

Impact of Elevated Atmospheric Carbon Dioxide on Yield, Vitamin C, Proximate, Fatty Acid and Amino Acid Composition of Capsicum (*Capsicum Annuum*)

Andaleeb Azam^{1*}, Abdul Hameed², Ikhtiar Khan³

¹Department of Chemistry, Women University Swabi, Swabi, KP, Pakistan

²National Center for Nanoscience and Technology, Haidian, Beijing, China

³Institute of Chemical Sciences, University of Peshawar, Pakistan

*Email: andaleebazam1@gmail.com

Abstract. A steady increase in the atmospheric CO₂ concentration due to human activities in the last few decades is largely believed to be a major cause of climate change. Since agriculture is a climate sensitive system, there is a growing concern that the CO₂ added to the air is changing the nutritional composition of fruits, grains and vegetables. Based on this hypothesis, the present experiment work was conducted to study the effect of enhanced atmospheric CO₂ on nutritional and biochemical composition of chili (*Capsicum annuum*). Five local varieties of capsicum (Angrika, Best Choice, Diana, Magama, and 99PE-1689) were grown under two concentrations of CO₂, i.e. 400 μmol mol⁻¹ (ambient) and 1000 μmol mol⁻¹ (elevated) under controlled conditions in green houses. Fruits were harvested at two stages of maturity and analyzed for proximate, elemental, fatty acid and amino acid concentration. Capsicum fruits grown under higher CO₂ concentration had different nutritional composition. Vitamin C, protein, ash and fat contents decreased significantly ($P \leq 0.05$), acidity decreased non-significantly ($P > 0.1$) whereas sugar and fiber contents increased significantly under enhanced CO₂. Elemental composition showed a significant increase in C, H, Fe, Mn and a decrease in N, Ca, Mg and Zn contents. Few elements (S, K and Cu) showed no significant trend with elevated CO₂. Fatty acids, with few exceptions, were not much affected by CO₂ enrichment. Amino acids decreased with elevated concentration of CO₂. The effect of enhanced CO₂ was more pronounced at the fully matured (red) stage as compared to the pre-mature (green) stage of capsicum fruit. Enhanced atmospheric CO₂ lowered the nutritional quality of capsicum fruit by decreasing its vitamin C, proteins, Ca, Mg and Zn contents.

Keywords: Atmospheric CO₂, capsicum, nutritional quality, elemental composition.

1 Introduction

Industrial revolution has changed the composition of natural atmosphere with increased amount of greenhouse gases and other pollutants. Concentration of carbon dioxide (CO₂) has increased from a pre-industrial value of 280 ppm to the present concentration of 380 ppm [1]. With human activities like combustion of fuel, deforestation and desertification continued at the current rate, concentration of atmospheric CO₂ is expected to cross 550 ppm by mid-century and could double by end of this century [2]. Agricultural products are expected to be affected by any change in the atmospheric composition particularly an increase in CO₂, which is used as a raw material in photosynthesis. An increase in the concentration of atmospheric CO₂ is expected to increase the edible biomass of agricultural products by increasing growth and yield of plants and vegetables [3-6]. Many chemical changes can also occur in plants as a result of elevated levels of CO₂. Studies related to this hypothesis have mostly been concentrated on the growth and yield of the food crops, whereas its possible impact on the nutritional balance and elemental composition has been ignored. Available studies are predominantly on crops like wheat, maize, rice and potato, and very little information is in fact available on vegetables.

Capsicum is a plant that belongs to the *Solanaceae* family and is classified under vegetable fruit. Capsicum varieties are used worldwide as raw or cooked food, for preparation of pickles, making pastas

and sauces, as well as to produce spice in the food. It is either used fresh or preserved by open air drying. Capsicum has a number of medicinal uses worldwide[7].

Capsicum is an important source of vitamin C. It is comparable to other vitamin C rich vegetables and even citrus fruits in some cases[8]. It also contains high levels of vitamin A, E and B complex as well as antioxidants, aromas and natural colors [9] [10]. The quality of capsicum greatly varies and is affected by a number of factors; one major factor is the environmental conditions under which it is cultivated[11].

Elevated atmospheric CO₂ affects *Capsicum annuum* in different ways. It increases the growth rate and yield of the plant but has no effect on the chlorophyll content[12]. An increase in CO₂ concentration in the ambient air is also known to reduce the rate of transpiration in pepper plants [13]. Pulse - rate enhancement in CO₂ concentration has been found to decrease growth and yield of capsicum and increase visual injuries in the plant as compared to a constant increase in CO₂, which increased the plant growth and yield but did not cause injuries[12]. The CO₂ enrichment in plastic greenhouse also increased fruit length, weight, and diameter and fruit number per plant of *Capsicum annuum* [14]. Elevated CO₂ concentration reduced sucrose concentration in the pollen of *Capsicum annuum* by increasing their utilization for pollen germination under high temperature stress[15]. CO₂ is known for its effect on physiology and mortality of insects. In case of *Capsicum annuum*, the effect was studied on thrips and whiteflies, and it was found that higher CO₂ levels do not affect the insects directly but decrease the insect population indirectly by changing the plant metabolism[16]. In the present experimental work the effect of enhanced atmospheric CO₂ on the quality parameters of *Capsicum annuum* including proteins, carbohydrates, fats, fiber, ash contents, vitamin C, acidity and sugars (i.e. total sugars, reducing and non-reducing sugars), elemental composition (C, H, N, S, Fe, Ca, Mg, Zn, Mn, K, Cu) fatty acid and amino acid composition was investigated.

