

Rice growth and yield characteristics under elevated carbon dioxide and nitrogen management

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Abstract. The atmospheric carbon dioxide (CO₂) concentration is increasing and the on crop production needs to be investigated. A pot experiment was conducted in open top chambers (OTC) to determine the response of rice to elevated CO₂ (eCO₂) under varying time of nitrogen (N) application. The results revealed that photosynthesis, root and shoot dry matter production, yield components and nutrient absorption were favored at eCO₂ when N applied up to flowering stage (FT) of rice. However, the N application up to FT of rice also significantly improved percent filled grain, reduce spikelet sterility and rice yield increased by 18 to 20% under eCO₂. Rice plant absorbed higher amount of Zn, Ca, Mg, and Fe at eCO₂ when N was applied up to FT. Amylose was higher but protein percentage was lower at eCO₂. These results indicate that to maximize rice yield under eCO₂, it is important to supply N up to FT of rice in order to increase grain fertility and reduce spikelet sterility.

Key words: crop yield, gas exchange, grain fertility, plant nutrition, rice, spikelets sterility.

INTRODUCTION

Currently, the atmospheric CO₂ concentration is increasing much faster than previous (Bereiter et al., 2015; IPCC, 2019) and it would have a significant impact food production of the world through affecting plant growth, development, grain yield and quality (Raj et al., 2019). The C₃ crops respond more strongly to the eCO₂ than C₄ crop (Jablonski et al., 2002). The photosynthesis (Pn) of crops increased significantly under eCO₂ resulting increase in biomass and yield of C₃ plants (Chunwu et al., 2016). On the other hand, stomatal conductance (Gs) and transpiration rate (Tr) of plants reduced under eCO₂ condition (Chunwu et al., 2016). Rice (*Oryza sativa* L.) is one of the most important

crops in Asia (FAOSTAT, 2017). As a C₃ plant, growth and yield of rice are markedly enhanced by eCO₂ (Pachauri et al., 2014). Previous studies have shown that leaf area, shoot dry matter (DM), panicle and grain number per unit area and grain yield of rice were enhanced due to eCO₂ (Yang et al. 2006; Sasaki et al. 2007; Satapathy et al., 2015). Increased growth of the crops under eCO₂ condition will require higher nutrient uptake and assimilation. Because, rice production is greatly influenced by nutrient management (Roy et al., 2015); and the nutrient absorption of a crop is determined by the time of nutrient application. The eCO₂ has a dilution effect on nutrient uptake by the crop and nutrient levels in plants is decreased (Li et al., 2013). On the other hand, plants removed greater amount of nutrients like P, K, Mg, Fe, B and Mo from soil and translocated to the grain under eCO₂. Rice crops exhausted soil minerals to great extent leading to very low available nutrient content in soil after harvesting of the crops (Wang et al., 2011). Therefore, the demand for nutrients by crops might also get changed in future under eCO₂ condition.

As mineral fertilizer, nitrogen (N) is the main nutrient associated with yield (Jing et al., 2016). For synchronizing plants growth and reducing loss, N is recommended to apply in splits starting from transplanting to flowering. The application of N at early growth stage will helps to produce sufficient shoot biomass, but application at panicle initiation (PI) stage increase rice grain yield (Mamun et al., 2016). Therefore, the time of application be adjusted to achieve high yield under eCO₂ condition and high proportion of N should be applied at the late growth stage of rice, especially after PI stage (Yang et al., 2007; Wang et al., 2011). Early studies also suggested that recommended N might not be enough and higher amount should be applied for getting high yield under eCO₂ condition (Razzaque et al., 2009). However, Pilipavicius et al. (2006) showed a negative effect of higher CO₂ concentration on the early growth of *Chenopodium album*. In this situation, this study was conducted to determine the effect of eCO₂ on photosynthetic traits, grain production and nutrient absorption in rice. Besides, this study will help to identify a suitable time of N application for higher grain production of rice under the world's climate change scenario especially, under the changing CO₂ concentration.

