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EFFECT OF UV-B ON LIPID CONTENT OF THREE ANTARCTIC MARINE PHYTOPLANKTON

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Key Word Index—*Phaeocystis antarctica* ; *Odontella weissflogii* ; *Chaetoceros simplex* ; algae ; diatoms ; UV-B ; ultraviolet radiation ; Antarctic ; lipids ; fatty acids.

Abstract—The effects of UV-B radiation on the fatty acid, total lipid and sterol composition and content of three Antarctic marine phytoplankton were examined in a preliminary culture experiment. Exponential growth phase cultures of the diatoms, *Odontella weissflogii* and *Chaetoceros simplex*, and the Haptophyte, *Phaeocystis antarctica*, were grown at 2 (± 1)° and exposed to 16.3 (± 0.7) Wm^{-2} photosynthetically active radiation (PAR). UV-irradiated treatments were exposed to constant UV-A (4.39 (± 0.20) Wm^{-2}) and low (0.37 Wm^{-2}) or high UV-B (1.59 Wm^{-2}). UV-B treatments induced species-specific changes in lipid content and composition. The sterol, fatty acid and total lipid content and profiles for *O. weissfloggi* changed little under low UV-B when compared with control conditions (PAR alone), but showed a decrease in the lipid content per cell under high UV-B treatment. In contrast, when *P. antarctica* was exposed to low UV-B irradiance, storage lipids were reduced and structural lipids increased, indicating that low UV-B enhanced cell growth and metabolism. *Phaeocystis antarctica* also contained a higher proportion of polyunsaturated fatty acids under low UV-B in comparison with PAR-irradiated control cultures. The flagellate life stage of *P. antarctica* dies under high UV-B irradiation. However, exposure of *P. antarctica* to high UV-B irradiance increased total lipid, triacylglycerol and free fatty acid concentrations, indicating that increases in lipid content were associated with the colonial life stage. Lipid concentrations per cell also increased when *C. simplex* was exposed to high UV-B irradiance. This resulted from increases in free fatty acid concentration, principally saturated fatty acids, and may indicate degradation of complex lipid during high UV-B treatment. Under low UV-B, there was no statistically significant difference when compared with the control. In comparison with the PAR-exposed controls, low UV-B benefited or did not affect cellular metabolism and growth of the three microalgae examined. Our results indicate that the effect of UV-B irradiances on the lipid content of Antarctic marine phytoplankton is species-specific. Changes in ambient UV-B may alter the nutritional quality of food available to higher trophic levels.

INTRODUCTION

Spring-time stratospheric ozone concentrations over Antarctica now fall to less than 30% of pre-ozone hole values [1] and significant ozone depletion persists until February [2, 3]. Thus, throughout the period of high productivity, Antarctic phytoplankton are exposed to enhanced UV-B. Phytoplankton blooms in the shallow mixed depth of the marginal ice zone, which comprises 25–67% of the primary production in the Southern Ocean [4], may be especially vulnerable to extended exposure from high UV-B radiation [5, 6]. Similarly, sea ice algae contribute 10–50% of the primary production in some areas during spring [7], but at this time of year ice may be sufficiently transparent to UV so that biologically significant doses of UV-B radiation are transmitted to the ice algal community [8].

UV-B has been shown to reduce the survival, growth and production of phytoplankton [9–15]. More recently, studies have examined the impact of UV irradiation on fatty acids and lipid concentrations in algae [16–18]. However, these studies have not examined the effects of UV-B radiation on the lipid composition of Antarctic phytoplankton isolates.

Phytoplankton contain high concentrations of fatty acids that are essential components in the diet of

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higher trophic levels [18, 19]. An increase in UV-B radiation may change the concentration of these essential fatty acids and thus the nutritional value of Antarctic phytoplankton. Phytoplankton species composition may also be affected by increased UV-B [15]. Changes to the nature, quantity and quality of material available to grazers, such as krill, are likely to affect the structure and function of the Antarctic marine food web.