2 Materials and Methods

2.1 Experimental Design

Five local varieties of *Capsicum* cvs.; Angrika, Best Choice, Diana, Magama, and 99PE-1689 were selected for the study. Seeds were grown and harvested in proper seasons. Clay pots with 30cm diameter and 45cm depth were used for growing the plants. Pots were placed in greenhouses made of glass. Two different CO₂ concentrations (400 and 1000 $\mu\text{mol mol}^{-1}$) were maintained in the greenhouses. Carbon dioxide gas was released from cylinder through a pipe alongside the plant rows. Pipe height from the ground level was regularly adjusted to the plant canopy. The concentration of CO₂ was measured frequently with a Gas Analyzer (MX-42 an Oldham, France).

Soil of sandy clay loam texture was used for growing plants. Soil characteristics like electrical conductivity (0.65 d S/m) and pH (7.9) were recorded. Organic matter in soil (5.6 g/Kg), total nitrogen (0.032%), available phosphorous (11.4 mg Kg⁻¹), extractable potassium(148 mg Kg⁻¹), EDTA Zn (1.8 mg Kg⁻¹), EDTA Cu (2.4 mg Kg⁻¹) and EDTA Mn in the soil (4.2 mg Kg⁻¹) were all measured

Temperature of the air inside the green houses was continuously monitored during the experimental period and recorded. Average temperature during the growing season was 23.2°C with a maximum temperature of 34.1°C and minimum of 14.2°C.

Details of the experimental design, soil characteristics, greenhouses construction, CO₂ treatment, plant cultivation and harvest procedures have been described earlier (Khan et al., 2013).

2.2 Fruit Harvest and Analysis

Fruits of *Capsicum annuum* were harvested at two stages of maturity, green and red. Fresh samples were used for analysis of total acidity and vitamin C, for the rest of analysis dried sample was used. Analysis of fruit sample was carried out using standard methods available in the literature. Analysis of ash, fat, vitamin C, total titrable acidity and sugars were carried out according to the methods of Association of Official Analytical Chemists (AOAC, 2000). Fat contents were determined by soxhlet extractor using n-hexane solvent and ash by muffle furnace. Crude fiber was determined from the defatted material left after fat extraction by digestion with dilute acid followed by dilute base. Protein contents were

determined on the basis of total nitrogen with a conversion factor of 6.25 and nitrogen was determined with the help of CHNS elemental analyser (Vario EL III CHNS-O Elemental Analyser GmbH). Total titrable acidity was determined by titration of aqueous extract against standard alkali solution. Vitamin C was extracted from the fruit with the 15% solution of metaphosphoric acid and titrated against 2, 6-dichlorophenol indophenol dye. The dye was standardized with standard ascorbic acid solution. Sugars were determined by Lane and Eynon titrimetric method as mentioned in AOAC (2000). Reducing sugars were determined by titrating aqueous extract of plant material against Fehling's solution. Non-reducing sugars were converted to reducing sugars with citric acid and the total sugars were determined by the same method. Non-reducing sugars were determined from the difference of the two.

CHNS elemental analyzer (Vario EL III CHNS-O Elemental Analyser GmbH) was used for the determination of C, N, S and H (AOAC, 2000) while the rest of the elements (Fe, Ca, Mg, Zn, Mn, K, Cu,) were determined with atomic absorption spectrophotometer (AAS- Perkin Elmer, Analyst 700) (Allen et al., 1986).

GC-MS (Shimadzu Model QP 2010 plus) was used for the determination of fatty acids by the internal standard method. Fatty acids present in oil of the fruit were first converted to methyl esters of fatty acids (FAME) and then injected to GC-MS, using tridecanoic acid methyl ester as internal standard (AOAC, 2000).

Amino acids were extracted from the sample using 0.1N HCl and analyzed. Analysis was carried out using HPLC based amino acid analyzer (Schimadzu, Model LC-20AD) equipped with fluorescent detector (RF-10AXL) (AOAC, 2000).

2.3 Statistical Analysis

Results of triplicate analysis are expressed as mean value \pm standard deviation. Significance of the data was found from the value of probability level (P) obtained by subjecting the data to Student's t-test with software SPSS 16.0. The results were considered significant at $P \leq 0.05$, trend at $0.05 < P \leq 0.1$ and non-significant at $P > 0.1$.

3 Results

Elevated CO₂ significantly increased the number of fruits per plant for all five varieties of capsicum. Increase in number of fruits per plant was in the range of 34.61 to 50.00% and increase in the fresh weight was in the range 43.80 to 59.55% (Fig. 1A& B)

Amongst the proximate composition parameters, protein and vitamin C were reduced, while sugars and fibers contents were increased by elevated atmospheric CO₂. Protein content was studied for three varieties of capsicum. Highest amount of protein was present in the Magama variety (21.01%) and lowest amount was present in 99PE-1689 (12.29%). Elevated CO₂ significantly decreased the protein content of capsicum (Fig. 2). The decrease was highest for Best Choice variety (31.62%) and lowest for Magama (25.10%).

The major sugars in capsicum are reducing sugars (glucose + fructose) with small quantity of non-reducing sugar (sucrose). Enhanced level of CO₂ significantly increased sugar contents of capsicum (Fig. 3A). Both reducing sugars and non-reducing sugars were increased for all five varieties of capsicum. The increase was non-significant for non-reducing sugars. Elevated CO₂ increased the fiber content of capsicum varieties. For red fruits, the increase ranged between 6.09 and 9.03%, and for green fruits it ranged between 3.19 and 4.71%. Increase in fiber content was significant for matured fruits and followed a trend for premature fruits (Fig. 3B).

Enhanced atmospheric CO₂ decreased vitamin C significantly in capsicum varieties (Fig. 4A). The decrease ranged between 11.84 and 15.84% for the matured stages and 8.98 to 12.12% for green stages. Total titratable acidity of the red fruits of capsicum was slightly higher than premature green fruits, and it decreased with elevated CO₂ non-significantly ($P > 0.1$) (Fig. 4B).

