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Article

Aerobic Rice (Cv. MRIA) Differs Temporarily by Environmental Stress during Seed Development and Maturation

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Abstract: Drought and high temperature often occur simultaneously will eventually affect rice (Oryza sativa L.) yield and seed quality. The objective of the study was to compare the effects of high temperature (HT) and water limitation (WL) during seed development and maturation on yield component, seed quality, and amylose content in aerobic rice cv. MRIA. Two experiments were conducted concurrently in greenhouse with ~35/26°C subjected to elevated HT and/or WL; 5 days 38/28°C at 10, 15, 29 DAA and/or HT; 7 days at 40/28°C at 8 and 21 DAA with 12h photoperiod in control environment growth chamber. Both experiments were respectively harvested at physiological maturity of 31 and 35 DAA. Grain yielded from the irrigated plant declined from over 27% to below 18% between 15 and 29 DAA. Combination stresses resulted in greater reduction of grain yield than single stress. Late seed development (29 DAA) had no effect either on yield component or seed quality as well as produce a high concentration in amylase content (16.7%) with greater germination capacity (91%). The non-stress plant harvested at 21 DAA resulted the highest percentage of germinated seeds (97%) whereas the vigour index (VI) was reduced to 70.3% and 58.9% at 8 and 21 DAA respectively. Extended plant stress to 7 consecutive days incurred the highest VI with low concentration of amylose content. There was a linear relationship incurred between seed amylase content with germination capacity and VI, however, there was an inverse relationship (R2 = 0.19) between amylase content with VI from plant treated with (HT and/or WL) for 7 days. The present results confirmed that the avoidance HT and WL during early (10 DAA) and mid (15 DAA) seed development with short duration of stress (5 days) obtained better in yield component, enhanced the seed quality and amylose concentration. The combined HT and WL damaged seeds' quality were more than each stress alone. Therefore, avoiding HT and WL can be practically applied by farmers by irrigating throughout the rice plant during histodifferentiation to save water usage during seed maturation.

Keywords: aerobic rice; high temperature; water limitation; yield component; seed quality; amylose content

1. Introduction

Climate change is now tremendously important to the agriculture sector worldwide with consequences for crop yield and seed quality. Environmental conditions during rice cultivation affect seed quality [1-3] with drought and high temperature are the important elements to consider in order reducing crop failure resulting from sowing poor-quality seed. Christoph Müller et al. [4] reported that by year 2100 the impact of climate change on crop yields for high-emission climate scenarios ranges between 20 and 30%. In Malaysia, for instance, temperature increases by 1% account over 3.44% reduction of rice yield and 0.03% decreases in the following season [5]. Drought and high temperature are particular problems occurring episodically in some regions and ubiquitously in others. The highest temperature recorded in Peninsular Malaysia for three consecutive days was in

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May 2019 is between 37°C to 40°C as reported by the Malaysian Meteorological Department. Prolong of elevated temperature can cause a high evaporation rate in the soil which will lead to water stress. Generally, Malaysia has two monsoon regimes namely, Southwest Monsoon from late May to September which relatively shows dry weather and the Northeast Monsoon from November to March which is the main rainy season in Malaysia (Malaysia Meteorology Department, 2019). Farmers are advised to start sowing paddy seeds in Northeast Monsoon from November to March to avoid the detrimental effects of high temperature at flowering and during seed set that can cause asynchrony between male and female reproductive development [6-8]. Therefore, increased attention has been paid to understand the temporal sensitivity of temperature change and drought in Malaysia.

Aerobic rice is cultivated in a production system in well drainage, non-puddle, and non-saturated soil. It was developed by the International Rice Research Institute (IRRI) purposely to maintain the sustainability of rice production under water limitation environment. According to Hussain et al. [9], the aerobic rice cv. Aeron 1 produces an average of 3.3 to 3.5 t/ha to 5 t/ha of rice planted three times a year and can be harvested in only 90 days after sowing. In addition, Souki and Othman [10], reported that the average yield for aerobic rice cv. Aeron 1 planted in Sabah was 3.79 t/ha in season 2/2011 and 4.07 t/ha during 1/2012. Aerobic rice paddy cv. MRIA that has been released in Malaysia since 2013 [11]. Unfortunately, aerobic rice cultivation is not popular among local paddy farmers perhaps it might due to its low yield production [12,9] or susceptible to weed [13], pest and disease [9] as compared with flooded grown rice. Many of previous work has focused on irrigated lowland rice but limited to non-puddled aerobic and limitation particularly its effects on seed quality due to environmental stress during seed development and maturity.

