



Responses of Growth and Grain Yield of IR50404 Rice to Temperature Stress

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A B S T R A C T

Climate changes, rising warmth, drought, and CO₂, are now seriously influencing agriculture. In this study, four separate greenhouses (labeled GH1, GH2, GH3, and GH4) were built with plastic roofs and walls, except GH1, which had three walls with mesh to evaluate the impact of temperature stress on growth, biomass, and yield of rice variety IR50404 under different temperature regimes. The control treatment group was grown ambient, next to these greenhouses. GH1, GH2, GH3, and GH4's temperatures were from 0.9 °C to 3.1 °C higher than the ambient (as control). Carbon dioxide concentrations in GH2, GH3, and GH4 were recorded higher than the ambient, from 34.1 ppm to 48.2 ppm. Total vegetative dry matter was reduced from 15.9% to 20.5%, while grain yield declined from 20.8% to 24.6% when the mean temperature increased from 2.9 °C to 3.1 °C. High temperature or a combination of high-temperature stress with elevated CO₂ concentration reduced the grain yield and total vegetative dry matter.

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INTRODUCTION

In terms of the global climate change scenario, Vietnam is one of the countries most affected by climate change (Vien, 2011). At recent decades, the temperatures have increased in most meteorological sites in Vietnam, with rising temperatures by 0.62 °C on average across the country from 1958 to 2014 (Khoi & Phi, 2018). For example, it rose by 0.42 degrees Celsius between 1985 and 2014 (Hiraishi et al., 2014). Vietnam's highest and lowest temperatures increased significantly between 1961 and 2014 (Ngo-Duc, Kieu, Thatcher, Nguyen-Le, & Phan-Van, 2014). The number of hot days with maximum temperatures above 35 °C has grown practically everywhere in Vietnam (Thuc et al., 2016).

Rice is the most significant food crop in Vietnam and many other countries. The International Rice Research Institute (IRRI) developed the IR50404 rice variety, which was introduced in Vietnam in the early 1990s. The variety has a growth period of 85-90 days, a high yield of 6-8 tons/ha, is easy to grow, is mildly infected with rice blast and brown planthopper, and has a reasonable price, so many farmers trust it.

The question is how the increase in air temperature would affect the growth, biomass, and yield of rice variety IR50404. Many studies around the world have shown that when the temperature increases, the yield of rice grain decreases (Arshad et al., 2017; Chaturvedi, Bahuguna, Shah, Pal, & Jagadish, 2017; Fahad et al., 2019; Morita, Wada, & Matsue, 2016; Usui et al., 2016; Xiong, Ling, Huang, & Peng, 2017). Increased CO₂ concentration

increases biomass and rice yield (Cheng, Sakai, Yagi, & Hasegawa, 2009; Hasegawa et al., 2013; Usui et al., 2016). The research question is that in farming, both temperature and CO₂ concentrations increase in growth; how will biomass and rice yield happen? Many studies in Vietnam have focused on farming techniques to set high yields, improve rice breeding, and on chemical pest management or integrated methods. However, research on the effects of climate change (in particular, temperature stress and elevated CO₂ concentrations) in Vietnam has been very limited. A study was conducted to evaluate the impact of temperature stress on growth, biomass, yield components, and yield of the IR50404, which has been widely grown in An Giang province, Vietnam.

METHOD

Experiment location and crop season: The experiment was conducted in greenhouses on the An Giang university campus, Viet Nam from June to September 2021 (Summer-Autumn). Ambient as a control was located adjacent to GH1, as shown in Figure 1.

