



The Effect of Different Nitrogen Levels and Enrichment CO₂ on the Nutrient Contents of Rice Cultivars

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Abstract

An experiment was conducted during the July -December of 2003 to determine the nutrient compositions of rice under CO₂ enrichment of different levels of nitrogen supply. Rice plants were grown from seedlings to maturity inside open top chamber under elevated CO₂ (570 ±50) ppm, ambient CO₂ (~360ppm) and open field condition. Leaves and root were analyzed for C, N, Zn and Mg. C content was higher in the all plant parts of rice grown at elevated CO₂ compare than ambient CO₂ and field grown rice. Increased N supplies also increase C content of the plants. Nitrogen concentration was reduced in elevated CO₂ compare than other grown condition. Modern variety (BRRI dhan 39) contained higher C than local cultivars (Khaskani and Shakkorkhora). Nitrogen concentration was decreased under elevated CO₂ compare to other treatments.

Key words : Rice cultivars, Enrichment CO₂, C, N, Zn, Mg

Introduction

The present increase of atmospheric CO₂ concentration is predicts to continue the century due to human activities. Increased CO₂ has been observed to affect chemical composition of plant Materials (Lincoln, *et al.*1993). In particular within the plant allocation of carbon (C) and nitrogen (N) may be modified by the greater C availability (Ceulemans and Mousseau, 1994) including increased C/N ratio (McGuire *et al.*, 1995 and Penuelas and Estiarte., 1997). Interaction between elevated CO₂ and different nitrogen doses showed that high CO₂ could increase the rubisco efficiency and cause mobilization of nitrogen for their growth and development (Theobald *et al.* 1998). Increased rubisco efficiency under elevated CO₂ may require less amount of N for optimum biomass production (Sage *et al.*, 1988). Increased atmospheric CO₂ are affected by the plant nutrient in the biosphere. C₃ plant has not only promoted growth but also consequence for the demand of other nutrients under enrichment CO₂ (Theobald *et al.* 1998). Continuous increase in atmospheric CO₂ leads to significant effect on carbon-nitrogen dynamics in plants, which results abundant in carbon and scarce in nitrogen. This is caused by dilution affect due to higher carbohydrate concentration as well as a decreased demand for nitrogen in green tissues. Therefore the experiment was under taken with different levels of nitrogen fertilizer on partitioning of essential nutrients in rice and the effect of nutritional balance was modified in plants grown under different CO₂ concentrations.

Materials and Methods

The pot experiment was conducted at Bangabandhu Sheikh Mujibur Rahman Agricultural University, Gazipur, Bangladesh during rainy season of 2003. Soil physical and chemical properties were presented in Table V. The experiment was conducted in complete randomized block design (CRD). Three rice varieties were grown with three levels of nitrogen fertilizer under three CO₂ conditions. The rice varieties were BRRI dhan 39 (modern), Khaskani (local) and Shakkorkhora (local). Three levels of nitrogen fertilizer were optimum dose, supra optimum dose and control (no nitrogen). Optimum dose for modern variety was 90 kg N ha⁻¹ and for local 60 kg N ha⁻¹. Supra optimum dose for modern and local variety was 135 kg N ha⁻¹ and 90 kg N ha⁻¹ respectively which was 1.5 time higher than their optimum dose. Three growing conditions with CO₂ were elevated CO₂, ambient CO₂ and open field. Crop under the 'elevated CO₂' was grown in open top chamber (OTC) at a CO₂ concentration of 570 ± 50 ppm, while the 'ambient CO₂' treatment was maintained at the ambient CO₂ concentration of ~360 ppm in OTC. The 'open' treatment contained crop grown under open field condition at ambient CO₂ concentration. It is made of an iron frame of 3m in diameter and 3m in height. It was installed on the ground and covered with transparent polyvinyl chloride sheet. The top of the chamber was open to ensure near natural conditions. The CO₂ gas was supplied to

