How to Cite:
Sreekumar, A., Sashna, N. C., & Harilal, C. C. (2022). A comparative assessment of the CO2 assimilation potentials of Simarouba glauca (D.C.) and Syzigium cumini (L.) skeels. *International Journal of Health Sciences*, *6*(S9), 3674–3689. Retrieved from https://sciencescholar.us/journal/index.php/ijhs/article/view/13432

A comparative assessment of the CO_2 assimilation potentials of *Simarouba glauca* (D.C.) and *Syzigium cumini* (L.) skeels

Aparna Sreekumar
Environmental Science Division, Department of Botany, University of Calicut, Thenhipalam, 673635, Malappuram, Kerala, India
Email: aparnaavani@gmail.com

Sashna N.C
Environmental Science Division, Department of Botany, University of Calicut, Thenhipalam, 673635, Malappuram, Kerala, India

Harilal C.C.
Environmental Science Division, Department of Botany, University of Calicut, Thenhipalam, 673635, Malappuram, Kerala, India

***Abstract*---**The study assesses the growth and biochemical responses of Simarouba glauca (D.C.) and Syzigium cumini (L.) skeels to elevated levels of carbon dioxide under controlled growth conditions. A standardization study, excluding plants, was also undertaken for a period of 15 days to assess the resultant flux of CO_2 associated with the growth chambers. The study revealed that the day flux of CO_2 in the treated chamber differed from the control, in both the plants. Percentage day and night flux in CO_2 in the treated chamber having Simarouba glauca was higher than that of Syzigium cumini. Simarouba glauca was noted to be more efficient in carbon assimilation than Syzigium cumini which is evident from the CO_2 consumption rate of the plant during the day, increased plant height, stem thickness, leaf area, carbohydrates, minerals, plant carbon, C/N ratios. Hence Simarouba glauca can be promoted as an avenue tree with higher potentialities in carbon sequestration.

***Keywords*---**Simarouba glauca, Syzigium cumini, controlled growth chambers, diurnal flux, biochemical responses.

Introduction

Due to global warming and climate change the average global temperature is increasing. The increased levels of greenhouse gases in the atmosphere are contributed by natural events and human activities. The major share of greenhouse gases is carbon dioxide. There are many ways to reduce carbon dioxide in the atmosphere. One of the methods is carbon capture and sequestration. Carbon sequestration in trees is a cost-effective method for mitigation of global warming.Trees are natural sequesters of carbon since they store it in the form of biomass or wood. The trees assimilate carbon and is retained for longer duration with little leakage into atmosphere [1]. Response of trees to elevated CO_2 levels has been attempted by many. Study by [2] revealed that saplings of *Swietenia mahagoni* captured more CO_2 in the wood, compared to leaves and bark. [3] reported a positive response with increase in seedling height due to increased use of CO_2 for carbon assimilation and reduced photorespiration in *Santalum album*.

Carbon offset planting envisions the selection of ideal species with higher sequestration efficiencies and their strategic planting in ideal locations. Only limited number of tree species have been subjected to efficiency studies and a sizable number are remaining. Rising levels of CO_2 is reported to alter plant carbon fluxes, and is observed in most of the studies conducted in growth chambers [4]. In this study, the responses of two tropical tree species, *Simarouba glauca* and *Syzigium cumini* were assessed in CO_2 enriched conditions in controlled growth chambers.

From several studies it is revealed that *S. glauca* (Family Simaroubaceae) is one of the most important avenue tree species in India for afforestation programme which contribute to mitigate the climate change impact. It is tropical in distribution.The tree is evergreen with well-developed root system. *S. glauca* has got high degree of adaptability and fast biomass producing potential. In India plantations of *S. glauca* are present in Orissa, Andhra Pradesh, Karnataka, Maharashtra and Tamil Nadu. *S. cumini* (Family Myrtaceae) is an evergreen and tropical tree favoured for its fruit, timber and ornamental value. It is commonly known as Java plum/ black plum. Being a myrtacean member, its leaves are aromatic. Wood of the tree is water resistant and hence used in railway sleepers. It is native to Indian subcontinent and adjoining regions of Southeast Asia including Myanmar, Sri Lanka and Andaman Islands.

Materials and Methods

Plantlets of *S. glauca* and *S. cumini* were raised from certified seeds procured from the Seed Centre, Kerala Forest Research Institute, Peechi, Thrissur, Kerala, India. They were then raised in grow bags having potting mixture (soil, sand and organic manure) in the ratio 2:1:1. They were then maintained in a polyhouse for 18 months, with adequate supply of water, nutrients and periodic monitoring of growth parameters.

