

Influence of Rising Atmospheric CO₂ Concentrations and Temperature on Morpho-Physiological Traits and Yield of Rice Genotypes in Sub Humid Climate of Eastern India

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Abstract

A possible scenario for the end of the 21st century is that the atmospheric CO₂ concentration will be in the range of 510 - 760 $\mu\text{L}\cdot\text{L}^{-1}$ and that the mean global temperature will be 1.5°C - 4.5°C higher than present day. One of the pre-eminent manifestations of climate change is the increase in atmospheric CO₂ concentration. Both CO₂ and temperature are the key variables of global climate and may cause significant changes in crop productivity. An experiment was conducted inside open top chamber (OTCs) in *kharif* season 2014 to evaluate the effects of CO₂ enrichment and temperature rise with condition OTC1 (ambient condition), OTC2 (25% higher CO₂ than ambient), OTC3 (25% higher CO₂ + 2°C > ambient temperature) and OTC4 (2°C > ambient temperature) on physiological traits and yield of rice genotypes to identify the suitable genotypes for changing climatic conditions. The study revealed that rice genotypes performed better under elevated CO₂, with slight changes in development, such as growth and in yield attributing traits, depending on the genotypes. However, the beneficial direct impact of elevated (CO₂) on crop yield can be counteract by elevated temperatures. Rice genotype IR83376-B-B-24-2 was highly responsive while IR84895-B-127-CRA-5-1-1 was least responsive toward elevated CO₂. Physiological traits like relative water content (RWC %), membrane stability index (MSI %), chlorophyll content, photosynthetic rate and TSS content were improved under elevated CO₂. However, responses of these traits were negative with elevated temperature. We point out that studies related to changes in crop physiology and

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yield as a consequence of global climatic changes should be a priority due to their association with food security.

Keywords

Climate Change, Physiological Traits, Rice Genotypes, Yield Attributes

1. Introduction

One of the greatest manifestations of climate change is the increase in atmospheric CO₂ concentration. During the last twelve years, the rate of increase of CO₂ is 1.9 ppm yr⁻¹ and is forecasted to be as high as 570 ppm by the middle and may reach 700 ppm or more by the end of this century [1]. Future climate change and associated impacts will vary from region to region around the globe. Projections suggest that the global temperature will increase by 1.8°C - 4.0°C, depending on the greenhouse emission scenario [1]. FAO and IPCC has estimated that cereals production in India would go down up to 125 mt. and an overall increase of 2.0°C in temperature may cause almost 8% loss in farm level net revenue and around 5% in GDP [2]. The climate change impact on the productivity of rice in Punjab (India) has shown that keeping all other climatic variables remaining constant, temperature increases of 1°C, 2°C and 3°C, would reduce the grain yield of rice by 5.4%, 7.4% and 25.1%, respectively [3]. More studies suggest a 2% to 5% decrease in yield potential of wheat and maize for a temperature rise of 0.5°C to 1.5°C in India [4]. Biomass and yield tend to decline with increasing temperature, as higher temperatures shorten crop duration, enhance respiration and reduce time for radiation interception [5]. Both CO₂ and temperature are the key variables of global climate and may cause significant changes in crop productivity. There is rising evidence suggesting that many C₃ crops, may respond positively to elevate atmospheric (CO₂) in the absence of other stressful conditions [6]. but the beneficial direct impact of elevated (CO₂) can be counteracted by other effects of climate change, such as elevated temperatures, higher troposphere ozone concentrations and altered patterns of precipitation [7] [8]. Studies on various plant species have suggested that climate changes will affect the development, growth and productivity of plants through alterations in their biochemical, physiological and morphogenetic processes [9]. The high yielding varieties of rice and wheat developed through modern technologies have contributed significantly in achieving good harvest [10]. Nevertheless, there is only limited information on dynamics of physiological parameters after the heading stage of rice under high air temperature and CO₂ [11] [22], especially the lack of researches on dynamics of physiological parameters after the heading stage. Therefore, there is a need to develop genotypes that are either tolerant to warming or to identify genotypes which perform better under predicted climate change scenarios. The aim of this study was to address the basic issues of to identify suitable rice and wheat genotypes and their physiological changes inside plant system under projected climate change using Open Top Chamber.

