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Yield and Seed Chemical Composition under Elevated CO_2 and Temperature Based on the RCP 4.5 Scenario of Important Thai Rice

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Abstract This research aimed to examine the effects of combining increased temperature and carbon dioxide (CO_2) under RCP 4.5 scenarios on yield and seed chemical composition of Thai rice cultivars; Chinat1, Phathumthani1, and Phitsanulok2. The field warming experiment was conducted at Phitsanulok from 2018 to 2020. Sixteen field-open top chambers (OTCs) were applied to simulate the expected future climate situation under RCP 4.5 (the elevated temperature at 2°C and CO_2 up to 800 ppm, respectively). The results revealed that the single factor of elevated temperature at 2°C led to statistically significant grain size reductions of 2 rice cultivars; Chinat1 and Phitsanulok2. We also found that the combination of elevated temperature and CO_2 induced a significant grain yield loss (ton.ha^{-1}) by 68% in Phitsanulok2 but an increase of 38% appeared in Chainat1. Considering results in seed chemicals, the statistically significant decrease trends in Amylose and Brix values were shown in 3 cultivars. These results suggest that elevated temperature and CO_2 caused the imbalance in seed nutrient contents and an increase in yield loss in some Thai rice cultivars.

Keywords Scenario, Rice, Yield, Amylose, Brix Value

1. Introduction

Since the pre-industrial period, anthropogenic emissions of carbon dioxide and other greenhouse gases have risen in the atmosphere. Their impacts have been seen throughout the climate system, and they are very likely to be the main factor in the warming that has been recorded since the middle of the 20th century [1]. Numerous data points also show that the earth as a whole has already warmed by several degrees centigrade over the previous few decades. The rising trend is anticipated to continue for the rest of this century, according to studies on the earth's climate using computer models, including new Representative Concentration Pathways (RCPs) predictions [2]. The Intergovernmental Panel on Climate Change (IPCC) said in its fifth assessment report that, with RCP4.5, the predicted increase in global surface temperature at the end of the 21st century [1,3]. Additionally, the Representative Concentration Pathway 4.5 predicts that carbon dioxide (CO_2) levels will reach 538 ppm (not to exceed 1,000 ppm) (RCP 4.5) [1,4]. Fairly conclusive evidence also indicates that the tropics will encounter more frequent high temperature extremes or unpredictable weather as the global mean temperature rises [5].

Thailand is situated in the tropical region between latitudes 5° 37° and 20° 27° N and longitudes 97° 22° and 105° 37° E. A total of 200,000 square miles or 518,000 square kilometers make up the area [6]. One of the tropical nations that is predicted to be most impacted by upcoming climate change is Thailand. The average yearly temperature in Thailand has dramatically increased by roughly 0.95°C between 1955 and 2009, according to the Climatological Center of Thailand. This is far higher than the global average temperature increase of 0.6°C. Over the previous 55 years, there has also been an increase in the annual peak, average, and lowest temperatures of roughly 0.86°C, 0.95°C, and 1.45°C, respectively [7]. These temperature trends are rising as a result of the nation's emissions of greenhouse gases. One of Southeast Asia's economies with the quickest rate of energy-intensive growth is Thailand. The burning of fossil fuels, particularly CO_2, was the main source of GHG emissions in the energy sector. In 2017, total CO_2 emission was 235.8 Mt-CO_2eq. In 2017, the majority of the 96.8 Mt-CO_2eq (41.1%) came from the generation of heat and electricity. Transportation and manufacturing industries contributed 78.4 Mt-CO_2eq (33.3%) and 45.2 Mt-CO_2eq (19.2%) of total CO_2emission, respectively [8].

Changes in the agriculture sector are among the most alarming effects of climate change. The most significant element affecting agricultural productivity is the climate. As a result, throughout the past several decades, significant changes in research have been driven by the negative consequences of climate change on agriculture. Many studies focus on the potential physical impacts of climate change (temperature and CO_2 effects) on agriculture, including changes in yield, certain physiological changes, and animal yields as well as the economic ramifications of these prospective yield changes [9]. Climate change is the main threat for the agriculture industry globally, including Thailand, according to the majority of published studies on the impacts of CO_2 and temperature stress on diverse species of plants. Both of these factors can have significant effects on plants and vital crops [10,11]. Numerous physiological and morphological features of phenology, growth, physiological processes, nutritional value in seeds, genetics, and yield output of crops can be significantly impacted by climate change and fluctuation [5]. It is projected that climate change will have a detrimental effect on food security, food quality, and production. Most of the world's crops have experienced heat stress at some point in their life cycles as a result of the high temperatures brought on by climate change [12].

Despite the issues brought on by the changing climate, an increase in global food production of around 70% on the basis of cereals (wheat, rice, or coarse grains) by 2050 is required to fulfill the projected demand. Climate change has caused enormous upheavals, but there shouldn't be any major obstacles to world agriculture supplying all the food required for the population of the future [13].

One of the world's starch-grain crops is rice (*Oryza sativa L.*). More people are directly nourished by it than any other crop, making it the most significant crop for human consumption. Additionally, rice is a staple diet in Asia, home to almost half of the world's poorest people, and is gaining popularity in Africa and Latin America [14]. One of the most important aspects affecting rice's seasonal growth is temperature. High temperatures have an influence on rice production in terms of both yield and grain quality [15]. The length of time from emerging to flowering and from flowering to maturity has a significant impact on rice's grain filling and yield components [16].

The ideal temperature for rice grain growth is around 30 degrees, and below 8 degrees and over 41 degrees, grain growth stops [16]. The reaction of grain weight to temperature has been observed to differ significantly between wheat and rice; in spring wheat, single-grain weight began to decline below 19°C, but rice's dropped above 25°C [17]. According to several experimental and analytical researches, a rise in CO_2 under climate change from 380 mol.mol-1 to 550 mol.mol-1 predicted for the year 2050 would boost C3 photosynthesis by 38% [18]. Typically, enhanced photosynthesis, better water usage efficiency, and less photorespiration might all boost rice productivity. Increased tillers, panicles, spikelets per panicle, the percentage of filled spikelets, and 1,000-grain weight have all been linked to higher rice output under long-term elevated CO_2. Rice cultivars respond to elevated CO_2 by producing more biomass and seeds, but conversely, rising air temperatures had the opposite effect [19]. Higher non-structural carbohydrate (NSC) concentration in a sink (seeds) among crop species was also a consequence of increased CO_2, which also increased photosynthetic rates and improved carbohydrate metabolism enzymatic activity in the source (leaf) tissue under non-stress conditions. This included promoting sucrose-P synthase (SPS) activity and boosting the accumulation of soluble sugars and starch [20]. However, these processes will cause an imbalance of carbon to nitrogen (C/N) ratio assimilation in a plant [22].

On the other hand, heat stress on rice reveals that warmth is the primary factor causing damage under conditions of both increased temperature and elevated (CO_2+HT) interaction [23]. Despite the fact that growing season is prolonged, increasing CO_2 has some positive effects on certain growth phases. However, post-heading excessive heat may have an influence on rice yield components and quality features [24]. According to Chaturvedi1 et al. [21], increasing CO_2 considerably enhanced rice grain weight, seed set, panicle weight, and photosynthesis. However, during blooming and early grain filling, high temperatures and increasing CO_2 dramatically decreased seed set and 1,000-grain weight, respectively.

Industrialization in Thailand during the recent decades has led to an increase in greenhouse gases in the atmosphere. Additionally, regional meteorological observatories in Thailand have altered the current mean temperature during the rice growing season. Given that Thailand is an agricultural nation, it is crucial to understand how climate change is affecting several significant varieties of Thai rice.

Therefore, the objectives of this paper are to summarize some of the major research findings from the future projected climate change situation under the RCP4.5 scenario on the effects of CO_2 and temperature on yield quantity and quality of some important Thai rice (*Oryza sativa* L.) cultivars.

2. Materials and Methods

The research was conducted to determine the effect of increased temperatures and elevated CO_2 levels under the Representative Concentration Pathway 4.5 (RCP4.5) on yield components and seed chemicals of 3 cultivars of Thai rice. The RCP4.5 is the scenario that stabilizes radiative forcing at 4.5 $W.m^{-2}$ in the year 2100 [4].