Increased atmospheric CO₂ affected the mineral composition of capsicum varieties. A significant increase was observed for some minerals like H, Fe and Mn while many other important minerals were reduced including Ca, Mg and Zn. No significant trend was observed for K and Cu. Cu tended to decrease in 99PE-1689 ($0.05 < P \leq 0.1$) (Fig 5, 6 and 7).

Probability level (P) obtained from t test

Parameter	Fruit no.	Fruit Weight
99PE-1689	(0.073)	0.001
Magama	0.031	0.008
Angrika	0.024	0.005
Best Choice	(0.072)	0.007
Diana	(0.069)	0.005

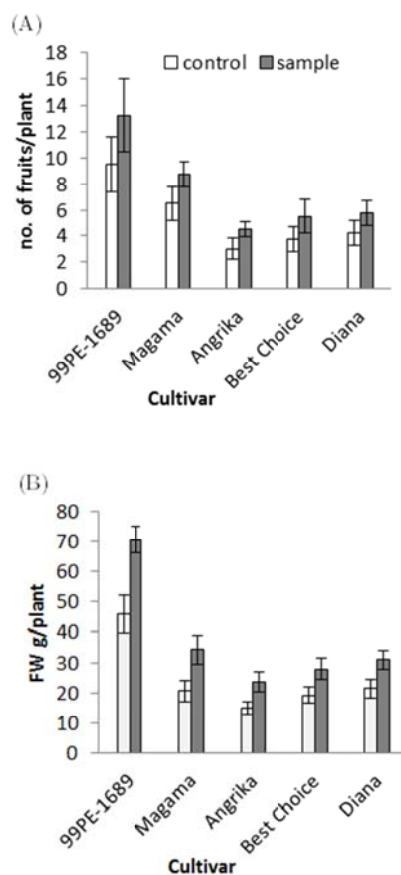


Figure 1. Effect of elevated atmospheric CO₂ on yield of capsicum fruits, (A) number of fruits per plant and (B) fresh weight in terms of g/plant. White histograms represent the data for control plants grown at 400 μmol mol⁻¹ and black histogram showing result for sample plants grown at 1000 μmol mol⁻¹. ns represents non-significant data ($P > 0.1$) and number in parenthesis indicate trend ($0.05 < P \leq 0.1$).

Probability level (P) obtained from t test

Parameter	99PE-1689 Red	Magama Red	Best Choice Red
Protein	0.024	0.004	0.002

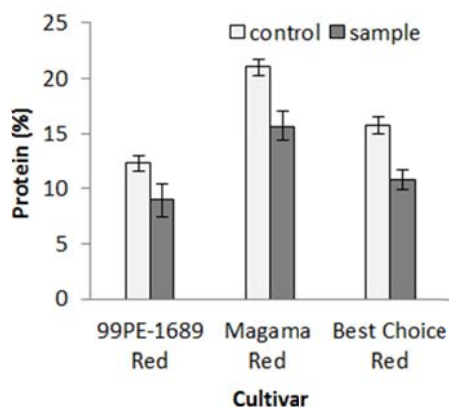


Figure 2. Effect of elevated atmospheric CO₂ on protein (%) of capsicum fruits. White histograms represent the data for control plants grown at 400 μmol mol⁻¹ and black histogram showing result for sample plants grown at 1000 μmol mol⁻¹. ns represents non-significant data (P>0.1) and number in parenthesis indicate trend (0.05<P≤0.1).

Fatty acids were determined for two varieties of capsicum, 99PE-1689 and Best Choice. Although the response of different fatty acids to elevated atmospheric CO₂ was not similar, generally a decreasing trend was observed (Table 1). Lauric acid increased very slightly for 99PE-1689 and remained unchanged for Best Choice. Myristic acid increased for both varieties. Pentadecanoic acid, palmitoleic acid, margaric acid and tricosanoic acid remained almost unaffected by elevated CO₂ for both varieties. Palmitic acid, stearic acid, oleic acid, linoleic acid, octadecadienoic acid, arachidic acid, behenic acid and tetracosanoic acid decreased for both varieties of capsicum. Elaidic acid and linolenic acid decreased for 99PE-1689 and increased for Best Choice.

Amino acids composition was determined for three varieties of capsicum. Almost all of the amino acids analyzed in capsicum varieties decreased under elevated CO₂ conditions. A slight increase was observed only for Val, Tyr and Arg in some varieties of capsicum (Table 2).

Probability level (P) obtained from t test

Parameter	Total sugars	Fibers
99PE-1689 Red	0.005	0.003
99PE-1689 Green	0.021	(0.056)
Magama Red	0.004	0.003
Magama Green	-	(0.069)
Angrika Red	0.007	0.013
Angrika Green	-	(0.067)
Best Choice Red	0.001	0.006
Diana Red	0.021	0.023

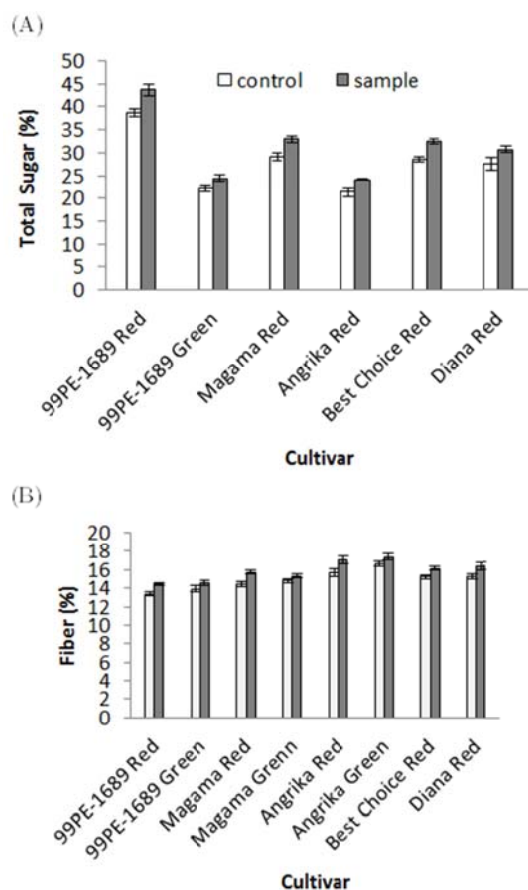


Figure 3. Effect of elevated atmospheric CO₂ on (A) total sugars (%) and (B) fiber content (%) of capsicum fruits. White histograms represent the data for control plants grown at 400 $\mu\text{mol mol}^{-1}$ and black histogram showing result for sample plants grown at 1000 $\mu\text{mol mol}^{-1}$. ns represents non-significant data ($P > 0.1$) and number in parenthesis indicate trend ($0.05 < P \leq 0.1$).