Instead of proper management practices [10] and suitable timing during harvesting process [14], environmental factors such as drought and, elevated temperature also need to be given attention too in order to produce high yield and, good seed quality as well as for the next growing season. Moreover, Abayawickrama et al. [15] found that grain quality was affected by grain filling characteristics formed under different temperatures. The amylose content in starch is considered an important factor for the evaluation of rice quality [16,17]. High temperature-induced expression of starch-lytic α-amylases during ripening is crucial for grain chalkiness [18]. Low seed filling rate and long seed filling duration are beneficial for starch accumulation, especially for the total starch content and amylose content [19]. This paper, therefore, seeks to further clarify the environmental effects on yield component and seed quality of aerobic rice cv. MRIA in relation to different seed development and maturation.

2. Materials and Methods

Two experiments were conducted concurrently and the plant was grown uniformly in the glasshouse using an aerobic rice cultivar. MRIA is an Indica cultivar, a moderate yielding with early harvest maturity. Seeds were obtained from the previous season which were cultivated in Universiti Teknologi MARA (UiTM) Kampus Jasin, Melaka, Malaysia and were tested for initial viability. More than 90% of the germination percentage was recorded; therefore, it was suitable to be selected as planting materials.

2.1 Plant husbandry

The plants were grown in 14 liters ERA plastic pots (30 top diameter x 20 bottom diameter x 28 cm vertical height) with drain holes. The growing media consisted of topsoil (Gajah mati series) and peat moss in the ratio 3:1. and was maintained for pH range 5.5-6.5. The pots with media were pre-soaked with tap water and then left to drain overnight. Seeds were soaked in water for 24 hours and direct sow in the seedling tray (64 holes) contained 100% peat moss. Watering was carried out once every two days until 6 DAS

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(Day after sowing). The transplanting process of the seedling was carried out on 18 August 2019 with four seedlings were planted in each pot. Then, 600g of peat moss were added on top of the soil surface to reduce the loss of soil moisture content. Pots were then irrigated by automatic drip irrigation three times each day, set up for 5-minutes nozzle discharging 200ml per minute. The application of fertilizer was followed as recommended by MARDI where granular fertilizer, NPK Green (Twin Arrow Fertilizer) containing N:P2O5:K2O (15:15:5) was added 1.4 g pot-1 at 10 and 50 DAS. Meanwhile, urea (Agrenas, Petronas) was respectively added at 25 and 50 DAS (0.6 g pot-1) [9]. Insecticide (WESCO Agencies) with 57% of Malathion was applied at 52 DAS to control aphids when detected. The timing of anthesis varied amongst the plants. Determination of 50% anthesis was recorded started from 60 DAS, with 50% anthesis was achieved at 67 DAS (0 DAA).

2.2 *Environmental stress*

The growth cabinet was maintained at 38/28°C (Exp.1) and 40/28°C (Exp.2) synchronized with 12 h photoperiod to provide high temperature. Therefore, the plant was exposed to respectively stress (HT at 38°C and/or WL) at 10, 15 and 29 DAA for 5-d (Exp. 1) or (HT at 40°C and/or WL) at 8 and 21 DAA (Exp. 2) for 7-d. The plant was brought back in greenhouse until its harvest maturity at 39 DAA (Exp.1) and 41 DAA (Exp.2). The RC-5 Data Logger was used to record the temperature meanwhile humidity was recorded by using CO2 Meter + Humidity/Thermometer (CO250) from Extech Instruments. The highest data of relative humidity recorded in the greenhouse was 93.5% while the lowest was 44.2%. Meanwhile, the highest greenhouse temperature recorded was 43.6°C and the lowest was 22.5°C.

3. Data Collection

3.1 *Dry matter production, yield and components of yield*

To minimize the variation among panicles, the only panicle emerged from 2 to 35 DAA was used for sampling. Four pots (four plants in each pots= 32 plants) were randomly selected and data for a component part of dry weights (shoot and root) were recorded. Shoot and root were dried using the constant oven (Binder ED115) at 60°C for 2 days. Panicle numbers per pot were recorded. Seeds were threshed gently by hand from panicles; remove empty seed and only filled seeds were counted and weighted. Harvest index was estimated as the ratio of seed yield to total dry matter yield.

3.2 *Seed quality assessment*

3.2.1. Seed fresh weight, seed moisture content and seed dry weight

Three samples of 10 fresh seeds were randomly sorted, counted and weighted to estimate mean seed fresh weight. Seed moisture content was estimated on the fresh weight basis using a constant temperature oven [20]. The mean of 100 seed dry weight was calculated from fresh weight and moisture content.

3.2.2. Normal germination capacity (%)

Harvested seeds were dried to 10–14% moisture content (estimated by change in sample weight). The ability of seed to germinate was assessed on samples of 150 seeds (three replicates of 50) in moist rolled paper towels at 34/11°C (16 h/8 h) for 21days [21]. Seedlings were assessed for normal seedling development [20]. Firm seeds that remained after 21 days were pricked and the test extended to 28 days to break dormancy.