The aim of the present investigation was to determine the effects of UV irradiation on the fatty acid, total lipid and sterol content of three Antarctic marine phytoplankton: *Odontella weissflogii*, *Chaetoceros simplex* and *Phaeocystis antarctica*. Preliminary experiments showed that these algal species were capable of surviving the experimental radiation treatments. To our knowledge, fatty acid profiles for *O. weissflogii* have not been published previously. The previously published fatty acid profile for *P. antarctica* [20] is similar to the results found in our study. Fatty acid profiles published for temperate *C. simplex* [19] are distinct from the Antarctic strain reported here.

Two regimes were chosen, low and high UV-B. The low UV-B treatment approximated to the radiation levels presently occurring at the Vestfold Hills, Antarctica (68S 77E) during the summer solstice. High UV-B was over four times this level. Photosynthetically active radiation (PAR) and UV-A, which are required for photoreactivation repair of UV-B induced damage [21], were included in the irradiance of experimental UV treatments.

RESULTS

Cell numbers

The three algae showed inter- and intraspecific changes in cell concentration as a result of the irradiance treatment (Table 1). *Chaetoceros simplex* exhibited significant and similar growth under control and low UV-B treatments but no growth during exposure to the high UV-B treatment. The *Phaeocystis antarctica* culture used in the experiments contained the two principal life stages, colonial and flagellate. These life stages differ greatly in their physiology and UV susceptibility [14, 22]. The control and low UV-B treatments contained both life stages of *P. antarctica* at the end of the exposure (Table 1). The flagellate stage of *P. antarctica* exhibited no significant growth during irradiation in the control and low UV-B treatments but suffered 100% mortality when exposed to high UV-B. Although the mean cell concentration of colonial stage *P. antarctica* increased markedly in control and low UV-B treatments, these changes were not statistically significant. High UV-B caused 75% mortality of the colonial stage and this life stage comprised all the live cells under this radiation treatment. *Odontella weissflogii* cell numbers were approximately an order of magnitude less than those measured for the other two species and, at this low cell abundance, no significant change in cell numbers could be discerned during the 2 days irradiation, irrespective of treatment.

Total lipids

Cellular lipid changes differed between algal species and irradiation treatments. Variations in both lipid concentrations and compositions were observed (Table 2). *Odontella weissflogii* cell concentrations did not change significantly between light treatments. Total cellular lipid concentration and lipid per cell in *O. weissflogii* decreased under high UV-B (195 ± 34 to 139 ± 4 pg cell^{-1}; Table 2). However, the lipid profile was unaffected by both UV-B treatments. Control and UV-B-exposed treatments showed between 93 and 95% polar lipid for all three treatments. Triacylglycerol and free fatty acids accounted for the remaining lipid (Table 2). This indicates that although lipid concentration decreased under high UV-B, it was not being stored or degraded and, thus, the *O. weissflogii* cells were experiencing little physiological stress under either low or high UV-B treatments.

Under control and low UV-B irradiance treatments, *C. simplex* exhibited significant growth but no significant difference in lipid profiles. Exposure to high

Table 1. Cell numbers for three Antarctic marine algae cultured under high UV-B, low UV-B and control treatments

Cell numbers (x10^5) per litre

Algal species	$T = 0$	Treatment Control	Low UV-B	High UV-B
Chaetoceros simplex	465 (89)	948 (152)	860 (142)	474 (72)
Phaeocystis antarctica (Flagellate)	227 (44)	233 (36)	205 (48)	–
Phaeocystis antarctica (Colonial)	82 (21)	122 (41)	184 (72)	38 (22)
Phaeocystis antarctica (total)	310 (65)	355 (78)	389 (120)	38 (22)
Odontella weissflogii	18 [3]	16 (3)	16 (4)	18 (3)

$T = 0$ refers to the cell concentration immediately prior to incubation. Figures in parentheses represent standard deviation for three replicates.