2. Material and Methods

2.1. Field Experiment

This study was conducted in the experimental farm of ICAR Research Complex for Eastern Region, Patna located at 25°35'37"N latitude and 85°05'E longitude and at an altitude of 51.8 m above mean sea level. The land area of open-top chambers (OTCs) had a level topography. The climate of the experimental site is semi-arid with dry hot summer and mild winters. The crop season of rice crop is from July to Oct. (*kharif* season). The soil at the experimental site belongs to the major group of Indo-Gangetic alluvium (Table 1).

Table 1. Soil characteristic of experimental site.

Year	Sand (%)	Silt (%)	Clay (%)	Organic carbon (%)	Soil pH	Bulk density (mg/m ³)	Electrical conductivity (dSm ⁻¹)	Available nitrogen (kg/ha)	Available phosphorus (kg/ha)	Available potassium (kg/ha)
2013-14	29.5	41.5	28.0	0.67	7.3	1.45	0.26	237	27.0	203.2

2.2. Crop Management

Four rice genotypes (R. Bhagwati, IR64, IR83376-B-B-24-2 and IR84895-B-127-CRA-5-1-1) were evaluated inside open top chambers (OTCs) at ICAR-RCER, Patna, in *Kharif* season 2014 with an objective to assess the impact of elevated CO₂ and temperature (2°C > ambient) on morpho-physiological traits and yield. The treatment condition in each OTC was OTC1 (ambient condition), OTC2 (25% higher CO₂ than ambient), OTC3 (25% higher CO₂ + 2°C > ambient temperature) and OTC4 (2°C > ambient temperature). Fields (inside the OTCs) were dry ploughed and leveled but not puddled during land preparation. Twenty one days (21 days) old seedlings from wet bed nursery were transplanted at the rate of 2 seedlings per hill at a spacing of 20 cm × 15 cm in plots. In each plot a uniform plant stand was maintained and standard agronomic practices were followed for raising and maintenance of plants. Plots were fertilized at the rate of 90:60:40 kg N:P:K ha⁻¹. Nitrogen was applied on three occasions (1/3 each at sowing/transplanting as a basal, at 30 days and at 60 days after transplanting), while the P₂O₅ and K₂O were applied as a basal application. The experimental plots were kept weed free by hand weeding. Three replications were maintained for each genotype in each open top chamber. Replications were randomized within the OTCs. The plot size for each replication was 1 m² and about 33 plants per square meter were available. The observations were recorded on ten randomly selected plants per genotype per replication for all the traits, plant height (cm), as well as grain yield (t/ha).

2.3. CO₂ Supply, Temperature Enrichment and Monitoring

All the OTCs were equipped with humidity, temperature and CO₂ sensors. Each open top chamber was divided into four equal quadrants with water proof brick partitioning. In each quadrant 3 replications were maintained. Pure CO₂ (99.7%, v/v CO₂ and less than 10 ppm CO) was released from a commercial grade cylinder fitted with a regulator. Carbon dioxide concentration of air within the elevated CO₂ chambers was maintained around the target concentration by a PC-based real-time data acquisition and control (DAC) system designed based on the principles described in [12]. The air sample from the middle of the chamber was drawn periodically into a CO₂ sensor (NDIR, make Topak, USA) to monitor CO₂ concentration. The set level of CO₂ was maintained with the help of solenoid valves which were controlled by Program Logic Control (PLC) and Supervisory Control and Data Acquisition (SCADA) system running Winlog software (Make SELCO, Italy). A data logger recorded the mean CO₂ within all chamber sat 15-min intervals. The CO₂ supply was switched on and temperature maintained only during the daylight hours (*i.e.* from 09:00 to 17:00 h). In the OTCs with elevated temperature, reference temperature was obtained from the control OTC and air temperature was increased 2°C above ambient chamber by infra red (IR) heating tubes, and controlled by the SCADA system. The chambers were washed regularly with a gentle stream of water to remove the dust and to maintain transparency.

2.4. Weather during Crop Season

Daily maximum and minimum temperatures, maximum and minimum relative humidity, daily rainfall were recorded from the meteorological observatory of the ICAR Research Complex, Patna. Mean daily maximum and minimum temperatures and relative humidity (RH) inside the OTC were recorded using data-logger (Figure 1).