Carbon dioxide- controlled chamber – Experimental setup

For experimentation with each plant, two controlled growth chambers, each with a size of length 1.8m, breadth 1.8m and height 2.4 m were used. The chambers were made with PVC pipes, covered with polyethylene sheets, permitting light inside. The control chamber was equipped with the facility for the supply of ambient air through an air compressor, whereas the treatment chamber was equipped with a CO_2 cylinder and an air compressor for the supply of CO_2 – air mixture in specific doses. Both the chambers were fitted with the facility for the analysis of CO_2 (ppm), temperature and humidity, together with an exhaust facility for controlling micro climatic conditions inside the chamber, if required. The chambers were also fitted with a semi-automated facility for the irrigation of plantlets during experimentation. The experimental setup for both the plants are given in figure 1 a&b.

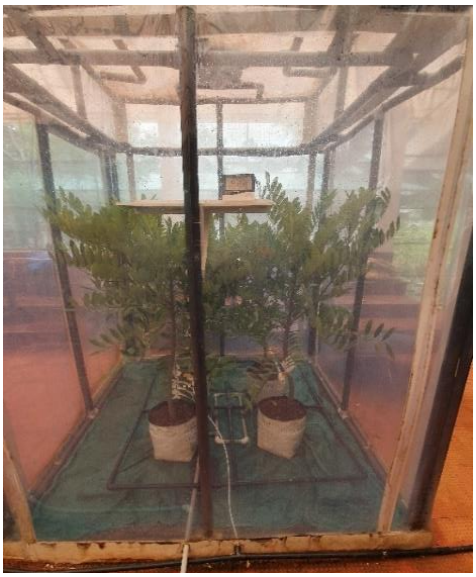

FIGURE 1a Experimental setup for control chamber

FIGURE 1b Experimental setup for treated chamber

CO_2 supply and monitoring of micro environmental conditions

Two sets of plants (four each) from *Simarouba glauca* and *Syzigium cumini* grown ideally for 18 months were selected, of which one set was taken to the control chamber having ambient air supply and the other to the chamber having elevated supply of CO_2.The chambers were then sealed to prevent the exchange of air from outside. To the treatment chamber, CO_2 mixed with air has been supplied for about 15 minutes, every day in the morning, maintaining a resultant level of $CO_2$to 990.5 ± 116.6 ppm. Similarly, the control chamber was supplied with ambient air daily, for 15 minutes maintaining a CO_2level of 654 ± 96.17 ppm. The magnitude of temperature, humidity, and carbon dioxide concentration associated with both the chambers were monitored twice a day at 9am and 6pm. Estimation of CO_2 associated with both the chambers were carried out through an automated CO_2 analyzer (NDIR type Infrared Gas Analyzer, Fuji Electric, Japan). Temperature and humidity were monitored using Billion bag digital wireless electronic Hygro-thermometer. Light intensity inside the control and treated chambers were assessed using solar radiation monitor. The experimentation was carried out for 15 days. The facilities associated with CO_2 supply and monitoring include CO_2 cylinder with regulator, compressor facility for CO_2 – air mixture and NDIR type Infrared Gas Analyzer.

Measurement of growth parameters

Growth attributes of *S. glauca and S. cumini* were assessed at two stages of experimentation, one on the initial day (0DoT) and the other on the final day of treatment (15DoT). Morphological parameters assessed include plant height, stem thickness and area of leaves. A measuring tape was used to measure the plant height from the level of soil to the region of active meristem. Stem thickness was measured at the collar level using a screw gauge. Total number of leaves were

counted and the length of leaves together with their breadth at the widest portion was measured. Leaf area was calculated from the above parameters.

Estimation of biochemical parameters

Biochemical parameters associated with *S. glauca* and *S. cumini* from the control and CO_2 treatment sets were recorded at 4 stages (0DoT, 5DoT, 10DoT and 15DoT). Parameters analyzed include leaf pigments (total chlorophyll and carotenoids), carbohydrates, phenol and protein. Pigments were estimated according to [5]. Carbohydrate was estimated following [6]. Phenol and protein content were estimated following [7] and [8] respectively.

Estimation of minerals

Estimation of the mineral contents of plants was undertaken at four stages of growth (0DoT, 5DoT, 10DoT and 15DoT). For the estimation of minerals, plant parts were separated, dried and subjected to perchloric acid - nitric acid - sulphuric acid digestion. Titration method was followed to determine the extent of calcium and magnesium. Elements such as sodium and potassium were estimated using a flame photometer (Systronics, 128). For determining the C/N ratio of plants, leaves after drying were ground to powder. Carbon and nitrogen contents of each sample was measured using instrument FLASH 2000 (Thermofisher).

Standardization studies of growth chambers

A standardisation study was undertaken in the same way for assessing the daily flux of gases associated with both the chambers, in the absence of plants. For this, the entire experiment was repeated in the absence of plants, with simultaneous supply of air / air - CO_2 mixture to the respective chambers and subsequent monitoring of temperature, humidity, and CO_2 associated with the chambers at specific time intervals of a day (9am and 6pm) for 15 days. The data so generated was used to assess the net flux in CO_2 in the presence and absence of plants within the growth chambers.

Statistical analysis

Two-way Analysis of Variance (ANOVA) was carried out and the significance was tested at 0.05 level of critical difference using SPSS version 27.