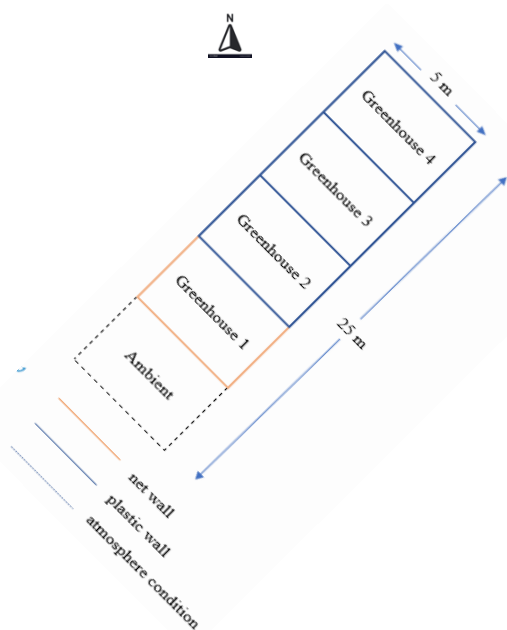


Figure 1. Diagram of the location of 4 greenhouses on the An Giang university campus (Vietnam)

Experiment design

The size of each greenhouse was 5m in length x 5m in width x 4m in height and made of a steel frame. The roofs and walls of all 4 GHs were plastic except for GH1, having three walls made of the net (18 mesh cell density) (Figure 1). The purpose of netting at GH1 was to lower the air temperature. All four GH1, GH2, GH3, and GH4 were ventilated with triangular air holes near the roofs. The size of this isosceles triangle air vent was 4.5m x 4.5m x 5m. A door was arranged at a corner of GH1. Each GH and the

ambient have three sensors to record hourly temperature, CO₂ concentration, and light intensity connected to a self-recording system.

Every location planted 42 pots of rice (34x28x28 cm), filled with loamy sand soil. These pots were arranged into three similar groups. We used two pots of each group to measure the parameter values in every observation. Planting density was three hills/pot with a fertilizer formula of 120-45-40 kg/ha NPK.

Parameters

Temperature (°C), CO₂ concentration (ppm) inside and outside the GHs, and light intensity (lux) were automatically recorded every 60 minutes.

Rice growth parameters (IRRI, 2002) were collected at 20, 40, and 60 days after planting (DAP) and harvest time: plant height (cm), number of tillers/pot, total vegetative biomass (TVB, above the ground, g/pot), total vegetative dry matter (TVDM, g/pot), grain yield (g/pot), and yield component (panicles/pot, spikelet/panicle, filled grain percentage, 1000-grain weight, and grain yield).

Statistical analysis

Minitab software was used to analyze variance (ANOVA) and Tukey test at a 5% level for the significant difference between mean values to test the impact of different temperature regimes and CO₂ concentrations on the parameters. The relationship between daily mean temperature, CO₂ concentration, grain yield, and total vegetative dry matter was determined using an exponential correlation of the power curve.

RESULTS AND DISCUSSION

Temperature in greenhouses

The experiment was conducted in the rainy season, changing high and low temperatures following the cloud covering or rain, with extraordinarily high daily temperature fluctuations in the four GHs. The temperature range wildly fluctuated from 16.6 °C to 21.1 °C inside GHs and outside, showing a lower one (6.7 °C). So, the differences compared to ambient were 0.9 °C, 3.1 °C, 3.1 °C, and 2.9 °C, respectively (Table 1 and Figure 1). Under the same conditions as the nylon roof, the mesh wall in GH 1 had a mean temperature lower than that of the GHs with plastic walls from 1.3 °C, 2.1 °C, 2.9 °C (Table 1).

The minimum temperatures did not significantly differ, but the maximum temperatures were very distinct, oscillating from 9.4 °C to 14.4 °C compared to ambient. In GH, almost all days in the growing period had $T_{max} \geq 34$ °C. At the high-stress temperature (≥ 45 °C), GH2, GH3, and GH4

had many days to reach and exceed the threshold temperature from 62 to 72 days (Table 1). It shows that under this condition, plants are frequently affected by temperature stress, including vegetative, reproduction, and grain-filling stages (Krishnan, Ramakrishnan, Reddy, &

Reddy, 2011). With the influence of temperature above 50 °C, the processes of metabolism and growth have been stopped, causing heat stress. At the grain-filling stage, rice grain may be flattened (Krishnan et al., 2011).

