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BRIEF COMMUNICATION

Influence of ultraviolet-B radiation on photosynthetic and biochemical characteristics of a mangrove *Rhizophora apiculata*

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Abstract

Changes in photosynthesis and biochemical constituents were studied in *R. apiculata* seedlings grown under solar and solar enhanced UV-B radiation, equivalent to 10, 20, 30, and 40 % stratospheric ozone depletion. The seedlings grown under 10 % UV-B radiation showed an increase of 45 % net photosynthetic rate (P_N) and 47 % stomatal conductance, while seedlings grown under 40 % UV-B radiation exhibited a decrease of 59 % P_N with simultaneous elevation of 73 % intercellular CO_2 concentration. Effects of UV-B on contents of lipids, saccharides, amino acids, and proteins were significant only at high doses of UV-B radiation. The concentration of anthocyanin was reduced with increasing doses of UV-B. The reverse was true with phenols and flavonoids.

Additional key words: anthocyanins; carotenoids; chlorophyll; flavonoids; intercellular CO_2 concentration; net photosynthetic rate; phenols; proteins; saccharides; stomatal conductance.

Ozone (O_3) is a principal gas of the atmosphere which absorbs solar UV-radiation at wavelengths shorter than 300 nm (Caldwell and Flint 1994). But, reduction of O_3 content due to anthropogenic influence has resulted in an increase of solar UV-B radiation (Caldwell *et al.* 1989). The UV-B has important photobiological effects, and the effects of solar enhanced UV-B radiation on lower and higher plants have been extensively reviewed (Caldwell 1971, 1977, 1991, Tevini and Teramura 1989,

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Murthy and Rajagopal 1995). The UV-B causes multitude effects on plant physiological responses, especially photosynthesis, targeting on the reaction centre of photosystem 2 (Noorudeen and Kulandaivelu 1982), carboxylating enzymes (Jordan *et al.* 1991), and polypeptides of thylakoid membranes (Nedunchezhian and Kulandaivelu 1991, Nedunchezhian *et al.* 1995). Most of the studies report only injurious effects of UV-B on plant growth and photosynthesis, but in some plants UV-B enhances photosynthesis (Beyschlag *et al.* 1988, Tezuka *et al.* 1993). Such studies are lacking in mangroves, a tropical coastal marine ecosystem; in marine ecosystems, studies of UV-B effects were primarily concerned with phytoplankton. Although mangrove growth and photosynthesis have been studied in several species, those in response to UV-B have not been done (Kathiresan and Moorthy 1993, 1994, Moorthy and Kathiresan 1993). The mangroves are prime target of the tropical coastline for the global climate changes and sea level rise. Therefore, we have quantified, in the present study for the first time, the changes in photosynthesis of *R. apiculata* grown under solar enhanced UV-B radiation.

Healthy propagules of *R. apiculata* L. Blume were collected from the Pichavaram mangrove forest (lat. 11°27'N; long. 79°47'E). Propagules 25 ± 2 cm long were used for UV-B treatment. The propagules were placed in 1000 cm^3 containers containing 500 cm^3 of Arnon-Hoagland (1940) nutrient medium with a salinity of 15 kg m^{-3}. The nutrient medium was replenished once in 3 d. After sprouting, the propagules were exposed to different UV-B irradiances. The propagules had only two leaves throughout the experimental period.

The UV-B radiation was supplied by *Philips* sun-lamps (*TL 12/20 W, Philips Glolampenfabriken*, The Netherlands). The sun-lamps were preburnt and matched for desired spectral irradiance (Teramura *et al.* 1980) prior to each treatment. Photosynthetically active radiation (400-700 nm) under the lamp was 90 %. The radiation was filtered through a 0.13 mm cellulose diacetate (pre-solarised) for transmission below 290 nm for treatment with supplemental UV-B radiation. The UV-B was checked using the generalised plant response action spectrum of Caldwell (1971) normalised at 300 nm. Therefore, the plants kept under UV lamps received supplemental doses of UV-B radiation in addition to ambient solar UV-B. The cumulative radiation (ambient + supplemental) was similar to that which would be received at the study site at Pichavaram mangrove (11°N) with 10, 20, 30, and 40 % depletion (marked further as UV-B_{10}, UV-B_{20}, UV-B_{30}, UV-B_{40}, respectively) of stratospheric ozone (Moorthy 1996) during a cloudless day on the summer solstice according to Green *et al.* (1980). Under such conditions the plants were irradiated for 6 h daily (10:00-16:00) for 14 d in an open field environment with the temperature at 28 ± 2 °C. Different UV-B irradiances were induced by altering the distance between the top of plants and the lamp frame. The required irradiance was monitored and maintained once in 7 d.