Results and Discussion

CO_2 flux in the growth chamber

Day flux of CO_2 was estimated as the difference in the extent of CO_2 supplied in the morning with that of the CO_2 retained in the evening. Similarly, night flux was assessed as the extent of CO_2 in the evening with that of its extent in the next day morning. The day flux of CO_2 associated with the control and the CO_2 treated chambers of standardisation studies and studies with *S.glauca* and *S.cumini* are represented in table I.

In both standardization studies and experimentation with the plants, a progressive reduction in the extent of CO_2 was noticed in the evening, compared to morning. In the case of standardisation study, the percentage decline in day flux in the control chamber was 4.618% and that of the treated chamber was 7.56 %. Reduction in day flux in the control chamber of *S. glauca* was 24.8% and that of its CO_2 treated chamber was 39.6%. Day flux in the control chamber of *S. cumini* decreased by 6.2%and treated chamber by 0.29%. Day flux are indications of the sequestration efficiencies of plants [9]. In the present study, plants within the CO_2 treated chamber assimilated a higher share of CO_2 compared to control and that of the standardization studies. The higher flux in CO_2 within the treated chamber can be due to the efficiency of plants growing within it. The night flux of CO_2 attributed by *S. glauca* and *S. cumini* in the control and treated chambers with that of the standardisation studies are represented in table II.

Night flux in standardisation study in control chamber is 5.7 % and 2.39 % in treated chamber. The results showed an increase in night flux of CO_2 in both control (33.5%) and treated chambers (62.73%) having *S. glauca*, and this might be due to the respiratory attribution by the plants during night. Similarly in *S. cumini*, night flux of CO_2 in control increased by 5.53% and 8.9 % in treated. According to [10], plants grown in elevated CO_2 have higher respiration rates than control, which substantiates the findings of the present study. Increase in CO_2 enhances respiration which is associated to higher carbohydrate concentration and higher substrate availability [4].

Table I day flux of CO_2 in control and treated chambers

Day flux (ppm)						
	Control			Treated		
Day	Std	*Simarouba glauca*	*Syzigium cumini*	Std	*Simarouba glauca*	*Syzigium cumini*
1	15	-179	17	-70	-425	-113
2	-38	97	-54	-58	-52	-189
3	-63	-300	-43	-57	-371	-206
4	-28	-240	-75	-73	-425	-238
5	-26	-56	-136	-85	-244	-231
6	-22	-295	-66	-76	-498	-320
7	-17	-267	-63	-81	-521	-332
8	-33	-263	-1	-56	-478	-418
9	-32	-36	4	-96	-450	-303
10	-41	-95	3	-109	-466	-234
11	-36	-190	-4	-68	-500	-316
12	-19	-233	-11	-94	-560	-258
13	-42	-239	-15	-95	-456	-294
14	-48	-219	-12	-61	-507	-271
Res	-30.7±17.93	-179.64±115.58	-36.38±42.23	-77.07±16.83	-425.21±131.85	-265.92±73.94
% change	-4.618	-24.8	-6.2	-7.56	-39.6	-0.29

'Std'- Standardisation, '+' - CO_2 attribution to the growth chamber, '-' - CO_2 reduction in the growth chamber

Table II night flux of CO_2 in control and treated chambers

Night flux (ppm)						
	Control			Treated		
Day	Std	*Simarouba glauca*	*Syzigium cumini*	Std	*Simarouba glauca*	*Syzigium cumini*
1	39	184	77	18	478	81
2	58	169	28	32	52	31
3	27	246	97	36	371	102
4	38	34	55	2	367	75
5	22	251	59	19	484	75
6	46	271	32	11	488	44
7	29	269	28	13	465	72
8	39	70	21	28	223	-1
9	48	15	-2	30	418	12
10	41	198	7	19	466	51
11	28	229	27	22	458	99
12	20	268	19	17	560	57
13	52	191	15	46	456	55
14	25	187	5	23	407	55
Res	36.6±11.5	184.4± 86.3	33.4 ±28.6	22.6 ± 11.2	406.6± 128.8	57.71 ± 29.75
% change	5.7	33.5	5.53	2.39	62.73	8.9

'Std'- Standardisation, '+' - CO_2 attribution to the growth chamber, '-' - CO_2 reduction in the growth chamber

Microclimatic conditions within growth chambers

Micro climatic conditions like temperature and humidity experienced in both the experimental conditions with and without plants are given in tables III and IV respectively.