The experiment was conducted in the Phitsanulok province (16°799790′, 100°225468′E), Northern region of Thailand from 2018 to 2020. The total study area covered about 300 m^2. The area has a tropical climate. The mean air temperature and mean maximum temperature in this area are approximately 27.3°C and 32.4°C in the growing season, respectively.

2.1. Temperature and CO_2-Controlled in Field Open Top Chambers

Exposure of rice plants to elevated air temperature and CO_2 was carried out in 16 field-open top chambers (OTCs). Each rice plant inside OTC is exposed to the same condition of solar radiation and soil nutrient including water quantity. Rectangle open-top chambers were built in 2018; they were used throughout this study period. Each chamber size was 3 m (width) x 4 m (length) x 30 m (height). It was constructed from transparent plastic hence the chamber did not reduce light quantity for photosynthesis inside OTCs (Figure 1).

Two climate situations of air temperature inside chambers) were set up to maintain at ambient level (30.3-40.5°C; CT and elevated air temperature (32.8-42.9°C; HT4.5 by black infrared bulbs and air conditioners that were linked to the automatic controlling system. The average daytime temperatures for 4 treatments were shown in Figure 2. The projected changes in the average temperature with 2 degrees Celsius of global warming in this research (HT4.5 and HT+$CO_2$4.5) compared to CT treatment were shown in Figure 3. The CO_2 generator tanks (5 kg) were applied to generate elevated CO_2 inside OTC Open Top Chambers. CO_2 levels in 2 treatments ($CO_2$4.5 and HT+$CO_2$4.5) were controlled by a CO_2 valve and measured by a CO_2 meter (Model testo 440-Climate Measuring Instrument) (Figure 1). Levels of CO_2 concentrations were maintained at ambient (~430 ppm; CT) and elevated levels (~800 ppm; $CO_2$4.5 and HT+$CO_2$4.5) for this field experiment based on the RCP4.5 scenario (Figure 4). The exposure period for both controlled temperature levels and CO_2 concentrations was set at 10 hrs. daily from 07.00 AM to 05.00 PM from the transplanted stage to the harvest stage.

Figure 1. Open Top Chamber for field studies of elevated air temperature and CO_2 concentration on rice

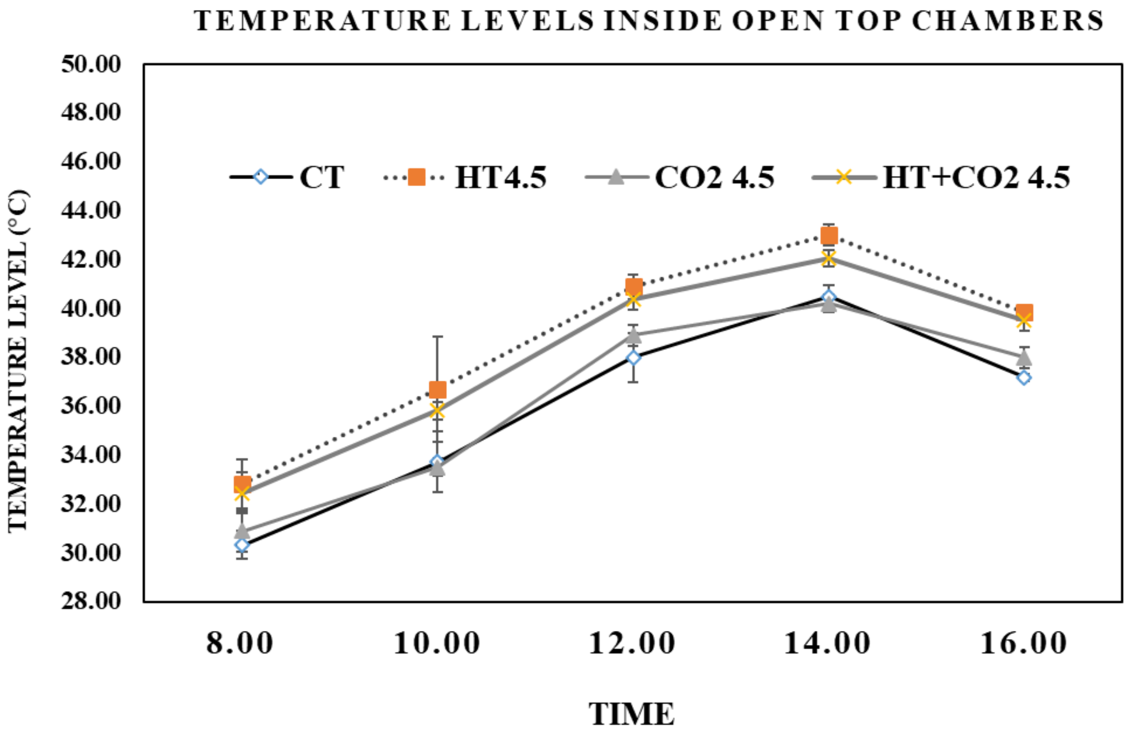


Figure 2. Average temperature levels (±SD) inside Open with 2 degrees Celsius of global warming in 4 treatments

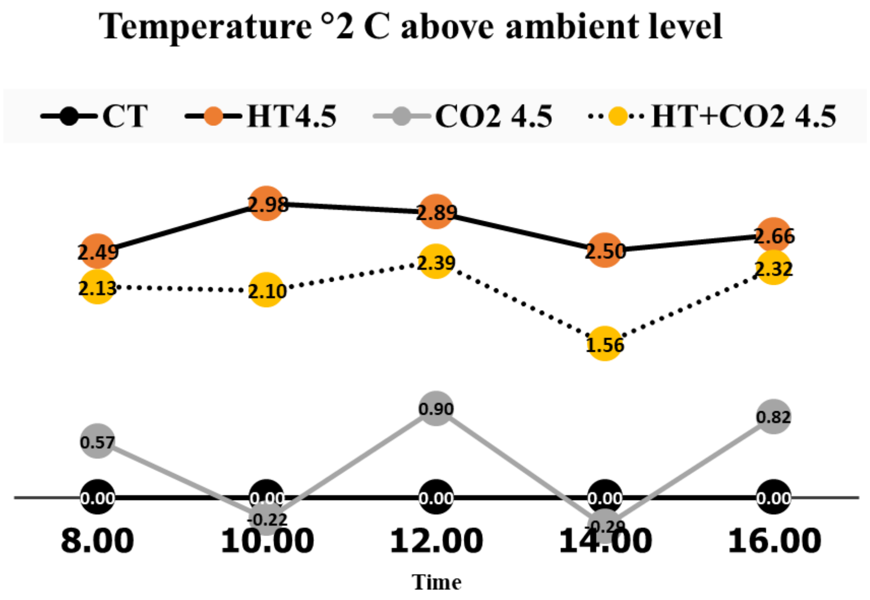


Figure 3. The average temperature with 2 degrees Celsius of global warming in HT4.5 and HT+$CO_2$4.5 treatments compared to CT treatment

