Growth and Water Use Response of Doubled-Haploid Rice Lines to Drought and Rewatering during the Vegetative Stage

Joel DLC. Siopongco, Akira Yamauchi, Hosseini Salekdeh, John Bennett and Len J. Wade

Abstract: We examined the responses of doubled-haploid lines (DHLs) of rice (Oryza sativa L.) to drought and rewetting in controlled rainfed lowland conditions, to test the hypothesis that the DHLs would permit trait comparisons with less confounding by unrelated traits than had been reported previously. IR62266 and four DHLs derived from the cross between IR62266 and CT9993 (DHL-32, 51, 54 and 79) were grown in pot experiments in the greenhouse at the IRRI, Los Baños, Philippines. Genotypic variation in leaf and tiller development, transpiration, water use efficiency, osmotic adjustment and leaf water potential was examined in relation to dry matter production. Results revealed that greater seedling vigor through continued leaf expansion in early drought was associated with greater dry matter production after rewetting. A higher water use efficiency was related to a greater increase in dry matter production during drought. Leaf water potential was correlated strongly with dry weight, not only during drought, but especially on rewetting. Therefore, we found that the ability to continue leaf expansion, higher water use efficiency, and a greater osmotic adjustment for maintenance of leaf water potential as drought progressed were desirable traits for improved performance under drought and improved ability to recover on rewetting. These relationships could be analyzed precisely using such genetically-related materials as DHLs, with less confounding effects of plant size and genetic background.

Key words: Drought, Drought recovery, Osmotic adjustment, Rainfed lowland, Rice, Water potential, Water use efficiency.

The rainfed lowland rice crop is grown in bunded fields occupying about 46 Mha in South and Southeast Asia, contributing about a third of the world rice area (MacLean et al., 2002). Because rain is the main source of water in the rainfed lowlands, drought is the major constraint to rice productivity, which averages 2.3 t ha\(^{-1}\). This is exacerbated by soil transitions from flooded and anaerobic to droughted and aerobic conditions (Wade et al., 1999), which also impacts on nutrient response (Wade et al., 1998).

Traits conferring tolerance to such adverse conditions may be better examined for responses in the greenhouse, in controlled conditions designed to closely follow the constraints faced by plants in the field. Wade et al. (2000), Azhiri-Sigari et al. (2000) and Kamoshita et al. (2000) developed a robust methodology for this purpose. Their studies, which included CT9993 and IR62226, which are parents of a regularly-studied population of doubled haploid lines (DHLs), demonstrated genotypic variation among cultivars examined in seedling vigor, characterized by robust tillering, leaf area expansion, root growth and water extraction at depth during late drought, recovery ability on rewetting, and osmotic adjustment (OA). These drought and recovery treatments simulated rainfed lowland soil drying and wetting patterns. Kamoshita et al. (2004) then argued that cultivars with enhanced seedling vigor and more rapid root exploration and water extraction from depth may be advantageous, as long as there were reserves in the subsoil and the drought was relieved before the water was exhausted. They concluded that selection for drought recovery ability should be advantageous for early season drought.

Osmotic adjustment is influenced by root traits that modify access to additional water resources, either from soil layers already explored or from deeper soil layers (Fukai and Cooper, 1995). Plant size influences...
these traits via radiation intercepted and the resulting heat load that drives evaporative demand. So for two plants with identical ability in capacity for OA, more water tends to be lost from the larger plant. This process is modified by stomatal control, perhaps under the influence of root signals emanating from root tips in drying soils, which are expected to be numerous in shallow soil layers of rainfed lowland rice (Fukai and Cooper, 1995). Consequently, the expression of traits for leaf water potential ($\Psi_L$) and OA, their consequences for maintenance of dry weight and leaf area as drought progresses, and for subsequent recovery on rewatering, are subject not only to resource availability (e.g. plant available water extraction from soil), but also to modification of water loss from the canopy via leaf area and stomatal regulation, which in turn are modified by plant size and genetic background.