3.2.3. Shoot and Root length, seed vigor index

Seven seedlings of germinated seeds in the paper towel were selected randomly and labeled by using white stickers. The final length of shoot and root (cm) of the seedlings had been recorded on day 12. The data for seed vigor index were calculated and derived from the root and shoot length and percentage of normal germination capacity.

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3.1 Amylose concentration

The percentage of amylose concentration was determined using the amylose-iodine colorimetry method performed using a spectrophotometer set up at 590nm, the corresponding of absorbance (590 nm) to 2.5mL of the test solution. Amylose content (%) = x mg amylose/100mL= x/2.5 x 100mg amylose. The result was expressed as a percentage by mass, in the milled rice on the dry basis [22].

4. Results

Table 1 shows the cumulative of aerobic rice cv. MRIA yield and its yield components for the different environmental stresses harvested at physiological maturity of 31 and 35 DAA during the experimental period. In the present study, grain yield (number of seed) in plants irrigated throughout declined from over 27% to below 18% between 10 and 29 DAA (Exp. 1), whereas combination stress (HT, WL-5 d) resulted a greater increase and lowest on plant exposed to single stress (WL for 5 d) (Table 1A). In Experiment 2, the highest grain yield was obtained from 21 DAA resulted 14% more as compared to plant irrigated throughout (control) at 8 DAA (Table 1B). The increased pattern was similar with other treatments of combination stress (HT, WL-7 d) and single stress (WL-7 d) increased to 24% and 8 % respectively. From the result, it is shown that during intermediate seed development (15 DAA), significant treatment variation was observed for combination stress (HT and WL at 5 days) resulted the lowest obtained 279 seed.

Seed fresh weight (SFW) at the end of the study from both experiments were significantly greater in the control than the plant treated with environmental stress (Table 1A & B). Reduction in SFW was pronounced in those plants exposed to combination stresses with obtained only 7.2g at 10 DAA (Exp. 1) and 9.26g at 8 DAA (Exp. 2). Significant difference ($P<0.05$) in the number of seeds produced per pot between treatments at the reproductive phase which is during early seed development (10 and 15 DAA). However, no significant difference ($P=0.686$) observed at late seed development (29 DAA). Meanwhile, the lowest number of seeds was observed under elevated temperature treatment (38°C) which is 271 and 279 at 10 DAA and 15 DAA compared to control and water limitation treatment.

The panicle number and root:shoot ratio, on a per stress basis, did not vary significantly between the DAA. However, root dry weight, harvested at 31 DAA (Exp. 1) and shoot dry weight harvested at 35 DAA (Exp. 2) showed a significant difference within environment stress variation. For the harvest index (HI), the present result showed a similar trend as the SFW whereas decreasing value of HI occurred in the plants treated with a combination of environmental stress for both experiments. The HI for plant treated to combining stress at 15 DAA (Table 1A) obtained the lowest value indicated 57% less as compared to control plant (Exp. 1). However, in Exp.2, the reduction was only 22% less - at 8 DAA (Table 2B).

Table 1. (A) Experiment 1. Yield component of Indica rice cv. MRIA freshly harvested at harvest maturity (31 DAA) from plants grown in glass house; irrigation throughout (control), combination of high temperature (HT) at 38°C and WL at 5-d and WL at 5-d respectively exposed at 10, 15 and 29 DAA. (B) Experiment 2. Yield component of Indica rice cv. MRIA freshly harvested at harvest maturity (35 DAA) from plants grown in glass house; irrigation throughout (control), combination of high temperature (HT) at 40°C and WL at 7-d and WL at 7-d respectively exposed at 8 and 21 DAA.

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DAA	Treatment	Grain yield	Seed weight Per Pot (g)	Number of Panicle	Root dry weight (g)	Shoot dry (g)	Root:Shoot	Harvest index
A								
10	Control	767a	18.9a	9	35.37a	26.90	1.42	75.55a
	HT, WL 5-d	271b	7.2b	6	14.73b	23.66	0.69	32.46a
	WL, 5-d	415ab	11.1a	6	19.85b	17.86	1.14	63.36a
15	Control	626a	19.7a	8	30.63a	27.50	1.12	74.14a
	HT, WL 5-d	279b	7.6b	6	25.43a	22.46	1.26	31.65b
	WL, 5-d	561a	16.8a	8	30.22a	22.70	1.32	74.13a
29	Control	560a	18.0a	9	31.07a	29.42	1.11	65.53a
	HT, WL5-d	528a	16.4a	10	40.80a	26.57	1.51	63.78a
	WL, 5-d	495a	15.4a	7	28.66a	24.76	1.17	62.39a
	DAA	ns	*	ns	ns	ns	ns	ns
	Treatment	**	**	ns	*	ns	ns	**
B								
8	Control	471.00	17.71a	9	41.54	63.51a	0.66	7.25
	HT, WL 7-d	301.75	9.26b	9	38.31	57.16a	0.68	9.33
	WL, 7-d	352.75	10.83b	7	45.80	51.58a	0.91	15.79
21	Control	549.75	18.74a	8	38.51	57.39ab	0.67	28.90
	HT, WL 7-d	397.25	13.69b	7	43.01	62.28a	0.70	17.70
	WL, 7-d	384.25	13.73b	8	32.80	40.75b	0.85	23.26
	DAA	ns	*	ns	ns	ns	ns	ns
	Treatment	ns	**	ns	ns	*	ns	ns

