

Short Communication

Impact of ultraviolet-B radiation on a marine red alga *Kappaphycus alvarezii* (Solieriaceae, Rhodophyta)

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Kappaphycus alvarezii (Doty) Doty, an exotic alga to Indian waters is one of the best sources of kappa carrageenan. It was cultured and 60 days old algal fragments were subjected to UV-B radiation (280-320 nm) for 30 to 180 minutes, in the laboratory and analysed for pigments and carrageenan content. A gradual decrease in pigment levels, soluble protein content, carrageenan yield, and in the properties of carrageenan was observed as compared to control. More than 150 min exposure to UV-B radiation strongly inhibited pigment accumulation (63.90 to 92.20%), carrageenan yield (21.68 to 33.82%), gel strength (81.17 to 82.90%) and protein (48.42 to 58.20%).

Stratospheric ozone, a gaseous layer, greatly attenuates the solar ultraviolet radiation, particularly wave lengths shorter than 310 nm and protects the world biological organisms from the adverse effects of solar UV-radiation. Since the industrial pollutants emitted into the atmosphere have been continuously depleting the ozone molecules, the changes of the ozone layer are expected to allow more UV-radiation in a wave length range of 280-320 nm, termed as UV-B radiation¹.

There are many experimental studies on the detrimental effects of UV-B radiation on growth², photosynthesis³, specifically photosystem-II⁴ and biochemical metabolisms^{5,6} of marine microalgae. But very few investigations on marine macroalgae are available⁷. The spectral characteristics of the UV-B photoinhibitory response in plants and algae suggest that targets of UV-B damage are DNA and photosynthetic components⁸ such as, DI polypeptides of the photosystem II reaction centre⁹. Evidence for the changes in the carbohydrate levels is not available where the seaweeds were subjected to UV-B stress. *Kappaphycus* crop after harvesting is generally dried under sunlight on seashore for further utilisation. While drying the alga in the sun there is every possibility of being exposed when it is wet immediately after harvest and also during process of

drying because the sunlight reaching the coastal belt always contain little ultraviolet radiation (Kulandaivelu, unpublished). Hence the present investigation was undertaken to study the effect of UV-B radiation on the pigments, carbohydrate content and protein of cultivated red macroalga *Kappaphycus alvarezii* (Doty) (Solieriaceae, Rhodophyta).

The young fragments of uniform (10-15cm) size of *Kappaphycus alvarezii* enclosed in perforated polythene bags were cultured by long line rope method¹⁰ in the shallow waters at Thonithurai (Gulf of Mannar, lat. 9°16'N; long 79° 12'E). After 60 days, when the plants reached harvestable size and started maximum accumulation of carrageenan (it is observed that from 60 days till 120 days the accumulation of carrageenan never changed) they were subjected to UV-B radiation exposure for 30, 60, 90, 120, 150 and 180 minutes in a culture room maintained at 25 ± 2°C. UV irradiance of TL 20/12 Philips sun lamp (N.V.Philips) Gleoelamponfa brieken, The Netherlands⁴, as measured at the plant surface was 2w/m². About 100 mg plant material was ground with 80% acetone, centrifuged at 5000 rpm, the clear solution was collected and the amount of chlorophyll *a* was quantified using the method of Arnon¹¹. The measurement of pigments and protein concentrations were quantified after taking the absorptions using the

spectrophotometer Hitachi-557 (Japan). Phycobiliproteins concentration was determined adopting the method of Kremer¹². The protein content was estimated following the method of Harrison & Thomas¹³. The carrageenan was extracted using the method of Criagie & Leigh¹⁴. The physical properties of carrageenan viz., gel strength, gelling and melting temperatures were determined for 1.5% solution by following the method of Thomas & Krishnamurthy¹⁵.

Changes in the levels of photosynthetic pigments in *Kappaphycus* plants exposed to UV-B radiation are shown in Table 1. UV radiation adversely affected the chlorophyll- *a* content; a significant inhibition (3.14 to 6.10% over control) was observed during prolonged exposure period after 150 min. Phycocyanin (PC) and Phycoerytherin (PE) showed a marked decrease (36.09 to 90.26% and 12 to 68.8% respectively) in their levels as compared to control.