2.5. Plant Sampling

Rice plants with uniform development process were tagged in each replication at heading stage in each plot. Tagged plants of three hills from each plot were sampled at anthesis stage, with flag leaves removed from plants for physiological parameter measurements as follows:

2.6. Relative Water Content

Leaf relative water content (RWC %) was estimated by recording the fresh weight, turgid weight of 0.5 g fresh leaf samples by keeping in water for 4 h, followed by drying in hot air oven till constant weight was achieved [13].

$$\text{RWC (\%)} = \left[\frac{(\text{Fresh wt.} - \text{Dry wt.})}{(\text{Turgid wt.} - \text{Dry wt.})} \right] \times 100$$

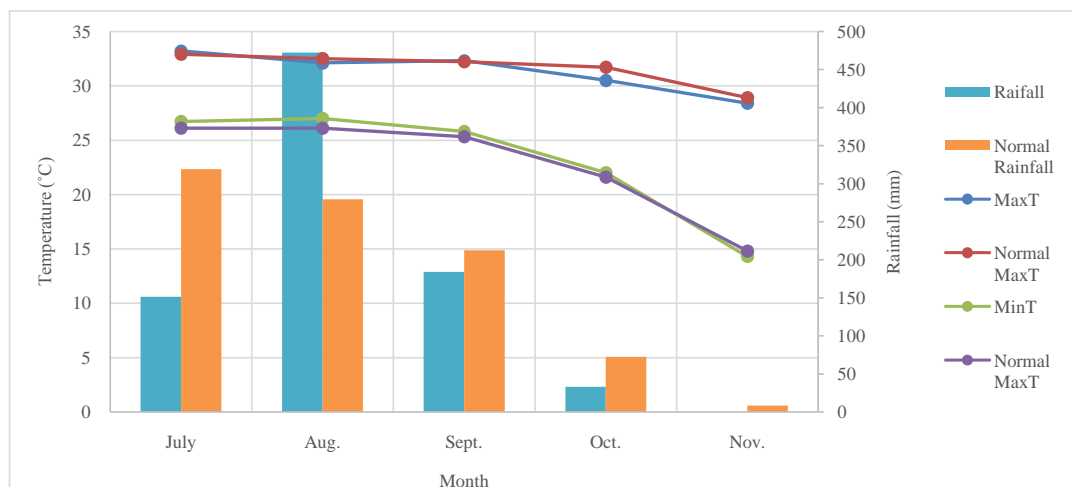


Figure 1. Average climatic condition of experimental site during *Kharif* season (July to October).

2.7. Membrane Stability Index

Membrane stability index (MSI %) was estimated as per [14]. Leaf material (100 mg), in two sets, was taken in test tubes containing 10 ml of double distilled water. One set was heated at 40°C for 30 min in a metabolic water bath, and the electrical conductivity of the solution was recorded by conductivity bridge (C_1). Second set was boiled at 100°C on a boiling water bath for 10 min, and its conductivity was measured by conductivity bridge (C_2). Membrane stability index was calculated as:

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

2.8. Total Chlorophyll Estimation

Estimation of chlorophyll content in plants is based on the absorption of light by chlorophyll extracts prepared by incubating the leaf tissues in DMSO (Dimethyl sulfoxide). DMSO renders plasmalemma permeable thereby, causing the leaching of the pigments [15]. Absorbance was recorded at 663 and 645 nm using DMSO as blank and was expressed as $\text{mg} \cdot \text{g}^{-1} \text{FW}$

$$\text{Total chlorophyll} = (20.2 \times \text{OD}_{645} + 8.02 \times \text{OD}_{663}) \times V/1000 \times w$$

2.9. Net Photosynthetic Rate

Rate of photosynthesis was measured on leaves using portable Infrared Gas Analyzer (IRGA LI-6400 Model). The rate of photosynthesis was measured by operating the IRGA in the closed mode. The photosynthetic rate was determined at anthesis stage in the upper most fully expanded leaf between 10 a.m and 11.30 a.m by providing artificial light source of light intensity $1200 \mu\text{mol} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$. The net photosynthetic rate was expressed as $\mu\text{mol} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$.

2.10. Total Soluble Sugars

Total sugar was determined the by Anthrone reagent method [16].

One ml of sugar sample was taken and to this 4 ml solution of anthrone reagent was added. The mixture is heated on a boiling water bath for 8 min followed by cooling. The optical density of green to dark green colour was read at 630 nm in UV-visible spectrophotometer (model Specord Bio-200, Analytik Jena, Germany). A blank and two freshly prepared glucose standards were also included with each set of samples.

2.11. Statistical Analysis

The data were analyzed statistically using factorial complete randomized design (CRD) and CD at 5% ($p = 0.05$) and ANOVA were calculated. The analysis was done using Statistics 8.1 software programme.

