

**Detrimental effects of heat stress on grain weight  
and quality in rice (*Oryza sativa* L.) are aggravated  
by decreased relative humidity**

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## Abstract

Increasing heat stress coupled with decreased relative humidity (RH) has raised concerns regarding the risk of rice production under global warming. However, it remains largely unknown how rice yield and quality are affected by heat stress coupled with decreased RH during grain filling stage. To investigate this, experiments were conducted under controlled environments of growth chamber on six rice cultivars varying in heat tolerance using 12 combinative treatments of three factors: two RH levels (75% and 85%)  $\times$  three temperature levels (daily maximum temperature at 33°C, 35°C and 37°C)  $\times$  two durations (8 d and 15 d after anthesis). Results show that RH75% at temperature treatments significantly reduced grain weight, which was higher than RH85%. The same trend was also observed for both head rice rate and chalkiness. Even for R168, the most heat-tolerant cultivar, there were some differences for these traits between the two RH regimes. The lower RH caused the most detrimental effect at 35°C, and to a lesser extent at 33°C, but a negligible effect at 37°C. The result of this study sheds some light on better understanding the interaction effects of temperature and RH on rice quality during the grain filling stage, suggesting that RH should be taken into consideration in heat tolerance screening and identification to facilitate rice breeding and genetic improvement.

## Introduction

~~Average~~The average global surface temperature was estimated to have risen by 0.85°C over the period of 1880–2012 (Stocker et al. 2013). As land is warming faster than the ocean, evaporation on land greatly is increased. But due to limitation of soil moisture, water supply, and crop transpiration, near-surface relative humidity (RH) over land tends to decrease substantially. As a result, most land areas, except coastal areas, will become drier (Byrne & ~~O'German~~O'Gorman 2016; Dai et al. 2018; Orimoloye et al. 2018; Po-Chedley et al. 2018).

Rice is one of the most important staple cereals, and it feeds more than half of the world's population (Seck et al. 2012). However, rice yield fluctuates considerably, as rice is susceptible to climate change (Jagadish et al. 2015; Yan et al. 2017). With each 1°C increment in whole-season minimum temperature, there is a 10% yield loss (Peng et al. 2004). In addition to this detrimental effect of heat stress on rice yield, heat-induced damage in grain quality is also a serious problem, leading to considerable economic losses (Lyman et al. 2013).

~~Grain~~The grain filling stage, one of the most critical periods for rice yield and quality formation, is sensitive to environmental stress (Yoshida 1981). High temperature during this period results in accelerated grain filling rate, shortened grain filling duration, lowered grain weight, and/or deteriorated milling quality from the increased amount of chalky and fissured grains, due to high temperature-forced maturity (Ambardekar et al. 2011; Bao 2019; Cooper et al. 2006; Zhou et al. 2019). This is particularly the case during the early and middle grain-filling periods, especially the first 15 days, which is thought to be most susceptible to high-high-temperature stress (Cooper et al. 2008; Cooper et al. 2006; Wu et al. 2016). Although an optimum temperature ranging from 25°C to 29°C in the first 15–20 d of the grain-filling stage has been suggested, it is subject to cultivars varying with their varietal tolerance (Abayawickrama et al. 2017; Morita et al. 2016; Wu et al. 2016).

Under high temperatures, the starch anabolic pathway is inhibited in the developing grains (Yamakawa & Hakata 2010; Yamakawa et al. 2007); many immature starch granules are observed in the endosperm cells (Zakaria et al. 2002). Also, altered expression of  $\alpha$ -amylase genes and increased enzymatic activity were detected, suggesting that the starch is degraded under elevated temperatures (Hakata et al. 2012). Besides, compared to a heat stress-tolerant cultivar, sucrose transport and metabolism

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in the phloem of the leaf, sheath, stem, and grains are inhibited in a heat stress-susceptible cultivar under ~~heat-heat~~-stressed environment (Tanamachi et al. 2016; Zhang et al. 2018a). Thus, heat stress causes a starch shortage, reduced grain weight, and poor quality.