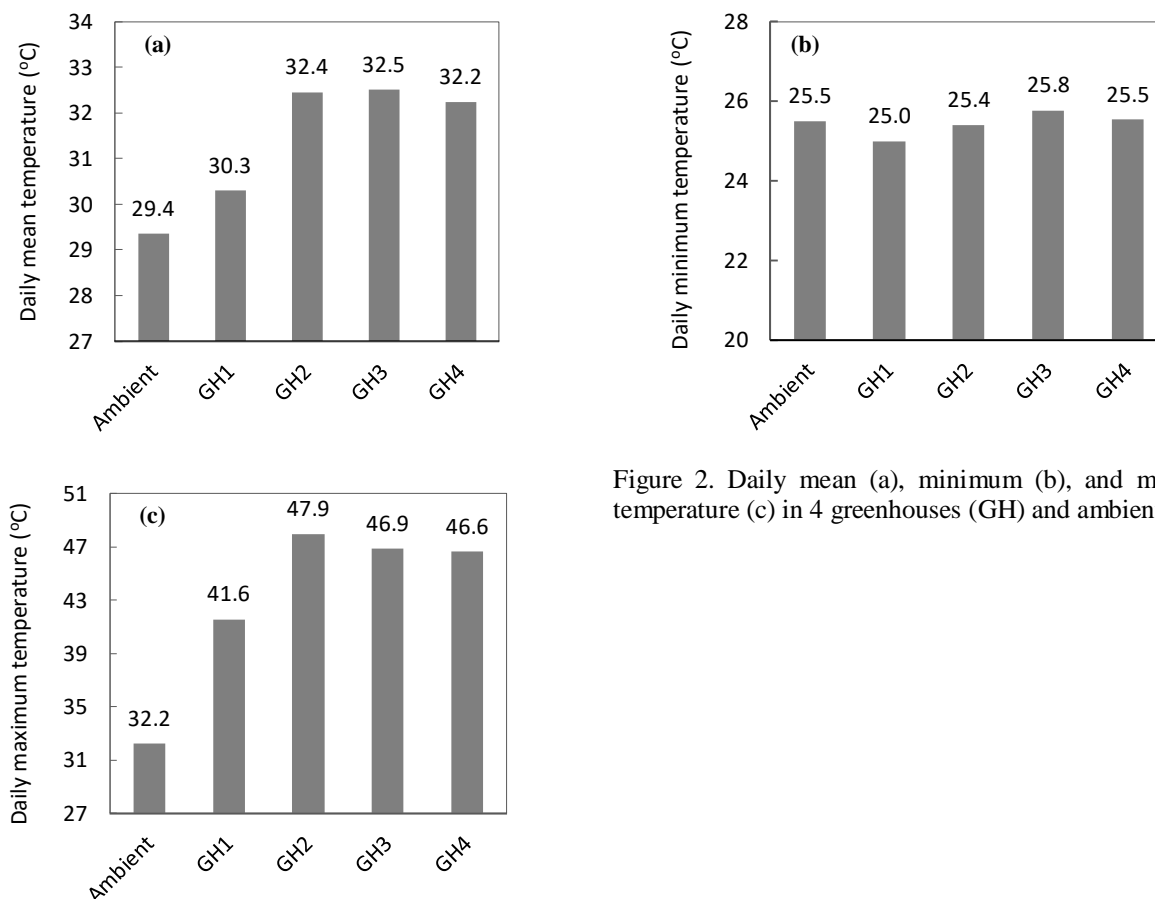


Figure 2. Daily mean (a), minimum (b), and maximum temperature (c) in 4 greenhouses (GH) and ambient

Table 1. Characteristic of temperature (°C) in 5 locations

Location	Range (min-max)	Mean Temperature ¹	Number of days ($T_{max} \geq 34$ °C)	Number of days ($T_{max} \geq 45$ °C)
Ambient	6.7	-	11	0
GH1	16.6	0.9	89	21
GH2	22.5	3.0	87	62
GH3	21.1	3.1	88	68
GH4	21.1	2.8	88	72