Marked decline was observed after 60 minutes in case of phycocyanin and after 150 minutes in phycoerythrin. The changes in chl- *a* reported in UV-B treated plants, altered the net photosynthetic and respiration rates³. UV radiation preferentially reduced the non-photosynthetic pigments in marine plants, whereas, it greatly enhanced their accumulation in higher plants¹⁶. The phycoerytherin, which is responsible for the major light harvesting function (>90%) is seriously affected by UV-B radiation. Beyond 30 min exposure to UV radiation an increase in PE/PC ratio was observed. This indicates that UV-B radiation strongly inhibited the accumulation of PC than PE. However, beyond 150 min the rate of inhibition of PC considerably reduced because the plant system has recovered from UV stress. Grobe & Murphy¹⁷ have reported that marine macroalgae particularly the red algae are very sensitive to UV

Table 1—Changes in pigment level in *Kappaphycus alvarezii* exposed to ultraviolet-B radiation for different periods. (Figures in parentheses indicate percent inhibition with references to respective controls n=3)

Treatment period	Chl <i>a</i> (mg/g.fr.wt)	Phycoerythrin (PC) (µg/g.fr.wt)	Phycocyanin (PE) (µg/g.fr.wt)	Protein (mg/g.fr.wt)	PE/PC
Control	0.952 ± 0.47	1554.63 ± 86.03	457.06 ± 67.06	0.844 ± 0.09	3.4
30 min	0.928 ± 0.02 (3.14)	1212.34 ± 73.3 (12.0)	292.07 ± 41.61 (36.09)	0.753 ± 0.03 (10.8)	4.15
60 min	0.776 ± 0.09 (8.5)	1117.44 ± 23.52 (28.1)	209.07 ± 31.4 (54.25)	0.640 ± 0.02 (24.2)	5.34
90 min	0.716 ± 0.09 (24.8)	1063.99 ± 57.35 (31.64)	187.63 ± 16.44 (58.94)	0.577 ± 1.73 (31.4)	5.67
120 min	0.702 ± 0.03 (26.3)	860.67 ± 65.84 (44.42)	179.69 ± 18.76 (60.31)	0.487 ± 0.12 (43.3)	4.8
150 min	0.344 ± 0.01 (63.9)	639.53 ± 95.14 (58.9)	158.99 ± 20.14 (65.21)	0.446 ± 0.34 (48.4)	4.02
180 min	0.314 ± 0.01 (67.1)	485.55 ± 31.9 (68.8)	44.78 ± 12.83 (0.2)	0.353 ± 0.01 (58.2)	3.35

Table 2—Changes in carrageenan yield and its properties of *Kappaphycus alvarezii* after Ultraviolet-B treatment. (Figures in parentheses indicate percent reduction or increase over control, N=3)

Treatment time	Carrageenan yield (%)	Gel strength of 1.5 carrageenan (g/cm ²)	Gelling temperature (°C)	Melting temperature (°C)
Control	58.8	244.44 ± 1.29	50.2 ± 0.21	90.0 ± 1.15
30 min	54.04	166.23 ± 1.58	56.66 ± 1.11 (12.86)	87.33 ± 1.32 (2.96)
60 min	49.26	159.64 ± 5.96	61.0 ± 0.66 (21.51)	81.3 ± 1.25 (9.7)
90 min	49.11	148.33 ± 8.45	64.83 ± 0.5 (29.14)	77.53 ± 0.89 (13.9)
120 min	48.77	86.33 ± 11.40	66.53 ± 1.04 (32.52)	53.03 ± 1.45 (41.1)
150 min	46.05	44.53 ± 4.81	67.33 ± 0.28 (34.12)	52.5 ± 1.95 (42.7)
180 min	38.91	41.56 ± 1.8	69.6 ± 0.28 (39.2)	49.66 ± 1.75 (54.9)

radiation, like *Ecklonia* and *Eucheuma*. The present investigation agrees with their observation in *Eucheuma* and UV-B radiation not only inhibits the accumulation of photosynthetic pigments but also the protein concentration of *Eucheuma* (= *Kappaphycus*) (Table 1). The effect of UV-B radiation has been related to the damage of DNA and protein¹⁸. However, the loss of chl- *a* molecules also is one of the reasons for the decrease in the protein concentration²⁰.

A short time UV-B exposure (even 30 min) had a large impact on carrageenan yield and its properties (Table 2), a gradual reduction in the yield (8.1 to 33.82%) and gel strength (31.99 to 82.99%) over the control was observed, when the UV-exposure time was increased from 30 min to 180 min. Similarly detrimental effect was observed on gelling and melting temperatures of carrageenan (Table 2). No experimental evidence is reported on the effect of UV-B radiation correlating with carrageenan yield and its properties. Ekman & Peterson²⁰ reported that the photon irradiance and day length seriously alter the carbohydrate (agar) yield and gel strength in *Gracilaria sordida* and *Gracilaria verrucosa*. Further studies are required to understand the responses of other agrophytes and carrageenophytes to UV-B radiation.

