

Species analyzed, common name and total number of MAAs isolated in each sample (continued)

Species	Common name	Number of MAAs
<i>Notasellus sarsi</i>	Isopod	6
<i>Euphausia superba</i>	Krill	8
Amphipod no. 2	Amphipod	6
Amphipod no. 3	Amphipod	5
Amphipod no. 4	Amphipod	7
Amphipod no. 6	Amphipod	6
Amphipod no. 8	Amphipod	7
Amphipod no. 10	Amphipod	8
Amphipod no. 13	Amphipod	6
<i>Beania livingstonei</i>	Bryozoan	2
<i>Inversiula nutrix</i>	Bryozoan	6
<i>Granaster nutrix</i>	Sea star	6
<i>Amphioplus affinis</i>	Brittle star	1
<i>Cucumaria cf. georgiana</i>	Sea cucumber	0
<i>Ekmocucumis steineri</i>	Sea cucumber	0
<i>Molgula enodis</i>	Sea squirt	7
<i>Salpa thompsoni</i>	Salp	0
Chaetognath no. 1	Chaetognath	0
Icefish no. 1 (larvae)	Icefish	6
<i>Curdiea racovitzae</i>	Red algae	6
<i>Iridea chordata</i>	Red algae	6
<i>Lithothamnion cf. antarcticum</i>	Red algae	2
<i>Palmaria decipiens</i>	Red algae	7
<i>Phyllophora appendiculata</i>	Red algae	5
<i>Desmarestia menziesii</i>	Brown algae	3
Algal mat	Filamentous greens	4
Algal mat	Filamentous diatoms	3

Molecular and biological responses of antarctic phytoplankton to ultraviolet radiation

DAVID L. MITCHELL and DENEK KARENTZ

Laboratory of Radiobiology and Environmental Health
University of California San Francisco, California 94143-0750

Air pollution has resulted in global decreases in stratospheric ozone concentrations and an increase in the amount of harmful solar radiation reaching the Earth's surface. The effects of increased ultraviolet-B (UV-B) radiation (290–320 nanometers) on the human population are complex. The obvious and direct consequences include increased incidence of skin cancer and accelerated aging; less obvious and more indirect effects include deterioration of natural systems such as marine plankton, integral to oxygen production and the base of the oceanic food chain. To assess the impact of ozone depletion on marine communities, it is necessary to define the biomolecular response of individual organisms to UV-B.

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References

- Chioccare, F., A. Della Gala, M. de Rosa, E. Novellino, and G. Prota. 1980. Mycosporine aminoacids and related compounds from the eggs of fishes. *Bulletin des Sociétés Chimique Belges*, 89, 1,101–1,106.
- Dunlap, W.C., B.E. Chalker, and W.M. Bandaranayake. 1988. *New sunscreens agents derived from tropical marine organisms of the Great Barrier Reef, Australia*. Proceeding 6th International Coral Reef Symposium.
- Dunlap, W.C., D.McB. Williams, B.E. Chalker, and A. Banaszak. 1989. Biochemical photoadaptation in vision: UV-absorbing pigments in fish eye tissues. *Comparative Biochemistry and Physiology*, 93B, 601–607.
- Karentz, D., F.S. McEuen, M.C. Land, and W.C. Dunlap. In press. A survey of mycosporine-like amino acid compounds in Antarctic marine organisms: Potential protection from ultraviolet exposure. *Marine Biology*.
- Mitchell, D.L. and D. Karentz. 1990. Molecular and biological responses of antarctic phytoplankton to ultraviolet radiation. *Antarctic Journal of the U.S.*, 25(5).
- Nakamura, H., J. Kobayashi, and Y. Hirata. 1982. Separation of mycosporine-like amino acids in marine organisms using reversed-phased high performance liquid chromatography. *Journal of Chromatography*, 250, 113–118.

Ultraviolet light is lethal to living systems. Due to its absorbance spectrum, DNA is considered the major cellular target (i.e., it is the primary chromophore). A portion of the energy absorbed by DNA is converted into stable structural damage, primarily involving interactions between adjacent pyrimidine bases (i.e., thymine and cytosine). The major photoproducts induced are the cyclobutane dimer and (6–4) photoproduct (so named for the chemical linkages between the dimerized bases). These lesions cause significant distortions in the phosphodiester backbone which inhibit transcription of essential genes as well as the onset and progression of DNA replication.

DNA repair mechanisms have evolved in response to this damage:

- photoreaction (PR) specifically splits cyclobutane dimers by the combined action of a simple enzyme and visible light;
- nucleotide excision repair (NER) is a more complex system involving recognition of a broader class of damage, assembly of a DNA repair complex at the site of damage, incision of the DNA backbone upstream from the damage, and the concomitant excision and resynthesis of the damaged strand by the action of DNA polymerase.

