Pertanika J. Trop. Agric. Sci. **32 (2)**: 293 - 303 (2009)
ISSN: 1511-3701
© Universiti Putra Malaysia Press

Pollen and Seed Yield Components of Water-stressed Cultivated and Weedy Rice

Puteh, A.B.*, Jali, N., Ismail, M.R., Juraimi, A.S. and Samsudin, N.
Department of Crop Science, Faculty of Agriculture,
Universiti Putra Malaysia, 43400 UPM, Serdang, Selangor, Malaysia
**E-mail: aputeh@yahoo.com*

ABSTRACT

Water stress occurring during the early phase of the reproductive growth stage may influence plant reproduction success. The objectives of this study were to evaluate the responses of pollen and seed yield components to water stress during anthesis in cultivated rice (*Oryza sativa* L.) varieties and weedy rice strains. Studies were conducted in 2007 using three cultivated rice varieties; MR 84, MR 219 and MR 232. In 2008 three weedy rice strains were obtained from Seberang Perak, Kuala Pilah and Tanjung Karang areas. Studies were conducted in the field where plants were grown in polybags and submerged in polyethylene tanks. Prior to anthesis, plants were subjected to water stress by taking the plants out of the polyethylene tanks for five days. Flag leaf relative water content (RWC) and relative injury (RI) were measured daily during the stressed period. Pollen production and pollen viability were measured upon re-watering. Seed yield components measured were grain number per panicle, 100-grain weight, percentage filled grain and percentage spikelet sterility at harvest. Water stress caused a 13 – 34% decline in the number of pollen grains per anther in cultivated varieties but increased pollen production in weedy strains. Percentage pollen viability declined by 40 – 45% in MR 219 and MR 232, but increased by 15% in MR 84 when plants were water-stressed. Percent pollen viability in weedy strains never exceeded 52% and significantly declined with water stress. For cultivated varieties, water stress increased total number of grain per panicle by 31%. In weedy strains, only Seberang Perak increased in the number of grain per panicle due to water stress. Spikelet sterility was relatively higher in cultivated varieties (16 – 50%) compared with weedy strains (10 – 23%). The 100-grain weight was not affected by water stress in both cultivated varieties and weedy strains. This study indicated that weedy rice strains would gain a competitive advantage by producing more filled grains when water deficit occurs during anthesis.

Keywords: Water stress, weedy rice, pollen, seed yield components

INTRODUCTION

For successful seed set in cereal crops, viable pollen, receptive stigma and well developed ovule are required. Unfavourable environments occurring during the early phase of the reproductive growth stage will interrupt normal pollen grain and ovule developments and subsequently will lead to reduced or complete failure in seed set. Water availability during early reproductive growth stage especially during anthesis is one of the major environmental limitations and has been implicated in inconsistent yield potential and reduced seed yields in several grain crops (Boyer and Westgate, 2004; Barnabas *et al*. 2008).

The degree of damage caused by water deficit in several grain crops is strongly dependent on the genotype, severity of water shortage and plant growth stage at which it occurs (Jongdee

Received: 23 February 2009
Accepted: 24 March 2009
*Corresponding Author

et al., 2002; Lafitte, 2002; Liu *et al*. 2006; Vanuprasad *et al*. 2007). During early growth in plant reproduction, different phases show different susceptibility to water deficits. The most sensitive growth stage to water stress in cereals falls between booting stage until anthesis. Water deficits within this short span of time, particularly during meiosis, anthesis or early zygote formation, may subsequently cause the failure of seed set in grain crops. Sheoran and Saini (1996) reported that the most damaging effect of water stress occurred during meiosis in pollen mother cells This clearly indicates that the early reproductive growth stage is the most critical time for reproduction success. Thus, avoiding water deficit through good water management system during this critical stage will likely improve yield potential.

It appears that water availability during pollen grain formation will directly disrupt the reproductive process. The incidence of pollen grains sterility had been reported in barley, maize, wheat and rice when low water potential occurred during microsporogenesis and microgametogenesis (Saini, 1997; Saini and Westgate, 2000). In cultivated rice varieties, reduction in seed yield due to drought or water stress during anthesis had been frequently associated with increased percentage of spikelet sterility (Fukai *et al.,* 1999; Jongdee *et al*., 2002; Liu *et al*., 2006) and spikelet number per panicle (Boonjung and Fukai, 1996). Prolonged stress occurring during anthesis causes changes in panicles and flag leaf transpiration which contributed to severe spikelet desiccation and white heads (Ekanayake *et al.,* 1993).