3. Results and Discussion

The concentration of CO₂ changed from morning to evening. The concentration of CO₂ was higher during morning hour after that due to increase in consumption by plants as the day progressed led to decrease in concentration inside the OTCs. Moreover, during evening hour the concentration of CO₂ again rises (**Figure 2**). The temperatures inside OTCs were different in each OTC. A summary of temperature condition inside OTCs were presented in **Table 2**.

3.1. Relative Water Content (RWC%)

Relative water content of rice genotypes was measured to assess the water status of the plants inside open top chambers (**Figure 3**). The mean RWC was increased 5% under elevated CO₂ condition across the cultivars while there was decline in mean RWC (21%) under elevated temperature condition as compared to ambient condition. Genotypic differences were also observed inside OTCs under different set of CO₂ and temperature condition. Rice plants IR83376-B-B-24-2 and R. Bhagwati showed enhancement in RWC by around 7% and 5% treated with elevated CO₂ as compared to plant grown under ambient condition while there was decline in RWC by around 4% and 13% grown under elevated temperature as compared ambient condition. While, in IR84895-B-127-CRA-5-1-1 and IR 64 enhancement was 2% and 6%, respectively. The decline in RWC was more in IR84895-B-127-CRA-5-1-1 and IR 64 rice genotypes, 29% and 18%, respectively due to elevated temperature. RWC declined more slowly at elevated CO₂ [17]. Higher CO₂ levels will influence stomatal behavior beneficially by reducing water loss through transpiration, thus increasing water use efficiency [18].

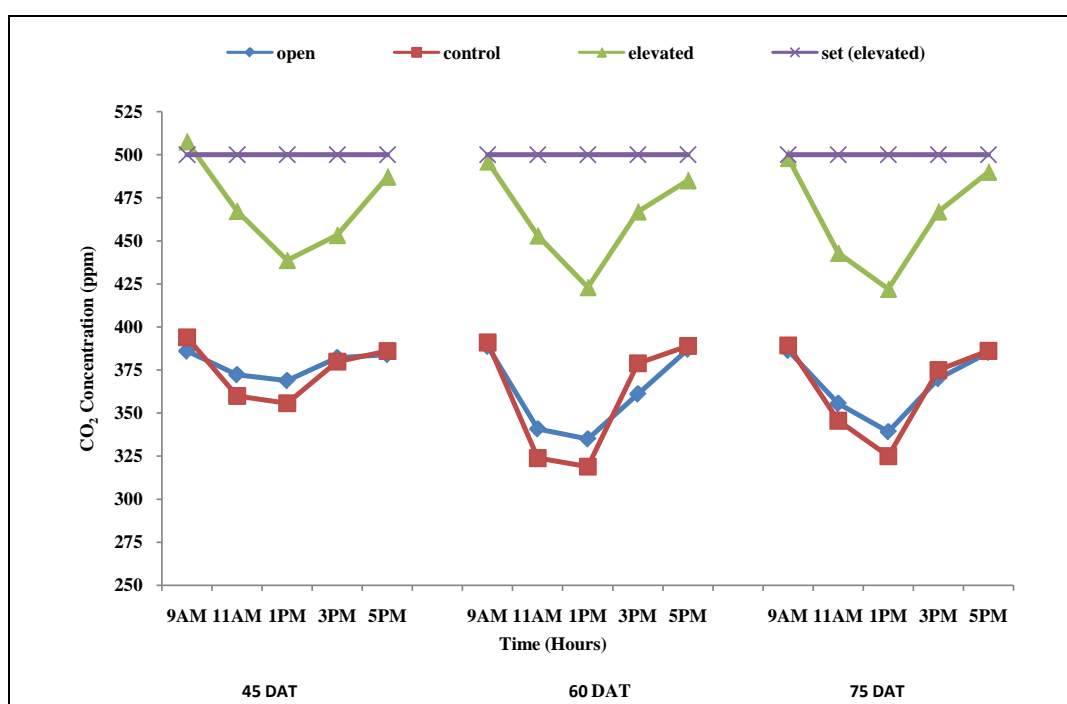


Figure 2. Dynamics of CO₂ concentration and consumption inside open top chamber (OTC 1, 2) and open field at 45, 60 and 75 days after transplanting.

Table 2. Average temperature variation inside open top chambers (OTCs) and field condition (met data) at 45, 60 and 75 days after transplanting (DAT).