[1] ns= no significance difference; *=significance difference at $p<0.05$; **= significance difference at $p<0.01$.

The results of the seed quality experiment as aerobic plant treated to environment stress for two different harvested physiological maturities (31 and 35 DAA) are presented in Table 2.

Seed dry weight was inhibited by HT and WL treatment (Table 2). After 10, 15 and 29 DAA of HT and WL treatment for five days, the seed dry weight of aerobic seed was reduced by 22.5%, 16.9% and 12.1%, respectively (Table 2 A). Meanwhile, after 8 and 21 DAA of similar combination stress for seven days, the seed dry weight of aerobic seed was reduced to 23.2% and 11%, respectively (Table 2B). The reduction of seed dry weight was more pronounced in combination environment stress (HT, WL) than that in single stress (WL), also pronounced in earlier plant exposed to stress (Table 2A, B).

Seed moisture content at harvest showed a slight decrease as the plants were exposed to stress. The range of entirely seed moisture content obtained was between 10.5%-13.8% (Table 2A) and 11.6-13.7% (Table 2B). Seed moisture content in the plants showed no difference from those plants exposed with environmental stress, whereas the combination of HT and ending irrigation at 5 days at 15 or 29 DAA resulted in greater declines compared with control or single stress (WL at 5 days). Combined stress at 15 or 29 DAA had led to rapid loss in moisture with seed from irrigation throughout and WL for 5 days equilibrating at about 12-13% moisture content.

No significance difference occurred for shoot length for the plants treated to environmental stress, however, it occurred in root length. In the HT and WL for 5 days treatment, root length was reduced up to 3.2%, 25.1% and 10.5%, respectively, as the plants were exposed to stress at 10, 15, and 29 DAA compared to the untreated plants (Table 2A). The reduction of root length was more severe as the plants treated for consecutive 7 days of stress with 48.9% and 55% for WL while for combination stress (HT and WL) with 59.4% and 48.9% as compared to control (Table 2B).

Germination capacity and vigour index were significantly affected by environmental stress treatment (Table 2A, B). However, either both or single stress did not reduce much while treated at 29 DAA. Interestingly, vigour index obtained the highest value (1434) once treated WL-7 days at 29 DAA, harvested at 31 DAA (Table 2A). The result in Table 2B shows that, the highest percentage for seed to germinate normally (harvested at 35 DAA) was 97% which was obtained from the untreated plants. In addition, vigour index showed a significant decrease with a reduction up 70.3% at 8 DAA and 58.9% at 21 DAA as a result of HT and WL for 7 days.

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Table 2. (A) Experiment. 1. Seed quality of Indica rice cv. MRIA freshly harvested at harvest maturity (31 DAA) from plants grown in glass house; irrigation throughout (control), combination of high temperature (HT) at 38°C and WL at 5-d and WL at 5-d respectively exposed at 10, 15 and 29 DAA. (B) Experiment 2. Seed quality of Indica rice cv. MRIA freshly harvested at harvest maturity (35 DAA) from plants grown in glass house; irrigation throughout (control), combination of high temperature (HT) at 40°C and WL at 7-d and WL at 7-d respectively exposed at 8 and 21 DAA.

DAA	Treatment	100 seed dry weight (mg)	Seed moisture content (%)	Shoot length (cm)	Root length (cm)	Germination capacity(%)	Seed vigor index
A							
10	Control	3.15	12.7	9.79	7.91	91.3	1622.9
	HT, WL 5-d	2.44	10.5	11.09	7.66	64.0	1182.3
	WL, 5-d	2.63	13.3	9.11	6.96	67.3	1102.6
15	Control	3.14	13.9	10.12	7.80	91.3	1553.2
	HT, WL 5-d	2.61	13.2	6.74	5.84	65.3	822.0
	WL, 5-d	2.87	13.8	8.02	6.69	71.3	1047.7
29	Control	3.06	13.8	8.45	6.66	90.7	1383.0
	HT, WL 5-d	3.02	11.8	9.20	5.96	90.7	1373.0
	WL, 5-d	3.06	12.6	9.13	6.75	87.3	1434.0
	DAA	**	ns	**	**	ns	**
	Treatment	**	**	ns	**	**	**
B							
8	Control	2.93	13.7	6.63	6.14	94.7	1209.2
	HT, WL 7-d	2.25	12.7	2.27	2.49	60.7	358.9
	WL, 7-d	2.89	12.3	2.48	2.76	91.3	478.2
21	Control	3.00	13.0	6.66	6.34	97.3	1265.6
	HT, WL 7-d	2.67	13.8	3.33	3.24	58.7	519.9
	WL, 7-d	2.81	11.6	2.94	3.14	92.0	558.8
	DAA	**	ns	**	**	ns	**
	Treatment	**	**	**	**	**	**