Probability level (P) obtained from t test

Parameter	Vitamin C	Total titratable acidity
99PE-1689 Red	0.001	Ns
99PE-1689 Green	0.008	Ns
Magama Red	0.001	Ns
Magama Green	0.003	Ns
Angrika Red	0.002	(0.083)
Angrika Green	0.002	Ns
Best Choice Red	0.004	Ns
Diana Red	0.005	Ns

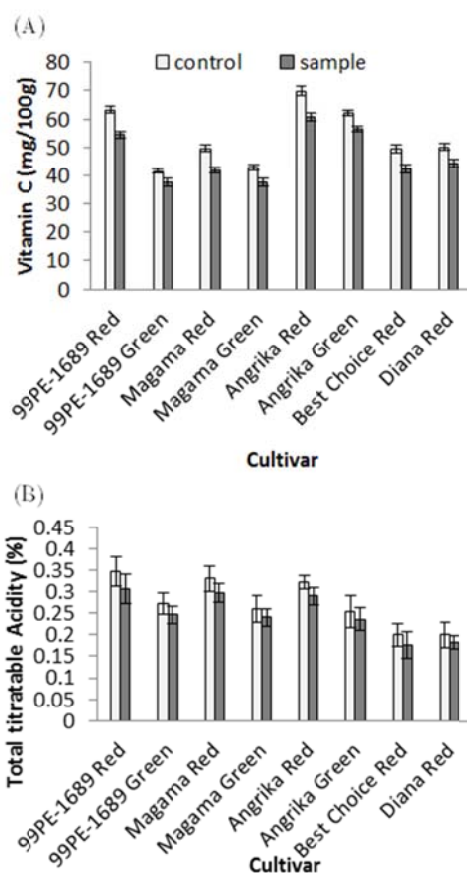


Figure 4. Effect of elevated atmospheric CO₂ on (A) vitamin C (mg/100g) and (B) total titratable acidity (%) of capsicum fruits. White histograms represent the data for control plants grown at 400 μmol mol⁻¹ and black histogram showing result for sample plants grown at 1000 μmol mol⁻¹. ns represents non-significant data (P>0.1) and number in parenthesis indicate trend (0.05<P≤0.1).

Probability level (P) obtained from t test

Parameter	Sulphur	Hydrogen
99PE-1689 Red	ns	0.001
Magama Red	ns	0.010
Best Choice Red	ns	0.004

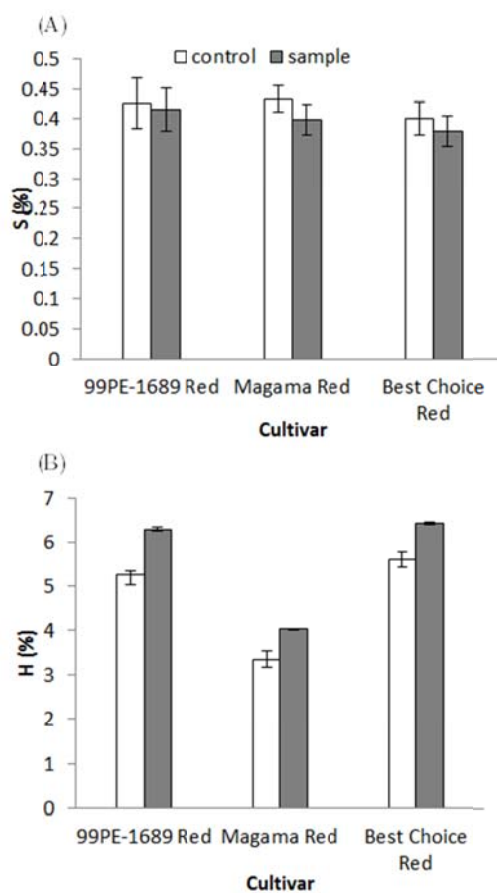


Figure 5. Effect of elevated atmospheric CO₂ on (A) sulphur (%) and (B) hydrogen (%) contents of capsicum fruits. White histograms represent the data for control plants grown at 400 µmol mol⁻¹ and black histogram showing result for sample plants grown at 1000 µmol mol⁻¹. ns represents non-significant data ($P > 0.1$) and number in parenthesis indicate trend ($0.05 < P \leq 0.1$).