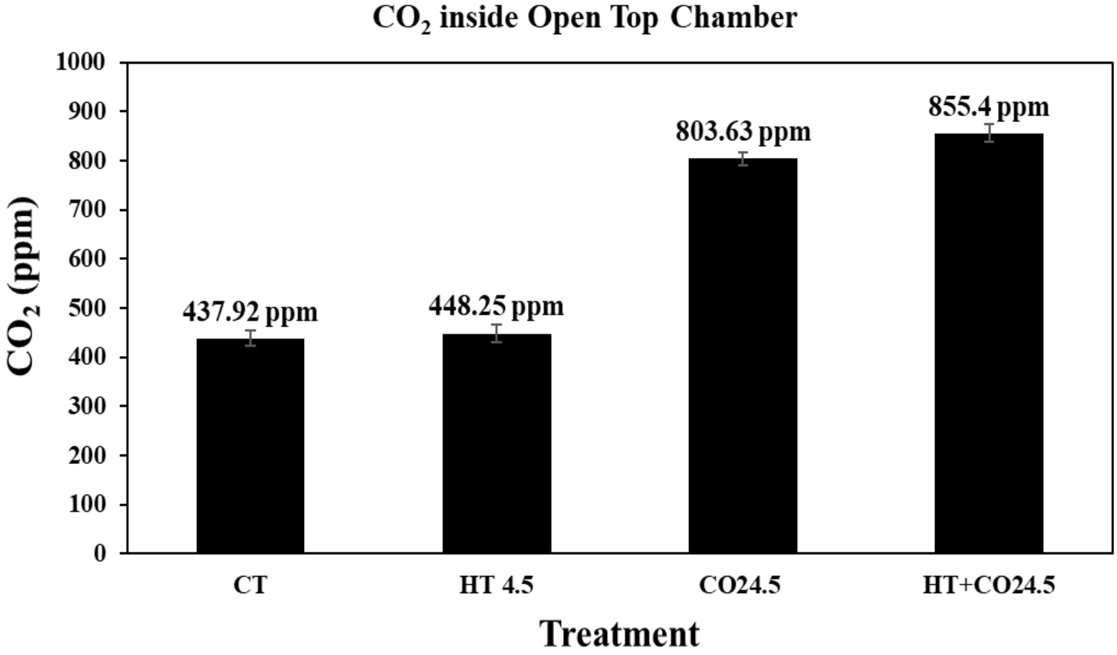


Figure 4. The average CO_2 (ppm) (±SD) with 2 degrees Celsius of global warming in $CO_2$4.5 and HT+$CO_2$4.5 treatments compared to CT treatment

2.2. Experimental Design and Rice Planting Management

This experiment comprised four treatments with four different levels of single and cofactor of temperature and CO_2. Sixteen OTCs (Figure 1) were used for the Randomized Complete Block Design (RCBD) with 4 replications. In the OTCs, the temperature and CO_2 levels remained constant for 10 hr.day^{-1}, however, these levels varied by the ambient weather conditions as follows:

(1) ambient temperature conditions (40.49/30.32°C) with ambient CO_2 (437 ± 15.5 ppm); CT treatment (Control Treatment)
(2) elevated temperature conditions (42.89/32.81°C) with ambient CO_2 (448.25 ± 18.2 ppm); HT4.5 treatment (High Temperature under RCP4.5 Scenario)
(3) ambient temperature conditions (40.20/30.89°C) with elevated CO_2 (863.68 ± 12.2 ppm); $CO_2$4.5 treatment (High CO_2 under RCP4.5)
(4) elevated temperature conditions (42.05/32.45°C) with elevated CO_2 (855.4 ± 17.8 ppm); HT+ $CO_2$4.5 treatment (High Temperature with High CO_2 under RCP4.5)

Thai rice (*Oryza sativa* L.) 3 cultivars viz. Chainat1, Phitsanulok2, and Pathumthani1 were selected to assess their responses to the interaction of elevated temperature and CO_2. In Northern Thailand, these cultivars are widely grown. The ideal growth season for Thai rice is from April to May, and rice planting was done during this time in 2019.

In the experiment, rice seeds were sown in soil block trays containing the same soil mixture. Watering was done daily in the soil block trays until the 10 DAE (10 days after emergence). To assess treatment duration effects, seedlings were transplanted to pots of 37 cm diameter (four plants per pot) and were then allowed to grow inside each chamber (3 pots per cultivar) under 4 conditions treatments. One day before transplantation, compound fertilizer (N-P2O-K2O: 16-16-16; 1.675 g per pot) was applied as basal dressing.

2.3. Yield Component Analysis

At the physiological maturity stage (~120-130 days), 12 plants per cultivar were harvested (random sampling) from each treatment. Selected panicles in each pot were counted as panicles per plant. Yield quality was analyzed by determining in yield component. The total number of grains, filled grains, and unfilled grains per panicle were counted manually from the panicles which were selected randomly. According to the given formula, the total number of filled and empty grains in each panicle was used to compute the percentage of filled grains in each panicle.

$$\textbf{Percentage of filled grains per panicle} = \frac{\textbf{Total filled grain x 100}}{\textbf{Total number of grains}}$$

The 1000 grains weight was determined by digital balance at about 14% moisture content. The thousand kernel weight was determined by randomly selecting one thousand grains from each rice sample and weighing them [25]. The following formula was used to convert the weight of a thousand grains, which was given in grams (gm), into a ton.ha-1 of grain production [26].

$$\textbf{Grain yield (t/ha) = No. of panicles.m}^{-2} \times \textbf{\% filled grains per panicle} \times \textbf{1000-grain weight(g)} \times 10^{-5}$$

2.4. Seed Chemical Composition

2.4.1. Amylose and Amylopectin

To assess the amylose and amylopectin concentrations as determined by colorimetry as stated by Juliano (1971) [26], starch in rice grains (3 cultivars) was determined. The embryo was removed from 100 hulled rice grains from each cultivar, and then the grains were ground into a fine powder using a grain crusher. For 24 hours, a thick, amber-colored container was used to store 40 g of dissolved potassium iodine. Additionally, 80 g of NaOH was dissolved in 2,000 ml of distilled water to create 1N NaOH solution. Then dilute acetic acid was prepared by diluting 115 mL of acetic acid in 2,000 ml of distilled water. Take 0.100 g of well powdered milled rice in a 100 ml volumetric flask and add 1 ml of 95% ethanol and 9.0 ml of 1N NaOH. Shake well and boil over a water bath for 10 minutes and make up the solution to 100 ml using distilled water. Pipette 5 ml from the 100 ml into another 100 ml volumetric flask. Add 1 ml I N acetic acid and then 2 ml iodide solution and make up the volume to 100 ml. Keep one flask blank without adding anything. Shake, stand for 20 minutes, and determine the percent Transmittance at 620 nm using a colorimeter by UV spectrophotometer. The percentage of amylose value will be shown on the screen.

Starch consists of a mixture of two polymers: amylose (linear chain) and amylopectin (branched chain) (Tester et al. 2004). Natural starches consist of about 10%–30% amylose and 70%–90% amylopectin [28]. Hence, the percentage of amylopectin in rice grain was calculated as the given formula.

%Amylopectin in grain = 100 - (% amylose)

2.4.2. Brix Value

By adding 1.5 ml of sulfuric acid (H2SO4) to 1.00 g of well-powdered, milled rice in a 30 ml glass beaker, shaking vigorously for 15 minutes, and then adding 3 ml of distilled water, the brix value was determined. At 100 °C for 10 minutes, the hydrolysis procedure (with H2SO4) was carried out. Once the filter paper was three-quarters full, the hydrolyzed solution was poured through it. Finally, a pocket refractometer was used to test the Brix value (Atago Inc., Tokyo, Japan).

2.5. Statistical Analysis

The experiment was designed as a Randomized Complete Block Design (RCBD) under 4 replications. Data of yield, yield component, amylose, amylopectin content, and Brix value were statistically analyzed by the analysis of variance (ANOVA). Significant differences in parameters were tested at $p<0.05$ (at a 95% confidence level) by DMRT.

3. Results

3.1. Temperature and CO_2 in Open Top Chambers

Daily measurements of CO_2 and air temperature were made within 16 OTCs from the seedling to the mature stages. According to the results, the mean air temperatures and CO_2 concentrations (plus or minus standard deviation) for 10 hours for each treatment are displayed (Figure 2 and Figure 4). The air temperature and CO_2 levels in the control treatment (CT) were 40.92°C 1.92°C and 437 15.5 ppm, respectively. The temperature level was 42.89/32.81°C, and the CO_2 concentration was 863.68/12.2 ppm above ambient.

In this experiment, the raised temperature and CO_2 content were regulated at 2°C (above ambient) and up to 800 ppm (around 400 ppm above ambient). As a result, the raised CO_2 and temperature in the experiment research were set relatively near to the IPCC's forecasted RCP4.5 scenario.