ns= no significance difference; *=significance difference at $p<0.05$; **= significance difference at $p<0.01$.

The amylose content was increased significantly in the seed while the plants were treated to single stress (WL for 5 days) for subsequent DAA – 10,15 and 29 DAA, whereas the highest increased was more pronounced in 29 DAA with 16% (Figure 1A). However, the amylose content did not differ between the untreated and treated plants in the seeds at 10 and 15 DAA (Figure 1A) or even prolonged the stresses for consecutive 7 days (Figure 1B) at 8 and 21 DAA. Based on these results, we inferred that the amylose content in the seeds was significantly build-up by the WL 5 -d treatment.

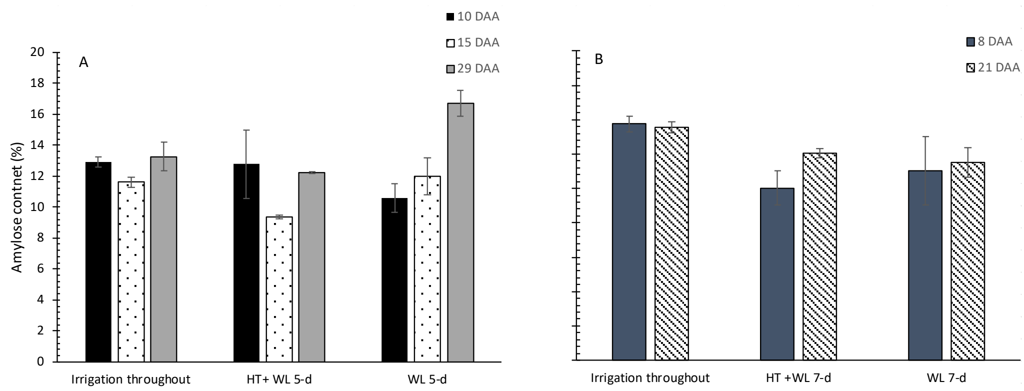

Figure 1: (A) Experiment 1. Seed Amylose content of Indica rice cv. MRIA freshly harvested at harvest maturity (31 DAA) from plants grown in glass house; irrigation throughout (control combination of high temperature (HT) at 38°C and WL at 5-d and WL at 5-d respectively exposed at 10, 15 and 29 DAA. (B) Experiment 2. Seed Amylose content of Indica rice cv. MRIA freshly harvested at harvest maturity (35 DAA) from plants grown in glass house; irrigation throughout (control), combination of high temperature (HT) at 40°C and WL at 7-d and WL at 7-d respectively exposed at 8 and 21 DAA.

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ns= no significance difference; *=significance difference at p<0.05; **= significance difference at p<0.01.

4.1 Relationship between Amylose content between germination capacity and seedling vigor index

In both experiments, the responses between the germination capacity (Figure 2A) and seedling vigour index (Figure 2B) and the percentage of amylase content were linear. The plant stress at consecutive 7 days exposed at 8 and 21 DAA were substantially more than in plant stress for 5 days at 10, 15 and 29 DAA. The slope of the lines was generally highest at plant treated for stress for 7 days at 8 and 21 DAA with R2 = 0.25 than during 10, 15 and 29 DAA for 5 d with R2 = 0.14.

Figure 2A indicates that there is an inverse relationship at p<0.05 between germination and amylase content percentage resulted from environment stress for 7 d at 8 and 21 DAA with R2 = 0.19. Meanwhile, a positive correlation at p<0.01 was present from in the plants treated for 5 days at 10, 15, and 21 DAA with R2 = 0.41

There was a positive correlation between Amylase concentration and germination capacity for MRIA1 with a significant value at p = 0.017. A higher germination capacity increases the total amylose content as shown in Figure 2 A. Meanwhile, Figure 2B shows a positive correlation between amylose concentration and seedling vigor index for MRIA1 with a significant value at p = 0.036 for the plants harvested at 89 DAS, however, showing a negative correlation for the plants harvested at 91 DAS.