Days after transplanting	OTC 1	OTC 2	OTC 3	OTC 4	Field condition
45 DAT	28.3	28.7	31.0	30.2	28.0
60 DAT	28.1	28.5	30.1	29.8	27.3
75 DAT	29.4	29.7	31.3	30.7	28.9

3.2. Membrane Stability Index (MSI%)

Membrane stability index (%) was measured to assess the stability of the cell membrane of the plants leaf inside open top chambers under elevated CO₂ and temperature condition (Figure 4). The mean MSI (11%) was increased under elevated CO₂ condition across the cultivars while there was decline in mean MSI (14%) under elevated temperature condition. Rice plants IR83376-B-B-24-2 and R. Bhagwati showed enhancement in MSI by around 14% and 10% treated with elevated CO₂ while there was decline in MSI by around 11% and 12% with elevated temperature as compared ambient condition. While, in IR84895-B-127-CRA-5-1-1 and IR 64 the increment in MSI due to elevated CO₂ was 7% and 12%, respectively. The injury of cell membrane structure and function due to elevated temperature was also reported [19]-[21].

3.3. Chlorophyll Content (mg/g DW)

The mean chlorophyll content was increased under elevated CO₂ across the genotypes while there was decline in mean chlorophyll content under elevated temperature condition (Figure 5). Plants of IR83376-B-B-24-2 and R.

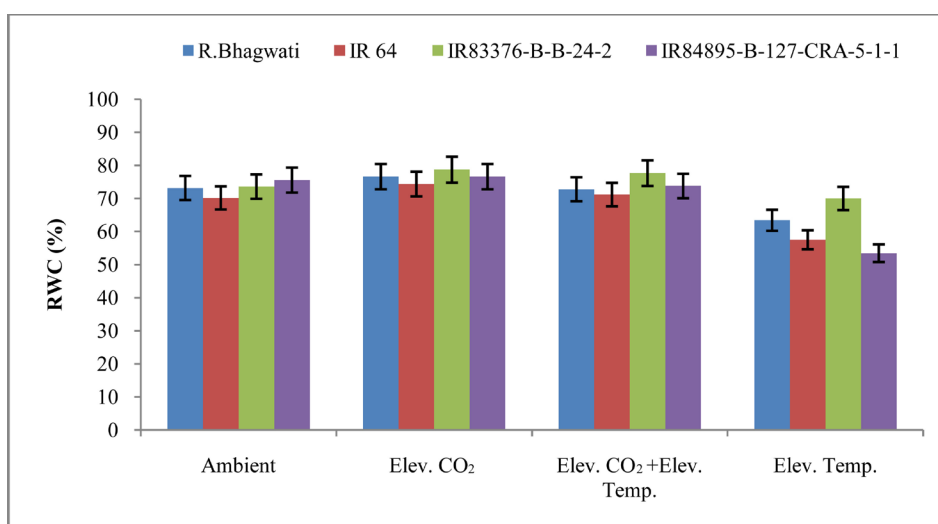


Figure 3. Relative water content (RWC %) of rice genotypes grown under different climatic condition inside open top chambers.

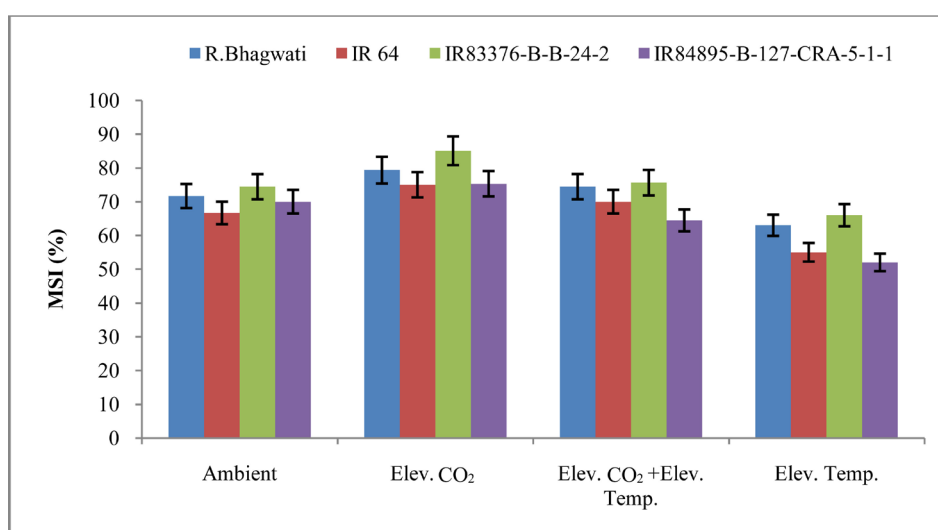


Figure 4. Membrane stability index (MSI %) of rice genotypes grown under different climatic conditions inside open top chambers.