MATERIALS AND METHODS

Experimental site and planting materials

A pot experiment was conducted inside OTC at Bangabandhu Sheikh Mujibur Rahman Agricultural University (24.09° N latitude and 90.26° E latitude), Bangladesh in boro. The OTCs (area 9 m²) were constructed with iron frame that installed on the ground according to Uprety (1998). For growing rice plants, 96 pots (size: 24 cm diameter and 27 cm height) were filled up with wet soil. Where amount of soil per plot was almost 13 kg other than water and blank pot weight was 0.5 kg. After potting, triple super phosphate, muriate of potash, gypsum, ZnSO₄ were added @ 0.65, 0.72, 0.52, 0.065 g pot⁻¹, respectively. Physic-chemical properties of initial soil was determined during pot preparation. Initial soil chemical parameters were pH 5.73, soil organic matter 1.70%, total N 0.082%, available P 10.22 µg g⁻¹, available K 0.41 meq per 100 g soil. Forty-day old seedlings of BRRI dhan28 were transplanted on 20 January 2018. Transplanting was done by hand with two seedlings hill⁻¹ pot⁻¹. All the fertilizers except N were applied before transplanting of rice seedlings.

Experimental design and setup

A randomized complete block design with eight replications was used for the experimentation. The treatments comprised of two factors. Factor A consisted of four growing conditions- i. OTC with 500 ppm (elevated CO₂) (eCO₂), ii. OTC with 450 ppm CO₂ (intermediate CO₂ concentration, iCO₂), iii. OTC with ambient CO₂ (400 ppm CO₂) (aCO₂) and iv. Open field (OF, 380 ppm CO₂). Factor B was the timing of N application, viz. i. N1 = 1/3rd N at early tillering (ET) + 1/3rd at active tillering (AT) + 1/3rd before PI, ii. N2 = 1/3rd N at ET + 1/3rd before PI + 1/3rd at booting stage, and iii. N3 = 1/3rd N at ET + 1/3rd before PI + 1/3rd at FT. The rate of N was 2.0 g plot⁻¹ (320 kg ha⁻¹). The CO₂ gas was supplied to the OTC chambers from CO₂ gas cylinder using a blower from 7 days after transplanting to physiological maturity of rice. A portable Pn system (model: LICOR 6200, Lincoln, Nebraska) was used to determine the CO₂ concentration inside the OTC regularly. In eCO₂ treatment, the CO₂ concentration fluctuated from 490 to 510 ppm while it was 440 to 460 ppm in case of iCO₂. The present atmospheric CO₂ concentration in crop field is 380 ppm and it is increasing @ 1.5 ppm year⁻¹. Therefore, the CO₂ concentration was considered as 450 and 500 ppm, so that we may predict the effect of eCO₂ on crop in near future. Month wise average temperatures were taken. The pots were irrigated after transplanting of rice seedlings. A floodwater depth of 2–3 cm was maintained in each pot until a week before maturity of the crop. Intercultural operations were done uniformly in each pot to ensure normal growth of the crop. Weeding was done in the experimental pot as and when necessary. Regular irrigation was also done to maintain a saturated condition in pot.

Experimental measurement

The Pn, Tr and Gs were determined during full flowering. A portable Pn system (model: LICOR 6200, Lincoln, Nebraska) was used to record Pn, Tr and Gs. Plant height, number of effective tillers hill⁻¹, filled and unfilled grains panicle⁻¹, grain size, grain yield, shoot and root biomass were taken during harvesting. Plant height was taken from base of the plant to the top of the plant by using a meter scale. Shoot and root weight was recorded at maturity stage. Samples were collected and oven dried at 70 °C for 72 hours and dry weight of each hill was recorded separately. In each case, the mean of eight hills was calculated. The total grain of each hill was weight and recorded as grain yield hill⁻¹. The grain yield was adjusted to 14% moisture content using the following formula:

$$\text{Adjusted weight} = \frac{W \times (100 - M_1)}{(100 - M_2)} \times 100$$

where, W, M₁ and M₂ were fresh weight, fresh and adjusted moisture percent of the grain, respectively.