The degree of seed yield reduction due to water deficit is highly dependent on the timing of stress (Garrity and O'Toole, 1994). Water deficit during meiotic stage had been reported to reduce seed set in some cultivated rice varieties (Sheron and Saini, 1996) and maize (Boyer and Westgate, 2004). In hybrid rice, water deficit during the grain filling stage enhanced grain yield due to remobilization of carbon reserves (Yang *et al*., 2003). Therefore, water deficit prior to or during anthesis in several cultivated varieties is well documented and as accepted evidence which reduces seed set in majority of cereal crops. However, little information are available on whether water deficit during early reproductive growth stage will affect pollen production and viability in weedy rice genotypes in comparison with cultivated rice varieties. The objectives of this study were to compare the differences in pollen and seed yield components response to water stress during anthesis in cultivated rice varieties and weedy rice strains.

MATERIALS AND METHODS

Plant Materials and Rice Culture

This study was conducted at the Faculty of Agriculture, Universiti Putra Malaysia in 2007 and 2008. In 2007 season, three cultivated rice varieties (the old popular MR 84 variety and the new widely planted MR 219 and MR 232 varieties) were used. In 2008, three wild strains of weedy rice seeds collected in November 2007 from three different rice growing areas in Malaysia were used.

Seeds of the cultivated varieties and wild strains of weedy rice were sown in black perforated plastic polybags of 30×30 cm size containing approximately 10 kg of soil obtained from a rice growing area. Several seeds were sown in each polybag, but only two emerging seedlings were allowed to grow to avoid overcrowding. The polybags were submerged in water in polyethylene tanks of 90×90 cm size. A total of 10 polybags were housed in each polyethylene tank. The polyethylene tanks containing seedlings in polyethylene bags were placed in an open field and were arranged in a completely randomized design with two replications and with several samplings per experimental unit. Standard procedures of rice growing culture were followed throughout the studies. Seedlings growth and development were monitored daily.

Water Stress Treatment

When a panicle was protruded through the flag leaf from any tiller within a polyethylene tank, all the 10 polyethylene bags were taken out to impose water stress treatment for five days. Re-watering of stressed plants by putting back the plants into the polyethylene tank was done on day 6. The control plants were left submerged in water in the polyethylene tank throughout the study. Within this imposition of water-stress period, the polybags were placed under the shade in the glasshouse to avoid additional water from rain. A tensiometer (Irrometer, Model SR, Riverside, CA, USA) was inserted into each polybag of the stressed plants at a depth of 15 cm to monitor soil moisture potential (ψ_w) during the water stress period (Table 1). The first reading of ψ_w was recorded one hour after imposition of water stress in the morning (9.00 – 10.00 h). The final ψ_w reading (day 6) was taken within one hour after the stressed plants were re-watered in the polyethylene tanks.

Measurement of Leaf Water Relations Parameters

Fully extended flag leaves were randomly cut from the stressed plants beginning from 1 day until 5 days stressed period to determine RWC. A total of three flag leaves from different plants were sampled daily at mid-morning (10.00 – 11.00 h). The last flag leaf samples (day 6) were taken within one hour after the stressed plants were re-watered in the polyethylene tanks. Flag leaf samples of the control plant were collected at the same time when flag leaf samples from stressed plants were collected. The cut pieces of 5 cm section flag leaves were put in vials and immediately brought to the laboratory to determine fresh weight (FW). Distilled water was added to the vials containing the flag leaf sections and were kept for 24 h at room temperature (25°C) in the dark. The flag leaf sections were weighed to determine turgid weight (TW). Dry weight (DW) of the leaf sections were obtained after oven-drying at 80°C for 48 h. The RWC was calculated using the formula:

$$RWC = [(FW - DW)/(TW\text{-}DW)] \times 100$$

Determination of cell membrane thermal stability (CMTS) of flag leaf for control and water stressed plants was done according to the procedures outlined by Prasad *et al.* (2006). The flag leaves collected for RWC determination were also used to determine CMTS. The CMTS was estimated by using the following formula:

$$\%\ CMTS = [1\text{-}\ (T_1/T_2)]/[1\text{-}\ (C_1/C_2] \times 100$$

where T and C refer to electrolytic leakage (conductivity) in the control and heat-treated samples and subscripts 1 and 2 refer to

TABLE 1

Soil water potential (ψ_w) at different days in polybags during stressed period for cultivated varieties (2007) and weedy rice strains (2008)