Bhagwati grown under elevated CO₂ conditions showed enhancement in chlorophyll content by around 24% and 23% while there was decline in chlorophyll content by around 16% and 15% when grown under elevated temperature. While, in IR84896-B-127-CRA-5-1-1 and IR 64 it was 6% and 12%, respectively. However under elevated temperature condition the decline in chlorophyll content in rice genotypes IR84895-B-127-CRA-5-1-1 and IR 64 was 27% and 18%, respectively. [22] reported that high air temperature during heading stage negatively influenced SPAD value (relative content of chlorophyll) in rice flag leaves, significant reduction occurring with the continuous increment of air temperature.

3.4. Photosynthetic Rate ($\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$)

Study revealed that photosynthetic rate of rice genotypes under elevated CO₂ treatment was significantly ($p < 0.05$) greater than the ambient condition (Figure 6). However, the maximum photosynthetic rate achieved by rice genotype IR83376-B-B-24-2 with elevated CO₂ (*i.e.* 28.3). Photosynthetic rate under ambient CO₂ was (20.1) significantly ($p < 0.05$) lower than that of elevated CO₂ (26.4) treatment. Elevated temperatures have negative effect on plant photosynthetic rate. The mean photosynthetic rate was reduced (21%) under elevated temperature

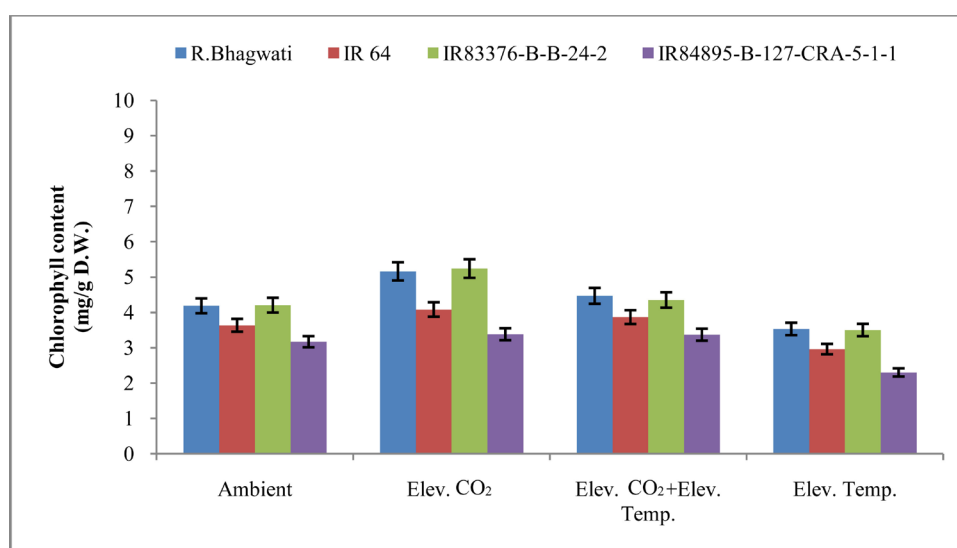


Figure 5. Chlorophyll content of rice genotypes grown under different climatic conditions inside open top chambers.