Grain amylose and protein content as were measured after harvesting of the crop and the concentration of grain N, Zn, Ca, Mg and Fe were determined. The samples were dried at 70 °C for 72 hrs and ground. The ground sample was digested in concentrated H₂PO₄ and total N concentration was determined by Kjeldahl method (Kjeldahl, 1883). The concentration of other nutrients, samples were taken and digested using mixture of HNO₃: HClO₄ (Bhargava & Raghupathi, 1993). The amylose content as determined by following the procedure as described by Cruz & Khush (2000). Protein content in rice samples were determined by macro Kjeldahl procedures.

Statistical analysis

Data gathered on different parameters were statistically analyzed using computer software package CropStat, version 7.2 (IRRI, 2007). Analysis of variance of the data was calculated and the significance of the factor (growing condition and timing of N application) was tested at the 5% level of probability. Treatment means were separated with Duncan's Multiple Range Test at 5% level of probability (Gomez & Gomez, 1984).

RESULTS AND DISCUSSION

Photosynthetic parameters

The combined effect of CO₂ and N application on Pn rate, Tr and Gs was statistically significant (Fig. 1). The Pn increased gradually from OF to eCO₂ in all N treatments. A significant reduction in Tr and Gs were observed in OTC as compared to OF in all N treatments. These two parameters were decreased gradually with the increasing of CO₂ concentration. The highest Tr was measured from OF with N3 treatment. On the contrary, the lowest Tr was recorded from eCO₂ with N1. The eCO₂ improved leaf Pn during flowering, while Tr and Gs declined in all N application treatments (Fig. 1). Nitrogen is one of the most important constituents of chlorophyll (Chl). Application of N at FT increased the concentration of N in leaf. The eCO₂ concentration increased leaf Chl also reported by De Costa et al. (2003) and Wang et al. (2014). Exogenous supply of CO₂ augmented its concentration inside rice leaf. As it is the substrate of Pn, it was expected that increasing CO₂ level in cellular level would increase Pn rate. Higher Pn at and iCO₂ and eCO₂ implies that rice plants had adopted a beneficial acclimation strategy for growth in CO₂ enhancement. Higher Pn rate of crops grown under eCO₂ have been well documented in earlier studies (Chen et al., 2014). Both Tr and Gs of rice plants at eCO₂ conditions were significantly lower than OF condition at FT of rice. At high CO₂ level, the Tr is reduced because of a direct effect of Gs. The lower Tr and Gs of rice under eCO₂ conditions could be beneficial for the growth of rice (Chunwu et al., 2016). The decreased Gs under eCO₂ was also reported by Wang et al. (2020) and Cai et al. (2018). Reduction in Gs might increase resistance to CO₂ diffusion into the leaf, thus partially offsetting the maximum stimulation of carboxylation rate.

Plant growth parameters

Plant height, shoot and root DM were significantly influenced by the interaction of CO₂ and N (Fig. 1). The tallest plant was measured in eCO₂ condition in all N application treatments. Significantly the shortest plant height was recorded from OF with all N treatments. Significantly the highest shoot DM production was recorded from eCO₂ with N1 (42.22 g plant⁻¹). The shoot DM obtained from N2 and N3 under eCO₂ were statistically similar. Though, shoot DM production under aCO₂ was numerically higher than OF, but they were not statistically different in all N treatments. The root DM of rice plant also increased gradually from OF to eCO₂ as well as N3 to N1 (Fig. 1). The highest root mass was recorded under eCO₂ with N1 (15.02 g plant⁻¹) which was followed by N2 (12.87 g plant⁻¹) and N3 (12.20 g plant⁻¹) under same environment. The lowest root mass was obtained from OF in all N treatments. The eCO₂ enhanced plant height, shoot and root DM of rice (Fig. 1). The result is in agreement with the findings of Haque et al. (2005). The eCO₂ increased the Pn rate of rice (Fig. 1) and the high Pn rate might have

contributed to production of taller plants under such condition. The shoot DM of the crop grown under eCO₂ increased as compared to that under OF (Satapathy et al., 2015).

