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

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Abstract. The atmospheric carbon dioxide (CO_2) 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 CO_2 (eCO_2) 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 eCO_2 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 eCO_2. Rice plant absorbed higher amount of Zn, Ca, Mg, and Fe at eCO_2 when N was applied up to FT. Amylose was higher but protein percentage was lower at eCO_2. These results indicate that to maximize rice yield under eCO_2, 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 CO_2 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 C_3 crops respond more strongly to the eCO_2 than C_4 crop (Jablonski et al., 2002). The photosynthesis (Pn) of crops increased significantly under eCO_2 resulting increase in biomass and yield of C_3 plants (Chunwu et al., 2016). On the other hand, stomatal conductance (Gs) and transpiration rate (Tr) of plants reduced under eCO_2 condition (Chunwu et al., 2016). Rice (*Oryza sativa* L.) is one of the most important crops in Asia (FAOSTAT, 2017). As a C_3 plant, growth and yield of rice are markedly enhanced by eCO_2 (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 eCO_2 (Yang et al. 2006; Sasaki et al. 2007; Satapathy et al., 2015). Increased growth of the crops under eCO_2 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 eCO_2 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 eCO_2. 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 eCO_2 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 eCO_2 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 eCO_2 condition (Razzaque et al., 2009). However, Pilipavicius et al. (2006) showed a negative effect of higher CO_2 concentration on the early growth of *Chenopodium album*. In this situation, this study was conducted to determine the effect of eCO_2 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 m^2) were constructed with iron frame that installed on the ground according to Uprety (1998). For growing rice plants, 96 pots (size: 24 cmin diameter and 27 cmin 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, $ZnSO_4$ 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 CO_2) (eCO_2), ii. OTC with 450 ppm CO_2 (intermediate CO_2 concentration, iCO_2), iii. OTC with ambient CO_2 (400 ppm CO_2) (aCO_2) and iv. Open field (OF, 380 ppm CO_2). 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 CO_2 gas was supplied to the OTC chambers from CO_2 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 CO_2 concentration inside the OTC regularly. In eCO_2 treatment, the CO_2 concentration fluctuated from 490 to 510 ppm while it was 440 to 460 ppm in case of iCO_2. The present atmospheric CO_2 concentration in crop field is 380 ppm and it is increasing @ 1.5 ppm year^{-1}. Therefore, the CO_2 concentration was considered as 450 and 500 ppm, so that the we may predict the effect of eCO_2 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, M_1 and M_2 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 H_2PO_4 and total N concentration was determined by Kjeldahl method (Kjeldahl, 1883). The concentration of other nutrients, samples were taken and digested using mixture of HNO_3: $HClO_4$ (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 CO_2 and N application on Pn rate, Tr and Gs was statistically significant (Fig. 1). The Pn increased gradually from OF to eCO_2 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 CO_2 concentration. The highest Tr was measured from OF with N3 treatment. On the contrary, the lowest Tr was recorded from eCO_2 with N1. The eCO_2 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 eCO_2 concentration increased leaf Chl also reported by De Costa et al. (2003) and Wang et al. (2014). Exogenous supply of CO_2 augmented its concentration inside rice leaf. As it is the substrate of Pn, it was expected that increasing CO_2 level in cellular level would increase Pn rate. Higher Pn at and iCO_2 and eCO_2 implies that rice plants had adopted a beneficial acclimation strategy for growth in CO_2 enhancement. Higher Pn rate of crops grown under eCO_2 have been well documented in earlier studies (Chen et al., 2014). Both Tr and Gs of rice plants at eCO_2 conditions were significantly lower than OF condition at FT of rice. At high CO_2 level, the Tr is reduced because of a direct effect of Gs. The lower Tr and Gs of rice under eCO_2 conditions could be beneficial for the growth of rice (Chunwu et al., 2016). The decreased Gs under eCO_2 was also reported by Wang et al. (2020) and Cai et al. (2018). Reduction in Gs might increase resistance to CO_2 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 CO_2 and N (Fig. 1). The tallest plant was measured in eCO_2 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 eCO_2 with N1 (42.22 g plant^{-1}). The shoot DM obtained from N2 and N3 under eCO_2 were statistically similar. Though, shoot DM production under aCO_2 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 eCO_2 as well as N3 to N1 (Fig. 1). The highest root mass was recorded under eCO_2 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 eCO_2 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 eCO_2 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 eCO_2 increased as compared to that under OF (Satapathy et al., 2015).