Probability level (P) obtained from t test

Parameter	Calcium	Magnesium	Potassium
99PE-1689 Red	0.003	0.007	Ns
Magama Red	0.019	0.001	Ns
Angrika Red	0.016	0.001	Ns
Best Choice Red	0.022	0.001	Ns
Diana Red	0.017	0.005	Ns

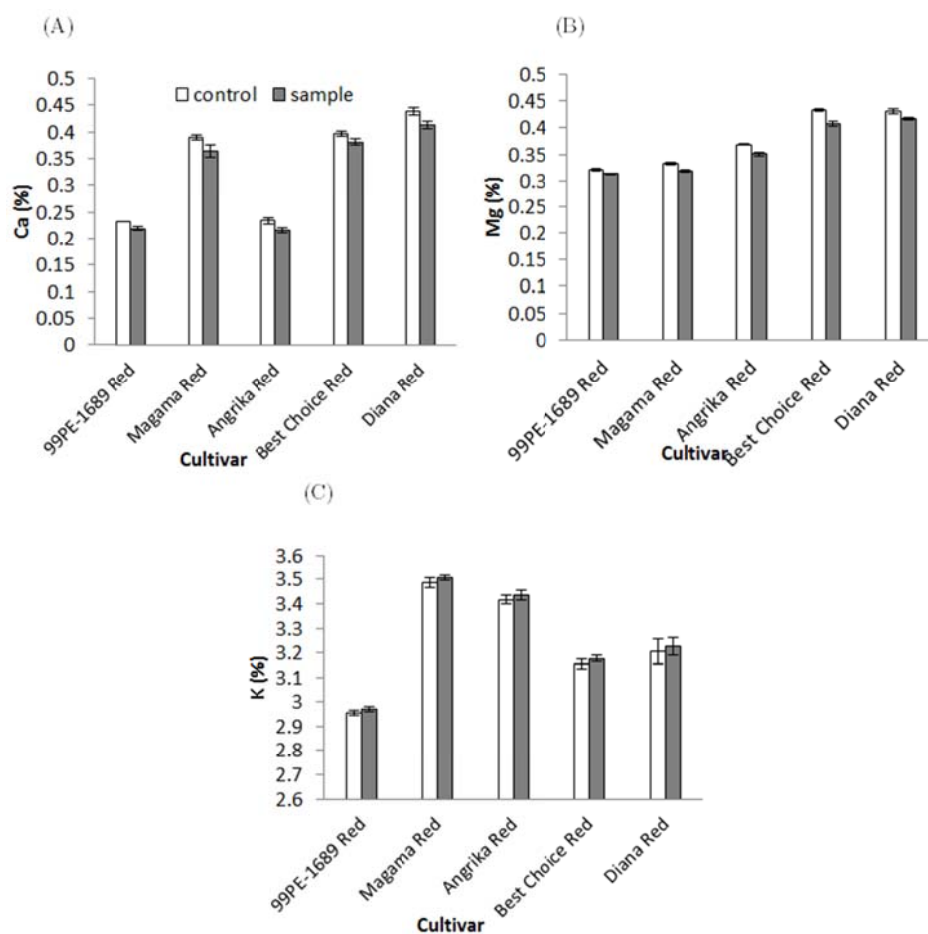


Figure 6. Effect of elevated atmospheric CO₂ on macro mineral content of capsicum fruits, (A) Calcium (%), (B) Magnesium (%) and (C) Potassium (%). White histograms represent the data for control plants grown at 400 μmol mol⁻¹ and black histogram showing result for sample plants grown at 1000 μmol mol⁻¹. ns represents non-significant data (P>0.1) and number in parenthesis indicate trend (0.05<P≤0.1).

Probability level (P) obtained from t test

Parameter	Zinc	Copper
99PE-1689 Red	0.004	(0.056)
Magama Red	0.021	Ns
Angrika Red	0.024	Ns
Best Choice Red	0.022	Ns
Diana Red	0.008	Ns

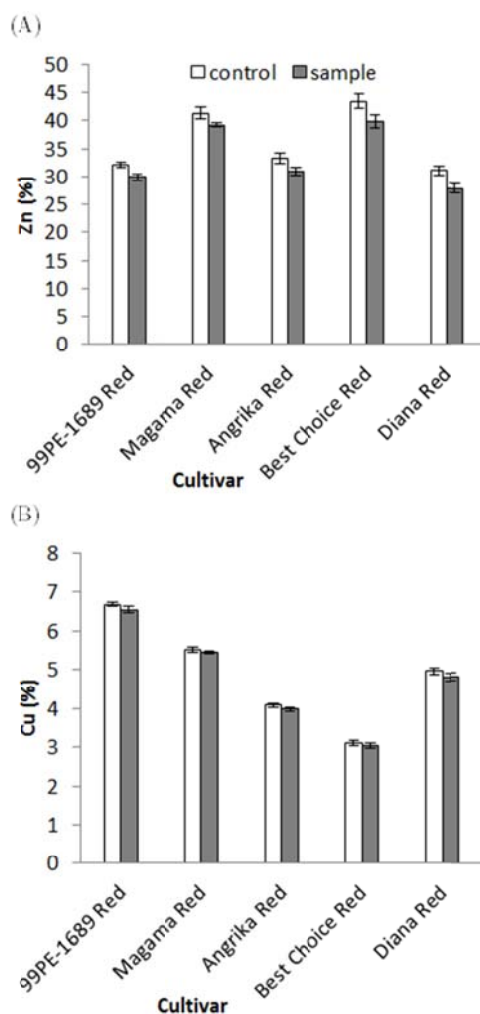


Figure 7. Effect of elevated atmospheric CO₂ on micro mineral content of capsicum fruits, (A) Zinc (%) and (B) Copper (%). White histograms represent the data for control plants grown at 400 µmol mol⁻¹ and black histogram showing result for sample plants grown at 1000 µmol mol⁻¹. ns represents non-significant data ($P > 0.1$) and number in parenthesis indicate trend ($0.05 < P \leq 0.1$).