In this aspect, cultivars were reported to vary in OA (Lilley and Ludlow, 1996; Zhang et al., 1999; Kamoshita et al., 2000; Jongdee et al., 2002), but no relationship with dry matter production in drought has yet been established (Pantuwan et al., 2002c; Kamoshita et al., 2004), despite reports in other species (Zhang et al., 1999). In contrast, an ability to maintain $\Psi_L$ as drought progressed seemed to reliably indicate cultivars that were more stable under drought conditions in rainfed lowland (Jongdee et al., 2002). However, whether such maintenance of $\Psi_L$ arose from improved water extraction via better root systems or from maintenance of cell turgor during partial dehydration via osmotic adjustment was not clarified. Such traits are highly interactive, not only with prevailing conditions, but also with plant size and genetic background.

In addition, water use (transpiration) and water use efficiency (WUE) in relation to dry matter production are also considered important agronomic characters for drought tolerance in crops (Blum, 1993). There is increasing interest in improving WUE of crop cultivars so that plants can grow and yield well under water deficient conditions (Boyer, 1996). In rainfed lowland rice, Wade et al. (2000) reported WUE as mildly contributing to higher plant growth rate in drought periods, and emphasized a need for further study as to how WUE contributes to dry matter production in rainfed lowland rice.

Most of the previous studies described above, however, have studied the responses of rainfed lowland rice to drought and rewatering by comparing cultivars differing widely in phenology, plant size and genetic background. These background traits are expected to confound the expression of traits more directly associated with improved adaptation to drought and recovery in rainfed lowland conditions. Difficulty in phenotyping, for instance, was recognized by Kamoshita et al. (2002a) as the greatest challenge in correctly identifying quantitative trait loci in abiotic stress, where both constitutive and adaptive traits are present, and each interacts with both the prevailing conditions and genetic background (Kamoshita et al., 2002b). Consequently, the research focus is shifting to studies involving improved genetic materials such as DHLs (Sarkarung et al., 1997) and recombinant inbred lines (Jongdee et al., 2002) so that traits conferring improved adaptation to drought can be studied with less confounding by genetic background. In this aspect, Siopongco et al. (2005) examined variation among DHLs in root traits, water extraction and the relationship between them, for rice grown under simulated rainfed lowland conditions in the greenhouse and were able to show the critical root density for water extraction that is different from that of other rice ecotypes.

In this study, we examined responses to drought and rewatering by using DHLs of rice in controlled greenhouse conditions designed to mimic conditions in rainfed lowland rice fields. We propose that the DHLs permit trait relationships to be examined more closely, with less confounding by various morphological traits such as plant size because of closer genetic background. Here, we seek to relate tiller production, leaf expansion, water use, water use efficiency, leaf water potential and osmotic adjustment to dry matter production during drought, and their subsequent recovery on rewatering. We conclude by considering traits conferring an adaptive advantage in drought and in recovery after rewatering, and the implications for genetic improvement and selection of better cultivars. Comparisons are also made with recent field evidence.

Materials and method

1. Cultural details

Drought-stress treatments followed the greenhouse protocol established by Wade et al. (2000), with cultural details as described in Siopongco et al. (2005). Briefly, two experiments were conducted in 2000 in the greenhouse at the International Rice Research Institute (IRRI), Los Baños, Philippines (14°11’ north, 121°15’ east, 23 m altitude). A split-plot design with three replicates was used with two water regimes (well-watered and no irrigation) as main plots and five rice genotypes as subplots; IR62266-42-6-2 (IR62266) and four DHLs: IR68586-F2-CA-51 (DHL-51), IR68586-F2-CA-54 (DHL-54), IR68586-F2-CA-79 (DHL-79) and IR68586-F2-CA-92 (DHL-32). These DHLs were chosen based on research by Kamoshita et al. (2002a) in the greenhouse and Babu et al. (2003) in the field, where these DHLs differed in root traits and osmotic adjustment.