Table 2. Cellular lipid concentration (pg cell^{-1}) and percentage composition of three Antarctic marine algae cultured under high UV-B, low UV-B and control treatments

		Concentration (pg cell^{-1})					
Algal species	Treatment	Hydrocarbon	Triacylglycerol	Free fatty acid	Sterol	Polar	Sum
**Chaetoceros simplex*	Control	nd	0.04 (0.0)	0.3 (0.02)	0.02	0.8 (0.2)	1.2
	Low UV	nd	nd	0.4 (0.1)	0.03	1.0 (0.2)	1.4
	High uv	nd	nd	0.9 (0.1)	0.03 (0.01)	1.1 (0.1)	2.0
**Odentella weissflogii*	Control	0.4 (0.2)	3.6 (0.6)	9.0 (1.1)	1.5 (0.2)	195 (34)	210
	Low UV	0.1 (0.004)	2.9 (1.0)	7.7 (3.2)	1.2 (0.3)	176 (29)	187
	High UV	0.4 (0.002)	2.9 (0.3)	4.8 (0.5)	1.2 (0.3)	139 (4)	148
Phaeocystis antarctica	Control	0.01 (0.01)	0.002 (0.001)	0.1 (0.03)	0.02 (0.02)	0.5 (0.2)	0.6
	Low UV	0.02 (0.01)	0.003 (0.0001)	0.1 (0.02)	0.01 (0.007)	0.7 (0.2)	0.8
	High UV	0.19 (0.08)	0.08 (0.030)	0.8 (0.5)	0.13 (0.1)	4.8 (3.0)	6.0
		Percentage composition					
**Chaetoceros simplex*	Control	nd	0.4	16 (2.4)	1.9 (0.8)	79 (3.0)	
	Low UV	nd	nd	18 (3.9)	1.9 (0.2)	78 (3.6)	
	High UV	nd	nd	28 (2.2)	1.9 (0.3)	69 (2.8)	
**Odontella weissflogii*	Control	0.2 (0.1)	1.2 (0.1)	2.5 (0.2)	0.8 (0.0)	93 (0.4)	
	Low UV	tr	0.9 (0.3)	2.5 (0.7)	0.7 (0.2)	94 (1.7)	
	High UV	0.3 (0.3)	1.2 (0.4)	1.7 (0.7)	0.8 (0.2)	93 (1.9)	
Phaeocystis antarctica	Control	2.0 (1.5)	0.1 (1.0)	6.9 (2.4)	2.8 (0.8)	90 (1.6)	
	Low UV	1.8 (2.2)	0.1 (0.1)	3.7 (0.6)	1.8 (1.2)	94 (0.9)	
	High UV	1.2 (3.6)	0.9 (0.1)	8.2 (1.9)	2.5 (2.6)	88 (3.1)	

Figures in parentheses indicate standard deviation of two replicates. nd : not detected. *Unidentified pigments also present

UV-B inhibited cell growth and cellular lipid concentration per cell increased. This increase was predominantly due to a greater than two-fold increase in free fatty acid concentration in comparison with the control (Table 2). The lipid profile of *C. simplex* also showed an increase in free fatty acid (16 ± 2 to $28 \pm 2\%$) and a decrease in the proportion of polar lipid (79 ± 3 to $69 \pm 3\%$) under high UV-B treatment when compared to the low UV-B treatment and the control (Table 2).

Under low UV-B, polar lipid composition of *P. antarctica* increased (90 ± 2 to 94 ± 1 %) and free fatty acid decreased (7 ± 2 to 4 ± 0.6% Table 2), when compared with the control. This indicates a small increase in the cell membrane components and a decrease in degradation products. Exposure of *P. antarctica* to high UV-B caused an increase in lipid concentration but there was high variability between replicates (0.6 ± 0.2 to 6 ± 4 pg cell^{-1}; Table 2). In addition, the flagellate stage died and the concentration of live colonial stage cells declined. Colonial cells are characteristically larger than the flagellates and dead cells may also have contributed to the higher lipid concentrates per cell. The concentration of free fatty acids also increased (0.07 ± 0.03 to 0.8 ± 0.5 pg cell^{-1}; Table 2) under the high UV-B irradiance, indicating that *P. antarctica* experienced physiological stress; however, free fatty acid and polar lipid percentage composition were similar to the control (8 ± 2 and 88 ± 3%, respectively). The lipid composition is therefore consistent with an enhancement of growth conditions of this alga under low UV-B radiation, compared with PAR light conditions alone.

The three algal species showed no statistically significant changes in lipid concentrations of the minor lipid components, sterols and hydrocarbons or unidentified pigments (from the TLC-FID analysis) for the two UV-exposed treatments (Table 2).