Day	Soil Water Potential (ψ_w)	
	Cultivated varieties	Weedy strains
	––––––––– kPa –––––––––	
1	5.0 ± 0.01 ‡	5.0 ± 0.02
2	13.13 ± 1.38	10.00 ± 0.57
3	43.75 ± 4.75	34.63 ± 4.5
4	78.13 ± 4.88	61.0 ± 6.01
5	83.0 ± 0.01	87.00 ± 5.00
6 (re-watered)	2.5 ± 0.50	2.0 ± 0.01

‡ ± values indicate standard error of the means.

conductivity before and after autoclaving. Relative Injury (RI) was then derived from the following formula:

$$\% \text{ RI} = 100 - \text{CMTS}$$

Determination of Pollen Quality

Pollen quality determined in this study was referred to as pollen number per anther and pollen viability. During the water stress period, stressed and control plants were monitored daily for anthesis. Prior to anthesis, a total of 6 matured anthers from 20 spikelets from different panicles of stressed plants were randomly selected within 1 h following re-watering (day 6). Spikelets of the control plants were collected on the same day as spikelets from stressed plants were collected.

Pollen number was estimated by placing an anther on a glass slide with grids and squashed with a needle to disperse pollen grains on the slide. Estimation of pollen grain number was done under a light microscope (Hirox Hi-Scope, KH-2700, Japan).

The remaining anthers after determination of pollen number were used for a pollen viability test. A total of six matured anthers were randomly selected from stressed and control plants. Tetrazolium chloride (tetrazolium 2,3,5-triphenyl chloride) test was used to test pollen viability for cultivated varieties. Pollen grains were dusted onto glass Petri dishes. Five to six drops of 0.5% Tetrazolium chloride solution were added. The Petri dishes containing pollen grains were than placed in an oven at 38˚C for two hours. At the end of two hours, a uniform red stain on pollen grains observed under a light microscope (Hirox Hi-Scope, KH-2700, Japan) were considered as viable. Percentage of pollen viability was estimated based on 300 pollen grains. For weedy rice strains, 1% IKI was used to estimate pollen viability (Prasad *et al.* 2006). IKI was used to avoid the damage to pollen grains at high temperature exposure (Huang *et al.,* 2006). A similar procedure was followed as used in cultivated varieties except that pollen grains were not exposed to high temperature, instead the pollen grains were left at room temperature, at approximately 25˚C, while staining in IKI solution.

Determination of Seed Yield Components

Seed yield components were determined when the seed reached full maturity (when seeds changed from green to yellow). The process of seed ripening took 25 – 35 days. A total of twelve randomly selected panicles were harvested from each experimental unit. Panicles were air-dried at room temperature for 24 h before seed yield components were recorded. The grains were separated from panicles to determine the number of grain and grain weight (filled and unfilled spikelets) per panicle. The grains from twelve panicles were then bulked. Percentage sterility and filled grain were calculated based on 15 g of bulked grains. Filled grains were separated from empty spikelets using a General Seed Blower (Hearson, MO., USA). Sub-samples of four random 100-grain samples were counted to determine 100-grains weight.

The data collected were analyzed for analysis of variances by means of SAS statistical analysis package (SAS, 1995). Data from each study date were analyzed separately. Multiple mean comparison analysis for treatment combinations of variety and stress treatment was performed by using least significant different at $\alpha = 0.05$ level when F-test was significant.

RESULTS

Soil Moisture Potential

Soil moisture potential in polyethylene bags of stressed plants increased from 5 kPa at the beginning of stress period (day 1) to 83 kPa and 87 kPa for cultivated and weedy rice, respectively (Table 1). The stressed plants started to show some curled leaves beginning at day 3. By day 4, all leaves on stressed plants were curled, but none of the plants died during the stressed period. The curled leaves of stressed plants recovered their normal shape and structure 3 – 4 h after re-watering.

Flag Leaf Relative Water Content

The RWC of flag leaf in cultivated rice varieties for water-stressed and control plants ranged between 89 – 96% and 82 – 99% at the beginning of stressed and unstressed (control) period, respectively (*Fig. 1*). The RWC for both stressed and control plants were maintained above 90% at the end of day 4. This indicates that stressed period for four days was not severe enough to cause differences in RWC.

