

## Effect of elevated carbon dioxide concentration and relative humidity on the growth of forest tree seedlings

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### ABSTRACT

Crop plants are shown to have high photosynthetic rate at elevated CO<sub>2</sub> concentrations. Experiments were conducted with an objective to study the influence of elevated CO<sub>2</sub> concentration and relative humidity (RH) on seedling growth of *Eucalyptus citriodora*, *Derris indica*, *Spathodea companulata* and *Acacia auriculiformis*. A simple cost effective technique was developed and standardised to increase the CO<sub>2</sub> concentration and RH around the seedlings in a forest nursery. Increase in the CO<sub>2</sub> concentration to about 700-800  $\mu\text{mol mol}^{-1}$  during the night and only a few hours during the day increased the biomass of all the species tested. Growth response to elevated CO<sub>2</sub> and RH was more in *Derris* and *Spathodea* than in the other two species examined. In all the species gas exchange studies showed increased photosynthesis with instantaneous increase in CO<sub>2</sub> concentration. High growth in *Acacia* was associated with better leaf area development whereas it was due to better photosynthetic efficiency in *Eucalyptus*. The experiment proved that the technique can be effectively used to enhance seedling growth in forest

**Key words:** Elevated CO<sub>2</sub>, Relative humidity, biomass, tree seedling growth.

### INTRODUCTION

There has been a considerable awareness about the population growth, deforestation and global warming, and afforestation programmes are assuming great importance, especially in tropical countries.

For large scale afforestation programmes, availability of seedlings is often a major constraint. In general, under normal conditions, to obtain tall and healthy transplantable seedlings of woody species, they have to be maintained in forest nurseries for long periods because of very low growth rates.

Plant growth and development depends on its genetic makeup as well as on environmental factors such as light, temperature, relative humidity, soil nutrient and water status and CO<sub>2</sub> concentration. Further, the total biomass production is mainly governed by photosynthesis. One of the major environmental factors that affects the photosynthesis and thus the biomass production is the ambient CO<sub>2</sub> concentration. It is possible to increase the biomass production by CO<sub>2</sub> fertilization (Nataraja 1991, Bowes 1993). Sasek and Strain (1991) observed 76 and 135 per cent increase in biomass in Japanese honey suckle vine at 675 and 1000  $\mu\text{mol mol}^{-1}$  of

CO<sub>2</sub> concentration, respectively. Similarly, doubling of biomass was also reported in sweet oranges at elevated CO<sub>2</sub> levels (Idso *et al.* 1991). In general, growth increment under elevated CO<sub>2</sub> will be maximum in C<sub>3</sub> plants compared to C<sub>4</sub> (Bazza 1990, Bowes 1993). Projected high CO<sub>2</sub> concentration in the atmosphere is likely to increase the productivity of forest ecosystems (Thornley and Cannell 1996).

The increase of biomass under elevated CO<sub>2</sub> concentration could be due to several reasons such as increased photosynthetic surface area and photosynthesis, reduction in photorespiration or dark respiration rates, etc. In the present study, we report the effect of elevated CO<sub>2</sub> concentration and relative humidity (RH) on single leaf photosynthesis and seedling growth in a few forest tree species. Since the methodology used in this study is simple, it can be easily adapted in forest nurseries to enhance seedling growth rates.

### MATERIALS AND METHODS

#### Plant material

Seedlings of *Eucalyptus citriodora*, *Derris indica*, *Spathodea companulata* and *Acacia auriculiformis* were grown in polythene bags containing sand, red soil and farm yard manure in 1:1:0.5 ratio, by weight. The plants were watered twice a day to maintain the soil water status at field capacity.

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### Method of CO<sub>2</sub> and Relative humidity (RH) enrichment

Four meter long trenches of 1m depth and 0.6m width were dug and decomposing organic matter was added at the rate of 13-16 kg to each trench and spread uniformly. Six months old healthy seedlings were selected and transferred to the trenches. To enhance RH and CO<sub>2</sub> levels around the seedlings, the trenches were covered with transparent polythene film (125 $\mu$  thick and with 80-85% transmission of the incident light), from 16.30 hrs. in the evening to 10.30 hrs. the next day. The polythene house was supported by a metal frame with 4.2 meter x 1.2 meter x 2 meter length, width and height respectively. Before closing the trench, water was sprinkled on the decomposing organic matter to stimulate soil microbial activity. The trenches were made air tight by sealing the free ends of the covers in to the soil. By this method CO<sub>2</sub> released by dark respiration of the seedlings and by the decomposing organic matter was trapped and made available to the plants for photosynthesis during morning hours between 08.30 - 10.30 hrs. There was simultaneous increase in RH (>90%) inside the covered trenches. Uncovered trenches acted as control. Fifteen seedlings were maintained in each treatment for 90 days.

The CO<sub>2</sub> concentration and RH inside trenches were monitored during the experimental period using portable infra-red gas analyzer (LCA-2, ADC, London, UK) by drawing air from the trenches at different intervals.