Fatty acids

Fatty acid profiles and concentrations reflected the total lipid results for the three algal species (Table 3). The fatty acid concentration decreased in *O. weissflogii* and increased in *P. antarctica* and *C. simplex*. Of the three algae, *O. weissflogii* contained the highest concentration and relative abundance of polyunsaturated fatty acids (PUFAs) (Tables 1 and 3). For all three treatments, PUFAs remained at 60% of the total fatty acid composition. $20:5\omega3$ comprised between 26 and 29% of the total fatty acids (Table 3). The fatty acid profile of *O. weissflogii* remained almost unchanged for both UV-B treatments and the control (Table 3). Although there was no change in the PUFAs, there was a small increase in the relative amounts of 22 : 1 and 20 : 1 under high UV-B (Table 3).

Under high UV-B, the percent contribution by PUFAs in *C. simplex* decreased from 37 to 31% (Table 3). C_{16}PUFAs and $18:3\omega6$ decreased (Table 3) and the contribution by $20:5\omega3$, though not significant,

Table 3. Total fatty acid percentage composition of three Antarctic marine algae cultured under high UV-B, low UV-B and control treatments

	Percentage Composition (%)								
	Chaetorceros simplex			*Odontella weissflogii*			*Phaeocystis antarctica*		
	Control	Low	High	Control	Low	High	Control	Low	High
Saturated									
14 : 0	7.5 (0.8)	8.5 (0.9)	7.9 (0.6)	5.4 (*)	5.3 (0.3)	5.8 (0.3)	28.0 (0.4)	23.5 (1.2)	24.9 (6.0)
15 : 0				0.2(*)	0.3(*)	0.2(*)			
16 : 0	5.8 (0.5)	5.8 (0.1)	8.7(1.1)	5.1 (0.1)	5.3 (0.1)	5.3 (0.2)	21.3 (6.1)	15.5 (1.1)	23.0 (3.7)
18 : 0	0.6 (*)	0.6 (*)	2.6 (2.1)	0.2 (*)	0.2 (*)	0.3 (*)	2.5 (1.1)	2.7 (2.2)	2.9 (0.8)
20 : 0	0.3 (*)	0.3 (*)	0.1 (0.2)						
SUM	14	15	19	11	11	12	52	42	51
Monounsaturated									
15 : 1	0.3 (*)	0.3 (*)	0.5 (*)	0.3 (*)	0.2 (*)	0.3 (*)	0.9 (*)	0.3(0.5)	0.4 (0.6)
16 : 1	0.2 (0.3)	0.2 (0.3)		0.3 (*)	0.4 (*)	0.3 (*)			
16 : 1ω7c†	38.9 (2.0)	38.2 (2.6)	37.6 (7.1)	19.4 (0.1)	19.3 (0.2)	19.8 (0.1)	6.0 (0.8)	7.5 (0.4)	6.3 (1.2)
16 : 1ω5*c*	0.5 (*)	0.5 (*)	0.4 (0.1)	0.8 (*)	0.7 (0.1)	0.4 (*)	0.8 (0.2)	0.7 (*)	0.9 (0.2)
16 : 1ω13*t*	1.4 (*)	1.4 (0.1)	1.5 (0.6)	2.3 (0.1)	2.1 (0.1)	2.3 (0.1)	1.0 (0.2)	1.4 (*)	0.6 (0.8)
18 : 1ω9	1.8 (0.2)	2.1 (*)	5.0 (1.3)	0.2 (*)	0.2 (*)	0.2 (*)	20.1 (0.2)	21.1 (0.2)	23.2 (1.6)
18 : 1ω7	4.6 (0.2)	4.3 (0.1)	4.5 (1.3)	4.1 (*)	4.1 (0.1)	3.9 (0.1)	7.3 (*)	7.6 (1.1)	7.9 (0.4)
18 : 1ω5							1.2 (0.6)	1.5 (0.1)	1.5 (0.5)
20 : 1				0.22 (*)	0.22 (*)	0.38 (*)			
22 : 1						0.20 (*)			
SUM	48	47	49	28	27	28	37	40	41