3.2. Seed Yield

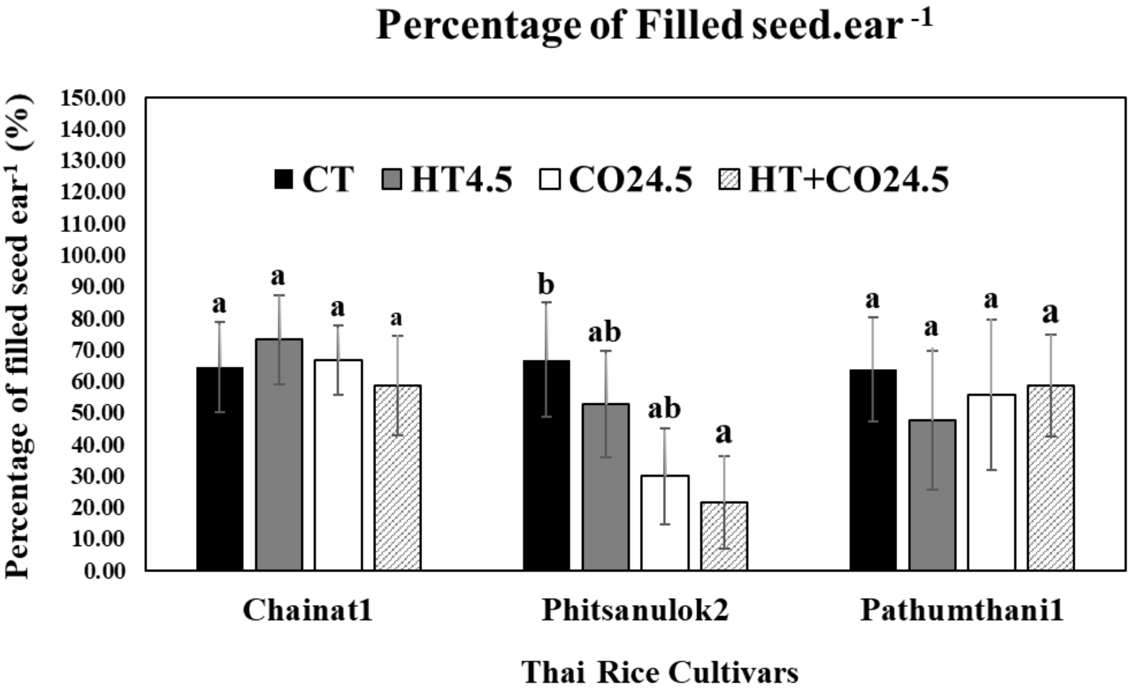
In this experiment, grain yields of 3 rice cultivars in 4 treatments were examined for the percentage of filled grains per panicle, 1000-seed weight, and grain yield ($t.ha^{-1}$) at the maturing stage. These parameters were used in our study for estimating rice yields.

The percentage of filled seed.ear-1 in 4 treatments for 3 rice cultivars was calculated by data of total rice grains and No. of filled $seed.ear^{-1}$. Results in Chainat1 and Pathumthani1 show less sensitivity to elevated temperature and CO_2 than Phitsanulok2. Figure 5 revealed that a significant difference ($P > 0.05$) was not found in both 2 cultivars, whereas there was a significant difference ($P < 0.05$) in Phitsanulok2. The interaction between elevated air temperature by 2°C and CO_2 at ~800 ppm was adequate to cause a piece of evidence of a significant decrease in the percentage of filled seed.ear-1 by 67% in $HT+CO_2 4.5$ treatment (compared to CT).

The results of 1000 seed weight (g) measurements were also revealed in this part. This parameter will vary with rice seed size. Hence, it is one of the important parameters for estimating the seed size including the quantity of grain yield. Figure 6 below shows the 1000 seed weight of 3 rice cultivars. The results were not similar to the percentage of filled $seed.ear^{-1}$ described above. Chainat1 and Phitsanulok2 were sensitive cultivars among rice cultivars because the significant difference ($P<0.05$) in 1000 seed weights was shown in these cultivars. Whereas there was no significant difference ($P<0.05$) in Pathumthani1. Both of single factor and cofactor of elevated temperature and CO_2 could cause a significant decrease in 1000 seed weight in Chainat1. Among rice planting under 3 treatments (except CT), 1000 seed weight was more sensitive to $CO_2$4.5 by the reduction of 46% (compared to CT) in Chainat1. Moreover, 1000 seed weights were also reduced by approximately 40% and 43% under HT4.5 and HT+$CO_2$4.5 treatments. In addition, the statistically significant difference reduction of 1000 seed weight by 8.7% was also shown in Phitsanulok 2 under HT+$CO_2$4.5 treatment.

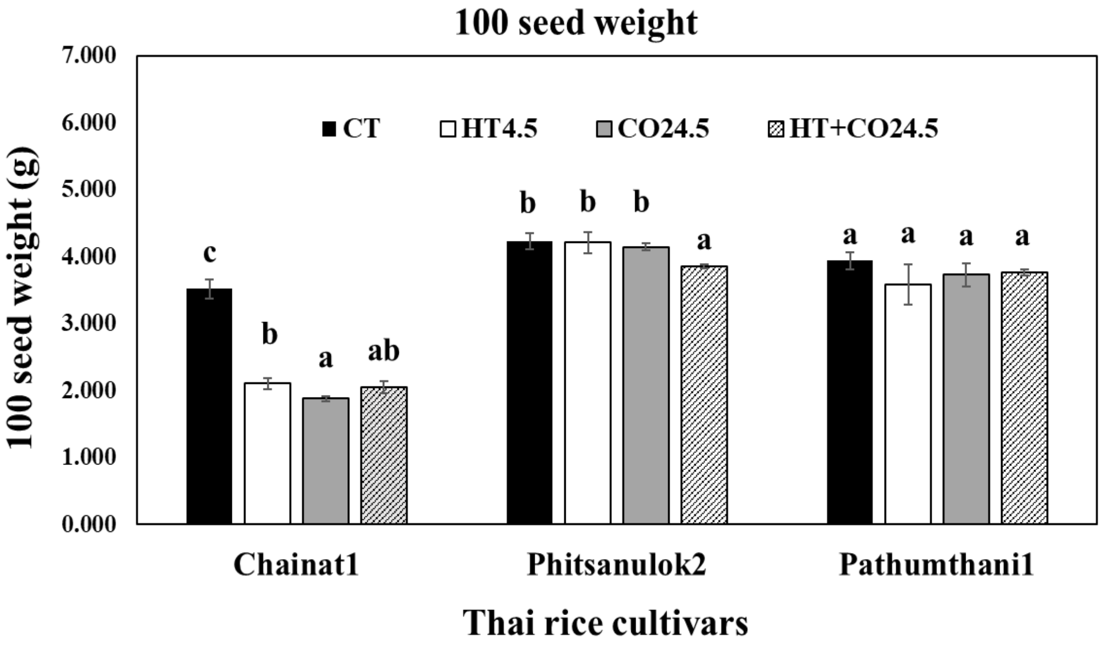
The result of grain yield was shown in Figure 7. The statistically significant difference (P <0.05) results among treatments in grain yield (t.ha^{-1}) were found in 2 rice cultivars; Chainat1 and Phitsanulok2. However, Phitsanulok2 seemed to be more sensitive to elevated temperature and CO_2 interaction than Chainat1 and Pathumthani1 because the 68% reduction appeared under the HT+$CO_2$4.5 treatment. It is estimated that a ~2°C with ~ 400 ppm (CO_2) increase may directly be caused 68% rice yield losses in Phitsanulok2. In contrast, the same situation may induce rice yield to increase by 38.5% for Chainat1. The result in Phitsanulok2 was consistent with the percentage of filled seed.ear^{-1}.

All results in yield components showed that Pathumthani1 was less sensitive to single or co-factor of elevated temperature and CO_2. There were not any statistically significant results in this cultivar.