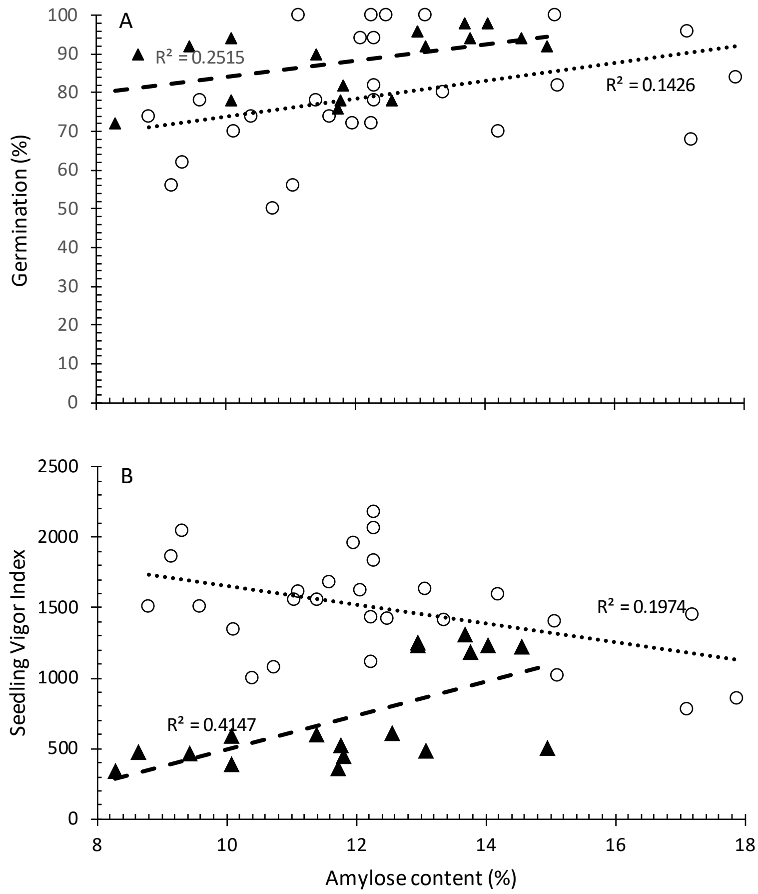

Figure 2: Comparison of Seed Amylose content in relation with ability to germinate (dried-seed) (a) and seedling vigor index (b) for seeds of Indica rice cv. MRIA harvested at maturity (89-91 DAS) from plants grown in glass house from all 15 rice seed lots produced in August 2018. Open symbols

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(○) are plants exposed for 5-days, at 10, 15, 29 DAA and solid symbol for 7 -days at 8 and 21 DAA (▲).

5. Discussion

Comparison amongst treatments showed considerably and consistent effects of the two environmental stresses namely the WL and HT treatments during seed development and maturation (Table 1A & B; Table 2A & B; and Figure 1 & B). The combination of WL and HT treatments resulted more detrimental effects; particularly, early seed development and considerably poorer seed quality later on.

The number of seeds produced was not affected by environmental stresses at 29 DAA. As expected, environmental effect pronounced in early of seed development (10 &15 DAA) and damaged by the combined WL and HT stresses. During a high temperature, generally soil will dry and plant will wilt due to high transpiration rate. In order to reduce the transpiration rate, stomata will close and lack of CO2 in plants to do photosynthesis to produce carbohydrate. In this state, we can see the plant suffer from water stress when the demand of water exceeds the available amount. In order to cater this problem, farmers are advisable to keep regular watering of rice plants: particularly during early seed development. However, farmers do not have to worry if the temperature rises up to 38ºC during late of seed development or lack of water for 5 days as the present study suggested that during late seed development, rice plants can adapt with those stresses. Othman et al. [23] mentioned that aerobic rice can be harvested as early as 90 DAS as it has reached its physiological maturity and fully ripe. On the contrary, in normal environmental (without environmental stress (<38ºC), the present study suggested to harvest cv. MRIA as early as 10 DAA (70 DAS) to get a higher yield production (Table 1A). However, these results still yet could not be considered as the final word on aerobic cultivar as a whole, as much more need to be known about this aerobic cultivation system before drawing firm and broadly generalized conclusions.

The greater part of the water required by crops is met by uptaking from soil through the root system. Greater root:shoot ratio means a greater root density and root interception for nutrient uptake. The present results (Table 2A & B) showed that root:shoot ratio had no significant difference to environment stress, not even in early or late seed development. However, this study showed that the combination of stresses (HT + WL) resulted the value of root:shoot ratio substantially increased by seed development maturity. As plant response to water stress, root elongates more compared to the shoot as deep root penetration increases the water and nutrient absorption to survive. This is one of plant mechanisms to cater the problem in a short period of time while treating water stress. Generally, root will elongate when it is in water stress in order to increase the absorption of water as mechanism of plant survival. Therefore, this study indicated that, MRIA is a type of cultivar that tolerates with water limitation which is supported the study conducted by [9,23].