Branched chain									
*i*14 : 1				0.3 (*)	0.2 (*)	0.2 (*)			
*i*14 : 0	0.3 (*)	0.1 (0.2)	0.3 (0.4)						
br 15 : 1				0.1 (*)	0.2 (*)	0.2 (*)			
br 15 : 1				0.4 (*)	0.3 (0.1)	0.2 (*)	0.7 (0.2)	0.8 (0.2)	0.4 (0.5)
*i*15 : 0				0.2 (*)	0.2 (*)	0.2 (0.1)	1.3 (0.4)	1.0 (*)	1.4 (0.2)
SUM	0.3	0.1	0.3	1.0	1.0	0.8	2.0	1.8	1.8
Polyunsaturated									
16 : 4+16 : 3	7.5 (0.8)	7.4 (0.5)	5.1 (0.5)	12.9 (0.2)	12.0 (*)	12.8 (0.1)			
C_{16} PUFA	0.3 (0.5)	0.3 (0.4)	0.2 (0.3)						
16 : 2+16 : 3	4.7 (0.6)	4.7 (0.3)	4.3 (0.2)	8.7 (0.1)	10.0 (0.5)	8.3 (0.1)			
C_{18} PUFA		0.2 (0.3)	0.2 (0.3)	0.4 (0.1)	0.4 (*)	0.4 (0.1)			
C_{18} PUFA	0.4 (*)	0.3 (*)	0.1 (0.2)	0.4 (*)	0.5 (*)	0.4 (*)	0.3 (0.4)	0.9 (0.1)	0.4 (0.5)
18 : 3ω6	2.7 (0.4)	2.0 (0.2)	0.8 (1.1)	3.2 (0.1)	3.0 (*)	3.1 (0.1)	1.4 (0.5)	1.4 (0.9)	0.3 (0.4)
18 : 4ω3	3.3 (0.5)	3.5 (0.4)	3.0 (0.4)	1.7 (0.2)	1.6 (*)	1.5 (*)	3.7 (3.9)	8.5 (1.5)	2.3 (3.2)
18 : 2ω6	0.3 (0.5)	0.3 (0.4)		1.3 (*)	1.2 (*)	1.2 (*)	0.6 (0.1)	1.2 (0.3)	0.4 (0.5)
18 : 3ω3	0.7 (0.4)	0.5 (0.4)	0.9 (1.3)	0.9 (*)	0.9 (*)	0.8 (*)	3.0 (0.8)	4.3 (0.2)	3.3 (0.5)
C_{18} PUFA				0.4 (*)	0.4 (*)	0.7 (*)			
18 : 2		0.3 (*)	1.6 (0.6)						
20 : 5ω3	17.3 (5.1)	17.3 (3.9)	14.4 (0.4)	28.2 (0.1)	28.3 (0.5)	28.6 (0.5)			
22 : 6ω3	0.5 (0.1)	0.5 (*)	0.4 (*)	2.3 (*)	2.3 (*)	2.1 (0.1)			
SUM	38	38	31	60	61	60	9	16	7

Fatty acids are designated as total number of carbon atoms : number of double bonds followed by the position of the double bond from the aliphatic end. The prefixes *i* and *a* indicates *iso*- and *anteiso*- branching, respectively. The suffixes *c* and *t* indicate *cis* and *trans* geometry. Figures in parentheses represent standard deviations of two replicates. *represents standard deviation less than 0.05%. †*Odontella weissflogii* contains some 16 : 2 which co-eluted with 16 : 1ω7c.

Table 4. Sterol composition of three Antarctic marine algae cultured under high UV-B, low UV-B and control treatments

	Percentage composition (%)		
	Treatment		
Sterol (trivial name)	Control	Low UV-B	High UV-B
Odontella weissflogii			
Cholesta-5,24-dien-3β-ol (desmosterol)	23 (3)	26 (3)	28 (2)
24-Methylcholesta-5,24(28)dien-3β-ol (24 methylene cholesterol)	37 (1)	38 (3)	38 (2)
Other	40 (2)	36 (1)	35 (1)
Chaetoceros simplex			
Cholesta-5,22E-dien-3 ol (*trans*-22 dehydrocholesterol)	28 (*)	25	28 (*)
Cholest-5-en-3βol (cholesterol)	16 (0.4)	13	13 (0.1)
24-Methylcholesta-5,22E-dien-3βol (brassicasterol)	52 (0.2)	50	52 (0.1)
24-Ethylcholesta-5,22E-dien-3βol (stigmasterol)	5 (0.2)	5	7 (0.1)
Phaeocystis antarctica			
24-Methylcholesta-5,22E-dien-3βol (brassicasterol)	100	100	100