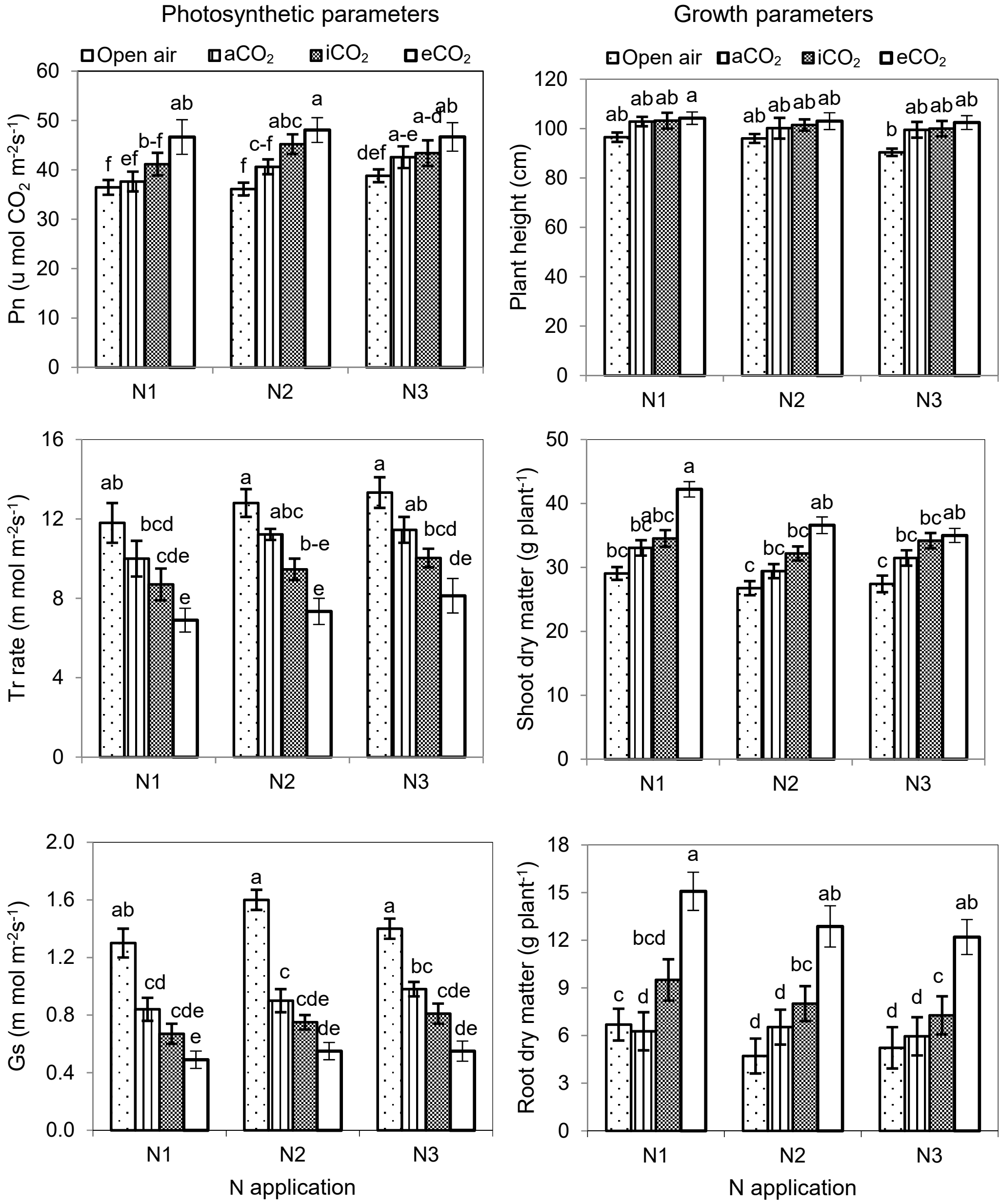

Figure 1. Photosynthetic and growth parameters of rice as affected by N fertilization under eCO_2 at FT. Bar graphs indicate mean value ± standard error and bars with similar letters did not vary significantly at 5% level of probability; eCO_2, iCO_2 and aCO_2 indicate 500, 450 and 400 ppm CO_2, respectively; 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 eCO_2 condition, thus the assimilated DM in plants under this condition enhanced the height of the plants. The eCO_2 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 eCO_2 (700 ppm) had detrimental effect on the early growth of *Chenopodium album*. However, they also showed that the increase of air temperature and CO_2 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 CO_2 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 eCO_2 with N1 as well as at eCO_2 with N3. The sterility of rice spikelets varied statistically due to combined effect of CO_2 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, aCO_2, iCO_2 and eCO_2, respectively. The highest number of filled grains were obtained from N2 (143) under eCO_2 condition which was statistically identical with N1 under iCO_2 and N3 under aCO_2 conditions (142). The production of total number of grains hill^{-1} was the highest under eCO_2 with N1 (Table 1). However, the percent filled grain was the highest in iCO_2 with N3, where the spikelet sterility was the lowest. The grain size did not affect significantly due to the interaction of eCO_2 and N application timing. Weight of 1,000-grains were numerically higher under iCO_2 condition with N3 followed by treatment N1 for aCO_2. A lower weight of 1,000-grain was recorded in the conditions of aCO_2 and eCO_2 with N2 treatment. The grain yield of rice varied significantly due to interaction of time of N application under eCO_2. The highest rice yield was recorded under eCO_2 (66.47 g) with N1 which was statistically identical with N3 treatment under same environmental condition.