Harvest index was estimated as the ratio of seed yield to total dry matter yield. Dry matter is produced as a consequence of photosynthetic uptake of carbon dioxide through stomata. This was supported by Yang and Zhang [24], who showed that water stress during grain filling increased the remobilization of assimilate from the rice straw to grains and resulted in early senescence. Furthermore, water stress during grain filling induces the conversion of stem reserves into soluble sugars and the transport of these sugars into the grains [25]. Nicolas et al. [26] reported seed dry weight was reduced in response to early drought due to the development of a smaller number of endosperm cells. In most cases, higher temperatures during the grain-filling period increase the grain dry matter accumulation rate but shorten the grain-filling period [1]. A study conducted by Zaman et al. [27] reported that yield components (panicle number, grain yield and 100-grain weight) for MRIA greatly decreased when water stress was imposed at the panicle initiation stage; however, not much effect during late seed development due to negligible effect on water loss and Water Use Efficiency (WUE) [28]. During seed development, starch content decreases significantly once it is subjected to drought due to insufficient supply of

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photoassimilation [29] resulted a reduction in grain weight [30] and low of amylose content [31].

IRRI [32] reported that rice seed harvested from paddy fields at optimum grain maturity have average moisture content about 20–25 %. Higher moisture content results in more losses from poor grain quality while lower moisture content results in more losses from shattering [33]. Since the seed in this study had already been dried naturally in planta to below 14% moisture content (Table 2), further desiccation was not required for storage. ISTA [34] reported that rice seeds, which can withstand drying down to low m.c. of around 5% to 10% and successfully stored at low freezing temperature for long periods. In addition, in terms of seed moisture content at harvest maturity, this study suggests that cv. MRIA was well adapted to drought and high temperature stresses. MRIA cultivar was more damaged by drought and high temperature stress combined, with only 60% (8 DAA) and 58% (21 DAA) obtained for normal germination (Table 2B) with seed moisture content of 12.0% and 13% respectively at harvest. This low viability after mother plant stress is supported by Martínez-Eixarch and Ellis [35] who reported that the viability of Gleva seeds was reduced by extreme-temperature (38/34°C) treatments applied in the 7 or 14 d immediately after anthesis. Similarly, another study conducted by Bukharov et al. [36] reported that the duration of heat at 40°C may lead to a decrease in viability of dill (Anethum graveolens) which vary from 1-5 days of exposure.

Seedling vigor index also was seriously affected under elevated temperature (38°C) during the early and mid-seed development at 10 DAA and 15 DAA (Table 2A & B). As the standard germination test does not consistently predict the field performance of a seedlot, seed vigor index can be used to determine seed quality as defined as potential to perform in rapid uniform emergence and development of normal seedlings under a wide range of field conditions' [37]. The maximum potential for seed vigour expression in most crops is achieved when the seed is at its maximum dry weight, a stage known as physiological maturity [38]. A similar result was discovered in this present study, where high seed dry weight and expressed good in seedling vigour index obtained. However, the seed vigour assessment requires ongoing studies in order to develop more thorough understanding of what causes seed deterioration that leads to low seed quality.

Environmental stress during late seed development (29 DAA) or maturation had no effect on rice yield production and plant harvest index. Therefore, during early and mid-seed development are important stages to avoid from any environmental stress similar reported by Abdul Rahman and Ellis [1]. The present study showed that the highest SDW incurred the highest GC and SVI (Table 2A & B). Besides, the present study also showed that the lowest 100- SDW respectively produced only 2.44g and 2.61g as exposed to elevated temperature treatment (38°C) while the highest 100- SDW for both experiments obtained 3.15g and 3.14g., respectively, which were dominated by control. From this control data, our present study obtained about 25% more weight as compared to a previous study conducted by Zaman et al. [12]. Physiological maturity (PM) was defined by Shaw and Loomis [39] as the stage in seed development when the seed reaches its maximum dry seed weight and yield. Therefore, this hypothesis was consistent with our present study, whereas, 100-SDW for control was not increased at 15 and 29 DAA (Table 2A), indicated that during 10 DAA, the plant has reached physiological maturity. As the PM reached, the seed is no longer affected by environmental stress [1].

The combination of drought and high temperature represented an excellent example of two different abiotic stress conditions that occur in the field simultaneously [40,41]. The combination stress (HT + WL) was detrimental damaging during seed development but had no effect at later seed development (29 DAA). This was consistent with the finding by Abdul Rahman and Ellis [1] which reported that combining drought and HT would damage seed quality more than each stress alone, and more so in the Japonica cv. Gleva than the Indica cv. Aeron 1. Seed quality was vulnerable to plant drought and HT at anthesis and histodifferentiation [42]. The seed quality of aerobic rice (Oryza sativa L. subsp. indica) cv. Aeron 1 seed is particularly sensitive to high temperatures – and thus climate change – during development and maturation and more resilient to damage from drought

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and HT stresses than the Japonica whereas combining both stresses would damage seed quality extremely than each stress alone [1].