Figures in parentheses represent standard deviations of two replicates; *represents less than 0.05%.

also decreased by 3% (Table 3). Saturated fatty acids increased from 14 to 19% (Table 3). This was primarily due to an increase in 16 : 0. Although 18 : 0 also increased, it was not statistically significant (Table 3). The monounsaturated fatty acid 18 : 1ω9 also increased under high UV-B (Table 3). The fatty acid profiles for the control and low UV-B treatment were similar and all other fatty acids remained at similar relative concentrations under the three light treatments.

The relative amounts of PUFAs in *P. antarctica* increased in the low UV-B treatment in comparison with the control and high UV-B treatments (Table 3). This was due to an increase in C_{18} PUFAs from 9 (control) to 16 (low UV-B) %. Under high UV-B, they decreased to 6.6% (Table 3). There was a corresponding small decrease in saturated fatty acids under low UV-B for *P. antarctica* (48%), when compared with the control (52%) and the high UV-B treatment (51%) (Table 3). Under high UV-B, 16 : 0 increased in comparison with both the control and the low UV-B treatment (15–23%, Table 3) and saturated fatty acids decreased (Table 3). 18 : 1ω9 also increased slightly at low UV-B irradiances.

Sterols

Although there was an increase in percentage composition in the C_{28} sterol 24-ethylcholesta-5,22E-dien-3-ol (stigmasterol) in *C. simplex*, overall the sterol profiles remained largely unchanged as a result of radiation treatment (Table 4). Although the sterol concentration varied slightly (Table 2), generally the sterols appeared to be unaffected by UV-B irradiance, irrespective of species or irradiance.

DISCUSSION

Avoidance, repair, screening and/or intracellular quenching mechanisms would mediate the susceptibility of individual phytoplankton species to UV-B [21, 23]. The limited number of Antarctic algal species examined to date have defence or repair mechanisms against the damaging physiological effects of UV [9, 12]. *Phaeocystis antarctica* and some Antarctic marine diatoms use molecular mechanisms and pigments to cope with UV radiation [9, 14, 24]. The effect of light on lipids of temperate algae has been shown to be species-specific [19, 25], as has the effect of UV-B irradiance [16–18]. Some temperate algae [*Chaetoceros calcitrans* (Paulsen) Takano and *Skeletonoma costatum* (Greville) Cleve] show no change in their fatty acid content after UV-B radiation [18]. Other species are affected detrimentally [18].

In some algal species, PUFA concentrations have been shown to decrease under UV-B, but can be restored to normal levels during periods of lower UV-B and high PAR [17, 18]. It therefore follows that the diurnal variation in UV radiation and PAR in Antarctica may allow Antarctic phytoplankton to restore essential PUFAs if they become depleted, despite experiencing continuous light during the Antarctic summer. It has also been suggested that ozone depletion over Antarctica for two decades may have selected species or strains that possess greater tolerance to UV-B exposure [15].

Odontella weissflogii survived high levels of UV irradiation at polar temperatures [13, Davidson, A.T., unpublished data]. The cell-size of *O. weissflogii* is an order of magnitude larger than the other phytoplankton species used in our study. The large cell-size, resultant slower division rate and low concentrations

of *O. weissflogii* cells meant that no significant growth could be discerned during the 2 days of experimental irradiance. In our study, *O. weissflogii* contained the highest cell lipid concentrations of the three algae under all treatments. The fatty acid composition of *O. weissflogii* did not change and PUFAs remained high across all treatments. However, there was a reduction in lipid concentration in *O. weissflogii* under high UV-B. This decrease, though small, may be important for higher trophic levels. A relatively less abundant but large cell may comprise a significant proportion of the carbon standing stock. Thus, a small decline in cell lipid concentration of such a species could significantly alter the overall nutritional quality of the phytoplankton community.