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References

- Madronich S, Implications of recent total atmospheric ozone measurements for biological active ultraviolet radiation reaching the Earth's surface *Geophy. Res Lett*, 19 (1992) 37-40.
- Henley W J, Growth and photosynthesis of *Ulva rotundata* (chlorophyta) as a function of temperature and square wave irradiance in indoor culture, *J Phycol*, 28 (1992) 625-634.
- Maegawa M, Kunieda M & Kida W, Difference of the amount of UV absorbing substance between shallow and deep water red algae, *Jpn J Phycol*, 41 (1993) 351-353.
- Muthuvelan B, Fujimori K, Murugan C & Kulandaivelu G, Influence of irradiation quality on photosynthetic pigments saceharides nitrate reductase activity thylakoid organisation and growth in the green algae *Ulva penusa*, *Biol Plant*, 40 (1997/98) 211-218.
- Premkumar A Eswaran K & Kulandaivelu G, Growth, biochemical and photosynthetic responses of greengram seedlings under water stress to ultraviolet-B radiation, *Plant Physiol & Biochem*, 20 (1993) 31.
- Cullen J J & Neale P J Ultraviolet radiation, ozone depletion and marine photosynthesis, *Photosynth Res*, 39 (1994) 303-320.
- Larkum A W D & Wood W F, The effect of UV-B radiation on photosynthesis and respiration of photoplankton, benthic macroalgae and sea grasses, *Photosynth Res*, 36 (1993) 17-23.
- Smith R C Prezelin B B Baker K S Bidigare R R Boucher N P Coley T Karentz D Maclantyre S Metlick H A Menzes D Ondrussek M Wan Z & Waters K J, Ozone depletion, ultraviolet radiation and phytoplankton biology in Antarctic waters, *Science*, 225 (1992) 952.
- Lasser M P Cullen J J & Neale P J, Carbon uptake in a marine diatom during acute exposure to ultraviolet-B radiation - Relative importance of damage and repair, *J Phycol*, 30 (1994) 183-192.
- Mairh O P Jodape S T Tiwari A & Rajyaguru M R, Culture of marine red alga *Kappaphycus striatum* (Schmitz) Doty on the Saurashtra region, west coast of India *Indian J Mar. Sci.*, 24 (1995), 24-31.
- Arnon D I, Copper enzymes in isolated chloroplasts, polyphenol oxidase in *Beta vulgaris*, *Plant Physiol*, 24 (1949) 1-15.
- Kremer B P, Electrophoric and spectral characterisation of algal phycobili proteins, in *Experimental phycology*, edited by Lobban C S, Chapman D J & Kremer B P, (Cambridge University Press, Cambridge) 1988, pp. 111-120.
- Harrison P J & Thomas T E, Biomass measurements: Protein determination, in *Experimental Phycology*. edited by Lobban C S, Chapman D J & Kremer B P, (Cambridge University Press, Cambridge) 1988, pp. 27-34.
- Craigie J S & Leigh C, Carrageenan and agars, in *Hand book of phycolological methods : Physiological and biochemical methods*, edited by Hellebust J A & Craigie J S, (Cambridge Univ. Press, Cambridge) 1978, pp. 109-132.
- Thomas P C, Krishnamurthy V, Agar from cultured *Gracilaria edulis* (Gmel) Silva, *Bot Mar*, 19 (1976) 115-117.
- Eswaran K, Premkumar A & Kulandaivelu G, Impact of enhanced UV-B on photosynthetic and biochemical characteristics of maize under water stress, *Plant Physiol & Biochem*, 20 (1993) 36-40.
- Grobe C W & Murphy T M, Inhibition of growth of *Ulva expansa* (Chlorophyta) by ultraviolet-B radiation, *J Phycol*, 30 (1994) 782-790.
- Giese A C, Studies on ultraviolet radiation action on animal cells, in *Photophysiol*, Vol.2, edited by Giese AC, (Academic Press, New York) 1964, pp. 203-245.
- Tzinis G Argyroudi-Akoyunoglou J H & Akoyunoglou G, The effect of dark intervals in intermittent light on thylakoid development: Photosynthetic unit formation and light harvesting protein accumulation, *Photosynthe Res*, 14 (1987) 241-258.
- Ekman P & Pedersen M, The influence of photon irradiation day length, dark treatment, temperature and growth on the agar composition of *Gracilaria soridiada* and *Gracilaria verucosa*, *Bot Mar*, 33 (1990) 483-495.