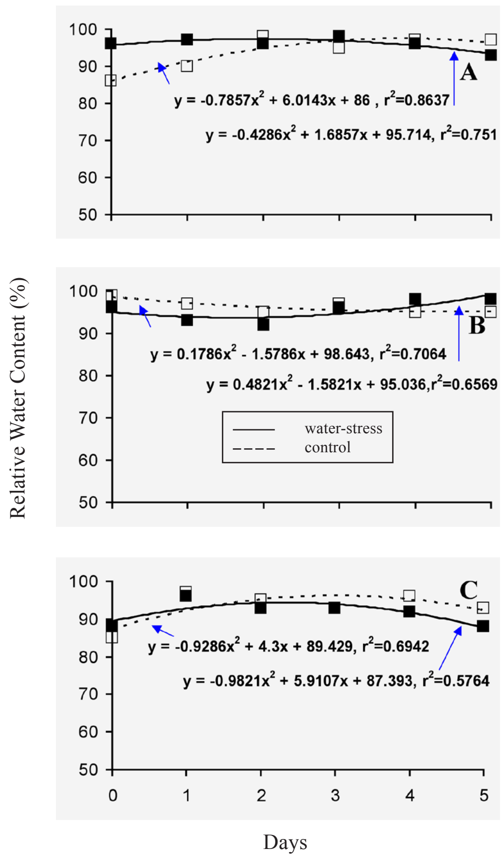

Fig. 1: Relative water content of MR 232 (A), MR 219 (B) and MR 84 (C) varieties for control (□) and water stress (■) treatments

The flag leaf RWC for Seberang Perak strain was between 55 – 60% at the beginning of the stress period (day 1) (*Fig. 2A*). RWC was >91% at day 1 for Kuala Pilah and Tanjung Karang strains (*Fig. 2B* and *2C*). Similar to cultivated rice varieties, RWC for both stressed and control weedy strains was maintained above 90% at the end of day 4.

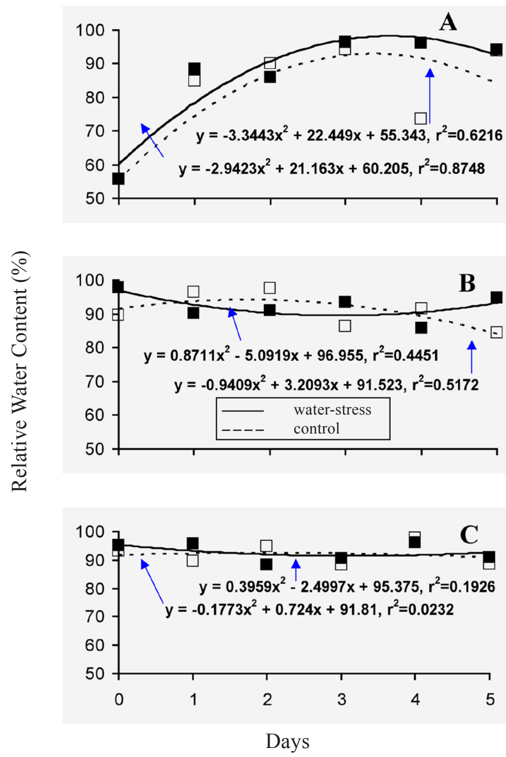

Fig. 2: Relative water content of Seberang Perak (A), Kuala Pilah (B) and Tanjung Karang (C) strains for control (□) and water stress (■) treatments

Flag Leaf Relative Injury

There were slight differences between cultivated varieties in %RI for water stressed and control plants. The stressed plants showed a declining pattern of RI with increased number of stressed period (*Fig. 3*). The RI due to water stress from day 1 to day 5 in MR 219 variety was in the range of 16 – 32%, and 37 – 47% for MR 84 variety. In MR 219 control plants, RI never exceeded 25% when recorded within same period of time. In MR 84 control plants, percent RI increased from 24% at day 1 to 55% at day 5.

RI for weedy rice strains never exceeded 15% during 5 days of stress (*Fig. 4*). Two strains, Seberang Perak and Kuala Pilah, showed increasing trend of % RI with increased days of stress period. Similarly, the control strains had <15% RI. Tanjung Karang strain showed the lowest % RI after 5 days of water stress compared with other two strains (*Fig. 4C*).

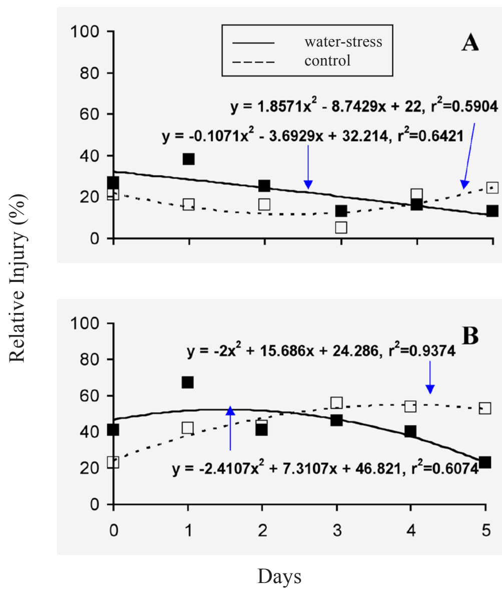

Fig. 3: Relative injury of MR 219 (A) and MR 84 (B) varieties for control (□) and for water stress (■) treatments