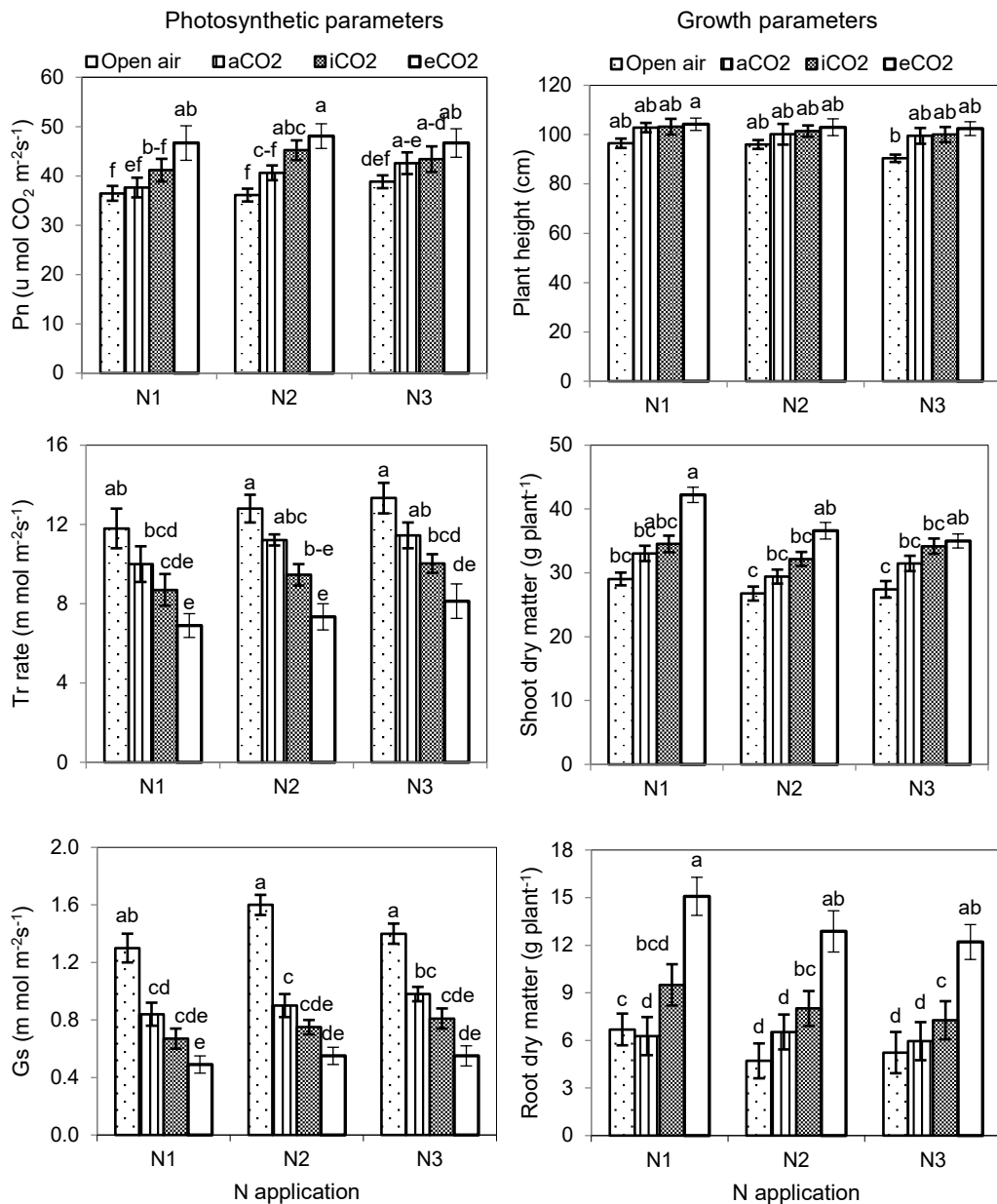


Figure 1. Photosynthetic and growth parameters of rice as affected by N fertilization under eCO₂ at FT. Bar graphs indicate mean value ± standard error and bars with similar letters did not vary significantly at 5% level of probability; eCO₂, iCO₂ and aCO₂ indicate 500, 450 and 400 ppm CO₂, respectively; N1 = 1/3rd of N at ET + 1/3rd at AT + 1/3rd before PI; N2 = 1/3rd of N at ET + 1/3rd before PI + 1/3rd at booting; and N3 = 1/3rd of N at ET + 1/3 N before PI + 1/3rd at flowering.