Details of soil and pot preparation, planting and environmental conditions during the experiments were presented in Siopongco et al. (2005). Briefly, the first experiment was conducted from 28 February to 25 April 2000 and the second from 24 June to 22 August
Siopongco et al. — Growth and Water Use of Rice DHLs under Drought and Rewatering

Pre-germinated seed was sown after puddling in PVC pots of 20-cm internal diameter and 55-cm height containing 23 kg of sandy loam soil. From 14 days after sowing (DAS), 2-cm of ponded water was maintained. In the drought-rewatered treatment, pots were drained at 21 DAS, water was withheld until about 4 kg of water was lost by transpiration, then pots were rewatered to maintain 2-cm of ponded water.

2. Measurements

(1) Transpiration and plant sampling

Daily transpiration was calculated from 21 DAS until the end of the experiments by measuring loss in pot weight in drought and water added in well-watered treatments. Cumulative transpiration was calculated as the sum of daily increments in each water regime.

Plants were sampled at 21 DAS, before water was withheld from drought treatments, between 31 and 34 DAS, when cumulative transpiration was about 2.0 kg, between 40 and 48 DAS when cumulative transpiration was about 4.0 kg, and between 50 and 58 DAS, at 10 days after the previous sampling.

To assess plant response during each stage of drought development and rewatering during the experiment, growth periods were divided into the early drought period (between samples 1 and 2), late drought period (between samples 2 and 3), and rewatering period (between samples 3 and 4).

Water use efficiency and transpiration rate were calculated during each drought period, from actual change in dry weight and in water use for each period. Plants in both water regimes were sampled at the same time. In experiment 2, additional sampling, which will be referred as the intermediate drought period, was done when cumulative transpiration was about 3.0 kg, between 36 and 39 DAS.

At each sampling occasion, tillers were counted and leaf area was measured using the Li-COR LI-3100 (LI-COR, Lincoln, NE, USA) area meter.

(2) Plant water status

At 33, 38, 43 and 53 DAS, $\Psi_L$ and osmotic potential at full turgor were measured at pre-dawn only for the second experiment. One leaf blade of the second-youngest fully expanded leaf on the main stem or on a primary tiller with similar size was inserted into a long plastic bag, severed just below the ligule, and covered with a wet cloth. The leaf blade was immediately taken to the pressure chamber (Soil Moisture Equipment Corporation, Santa Barbara, CA, USA) for measurement of $\Psi_L$. Another leaf blade of the second-youngest fully expanded leaf was taken in the same way, soaked in water in the refrigerator for 24 hours to

Fig. 1. Time course of shoot dry weight (g plant$^{-1}$) in well-watered (a, c) and stress (b, d) treatments of experiments 1 (a, b) and 2 (c, d). Solid lines represent well-watered and dotted lines represent drought treatments. The second arrow for rewatering is for DHL-32, which transpired water more slowly. Bars represent LSD$\text{0.05}$ for shoot dry weight.
saturate the leaf tissue, then placed in an eppendorf tube, frozen with liquid nitrogen for 30 seconds to stop the physiological function of its cells, and stored in the deep freeze. Osmotic potential at full turgor was measured with the freezing-point osmometer, micro-Osmette (Precision System Inc. Natick, MA, USA). Osmotic adjustment was calculated as the difference between the turgid osmotic potential in the well-watered treatment and in the stress treatment on the same sampling date (Babu et al., 1999).

(3) Statistical analysis
Analyses of variance were conducted for each water regime and least significant differences at a probability of 5 % were determined using Systat 7.0 (SPSS Inc. 1997). Correlations among dry weight or increments in dry weight at the various growth stages, tiller number increment, leaf area increment, water use, WUE, OA and $\Psi_L$ were also obtained.