Table III range of temperatures (minimum, maximum and mean) noticed within the chambers under varying experimental conditions

Experimental condition	Standardisation studies		*Simarouba glauca*		*Syzigium cumini*	
	Morning temperature ^{0}C (Range and Mean)	Evening temperature ^{0}C (Range and Mean)	Morning temperature ^{0}C (Range and Mean)	Evening temperature ^{0}C (Range and Mean)	Morning temperature ^{0}C (Range and Mean)	Evening temperature ^{0}C (Range and Mean)
Control chambers	37.5 – 44.0	32.3 – 37.3	28.6-37.4	27-35.1	29.6-38.1	29.5-34.9
	40.3±1.9	35.30±1.78	32.29±2.62	32.29±2.28	33.93±3.47	32.08±1.61
Treated chambers	39.3-44.4	33.1-37.1	28.1-37.9	27-34.5	29-38.8	29.8-34.3
	40.78±1.8	35.12±1.55	32.39±2.67	32.12±2.12	32.71±3.33	31.98±1.37

Table IV Range of humidity (minimum, maximum and mean) noticed within the chambers under varying experimental conditions

Experimental condition	Standardisation studies		*Simarouba glauca*		*Syzigium cumini*	
	Morning Humidity % (Range and Mean)	Evening Humidity % (Range and Mean)	Morning Humidity % (Range and Mean)	Evening Humidity % (Range and Mean)	Morning Humidity % (Range and Mean)	Evening Humidity % (Range and Mean)
Control chambers	44-56	51-70	99	89-99	99	87-99
	51.08± 3.65	57.57 ± 6.02	99	95.69±3.4	99	97.93±3.2
Treated chambers	47-59	59-79	99	99	74-99	93-99
	54±3.8	68.64±6.01	99	99	97.33±6.45	98.6±1.55

The results showed variations in microclimatic conditions over a range and can be attributed to the varying concentrations of CO_2 inside the chambers. In the standardisation studies, morning temperatures in the control and treated chambers (^oC) were 40.3 ± 1.9 and 40.78±1.8 and those of evening temperatures were 35.3±1.78 and 35.12± 1.55, respectively. However, the morning and evening temperatures in the control chamber having *S. glauca* were 32.29±2.62 and 32.29±2.28 and that of the treated chamber was 32.39±2.67 and 32.12±2.12, respectively. In control chambers of *S. cumini* morning and evening temperatures are 33.93±3.47 and 32.08±1.61 and in treated chambers morning temperature is 32.71±3.33 and evening temperature is 31.98±1.37. Carbon dioxide, absorbs and emits radiation in the thermal infrared range and can greatly influence temperatures. In this study, stimulation in CO_2 showed a slight deduction in temperature in chambers with *S. cumini*.

Percentage humidity (table IV) also showed variations, which are attributed to varying concentrations of CO_2. In the standardisation study, morning and evening humidity in the control chamber was 51.08±3.65 and 57.57± 6.02, whereas it was 54± 3.8 and 68.64± 6.01 respectively in the treated chamber. The humidity in the control chamber having *S. glauca* was 99% (morning) and 95.69±3.4 (evening), and in the treated chamber, average morning and evening humidity was found to be the same (99%). In control chambers with *S. cumini*, morning and evening humidity was found to be 99%and 97.93±3.2 respectively whereas in treated chambers, humidity is 97.33±6.45 in the morning and 98.6±1.55 in the evening. Present study showed slight increase in humidity under stimulated CO_2 in *S. glauca* which is consistent with the results of [11]. It is postulated that the metabolic status and resultant evapo-transpiration by the aerial and belowground biomass of plants, evaporation from soil, activity of soil microorganisms etc. can attribute humidity to external environmental conditions. Light intensity inside the control and treated chamber measured using solar radiation monitor was found to be within the range of 100- 250µ mol/m^2/sec.

Growth responses of S. glauca and S. cumini under CO_2 treatment

Data regarding the growth attributes of *S. glauca and S. cumini* subjected to experimental conditions are presented in Table V.

Table V Variations in growth parameters of *s. glauca and s. cumini* under control and CO_2 treated conditions

	Simarouba glauca						*Syzigium cumini*					
	Control			Treatment			Control			Treatment		

Mean values of growth parameters such as plant height, stem thickness and leaf area increased under elevated CO_2 in both the plants. After 15 days of experimentation, an increase of 0.54%, 2.79% in plant heights were noticed in *S. glauca* and *S. cumini* under treatment with CO_2. In a study conducted in cassava plants, [12] noticed an increase in plant height under elevated CO_2. Stem thickness increased by 5.22% and 1.96% in *S. glauca* and *S. cumini.* An increase in stem thickness was reported in *Trifolium alexandrium* and *Theobroma cacao* when exposed to elevated CO_2 [13 and 19]. In the present study, an increase of 58.78% in leaf area was observed under elevated CO_2 in *S. glauca* and an increase of 14.54% was observed in *S. cumini*. Changes in cell wall loosening or extensibility causes the stimulation in leaf expansion due to elevated CO_2. It is not related to cellular water content in growing leaves [14]. Increased leaf area under elevated CO_2 might be due to increased nitrogen input, cell expansion, leaf thickness, and the number of palisade cell layers [12].