With regards to heat-induced damage in grain yield, panicle, a major determinant for yield stability and grain quality, is greatly conditioned by ~~high-high~~-temperature stress during the grain filling stage. Physiologically, panicle development is not only influenced by air temperature but also by air humidity (Weerakoon et al. 2008). Compared with humidity conditions in Hubei, China, lower RH conditions (RH15%) in New South Wales, Australia, to some extent were reported to alleviate heat-induced damage in rice through '~~transpiration cooling~~'-cooling' during the flowering stage (Matsui et al. 2014; Tian et al. 2010; Yoshimoto et al. 2012). But in the grain filling stage, hot and dry wind conditions can accelerate water loss in panicles and increase the chalky grain rate (Hiroshi et al. 2012; Kang et al. 2003). Oya & Yoshida (2008) also found a clear varietal difference in chalky formation among varieties with wind treatments. It is still controversial how rice yield and quality ~~is-are~~ affected by heat stress, coupled with different RH during the grain filling stage.

Endosperm cells of rice grain rely on osmotic adjustment to maintain cell turgor (Wada et al. 2019) under heat stress, which is similar to rice grown under dry wind (Wada et al. 2014; Wada et al. 2011). This suggests the existence of a similar mechanism between high temperature and dry wind responses. Besides, lower RH in the air also reduces head rice yield (F. Thompson & G. Mutters 2006). For example, in northern India, RH showed a significant decline by 1.44% per year, which in turn significantly affected the yield of almost all crops (Chakraborty & Hazari 2017). It is estimated that for every 1% decrease in average RH during the growing season in China, rice yield decreases by 0.75% (Zhang et al. 2017). These studies indicate that RH variation during grain filling stage may make a big difference to rice yield and quality formation, but whether such difference would be aggravated by the interaction of RH with temperature among cultivars remains to be tested.

In the Jiangnan basin of the middle reaches of the Yangtze River valley of central China, mid-season *indica* rice varieties often suffers yield loss from heat stress (Tian et al. 2010), and increased temperature in summer is always accompanied with decreased

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humidity (Gong et al. 2006; Guo et al. 2016). For example, when the maximum temperature of approximately 35°C or daily mean temperature of 30°C lasts for more than ~~3~~three days, RH decreases from ~85% to ~70% (Tian et al. 2010). With climate change, such a high temperature in this region has become longer in recent years (Liu et al. 2019; Tan & Shen 2016). This study hypothesized that high temperatures~~s~~s, coupled with decreased RH~~s~~s, aggravates the adverse effects on rice grain quality. To verify this hypothesis, experiments were conducted in a controlled environment with six rice cultivars varying in heat stress tolerance. The objectives were to investigate 1) the effect of RH on high temperature-induced grain weight and quality loss; 2) varietal differences under combinations of RH temperature treatments.

## Materials & Methods

### Plant materials

Six rice cultivars with different heat tolerance during the grain filling stage were sourced from our previous studies. They include three rice hybrids – Liangyou27 (LY27), Liangyou6 (LY6), Zhuliangyou47 (ZLY47), and three conventional varieties – 16343, R168, and IR64. R168 showed smaller grain weight and quality difference between temperature conditions, IR64, and the other four varieties showed sensitivity to high temperature (data showed in Table S1).

### Growth conditions

A pot experiment was conducted in 2017 on the experimental farm of Yangtze University (Jingzhou City, 112°09'E, 30°21'N, 32 masl) in the western part of the Jiangnan Basin in China.

Seeds were sown on April 8, 2017; 20-day-old seedlings were transplanted to each plastic pot (inner diameter 30 cm, height 30 cm) containing 12.5 kg soil and 8 g N:P:K compound fertilizer (26:10:15). After transplantation, the soil surface in the pots was kept submerged until maturity in the natural conditions. Tillers were cut off once emerged, leaving only the main stem in each plant throughout the whole growth duration.