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the chamber from gas cylinder using a manifold gas regulator, pressure gauge and underground pipeline for using natural air with the help of a blower. The blower 30cm diameter thoroughly mixes with the supplied CO₂ gas with atmospheric air and blew to the chamber. The rice plants were grown in plastic pots containing approximately 12kg clayed soil. The treatments were replicated thrice and each pot had one seedling. Thirty - days old seedling in each variety were planted on August 2, 2003. Except N a fertilizer doses of 20 kg P, 60 kg K 20 kg S and 3.5 kg Zn per ha was applied (source of NPKSZn from urea, triple super phosphate, muriate of potash, Gypsum and Zinc sulphate) prior to transplanting, while N was applied in three installment at the time of 4, 21 and 52 days after transplanting. The source of N from urea. Several cultural practice were done as and when necessary. Standing water of 2 cm above the soil was maintained until the crops attained hard dough stage. The concentration of CO₂ in the chamber was monitored using infrared gas analyzer (Model LI 6200, Licon, Lincoln, USA). BRRIdhan 39 harvested 16 November, Khaskani Harvested 30 December and Shakkorkhora harvested 15 December of 2003. After harvesting root of the plants was removed by washing the soil of the pots without disrupting the small branches of the roots. Each plant was separated into leaves, stem, roots and yield and dried at 70°C for 72 hours. Dry mass of plants parts was recorded by electronic balance (AND, Counting, FX-300).

Total N determination

Total nitrogen contents were determined by modified Kjeldahl digestion (colorimetric) method. For determination of total nitrogen, oven dried plant parts were ground and samples were taken in Kjeldahl digestion flasks and digestion catalyst added in the sample. Salicylic- H₂SO₄ was added slowly. Samples were digested in gas burner more than 2 hours. 1 ml of digested sample was taken in 50 ml volumetric flask and 4 ml of H₂O was added. 1 ml of solution of EDTA, methyl red and ethanol, 7-10 ml (amount depends on titration where colour changes from pink to yellow) of solution of 0.3 N NaOH, 5 ml of solution of phenol and sodium

nitroprusside, 5 ml of solution of disodium hydrogen phosphate, trisodium phosphate and sodium hypochlorite were added. The volume was made up to 50 ml with H₂O. The flasks were shaken vigorously. After 2 hours absorbance was measured at 625 nm in a double beam spectrophotometer (Hitachi model 200-20).

Carbon estimation

Total carbon content was determined by wet digestion following the modified Walkley -Black method (Walkley and Black, 1934). The dried sample (3 mg) was oxidized with a mixture of potassium dichromate and concentrated sulphuric acid using the heat of dilution of acid. The unused potassium dichromate was estimate by back titration with ferrous ammonium sulphate.

Zn and Mg estimation

Samples were taken and digested using mixture of HNO₃ and HClO₄ (9:4) according to Bhargava and Raghupathi (1993). The digested material was used for the estimation of Mg and Zn and were determined in an atomic absorption spectrophotometer (Hitachi 170 -30, Japan). For estimation of nutrient plant samples were mixed together and then required amount was weighed in triplicate. The data were analyzed by partitioning the total variance with the help of computer using MSTATC program. The treatment means computed using Duncans Multiple Range test (DMRT).

Result and Discussion

The carbon content was higher in plants at elevated CO₂ compare to ambient CO₂ and field grown rice. The maximum carbon content was recorded in leaves at elevated CO₂ and lowest in grain in the field grown rice (Table Ia and Ib). Elevated CO₂ increased leaf carbon content by 36.14 % and 33.03% and root carbon content by 5.84% and 4.71% respectively over ambient CO₂ and field grown rice. This result agreed with Pal *et al.* (2003) in roots, stem and leaves of wheat in response to elevated CO₂. Jitla *et al.* (1997) also

Table I a. Effects of elevated CO₂ and nitrogen levels of leaf carbon content (%) of rice cultivars