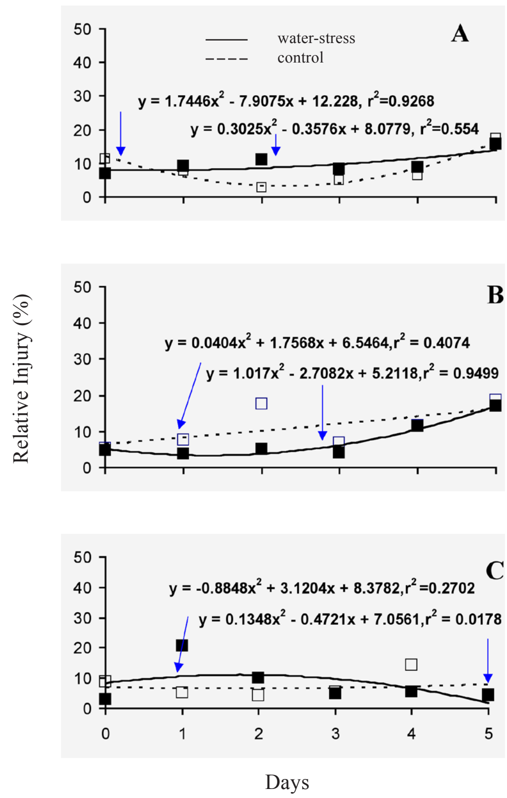

Fig. 4: Relative injury of Seberang Perak (A), Kuala Pilah (B) and Tanjung Karang (C) strains for control (□) and water stress (■) treatments

Pollen Quality

Water-stressed plants of cultivated varieties showed a decline in pollen number per anther by an average of 20% over the control. The number of pollen per anther for cultivated varieties ranged between 448 and 635 for the control plants and declined to 382 – 553 when the plants were water-stressed (Table 2). Only the MR 219 variety showed significant decline in pollen number per anther by 35% when plant were stressed for 5 days. The decline in pollen number per anther due to water stress for MR 232 and MR 84 was on the average of 14%.

Pollen viability as measured by TTC staining significantly declined in MR232 and MR219 (Table 2). The decline in pollen viability in MR232 variety was 45% and 40% for MR219.

Total pollen production in weedy rice strains was much higher than in cultivated varieties for both the control and water stressed plants. The pollen number per anther recorded in control plants ranged between 1276 and 1806 and 1529 – 1888 in water-stressed plants (Table 3). Only Seberang Perak strain significant increased (by 29%) pollen production when the plants were water-stressed during anthesis. The increase in pollen production when subjected to water stress for Kuala Pilah and Tanjung Karang strains was <5%. An average increase of pollen number per anther for water stressed plants was 10% over the control. This indicates that when weedy rice strains experience water stress during anthesis, it will trigger plants to produce more pollen.

Pollen viability was low (21 – 52%) for all weedy rice strains regardless of water stress

TABLE 2
Number of pollen and pollen viability of cultivated varieties of unstress (control) and water stress treatments

Variety	Treatment	No. pollen anther^{-1}	Viability (%)
MR232	Control	635	90
	Water stress	553	49
MR219	Control	511	71
	Water stress	336	42
MR84	Control	448	58
	Water stress	382	67
LSD		152	22
ANOVA		F-value (P- value)	
Stress		8.94 (0.024)	8.45 (0.027)
Variety		10.61 (0.011)	1.16 (0.037)
Stress x variety		0.90 (0.456)	4.64 (0.061)

TABLE 3
Number of pollen and pollen viability of weedy rice strains of unstress (control) and water stress treatments

Strain	Treatment	No. pollen anther^{-1}	Viability (%)
Seberang Perak	Control	1276	21
	Water stress	1650	16
Kuala Pilah	Control	1806	52
	Water stress	1888	37
Tanjung Karang	Control	1519	52
	Water stress	1529	49
LSD		116	4
ANOVA		F-value (P-value)	
Stress		15.33 (0.008)	37.44 (<0.001)
Strain		38.44 (0.004)	236.66 (<0.001)
Stress x strain		8.34 (0.019)	8.15 (0.019)

treatments. Seberang Perak strain had only 21% pollen viability and significantly declined to 16% viability when plants were stressed. Similarly, Kuala Pilah strain showed a significant decline in pollen viability from 52% to 37% when plants were subjected to stress. Tanjung Karang strain did not decline in pollen viability when the plants were stressed for 5 days during anthesis.