In this experiment, taller plants were measured under eCO₂ condition, thus the assimilated DM in plants under this condition enhanced the height of the plants. The eCO₂ induced increase in biomass was also observed in mungbean (Chowdhury et al., 2005) and rice (Razzaque et al., 2009). On the other hand, Pilipavicius et al. (2006) showed that eCO₂ (700 ppm) had detrimental effect on the early growth of *Chenopodium album*. However, they also showed that the increase of air temperature and CO₂ concentration had damaging tendency for above and below ground fresh and dry weight of *Chenopodium album*.

Yield characteristics

Production of panicle hill⁻¹ varied due to interaction of CO₂ concentration and timing of N application (Fig. 2). The highest panicles production was recorded at OF with N2 which was statistically identical with the panicle production at eCO₂ with N1 as well as at eCO₂ with N3. The sterility of rice spikelets varied statistically due to combined effect of CO₂ and N application. Rice grown in N1 treatment produced higher percentage of unfilled grains in all growing conditions and lowest in N3 treatment. In case of N3 treatment, production of unfilled grains were 20, 19, 14, and 20 under OF, aCO₂, iCO₂ and eCO₂, respectively. The highest number of filled grains were obtained from N2 (143) under eCO₂ condition which was statistically identical with N1 under iCO₂ and N3 under aCO₂ conditions (142). The production of total number of grains hill⁻¹ was the highest under eCO₂ with N1 (Table 1). However, the percent filled grain was the highest in iCO₂ with N3, where the spikelet sterility was the lowest. The grain size did not affect significantly due to the interaction of eCO₂ and N application timing. Weight of 1,000-grains were numerically higher under iCO₂ condition with N3 followed by treatment N1 for aCO₂. A lower weight of 1,000-grain was recorded in the conditions of aCO₂ and eCO₂ with N2 treatment. The grain yield of rice varied significantly due to interaction of time of N application under eCO₂. The highest rice yield was recorded under eCO₂ (66.47 g) with N1 which was statistically identical with N3 treatment under same environmental condition.

In N3 treatment, rice grown under eCO₂ produced 65.85 g grain plant⁻¹, which was 18, 14 and 20% higher than that of OF, aCO₂ and iCO₂ conditions, respectively. Similarly, in both N1 and N2 treatments, significantly higher grain yield also recoded from eCO₂ as compared to other environmental conditions. The highest grain under

Table 1. Effect of eCO₂ and N fertilization on grain production of rice

Growing conditions	N application	Total spikelets (no. hill ⁻¹)	Filled grains (%)	1,000-grain weight (g)
sOpen field	N1	3,490 ^{def}	74 ^e	20.31
	N2	3,991 ^{abc}	75 ^e	20.67
	N3	3,173 ^{fg}	85 ^{abc}	20.80
aCO ₂	N1	3,453 ^{def}	80 ^{cde}	20.88
	N2	3,570 ^{c-f}	83 ^{bcd}	19.31
	N3	3,375 ^{ef}	88 ^{ab}	19.45
iCO ₂	N1	3,633 ^{b-f}	80 ^{cde}	20.41
	N2	3,368 ^{ef}	86 ^{abc}	20.61
	N3	2,832 ^g	90 ^a	21.56
eCO ₂	N1	4,205 ^a	77 ^{de}	20.51
	N2	3,900 ^{a-d}	80 ^{cde}	19.31
	N3	3,726 ^{a-e}	87 ^{ab}	20.40
CV (%)		11.8	6.4	3.2