Probability level (P) obtained from t test

Parameter	Iron	Manganese
99PE-1689 Red	<0.001	0.006
Magama Red	<0.001	<0.001
Angrika Red	<0.001	<0.001
Best Choice Red	<0.001	<0.001
Diana Red	<0.001	<0.001

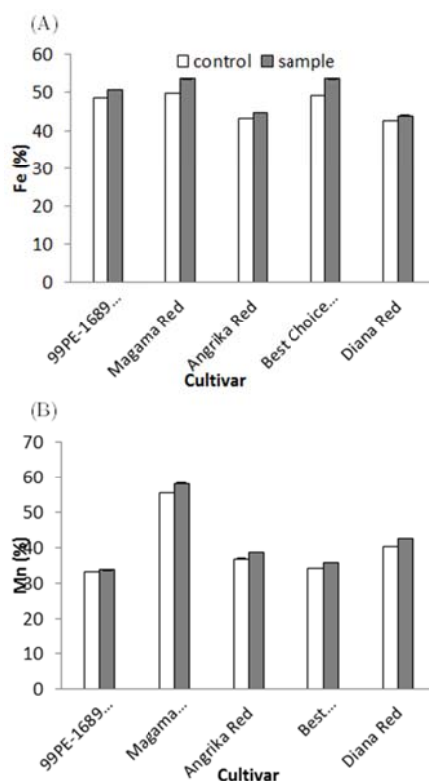


Figure 8. Effect of elevated atmospheric CO₂ on micro mineral content of capsicum fruits, (A) Iron (%) and (B) Manganese (%). White histograms represent the data for control plants grown at 400 μmol mol⁻¹ and black histogram showing result for sample plants grown at 1000 μmol mol⁻¹. ns represents non-significant data (P>0.1) and number in parenthesis indicate trend (0.05<P≤0.1).

Table 1. Fatty acids (%) composition of capsicum varieties grown at two levels of CO₂

Fatty acids	99PE-1689		Best Choice	
	concentration (μmol mol ¹)			
	400	1000	400	1000
C12:0; Lauric acid	0.007	0.009	0.002	0.002
C14:0; Myristic acid	0.036	0.045	0.020	0.038
C15:0; Pentadecanoic acid	0.002	0.002	0.002	0.003
C16:0; Palmitic acid	0.231	0.197	0.391	0.345
C16:1c; Palmitoleic acid	0.007	0.006	0.006	0.005
C17:0; Margaric acid	0.006	0.005	0.007	0.007
C18:0; Stearic acid	0.077	0.052	0.073	0.067
C18:1c; Oleic acid	0.067	0.041	0.068	0.058
C18:1n9t; Elaidic acid	0.011	0.008	0.007	0.011
C18:2c; Linoleic acid	0.850	0.738	1.147	0.833
C18:2t; Octadecadienoic acid	0.012	0.009	0.023	0.011
C18:3n3; Linolenic acid	0.312	0.188	0.203	0.490
C20:0; Arachidic acid	0.026	0.017	0.029	0.022
C22:0; Behenic acid	0.020	0.014	0.026	0.016
C23:0; Tricosanoic acid	0.004	0.002	0.004	0.003
C24:0; Tetracosanoic acid	0.019	0.014	0.019	0.012

Table 2. Amino acids (%) composition of capsicum varieties grown at two levels of CO₂

Amino acids	99PE-1689		Magama		Best Choice	
	CO ₂ concentration ($\mu\text{mol mol}^{-1}$)					
	400	1000	400	1000	400	1000
Aspartic acid	0.085	0.079	0.227	0.202	0.237	0.140
Threonine	0.054	0.039	0.269	0.124	0.212	0.084
Short consensus repeats	0.051	0.050	0.149	0.147	0.212	0.147
Glutamic acid	0.150	0.117	0.366	0.227	0.467	0.332
Proline	0.024	0.020	0.033	0.047	0.050	0.041
Glycine	0.016	0.013	0.083	0.028	0.033	0.020
Valine	-	0.039	0.089	0.034	0.053	0.070
Methionine	0.025	0.073	0.110	0.075	0.305	0.150
Isoleucine	0.221	0.190	0.230	0.167	0.331	0.141
Leucine	0.209	0.178	0.389	0.259	0.127	0.107
Tyrosine	0.093	0.078	0.157	0.120	0.046	0.075
Phenylalanine	0.011	0.012	0.139	0.039	0.047	0.033
Lysine	0.021	0.013	0.062	0.031	0.049	0.039
Arginine	0.016	0.023	0.079	0.081	0.077	0.043

4 Discussion

The purpose of the present study was to evaluate the effect of enhanced atmospheric CO₂ on the nutritional composition of different varieties of chili (*Capsicum annuum*) at two stages of maturity i.e. green and red. It was found that increased atmospheric CO₂ caused a nutritional imbalance in all varieties of capsicum at both stages of maturity by increasing sugar content, decreasing protein as well as vitamin C and disturbing the concentration of many important minerals. Since data on these particular local cultivars is not available, a point to point comparison is not possible, but the trend of change is similar to other plants studied earlier.

Capsicum fruits were picked at maturity. The plants grow and mature faster in elevated CO₂ compared to ambient plants. Analyzing fruits of same age for control and sample plants is in fact a comparison between plants of different maturity levels, which surely will be different in their nutritional composition, and will not specifically reflect the effect of elevated CO₂ [17]. In present experiment, such problems were avoided by comparing fruits harvested at same maturity level and not same age. Fruits were picked at two maturity levels, a fully matured red stage and a premature green stage.

Elevated CO₂ increased the number of fruits produced per plant as well as fresh weight of the capsicum fruits, thus increasing the yield substantially. Increase in the number and fresh weight of fruits was different for all the five varieties of capsicum, however, qualitative response of all varieties to the elevated CO₂ was similar. The results are in agreement with the previous experiments where growth, yield and root/shoot ratio of capsicum plants increased with the enhanced atmospheric CO₂ [12, 14]. This is quite expected as any increase in concentration of atmospheric CO₂, a raw material for photosynthesis, will increase the rate of photosynthesis, thus increasing the edible biomass, the final product of photosynthesis.