UV-B exposure has been shown to reduce biosynthesis of essential fatty acids rather than the growth of temperate phytoplankton [18]. This indicates that although algal growth continues unabated in some UV tolerant species, the nutritional content may decline. The production of the essential fatty acids, $20:5\omega3$ and $22:6\omega3$, appears to decline after UV irradiance in many UV-sensitive temperate species [17, 18]. In some species, the reduction in fatty acid concentration was based solely on the decrease in PUFA production [17]. A temperate strain of *C. simplex* has been shown to double the production of $20:5\omega3$ between high and low natural PAR conditions, respectively [19], reflecting a relationship between light and fatty acid composition. Our data showed that PUFA concentrations in *C. simplex* did not change markedly under the two UV treatments, but PUFA percentages decreased in *C. simplex* under high UV-B. The increase in saturated fatty acid concentrations under high UV-B exposure indicates oxidative degradation of total lipid to form free fatty acids. The variation noted between the temperate strain of *C. simplex* [19] compared with the Antarctic strain reported here may be attributed to the species coming from different regions and incubation temperatures as polyunsaturated fatty acids were much higher in the polar species.

The tolerance to UV-B exposure by phytoplankton cells changes with life stage with cells more vulnerable during logarithmic growth [18]. It is likely that differing UV-B sensitivity between life stages contributed to changes in the cellular lipid concentration of *P. antarctica*. Under high UV-B, the flagellate phase was absent. These results concur with the study by Marchant *et al.* [5] that reported that the flagellate phase died under similar high UV treatment; the colonial phase was more tolerant of UV-B exposure. The decrease in flagellates under UV radiation has also been observed in other long-term studies [26]. However, the high standard deviation between replicates and the potential inclusion of dead cells in the lipid profile could also have contributed to the observed results.

Growth of *P. antarctica* is enhanced by exposure to near surface Antarctic UV-B irradiances [14, 26]. The total lipid profile and higher PUFA composition for *P. antarctica* under low UV-B support these findings. Studies of the nutritional characteristics of the flagellate vs colonial stage have reported that animals avoid the consumption of colonial *P. antarctica* [28]. *Phaeocystis antarctica* is one of the first species to proliferate in Antarctic waters, forming shallow blooms in the marginal ice zone as it retreats southward during spring and summer [5, 22]. Thus, *P. antarctica* occupies a habitat where it will be susceptible to UV-B exposure [5]. The effect of the susceptibility of the flagellate stage on algal species composition and grazers is yet to be ascertained.

In our study, C_{18} PUFAs, $18:3\omega3$ and $18:4\omega3$, increased in *P. antarctica* when exposed to low UV-B irradiance. A similar increase was noted in $18:3\omega3$ in Antarctic field ice diatom communities exposed to increasing ambient light (McMinn, A. *et al.*, 1997; unpublished data). These observations suggest that biosynthesis of C_{18} PUFA, for certain species of algae may be enhanced under higher UV light intensities.

Sterols are membrane constituents whose structure can absorb in the UV-B region of the spectrum. Thus, they could be expected to be affected by UV-B radiation. However, no significant changes were observed in the present study.

Lipids of the three phytoplankton exhibited the same or higher degrees of unsaturation under low UV radiation when compared with control cultures. Increased production of $20:5\omega3$ under elevated UV-B has been noted previously for the diatom, *Ditylum brightwellii* (West) Grunow [16]. However, the increase in C_{18} PUFAs observed in our study for *P. antarctic* has not been reported previously. The lack of change in the fatty acid profile for *C. simplex* and *O. weissflogii* under low UV-B indicates substantial tolerance to UV-B irradiance. Our results, like those of Davidson and co workers [14, 15, 27], indicate that such UV-B fluxes may be beneficial. The three polar marine phytoplankton species examined in our study are resistant to exposure to UV-B irradiances similar to Antarctic near-surface fluxes at the summer solstice. However, all three algae showed signs of stress under high UV-B, as indicated by an decrease in the total lipid concentration in *O. weissflogii* and increase in free fatty acid (i.e. degradation products) in *C. simplex* and *P. antarctica*. This was not due to changes in cell-size, because no significant changes in cell volume were observed.