Variations in pigments and metabolites in S. glauca and S. cumini under experimentation

Plants respond to changes in its external environment by adjusting biochemically [15]. The changes in pigments and other biochemical components in *S. glauca and S. cumini* in response to elevated levels of CO_2 has been assessed at four stages of growth. Mean values of pigments such as total chlorophyll, carotenoids and other biochemical components like carbohydrates, protein and phenol contents in plants from the control and elevated CO_2 supply are estimated on the initial, 5^{th}, 10^{th} and 15^{th} day of experimentation and the results are presented in table VI. In the present study with *S. glauca,* total chlorophyll and carotenoids decreased by 8.02%, 13.04% in treated compared to control. The study conducted by [16] in *Medicago sativa* reported increased chlorophyll content under elevated levels of CO_2 due to improved substrate availability for assimilation and reduced water loss due to lower stomatal conductance. In *S. cumini* all the pigments showed a decline. It is found that in *S. glauca and S. cumini,* chlorophyll content decreased with the accumulation of non-structural carbohydrates under elevated CO_2 concentration and temperature which in accordance with the study of [17].

Table- V1 Biochemical responses of *simarouba glauca* and *syzigium cumini* to elevated levels of CO_2

Simarouba	Days	Total chlorophyll (mg/g)	Carotenoids (mg/g)	Carbohydrate (mg/g)	Protein (mg/g)	Phenol (mg/g)
Control	DOT 1	1.008±0.259	0.191±0.038	156.58±19.81	740.73±99.32	421.08±47.23
	DOT 5	2.122±0.612	0.323±0.091	147.44±34.69	638.52±135.82	285.57±53.6
	DOT 10	1.129±0.350	0.20±0.057	138.91±44.40	354.2±113.21	301.69±60.59
	DOT 15	1.278±0.446	0.22±0.067	146.01±34.67	692.61±110.66	297.03±32.26
Mean		1.378	0.233	147.2	606.5	326.275
SE		0.254	0.030	3.615	3.615	3.615
Treated	DOT 1	1.061±0.290	0.1887±0.057	175.83±16.73	606.37±108.37	352.54±210.38
	DOT 5	1.675±0.460	0.260±0.061	140.42±25.46	522.06±161.83	257.8±93.46
	DOT 10	1.198±0.250	0.196±0.038	140.35±35.75	565.93±156.03	323.73±88.10
	DOT 15	1.158±0.086	0.193±0.021	137.86±26.52	547.27±105.14	273.11±109.93
Mean		1.2675	0.205	148.615	560.4075	301.795
SE		0.137	0.018	9.09	17.76	22.01
% change		-8.02	-13.04	+0.95	-7.6	-7.5
P value		NS	NS	NS	< 0.05	NS
Syzigium						
Control	DOT 1	0.92±0.16	0.16±0.03	55.06±19.29	297.73±185.82	230.21±147.21
	DOT 5	1.39±0.23	0.37±0.06	85.66±39.5	290.80±111.43	257.2±90.26
	DOT 10	1.78±0.08	0.27±0.07	66.93±27.81	666.05±70.90	241.78±69.06
	DOT 15	1.54±0.51	0.25±0.06	82.34±7.54	1015.37±193.58	351.10±231.61
Mean		1.408	0.263	72.498	567.450	270.050
SE		0.181	0.043	7.1	173.1	27.57
Treated	DOT 1	1.15±0.06	0.236±0.01	53.85±5.02	683.24±59.96	336.12±122.62
	DOT 5	1.29±0.29	0.33±0.05	59.39±16.08	357.79±217.09	214.63±20.57
	DOT 10	1.4±0.21	0.16±0.03	58.38±7.16	985.59±99.2	379.43±27.87
	DOT 15	1.62±0.103	0.31±0.01	67.95±8.40	900.13±176.69	327.36±61.25
Mean		1.365	0.258	59.893	731.625	314.35
SE		0.099	0.039	2.94	139.94	35.14
% Change		-2.85	-3.84	-17.38	+28.93	+16.4
P value		NS	< 0.05	NS	< 0.05	NS

Chlorophyll decline is more pronounced in plants grown under elevated CO_2 levels, which is due to the dilution of chlorophyll and degradation by excess utilisation under higher levels of CO_2 [18]. [19] also reported decrease in chlorophyll concentration in *Quercus suber* at elevated CO_2, which is explained by the dilution effect caused by the accumulation of starch. Chlorophyll reduction at elevated CO_2 can also be related to the diminution in nitrogen uptake [20]. According to [21] under elevated CO_2 conditions, increased plant nutrients, water, and light efficiency helped in the alleviation of stress, which in turn results in the down regulation of carotenoid biosynthesis. The decline in carotenoid content in the present study also might be a stress response.