Seed Yield Components

Water stress during anthesis for four days increased total number of grain per panicle for all the three cultivated varieties. In MR 84 variety, short period of water stress caused the plant to produce less pollen grains but without a significant reduction in percentage filled grain although high spikelet sterility. The significant increase in total grain number was only observed in MR 84 variety which was increased by 58% (Table 4). Control plants of MR 232 variety had the highest number of grains per panicle and the lowest was recorded in MR 219. The 100-seed grain weight was not affected when plants were subjected to water stress for 4 days during anthesis. The 100-grain weight were on an average of 2.14 g for MR 219 and MR 232 and 1.72 g for MR 84. Percentage filled grain showed no significant difference between control and water stressed plants for all three varieties. The average percentage filled grain was highest in MR 232 which was 84% compared with 55% in MR84. Percentage sterility (empty spikelets) per panicle was not severely affected when cultivated varieties were stressed for 5 days during anthesis. MR 84 variety had 46% spikelet sterility average across stress treatment. MR 219 showed insignificant increased in percentage sterility from 37% for control to 50% when water-stressed. This indicates that MR 219 was the most sensitive to water stress among the three varieties tested.

Water stress for 5 days during anthesis showed inconsistent and no significant effect on total number of grain per panicle for all three weedy rice strains (Table 5). The total number of grain per panicle was increased in Seberang Perak strain but was decreased in Kuala Pilah strain when plants were water stressed. The 100-seed grain weight for all three weedy rice strains on the average was much higher than the cultivated rice varieties. Only Seberang Perak strain showed a significant increase in 100-grain weight following stress treatment. Percentage filled grain was not affected by water stress in Kuala Pilah and Tanjung Karang strains which was on the average of 89%. Similarly, percentage sterility was not affected by water stress in Kuala Pilah and Tanjung Karang strains. Increase in percentage filled grain from 77% in control plants to 83% was observed in Seberang Perak strain. Concomitant decline in percentage sterility from 23% to 17% was also observed in Seberang Perak strain.

DISCUSSION

Leaf relative water content (RWC) is one of the commonly use indicators of leaf water status when plants are subjected to water stress. Measurement of leaf relative water content is easy to perform; however, inconsistent and

TABLE 4
Seed yield components of cultivated rice varieties of unstress (control) and water stress treatments

Variety	Treatment	No. of grain panicle^{-1}	100-grain wt. (g)	Filled grain (%)	Sterility (%)
MR232	Control	125	2.11	83	18
	Water stress	146	2.20	85	16
MR219	Control	90	2.10	63	37
	Water stress	105	2.17	50	50
MR84	Control	105	1.70	56	45
	Water stress	166	1.74	53	47
LSD		43	0.22	18	15
ANOVA		F-value (P-value)			
Stress		4.81 (0.071)	0.23 (0.648)	0.06 (0.813)	6.21 (0.048)
Variety		4.43 (0.066)	26.39 (0.001)	2.44 (0.168)	13.32 (0.006)
Stress x variety		0.12 (0.889)	0.70 (0.532)	0.61 (0.573)	1.53 (0.290)

TABLE 5
Seed yield components of weedy rice strains subjected to unstress (control) and water stress treatments

Strain	Treatment	No. grains panicle^{-1}	100-grain wt. (g)	Filled grain (%)	Sterility (%)
Seberang Perak	Control	172	2.86	77	23
	Water stress	198	3.37	83	17
Kuala Pilah	Control	150	2.63	90	10
	Water stress	131	2.80	89	11
Tanjung Karang	Control	160	2.81	90	10
	Water stress	159	2.68	87	13
LSD		27	0.60	4	4
ANOVA		F-value (P-value)			
Stress		0.12 (0.744)	0.81 (0.487)	0.08 (0.0790)	0.17 (0.690)
Strain		16.49 (0.003)	1.63 (0.271)	6.09 (0.035)	9.97 (0.012)
Stress x strain		4.23 (0.072)	0.84 (0.487)	1.04 (0.408)	7.75 (0.255)