Before initiation of the treatments, ten plants from each species were harvested for recording initial growth. At harvest, plants were divided into stems, leaves and roots, and dry weight was recorded after drying to a constant weight at 80°C. Leaf area was recorded using a leaf area meter (Li-3000, Li-COR Inc., Nebraska, USA).

### Measurement of photosynthesis

Photosynthetic rates at ambient and elevated CO<sub>2</sub> concentrations were measured using portable photosynthetic system (ADC, LCA-2, London, UK) on youngest fully expanded leaves at saturating light intensities (1800  $\mu$ Ein m<sup>-2</sup> s<sup>-1</sup>). The leaf-air vapour pressure deficit was maintained around 2 kPa by regulating the air flow rates through the leaf chamber. Air enriched with CO<sub>2</sub> was generated by adapting the methodology developed by Sheshshayee *et al.* (1992). Each leaf was allowed to stabilise for 5-8 minutes before measuring the steady state of CO<sub>2</sub> uptake. Twelve measurements were

made on each species.

Collected data were subjected to statistical analysis and significant differences were tested using an analysis of variance (ANOVA) procedure.

## RESULTS

The CO<sub>2</sub> concentration inside the covered trench increased linearly from 18.00 hrs. to 06.00 hrs. and it was around 750 $\pm$ 55  $\mu$ mol mol<sup>-1</sup> at 06.00 hrs. In addition to this, there was an increase in RH also (>90%). With the increase in light intensity in the morning hours from 7.30 to 10.30 hrs., a steady reduction in CO<sub>2</sub> concentration inside the covered trenches was observed and it reached ambient CO<sub>2</sub> levels at around 10.30 hrs.

### Growth parameters

Plant height at the end of the experiment showed significant increase due to CO<sub>2</sub> and RH enrichment. Exposure of plants to elevated CO<sub>2</sub> and RH resulted in 39 per cent increase in plant height compared to the plants grown at ambient CO<sub>2</sub> levels (Table 1). In *D. indica* the response was very marked. High CO<sub>2</sub> and RH treatment increased total plant leaf area also.

Table 1: Effects of elevated CO<sub>2</sub> concentration and relative humidity on plant height, leaf number and leaf area of seedlings in four tree species after 90 days. Each value is the mean of 12 seedlings.

Species	Plant height, cm		Leaf number, plant <sup>-1</sup>		Leaf area, cm <sup>2</sup> plant <sup>-1</sup>	
	Control	Elevated CO <sub>2</sub> and RH	Control	Elevated CO <sub>2</sub> and RH	Control	Elevated CO <sub>2</sub> and RH
<i>A. auriculiformis</i>	63.72	75.03	36	47	226	615
<i>D. indica</i>	25.40	47.97	10	17	449	1451
<i>E. citriodora</i>	69.00	84.47	30	50	509	745
<i>S. samanulata</i>	29.20	36.06	11	15	357	741
LSD <sub>0.05</sub> (Treatment)	12.4		8.6		108	

Table 2: Effects of elevated CO<sub>2</sub> concentration and relative humidity on leaf biomass, total biomass and root/shoot ratio in seedlings of four tree species after 90 days. Each value is the mean of 12 seedlings.

Species	Leaf biomass, g plant <sup>-1</sup>		Total biomass, g plant <sup>-1</sup>		Root/Shoot ratio	
	Control	Elevated CO <sub>2</sub> & RH	Control	Elevated CO <sub>2</sub> & RH	Control	Elevated CO <sub>2</sub> & RH
<i>Acacia</i>	3.56	7.80	16.88	26.36	0.943	0.727
<i>Derris</i>	2.50	9.52	7.67	26.16	0.641	0.559
<i>Eucalyptus</i>	4.58	8.54	14.91	28.13	0.615	0.636
<i>Spathodea</i>	2.92	6.04	12.73	27.38	0.923	0.828
LSD <sub>0.05</sub> (Treatment)	1.24		2.33		N <sub>s</sub>	

Among the four species studied, *D. indica* showed 223% increase in leaf area under elevated CO<sub>2</sub> and RH treatment. In the other species the increase ranged from 46 to 173 per cent (Table 1, Fig. 1a).

Exposing the plants to elevated CO<sub>2</sub> and RH resulted in a significant increase in total biomass in all the four species. In *D. indica* there was a four fold increase in biomass due to CO<sub>2</sub> and RH enrichment, and in the other species it ranged from one and half to two folds (Table 2). *Acacia*, *Derris* and *Spathodea* showed a marginal and statistically non-significant reduction in root to shoot ratio under high CO<sub>2</sub> and RH treatment (Table 2).

Table 3. Net Photosynthetic rates ( $\mu\text{mol m}^{-2} \text{s}^{-1}$ ) measured at 330 and 660  $\mu\text{mol mol}^{-1}$  reference  $\text{CO}_2$  concentration in seedlings of four tree species. Measurements were made on the uppermost fully expanded leaf under saturated light intensity (n=15 for *Eucalyptus* and n=12 for other species).