We have initiated studies on the induction and repair of UV-B damage in various antarctic phytoplankton species using

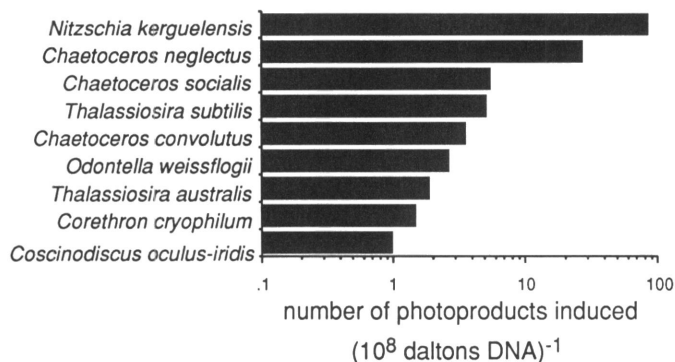


Figure 1. Total number of photoproducts induced in each diatom species by exposure to 2,500 joules per square meter.

radioimmunoassays which detect cyclobutane dimers and (6–4) photoproducts in DNA (for technique see Mitchell, Haipek, and Clarkson 1985). Species were collected at Palmer Station and maintained in the laboratory for photobiological studies. The preliminary results indicate a great diversity in the photobiology of individual phytoplankton species. In figure 1, the combined induction of cyclobutane dimers and (6–4) photoproducts (induced at about equal rates) is shown after 2,500 joules per square meter UV-B light emitted by a filtered sunlamp. The difference in the amount of photodamage induced per unit length DNA (10^8 daltons = 150,000 base pairs) in the individual species was nearly 100-fold, ranging from 0.9 lesions per 10^8 daltons in *Coscinodiscus oculus-iridis* to 84 lesions per 10^8 daltons in *Nitzschia kerguelensis*.

Comparable differences in the ability of individual species to repair ultraviolet damage was also observed (figure 2). Diatoms irradiated with 2,500 joules per square meter UV-B light were incubated for 6 hours in either yellow or white light conditions prior to DNA isolation. Under the yellow lights, photoreactivation was minimized and repair was restricted to nucleotide excision; under white lights, the contribution of photoreactivation could be assessed. The capacity of individual diatoms to excise damage ranged from negligible to near complete removal within 6 hours; this extreme variation was even

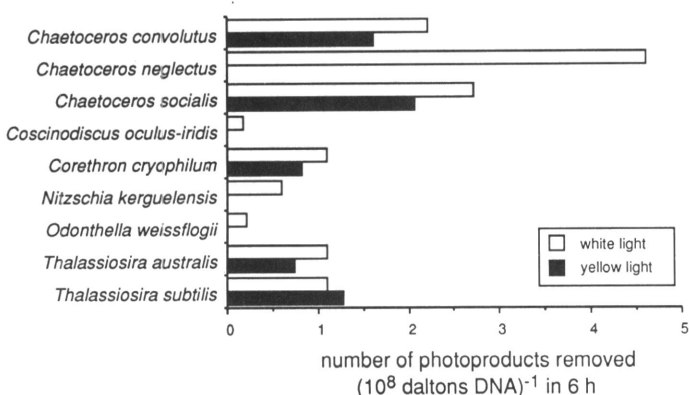


Figure 2. Number of photoproducts removed under white and yellow light post-irradiation incubation after exposure to 2,500 joules per square meter.

D_{37} values for each species under white and yellow light

Species	D_{37}	
	White	Yellow
<i>Coscinodiscus oculus-iridis</i> (3)	8,243	1,180
<i>Coscinodiscus oculus-iridis</i> (12)	12,582	3,424
<i>Coscinodiscus oculus-iridis</i> (80)	12,647	934
<i>Corethron cryophilum</i>	25,338	312
<i>Licmophora decora</i>	4,057	1,173
<i>Thalassiosira australis</i>	681	84
<i>Odontella weissflogii</i>	3,695	507
<i>Eucampia antarctica</i>	2,915	1,680
<i>Porosira pseudodenticulata</i>	2,196	308
<i>Thalassiosira subtilis</i>	1,844	252

observed with a single genus (e.g., *Chaetoceros*). White light enhanced repair in nearly all species, suggesting that photoreactivation may play a significant role in damage tolerance by marine phytoplankton. We hope to elucidate this question more definitively in future experiments by separating photoreactivation from generalized effects on cell metabolism caused by white light (i.e., photosynthesis).

Consistent with the diverse molecular response to ultraviolet light, the ability of phytoplankton to survive the effects of UV-B light, according to our findings, was extremely variable. The UV-B doses required to kill, on average, one cell under the different light regimes (the D_{37} value) are shown in the table. It is apparent that, within this cross-section of the antarctic marine community, relatively sensitive (e.g., *Thalassiosira* spp.) and resistant (e.g., *Coscinodiscus* spp.) phytoplankton species co-exist.

Marine phytoplankton are an important component of the oceanic food chain and provide much of the world's oxygen. From our studies, it is evident that to understand the consequences of ozone depletion on marine environments, it is essential to define the photochemistry and photobiology of the individual organisms constituting natural communities. Knowledge of the molecular and biological responses to UV-B light at the species-specific level will enable us to predict changes in the biomass and species composition of marine ecosystems.

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Reference

Mitchell, D.L., C.A. Haipek, and J.M. Clarkson. 1985. Further characterization of a polyclonal antiserum for DNA photoproducts: The use of different labeled antigens to control its specificity. *Mutation Research*, 146, 129–133.