

Growth and photosynthetic characteristics of *Ceriops roxburghiana* under NaCl stress

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Abstract

The plant growth, net photosynthetic rate (P_N), intercellular CO_2 concentration (c_i), and dry matter production of *Ceriops roxburghiana* Arn. were significantly increased with increasing salinity from 0 to 400 mM NaCl. At 600 mM NaCl, shoot and root lengths, and dry mass were significantly depressed with respect to control. Absence of diurnal fluctuation of concentrations of organic acids, and the low activity of phosphoenolpyruvate carboxylase and high activity of ribulose-1,5-bisphosphate carboxylase confirmed the operation of C_3 pathway in *Ceriops* even at increasing salinity.

Additional key words: carbon assimilation pathway; chlorophyll; intercellular CO_2 concentration; phosphoenolpyruvate carboxylase; ribulose-1,5-bisphosphate carboxylase; salinity.

Mangroves on the coastal region represent fragile ecosystems. They are important, because they possess enormous biodiversity and provide a biologically most productive habitat. Salinity is the main stressor and regulator of the development of mangrove forests. Among the various metabolic processes influenced by salt stress, photosynthesis merits priority attention as it is a key to plant productivity. P_N of many plant species declines with increasing rhizosphere salinity. C_3 photosynthetic pathway is typical for mangrove species. However, little is known as to how salinity may influence the plant's distribution in inland and saline areas through an effect on their photosynthesis. The present study therefore evaluates growth and photosynthetic performance of *C. roxburghiana* under different concentrations of NaCl.

Young seedlings of *C. roxburghiana* were collected from the mangrove forest of Pichavaram which is located at the southeast coast of India (11°24'N and 79°44'E), about 16 km east of Annamalai University campus, spread to an area of 1100 ha. Seedlings in polyethene sleeves containing homogeneous mixture of garden soil

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were treated with 0-1000 mM NaCl. The seedlings treated with NaCl above 600 mM did not survive. Samplings were randomly collected on the 90th d after salt treatment. Root and shoot lengths were measured. Leaves, stems, and roots were separated and fresh and dry (80 °C, 48 h) masses were determined.

The chlorophyll (Chl) content was determined spectrophotometrically according to Arnon (1949). P_N and c_i were measured using a portable infrared gas analyser (LCA 2, ADC, UK) at temperature of 30±2 °C, CO₂ concentration of 350 mg m⁻³, irradiance of 450±25 W m⁻², and relative humidity of 60±10 %. Phosphoenolpyruvate carboxylase (PEPC; EC.4.1.1.31) and ribulose-1,5-bisphosphate carboxylase (RuBPC; EC.4.1.1.39) activities were assayed by the methods of Sadasivam and Gowri (1981). The enzymes' extraction was achieved by crushing 1 g of lamina with 10 amounts (m/v) of extraction medium which contained 50 mM Tris-HCl (pH 7.5), 2 mM MgCl₂, 1 mM EDTA, 40 mM mercaptoethanol, 1.5 % polyvinylpyrrolidone, and 10 mM sodium metabisulphite. PEPC was assayed spectrophotometrically, where the assay mixture contained 0.1 M Tris HCl (pH 7.8), 0.4 µM NADH, 5 mM phosphoenolpyruvate, and 10 mM sodium bicarbonate; the activity was measured by the oxidation of NADH at 340 nm. RuBPC was assayed radiometrically using a scintillation counter, where the assay mixture contained 0.25 mM HEPES buffer (pH 7.8), 1 mM RuBP, 100 mM MgCl₂, 50 mM dithiothreitol, and 10 µm³ of NaH¹⁴CO₃ (specific activity 37 GBq mol⁻¹). Specific activity is expressed as the amount of enzyme required to fix 1 nmol(CO₂) s⁻¹ kg⁻¹ (protein) under the experimental conditions. Titratable acidity and stomatal aperture were determined according to Spalding and Edwards (1978) and Raghavendra and Das (1972), respectively.

The NaCl treatment enhanced the growth of *Ceriops* up to 400 mM concentration by increasing the fresh mass, dry mass, shoot and root length, leaf succulence, leaf area, P_N , c_i , and Chl content (Table 1). However, these characteristics were lower beyond 400 mM NaCl, and at 600 mM NaCl shoot and root lengths, and dry mass were lower than in control (0 mM). Similar changes in growth and biomass production have been reported in *Atriplex patula* (Ungar 1996) and some other halophytes (Adam 1990, Ungar 1991). Similar observations for Chl content were reported, *e.g.*, in *Sesuvium portulacastrum* (Venkatesalu and Chellapan 1993), *Salicornia brachiata* (Reddy *et al.* 1993), and *Ipomoea pescaprae* (Venkatesan *et al.* 1995). In non-halophytes, toxic concentrations of NaCl are much lower (see, *e.g.*, Rajasekaran *et al.* 1997).

