

# Research Journal of Pharmaceutical, Biological and Chemical Sciences

## Time Dependent Effect Of UV-B Radiation On The Photosynthetic Electron Transport Activities In The Cyanobacterium, *Spirulina platensis*.

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### ABSTRACT

In this investigation an attempt has been made to study the time dependent effect of UV-B radiation on the photosynthetic electron transport activities of the cyanobacterium, *Spirulina platensis*. To achieve this, cells were exposed to UV-B radiation ( $2 \text{ Wm}^{-2}$ ) for different intervals (20-80 min). The length of incubation gradually caused inhibition in both whole chain as well as PS II catalyzed electron transport. Between two photosystems, PS II is more sensitive than PS I. Light intensity measurements clearly demonstrated that light harvesting complex is main target for UV-B stress.

**Keywords:** electron transport, photosystem, *Spirulina*, UV-B Radiation.

<https://doi.org/10.33887/rjpbcs/2022.13.4.17>

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## INTRODUCTION

UV radiation is one of the serious issues since past few decades due to industrialization. Increase in the industrialization results in the increase in anthropogenically important atmospheric pollutants such as chlorofluorocarbons (CFCs), halocarbons, chloroform (MCF) and dioxins (NO<sub>x</sub>). Considerable amounts of natural production of reactive nitrogen species (RNS) such as nitric oxide (NO<sup>•</sup>), peroxyxynitrate (ONOO<sup>•</sup>) and nitrous oxide (N<sub>2</sub>O) from unpolluted aquatic and terrestrial ecosystems also contribute to the depletion of ozone layer.

Cyanobacteria is a primitive group of gram negative, ubiquitous in nature, oxygenic photoautotrophic prokaryotes which have wide distribution ranging from hot springs to Arctic and Antarctic regions and are important biomass producers in both aquatic and terrestrial ecosystems [1]. They are the valuable sources of natural products of medicinal and industrial importance and also they are the ecologically important organisms which act as natural biofertilizers which fix the atmospheric nitrogen as they consists of enzyme nitrogenase. During this nitrogen fixation and photosynthetic processes they absorb light because of their light absorbing nature they can easily absorb the harmful UV radiation which leads to lethal effects [2]. PS II is the membrane protein complex found in oxygenic photosynthetic organisms (higher plants, green algae and cyanobacteria), which harnesses light energy to split H<sub>2</sub>O into O<sub>2</sub>, protons and electrons [3]. There is general consensus that UV-B radiation influences primarily PS II, there are many different reports on possible targets [4, 5]. Different techniques were used to reveal the possible target sites of UV-B radiation such as fluorescence induction, flash-induced absorption changes, and measurement of O<sub>2</sub> evolution. However it seems to be well established that the redox components of PS II are affected by UV-B to some degree. Most observations support the notion that UV-B preferentially inactivates the water oxidizing complex with additional effects on the Q<sub>A</sub> and Q<sub>B</sub> acceptors, as well as on the Tyr-Z and Tyr-D donors [6]. The reasons for selecting *Spirulina platensis*, it is a blue green algae. It is now using as an important single cell protein as an edible source of protein (7). Our objective in this investigation is to identify the changes in the photosynthetic electron transport and energy transfer properties of PS II in the above cyanobacterium, *Spirulina plantensis* under the influence of UV-B.

## MATERIALS AND METHODS

*Spirulina platensis* is a trichomatous non-nitrogen fixing cyanobacterium. The trichomes consist of a number of cells (6-10) covered by a mucilaginous sheath and are spirally twisted. The cells were cultured in Zarrouk media at 25°C under illumination of white light with intensity, 20 Wm<sup>-2</sup>. The log phase cells were harvested in to fresh growth medium in to petri plates and exposed to UV-B radiation at influence rate of 2 Wm<sup>-2</sup> (obtained from a Philips TL 20 type in the spectral range of 280- 320 nm and with a peak at 312 nm) for different intervals (20-80 min). Whole chain electron transport assay (H<sub>2</sub>O→MV) was studied in terms of O<sub>2</sub> consumption due to photoreduction of Methyl viologen (MV) and its subsequent auto oxidation. The reaction mixture contained reaction buffer (25 mM Hepes- NaOH (pH 7.5), 20 mM NaCl), 0.5 mM MV, 1 mM sodium azide and the intact cells equivalent to 12 to 15 µg Chl *a*.