¹Increasing from the ambient

When raining, the ambient was cloudy, the temperature decreased very quickly, and the temperature amplitude was 6.7 °C. It was quite “stable” compared to the inside GHs. Nylon retained heat longer and lasted longer due to the greenhouse effect, so heat stress manifested itself more strongly (Khoshnevisan, Shariati, Rafiee, & Mousazadeh, 2014). Outside, there are also unfavorable conditions when having heavy rain (powerful winds on stormy days or tropical depressions) coinciding with pollination and seed

formation, which causes a decrease in rice growth and yield (C. Cai et al., 2016; JT & LH Jr, 1993; Krishnan et al., 2011; Saseendran, Singh, Rathore, Singh, & Sinha, 2000; Singh, Prasad, & Reddy, 2013). However, for rice grown in An Giang province, the yield loss was not due to an effect on pollination but mainly because of the rainstorm, reducing the quality of the milling.

Fig. 2 shows a much more variable temperature amplitude at GH2, GH3, and GH4 than ambient. The red dashed line

in each line graph indicates the average optimum temperature range for rice growth (30 °C) (Chen, Tang, Xu, Lan, & Cao, 2019). This temperature was lower than the

daily average in GH2, 3, and 4 but slightly higher in GH1. The maximum temperature fluctuates wildly on hot sunny days interspersed with rain or heavy cloud days.



Figure 3. Temperature variations of daily mean, minimum, and maximum temperature (°C). The red horizontal dashed line at 30 °C of every line chart shows the optimum temperature according to the literature (Chen et al., 2019).

IR50404 variety ending its growth period in 4 GHs and ambient were 89, 87, 88, 88, and 91 days, respectively. It is clear that with the higher temperature in this experiment, the rice growing period shortened from 1-3 days when cultivated in hotter conditions. It is consistent with the studies of Moldenhauer and Slaton (2001) and Oh-e, Saitoh, and Kuroda (2007).

Carbon dioxide concentration ([CO₂])

The average CO₂ concentration (ppm) in the four GHs was slightly higher than ambient, from 18.2 to 48.2 ppm (Table 2), corresponding to an increase of 4.7%, 8.4%, 9.8%, and 11.3%. Increasing minimum [CO₂] from the ambient was not considerable; however, increasing maximum [CO₂] from the ambient resulted in 325 to 459 ppm, the gradual increase of GH1, 2, 3, 4 compared with ambient (Table 2).

Table 2. Characteristics of carbon dioxide concentration (ppm) in 5 locations

Location	Average [CO ₂]	Min [CO ₂]	Max [CO ₂]	¹ Increasing average [CO ₂]	¹ Increase min [CO ₂]	¹ Increase max [CO ₂]	Light intensity (Lux)
Ambient	389.3 ± 7	385	445	-	-	-	48.890 ± 28.890
GH1	407.5 ± 44	386	770	18.2	1	325	43.310 ± 27.450
GH2	423.4 ± 57	385	793	34.1	0	348	43.370 ± 26.630
GH3	430.8 ± 71	386	868	41.5	1	423	43.970 ± 25.920
GH4	437.5 ± 89	370	904	48.2	5	459	44.010 ± 26.170

¹from ambient

This experiment assessed rice plants' biomass, growth, and yield were the combined effects of both temperature variables and different [CO₂] by day. However, the contribution of these two factors is different and depends on the threshold (IRRI, 2002). Grain yields are insensitive to [CO₂] at higher growth temperatures (Morita et al., 2016). Elevated [CO₂] and average temperature both affected grain yield and quality (e.g., protein content) (Jing et al., 2016). The confluence of [CO₂] and warmth impacts implies that both quantitative and qualitative changes in rice supply are feasible in warmer places (i.e., >34 °C) where rice was produced (Jing et al., 2016; Pal Madan et al., 2012).

Plant height and tiller number

The rice plant height observed at 20, 40, and 60 days after planting (DAP) for IR50404 rice variety in Table 3 showed that the plant height was significantly higher in GH1 and ambient compared with three other GHs. The rice plants rapidly grew to 40 DAP and slightly increased to 60 DAP (Table 3). The height of rice plants in 3 houses, GH2, GH3, and GH4, with higher temperatures, had a statistically significant difference compared to GH1 and ambient (Table 3).