Table 1. Effect of eCO_2 and N fertilization on grain production of rice

Growing conditions	N application	Total spikelets (no. hill^{-1})	Filled grains (%)	1,000-grain weight
Open 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$^{c\text{-}f}$	83^{bcd}	19.31
	N3	3,375^{ef}	88^{ab}	19.45
iCO_2	N1	3,633$^{b\text{-}f}$	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}	77de	20.51
	N2	3,900$^{a\text{-}d}$	80^{cde}	19.31
	N3	3,726$^{a\text{-}e}$	87^{ab}	20.40
CV (%)		11.8	6.4	3.2

Figures with similar letters did not vary significantly at 5% level of probability; eCO_2, iCO_2 and aCO_2 indicate 500, 450 and 400 ppm CO_2, 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 N3 treatment, rice grown under eCO_2 produced 65.85 g grain plant^{-1}, which was 18, 14 and 20% higher than that of OF, aCO_2 and iCO_2 conditions, respectively. Similarly, in both N1 and N2 treatments, significantly higher grain yield also recoded from eCO_2 as compared to other environmental conditions. The highest grain under

eCO_2 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 iCO_2 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 CO_2. Higher shoot DM was recorded from eCO_2; thus, it was expected to increase number of panicles from this treatment. In general, growth and yield of rice were expected to increase with CO_2 elevation. Previous experiments have demonstrated that rice produced higher number of tillers and panicle at eCO_2 condition (Yang et al., 2006). The main reason for the increases in yield with rising CO_2 were due mainly to the increased panicle number per unit area, as was also observed for rice grown in CO_2 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 eCO_2 (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).