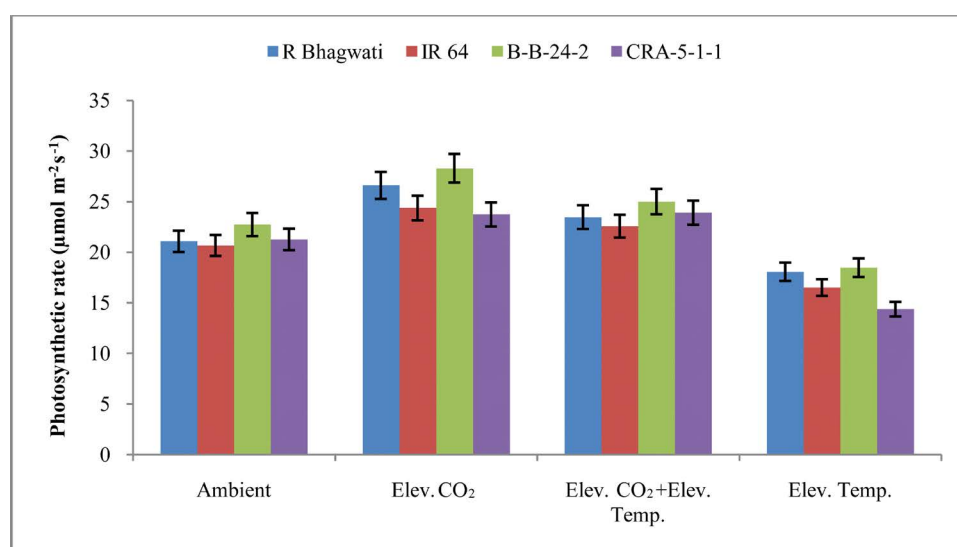


Figure 6. Photosynthetic rate of rice genotypes grown under different climatic conditions inside open top chambers.

across the genotypes, however rice genotype IR83376-B-B-24-2 was least affected due to elevated temperature. Elevated temperature have pronounced negative effect on the genotypes IR84895-B-127-CRA-5-1-1 and showed maximum decline (32%) in photosynthetic rate as compared to ambient condition. [23] [24] also reported that elevated CO₂ concentrations stimulate photosynthesis, leading to increased plant productivity and modified water and nutrient cycles. Elevated CO₂ concentrations may enhance potential net photosynthesis of C₃ plants because ribulose-1, 5-bisphosphate carboxylase/oxygenase (rubisco), an enzyme involved in both CO₂ fixation and photorespiration [25]. Thus, an increase in ambient CO₂ raises the leaf internal CO₂ concentration and the CO₂/O₂ ratio at the rubisco site, favoring carboxylation over oxygenation in ribulose-1, 5-bisphosphate (RuBP). In general, approximately 60% of assimilates demanded by rice grain-filling are derived from post-anthesis photosynthetic production produced by flag leaves, which prominently contributes to grain filling. Therefore, the photosynthetic ability of flag leaves is crucial for the determination of grain yield. [11] found that the impaired net photosynthetic rate by high temperature was mainly attributed to the reduction of chlorophyll content as well as activities of activating enzyme (RuBisCO) and carboxylase (RuBP) involved in photosynthesis in flag leaves.

3.5. Total Soluble Sugar Content (mg/g DW)

Total soluble sugar content of the elevated CO₂ treatment was significantly ($p < 0.05$) greater than the control OTC at all times (Figure 7). The mean total soluble sugar content was increased under elevated CO₂ across the genotypes. The maximum total soluble sugar content achieved by rice genotype IR83376-B-B-24-2 with elevated CO₂ (*i.e.* 4.62). Elevated temperatures have negative effect on total soluble sugar content. Rice genotype IR83376-B-B-24-2 was least affected due to elevated temperature. Elevated temperature have pronounced negative effect on the genotypes IR84895-B-127-CRA-5-1-1 and showed maximum decline in total soluble sugar content as compared to ambient condition. Heat triggered a significant decrease in sugar contents of flag leaves, implying that photosynthetic mechanism was severely impaired along with the reduction of photosynthesis under high temperature, which was in agreement with the reports by [11].

3.6. Yield and Yield Attributes

Results revealed that yield and yield attributes traits of all the genotypes of rice showed positive response with elevated CO₂ and negative with elevated temperature (Table 3). The grain yield increased with elevated CO₂ but declined with elevated temperature across the genotypes. Rice genotype IR83376-B-B-24-2 (4.81 t·ha⁻¹) followed by Rajendra Bhagwati (4.52 t·ha⁻¹) produced higher yield in 25% higher CO₂ concentration (500 ppm) than other two varieties. Higher reduction in yield was observed in genotype IR84895-B-127-CRA-5-1-1 under