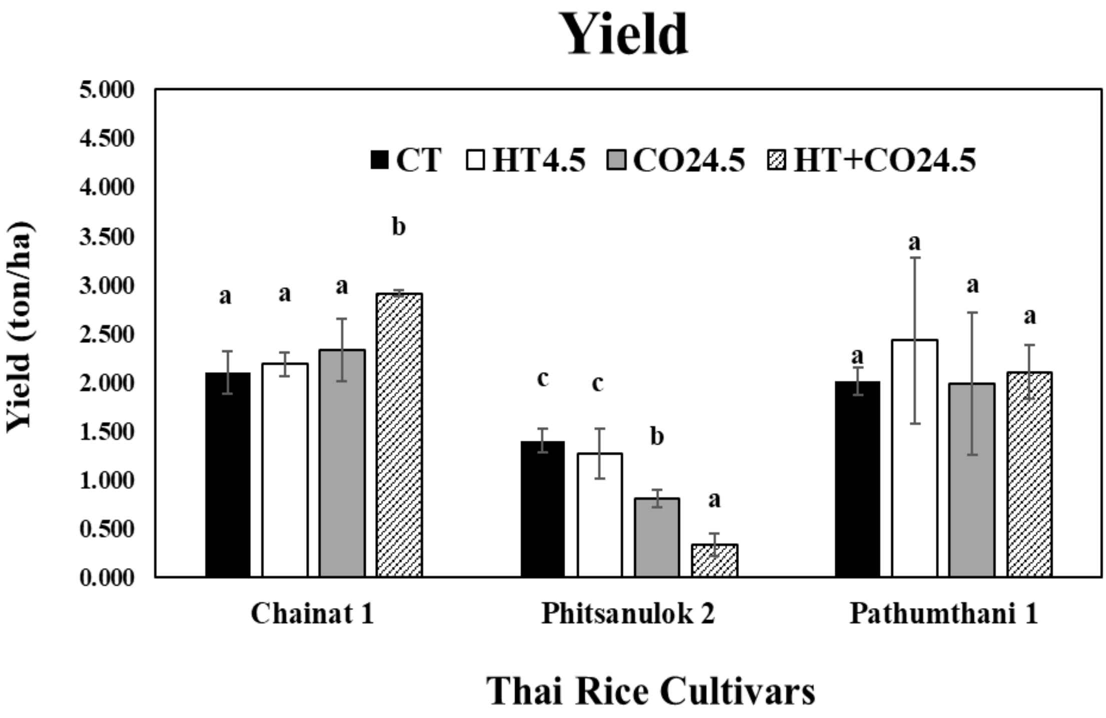
Note: The different letters for each treatment indicate a significant difference at P <0.05. Error bars above each histogram indicated standard deviations (S.D.) observed from samples of each treatment

Figure 5. Mean values (± SD) of filled seed per ear measured at harvest stage of growth and exposed to elevated air temperature levels and CO_2 concentrations in 3 rice cultivars



Note: The different letters for each treatment indicate a significant difference at *P <0.05*. Error bars above each histogram indicated standard deviations (S.D.) observed from samples of each treatment.

Figure 6. Mean values (± SD) of 1000 seed weight (g) measured at harvest stage of growth and exposed to elevated air temperature levels and CO_2 concentrations in 3 rice cultivars



Note: The different letters for each treatment indicate a significant difference at *P*<0.05. Error bars above each histogram indicated standard deviations (S.D.) observed from samples of each treatment.

Figure 7. Mean values (± SD) of grain yield (t/ha) measured at harvest stage of growth and exposed to elevated air temperature levels and CO_2 concentrations in 3 rice cultivars

3.3. Amylose and Amylopectin Content

Amylose and amylopectin contents in rice grains are very important for the textural properties of cooked rice grains. Results of amylose content, amylopectin content, and amylose: amylopectin ratio under the RCP 4.5 situation are shown in figures 8-10.

For all 3 rice cultivars, there was a significant difference (P 0.05) found in these parameters (rice starch). Amylose concentration in 2 cultivars increased in response to simulated temperature or CO_2 increases (Chainat1 and Pathumthani1). In contrast, rice cultivation with the cofactor (HT+CO_2) can reduce amylose content by 1.55%, 3.13%, and 11.52% compared to CT in Chainat1, Phitsanulok2, and Pathumthani1, respectively (figure 8). In this case, Pathumthani1 was reflected mostly under HT+$CO_2$4.5 treatment. In the case of HT4.5 treatment, we observed the increase of amylose content in Chainat1 (Compared to CT) by significantly different ($P < 0.05$).

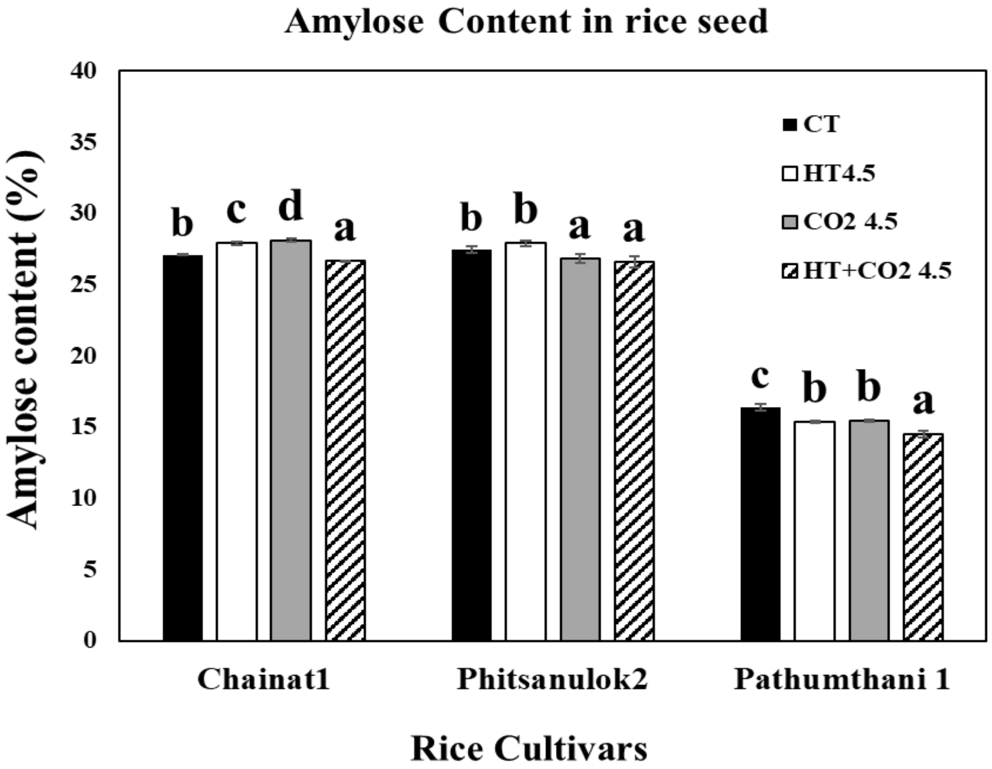
Figure 9 shows amylopectin content in 3 cultivars under these situations. All opposite statistically significant results compared to amylose results were shown in all 3 cultivars. The highest % increase was also observed in Pathumthani1 by 2.27%.

Normally, the ratio of amylose and amylopectin in rice starch is around 0.137±0.001 [28]. Starches with lower amylose content will have higher glycemic indexes [28]. The ratio of amylose: to amylopectin under CT treatment was 0.371±0.001, 0.3784±0.0038, and 0.1961±0.0033 in Chainat1, Phitsanulok2, and Pathumthani1, respectively (Figure 10). A comparison of the results from total treatments revealed all significantly different ($P < 0.05$) reductions of amylose: amylopectin ratios in all 3 cultivars. The highest reduction was observed in HT+$CO_2$4.5 treatment compared to other treatments by 2.12%, 4.28%, and 13.46% in Chainat1, Phitsanulok2, and Pathumthani1, respectively.