Variety	Nitrogen levels	Elevated CO ₂	Ambient CO ₂	Field condition
BRRIdhan 39	Control	44.75 cA	38.25 dCe	40.75 cB
	Optimum	52.00 bA	45.25 bC	46.75 bB
	Supra optimum	54.75 aA	48.50 aB	48.00 aB
Khaskani	Control	41.50 eA	37.50 Be	38.50 dB
	Optimum	42.75 dA	39.50 dB	40.00 cdB
	Supra optimum	46.00 cA	41.00 cdB	41.70 cB
Shakkorkhora	Control	41.25 eA	39.00 dB	39.50 dB
	Optimum	43.50 cdA	41.75 cB	42.50 cB
	Supra optimum	45.25 cA	42.75 cC	44.25 bcB

Means followed by same small letter (column) and capital letter (row) did not differ significantly

reported similar result in *Lolium perenne*. Interaction between elevated CO₂ and different nitrogen supplies showed greater carbon availability in plant organs, including increased carbon-nitrogen ratio. Increased C/N ratio also increased photosynthesis and decreased respiration. Several authors confirmed this finding (Pal, *et al.*, 2003, Jitla *et al.* 1997, Tognetti, and Penuelas 2003).

Lower nitrogen content in leaves, and roots were recorded when rice plant were grown under elevated CO₂ compared to ambient CO₂ and field grown rice. Maximum reduction of N in all the plant parts occurred at no nitrogen supply (Table II a, IIb). When N supply increased, N concentration also increased all the plant parts of all the growing condition. Decreased N concentration under elevated CO₂ by 9.5% and

Table I b. Effects of elevated CO₂ and nitrogen levels of root carbon content (%) of rice cultivars

Variety	Nitrogen levels	Elevated CO ₂	Ambient CO ₂	Field condition
BRRIdhan 39	Control	39.90 dA	36.70 cB	37.22 dB
	Optimum	43.91 bcA	41.24 bB	40.75 cB
	Supra optimum	46.92 aA	43.49 aB	41.84 bcC
Khaskani	Control	36.56 dB	35.62 cB	38.36 cdA
	Optimum	41.85 cA	40.75 bB	40.51cB
	Supra optimum	44.67 abA	41.21 bB	42.36 bB
Shakkorkhora	Control	38.94 dA	36.31 cB	36.86 cdB
	Optimum	43.45 bcA	41.19 bB	43.75 abA
	Supra optimum	45.46 abA	42.16 abB	44.84 abA

Means followed by same small letter (column) and capital letter (row) did not differ significantly

The result also revealed that accumulation of more carbohydrate under elevated CO₂ helps in development of more new sinks and acquisition of other nutrients to sustain greater plant growth and productivity. The acquisition of carbon by different organs of rice plant varied among the varieties over the nitrogen levels. The modern variety BRRIdhan 39 contained highest carbon in leaves (54.75%), and roots (35.62%) under elevated CO₂ and high nitrogen level. Local varieties of rice contained lower proportion of carbon in different organs, which reduced further at field and lower nitrogen condition. As modern rice variety is more responsive to applied nitrogen, acquisition of more nitrogen under supra optimum nitrogen treatment might favor to increase carbon content of modern rice at elevated CO₂ concentration (Pal, *et al.*, 2003). Among the variety and nitrogen interaction modern variety (BRRIdhan 39) contained higher C compare than local cultivars.

14.62% in leaf and 7.38% and 6.83% in roots as compared to ambient CO₂ and field grown rice. Similarly lower N concentration under elevated CO₂ has been reported in wheat and soybean (Pal *et al.*, 2003, Hocking and Meyer 1991, Reeves *et al.* 1994). However reduction in nitrogen concentration under elevated CO₂ was less when N supply in soil has been increased. Rogers *et al.* (1996) have been reported that high CO₂ induced reduction in leaf N concentration disappeared when N supply was enhanced in cotton (Rogers *et al.* 1996).