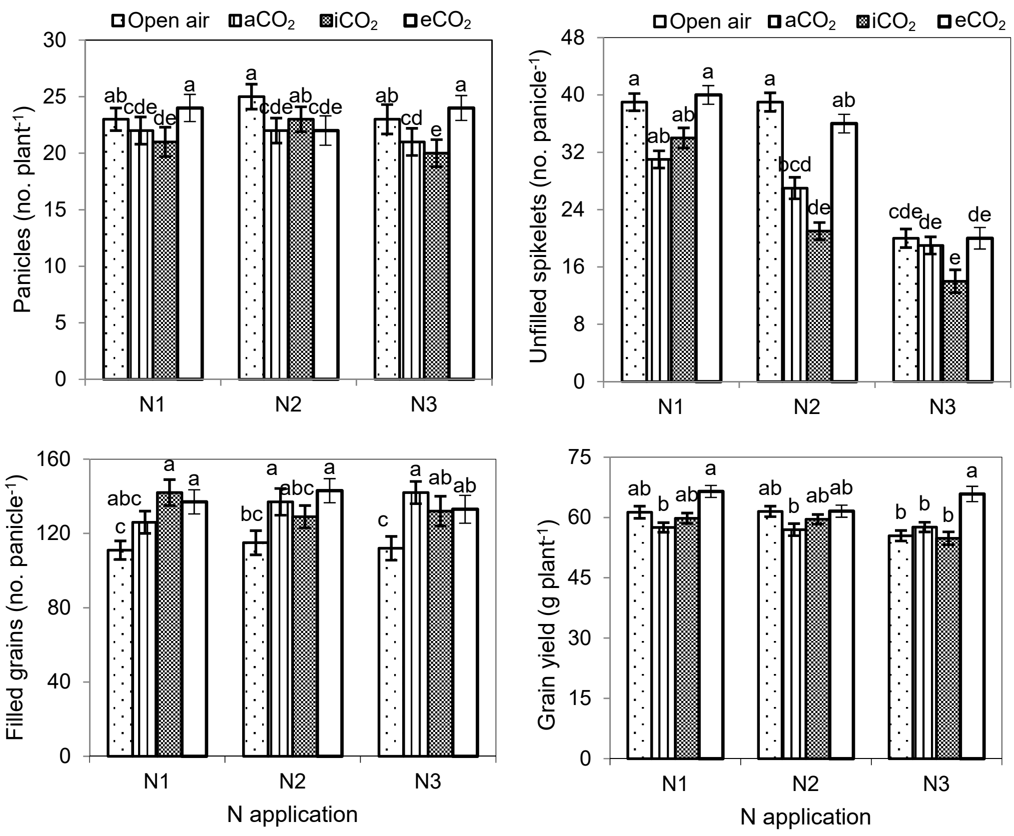

Figure 2. Panicles and grain production of rice as affected by N fertilization under eCO_2.

Bar graphs indicate mean value ± 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; 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 eCO_2 condition as compared to aCO_2, iCO_2 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 eCO_2 as compared to iCO_2 and aCO_2. Similarly, N3 facilitated in absorption of more Zn in grain followed by N2. In case of Calcium, rice plant uptake more Ca at eCO_2 and N3 facilitated in absorption of more Ca in grain followed by N2. In case of Magnesium, rice plant uptake more Mg at eCO_2 and N3 facilitated in absorption of more Mg in grain followed by N2. In case of iron (Fe), rice plant uptake more Fe at eCO_2 and N3 facilitated in absorption of more Fe in grain followed by N1 (Table 2).

Table 2. Effect of eCO_2 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^{cde}
	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; eCO_2, iCO_2 and aCO_2 indicate 500, 450 and 400 ppm CO_2, 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 CO_2 concentrations and time of N application (Fig. 3). Amylose percentage was found higher at eCO_2 concentration i.e., iCO_2 with N3 followed by eCO_2. The aCO_2 with N2 showed the lowest percentage of amylose. Protein percentage was found lowest at eCO_2 with N1 and the highest amount of protein percentage was found in OF condition with N3. The pants grown under iCO_2 condition accumulated the highest amount of N in grain, while the lowest at eCO_2 in all N treatments. Under eCO_2 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 eCO_2 by 9.8% and 14.62% in leaf and 7.38% in roots as compared to ambient CO_2 and field condition (Razzaque et al., 2009). Similarly lower N concentration under eCO_2 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 iCO_2 and eCO_2

condition, though protein percent was reduced. However, protein content at iCO_2 slightly increased with N2 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 N2 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).

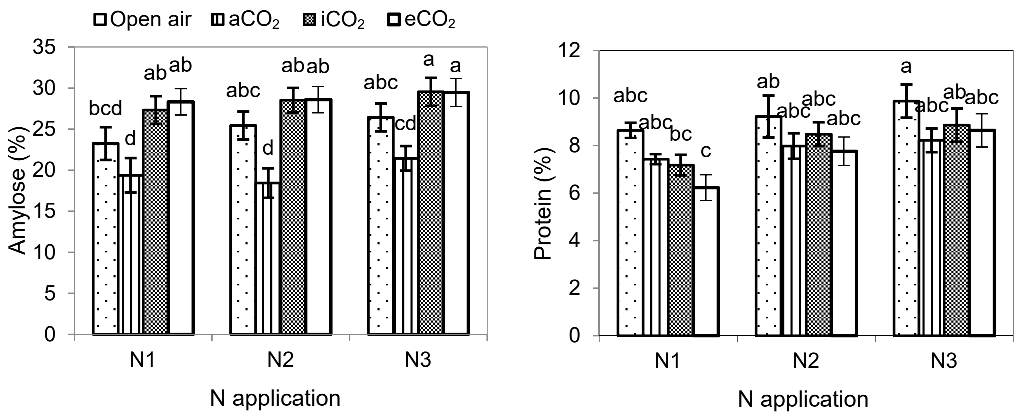

Figure 3. Grain quality properties of rice as affected by N fertilization under eCO_2.

Bar graphs indicate mean value ± 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; 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.

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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