varying results in relation to seed yields was reported in rice (Jongdee *et al*., 2002; Lafitte, 2002). Similarly, in this study measurement of flag leaf RWC during the stressed period was inconsistently correlated to most of the parameters measured. These results suggest that other factors are as important as RWC in determining response to water stress during flowering. Therefore, RWC of flag leaf recorded during stressed period appeared to be an unreliable plant water stress indicator. The concept of leaf membrane thermal stability to measure plant relative injury to water stress was based on the leaf cell membrane integrity by measuring electrolytes leakage following hot and cold exposure of leaf cells (Agarie *et al*., 1995). The flag leaf RI in MR 219 observed in this study was lower than that in MR 84, but the low percentage value of RI in MR 84 did not contribute to an increase in grain production per panicle. In an earlier study (Prasad *et at*., 2006), rice varieties grown at higher temperature and had lower RI also did not contribute to higher percent spikelet fertility and grain yields. This observation suggests that RI may not be a reliable selection criteria for plant stress in rice. Furthermore, the versatility of leaf membrane thermal stability which measures plant responses to water deficits has yet to be tested on several crop species.

Withholding water from rice plants for five days appeared not to be severe enough to have any critical effect on pollen production in cultivated varieties, instead the weedy strains tended to produce more pollen. In general, pollen production in weedy rice strains was increased by an average of 10% when water-stressed; however, in cultivated varieties it was reduced by an average of 20%. Only the MR 219 variety decreased in pollen production following water stress during anthesis. For MR 219, not only pollen production was reduced but pollen viability was also significantly lowered. Reduction in pollen viability in this particular variety led to reduced proportion of filled grain on panicle in response to higher percentage of spikelet sterility. It indicates that this variety was sensitive to even a short period of water stress, especially during anthesis compared with other two cultivated varieties. Wilcox and Neiland (2002) reviewed several stages which lead to pollination failure and one of them was due to insufficient pollen quantity. Liu *et al*. (2006) implicated increased spikelet sterility in rice after six days of water stress at anthesis to reduced anther dehiscence and low stigmata-pollen density rather than due to reduced pollen viability. For MR 232 variety, pollen viability was affected but the proportion of filled grain on the panicle was not affected because of low percentage of spikelet sterility. The ability of MR 232 to produce high number of pollen, >550 pollen per anther, under water deficit environment appears to compensate for the reduction in pollen viability and subsequently resulted in high percentage of filled grain. This suggests that high amount of pollen in an anther may give a high chance of successful fertilization. A minimum of 20 pollen grains per stigma are required for a successful fertilization and seed set in rice (Matsui and Kagata, 2003). In this study, it was found that seed size as measured by 100-grain weight was not affected by water stress in both cultivated varieties and weedy strains. This indicates that accumulation of dry weight by seed which determines the final seed size is not affected if water stress occurred within five days during early part of reproductive growth stage in rice.

In general, percentage spikelet sterility in weedy strains was relatively lower (10 – 23%) compared with cultivated varieties (16 – 50%). Pollen viability in weedy strains was relatively low compared with cultivated varieties. However, number of grain and the proportion of filled grain formed in weedy strains were higher than in cultivated varieties. In one of the strains, Seberang Perak, a short period of water stress significantly increased percentage of filled grain. A possible explanation for the increase in the proportion of filled grain in this particular strain was higher pollen number being produced which subsequently caused a greater chance of successful fertilization. Another possibility is that the nature of those weedy strains which can tolerate water stress without affecting the yield components. In hybrid rice, mild water stress prior to anthesis facilitates remobilization of C reserves, increase grain-filling rate and increase grain yield (Yang *et al*., 2003).

CONCLUSIONS

Tolerance to water stress based on RWC and flag leaf percentage relative injury seem to be unreliable indices of plant stress on which to select new rice variety. The data suggest that water stress for less than five days will enhance weedy rice strains to produce higher amount of seeds. Water stress for less than five days during early reproductive growth stage will contribute to a competitive advantage for weedy rice. This study also suggests that longer duration of water stress (>5 days) should be conducted to confirm the differences in the effect of water stress on pollen and seed yield in cultivated and weedy rice.

High amount of pollen per anther and high pollen viability appeared to be important genotypic factors and should be included as a selection criterion for a new rice variety. The new cultivated varieties with high number of pollen per anther can withstand short water stresses, especially during the early reproductive growth stage without negative effect on seed set and seed yields.

The presence of weedy rice in the field and a short period of water stress occurring prior to or during anthesis may contribute to higher incidence of weedy rice infestation in the following growing season.

ACKNOWLEDGEMENTS

The authors are grateful to the Malaysian government for providing Fundamental Research Grant Scheme (FRGS) to carry out this project. The authors also wish to acknowledge Prof. Shamshuddin Jusop from Faculty of Agriculture, UPM for his constructive comments and suggestions.