Results
1. Total dry weight
Total dry weights were larger in well-watered than drought treatments (Fig. 1). In well-watered treatments, total dry weights were larger in experiment 1 than experiment 2. In general, IR62266 accumulated dry matter more quickly and DHL-32 more slowly in well-watered treatments in both experiments. In experiment 1, lines did not differ significantly during drought, however, they differed on rewatering.
Specifically, DHLs 79 and 54 produced a larger dry matter response to rewatering than DHL-51 and IR62266. DHL-32 accumulated dry matter more slowly than the other lines, and its dry matter increased less than in other lines after rewatering, although there was no significant difference between the lines in late drought.

2. Tillers, leaf area and total dry weight
   Experiment 1 had larger initial values (1st sampling) for tiller number and leaf area than experiment 2 (Table 1). Tiller numbers in experiment 1 and leaf area in both experiments were reduced in early drought. IR62266 consistently had the most tillers in both experiments under well-watered and early drought. Except for the initial sampling in experiment
1. leaf area of IR62266 was highest under well-watered treatments. DHL-32 generally had fewer tillers and less leaf area under early drought. Increments in leaf area in late drought were inconsistent between experiments, although a high leaf area increment in rewatering was apparently associated with a low leaf area increment in late drought (Table 2). However, in both experiments, DHLs 54 and 79 had significantly larger increments in total biomass on rewatering than the other genotypes. On the other hand, DHL-32 had the lowest total biomass and leaf area increments in both experiments.

3. Total dry weight, transpiration and WUE during drought

Increments in dry weight were generally higher in late drought than in early drought in experiment 1, but not in experiment 2 (Table 3). The increment in total dry weight in the late drought interval was significantly higher in DHL-79 in experiment 1 and in DHL-32 in experiment 2, although the latter was associated with a longer duration. Daily transpiration was not closely related to the increments in dry weight. Lines differed in WUE, especially in late drought where DHL-79 was significantly higher and DHL-54 significantly lower in experiment 1, while DHL-32 was significantly higher and DHL-79 significantly lower in experiment 2.

4. Total dry weight increment in relation to tiller number and leaf area

There was no significant correlation between tiller number increment and total dry weight increment in all drought and rewatering treatments for both experiments (Table 4). On the other hand, correlation of leaf area increment with total dry weight increment was significant at early drought (0.83*) in experiment 1, mildly correlated (0.80 ns) in experiment 2 and significant at rewatering (0.86*) in experiment 2. Leaf area increment was consistently not significantly correlated with late drought total dry weight increment in both experiments.

5. Total dry weight increment in relation to transpiration and water use efficiency

Correlations between transpiration and total dry weight increment were generally low and even negative except at early drought in experiment 2 where transpiration was significantly correlated (0.89*) with total dry weight increment (Table 5). Correlations of WUE with total dry weight increment were highly significant (P=0.01) in all cases except at early drought (0.56 ns) in experiment 2.

6. Total dry weight in relation to ΨL and OA

Correlations between OA in early drought, intermediate drought and late drought, and either dry weight or change in dry weight in the various periods, were generally not high (Table 6). Specifically, while OA at early drought was mildly correlated with total

Table 5. Correlation between transpiration, water use efficiency, and total dry weight increment among 5 genotypes during early and late drought.

| Drought period | Transpiration | Water use efficiency |
|----------------|---------------|---------------------|
| Experiment 1   |               |                     |
| Total Dry      | Early         | −0.64               | 0.96**              |
| Weight Increment | Late       | −0.96**             | 0.97**              |
| Experiment 2   |               |                     |
| Total Dry      | Early         | 0.89*               | 0.56                |
| Weight Increment | Late       | −0.28               | 0.95**              |

Correlation at 1%** level and 5%* level of significance.