### Treatment

Six controlled environments with the combination of air temperature and relative humidity (RH) were simulated and implemented in a growth chamber (AGC-MR, Zhejiang Qiushi Environment Co., Ltd., Zhejiang, China). Experimental treatments were two RH levels (i.e., RH75% and RH85%), three temperature levels (daily maximum temperatures of 33°C, 35°C, and 37°C), and two durations (8 and 15 days after anthesis), which are shown in Fig. 1. Hourly temperature changes during the day are shown in Fig. 1. RH in each treatment environment was kept constant.

Panicles heading on the same day were marked for treatments after anthesis. Six pots of each cultivar were subjected to each combination of the controlled environment (temperature × RH combinations) for eight and 15 days, respectively. The plants grown under natural conditions (six pots per cultivar) were used as controls; the temperature and RH are shown in Table S2.

#### Measurements of grain weight, milling quality, and chalkiness

Filled grains were collected and sun-dried to the moisture content of 13% after harvest. Thousand-grain weight with three replications was measured. After 3-month storage (room temperature: 15–20°C, RH: 10–20%), milling quality and chalkiness were measured. Rice grain samples (30 g for each cultivar per treatment) were dehulled and then polished for 30s (JDMZ100, Beijing Dongfu Jiuhe Instrument Technology Co., Ltd., China) to obtain milled rice, from which head rice was separated and then weighed. The weight of the head rice to the sample weight (30 g) is calculated as the head rice rate. The Chalkiness of head rice was evaluated with a rice appearance quality tester (JMWT12, Beijing Dongfu Jiuhe Instrument Technology Co., Ltd., China).

#### Stress tolerance estimation

Stress tolerance was evaluated by the membership function value (MFV) based on the theory of fuzzy mathematics (Zadeh 1965). The method was modified from by Chen et al. (2012) and Liu et al. (2015). The heat-tolerant coefficient (HC) was calculated as the ratio of the value in the combinative treatment of temperature × RH × duration to that in the control of the same cultivars for individual traits, using the following equation.

$$HC_{ijk} = \frac{X_{ijk}}{CK_{ij}}$$

Where  $HC_{ijk}$  is the heat-tolerant coefficient of the trait (i) for cultivar (j) in treatment (k),  $X_{ijk}$  is the value of the trait (i) for the cultivar (j) in the treatment (k),  $CK_{ij}$  is the value of the trait (i) for the cultivar (j) under the control condition.

As with HC, MFV for grain weight and head rice rate were calculated, following the equation of:

$$MFV_{ijk} = \frac{HC_{ijk} - \min(HC_i)}{\max(HC_i) - \min(HC_i)}$$

Where  $MFV_{ijk}$  is the membership function value of heat tolerance of the trait (i) for the cultivar (j) in treatment (k),  $HC_{ijk}$  is the same as earlier defined,  $HC_i$  is the heat-tolerant coefficient value of the trait (i) over all cultivars and all treatments.

Considering that the value of chalkiness tends to increase after treatment, which is

contrary to changes in grain weight and milled rice rate, and that the absolute value of chalkiness under control conditions is usually very small, MFV for chalkiness was calculated following the equation of:

$$MFV_{ijk} = 1 - \frac{X_{ijk} - \text{Min}(X_i)}{\text{Max}(X_i) - \text{Min}(X_i)}$$

Where  $X_i$  is the value of the trait (i) over all cultivars and all treatments.

For these three traits, MFVs are dimensionless, real number interval [0,1], standing for individual ~~cultivar's~~ cultivar's heat tolerance under treatments.