Species	Net Photosynthetic rate	
	330 $\mu\text{mol mol}^{-1}$	660 $\mu\text{mol mol}^{-1}$
<i>Acacia</i>	12.0	21.5
<i>Derris</i>	10.8	17.3
<i>Eucalyptus</i>	15.5	24.5
<i>Spathodea</i>	7.1	13.4
LSD <sub>05</sub> (Treatment)	4.9	

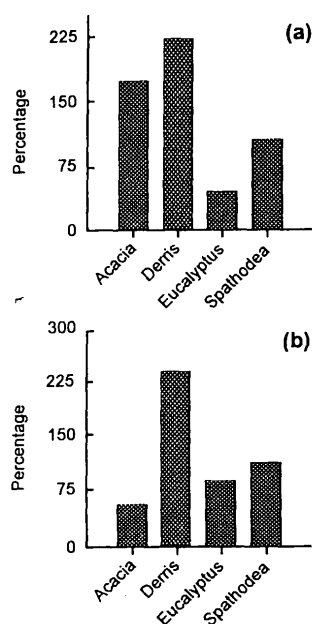


Fig.1: Percent increase in leaf area (a) and total biomass (b) in different tree seedlings grown under elevated  $\text{CO}_2$  concentration and relative humidity for 90 days

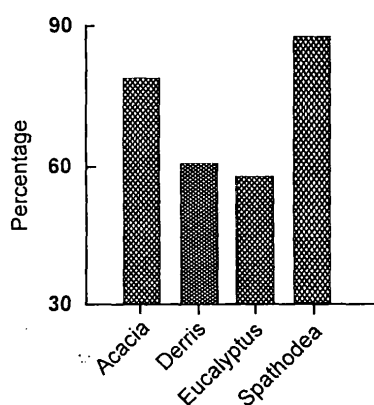


Fig. 2. Percent increase in photosynthesis when measured at elevated  $\text{CO}_2$  concentration of 660  $\mu\text{mol mol}^{-1}$  in four different tree seedlings.

## Photosynthesis

*Eucalyptus* had high rates of photosynthesis and *Spathodea* showed the least (Table 3) when

measured at ambient  $\text{CO}_2$  concentration. There was a significant increase in photosynthesis with increase in  $\text{CO}_2$  concentration. The percent increase in photosynthesis at elevated  $\text{CO}_2$  concentration ( $\text{CO}_2$  sensitivity) were to an extent of 79, 88, 58 and 60 in *Acacia*, *Spathodea*, *Eucalyptus* and *Derris* respectively (Fig.2).

## DISCUSSION

The results of the experiment conducted to study the influence of elevated  $\text{CO}_2$  concentration and RH during night and for a brief period during morning hours on growth of tree species seedlings showed significant increase in leaf area and total biomass accumulation (Fig. 1a&b). Increase in  $\text{CO}_2$  concentration alone was shown to increase leaf area and biomass in a number of plant species including tree species (Baker *et al.* 1990; Sasek and Strain 1991, Idso *et al.* 1991; Idso and Kimball 1991, Tissue *et al.* 1993, Lewis *et al.* 1996, Roden and Ball 1996). In *Pinus radiatus* increased wood production was also reported (Conroy *et al.* 1990). In some cases increased biomass production was associated with increased branching in woody species (Sionit *et al.* 1985). In this study also, *Acacia*, *Eucalyptus* and *Spathodea* showed substantial increase in branch number at elevated  $\text{CO}_2$  concentration and RH (data not shown).

In *D. indica* photosynthetic surface area was relatively more at elevated  $\text{CO}_2$  concentration and RH compared to the other species examined. The faster leaf area development probably enhanced the canopy net carbon gain under elevated  $\text{CO}_2$  concentration and RH environment. Therefore the per cent increase in biomass accumulation was more in *D. indica* than the other species examined. Even though *Eucalyptus* showed only a small per cent increase in leaf area (Fig. 1a), a good biomass accumulation was noticed (Fig.1b). This suggests that in *Eucalyptus*, better net carbon fixation ability resulted in high biomass under elevated  $\text{CO}_2$  concentration and RH treatment. In fact, *Eucalyptus* had significantly high photosynthetic rate when measured either at ambient or elevated  $\text{CO}_2$  concentration (Table 3). Similar to *Derris*, *Acacia* also showed enhanced leaf area but it did not result in high biomass production. Higher leaf area development under elevated  $\text{CO}_2$  concentration and RH environments may be due the direct effect of RH which helps in maintaining cell turgidity and thus helps optimum cell metabolic activity and cell expansion.

## CONCLUSION

The findings of the present investigation indicate that a short term exposure of tree species seedlings to elevated CO<sub>2</sub> concentration and RH can boost the growth. High RH helps in maintaining cell turgidity and hence helps in leaf expansion as indicated by the increased leaf area. Species variation in their response to elevated CO<sub>2</sub> concentration and RH was observed. The technique used in the present investigation for growing seedlings under elevated CO<sub>2</sub> concentration and RH is simple and cost effective and therefore, can be easily adapted in forest nurseries. Rapid initial growth in nursery can reduce the nursery duration substantially.

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