REFERENCES

Agarie, S., Hanoaka, N., Kubota, F. Agata, W. and Kaufman, P.B. (1995). Measurement of cell membrane stability evaluated by electrolyte leakage as a drought and heat tolerance test in rice (*Oryza sativa* L.). *Journal of the Faculty of Agriculture, Kyushu University, 40,* 233 – 240.

Barnabas, B., Jager, K. and Feher, A. (2008). The effect of drought and heat stress on reproductive processes in cereals. *Plant, Cell and Environment, 31,* 11 – 38.

Boonjung, H. and Fukai, F. (1996). Effects of soil water deficit at different growth stage on rice growth and yield under upland conditions. 2. Phenology, biomass production and yield. *Field Crop Research, 48,* 47 – 55.

Boyer, J.S. and Westgate, M.E. (2004). Grain yields with limited water. *Journal of Experimental Botany, 55,* 2385 – 2394.

Ekanayake, I.J., De Dat, S.K. and Steponkus, P.L. (1993). Effect of water deficit stress on diffusive tolerance, transpiration, and spikelet desiccation in rice (*Oryza sativa* L.). *Annals of Botany, 72,* 73 – 80.

Estrada-Campuzano, G., Miralles, D.J. and Slafer, G.A. (2008). Genotypic variability and response to water stress of pre- and post-anthesis phases in triticale. *European Journal of Agronomy, 28,* 171 – 177.

Fukai, S., Pantuwan, G., Jongdee, B. and Cooper, M. (1999). Screening for drought resistance in rainfed lowland rice. *Field Crop Research, 64,* 61 – 74.

Garrity, D.P. and O'toole, J.C. (1994). Screening rice for drought resistance at the reproductive phase. *Field Crop Research, 39,* 99 – 110.

Hirayama, K., Ishida, K. and Tomaru, N. (2005). Effect of pollen shortage and self-pollination on seed production of endangered tree, *Magnolia stellata*. *Annals of Botany, 95,* 1009 – 1015.

Huang, Z., Zhu, J., Mu, X. and Lin, J. (2006). Pollen dispersion, pollen viability and pistil receptivity in *Leymus chinensis*. *Annals of Botany, 93,* 295 – 301.

Jongdee, B., Fukai, S. and Cooper, M. (2002). Leaf water potential and osmotic adjustment as physiological trains to improve drought tolerance in rice. *Field Crop Research, 76,* 153 – 163.

Lafitte, R. (2002). Relationship between leaf relative water content during reproductive stage water deficit and grain formation in rice. *Field Crops Research, 76,*165 – 174.

Liu, J.X., Liao, D.Q., Oane, R., Estenor, L., Yang, X.E., Li, Z.C. and Bennett, J. (2006). Genetic variation in the sensitivity of anther dehiscence to drought stress in rice. *Field Crop Research*, *97,* 87 – 100.

Matsui, T. and Kagata, H. (2003). Characteristics of floral organ related to reliable self-pollination in rice (*Oryza sativa* L.). *Annals of Botany, 91,* 473 – 477.

Moinuddin, R.A., Fischer, R.A., Sayre, K.D. and Reynolds, M.P. (2005). Osmotic adjustment in wheat in relation grain yield under waters deficit environments. *Agronomy Journal*, 1062 – 1071.

Prasad, P.V.V., Boote, K.J., Allen, Jr., L.H., Sheehy, E. and Thomas, J.M.G. (2006). Species, ecotypes and cultivar differences in spikelet fertility and harvest index of rice in response to high temperature stress. *Field Crop Research, 95,* 398 – 411.

Sheoran, I.S. and Saini, H.S. (1996). Drought-induced sterility in rice: changes in carbohydrate levels and enzymes activities associated with the inhibition of starch accumulation in pollen. *Sexual Plant Reproduction, 9,* 1661 – 1669.

Saini, H.S. (1997). Effect of water stress on male gametophyte development in plants. *Sexual Plant Reproduction, 10,* 67-73.

Saini, H.S. and Westgate, M.E. (2000). Reproductive development in grain crops under drought. *Advances in Agronomy, 68,* 59 – 96.

Vanuprasad, R., Lafitte, H.R. and Atlin, G.N. (2007). Response of direct selection for grain yield under drought stress in rice. *Crop Science*, *47,* 285 – 293.

Wilcox, C. and Neiland, R. (2002). Pollination failure in plants: why it happens and when it matters. *Trends in Plant Science, 7,* 270 – 277.

Yang, J., Zhang, J., Wang, J.Z., Liu, L. and Zhu, Q. (2003). Post-anthesis water deficits enhance grain filling in two-line hybrid rice. *Crop Science, 43,* 2099 – 2108.