Table 3. Plant height (cm) and the number of tillers under different temperature regimes

Location	Plant height			Tiller number/plant		
	20 DAP	40 DAP	60 DAP	20 DAP	40 DAP	60 DAP
Ambient	52.3 a	90.3 a	95.2 a	37.2 a	66.5 ab	60.7 c
GH1	53.0 a	89.7 a	94.2 a	35.8 ab	55.8 c	60.8 c
GH2	46.5 b	83.7 ab	89.0 b	32.7 b	58.0 bc	70.8 a
GH3	48.4 b	81.5 b	84.0 b	37.0 a	70.8 a	71.8 a
GH4	40.9 c	84.3 ab	88.4 b	29.0 b	65.5 abc	77.8 a
F	*	*	*	*	*	*

DAP: days after planting. Numbers followed by the same letter in a column are not significantly different at the 5% level by Tukey's test.

Up to 40 DAP, plants increased tillers quickly, reaching 66.5 tillers/pot in ambient and 55.8 tillers/pot in GC1. It is worth mentioning that the number of tillers increased quickly in GH2, 3, and 4 were hotter than ambient. At 60 DAP, the number of tillers in the GHs with the higher temperature still maintained a significantly higher one ($P < 0.05$). The number of tillers increased with increasing temperature. Some researchers have proved that heat increased the number of tillers, but those gave lower spikelets/panicles (Krishnan et al., 2011; Xu, Henry, & Sreenivasulu, 2020). Moreover, under severe stress, the spikelet is flattened at the filling grain stage, increasing the number of tillers and panicles per unit area but inefficient in grain yield (Krishnan et al., 2011; Xu et al., 2020).

Total vegetative biomass (above the ground)

Total vegetative biomass (TVB) above the ground (g/pot) significantly differed between 5 locations at a 5% level. At

the tillering stage, at about 20 DAP, TVB did not differ significantly between inside and outside the greenhouses. From 40-60 DAP in the stem elongation to panicle formation stage, TVB increased rapidly, especially GH1 had the highest TVB at 40 DAP and higher than GH3 at 60 DAP ($P < 0.05$). At harvest time, TVB was differentially high in GH1, GH4, and ambient, while GH2 and GH3 had statistically significantly lower TVB at a 5% level (from 274.2 g/pot to 304.7 g/pot). Therefore, the percentage of TVB decreased from ambient at harvest of GH2 and GH3 was the highest, dropping by -28.4% and -20.5%, respectively (Table 4).

On the other hand, total vegetative dry matter total (TVDM) in GH2 and GH3 at harvest was lower than GH1 and ambient, although periods from 20 DAP to 40 DAP were not significantly different. The percentage of TVDM decreased from ambient at harvest at GH2 and GH3 were -15.9 and -20.5%, respectively. Greenhouse 4, with an even

higher TVDM was 0.7% grown outside (Table 5). GH4 still had high temperatures but lower than GH2 and GH3, showing the highest CO₂ (437.5 ± 89 ppm compared to the ambient of 389.3 ± 7 ppm).

This demonstrated that when increasing temperatures and [CO₂] concentration, rice plants show a drop in TVB and an increase in tillers but a loss in grain production (Table 3, 6).

Table 4. Total vegetative biomass (TVB, g/pot)

Location	20 DAP	40 DAP	60 DAP	Harvest	% TVB decreased from ambient at harvest
Ambient	33.8 ab	244.0 b	318.8 ab	383.2 ab	-
GH1	31.2 ab	314.8 a	345.2 a	405.3 a	5.8
GH2	39.5 a	285.7 b	320.2 ab	274.2 b	-28.4
GH3	30.0 b	199.2 c	297.5 b	304.7 b	-20.5
GH4	32.0 ab	260.0 b	345.8 a	372.8 ab	-2.7
F	*	*	*	*	

Tukey's test finds that numbers in a column followed by the same letter are not significantly different at the 5% level.