3.4. Brix Value

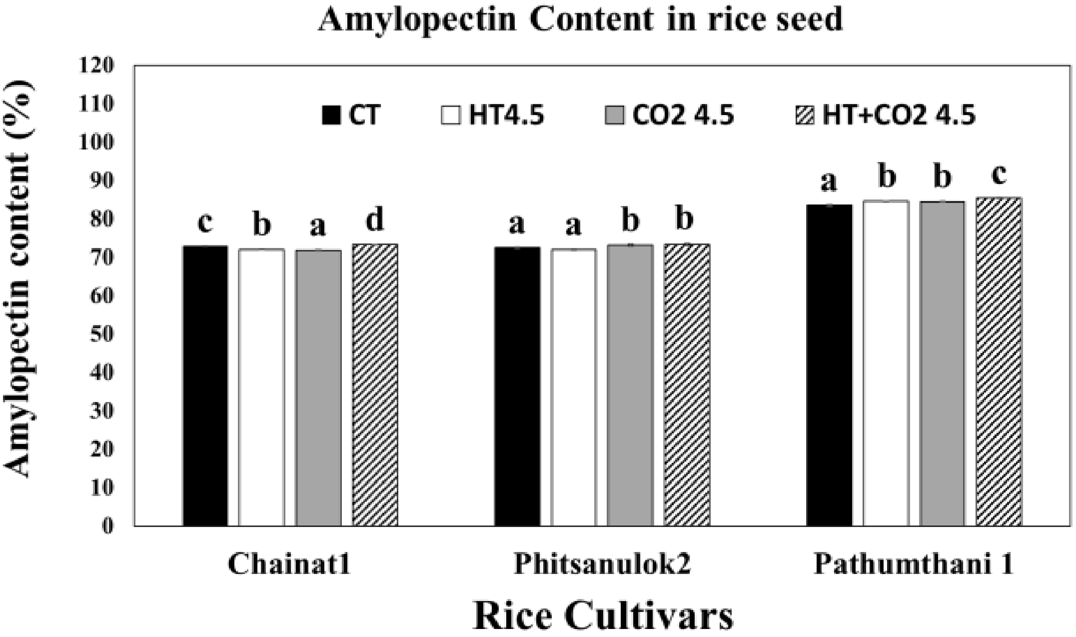
Three cultivars' Brix readings differed considerably (P 0.05) across the four treatments (Figure 11). These findings also corresponded to amylose contents during HT+$CO_2$4.5 treatment as compared to CT. Under the co-factor scenario, the brix values of 3 cultivars, Chainat1, Phitsanulok2, and Pathumthani1, decreased by 33.48%, 11.44%, and 9.29%, respectively. The results also showed that all cultivars' Brix levels might significantly drop as a result of both the single factor and the combination of high temperature and CO_2. In comparison to the other 2 rice cultivars, Pathumthani1 appeared to respond to the HT+$CO_2$4.5 treatment more effectively in terms of amylose content, amylopectin content, and Brix value.

Results in Pathumthani1 revealed that it was the resistant cultivar to cofactor of elevated temperature and CO_2 when considered in yield loss. In contrast, it was the susceptible cultivar when considered in seed chemical under the same situation as the other 2 cultivars.



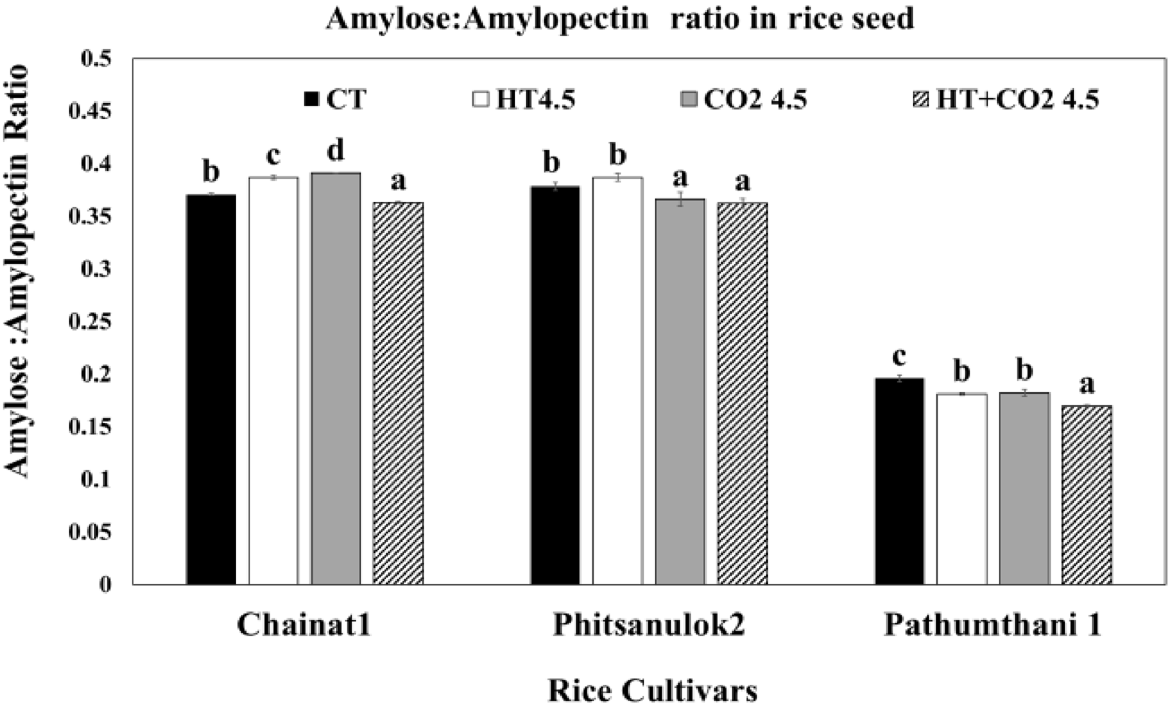
Note: The different letters for each treatment indicate a significant difference at *P <0.05*. Error bars above each histogram indicated standard deviations (S.D.) observed from samples of each treatment.

Figure 8. Treatment means (± SD) of amylose content (%) of 3 rice cultivars affected by elevated air temperature levels and CO_2 concentrations



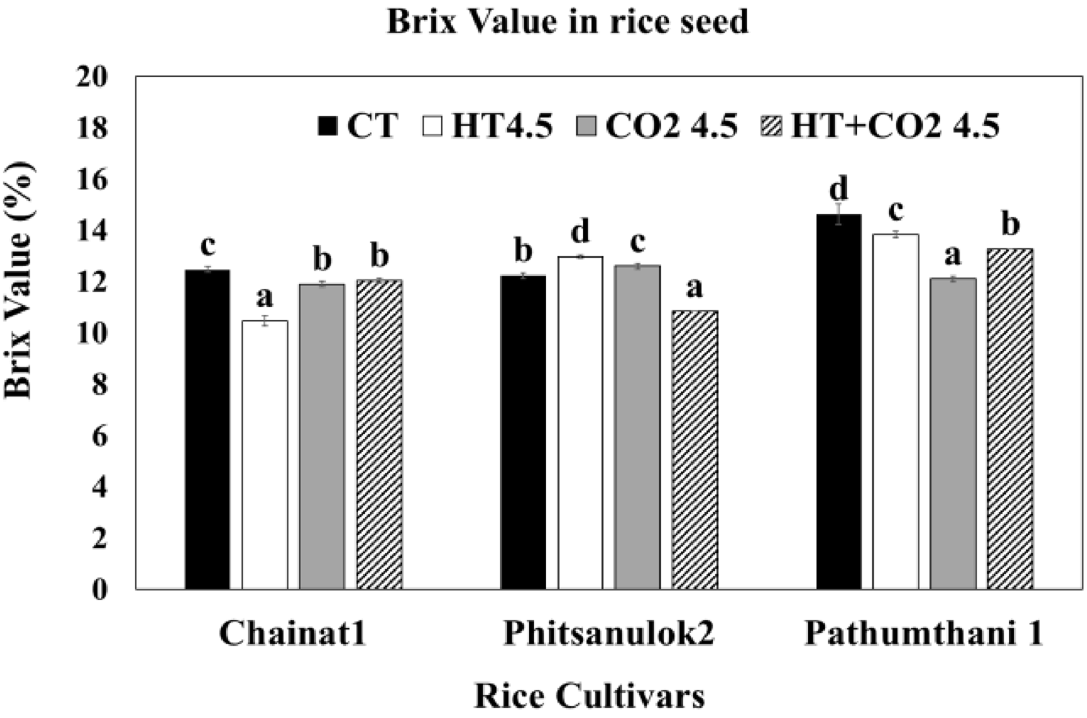
Note: The different letters for each treatment indicate a significant difference at $P < 0.05$. Error bars above each histogram indicated standard deviations (S.D.) observed from samples of each treatment.

