPa) conditions exhibited a greater variation in the amount of water uptake with smaller variation of WUE. These results indicate that TDW, the product of the amount of water uptake and WUE, is primarily dependent on the amount of water uptake under adequate soil moisture conditions (flooded and −0.10 MPa), but it is considerably dependent on WUE as well under the water-deficient condition (−0.52 MPa).

The cultivars that exhibited superior TDW under the unflooded conditions had a higher water uptake ability and/or WUE (Table 1). For example, Shwe Nang Gyi (indica II) and Radin Goi Sesat (indica II) under the −0.10 MPa regime, and Chin Galay (indica II) and Nepal 555 (indica I) under the −0.52 MPa regime were found to have higher WUE but lower water uptake ability. In contrast, Bingala (indica II) and Shusuonshu (indica II) under the −0.10 MPa regime, Jaguary (japonica) and Azucena (japonica) under the −0.52 MPa regime exhibited comparably higher water uptake ability with lower WUE. It is notable that there existed several cultivars which showed superior water uptake ability and WUE. Pululik Arang (indica II) under the two unflooded regimes, and Badari Dhan (indica I) under the −0.10 MPa regime and Davao 1 (indica II) under the −0.52 MPa regime were found to be this type of cultivars. Thus, in breeding programs aimed at enhancing the genetic adaptability of rice cultivars to water-limited conditions, water uptake capacity should be a primary target trait in most cases, although WUE should also be a target in specific cultivars as well.

3. Relationship between water uptake ability, WUE and morphological traits

RDW was positively correlated with SDW among all the cultivars tested under the three soil moisture regimes (Fig. 5). There existed several cultivars that exhibited RDW under the −0.52 MPa regime comparable to that under the flooded or the −0.10 MPa regime, whereas SDW under −0.52 MPa generally declined to ca. 50% of that under the flooded or −0.10 MPa regime (Fig. 5). This result suggests that, under the water-deficient conditions, genotypic variation among rice cultivars is expressed more substantially in root growth than in shoot growth, probably due to the greater adaptability of root growth to water-deficient conditions.
conditions. Regardless of soil moisture regimes, RDW was significantly correlated with the amount of water uptake (Fig. 6). It is remarkable that there existed wide genotypic differences in the amount of water uptake per unit RDW (Fig. 7). Specifically, Azucena, a drought tolerant cultivar, exhibited greater water uptake per RDW in all the moisture regimes. The amount of water uptake of this cultivar under the −0.52 MPa regime was the greatest among all the cultivars examined, while its RDW was relatively light compared with the other cultivars (Table 1).

Water uptake by plants is strongly dependent on the morphological and physiological attributes of roots. Regarding rice root morphology, which is adaptable to flooded conditions, Yamauchi et al. (1987, 1988) speculated that those cultivars with a concentrated type of root system, which results in a large number of short individual crown roots, might be more adaptable to flooded conditions, because such a root system might be capable of supplying adequate oxygen to the root apex which requires a large amount of oxygen for cell division. In contrast, root penetration into a deeper soil layer (Araki and Iijima, 1998; Hirasawa et al., 1998; Kato et al., 2006a, 2007), or lateral root development (Kano et al., 2006; Suralta et al., 2008), was indicated as a critical root trait for adaptability to water-deficient conditions. Furthermore, the plasticity of root development was emphasized as an important trait for the adaptability to soil moisture changes (Kano et al., 2011). In this study, root number did not seem to be correlated with TDW among the top 20 cultivars that exhibited superior biomass production under the −0.10 and −0.52 MPa regimes (Table 1). Thus the quantity of root of these drought-tolerant cultivars did not appear to be associated with biomass production and water uptake ability under the water-deficient conditions. As described above, the morphology of individual crown roots (lateral root development and/or root length) probably plays an important role in water uptake ability under water-deficient conditions. In this regard, Gowda et al. (2011) emphasized, from a review of previous studies on root biology and its relation to drought, the importance of further understanding of root function and systems for improving water uptake capacity, hence achieving the drought avoidance in rice. Further studies are needed to identify the morphological and/or physiological root attributes regulating the water uptake ability per unit root length/volume under the water-deficient conditions.

Shoot traits may also play important roles in determining biomass production under water-deficient conditions. Leaf area was positively correlated with biomass production (TDW) and the cumulative amount of water uptake regardless of soil moisture regimes in our previous study (Matsunami et al., 2010). The capacity of leaf expansion was previously identified to be one of the important traits to maintain or increase the biomass production under upland conditions, because leaf expansion directly affects biomass production through photosynthesis and water uptake through transpiration. For example, Okami et al. (2008) found a significant cultivar difference in leaf expansion capacity among several upland and lowland rice cultivars under the irrigated upland condition, and a positive correlation between their leaf area and biomass production. They also found, in another study in which cultivar differences were examined under water-deficient conditions, that an increase in leaf area was regulated by an increase either in the area of individual leaves or in tiller number, depending on the cultivar (Okami et al., 2010).

Although further studies are necessary for precise identification of phenological, morphological, and physiological traits that contribute to adaptability to water-deficient conditions, our results indicate that a marked genotypic difference exists in biomass production at the early vegetative growth under water-deficient conditions, and that this difference may be ascribed primarily to greater water uptake capacity, and additionally to higher WUE in the drought-tolerant cultivars.

Acknowledgements

We thank Mr. H. Matsuda (Akita Prefectural University) for performing chemical analysis of the soil and Dr. K. Ebana (NIAS) for providing the information on the classification of RDRS. This work was supported in part by the Japanese Society for the Promotion of Science. Special thanks go to the late Dr. S. Masaki (Agricultural Experiment Station, Akita Prefectural Agriculture, Forestry and Fisheries Research Center) for his helpful advice on the implementation of this study.

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