Protein content was different in different varieties of capsicum. Variation in the protein content of different varieties of capsicum was also noted by [18]. Nutritional quality of capsicum was negatively affected by enhanced CO₂, in terms of protein content. No such reduction in protein has been previously reported for capsicum, however it was observed for a number of other food plants like potato, rice and soybean [19, 20]. This reduction could be a result of increased amount of nonstructural carbohydrates due to increased photosynthesis leading to dilution as well as increased water use efficiency. This could be coupled with decreased rate of transpiration leading to a reduction in nitrogen uptake by leaves, as a result decreasing protein content of crops and vegetables.

Carbohydrates are major components of the capsicum. Reducing sugars (glucose and fructose) are the main sugars in capsicum as compared to non-reducing sugars (sucrose). The green stage sugar content was determined only for 99PE-1689. It was observed that red stage has higher amount of sugar content as compared to green stage. The result is in agreement with the previous studies by Martinez et al.[21] who proposed that with fruit maturity, changes in action of cell wall degrading enzymes result in accumulation of sugars and carbohydrates in the cell walls. Sugar content of the capsicum was significantly increased by elevated CO₂, however the increase was non-significant for non-reducing sugars. Increase in sugars is higher for red stage of 99PE-1689 as compared to its green stage.

Effect of enhanced CO₂ on sugar and carbohydrate content of capsicum is not reported, however increase is observed for other crops and vegetables[20, 22]. Increase in carbohydrate concentration is not unexpected as enhanced atmospheric CO₂ leads to increase in photosynthesis, resulting in greater amount of plant non-structural carbohydrates leading to relatively more carbon based secondary compounds.

Fiber content of capsicum was higher and was almost similar for green and red stages of all varieties, results being in agreement with the previous work by Martinez et al. [21] Increase in the fiber content followed a trend for green stages and was significant for red stages. Although the fiber contents of red and green stages were not much different, the effect of elevated CO₂ on fiber content was much pronounced for red stage as compared to the green stage.

Vitamin C is a very important constituent of capsicum, found in more quantity in red stage as compared to the green stage. Vitamin C contents of two varieties, among a total of five, fall within the range of Recommended Dietary Allowance (RDA) (60-90 mg/day). Vitamin C contents of the rest of the three varieties were slightly below the required amount. Enhanced atmospheric CO₂ decreased vitamin C contents of all varieties significantly. The reduction for red stage was slightly more than the green stage of the corresponding variety. Capsicum is a very important source of vitamin C and its vitamin contents are comparable to the other recognized good sources this vitamin[23]. In our experiments, elevated CO₂ adversely affected nutritional quality by decreasing Vitamin C content of capsicum fruit.

Total titratable acidity, which represents organic acid content in the fruit, was higher at the red stage as compared to green stage, results similar to previous findings [21]. Acidity of capsicum was not affected by atmospheric CO₂, although a small decrease in acidity of both red and green stages was observed but the decrease was statistically non-significant.

Mineral composition of capsicum varieties was altered by elevated CO₂. Hydrogen was increased, while Ca and Mg decreased, while no effect was observed on S and K concentration. Amongst the micro minerals, Zn decreased, Fe and Mn increased while no significant effect was observed on Cu content. Cu followed a trend ($0.05 < P \leq 0.1$) in 99PE-1689. Hydrogen along with C is the main constituents of carbohydrates and increase in H may be due to increase in carbohydrates concentration with enhanced atmospheric CO₂. Reduction in Nitrogen and other minerals could be a result of dilution caused by increase in non- structural carbohydrates, or a decrease in the uptake of minerals due to reduced transpiration from leaves with elevated CO₂ [24]. Increase in important nutrients like Fe and Mn with elevated CO₂ is favorable, but at the same time reduction in Ca, Mg and Zn could adversely affect the nutritional quality of capsicum fruit.

In our experiment, the major fatty acids found in capsicum were Palmitic, linoleic and linolenic acid. These results are in agreement with previous work of Cook et al.[18]. He determined chemical composition of 13 varieties of capsicum and found that linoleic and linolenic acid were major fatty acids. Enhanced atmospheric CO₂ affected all the three major fatty acids of capsicum. Palmitic and linoleic acids decreased for both varieties while linolenic acid decreased for one variety only. Linoleic and linolenic acids are essential fatty acids, as they are not only used as fuel but also play important role in biological processes in human body. Human body is unable to synthesize these acids and they must be taken in the diet. Decrease in concentration of fatty acids by elevated CO₂ is also in agreement with the reduction in crude fat content of capsicum varieties. Decrease in concentration of essential fatty acids will lower the nutritional quality of capsicum fruits in future CO₂ enriched atmosphere.

The decrease in amino acid concentration with elevated CO₂ observed in this work has not been reported for Capsicum. The reduction in present experiment is in accordance with the earlier reported decrease in amino acid contents of potato tuber [20] and wheat grains [25]. Reduction in amino acid

contents, especially the essential amino acids like isoleucine, leucine, phenylalanine, lysine, threonine and tyrosine under elevated CO₂ conditions will lower the nutritional quality of capsicum.

5 Conclusions

The nutritional quality of the capsicum was affected by elevated CO₂ in the present experiment. The general trend is that important nutritional parameters including protein and vitamin C were negatively affected by enhanced atmospheric carbon dioxide, while sugars and fibers were increased. This is an alarming factor for the modern world where most of the people search for protein rich diet and want to avoid sugars and carbohydrates especially in case of diabetic patients. Mineral and essential fatty acid content of capsicum varieties was also altered with enhanced atmospheric CO₂ with a general reducing trend. Among minerals, Ca, Mg and Zn contents of all five capsicum varieties are reduced which may increase the chances of hidden hunger. Reduction in important minerals is usually thought to be a result of nutritional dilution which is caused by increase in non-structural carbohydrates in CO₂ enriched air. Another reason might be reduced transpiration which in turn decreased the rate of uptake of important minerals from soil. Important essential oils like Palmitic, linoleic and linolenic acid were also negatively affected in CO₂ enriched atmosphere. Essential fatty acids must be taken through diet as human body is unable to synthesis them. Decrease in concentration of essential fatty acids will lower the nutritional quality of capsicum fruits in future CO₂ enriched atmosphere. The biochemical response of different varieties of capsicum to CO₂ enrichment is the same qualitatively; however its quantitative response is not exactly the same. Although our data is on a limited scale, the message is loud and clear that food quality will be negatively affected by the CO₂ build up in the atmosphere. The data on CO₂ enriched air obtained so far support the hypothesis that breeding new crop varieties along with better crop management are possible measures to combat the eminent food security threat with the climate change.