Table 6. Correlation between osmotic adjustment, leaf water potential and dry weight among 5 genotypes during early, intermediate and late drought and rewatering.

| Drought period | Osmotic adjustment | Leaf water potential |
|----------------|--------------------|----------------------|
|                | Early              | Intermediate | Late | Early | Intermediate | Late |
| Total          | Early              | 0.54        | n/a   | n/a   | −0.25 | n/a | n/a |
| Dry            | Intermediate       | 0.03        | −0.07  | n/a   | −0.72 | 0.00 | n/a |
| Weight         | Late               | −0.96**     | 0.16   | −0.16  | −0.57 | −0.90* | −0.38 |
|                | Rewatering         | 0.26        | −0.21  | 0.12   | −0.49 | 0.29 | 0.91** |
| Change in      | Early              | 0.55        | n/a   | n/a   | −0.24 | n/a | n/a |
| Total          | Late               | −0.62       | 0.11   | −0.27  | 0.15  | −0.53 | −0.89* |
| Dry weight     | Rewatering         | 0.29        | −0.22  | 0.12   | −0.47 | 0.31 | 0.91** |

Correlation at 1%** level and 5%* level of significance.

Relationship of these values are plotted in Fig. 2.

n/a, not applicable due to chronology.
In contrast, relationships between $\Psi_L$ and dry weight were strong, accounting for about 52, 81 and 83 % of variation in the early, intermediate and late drought periods, respectively (Fig. 2d-f). Specifically, $\Psi_L$ in dry weight in early drought (0.54 ns), there was no relationship between OA in intermediate and late drought with total dry weight in late drought (0.16 ns) and rewatering (0.12 ns), respectively (Fig. 2a-c).
late drought was significantly correlated with total dry weight on rewatering ($0.91^{**}$), when total dry weights differed significantly among lines.

### 7. Leaf water potential, osmotic potential and osmotic adjustment

In well-watered conditions, $\Psi_L$ was about $-0.1$ MPa and declined slightly with plant age, especially in DHL-32 at day 43 (Fig. 3a). Osmotic potential was $-1.0$ to $-1.2$ MPa in well-watered conditions.

Leaf water potential decreased under drought before recovering on rewatering, but the extent of the changes differed among lines. In the early drought period, $\Psi_L$ dropped rapidly to about $-0.4$ MPa in DHL-54, DHL-51 and IR62266, while the decline was slower in DHL-32 and DHL-79. However, as drought intensified, $\Psi_L$ in DHL-32 continued to decline to $-0.6$ MPa, while $\Psi_L$ stabilized around $-0.4$ MPa in the other lines (Fig. 3a). Osmotic potential decreased more strongly as drought intensified in DHL-51 and IR62266 than the other genotypes, which had an intermediate response (Fig. 3b). Osmotic adjustment was high in DHL-51 and low in DHLs 32 and 54 (Fig. 3c). These relationships are summarized in Fig 3d, where the increase in OA of DHL-32 to about 0.35 MPa failed to prevent its $\Psi_L$ dropping to $-0.6$ MPa, while in other lines, $\Psi_L$ remained at $-0.4$ MPa as drought intensified.

### Discussion

#### 1. Shoot hydration - leaf water potential and osmotic adjustment

The range in osmotic adjustment reported here is comparable with other reports for rainfed lowland rice (Kamoshita et al., 2000; Jongdee et al., 2002;
Kamoshita et al., 2004), but less than reports for rice from other ecosystems (Lilley and Ludlow, 1996; Babu et al., 1999; Zhang et al., 1999). Ranges in $\Psi_L$ were comparable with other reports for rice.

As expected, $\Psi_L$ was stable in well-watered conditions (Fig. 3), though it dropped a little in DHL-32 with age. The lines differed strongly in the extent of their OA in drought, with $\Psi_L$ more stable in severe drought in lines with stronger OA. Interestingly, $\Psi_L$ was also stable in DHL-79 which was intermediate in OA, but very effective in water extraction (Siopongco et al., 2005). This result is consistent with stability in plant water status under drought being attained via the balance of a number of differing mechanisms, favoring either the capture of additional water resources, adjustment of cellular properties as dehydration progresses, or a combination of both. This is discussed further below.