In *S. glauca*, Carbohydrate content increased by 0.95% in treated compared to control. [22] reported higher carbohydrate content under elevated CO_2 levels in *Solanumly copersicum, Gossipium hirsutum*, *Raphanus sativus*, *Elaeis guineensis* and *Glycine max*. Carbohydrate in *S. cumini* decreased by 17.38% in treated compared to control. A study conducted in *Melia dubia* by [15] showed a reduction in the amount of protein, since the plants lost the ability to take up soil nitrate and convert it to protein at enriched levels of CO_2. The reduction was due to significant leaf N- re allocation to supplemental sinks [23]. Reduced protein under

CO_2 enrichment was also reported by [24] which is in line with the results of the present study, where the protein content decreased by 7.6% under enriched CO_2 in *S. glauca* whereas in *S. cumini* protein content increased by 28.93% in treated compared to control. The results of protein content of *S. cumini* are in accordance with the study of [25] and the increase in protein content is due to the increase in nitrogen content present in soil.
Phenol content of *S. glauca* decreased by 7.5% in treated chamber, with respect to control chamber whereas an increase of 16.4% was observed under treated in *S. cumini*. Increased phenol content under elevated CO_2 was reported by [26] in *Zingiber officinale*.

Effect of minerals under CO_2 treatment in S. glauca and S. cumini

Variations in minerals under elevated CO_2 is depicted in table VII.

Table VII Variations In Minerals Under Elevated CO_2

		Minerals (%)							
		Simarouba glauca				*Syzigium cumini*			
	Days	Calcium	Magnesium	Sodium	Potassium	Calcium	Magnesium	Sodium	Potassium
Control	DOT1	0.84±0.20	0.51±0.16	0.15±0.12	0.48±0.05	0.88±0.11	0.29±0.1	0.01±0.16	0.05±0.13
	DOT5	1.28±0.32	0.16±0.05	0.22±0.15	0.47±0.1	0.86±0	0.3±0	0.09±0.18	0.06±0.1
	DOT10	1.01±0.18	0.32±0.20	0.22±0.15	0.056±0.2	0.8±0	0.58±0.14	0.11±0.18	0.05±0.02
	DOT15	1.28	1±0.42	0.12±0.09	0.05±0.2	0.96±0.22	0.54±0.07	0.11±0.11	0.89±0.14
Mean		1.1	0.5	0.18	0.26	0.875	0.4275	0.08	0.2625
SE		0.11	0.18	0.03	0.12	0.03	0.08	0.02	0.21
Treated	DOT 1	1.60±0.87	0.92±1.39	0.37±0.06	0.518±0.02	0.72±0.11	0.34±0.07	0.1±0.14	0.02±0.18
	DOT 5	1.17±0.09	0.22±0.05	0.22±0.14	0.04±0.03	0.75±0	0.39±0	0.14±0.14	0.04±0
	DOT10	1.06±0.49	0.58±0.54	0.24±0.14	0.05±0.1	0.8±0	0.19±0	0.19±0.16	0.056±0.15
	DOT15	1.36±0.11	1.05±0.07	0.13±0.1	0.07±0.20	0.8± 0	0.63±0.21	0.1±0.14	0.02±0.12
Mean		1.3	0.69	0.24	0.17	0.7675	0.3875	0.165	0.0325
SE		0.12	0.19	0.05	0.11	0.02	0.09	0.02	0.01
% change		18.18	38	33.3	-52.9	-12.64	-9.52	100	-88.46
P value		NS	NS	NS	NS	NS	NS	NS	NS

In *S. glauca***,** calcium and magnesium content increased under CO_2 treated, compared to control by18.18% and 38.18% respectively which is in agreement with the study of [27] where calcium and magnesium increased in bamboo species under excess CO_2 whereas in *S. cumini*, calcium and magnesium content decreased by 12.64% and 9.52% respectively. Sodium increased under treated compared to control by 33.3% in *S. glauca* and 100% in *S. cumini*, which is in accordance with the

study conducted in bamboo by [27]. Potassium decreased by 52.9% and 88.46 % in *S. glauca* and *S. cumini* respectively. Potassium and magnesium are essential plant nutrients that critically contribute to photosynthesis and long-distance transport of photo assimilates. Deficiency of either potassium or magnesium decreases the CO_2 assimilation. From the study of [28] it was found that cotton

grown under elevated CO_2 was more susceptible to potassium deficiency and it affected the photosynthesis of plants.

Table VIIII depicts the % change in carbon, nitrogen and C/N ratios of *S. glauca* and *S. cumini.* In *S. glauca* carbon concentration increased by 1.2 % in treated and nitrogen concentration decreased by 18.9 % in treated. C/N ratio decreased by 19.05 % in control whereas in treated it increased by 24.83 %. In *S. cumini* carbon and nitrogen concentration decreased by 33.9% and 1.58% respectively in treated. C/N ratio increased by 74.14 % in control while in treated it decreased by1.84 %. [29] reported increase in the C/N ratio of plant tissue. Dilution of nitrogen concentration by increased non-structural carbohydrates and increased carbon based metabolic products lead to the increase in C/N ratio of plants [30].

Table VIII: carbon, nitrogen and C/N ratio of plants (%) in control and elevated CO_2.