Figures with similar letters did not vary significantly at 5% level of probability; eCO₂, iCO₂ and aCO₂ indicate 500, 450 and 400 ppm CO₂, respectively; N1 = 1/3rd of N at ET + 1/3rd at AT + 1/3rd before PI; N2 = 1/3rd of N at ET + 1/3rd before PI + 1/3rd at booting; and N3 = 1/3rd of N at ET + 1/3 N before PI + 1/3rd at flowering.

eCO₂ with N3 treatment due to production maximum number of productive panicles and a greater number of grains panicle⁻¹. The lowest grain yield was obtained from iCO₂ with N3 (54.82 g). Rice yield is the product of number of panicles hill⁻¹, number of grains panicle⁻¹ and weight of individual grain. In this experiment, the production of bearing tillers increased at higher level of CO₂. Higher shoot DM was recorded from eCO₂; thus, it was expected to increase number of panicles from this treatment. In general, growth and yield of rice were expected to increase with CO₂ elevation. Previous experiments have demonstrated that rice produced higher number of tillers and panicle at eCO₂ condition (Yang et al., 2006). The main reason for the increases in yield with rising CO₂ were due mainly to the increased panicle number per unit area, as was also observed for rice grown in CO₂ enriched enclosures (Moya et al., 1998). Our results indicated that greater panicle number hill⁻¹ was clearly due to the increases in maximum shoot DM (tiller number) with eCO₂ (Fig. 2). Mamun et al. (2017) also found that the panicles production rice was also increased due to late application of N fertilizer. However, application of N before panicle application increased grain number panicle⁻¹ as reported by Abedin et al. (2015).

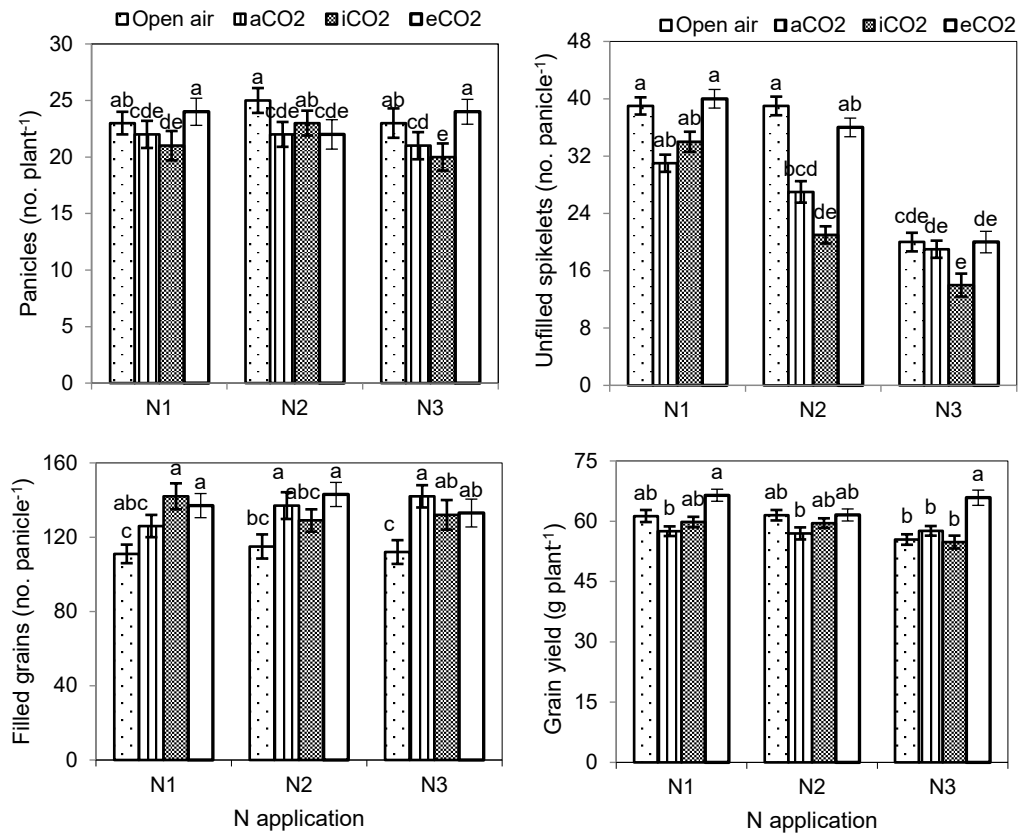


Figure 2. Panicles and grain production of rice as affected by N fertilization under eCO₂.