Pigments were extracted from two leaves and quantified for chlorophylls, Chl (Arnon 1949), carotenoids, Car (Ridley 1977), anthocyanins (Mancinelli 1983), flavonoids (Mirecki and Teramura 1984), and total phenols (Bray and Thorpe 1954). Net photosynthetic rate (P_N), stomatal conductance (g_s) and intercellular CO_2 concentration (C_i) were determined in intact leaves by using the *Li-Cor 6200* portable photosynthesis system. Leaves were analysed for total free amino acids (Moore and Stein 1948), saccharides (McCreedy *et al.* 1950), lipids (Bligh and Dyer 1959), and proteins (Lowry *et al.* 1950). Three replicates of 10 propagules each were analysed for each treatment. Different treatments were statistically analysed by the Student's *t*-test, and least significant differences (LSD) were calculated and defined either at 95 or 99 % confidence levels.

Table 1. UV-B induced changes in contents of chlorophylls (Chl) [g kg^{-1}(FM)] and carotenoids (Car) [mmol kg^{-1}(FM)], net photosynthetic rate (P_N) [μmol(CO$_2$) m^{-2} s^{-1}], stomatal conductance (g_s) [μmol m^{-2} s^{-1}], intercellular CO$_2$ concentration (C_i) [cm^3 m^{-3}], and relative contents of anthocyanin [A_{530}], flavonoids [A_{315}], and phenol [mg kg^{-1}(FM)] in *R. apiculata* seedlings. Values in parentheses are percent increase or decrease over control. *Significant at 5 % level.

	UV-B radiation [%]				
	0	10	20	30	40
Chl *a*	1.02±0.21	0.97±0.05 (-6)	1.00±0.06	0.99±0.08 (-3)	0.73±0.06 (-28)*
Chl *b*	0.46±0.07	0.46±0.08 (0)	0.37±0.02 (-18)	0.38±0.02 (-17)	0.36±0.05 (-21)*
Chl (*a*+*b*)	1.48±0.27	1.42±0.09 (-4)	1.32±0.14 (-11)	1.36±0.10 (-8)	1.09±0.11 (-26)*
Chl *a/b*	2.22±0.16	2.18±0.18 (-2)	2.68±0.04 (20)	2.63±0.15 (19)	2.06±0.15 (-7)
Car	0.28±0.05	0.36±0.02 (24)*	0.30±0.01 (7)	0.32±0.01 (13)	0.31±0.03 (9)
P_N	4.39±0.21	6.37±0.43 (45)*	4.73±0.33 (8)	3.30±0.45 (-25)	1.80±0.27 (-59)*
g_s	0.05±0.00	0.07±0.01 (47)	0.05±0.00 (0)	0.05±0.00 (8)	0.05±0.00 (17)
C_i	159±15	170±16 (7)	193 (21)	266±18 (67)*	275±22 (73)*
Anthocyanins	0.30±0.02	0.30±0.02 (0)	0.24±0.02 (-18)*	0.26±0.01 (-13)	0.25±0.03 (-16)*
Flavonoids	1.37±0.40	1.63±0.19 (19)	2.00±0.02 (47)*	2.22±0.36 (62)*	2.33±0.37 (71)*
Phenol	2.17±0.28	4.45±0.23 (105)*	4.49±0.15 (106)*	4.11±0.21 (89)	5.52±0.19 (153)*

UV-B treatments significantly reduced the concentrations of Chl (Table 1), but only the UV-B$_{40}$ effects (decline by about a quarter) were statistically significant. The Chl *a/b* ratio in seedlings was higher at UV-B$_{20}$ and UV-B$_{30}$ than at lower dose (Table 1). Several authors have already shown the reduction of Chls in many plant species following the treatment with both high (Sisson and Caldwell 1976, Brandle *et al.* 1977, Teramura *et al.* 1980, Petropoulou *et al.* 1995) or low (Vu *et al.* 1981) doses of UV. But an unaltered content of photosynthetic pigments in response to UV-B was also found (Sullivan and Teramura 1988, Ziska and Teramura 1992). Reduction in Chl content might be due to inhibition of biosynthesis or degradation of Chl and their precursors (Teramura 1983), accumulation of chlorophyllides *a* and *b* (Strid and Porra 1992), or inhibition of the *cab* gene, which codes for Chl proteins (Jordan *et al.* 1991). The content of Cars increased after UV-B treatments, but statistically significant was only the 24 % increase at UV-B$_{10}$ (Table 1). This is in accordance with the findings of Petropoulou *et al.* (1995) in *Pinus halepensis* and Kulandaivelu *et al.* (1989) in *Vigna sinensis*.