Table 5. Total vegetative dry matter (TVDM, g/pot)

Location	20 DAP	40 DAP	60 DAP	Harvest	% TVDM decreased from ambient at harvest
Ambient	5.1 a	61,2 a	88,6 b	149.3 a	-
GH1	4.7 a	69,3 a	108,6 a	148.9 a	- 0,3
GH2	6.3 a	59,2 a	105,2 a	125.6 b	- 15,9
GH3	5.2 a	60,8 a	87,6 b	118.6 b	- 20,5
GH4	3.6 b	38,5 b	77,6 b	140.3 ab	0,7
F	*	*	*	*	*

Tukey's test finds that numbers in a column followed by the same letter are not significantly different at the 5% level.

The field experiment result of Chaturvedi, Bahuguna, Pal, et al. (2017) using open-top field chambers proved that “increasing CO₂ concentration alone (+ 200 µL) boosted IR72 rice yield by 15% and total vegetative biomass by 31%” and “simultaneous increases in CO₂ and air temperature did not alter the biomass at maturity (relative to elevated CO₂ alone)”. On the other hand, grain yield remained [CO₂] insensitive at greater growth temperatures (Hoffman, Kemanian, & Forest, 2018). In the growing season, plants in 34/27/31°C temperature treatments gained more biomass and leaf area than plants in the 28/21/25°C temperature treatments (Hoque et al., 2020). Tillering increased with increasing temperature (Moldenhauer & Slaton, 2001).

Carbon dioxide increase may boost rice total biomass (Cheng et al., 2009; Hasegawa et al., 2013; H. Y. Kim, Lieffering, Kobayashi, Okada, & Miura, 2003; Kimball, 2016; Krishnan, Swain, Bhaskar, Nayak, & Dash, 2007; P. Madan et al., 2012). However, when the temperature is too high (exceeding 45 °C), it inhibits growth and reduces biomass and absorption of water (Mahmood et al., 2021; Xiong et al., 2017). Temperature is the critical limiting factor to the growth and yield of C3 plants (Salvucci & Crafts-Brandner, 2004). An elevation in [CO₂] was inadequate to account for the unfavorable impact of rising temperatures on wheat and rice biomass and yield (C. Cai et al., 2016; Chaturvedi, Bahuguna, Pal, et al., 2017). Wheat and rice yields were reduced by 10-12 percent and

17-35 percent, respectively, when high CO₂ and warmth were combined (C. Cai et al., 2016).

Yield component and grain yield

In lower temperatures and [CO₂] than GH2, GH3, and GH4, the ambient and GH1 got the number of panicle/pot, spikelet/panicle, and percentage of filled grain higher (Table 6, 2 and Figure 1). Filled grain (%) in GH3 and GH4 was only 59.3 and 68.0%, respectively, compared to ambient 13.5% and 24.6% in reduction. High temperature during the ripening stage (32-40 °C) increased unfilled grain and stress temperature beginning on the fourth day after heading, resulting in a drop in panicle weight in all cultivars (Zakaria, Matsuda, Tajima, & Nitta, 2002). Heat stress causes another dehiscence to be disrupted, resulting in poor pollen distribution and less pollen on the stigma. Furthermore, heat stress inhibited starch production in emerging grain, resulting in less starch buildup (Arshad et al., 2017).

Similarly, spikelet/panicle had less decline, only 8.1% and 10%, respectively, at GH3 and GH4 compared to ambient (calculated from Table 6). The final grain yield obtained at GH2, GH3, and GH4 was lower than at GH1 and ambient. The percentage yield loss was generally compared to the ambient of 0.9%, 20.8%, 24.6%, and 21.2%, respectively (Table 6). Consequently, the high-stress temperature factor was unambiguously the most important. It explicitly reduced spikelet/panicle, percentage of filled grain, and

final grain yield of rice variety IR50404 in this experiment.