### Data analysis

Microsoft Excel 2019 was used for data entry and collating. Analysis of variance (ANOVA) by using the R package ~~'agricolae'~~ 'agricolae' in R 3.6.0 was used to determine the effect of treatment factors on grain weight and quality. Comparisons between treatment means were performed using the least significant difference test (LSD) at  $P \leq 0.05$ . Function ~~'lm'~~ 'lm' in R package ~~'stats'~~ 'stats' in R 3.6.0 was used for multiple linear regression analyses between grain weight and treatment factors, head rice rate and treatment factors, and chalkiness and treatment factors, respectively. Multiple linear regression formula was  $Y = a*X1 + b*X2 + c*X3 + d*X4 + e$ , in which  $Y$  represents grain weight, head rice rate, and chalkiness in each treatment, respectably;  $X1$ , the max daily temperature in each treatment;  $X2$ , relative humidity in each treatment;  $X3$ , treatment duration days in each treatment; and  $X4$ , grain weight, head rice rate and chalkiness of each cultivar under controlled condition to eliminate the differences in the variety itself; with  $e$  representing the intercept.



## Result

### Grain weight

~~Effects~~The effects of temperature, RH, and temperature  $\times$  RH interaction on grain weight were significant among six cultivars (Table 1). Durations also had a significant effect on grain weight except for R168. Grain weight was significantly reduced in cultivars at RH 75% coupled temperature treatments, and the reduction was higher than that in RH 85% over the same three temperature treatments (Fig. 2). Under the same temperature and RH conditions, grain weight significantly decreased from 8 d to 15 d of the duration treatment (Fig. 2). The impact of different RH regime treatments (RH75% vs. RH85%) on grain weight was smaller at 37°C than at 33°C and 35°C (Fig. 3).

Multiple regression analysis showed that grain weight decreased when temperature and durations increased at RH75%, which was just the opposite to that at RH85% (Table 2).

MFVs of each trait in Table 3 showed heat tolerance of each cultivar under treatments, with the mean value as a comprehensive index for evaluation of heat stress tolerance of each cultivar. R168 was found to have the highest mean MFV (0.70), followed by IR64 (0.55), then LY27, LY6, ZLY47, and 16343 (0.54, 0.50, 0.45, and 0.44, respectively). R168 also showed the smallest difference of MFVs between RH conditions. Of all 12 treatment conditions, the treatment of 35°C by RH75% by 15 days showed the largest mean MFV difference among cultivars, which could be used to evaluate high-temperature tolerance.

R168 and IR64 were also the two cultivars with the highest mean MFVs of grain weight and the smallest difference between RH conditions (Table 3). Their grain weight decreased  $>2$  g only at 37°C (Fig 2). ZLY47 and 16343 showed high MFVs under treatments of 33°C, and their grain weight decreased  $>2$  g at 35°C or 37°C. LY27 and LY6 showed small MFVs at RH75% over temperature treatments; ~~their~~ their grain weight decreased  $> 2$  g at all three temperature regime treatments.

### Head rice rate

The Head rice rate of all cultivars was significantly affected by temperature and RH (Table 1). Durations also showed a significant detrimental effect on the head rice rate except in cultivar ZLY47. Interactive effects of temperature  $\times$  durations and RH  $\times$  durations were significant for the head rice rate among cultivars except for LY6. Compared to the control, the head rice rate significantly decreased in cultivars at RH75%

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over temperature treatments, and the reduction was higher than that at RH85% over the same temperature treatments (Fig. 4). ~~Difference~~The difference in head rice rate between these two RH treatments (RH75% vs. RH85%) was the greatest at 35°C and the lowest at 37°C (Fig. 3), suggesting that RH has a pronounced effect on temperature-induced head rice loss at 35°C. At the treatment of 33°C, a pronounced difference was found only in cultivars whose head rice rate was dramatically decreased (LY6 and ZLY47, Fig 4). R168 showed the most heat-tolerant in terms of head rice rate, except for the treatments of 37°C × RH 75%, with an MFV of >0.70 (Table 3). In addition, R168 showed a stable and higher head rice rate than other cultivars, even at 37°C (Fig 4). The MFVs of head rice rate for 16343 was ≥0.85 at 33°C but dropped sharply at 35°C in both humidity treatments (Table 3). For other cultivars, the temperature caused a sharp drop in response to variable RH: under RH 75% over temperature treatments, the temperature was 33°C for LY6 and ZLY47, or 35°C for LY27; under RH85%, the temperature was 35°C for LY6 and ZLY47, or 37°C for LY27.