In PS II mesurments, para benzoquinone (pBQ) was used to measure the PS II catalyzed electron transport (H<sub>2</sub>O→pBQ) in the intact cells. Being a lipophilic compound pBQ enters into the intact cells and accepts electrons at plastoquinone (PQ) position [8]. The reaction mixture contained reaction buffer (same as used in cell harvesting), 0.5 mM freshly prepared pBQ and the intact cells equivalent to 12 to 15 µg Chl *a*. PS I catalyzed electron transport assay was measured as O<sub>2</sub> consumption. The 2 ml reaction mixture contains reaction buffer [50 mM HEPES-NaOH (pH 7.5), 100 mM sucrose, 2 mM MgCl<sub>2</sub> and 5 mM KCl], 0.1 mM 2,6- dichlorophenol indophenol (DCPIP), 0.5 mM MV, 5 mM ascorbate, 1 mM sodium azide, 10 µM DCMU and thylakoid membranes equivalent to 40 µg of Chl.

## RESULTS AND DISCUSSION

To analyze the effect of UV-B radiations on primary reaction of photosynthesis, intact cells of *Spirulina platensis* were exposed for different time intervals of UV-B radiation (20-100 min) with the intensity of 2 Wm<sup>-2</sup>. After giving the treatment initially whole chain electron transport activity (H<sub>2</sub>O →MV) was measured using MV as terminal electron acceptor. Control cells exhibited the whole chain electron transport activity equal to that of 193 µmoles of O<sub>2</sub> consumed mg<sup>-1</sup> Chl h<sup>-1</sup> (Table-1). The increase in the duration of UV-B exposure from 20-80 min under conditions stirred caused a time dependent inhibition

in whole chain electron transport activity. There was a 52% inhibition was noticed in the whole chain electron transport activity after 40 min of incubation with UV-B (Table 1). Further increase in the incubation period to 80 min caused an enhancement in the inhibition to 81%. This inhibition in whole chain electron transport could be due to the alterations at two levels i.e. inhibition at PS II level or at PS I level as has been earlier discovered by several workers [9].

**Table 1: Effect of UV-B radiation on whole chain electron transport ( $H_2O \rightarrow MV$ ) on the intact cells of cyanobacterium, *Spirulina platensis*.**

UV-B exposure, min	Whole chain electron transport activity ( $H_2O \rightarrow MV$ ) $\mu$ moles of $O_2 \downarrow$ $mg\ Chl^{-1}\ h^{-1}$	Percentage inhibition
Control	193 $\pm$ 15	0
20	130 $\pm$ 11	33
40	93 $\pm$ 7	52
60	52 $\pm$ 4	74
80	37 $\pm$ 3	81

To prove this, time dependent effect of UV-B was studied using pBQ as Hill acceptor. UV-B radiation was able to induce almost 50% inhibition in Hill activity after 40 min of incubation only. Further increase in the exposure caused additional loss of PS II catalysed electron transport activity to 78% at the end of 80 min of exposure (Table 2). The inhibition in the PS II catalyzed electron transport activity could be due to alterations at the level of oxidizing site or changes at D1, D2 proteins or changes in the reducing side of PS II as has been suggested earlier by others [10-13]. When compare to the whole chain electron transport and PS II electron transport, PS I catalyzed reactions could not be assayed in intact cells of *Spirulina* as reduced DCPIP/TMPD/DAD did not readily enter in to intact cells.