Table 6. Yield component and grain yield of IR50404

Location	Panicle/pot	Spikelet/panicle	Filled grain (%)	1000-grain weight (g)	Grain yield / (g/pot)	% decreased from an ambient grain yield
Ambient	53.1 a	126.2 a	78.6 ab	22.2	92.4 a	-
GH1	58,5 a	132.2 a	81.3 a	22.6	91.3 a	-0.9
GH2	55.0 a	114.8 b	86.9 a	21.6	73.0 b	-20.8
GH3	47.0 b	113.5 b	59.3 c	21.0	69.5 b	-24.6
GH4	57,5 a	116.0 b	68.0 b	21.4	72.7 b	-21.2
F	*	*	*	ns	*	

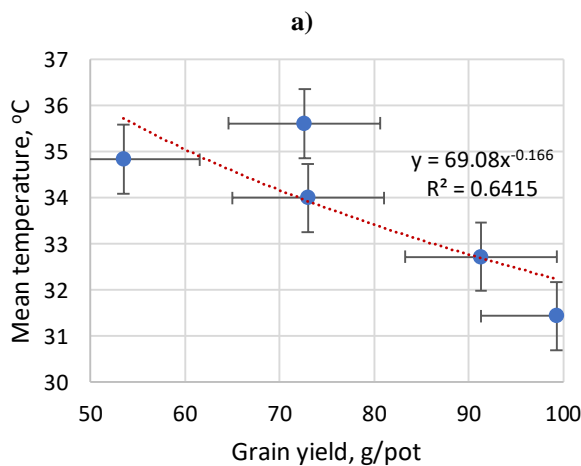
Tukey's test finds that numbers in a column followed by the same letter are not significantly different at the 5% level.

Findings in our experiment showed that increasing temperature from 0.9 °C to 3.1 °C reduced IR50504 grain yield from 0.9% (GH1) to 24.6% (GH3), although there was an increase of 4.7% – 11.2% [CO₂]. In addition, TVB and TVDM still decreased with increasing these two factors. The reasons that might impede the plant's development include sympathies, respiration, and metabolism.

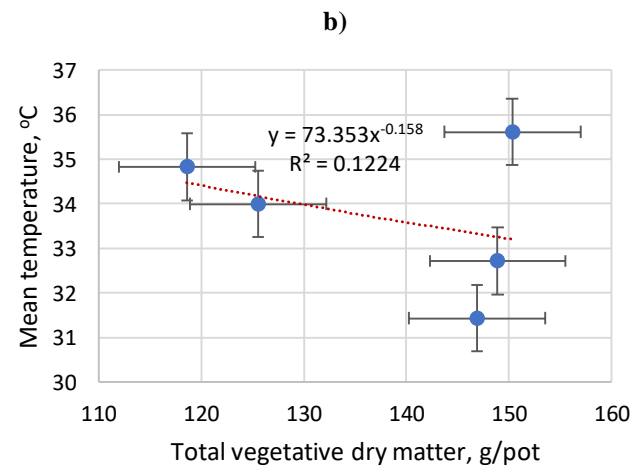
Correlation between temperature, [CO₂] with biomass, and yield

In many studies, it has been found that an increase in [CO₂] would improve grain yield (Chaturvedi, Bahuguna, Pal, et

al., 2017; Cheng et al., 2009; Jing et al., 2016; Kadam et al., 2014; H. Kim & You, 2010). In this experiment, the yield was significantly reduced under high-stress temperatures, especially in pre-healing, pollination, and seed production. In particular, the correlation graph in Figure 3 shows that increasing temperature in this experiment reduced grain yield for rice and TVDM, although there was an addition of 18 – 48 ppm [CO₂]. The yield reduction due to the temperature increase was more substantial than the decrease in accumulated biomass due to the increase [CO₂]. This is the combined effect.