Figure 9. Treatment means (± SD) of amylopectin content (%) of 3 rice cultivars affected by elevated air temperature levels and CO_2 concentrations



Note: The different letters for each treatment indicate a significant difference at $P < 0.05$. Error bars above each histogram indicated standard deviations (S.D.) observed from samples of each treatment.

Figure 10. Treatment means of amylose:amylopectin ratio of 3 rice cultivars affected by elevated air temperature levels and CO_2 concentrations



Note: The different letters for each treatment indicate a significant difference at *P <0.05*. Error bars above each histogram indicated standard deviations (S.D.) observed from samples of each treatment.

Figure 11. Treatment means of brix value of 3 rice cultivars affected by elevated air temperature levels and CO_2 concentrations

4. Discussion

The findings from our experiment are discussed below.

The overall results indicated that the interaction between elevated temperature (~2°C increase) with CO_2 up to 800 ppm (~400 ppm increase). This condition was the projected temperature and CO_2 increase under the emission IPCC scenario (RCP4.5) in the future. It was adequate to directly cause grain size reduction, rice yield losses (in Chainat1 and Phitsanulok2), and alteration in seed chemical (amylose, amylose: amylopectin ratio, and Brix value) in all 3 cultivars (Chainat1, Phitsanulok2, and Pathumthani1). Alteration of seed chemical composition in 3 cultivars in this experiment would lead to a nutrient imbalance in rice seed. These data suggest also that Pathumthani 1 is highly tolerant to thermal with CO_2 stress during the grain filling stage because there was no significant difference ($P < 0.05$) appeared in all yield components (Figures 5-7). However, it was the more susceptible cultivar than the other 2 cultivars when considering yield quality: amylose content, amylopectin content, and Brix value.

Results in Chainat1 and Phitsanulok2 demonstrate that they were susceptible to heat with CO_2 stress during the grain filling stage. Grain starch synthesis was also suppressed at the filling stage. In addition, only temperature or CO_2 increase conditions could also reduce the yield component. In consequence, Chainat1 and Phitsanulok2 were also sensitive cultivars, because yield quality and quantity reduction were shown under elevated temperature and CO_2.

According to our hypothesis, higher temperatures and CO_2 during the grain synthesis phase would affect grain formation and reduce starch production in grains that were already developing. The results are consistent with other studies showing that temperature and CO_2 rises will have a detrimental impact on rice output in many regions of the world because high temperatures damage crops' physiological systems [11]. Only a modest temperature variation of 1°C might result in a significant loss in yield due to a decreased number of grains produced. [29].

Temperatures above 32°C have been observed to negatively influence all phases of rice plant growth and development, with 33°C being the most crucial temperature during the blooming stage, according to Wu et al. Most physiological activities, including stomatal opening, photosynthesis, growth, and grain yield, are harmed by high temperatures [30].

Raj et al. [31] studied high-temperature stress of 3.9ºC on rice crops. Scientists discovered that increasing the daily mean temperature of rice from 28°C to 32°C dramatically lowered grain and biomass output. Temperature increases throughout the vegetative and early grain-filling stages of many rice types resulted in a reduced photosynthetic rate in the crop [32].

Many studies have proven that, in the absence of temperature increases, raising CO_2 has the overall impact of increasing crop yields [33]. Increased CO_2 levels in the atmosphere typically boost crop yields by stimulating photosynthetic activities, suppressing photorespiration, and improving water use efficiency. C3 crops, such as rice, typically directly benefit from increased CO_2. Rice plants respond to increased CO_2 levels by decreasing the oxygenase activity of RuBP carboxylase/oxygenase, an enzyme that suppresses photorespiration [20].

Since the rate of photosynthesis in C3 species, such as rice, is still below physiological saturation levels under present levels of atmospheric CO_2, it is expected that rising atmospheric CO2 levels will enhance photosynthesis and therefore productivity in most crops [22].

Horie et al. [34] confirmed that doubling CO_2 concentration resulted in a 30% increase in rice output. Various studies on rice reported that increased CO_2 levels enhanced seedling numbers, photosynthesis, biomass production, and grain production.

Conversely, a concurrent increase in ambient temperature and CO_2 has been shown to reverse the favorable effect of CO_2 [35]. Our results in this experiment are consistent with the above-mentioned. The negative results of rice yield under elevated CO_2 and temperature on rice yield have been reported by several studies.

The modeling studies from India and Bangladesh were applied to assess this point. Lal et al. [36] used the CERES rice and predicted a 20% decline in rice yields in northwestern India due to elevated CO_2 and temperature. Krishnan et al. [37] also used crop simulation studies to the assessed impact of elevated CO_2 and temperature on Indian rice yield. Results showed that increases in the CO_2 concentration up to 700 ppm with +4 °C above the ambient level induced yield loss by 7.63%, 9.38%, and 15.86% under GDFL, GISS, and UKMO scenarios, respectively.

Maniruzzaman et al. [38] studied six regions of Bangladesh using the CERES-Rice model. They evaluated the correlations between rice yield of dry season irrigated rice (Boro) cultivars (BRRI dhan28, BRRI dhan29, and BRRI dhan58) with higher temperatures and CO_2. Findings indicated that grain yield loss would be the largest (13% - 23%) if the temperature rises by 4°C and growth time decrease would be 23 - 33 days. Grain yield declines with rising temperatures of 1°C, 2°C, and 3°C are anticipated to be accounted for by elevated CO_2 levels of 421, 538, and 670 ppm, respectively

The field experiment was also applied to assess. Basmati rice (2 cultivars; PRH-10 and PS-2) was grown under elevated CO_2 (550 mmol/mol) and temperature 35/28°C) conditions to assess photosynthesis and carbohydrate metabolism. At elevated CO_2 with temperature increase induced reduction in photosynthesis of PRH-10 by 11% at the heading stage including a reduction in Sucrose Phosphate Synthase by 25% was also found [20].

Elevated growing temperatures or high CO_2 (with heat stress) have been shown to cause alterations in the importance of grain filling physiological and physicochemical properties of rice starches related to food-processing quality [39,40]. Hence, we hypothesized that heat stress or a combination of temperature and CO_2 increase can affect grain development such as grain filling or reduced grain quality.

Stressful situations under single and combined temperature and CO_2 on plants such as rice may be caused by any disturbing physiological processes including imbalances in the mechanisms of plants; as follows.

Rice grain productivity behaves variably to temperature, with various underlying processes at different phases of reproduction. Heat stress during blooming is a larger danger to rice grain production than heat stress during other phases of development [41]. High temperatures during blooming and the grain-filling phase, according to Dubey et al. [42], diminish yield by producing spikelet sterility and decreasing the length of the grain-filling phase. Due to the elevated temperatures during the ripening stage, aberrant morphology and colour develop in rice, most likely due to reduced enzymatic activity linked to grain filling, respiratory consumption of assimilation products, and decreased sink activity, which may result in grain powdery texture. It causes physiological anomalies in the male organs, lowering seed setting rates; and heat stress during grain filling disrupts carbohydrate metabolism, lowering grain weight [30,41].

Considering some heat stress-tolerant rice cultivars, they may have the potential controlling heat tolerance genes during the reproductive stage [43]. Hence, the differential responses in 3 cultivars under elevated temperature conditions suggest possible unique mechanisms in response to high-temperature stress. In addition, scientists have studied the physiological mechanisms underlying heat stress response (HSR) and tolerance in plants. They discovered the molecular responses and regulation of heat shock proteins (HSPs) under high-temperature conditions [44]. Gorantla et al. [45] reported that heat shock proteins (HSPs) were expressed significantly in the heat Stress tolerance of Rice (*Oryza sativa*). The function of heat shock proteins in heat stress tolerance is well known. It is possible to present candidate genes for differential heat stress tolerance between Pathumthani 1 and other cultivars.

Among the physiological processes in plants, photosynthesis is one of the most vulnerable to heat stress. Rice leaf photosynthesis is significantly decreased at temperatures over 35°C [46]. Yin et al. [47] reported that heat stress has been shown to influence mechanisms such as the rubisco activation state (essential for photosynthetic processes), the maximum efficiency of PSII photochemistry (Fv/Fm), the actual PSII efficiency in the light-adapted state, and/or non-photochemical quenching in rice leaves. On the contrary side, various adverse environmental variables such as dryness, low solar radiation, N deficiency, and low or high temperatures in the onset of panicle primordial and/or spikelet filling stage might boost spikelet sterility and subsequently crop yield [46,48].