Bar graphs indicate mean value \pm standard error and bars with similar letters did not vary significantly at 5% level of probability; eCO₂, iCO₂ and aCO₂ indicate 500, 450 and 400 ppm CO₂, respectively; N1 = 1/3rd of N at ET + 1/3rd at AT + 1/3rd before PI; N2 = 1/3rd of N at ET + 1/3rd before PI + 1/3rd at booting; and N3 = 1/3rd of N at ET + 1/3 N before PI + 1/3rd at flowering.

Nutrient accumulation and grain quality of rice

Rice plants accumulated higher amount of N in grain at eCO₂ condition as compared to aCO₂, iCO₂ and OF (Table 2). Similarly, N₃ facilitated in absorption of more N in grain followed by N₂. For Zinc, rice plant uptake more Zn at eCO₂ as compared to iCO₂ and aCO₂. Similarly, N₃ facilitated in absorption of more Zn in grain followed by N₂. In case of Calcium, rice plant uptake more Ca at eCO₂ and N₃ facilitated in absorption of more Ca in grain followed by N₂. In case of Magnesium, rice plant uptake more Mg at eCO₂ and N₃ facilitated in absorption of more Mg in grain followed by N₂. In case of iron (Fe), rice plant uptake more Fe at eCO₂ and N₃ facilitated in absorption of more Fe in grain followed by N₁ (Table 2).

Table 2. Effect of eCO₂ and nitrogen application on nutrient accumulation in rice grain

Growing conditions	Nitrogen application	Nitrogen (%)	Zinc (ppm)	Calcium (%)	Magnesium (%)	Iron (ppm)
Open field	N1	1.08 ^h	12.15	0.09	0.28	71.66 ^f
	N2	1.35 ^{ef}	13.10	0.11	0.30	78.87 ^f
	N3	1.50 ^{cd}	13.48	0.10	0.29	74.96 ^f
aCO ₂	N1	1.29 ^{fg}	13.16	0.09	0.29	111.40 ^{cd}
	N2	1.39 ^e	14.18	0.10	0.31	100.43 ^e
	N3	1.43 ^d	14.52	0.11	0.30	104.94 ^{de}
iCO ₂	N1	1.25 ^g	14.17	0.11	0.30	120.24 ^{cd}
	N2	1.47 ^{cd}	15.32	0.11	0.31	124.28 ^{bc}
	N3	1.54 ^{bc}	15.56	0.12	0.32	119.93 ^{cd}
eCO ₂	N1	1.50 ^{cd}	16.33	0.11	0.33	142.22 ^a
	N2	1.60 ^{ab}	16.65	0.12	0.34	139.23 ^{ab}
	N3	1.72 ^a	17.63	0.13	0.36	149.92 ^a
CV (%)		8.6	9.8	8.7	6.9	12.3

Figures with similar letters in a column did not vary significantly at 5% level of probability; eCO₂, iCO₂ and aCO₂ indicate 500, 450 and 400 ppm CO₂, respectively; N₁ = 1/3rd of N at ET + 1/3rd at AT + 1/3rd before PI; N₂ = 1/3rd of N at ET + 1/3rd before PI + 1/3rd at booting; and N₃ = 1/3rd of N at ET + 1/3 N before PI + 1/3rd at flowering.