### Chalkiness

For all cultivars, the temperatures, RH, and the interaction of temperature × RH had a significant effect on chalkiness (Table 1). The durations showed a significant effect on chalkiness for cultivars except 16343. For most cultivars, even with the same temperature and duration treatment, chalkiness was higher at RH75% than at RH85% (Fig. 5). The difference in chalkiness between RH treatments (RH75% vs. RH85%) was most pronounced at 35°C, followed by that at 33°C (Fig. 3). But at 37°C, the difference was the least. From the multiple regression analysis, chalkiness would be higher when the temperature rose, RH decreased, or durations increased (Table 2). R168 and IR64 showed the highest MFVs of chalkiness in all treatments (Table 3). R168 also showed the smallest chalkiness difference between RHs (Fig 5). Compared to the control, the chalkiness of R168 sharply increased (more than 10%) only at 37°C by the RH treatments; this was the case for IR64 at 35°C and 37°C, or for the remaining cultivars at all three temperature treatments (Fig 5).

## Discussion

An upward trend of long-term ~~high-high~~-temperature stress coupled with decreased RH would occur during rice grain filling in the Yangtze River basin, due to the climate change (Guan et al. 2015; Tan & Shen 2016; Zhang et al. 2018b). Although some studies have shown that hot and dry wind resulted in rice yield and quality loss (Hiroshi et al. 2012; Wada et al. 2014), the sole effect of RH remains unknown. Besides, meteorological factors are interrelated under field conditions; for example, in the summer season between late July and middle August in the middle reaches of the Yangtze valley, high temperature generally is coupled with low RH, and rainy or cloudy days come with high RH (Guo et al. 2016). Therefore, it is difficult to separate the effect of humidity on crop yield from the complex meteorological factors in a field experiment (Yoshida 1981; Zhang et al. 2017; Zhao & Fitzgerald 2013). In the current study, controlled environmental conditions were used to simulate three temperature with two RH regimes, which provided conditions to evaluate the effects of RH from those of air temperatures on high temperature-induced rice yield and quality loss during the grain-filling stage. Both temperature and RH ~~showed-had significantly main-~~ pronounced effects on grain weight, head rice rate, and chalkiness; temperature  $\times$  RH combination showed significantly interactive effects on grain weight and chalkiness (Table 1). Temperature treatments at RH75% significantly reduced grain weight, and the reduction was greater at RH85%. The same trend was also observed for grain quality loss. Our study is consistent with the work of Wada et al. (2011), who found grain appearance quality loss under the condition of dry, hot wind.

### The interaction effect of temperature and RH

During the flowering stage, panicle temperature rather than air temperature was curvilinearly related to spikelet fertility (Weerakoon et al. 2008). Rice could adjust panicle temperature homeostatic via transpiration cooling for its optimal growth. The air-panicle temperature difference altered under different RH conditions (Fukuoka et al. 2012; Yoshimoto et al. 2011). Yan et al. (2008) showed that under the temperatures of 31.5–33.5°C, the temperature difference between air and panicle was about 2°C under humid atmospheric conditions (~86% RH) or about 5°C under a dry atmospheric condition (~48% RH). Rice panicle benefits from its transpiration cooling under heat stress at flowering (Matsui et al. 2014). However, our results showed that a decreased

RH coupled with a high temperature at grain filling stage reduced grain weight and lowered grain quality. It means the benefit from the decreased RH by transpiration cooling may not be the only scenario of RH and temperature interaction.