Plant tissue increased the C/N ratio when increased C and decreased N concentration. Monge and Bugbee reported similar result in wheat at elevated CO₂ (Monge and Bugbee. 1998). The reduction in tissue nitrogen concentration might be due to the accumulation of more carbohydrates under

Table II a. Effects of elevated CO₂ and nitrogen levels of leaf nitrogen content (%) of rice cultivars

Variety	Nitrogen levels	Elevated CO ₂	Ambient CO ₂	Field condition
BRRIdhan 39	Control	0.76 eB	0.96 bcA	0.94 dA
	Optimum	0.92 bcB	0.98 bB	1.05 bcA
	Supra optimum	0.97 aB	0.98 bB	1.11 abA
Khaskani	Control	0.85 dB	0.87 dA	0.85 eB
	Optimum	0.90 cB	0.97 bA	0.93 dB
	Supra optimum	0.93 bB	1.02 bA	1.07 bcA
Shakkorkhora	Control	0.81dB	0.94 bcA	0.95 dA
	Optimum	0.91 cC	0.97 bB	1.13 aA
	Supra optimum	0.95 bC	1.07 aB	1.14 aA

Means followed by same small letter (column) and capital letter (row) did not differ significantly

Table IIb. Effects of elevated CO₂ and nitrogen levels of root nitrogen content (%) of rice cultivars

Variety	Nitrogen levels	Elevated CO ₂	Ambient CO ₂	Field condition
BRRIdhan 39	Control	0.53 eB	0.61cdA	0.61cdA
	Optimum	0.60 cB	0.63 cdA	0.63 cA
	Supra optimum	0.66 abB	0.71abA	0.71 abA
Khaskani	Control	0.57 dB	0.62 cdA	0.61 cdA
	Optimum	0.67 abB	0.70 bA	0.68 bB
	Supra optimum	0.68 aB	0.73 aA	0.72 aA
Shakkorkhora	Control	0.53 eB	0.56 eA	0.56 eA
	Optimum	0.56 dB	0.60 dA	0.61 cdA
	Supra optimum	0.62 bB	0.66 cA	0.66 cA

Means followed by same small letter (column) and capital letter (row) did not differ significantly

high CO₂ concentration (Wong, 1990). Conroy and Hocking(1993) also reported the reduced nitrogen concentration under CO₂ enrichment is optimization of the photosynthetic apparatus under high CO₂ by which less nitrogen is invested in rubisco during photosynthesis. Elevated CO₂ causes increased growth per unit uptake of other nutrients, which dilutes the nitrogen and concentration in plant tissues. Increased nitrogen also increased tissue nitrogen concentra-

tion although it was lower in modern variety grown under elevated CO₂ conditions. Accelerated growth of modern rice variety (BRRIdhan 39) under elevated CO₂ and lower N content in different plant organs of rice might be maximum N was invested in photosynthesis. This result agreed with Uprety and Mahalaxmi (2000) in mustard plant. The elevated CO₂ also affect the content of other macro and micronutrients like Mg and Zn in leaves and root of rice.

Table III a. Effects of elevated CO₂ and nitrogen levels of leaf zinc content (%) of rice cultivars

Variety	Nitrogen levels	Elevated CO ₂	Ambient CO ₂	Field condition
BRRIdhan 39	Control	29 dA	30 deA	27 eB
	Optimum	33 bcA	29 eB	33 bcA
	Supra optimum	36 aA	33 abB	36 aA
Khaskani	Control	27 dA	25 eB	27 eA
	Optimum	34 bA	31 cdC	33 bcB
	Supra optimum	35 bA	34 aB	33 bC
Shakkorkhora	Control	28 cdA	26 eB	25 eB
	Optimum	33 bcA	32 bcAB	30 deB
	Supra optimum	36 aA	34 aB	34 bB

Means followed by same small letter (column) and capital letter (row) did not differ significantly

Table III b. Effects of elevated CO₂ and nitrogen levels of root zinc content (%) of rice cultivars