2. Consequences of shoot hydration for dry matter production

In other species, it is reported that OA can assist dry matter production under drought (Zhang et al., 1999), but this has not been reported in rice (Pantuwan et al., 2002c; Kamoshita et al., 2004). In this experiment, there was some relationship between OA and total dry weight at early drought ($r=0.54$ ns), but the relationship weakened as drought intensified and after rewatering. This result is consistent with Pantuwan et al. (2002c) who reported that OA was not associated with yield or drought response index under drought, due to limited genotypic variation and difficulty in estimating OA precisely.

However, $\Psi_L$ at late drought was strongly correlated with total dry weight at rewatering (Table 6). While total dry weight at intermediate and late drought was not significantly reduced in lines with more negative water potential at early and intermediate drought (Fig. 2d-e), those lines that were able to maintain shoot hydration at the end of the drought period had the greater increase in total dry weight on rewatering (Fig. 2f). Although Figs. 2e and 2f have similarly significant correlations of -0.90 and 0.91, Fig. 2f is more biologically important, as there is a greater range in dry weight increase at rewatering in response to maintenance of leaf water potential in late drought. This result is consistent with the report by Jongdee et al. (2002) that a capacity to maintain $\Psi_L$ is a desirable trait for improved tolerance to drought and for improved ability to recover on rewatering (Fig. 2). However, it is important to note, that different DHLs attained that outcome in different ways. DHL-79 had a greater reliance on extraction of water from deeper soil layers as drought progressed, thus maintaining its $\Psi_L$ more from shoot hydration than from physio-morphological adjustments, such as OA. In contrast, DHL-51 had a greater reliance on extraction of surface water (Siopongco et al., 2005) and osmotic adjustment (Fig. 3). This result, combined with recent field evidence (Samson et al., 2002), suggests that deep roots are not always an advantage. For the sandy soils of northeast Thailand, where Jongdee et al. (2002) took their measurements, little soil water storage was available, so it is not surprising that greater root dry weight or greater root pull resistance were of little benefit (Pantuwan et al., 2002 a and b). Presumably, only a small root length density was needed to extract water from deeper soil layers, but it should be important to at least access that water. At Rajshahi in Bangladesh, where the silty soils hold considerable stored water at depth, the presence of some roots below the surface layer was advantageous (Samson et al., 2002).

3. Consequences of water use efficiency and leaf expansion for dry matter production

This study showed that dry matter accumulation was highly correlated with leaf area expansion at early drought and at rewatering (Table 4). These results are consistent with observations that lines with a greater seedling vigor and a greater ability to extract water during drought (Kamoshita et al., 2000, 2004), had a greater capacity to recover on rewatering, with a greater biomass at the end of the drought period (Mitchell et al., 1998), and a greater increment in leaf area (Wade et al., 2000) or dry weight (Table 2, Fig. 1) on rewatering, which are unique responses found for rainfed lowland rice but not for other rice ecotypes.
Further research is needed to explain the basis of this phenomenon, and why it is important only in the anaerobic-aerobic transitions encountered in the rainfed lowland rice ecosystem.

Yield under drought may be viewed as the product of the water extracted, the efficiency of its conversion to dry matter, and the proportion of that dry matter allocated to grain (Passioura, 1977). In every situation, the best outcome is an optimization of all three parameters. In this study, we found that WUE was highly correlated with total dry weight increment, especially in late drought (Table 5), which means the line with higher WUE produced more dry matter as drought intensified (Table 3). This response was not detected by Wade et al. (2000) and Kamoshita et al. (2004). However, in their studies, they used 8 and 6 genotypes of diverse backgrounds, which may explain their identifying differences in water use, but not WUE in late drought. These results suggest a greater precision was possible by using genotypes of similar genetic background.