At external NaCl concentration of 600 mM, the sodium concentration in the leaves was 16.1 g kg⁻¹(d.m.). This concentration lowered both growth and gas exchange. Although, the NaCl treatments beyond 400 mM decreased P_N in relation to those of 300 and 400 mM, there was no change in photosynthetic pathway. The opening of stomata during day, their closure during night, and the absence of diurnal fluctuation of organic acids (values not presented) excludes the possibility of this species being a CAM plant. Moreover, the determined enzyme activities confirmed the C₃ pathway of photosynthesis in *Ceriops*. Low activities of PEPC and high activity of RuBPC (Table 1) indicated the operation of C₃ carbon pathway.

Table 1. Effect of NaCl concentration on shoot or root length [cm plant⁻¹], fresh or dry mass [g plant⁻¹], leaf area [cm plant⁻¹], net photosynthetic rate, P_N [μmol(CO₂) m⁻² s⁻¹], intercellular CO₂ concentration, c_i [μmol mol⁻¹], chlorophyll content, Chl (a+b) [g kg⁻¹(d.m.)], and activities of carboxylases, phosphoenolpyruvate carboxylase (PEPC) and ribulose-1,5-bisphosphate carboxylase (RuBPC) [mmol(CO₂) kg⁻¹(protein) s⁻¹] in *C. roxburghiana*. Samples were taken on the 90th d after treatment. Values are means of 10 (growth) or 5 (photosynthetic characteristics) samples. CD for $p=0.05$.

	NaCl [mM]						CD
	0	100	200	300	400	500	
Shoot length	15.7	17.2	17.7	17.9	18.2	16.5	15.2 0.160
Root length	13.1	16.8	17.1	17.3	17.8	13.3	12.1 0.447
Fresh mass	5.87	7.02	7.45	8.96	8.26	7.77	7.19 0.056
Dry mass	3.25	3.34	3.42	3.60	3.71	3.24	2.10 0.018
Leaf area	8.85	10.59	13.58	19.18	21.77	14.51	9.90 0.355
P_N	6.6	7.2	7.8	8.2	9.1	7.4	6.9 0.133
c_i	0.533	0.580	0.601	0.630	0.681	0.582	0.568 0.091
Chl (a+b)	2.91	3.42	3.74	4.33	4.64	4.13	3.74 0.109
PEPC	2.40	2.34	2.28	2.46	2.40	2.28	2.34 0.001 ^{NS}
RuBPC	7.80	8.40	8.40	7.80	9.00	7.20	7.20 0.002 ^{NS}

References

Adam, P.: Saltmarsh Ecology. - Cambridge University Press, New York 1990.

Arnon, D.I.: Copper enzymes in isolated chloroplasts. Polyphenoloxidase in *Beta vulgaris*. - Plant Physiol. **24**: 1-15, 1949.

Raghavendra, A.S., Das, V.S.R.: Control of stomatal opening by cyclic photophosphorylation. - Curr. Sci. **41**: 150-151, 1972.

Rajasekaran, L.R., Kriedemann, P.E., Aspinall, D., Paleg, L.G.: Physiological significance of proline and glycinebetaine. Maintaining photosynthesis during NaCl stress in wheat. - Photosynthetica **34**: 357-366, 1997.

Reddy, M.P., Sanish, S., Iyengar, E.R.R.: Compartmentation of ions and organic compounds in *Salicornia brachiata* Roxb. - Biol. Plant. **35**: 547-553, 1993.

Sadasivam, S., Gowri, G.: Enzymes. - In: Sadasivam, S., Manickam, A. (ed.): Biochemical Methods. Pp. 90-134. New Age Int. Publishers, New Delhi 1981.

Spalding, M.H., Edwards, G.E.: Photosynthesis in enzymatically isolated leaf cells from the CAM plant *Sedum telephium* L. - Planta **141**: 59-63, 1978.

Ungar, I.A.: Ecophysiology of Vascular Halophytes. - CRC Press, Boca Raton 1991.

Ungar, I.A.: Effect of salinity on seed germination, growth and ion accumulation of *Atriplex patula* (Chenopodiaceae). - Amer. J. Bot. **83**: 604-607, 1996.

Venkatesalu, V., Chellappan, K.P.: Photosynthetic characteristics of *Sesuvium portulacastrum* L. under salt stress. - Photosynthetica **28**: 313-316, 1993.

Venkatesan, A., Venkatesalu, V., Chellappan, K.P.: Photosynthetic characteristics of *Ipomoea pes-caprae* Forsk. under NaCl stress. - Photosynthetica **31**: 631-634, 1995.