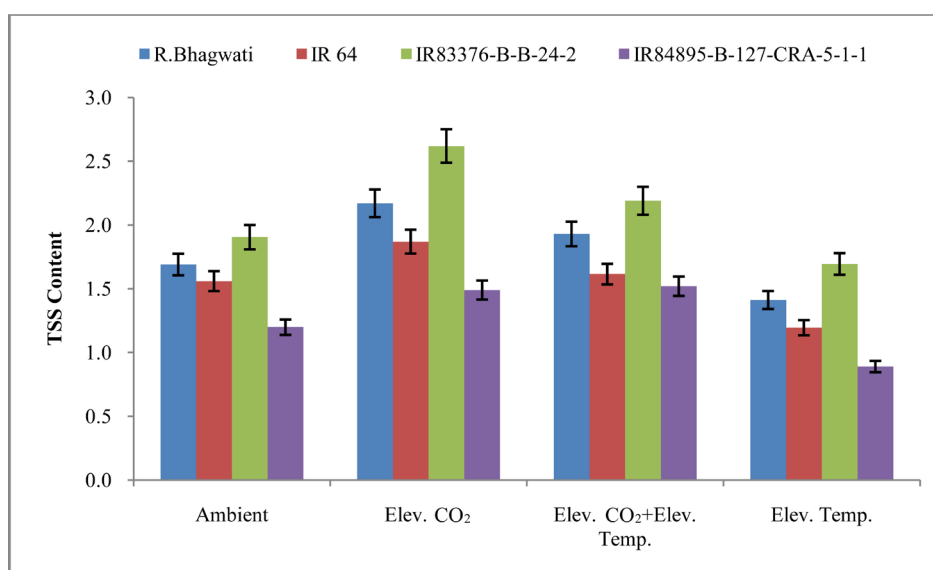


Figure 7. Total soluble sugar (TSS) of rice genotypes grown under different climatic conditions inside open top chambers.

Table 3. Yield contributing traits and yield of rice inside open top chambers (OTCs).

Cultivar	Treatment	Plant height (cm)			Panicle length (cm)			Grains panicle ⁻¹			1000 grain weight (g)			Grain yield (t·ha ⁻¹)		
R. Bhagwati	Ambient CO ₂	129			28.12			198			22.19			3.91		
	Elevated CO ₂ (25% higher > Ambient)	137			30.48			215			23.90			4.52		
	Elevated CO ₂ + Elevated Temp (2°C)	157			29.94			207			22.91			4.26		
	Elevated Temp (2°C) > Ambient	149			27.96			194			17.48			3.19		
IR64	Ambient CO ₂	132			28.56			218			23.68			3.78		
	Elevated CO ₂ (25% higher > Ambient)	142			30.74			242			25.03			4.23		
	Elevated CO ₂ + Elevated Temp (2°C)	160			29.46			219			24.14			4.02		
	Elevated Temp (2°C) > Ambient	155			28.6			198			18.69			3.42		
IR83376-B-B-24-2	Ambient CO ₂	153			31.32			228			24.09			4.18		
	Elevated CO ₂ (25% higher > Ambient)	166			35.14			245			27.01			4.81		
	Elevated CO ₂ + Elevated Temp (2°C)	177			32.16			233			25.08			4.49		
	Elevated Temp (2°C) > Ambient	170			28.96			231			20.52			3.9		
IR84895-B-127-CRA-5-1-1	Ambient CO ₂	93			19.88			109			21.77			3.56		
	Elevated CO ₂ (25% higher > Ambient)	95			20.42			115			22.07			3.88		
	Elevated CO ₂ + Elevated Temp (2°C)	104			20.14			113			22.04			3.70		
	Elevated Temp (2°C) > Ambient	103			19			104			17.10			2.91		
Factors		O	V	OXV	O	V	OXV	O	V	OXV	O	V	OXV	O	V	OXV
LSD (p = 0.05)		3.57	3.57	7.14	1.14	1.14	2.29	8.97	8.97	17.94	0.53	0.53	1.06	0.10	0.10	0.20
SEm+		1.78	1.78	3.57	0.57	0.57	1.15	4.49	4.49	8.98	0.24	0.24	0.49	0.04	0.04	0.09
CV (%)		4.06			6.60			7.39			2.23			2.47		

O—OTC; V—Variety; OxV—Interaction.

elevated temperature. [26] also reported that increasing CO₂ concentration in the atmosphere could lead to higher crop yields. [27] found that grain yield of rice was declined by 10% for each 1°C increase in the growing season minimum temperature above 32°C. [28] analyzed the impacts of elevated CO₂ and temperature on irrigated rice yield in eastern India by ORYZAI and info crop-rice models, and the result shows that increased CO₂ concentration can increase the rice yield, which is concerned with the sterility of rice spikelet's at higher temperature, the sowing time and the selection of genotypes.

3.7. Conclusions and Implications

Improved physiological traits (RWC, membrane stability, chlorophyll content, photosynthetic rate and TSS) may benefit rice genotypes under elevated CO₂ and temperature conditions. By virtue of greater membrane stability and photosynthetic rate, IR83376-B-B-24-2 and Rajendra Bhagwati may be assumed as tolerant among the studied rice genotypes for growing in changing climatic conditions. Since rice is a staple food crop, long term studies may provide better understanding on the efficiency of such genotypes which are increasingly associated with food security.

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