CYCLOBUTANE PYRIMIDINE DIMERS DETECTION IN MARINE PHYTOPLANKTON *ISOCHRYSIS GALBANA* FOLLOWING UV IRRADIATION

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Abstract

The effect of artificial and solar UV irradiation on DNA of marine phytoplankton *Isochrysis galbana* was investigated. Cultures grown under 16/8 light/dark regime contained smaller amount of photosynthetic pigments and UV absorbing compounds and showed greater sensitivity to UV light than the cultures grown under 24/0 light regime. For specific detection of cyclobutane pyrimidine dimers (CPDs) in *I. galbana* alkaline DNA filter elution using T4 endonuclease V was adapted. Artificial UV irradiation increases CPDs in dose-dependant manner. After 1 h of sunlight exposure, the level of DNA damage increase significantly but prolonged exposure decreased it due to the efficiency of *I. galbana* DNA repair mechanisms.

KEYWORDS: phytoplankton, pigments, UV, sunlight, cyclobutan pyrimidine dimers

Oxygen and sunshine (UV) are two major genotoxic agents that all photosynthetic organisms are obliged to be exposed to. One way to assess the importance of enhanced UV irradiation as a potential source of biological damage in marine organisms is to determine UV- induced DNA damage and the capacity for DNA repair. Cyclobutane pyrimidine dimers (CPDs) are exclusive UV product and make 75% of the UV-induced DNA damage products (1), having an inhibitory effect on transcription and replication as well as mutagenic effect. We measured incidence of CPDs in marine phytoplankton *Isochrysis galbana* cultures (2) grown under different light/dark regimes after artificial UV exposure. CPD content in *I. galbana* cells was expressed as SSF (strand scission factor) - measure of CPD sites as single strand breaks (3) after T4endonuclease V digestion. In cells grown under 16/8 h (light/dark) regime after exposure to artificial UV (0-37.5 kJ/m^2) more CPDs was detected than in the cells grown under constant illumination (24 h) (Fig.1).The latter contained higher amount of photosynthetic pigments (chlorophyll a) as well as UV absorbing compounds (Table 1). These results revealed correlation between light exposure, UV-absorbing compounds content and sensitivity to UV light, suggesting strong influence of photoprotective compounds in reducing the susceptibility to UV damage in phytoplankton cells.

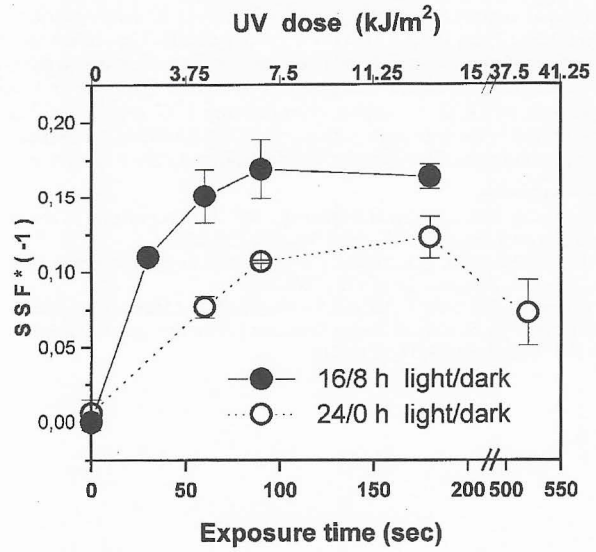

Fig. 1. Dependence of SS to artificial UV radiation for *Isochrysis galbana* cultures grown under differents light/dark regimes

Table1. Chlorophyll a and UV-absorbing compounds content in *Isochrysis galbana* cells grown under different light/dark regimes.

Illumination light/dark (h)	Chlorophyll *a* (μg/10^6 cells)	UV - absorbing components (A_{300}/10^7 cells)
24/0	0.353	0.792
16/8	0.192	0.158

Besides protection there is a great importance of the existence of DNA repair mechanisms as a part of biological response of phytoplankton cells to UV irradiation. DNA photodamage and DNA repair capacity in *I. galbana* cells exposed to ambient solar irradiation was measured on August 6 and 20,1996 (Table 2). After 1 hour, DNA damage increased and after 3 hours of exposure, it decreased to the 40% of the damage obtained after first 1 hour. Thus, DNA protection and damage elimination mechanisms successfully lowered CPDs accumulation, suggesting that sunlight exposure induces DNA damage that could be repaired even when the DNA damage and repair are happening simultaneously.

Table 2. CPDs accumulation and their repair in *Isochrysis galbana* cells during sunlight exposure in the Northern Adriatic (45o 05` N, 13o 30` E, Rovinj, Croatia).

SUNLIGHT EXPOSURE	August 6, 1996	August 20, 1996
Time/h	SSF	
11:00	-0.006 ± 0,019	-0,006 ± 0,019
12:00	-0,055 ± 0,015	-0,037± 0,002
14:00	-0,007 ± 0,018	-0,015 ± 0,003

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