Rice grain quality was influenced by the interaction of various CO₂ concentrations and time of N application (Fig. 3). Amylose percentage was found higher at eCO₂ concentration i.e., iCO₂ with N₃ followed by eCO₂. The aCO₂ with N₂ showed the lowest percentage of amylose. Protein percentage was found lowest at eCO₂ with N₁ and the highest amount of protein percentage was found in OF condition with N₃. The plants grown under iCO₂ condition accumulated the highest amount of N in grain, while the lowest at eCO₂ in all N treatments. Under eCO₂ condition, enhanced carbohydrate accumulation and accelerated growth would dilute the nutrient concentration in plants (Chunwu et al., 2016). Similarly, lower Tr and Gs may limit the Tr-mass flow of nutrients, lowering the nutrient accumulation in shoot (McGrath & Lobell, 2013; Houshmandfar et al., 2015). The N₃ facilitated in absorption of more N, Zn, Ca, Mg and Fe in grain. Decreased N concentration under eCO₂ by 9.8% and 14.62% in leaf and 7.38% in roots as compared to ambient CO₂ and field condition (Razzaque et al., 2009). Similarly lower N concentration under eCO₂ has been reported in wheat and soybean (Pal et al., 2003). Under late application of N, higher grain N content was also found by Mamun et al. (2020). Amylose content slightly increased under both iCO₂ and eCO₂

condition, though protein percent was reduced. However, protein content at iCO_2 slightly increased with N_2 treatment. Moreover, the percentage of the both characters decreased under aCO_2 condition. Amylose content determines the cooking and eating quality of rice. Khanam et al. (2004) reported that eCO_2 adversely affect the amylose content of rice. Although eCO_2 causes a decrease in grain protein concentrations of rice, grain protein yield remains stable or even increases in future high CO_2 environment because of the significant increase in grain yield due to CO_2 enrichment with N_2 treatment (Yang et al., 2007). Under control condition, the grain yield of rice increased when N applied at PI stage of rice (Mamun et al., 2018).

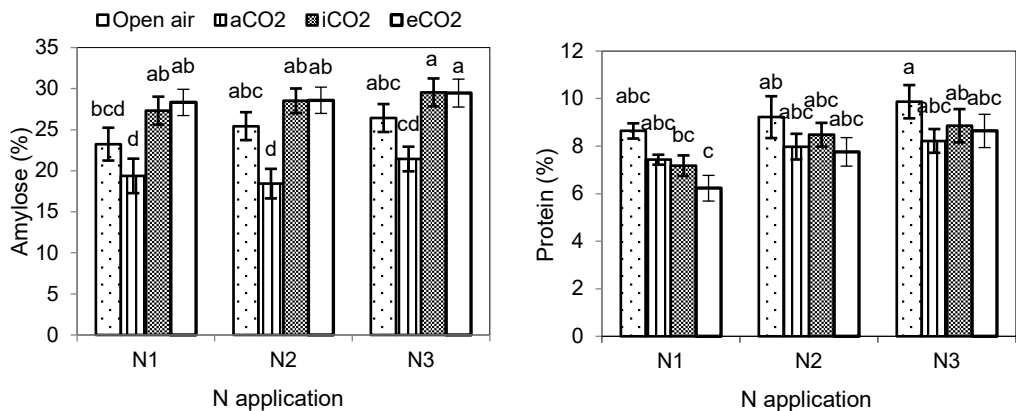


Figure 3. Grain quality properties of rice as affected by N fertilization under eCO_2 .

Bar graphs indicate mean value \pm standard error and bars with similar letters did not vary significantly at 5% level of probability; eCO_2 , iCO_2 and aCO_2 indicate 500, 450 and 400 ppm CO_2 , respectively; $N_1 = 1/3^{rd}$ of N at ET + $1/3^{rd}$ at AT + $1/3^{rd}$ before PI; $N_2 = 1/3^{rd}$ of N at ET + $1/3^{rd}$ before PI + $1/3^{rd}$ at booting; and $N_3 = 1/3^{rd}$ of N at ET + $1/3$ N before PI + $1/3^{rd}$ at flowering.

CONCLUSIONS

The results of this study revealed that the eCO_2 (450 to 500 ppm) with N application up to flowering stage favored Pn, shoot and root biomass production. Rice produced higher number of panicle and grains, gave better grain size and accumulated greater amount of nutrient in plants under eCO_2 with application of N up to flowering stage, resulting higher grain yield. Therefore, under future elevated eCO_2 conditions, rescheduling of N for rice is need. In addition, the amount of N supplied after initiation stage should be sufficient to maintain the extra dry matter produced under the eCO_2 conditions.

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