Variety	Nitrogen levels	Elevated CO ₂	Ambient CO ₂	Field condition
BRRIdhan 39	Control	47 bcA	43 cdB	43 cdB
	Optimum	47 bcA	44 cB	45 cB
	Supra optimum	55 aA	50 abB	52 aB
Khaskani	Control	42 dA	40 dB	41 dA
	Optimum	46 cA	45 cB	45 cB
	Supra optimum	49 bcA	42 cC	44 cB
Shakkorkhora	Control	50 bA	48 bB	47 bcB
	Optimum	55 aA	50 abB	49 bB
	Supra optimum	55 aA	51 aB	50a bB

Means followed by same small letter (column) and capital letter (row) did not differ significantly

Table IV a. Effects of elevated CO₂ and nitrogen levels of leaf magnesium content (%) of rice cultivars

Variety	Nitrogen levels	Elevated CO ₂	Ambient CO ₂	Field condition
BRRI dhan39	Control	0.21 abA	0.22 abA	0.20 abB
	Optimum	0.23 abA	0.22 abA	0.21 abB
	Supra optimum	0.23 abA	0.23 abA	0.22 abB
Khaskani	Control	0.21abB	0.22 abA	0.20 abB
	Optimum	0.21 abB	0.22 abA	0.23 abA
	Supra optimum	0.24 aA	0.24 aA	0.24 aA
Shakkorkhora	Control	0.20 abB	0.22 abA	0.21 abB
	Optimum	0.21 abB	0.22 abA	0.20 abB
	Supra optimum	0.22 abB	0.21abB	0.23 abA

Means followed by same small letter (column) and capital letter (row) did not differ significantly

Table IV b. Effects of elevated CO₂ and nitrogen levels of root magnesium content (%) of rice cultivars

Variety	Nitrogen levels	Elevated CO ₂	Ambient CO ₂	Field condition
BRRI dhan 39	Control	0.32 deA	0.28 cdB	0.22 dC
	Optimum	0.35 cdA	0.28 cdC	0.30 cB
	Supra optimum	0.36 bcA	0.30 cdB	0.35 aA
Khaskani	Control	0.35 cdA	0.31 cdB	0.32 bB
	Optimum	0.39 bcA	0.35 cB	0.32 bC
	Supra optimum	0.40 abA	0.32 cdB	0.32 bB
Shakkorkhora	Control	0.32 deA	0.30 cdB	0.30 cB
	Optimum	0.40 abA	0.37 bB	0.35 aB
	Supra optimum	0.42 aA	0.39 aB	0.35 aC

Means followed by same small letter (column) and capital letter (row) did not differ significantly

Table V. Physical and chemical properties of soil

Properties	Values
Textural class	Silty clay
Sand (%)	26
Silt (%)	30
Clay (%)	33
pH	6.5
Organic matter (%)	0.76
Exchangeable bases (%) Mg	0.54
Exchangeable bases (%) K	0.10
Available nutrient (μ /ml) : NH ₄ + -N	25.00
Available nutrient (μ /ml) : P	0.09
Available nutrient (μ /ml) : S	7.00
Available nutrient (μ /ml) : Zn	1.00

Increased N supply increased Zn content in leaf but no significant variation among the growing condition. (Table IIIa). In case of root elevated CO₂ increased Zn content simultaneously increased nitrogen supply. This result agreed with Pal *et al.* (2003) in wheat. He observed that when nitrogen supply increased root Zn content also increased in wheat.

No significant variation in Mg content was observed in leaf under different CO₂ and nitrogen treatments (Table IV a). Baxter *et al.* (1994) have also reported similar increase in P, Mg and K content in three grass species under elevated CO₂. Root Mg content was higher in elevated CO₂ than other treatments. No significant variation was found with increased N supply.

Conclusion

Elevated CO₂ increased carbon content but decreased nitrogen content in all the rice cultivars. Nitrogen was used photosynthesis as a result low nitrogen remains in rice plants. Higher nitrogen increased magnesium content in leaf and root which enhanced plant growth.

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