(c)



(d)

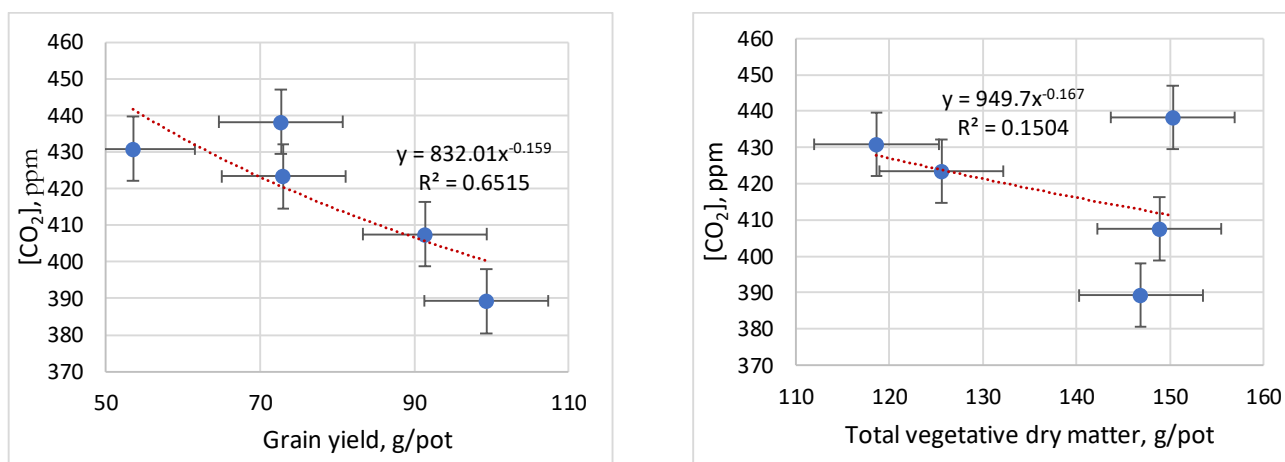


Figure 4. (a and b): the regression of impact of elevated temperature on grain yield and total vegetative dry matter
(c and d): the regression of impact of elevated [CO₂] concentration on grain yield and total vegetative dry matter

Atmospheric CO₂ enriched from 330 to 660 ppm enhanced grain production by raising the number of panicles/hill. Still, increasing temperature treatment above 28/21/25 °C lowered grain output by decreasing the number of filled grains/panicles (Chuang Cai et al., 2016). With rising [CO₂] treatment, evapotranspiration reduced, and water-use efficiency rose, but the opposite trends were seen with increasing temperature treatment. These findings suggest that future [CO₂] increases would enhance rice production by improving photosynthesis, growth, and grain output while decreasing water needs. Future rises in air temperature may result in yield declines and increasing water requirements in warmer places (Wang, Cai, Lam, Liu, & Zhu, 2018).

The number of filled grains per unit area for wheat and rice was the most relevant yield component after accounting for the impacts of high [CO₂] and temperature (C. Cai et al., 2016). The complicated treatment impacts the interaction of pre-heading length, nitrogen absorption, tillering, leaf area index, and radiation-use efficiency, and hence on yield and yield components (C. Cai et al., 2016).

More specifically, an increase in CO₂ resulted in better grain output, but an increase in temperature severely impacted rice plant grain yield. The temperature had a far more significant effect on grain yields than [CO₂] treatment (Chuang Cai et al., 2016). Grain yields decreased by an average of 7-8% for each 1°C increase from 28/21/25 to 34/27/31°C. Reduced grain yields with rising temperature treatment point to possible negative consequences on rice production in some places if air temperatures rise, particularly in areas with limited sun irradiation (Zhao et al., 2017).

CONCLUSIONS

Both biomass and grain yield of rice cultivar IR50404 decreased with increasing temperature. When raising the average temperature to about 3 °C, TVB decreased from 20.5% to 28.4%, and grain yield decreased from 20.8% to 24.6% even though small-scale rises in CO₂ concentrations from 34.1 ppm to 48.2 ppm.

In addition, elevated temperatures shortened the growth period by three days, mainly in the vegetative stage. The percentage of filled grain decreased proportionally with very high-stress temperatures. Filled grain decreased from 13.5% and 24.6% compared to the control when it increased to about 3 °C.

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