UV-B impacts directly upon the planktonic community by affecting growth, production and survival [15, 29, 30], thereby affecting the quantity and nature of food available to higher trophic levels. Lipids are an essential component in the nutrition of consumers [18, 19]. Our experiments were conducted under conditions of nutrient replete culture and artificial UV-B irradiance conditions. However, results showed that UV-B may alter the lipid composition of phytoplankton and that the magnitude of these changes was species-specific. Thus, the structure and function of

the Antarctic marine food web may also be affected by UV-B-induced changes in the quality of food source for the higher trophic levels.

EXPERIMENTAL

All measurements of irradiance were made with an International Light IL 1700 Radiometer equipped with broad band detectors to measure PAR, UV-A and erythermal UV-B [14]. A National Institute of Standards and Technology intercomparison package (NIST Test #534/240436-88) was used to calibrate each light sensor.

Unialgal cultures of the diatoms *Odontella weissflogii* (Janison) Grunow and *Chaetoceros simplex* Ostenfeld were isolated from sea ice collected in Prydz Bay, Antarctica, during the 1990/91 austral summer. *Phaeocystis antarctica* (Karsten) was isolated from Prydz Bay in the 1982/83 summer season. Cultures of diatoms and *P. antarctica* were maintained in 2 l glass flasks using f/2 growth medium [31] and GP5 medium [32], respectively, at $2 \pm 1^{\circ}$. Cool white fluorescent lights provided photosynthetically active radiation (PAR) intensity of 17.08 $Jm^{-2}\ s^{-1}$ (84.7 $\mu Em^{-2}\ s^{-1}$), with no UV-B enhancement, on a 12 h light : 12 h dark cycle. Immediately before experimental irradiation, three replicate subsamples of ca 15 ml were obtained from each parental culture and fixed with Lugol's iodine, a known sample volume sedimented and cells counted over 15 replicate fields using a Labovert inverted microscope. Mean cell concn and standard deviation were computed.

Each exponential growth phase parental culture was thoroughly mixed and 3 replicate 300 ml Costar polystyrene culture flasks (which completely absorbed wavelengths below 295 nm) established for each light treatment (control, low and high UV exposures). Cultures were irradiated for 24 h in a 48 h experimental period (6 h light : 12 h light : 12 h dark : 6 h light) [14, 24]. Exposures were conducted in a Thermoline controlled environment cabinet at $2 \pm 1^{\circ}$ with cool white fluorescent tubes to provide PAR and UV-A (320–400 nm), with UV-B provided by FS20T 12 UV-B Westinghouse sunlamps. PAR and UV-A irradiances were $16.3 \pm 0.7\ Wm^{-2}$ ($81.3 \pm 3.4\ \mu Em^{-2}\ s^{-1}$) and $4.39 \pm 0.20\ Wm^{-2}$, respectively. The spectral distribution and UV-B irradiance were varied by attenuation with glass filters [5] to provide low (0.37 Wm^{-2}) or high UV-B (1.59 Wm^{-2}). Sensors were each covered by an attenuating glass screen and a single layer of Costar culture flask to measure the experimental irradiances to which the algae were exposed. UV-B irradiances were chosen to reflect less than (74%) and greater than (318%) peak UV-B exposure as measured at an Antarctic coastal site [unpublished data : Casey station, 66°S, Roy, C. (1994)].

Following irradiation, each culture was well mixed and ca 15 ml was fixed with Lugol's iodine for subsequent estimation of cell concn (as above). Chlorotic and greatly vesicularised cells were considered to be dead [24]. The remainder of each experimental culture was filtered through Whatman GF/F filters. On completion of filtration, the filters were stored at -20° overnight before extraction of lipids the following day.

Replicate lipid extractions were performed as described in ref. [33]. A subsample of the total lipid extract was analysed with an Iatroscan Mark V TH-10 TLC-FID analyser [34]. The sterol frs were obtained by saponification of another subsample and were converted to TMSi ethers by reaction with *bis*(trimethylsilyl)trifluoroacetamide. Another sub-sample was taken for direct trans-esterification with MeOH–HCl–$CHCl_3$ (10 : 1 : 1, 3 ml) to produce the corresponding fatty acid Me esters which were extracted with Hexane–$CHCl_3$ (4 : 1). GC and GC-MS conditions were as described in ref. [33] and ref. [35], respectively.

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