Theoretically, there may be a balance between transpiration cooling and water deficit in rice panicles exposed to heat. Zhao & Fitzgerald (2013) reported that at daily maximum temperatures ranged from 30 °C to 33 °C, a lower RH led to higher head rice yield and lower chalkiness. Meanwhile, Wada et al. (2011) showed at day/night temperature of 34/26 °C, the dry wind caused water deficiency in panicles and starch accumulation restricted, which can lead to rice quality decline. In this study, decreased humidity coupled with temperature treatments caused grain weight reduction and quality loss, being the most pronounced at 35°C, and to a lesser extent at 33°C, but insignificant at 37°C. This indicates that transpiration cooling may not be enough to compensate for the negative effects of water deficit when rice is exposed to high-high-temperature stress (35°C and 33 °C).

#### Physiological mechanism of temperature and RH interaction

As water fluxes for growth and transpiration are linearly superimposed (Nonami & Hossain 2010), the impaired stomatal regulation ability of rice spikelets makes a greater rate of evaporative demand under high temperature (Garrity et al. 1986), thus leaving the spikelets and grains at risk of water deficit (Tanaka & Matsushima 1971). Water deficiency in the panicle caused by the dry, hot wind was detrimental to rice grain weight and quality formation (Hiroshi et al. 2012; Kang et al. 2003; Wada et al. 2011). Low relative humidity may reduce leaf photosynthesis assimilation capacity (Tanaka & Matsushima 1971), and the ordinary maturing process of grains is dependent on sufficient water supply (Cochrane et al. 2000; Ferrise et al. 2015). Thus, high temperatures with low humidity may induce high temperature-forced maturity.

Under dry, hot wind stress, the panicle water potential is temporarily reduced (Wada et al. 2011). Osmotic adjustment of endosperm cells with increased transport of assimilating was activated to maintain kernel growth, but starch biosynthesis was slowed down (Wada et al. 2014). The vacuolar structures in the cytosol, because of osmotic adjustment, were preserved during maturity, resulting in ring-shaped chalkiness (Hatakeyama et al. 2018). Under high-high-temperature stress, substantial solutes were accumulated in endosperm cells for osmotic adjustment, accompanied by

partial inhibition of amyloplast development and formation of protein bodies, caused air spaces remained in endosperm cells during grain dehydration, leading to a chalky appearance (Wada et al. 2019). Overall, similar mechanisms of caused starch synthesis restriction were also found under both high temperature and dry wind conditions, meaning that decreased RH may aggravate the high-high-temperature effect on grain weight and quality loss.

#### **Varietal differences under RH in combination with temperature treatments**

Daily mean air temperature over 25°C during the grain filling stage can cause rice grain quality loss (Morita et al. 2016; Wu et al. 2016). However, the magnitude of heat stress-induced damage varied with genotypes. Cooper et al. (2008) reported grain quality in susceptible cultivars significantly reduced at the nighttime temperature of 20°C, but such a phenomenon did not show in heat-tolerant cultivars at the nighttime temperature of 30°C. In this study, grain weight and quality traits did not change linearly with increasing temperatures. Instead, it only changed dramatically at a certain temperature, being different remarkably among cultivars (Fig. 2, 4 and 5). This was similar to the response of spikelet fertility to high temperatures during flowering (Jagadish et al. 2007).

The identification of heat-tolerant rice germplasm resources (e.g., N22) provided opportunities to breed heat tolerance rice at the flowering stage (González-Schain et al. 2015; Tetsuo & Shouichi 1978). Compared with the mechanism of heat-induced yield loss, the mechanism leading to grain quality losses is more complex (Jagadish et al. 2015), which brings more challenges for tolerant variety screening and identification. Most previous studies paid closer attention to temperature settings under controlled environment when compared to rice heat tolerance at the grain filling stage (Chen et al. 2017; Shiraya et al. 2015; Tanamachi et al. 2016). This study showed changes in grain weight and quality are affected by the interactions between temperature and humidity, which means that the setting of humidity also bears importance in evaluating varietal heat tolerance. We ~~therefore, therefore,~~ suggest that the effect of humidity should be considered in multi-variety tolerance screening and identification. Among those factors, the optimum combinative 35°C by RH75% by 15 days could be recommended to screen for heat tolerance of rice. Our results would attract broad attention to the detrimental interactive effects of high temperature and humidity on rice yield and quality and would