	Simarouba glauca						*Syzigium cumini*					
	Control			Treatment			Control			Treatment		

Statistical analysis

The decrease in total chlorophyll, carotenoids, protein and phenol in *S. glauca* after the exposure of CO_2 is observed in treated compared to control. From the results of statistical analysis (table 6) it is proved that the decrease in protein is significant. In *S. cumini* protein and phenol content increased under enhanced CO_2. The increase in protein content is statistically significant which is represented as $P<0.05$. Chlorophyll, carotenoid and carbohydrate content of *S. cumini* decreased in treated compared to control. The decrease in Carotenoid content of *S. cumini* is also statistically significant which was represented as $p < 0.05$ (Table 6).

Conclusion

The percentage day flux of CO_2 inside the treated and control chamber with *S. glauca* is higher than *S. cumini* which is higher than the standardisation study. The day flux of CO_2 in the treated chamber can be ascribed to the plants growing within it and is confirmed from the results of the standardisation experiment. However, in standardisation studies the percentage night flux of CO_2 inside the control chamber was higher than treated whereas in the treated chambers with *S. glauca* and *S. cumini* the percentage night flux of CO_2 is higher than in control indicating higher respiratory release of plants to the treated chamber. Uptake and assimilation of CO_2 by *S. glauca* is apparent from the increased morphological attributes of growth like plant height, stem thickness, leaf area, carbohydrate, mineral content. Similarly in *S. cumini* increased protein and sodium, are observed as a result of CO_2 uptake by the plant. Increased phenol content under elevated CO_2 are indications of stress to which *S. cumini* are subjected to under higher levels of CO_2 supply. The results indicate that *S. glauca* is expected to acclimatize under elevated CO_2 concentrations and adapts better than *S. cumini* suggesting good indication of establishment of *S. glauca* in potentially changed climatic condition.

References

1. Sharma, R., Pradhan, L., Kumari, M., & Bhattacharya, P. 2020. Assessment of Carbon Sequestration Potential of Tree Species in Amity University Campus

Noida. In *Environmental Sciences Proceedings* (Vol. 3, No. 1, p. 52). Multidisciplinary Digital Publishing Institute.

2. Superales, J. B. 2016. Carbon Dioxide Capture and Storage Potential of Mahogany (Swietenia macrophylla) Saplings. *International Journal of Environmental Science and Development*, *7*(8): 611.
3. Lamani, N., Shivaprasad, D., & Swamy, K. R. 2016. Effect of elevated carbon dioxide concentration on photosynthetic and transpiration rate in Sandal (*Santalum album* L.) *Proceedings of the International Academy of Ecology and Environmental Sciences*, *6*(2), 44-62.
4. Dusenge, M. E., Duarte, A. G., & Way, D. A. 2019. Plant carbon metabolism and climate change: elevated CO_2 and temperature impacts on photosynthesis, photorespiration and respiration. *New Phytologist*, *221*(1): 32-49.
5. Shoaf, W. T., and Lium, B. W. 1976. Improved extraction of chlorophyll a and b from algae using dimethyl sulfoxide. Limnology and Oceanography, *21*(6): 926-928.
6. Dubois, M., Gilles, K. A., Hamilton, J. K., Rebers, P. T., and Smith, F. 1956. Colorimetric method for determination of sugars and related substances. Analytical chemistry, *28*, 350-356.
7. Malick, C. P., and Singh, M. B. 1980. Estimation of total phenols in plant enzymology and histoenzymology. Plant enzymology and histoenzymology*:* A text manual. Kalyani Publishers. New Delhi.
8. Lowry, O. H., Rosebrough, N. J., Farr, A. L., and Randall, R. J. 1951. Protein measurement with the Folin phenol reagent. J. of biological chemistry, 193, 265-275.ncbi.nlm.nih.gov/14907713
9. Lahive, F., Hadley, P., and Daymond, A. J. 2018. The impact of elevated CO_2 and water deficit stress on growth and photosynthesis of juvenile cacao (Theobroma cacao L.) Photosynthetica, *56*, 911-920.10.1007/s11099-017-0743-y
10. Wang, X., Lewis, J. D., Tissue, D. T., Seemann, J. R. and Griffin, K. L. 2001. Effects of elevated atmospheric CO2 concentration on leaf dark respiration of Xanthium strumarium in light and in darkness. *Proceedings of the National Academy of Sciences*. *98*(5) :2479-2484.
11. Elvira, A. I. and Vasenev, I. I. 2020. Changes in CO_2 Flux Conditions in the Spruce Forest of Various Ages in the Central Forest Reserve of Russia. *European Journal of Molecular & Clinical Medicine, 7*(7): 693-704.
12. Ruiz-Vera, U. M., De Souza, A. P., Ament, M. R., Gleadow, R. M., and Ort, D. R. 2021. High sink strength prevents photosynthetic down-regulation in cassava grown at elevated CO_2 concentration. J. of experimental botany, *72*: 542-560.
13. Pal, M., Karthikeyapandian, V., Jain, V., Srivastava, A. C., Raj, A and Sengupta, U. K. 2004. Biomass production and Nutr.al levels of berseem (*Trifolium alexandrium*) grown under elevated CO_2. Agric. Ecosystems and Env, 101: 31-38.
14. Taylor, G., Ranasinghe, S., Bosac, C., Gardner, S. D. L., and Ferris, R. 1994. Elevated CO_2 and plant growth: cellular mechanisms and responses of whole plants. J. of Experimental Botany, 1761-1774.
15. Janani, S., Priyadharshini, P., Jayaraj, R. S. C., Buvaneswaran, C. and Warrier, R. R. 2016.Growth, physiological and biochemical responses of