The P_N was significantly enhanced in response to UV-B$_{10}$ (Table 1) and significantly declined at UV-B$_{40}$. Daley *et al.* (1978) showed that a small flux of UV stimulated CO$_2$ fixation. Sisson and Caldwell (1976) also recorded increased P_N at early stages of growth of *Rumex patientia* in response to UV-B. Bornman and

Vogelmann (1991) determined an increase of 38 % P_N with a concomitant reduction of 13 % Chl content in UV-exposed *Brassica carinata*. Murali and Teramura (1985) suggested that photosynthetically active radiation might counteract the UV-induced inhibitory effects on photosynthesis. Caldwell (1991) proposed that mechanisms other than photoreactivation may be operative in the UV-induced effect on photosynthesis. Tezuka *et al.* (1993) found promoted P_N in tomato and suggested that UV-B might be an essential factor for growth and photosynthesis. Beyschlag *et al.* (1988) found no inhibition of P_N with 30 and 45 % ozone depletion in greenhouse grown wheat and wild oat.

The g_s for CO_2 was significantly enhanced at UV-B_{10} (Table 1). This is in parity with the observations of Sullivan and Teramura (1988) in loblolly pine. Negash (1987) suggested that the g_s varied with irradiance, and high energy would close the stomata immediately. Since the present study was performed at high PAR, the effects of UV-B on g_s could be negligible or the presence of natural solar blue radiation, which is a potential generator of energy through respiration, might have alleviated the UV-effects on guard cells, and thus mostly an insignificant effect on g_s was noticed. The C_i was low at UV-B (Table 1), but an increase in residual level of CO_2 was observed at high doses of UV-B. This result is in accordance with Brandle *et al.* (1977) who reported an increased CO_2 in pea seedlings exposed to UV-B.

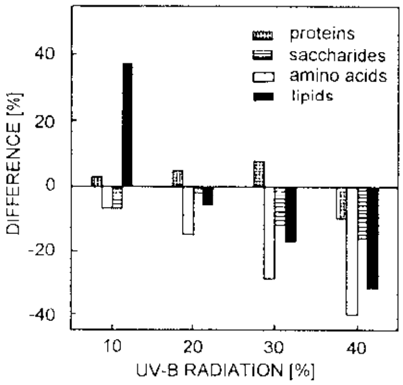

Fig. 1. UV-B induced changes in the contents of proteins, amino acids, saccharides, and lipids in leaves of *Rhizophora apiculata*. Values represent differences over untreated control.

The concentration of leaf proteins was improved following the UV-B treatments, but it was reduced by 10 % in UV-B_{40} (Fig. 1). A similar observation was recorded in field grown potato, radish, bean, and spinach (Esser 1980), but only during early leaf development (Vu *et al.* 1982). The increase in leaf protein was associated with a decline in total free amino acids, in response to UV-B (Fig. 1). The level of total amino acids was 40 % lower at UV-B_{40} than in control plants. The decrease in amino acids could be due to their utilization for protein synthesis, rather than due to their degradation or inhibition of their biosynthesis. UV-B reduced the concentration of saccharides (Fig. 1) only insignificantly. However, Garrard *et al.* (1976) recorded a decline in saccharide content in tomato, cabbage, and collard. The total lipid concentration was enhanced by 37 % at UV-B_{10}, but it was inhibited by 31 % at UV-B_{40} (Fig. 1). Similar results in seedlings grown under low UV-B fluence have been recorded by Tevini *et al.* (1981). Flavonoid concentration was significantly raised with the doses of UV-B radiation (Table 1). Tevini *et al.* (1981) also observed an increase of flavonoids in barley leaves attributed to phytochrome dependent flavone synthesis (Wellmann 1971). On the contrary, the anthocyanin content was reduced in all UV-B treatments except the lowest one (Table 1). Accumulation of anthocyanins is a specific effect of UV-B in many higher plants, despite of the fact that anthocyanin has little absorption in the UV-B range (Teramura 1983). Synthesis of anthocyanin may regulate phenol synthesis (Zaprometov and Zagoskina 1987). We have also observed a negative relationship between the concentrations of phenols and anthocyanins (Table 1). The phenol contents were significantly enhanced in all UV treatments. A similar result was noted in *Vigna sinensis* under water stress and UV-B radiation (Balakumar *et al.* 1993).

Generally, the UV-B at 10 and 20 % enhanced photosynthetic characteristics of *R. apiculata*, but inhibited them at 30 and 40 % doses. Hence, this species may be selected for vegetating the tropical coastal environment where the incidence of UV-B is a growing threat.

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