360 also be interesting to both breeders and agronomists for the adjustment of breeding  
361 targets. R168, the most heat-tolerant cultivar used in this study, showed smaller  
362 differences in grain weight and quality between two RH regimes. The variety like this  
363 could be selected as a heat-tolerance variety that can improve rice yield and quality  
364 under climate change.  
365

## **Conclusion**

We found a decreased RH aggravated the detrimental effects of high temperature on grain weight and quality, and such effects were the most pronounced at 35°C, and to a lesser extent at 33°C, but were not significant at 37°C. Heat tolerant cultivars were identified and verified to be less affected by the treatments.

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**Figures**

**Figure 1: Diagram of all treatments (A), temperature and RH diurnal variation, maximum temperature, average temperature, minimum temperature and average RH in each treatment (B-G).** The measured value was the average of 15 d, and the shading around the measured value indicates the standard error of the measured values of 15 d.

**Figure 2: Grain weight responses to different treatments.** (A) LY27, (B) LY6, (C) ZLY47, (D) R168, (E) IR64 and (F) 16343. Vertical bars denote standard deviations ( $n = 3$ ), a different letter in the lowercase indicates significant differences among means of different treatments as determined by the LSD test ( $p < 0.05$ ).

**Figure 3: Boxplot shows the difference between relative humidity treatments (RH75% vs. RH85%) in different temperature treatments and durations.** (A) grain weight, (B) head rice rate and (C) Chalkiness. Data for each boxplot are the value at RH75% minus the value at RH85% under each temperature  $\times$  duration treatment of 6 cultivars.

**Figure 4: Head rice rate responses to different treatments.** (A) LY27, (B) LY6, (C) ZLY47, (D) R168, (E) IR64 and (F) 16343. Vertical bars denote standard deviations ( $n = 3$ ), a different letter in the lowercase indicates significant differences among means of different treatments as determined by the LSD test ( $p < 0.05$ ).

**Figure 5: Chalkiness responses to different treatments.** (A) LY27, (B) LY6, (C) ZLY47, (D) R168, (E) IR64 and (F) 16343. Vertical bars denote standard deviations ( $n = 3$ ), a different letter in the lowercase indicates significant differences among means of different treatments as determined by the LSD test ( $p < 0.05$ ).

605 **Tables**

606

607 **Table 1:**

608 **Summary of ANOVA for grain weight, head rice rate and chalkiness in each cultivar.** Factors:  
609 Temperature (T), Relative humidity (RH), Duration days (D). Data are F-values with indication  
610 of significance levels (\* P < 0.05; \*\* P < 0.01; \*\*\* P < 0.001).

611

612 **Table 2:**

613 **Parameter estimation, r square and F-value of multiple regression.** In formula  $Y = a \cdot X_1 + b \cdot X_2 + c \cdot X_3 + d \cdot X_4 + e$ , where Y representing grain weight, head rice rate and chalkiness; X1  
614 representing daily maximum temperature in different treatment, X2 representing relative humidity  
615 in different treatment, X3 representing treatment days in different treatment, X4 representing the  
616 grain weight, head rice rate and chalkiness of each cultivar under controlled condition; \*, \*\* and  
617 \*\*\* indicate significance levels at P < 0.05; P < 0.01 and \*\*\* P < 0.001.

619

620 **Table 3:**

621 **Membership function values (MFVs) of grain weight, head rice rate and chalkiness for each**  
622 **cultivar in each treatment.**

623

624

625 **Supplementary Tables**

626

627 **Table S1 Heading date, daily mean temperature during 15 days after heading (T15), daily**

628 **mean relative humidity during 15 days after heading (RH15), grain weight, head rice rate**

629 **and chalkiness of each cultivar in field trial in 2016.**

630

631 **Table S2 Heading date, daily mean temperature during 15 days after heading (T15) and daily**

632 **mean relative humidity during 15 days after heading (RH15) of each cultivar.**