Conclusions

The use of DHLs in these studies was advantageous in allowing trait comparisons with greater precision than would be possible using unrelated cultivars, because of the common genetic background. Furthermore, the DHLs were more similar than unrelated cultivars in plant size, reducing confounding by extraneous environmental effects. Thus, the identified relationships were stronger than with unrelated cultivars, as concluded by Lafitte (2002). Through the use of such genetically-related materials and close examination on different stages of stress development including recovery at rewatering, the precision of the relationships obtained between (1) leaf expansion in both early drought and at rewatering and dry matter accumulation at rewatering, (2) water use efficiency during drought and dry matter accumulation, (3) late drought leaf water potential maintenance and total dry matter accumulation at rewatering, has been possible. We proposed these as additional desired traits for drought tolerance as stress progresses in the rainfed lowlands. Specifically, we want plants with a certain phenology and plant size, with avoidance traits to extract water, leafiness for seedling vigor, osmotic adjustment to maintain leaf water potential and with a high WUE as the drought stress intensifies. Nevertheless, we recognize it is important to validate these relationships in field studies.

Acknowledgments

Mr. Rene M. Panopio, Mr. Donato V. Lanwang and Mr. Ramon B. Masajo helped in the experimental operations. Dr. Surapong Sarkarung provided the seeds.

References

Azhiro-Sigari, T., Yamauchi, A., Kamoshita, A. and Wade, L.J. 2000. Genotypic variation in response of rainfed lowland rice to drought and rewatering. II. Root growth. Plant Prod. Sci. 3 : 180-188.

Babu, R.C., Nguyen, B.D., Chamarrera, V., Shamugasundaram, P., Ghezhi, I., Jayaprakash, P., Ganesch, S.K., Palchamy, A., Sadasivam, S., Sarkarung, S., Wade, L.J., Nguyen, H.T. 2003. Genetic analysis of drought resistance in rice by molecular markers: association between secondary traits & field performance. Crop Sci. 43 : 1457-69.

Babu, R.C, Pathan, M.S., Blum, A. and Nguyen, H.T. 1999. Comparison of measurement methods of osmotic adjustment in rice cultivars. Crop Sci. 39 : 150-158.

Blum, A. 1993. Selection for sustained production in water deficit environments, In D. Buxton III ed. International Crop Science I, CSSA, Madison, WI. 343-347.

Boyer, J.S. 1996. Advances in drought tolerance in plants. Adv. Agron. 56 : 187-218.

Fukai, S. and Cooper, M. 1995. Development of drought-resistant cultivars using physiomorphological traits in rice. Field Crops Res. 40 : 67-86.

Jongdee, B., Fukai, S. and Cooper, M. 2002. Leaf water potential and osmotic adjustment as physiological traits to improve drought tolerance in rice. Field Crops Res. 76 : 153-163.

Kamoshita, A., Wade, L.J. and Yamauchi, A. 2000. Genotypic variation in response of rainfed lowland rice to drought and rewatering. III. Water extraction during drought period. Plant Prod. Sci. 3 : 189-196.

Kamoshita, A., Zhang, J., Siopongco, J., Sarkarung, S., Nguyen, H.T. and Wade, L.J. 2002a. Effects of phenotyping environment on identification of QTL for rice root morphology under anaerobic conditions. Crop Sci. 42 : 255-265.

Kamoshita, A., Wade, L.J., Ali, M.L., Pathan, M.S., Zhang, J., Sarkarung, S. and Nguyen, H.T. 2002b. Mapping QTLs for root morphology of a rice population adapted to rainfed lowland conditions. Theor. Appl. Genet. 104 : 880-893.

Kamoshita, A., Rodriguez, R., Yamauchi, A. and Wade, L.J. 2004. Genotypic variation in response of rainfed-lowland rice to prolonged drought and rewatering. Plant Prod. Sci. 7 : 406-420.