Meliaceae species-Azadirachta indica and *Melia dubia* to elevated CO_2 concentrations. *J. of Appl Biology & Biotechnology. 4*: 052-060.

16. Al-Rawahy, S. H., Sulaiman, H., Farooq, S. A., Karam, M. F., & Sherwani, N. 2013. Effect of O3 and CO2 levels on growth, biochemical and nutrient parameters of alfalfa (Medicago Sativa). *APCBEE procedia*, *5*, 288-295.
17. Jeong, H. M., Kim, H. R., Hong, S., and You, Y. H. 2018. Effects of elevated CO_2 concentration and increased temperature on leaf Qual. responses of rare and endangered plants. J. of Ecol and Env, *42*, 1-11.
18. Kumari, M., Verma, S. C., and Bhardwaj, S. K. 2019. Effect of elevated CO_2 and temperature on crop growth and yield attributes of bell pepper (Capsicum annuum L.). J. of Agrometeorology, *21*, 1-6.
19. Faria, T., Wilkins, D., Besford, R. T., Vaz, M., Pereira, J. S., and Chaves, M. M. 1996. Growth at elevated CO_2 leads to down-regulation of photosynthesis and altered response to high temperature in Quercus suber L. seedlings. J. of Experimental Botany, *47*, 1755-1761.doi: 10.1093/jxb/47.11.1755.
20. Delucia, E. H., Sasek, T. W., and Strain, B. R. 1985. Photosynthetic inhibition after long-term exposure to elevated levels of Atoms. carbon dioxide. Photosynthesis Res, *7*, 175-184.
21. Loladze, I., Nolan, J. M., Ziska, L. H., and Knobbe, A. R. 2019. Rising Atoms. CO_2 Lower Concentrations of Plant Carotenoids Essential to Human Health: A Meta-Analysis. Molecular Nutr. and food Res, *63*, 1801047.
22. Ibrahim, M. H., Nulit, R., Wahab, P. E. M., Najihah, T. S., Razak, A., and Zain, N. A. M. 2018. Impacts of Elevated CO_2 on Growth, Carbohydrate Assimilation and Nutrient Uptake of Elaeisguineesis Jacq Seedlings. *Annual Res., and Review in Biol,* 1-9
23. Norby, R. J., Wullschleger, S. D. and Gunderson, C. A. 1996. Tree responses to elevated CO2 and implications for forests. *Carbon dioxide and terrestrial ecosystems.* 1-21.
24. Dong, J., Gruda, N., Lam, S. K., Li, X., and Duan, Z. 2018. Effects of elevated CO2 on Nutr.al Qual. of vegetables: a review. *Frontiers in plant Sci*, *9*, 924.
25. Ainsworth, E. A., Davey, P. A., Bernacchi, C. J., Dermody, O. C., Heaton, E. A., Moore, D. J. and Long, S. P. 2002. A meta-analysis of elevated CO_2 effects on soybean (Glycine max) Physiol. growth and yield. Global change Biol, *8*, 695-709.
26. Ghasemzadeh, A., Jaafar, H. Z., and Rahmat, A. 2010. Elevated carbon dioxide increases contents of flavonoids and phenolic compounds, and antioxidant activities in Malaysian young ginger (Zingiber officinale Roscoe.) varieties. *Molecules*, *15*(11), 7907-7922.
27. Guo, Z., Zhuang, M., Yang, L., Li, Y., Wu, S., and Chen, S. 2021. Differentiated mineral nutrient management in two bamboo species under elevated CO_2 Env. *J. of Env. Management*, *279*, 111600.
28. Reddy, K. R., and Zhao, D. 2005. Interactive effects of elevated CO2 and potassium deficiency on photosynthesis, growth, and biomass partitioning of cotton. *Field Crops Res.,*, *94*(2-3), 201-213.
29. Lotfiomran, N., Köhl, M., & Fromm, J. (2016). Interaction effect between elevated CO2 and fertilization on biomass, gas exchange and C/N ratio of European beech (Fagus sylvatica L.). *Plants*, *5*(3), 38.
30. Gifford, R. M., Barrett, D. J., & Lutze, J. L. (2000). The effects of elevated [CO2] on the C: N and C: P mass ratios of plant tissues. *Plant and Soil*, *224*(1), 1-14.