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References

1. Keeling, R.F., et al., Carbon Dioxide Research Group., 2008, Scripps Institution of Oceanography, University of California: California.
2. IPCC, IPCC(2007), in Climate Change. The Physical Science Basis. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change. Intergovernmental Panel on Climate Change, Cambridge Univ. Press 2007.
3. Allen Jr., L.H., et al., Advances in Carbon Dioxide Research. ASA Special Publication 611997. 179–228.
4. DaMatta, F.M., et al., Impacts of climate changes on crop physiology and food quality. Food Res. Int., 2010. 43(7): p. 1814–1823.
5. Kimball, B.A., K. Kobayashi, and M. Bindi, Responses of agricultural crops to free-air CO₂ enrichment. Adv. Agron., 2002. 77: p. 293-368.
6. Leakey, A.D.B., et al., Elevated CO₂ effects on plant carbon, nitrogen, and water relations: Six important lessons from FACE. J. Exp. Bot., 2009. 60(10): p. 2859–2876.
7. Bosland, P.W., Capsicums: Innovative uses of an ancient crop. Progress in new crops. ASHS Press, Arlington, VA, 1996: p. 479-487.
8. Hägg, M., S. Ylikoski, and J. Kumpulainen, Vitamin C and alfa- and beta-carotene contents in vegetables consumed in Finland during 1988-1989 and 1992-1993. J. Food Comp. Anal, 1994. 7: p. 252-259.
9. Howard, L.R., et al., Changes in phytochemical and antioxidant activity of selected pepper cultivars (Capsicum species) as influenced by maturity. J. Agric. Food. Chem., 2000. 48: p. 1713-1720.
10. Mulchi, C.L., et al., Growth and physiological characteristics of soybean in open-top chambers in response to ozone and increased atmospheric CO₂. Agr. Ecosyst. Environ., 1992. 38(1-2): p. 107-118.
11. Rajput, J.C. and Y.R. Parulekar, El pimiento, in Tratado de Ciencia y Tecnologia de lashortalizas, D.K. Salunkhe, Editor 2004, Zaragoza, Spain: Acriba. p. 203-225.

12. Penuelas, J., et al., Detrimental effects of fluctuating high CO₂ concentrations on peppers. *Photosynthetica*, 1995. 31: p. 361-370.
13. Janes, B.E., Effect of carbon dioxide, osmotic potential of nutrient solution, and light intensity on transpiration and resistance to flow of water in pepper plants. *Plant Physiol.*, 1970. 45: p. 95-103.
14. Furlan, R.A., et al., Effect of irrigation water depth and CO₂ application on sweet pepper yield cv. Mayata in plastic greenhouse. *Horticultura Brasileira*, 2002. 20(4): p. 547-550.
15. Aloni, B., et al., The effect of high temperature and high atmospheric CO₂ on carbohydrate changes in bell pepper (*Capsicum annuum*) pollen in relation to its germination. *Physiol. Plantarum* 2001. 112: p. 505-512.
16. Grodzinski, B., et al., Regulating plant/insect interactions using CO₂ enrichment in model ecosystems. *Adv. Space Res.*, 1999. 24: p. 281-291.
17. Taub, D.R. and X. Wang, Why are Nitrogen Concentrations in Plant Tissues Lower under Elevated CO₂? A Critical Examination of the Hypotheses. *J. Integr. Plant Biol.*, 2008. 50(11): p. 1365-1374.
18. Cook, J.A., et al., Nutrient and chemical composition of 13 wild plant foods of Niger. *J. Food Comp. Anal.*, 2000(13): p. 83-92.
19. Taub, D.R., B. Miller, and H. Allen, Effects of elevated CO₂ on the protein concentration of food crops: a meta-analysis. *Global Change Biol.*, 2008. 14(3): p. 565-575.
20. Högy, P. and A. Fangmeier, Atmospheric CO₂ enrichment affects potatoes: 2. Tuber quality traits. *Eur. J. Agron.*, 2009. 30(2): p. 85-94.
21. Martinez, S., et al., The composition of Aronia peppers (*Capsicum annuum* L.) at different stages of maturity. *Int. J. Food Sci. Nutr.*, 2007. 58(2): p. 150-161.
22. Islam, M.S., T. Matsui, and Y. Yoshida, Effect of carbon dioxide enrichment on physico-chemical and enzymatic changes in tomato fruits at various stages of maturity. *Sci. Hort.*, 1996. 65(2-3): p. 137-149.
23. Hägg, M., S. Ylikoski, and J. Kumpulainen, Vitamin C and alfa- and beta-carotene contents in vegetables consumed in Finland during 1988-1989 and 1992-1993. *J. Food Comp. Anal.*, 1994. 7(252-259).
24. Gifford, R.M., D.J. Barrett, and J.L. Lutze, The effects of elevated [CO₂] on the C:N and C:P mass ratios of plant tissues. *Plant. Soil*, 2000. 224(1-14).
25. Högy, P., et al., Grain quality characteristics of spring wheat (*Triticum aestivum*) as affected by free-air CO₂ enrichment. *Environ. Exp. Bot.* DOI: 10.1016/j.envexpbot.2011.12.007, 2011.