Lafitte, R. 2002. Relationship between leaf relative water content during reproductive stage water deficit and grain formation in rice. Field Crops Res. 76 : 165-174.

Lilley, J.M. and Ludlow, M.M. 1996. Expression of osmotic adjustment and dehydration tolerance in diverse rice lines. Field Crops Res. 48 : 185-197.

Maclean, J.L., Dawe, D.C., Hardy, B. and Hettel, G.P. eds. 2002. Rice Almanac: Sourcebook for the Most Important Economic Activity on Earth, third ed. CAB Publishing, Wallingford, England, Published in association with: International Rice Research Institute, West Africa Rice Development Association, International Center for Tropical Agriculture, and Food and Agriculture Organization of the United Nations, 253.

Mitchell, J.H., Siamhan, D., Wamala, M.H., Risimberi, J.B., Chinyamakobvu, E., Henderson, S.A. and Fukai, S. 1998. The use of seedling death score for evaluation of drought resistance in rice. Field Crops Res. 55 : 129-139.

Pantuwan, G., Fukai, S., Cooper, M., Rajatasereekul, S. and
O'Toole, J.C. 2002a. Yield response of rice (Oryza sativa L.) genotypes to drought under rainfed lowland 1. Grain yield and yield components. Field Crops Res. 73 : 153-168.

Pantuwan, G., Fukai, S., Cooper, M., Rajatasereekul, S. and O'Toole, J.C. 2002b. Yield response of rice (Oryza sativa L.) genotypes to drought under rainfed lowland 2. Selection of drought resistant genotypes. Field Crops Res. 73 : 169-180.

Pantuwan, G., Fukai, S., Cooper, M., Rajatasereekul, S. and O'Toole, J.C. 2002c. Yield response of rice (Oryza sativa L.) genotypes to drought under rainfed lowland 3. Plant factors contributing to drought resistance. Field Crops Res. 73 : 181-200.

Passioura, J.B. 1977. Grain yield, harvest index and water use of wheat. J. Aust. Inst. Agric. Sci. 43 : 117-120.

Samson, B.K., Hasan, M. and Wade, L.J. 2002. Penetration of hardpans by rice lines in the rainfed lowlands. Field Crops Res. 76 : 175-188.

Sarkarung, S., Pantuwan, G., Pushpavesa, S. and Tanupan, P. 1997. Germplasm development for rainfed lowland ecosystems: breeding strategies for rice in drought-prone environments. In S.Fukai, M. Cooper, and J. Salisbury eds., Breeding Strategies for Rainfed Lowland Rice in Drought-Prone Environments. Proc. no. 77. Australian Centre for International Agricultural Research, Canberra, Australia. 43-49.

Siopongco, J.DLC., Yamauchi, A., Salekdeh, H., Bennett, J. and Wade, L.J. 2005. Root growth and water extraction response of doubled-haploid rice lines to drought and rewatering during the vegetative stage. Plant Prod. Sci. 8 : 497-508.

Wade, L.J., George, T., Ladha, J.K., Singh, U., Bhuiyan, S.I. and Pandey, S. 1998. Opportunities to manipulate nutrient by water interactions in rainfed lowland rice systems. Field Crops Res. 56 : 93-112.

Wade, L.J., Fukai, S., Samson, B.K., Ali, A., Mazid, M.A. 1999. Rainfed lowland rice: physical environment and cultivar requirements. Field Crops Res. 64, 3-12.

Wade, L.J., Kamoshita A., Yamauchi, A. and Azhiri-Sigari, T. 2000. Genotypic variation in response of rainfed lowland rice to drought and rewatering. I. Growth and water use. Plant Prod. Sci. 3 : 173-179.

Zhang, J., Nguyen, H.T. and Blum, A. 1999. Genetic analysis of osmotic adjustment in crop plants. J. Exp. Bot. 50 : 291-302.