**Table 2: Effect of UV-B radiation on PSII catalyzed electron transport ( $H_2O \rightarrow p\text{-BQ}$ ) on the intact cells of *Spirulina platensis***

UV-B exposure, min	PS II electron transport activity ( $H_2O \rightarrow p\text{-BQ}$ ) $\mu$ moles of $O_2 \uparrow$ $mg\ Chl^{-1}\ h^{-1}$	Percentage inhibition
Control	314 $\pm$ 24	0
20	240 $\pm$ 23	24
40	164 $\pm$ 16	48
60	118 $\pm$ 11	63
80	62 $\pm$ 4	79

After giving the UV-B treatment we have studied the effect of UV-B on PS I catalyzed electron transport activities using thylakoids isolated from intact cells. The electron transport measurements indicated marginal inhibition in PS I activity was noticed (Table 3). The UV-B radiation affects the photosynthetic electron transport in a preferential manner. Between two photosystems, PS II is more susceptible than PS I. To identify the possible target site in PS II, a study has been made to analyze the effect of UV-B (40 min of exposure) at both light saturating ( $420\ Wm^{-2}$ ) and light limiting ( $2\ Wm^{-2}$ ) conditions (Table 4). The inhibition in PS II activity was more at light saturating conditions than that of light limiting conditions. The inhibition at light limiting conditions of UV-B treated sample could be due to alterations in the light harvesting complex pigment proteins of this cyanobacterium. The possible reason for the enhancement of inhibition at light saturating conditions by UV-B radiation in Hill reaction could be due to the presence of additional inhibitory site near PS II. Thus, when UV-B radiation is applied it exerts inhibitory effects on PS II photochemistry.

**Table 3: Effect of UV-B radiation on PSI catalyzed electron transport (DCPIPH<sub>2</sub>→ MV) on the intact cells of *Spirulina platensis***

UV-B exposure, min	PS I electron transport activity DCPIPH <sub>2</sub> → MV $\mu$ moles of O <sub>2</sub> ↓mg Chl <sup>-1</sup> h <sup>-1</sup>	Percentage inhibition
Control	372 ± 25	0
20	361 ± 24	3
40	343± 23	8
60	331± 22	12
80	316 ±21	16

**Table 4: Effect of different illuminated light intensities on UV-B induced PSII catalyzed electron transport activity inhibition in the cyanobacterium *Spirulina platensis*.**

Light intensity (Wm <sup>-2</sup> )	PS II activity ( $\mu$ mol O <sub>2</sub> evolved mg Chl <sup>-1</sup> h <sup>-1</sup> ) (H <sub>2</sub> O → pBQ)		Percentage inhibition
	Control	UV-B Treated (2.1 Wm <sup>-2</sup> )	
12	55 ± 4	32 ±3	42
110	140 ± 12	80 ± 7	43
220	189 ± 16	104± 9	45
420	305 ± 26	162 ± 13	47

#### REFERENCES

- [1] Stanier RY, Cohen-Bazire G. Ann Rev Microbiol 1977; 31:225-274.
- [2] Fernanda Pessoa M. Emir J Food Agric 2012;24:527-545
- [3] Yunsheng Lou, Ren L, Li Z, Cheng H, and Zhang T. Water, Air, and Soil Pollution 2011;219:501-506.
- [4] Tevini M. 2004, Plant responses to ultraviolet radiation stress. In: Chlorophyll a Fluorescence a Signature of Photosynthesis (Papageorgiou GC, Govindjee eds.), pp. 605-621, Springer, Dordrecht.
- [5] Imre Vass. Biochim Biophys Acta 2012;1817: 209-217.
- [6] Tyystjarvi E. Coord Chem Rev 2008;252: 361-376
- [7] Venkatraman LV. 1983, A monograph on *Spirulina platensis*. Biotechnology and application, CFTRI, Mysore, India.
- [8] Trebst, A. Ann Rev Plant Physiol 1974;25: 423-458.
- [9] Kulandaivelu G, Geetha V and Periyanan S. (1989) Inhibition of energy transfer reactions in cyanobacteria by different ultraviolet radiation. In: Photosynthesis- Molecular Biology and Bioenergetics. (Singhal GS, Barber J, Delley RA, Govindjee, Haselkorn R and Mohanty P. eds), pp.305-313, Narosa publishing House, New Delhi.
- [10] Renger G, Völker M, Eckert HJ, Fromme R, Hohm-Veit S, and Gräber P. Photochem Photobiol 1989;49: 97-105.
- [11] Wilson WH, Joint IR, Carr NG, and Mann NH. Apple environ Microbiol 199359:3736—3743.
- [12] Rajagopal S. and Murthy SDS. Bio Plant 1996;38:129-132.
- [13] Murthy SDS, and Rajagopal S. Photosynthetica 1995;31: 481-487.