Those plants were treated with the combination of stresses (HT+WL) for 7 consecutive days exposure. This present finding is similar with a study conducted by Martinez-Eixarch and Ellis [36], which reported that seed viability was most vulnerable by 7 days of exposure to high temperature (38C) in the 7 or 14 DAA, when histodifferentiation occurs. During seed development, seed are in the process into filling with its composition and this is influenced by various metabolic processes. At these processes, photoassimilate production and translocation are highly sensitive to drought and elevated temperature due to involvement of array of diverse enzymes [43]. The severity of seed quality damage by brief period of HT also depends on the duration imposed. In rice, for example, spikelet fertility was reduced from 90% to 20% by only 2h exposure to 38°C and 10% by <1h exposure to 41°C; therefore, seed quality in rice may reduce [7].

Water limitation at 5 days during late seed development obtained the highest amylase concentration. Aerobic rice used in this study (MRIA) was created to be tolerant to drought as well as high temperature, however, the present study proved that this cultivar was only tolerate with water limitation during late seed development. This might occur due to the plants had achieved physiological maturity and the seeds had been fully filled.

Highest germination percentage (97%) was obtained by the seed harvested at later harvest (25 DAA) from the control plants. Similar reports by Abdul Rahman and Ellis [1] highlighted that early harvest reduces ability for seeds to germinate normally and increases seed dormancy [44]. During rice seed germination, amylase concentration in aleurone layer is essential to perform starch degradation therefore provides the energy to initiate germination and seedling establishment [45,46]. This present study proved that elevated temperature (Figure 1B) during seed development resulted a low in amylose concentration. As this happen, it led to decrease in starch accumulation which important for initiate shoot and root emergence. This result similar with study reported by [47] as amylose content was reduced to 22% as exposed to high temperature during gain filling stage. Drought during anthesis stage (Figure 1A &B) affected the process of amylose accumulation and reduced the total amylose accumulation. The result of this study also consistent with Singh et al. [32] who discovered that, the drought stress resulted in small seed size and lowered amylose contents in starch grains. Starch content of paddy grain production decreases significantly once plants have been subjected to drought stress due to insufficient supply of photoassimilation or direct effect on starch synthesis in grains which are affected by sink dehydration [30].

Environmental stresses (HT and drought) affect to starch accumulation, particularly for the total amylose content. Moreover, the results also showed that the amylose content is positively related to GP and seed vigor in MRIA1. This finding was similar with a study reported by Wang et al. [48], whereas the higher GP obtained from seed containing high of amylose content. Thus, the high amylose content in seeds was responsible for the higher GP and VI of MRIA1. This indicated that, prolonged stress pronounced the highest seedling vigor index when the amylose content was low in concentration. In contrast, it was vice versa for 5 days stresses, whereas the highest germination percentage was obtained from the highest amylase content concentration. Tang et al. [49] found that temperatures during grain development had a significant effect on the accumulation of amylose and amylopectin. Kato et al. [50] reported that the higher temperature decreased the apparent amylose content and increased the ratio of short to long chains of amylopectin. Aerobic rice cv. MRIA can be confirmed as plant tolerate with water limitation during seed maturity development and elevated temperature, produced better in yield component and enhanced seed quality development as well as amylase content concentration.

In conclusion, 5 or 7-days exposures to WL or HT at 38 ºC or 40ºC were damaging yield and seed quality of aerobic rice cv. MRIA. The negative effects on final yield component, seed quality and amylase content were through effects on seed set, with little evidence of any direct effect either 5 or 7 days exposure to WL or HT during the seed-filling

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periods. The aerobic rice is considered a promising cultivation production system in water scarce environments.

6. Conclusions

In conclusion, this study suggests that, seed quality of seed lots of MRIA is reduced considerably more by the combined stresses than by either stress alone. The combination of both stresses had a significantly greater detrimental effect on yield component and rice seed quality as well as amylase content, particularly during histodifferentiation- early seed development while successively later exposures were harmless. Greater tolerance to heat and drought stress are priority strategies for rice seed quality in order to maintain future yield by adapting to climate change. Hence, overcoming the effects of high temperature and water stress on rice production is essential for food security in the future. As recommendations, aerobic rice cv. MRIA can be cultivated in water limitation areas. It is recommended to avoid HT and ensured to irrigate throughout the MRIA rice plant during histodifferentiation as well as to save water usage during early seed maturation.

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