Several studies have found that higher temperatures during grain loading have an effect on rice quality. The increasing temperature during the grain-filling time reduced grain weight, implying that physiological sink activity decreased under high-temperature stress. The fast decline in photosynthesis caused by increased temperatures may restrict source activity. [39]. Furthermore, high temperatures during the early phases of grain filling enhance the rate of endosperm cell division, resulting in flimsy cell membranes, fewer granules started, and starch accumulation on fewer building blocks inside thin-walled cells, resulting in greater PGWC and wide chalky patches [15].

Surprisingly, under high CO_2, photosynthesis and net primary output are directly connected to sink capacity in consuming or storing the extra non-structural carbohydrate, which may otherwise lead to photosynthetic adaptation in the source tissue. As a result, cultivars with increased sink size and strength to store or utilize photo-assimilate might profit from higher CO_2 levels by avoiding photosynthetic adaptation and retaining greater leaf level output [21].

Heat stress exposure during the reproductive and grain filling stages, on the other hand, has been shown to diminish rice output by reducing the fraction of viable spikelets, shortening grain filling durations, and reducing sink activity. Furthermore, under heat stress, leaf senescence, which reduces net photosynthesis, and decreased sucrose-starch conversion enzymatic activity are considered to cause ultimate grain weight in rice and other crops such as maize and wheat [21].

Moreover, heat stress during the grain filling stage also leads to reduced utilization of additional non-structural carbohydrates (NSC) in the sink despite increased assimilation supply from leaves, under elevated CO_2. For example, heat stress resulted in reduced grain weight or grain starch content even with a higher sucrose supply from source tissue under elevated CO_2 in rice [21,49,50].

An increase in soil temperature is also caused by the increased air temperature. This change is one of the environmental factors that affect physiological processes in plants. Warming increases the overall availability of nitrogen (N) in soils. However, soil temperature can affect plant N uptake and N form preference [51]. Changes in seed nutrient quality components may be caused by rising soil temperatures. Plant nitrogen (N) absorption and N form choice can be affected by soil temperature. Rates of change in uptake may not be uniform among N forms, however, because NO3- uptake is more sensitive to temperature than NH4+ uptake [52]. Increased soil temperature affects the rate of organic matter breakdown and mineralization of various organic components. It also has an impact on soil water content, conductivity, and plant availability. As a result, soil temperature has a significant impact on plant development through altering water and nutrient absorption [53].

In case of a combination of CO_2 and temperature increase up to stress, the condition induces alteration in plant physiology, this was probably due to the change in kinetic parameters of Rubisco (inactivation of Rubisco) and decreased solubility of CO_2 compared to O_2 that increase photorespiration, decreases in photosynthetic rates, reduction of plastoquinone (PQ), and increase in cyclic electron flow (CEF) around photosystem I (PSI) [20,54].

Many lines of evidence from field or laboratory studies demonstrate that only elevated CO_2 concentrations in the atmosphere also affect plant growth, yield production, or some mechanisms. Rising CO_2 concentrations directly affect plant metabolism, higher CO_2 stimulates net photosynthesis by increasing CO_2 substrate availability for Rubisco and simultaneously suppressing photorespiration. However, at low internal CO_2 concentrations (Ci), Rubisco carboxylation rates limit photosynthesis, and net CO_2 assimilation rates rise steeply as Ci increases. As CO_2 concentrations increase further, photosynthesis becomes limited by the ability to regenerate RuBP and then by the ability to use triose phosphates to produce starch and sucrose [55].

The photosynthesis process is stimulated more when CO_2 concentrations are low than when CO_2 concentrations are higher [56]. Elevated CO_2 reduces stomatal conductance, which affects both the carbon and the water dynamics of vegetation. A study across 40 species grown under 12 Free Air CO_2 Enrichment (FACE) experiments sites demonstrated that increased growth of CO_2 resulted in a 20% decrease in stomatal conductance [57]. A decrease in stomatal conductance induces suppressing nutrient uptake capacity (such as nitrate) leading to inhibiting leaf nitrate (NO_3^-) assimilation in C3 plants then growth declines [58].

Numerous pieces of research have revealed that under elevated CO_2, competing energy requirement of carbon and nitrogen assimilation in plants leads to higher tissue C: N ratio under elevated CO_2, leading to the release of organic matter content, decreasing grain protein and other associated quality parameters in rice [59].

Moreover, exposure to elevated CO_2 and high temperature led to a significant reduction in seed-set and sink starch metabolism enzymatic activity [21].

These alterations under elevated CO_2 and temperature lead to a reduction in starch biosynthesis in developing grain [40]. Plants' main reserve carbohydrate is starch. The starch held in the seeds and tubers of many crops such as rice, maize, wheat, barley, potato, and cassava serves as the primary source of energy in the human nutrition. As previously stated, starch, which is kept in the form of water-insoluble granules, is principally made of two forms of glucose polymers: amylose and amylopectin [39].

Starch is made up of a combination of amylose (linear chain) and amylopectin (branched chain) [60]. The capacity of meal preparation to elevate blood glucose levels is referred to as the glycemic index. The amylose-amylopectin ratio is a Glycemic Index indicator. Glycemic indices will be greater in starches with reduced amylose concentration [28].

Amylose concentration in rice grain is also essential, and temperature during grain filling, in particular, is a key element determining grain quality. Umemoto et al. [61] reported the content of amylose in the endosperm of non-waxy japonica rice (Oryza sativa cv Akitakomachi) under high temperatures. In this experiment, rice plants grown under high temperatures were reduced compared to rice grown under low temperatures. Whereas, Jiang et al. [62] investigated the effects of high temperatures (29/35°C) on Rice (Oryza sativa L.) grain, a non-waxy indica rice. The results suggest that under elevated temperature increased amount of long chains of amylopectin of endosperm in rice grain at high temperature (reduction in amylose). These results were consistent with our results in Thai rice cultivars. Alterations in amylose and amylopectin may be discussed by the following mechanisms.

The remaining starch is amylose, which is made up of linear glucose chains with a few lengthy branches and amylopectin, which form a helical structure inside the granule matrix and are produced by a single enzyme known as granule-bound starch synthase (GBSS) [39]. GBSS, a member of the GT5 glucosyltransferase family, is a crucial enzyme in amylose production [63]. The lowered activity of granule-bound starch synthase in rice endosperms at high temperatures may be primarily responsible for the lower amylose concentration, however other enzymes in the starch biosynthetic pathway are also influenced by high growth temperatures [39].

Rice is made up of several components, but starch is the predominant one, accounting for 80-90% of the overall elements. Starch is a glucose polymer connected together by -D-(1-D-4) or a-D-(1-6) glycosidic linkages [64]. Our results show that under cofactor caused a reduction in both amylose contents and Brix values for all 3 cultivars. We hypothesized that the alteration in seed starch seems to affect Brix values.

As a result, more study is needed to give proof of the effects of climate change on grain formation and starch biosynthesis in the developing grain stages, allowing Thai rice to cope with and offset the negative consequences of climate change.

5. Conclusions

In conclusion, this study was conducted to determine the effect of increased temperatures and elevated CO_2 levels on yield components and seed chemicals of 3 cultivars of Thai rice. The temperature increase was ~+2˚C, with elevated CO_2 concentrations of ~800 ppm based on RCP4.5. Results from the experiment showed that rice yield and seed chemicals reduced under high temperatures along with high CO_2 based on the RCP4.5 scenario. This conclusion was observed in terms of reduced grain yield, seed size, filled seed, amylose content, and Brix value. We found that all 3 cultivars were sensitive to thermal with CO_2 stress because the significant reduction of amylose contents and Brix values appeared. However, considering yield quantity, it seemed that Phitsanulok2 and Chainat1 are more susceptible to elevated thermal and CO_2 stress during the grain filling stage than the Pathumthani1 cultivar.

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