

## Consequences of UV-enhanced community respiration for plankton metabolic balance

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### Abstract

The net community production (NCP) of plankton communities affects their role as sources or sinks of atmospheric CO<sub>2</sub>. Most estimates of NCP have been made by enclosing communities in bottles, generally glass borosilicate, that remove ultraviolet (UV)B and part of UVA wavelengths. A series of experiments were conducted to test whether NCP values from communities incubated excluding UVB (+ part of UVA) radiation (i.e., in glass borosilicate) differ from those of communities receiving the full solar radiation spectrum (i.e., incubated with quartz bottles) and to explore the effect of UV radiation on the respiration rates and bacterial production in these communities. Plankton NCP tended to be 43% lower, on average, when the rates were measured under full solar radiation than when UVB (+ part of UVA) was removed. Dark respiration was significantly enhanced after exposure to the full solar spectrum for most communities, showing lower values when previously incubated in a light environment free of UVB (−50%) or in the dark (−62%). Bacterial production was inhibited by natural sunlight but increased, as observed for community respiration, when transferred to the dark. Communities previously exposed to full solar spectrum showed the greatest increase in bacterial production when allowed to recover in the dark. The net result of these responses were an increase in community respiration and decline in net community production over 24 h, indicating that UVB radiation plays a major role in the metabolic balance of the ocean's surface ecosystem.

Solar ultraviolet (UV) radiation, in particular the UVB band (280–315 nm), has deleterious effects in marine organisms (Häder et al. 2007; Llabrés et al. 2013), affecting both photosynthesis (Cullen et al. 1992) and respiration of plankton communities (Larkum and Wood 1993; Banaszak 2003). Therefore UVB has the potential to affect net community production (NCP), the net balance between production and respiration in the community, which is a fundamental property describing the role of plankton communities in carbon balance, by indicating whether they act as sinks for CO<sub>2</sub> and sources of organic matter (autotrophic communities, NCP > 0) or as sources of CO<sub>2</sub> to the atmosphere and sinks of organic matter in the system (heterotrophic communities, NCP < 0; Duarte et al. 2011). At present, there are few published assessments on the effect of UV irradiance on plankton NCP (Vidussi et al. 2011; Godoy et al. 2012), and most estimates from oceanic and freshwater systems derive NCP through the incubation of plankton in borosilicate glass Winkler bottles, which filter out UVB and part of UVA radiation. Thus, most published data on NCP have been obtained by removing UV (mostly the UVB band) radiation during incubation, which may bias estimates of plankton metabolic balance in surface waters. Indeed, Godoy et al. (2012) reported that removal of UVB radiation affected NCP estimates in the southeastern Pacific, typically leading to the overestimation of NCP. However, Godoy et al. (2012) also report that UVB radiation can increase planktonic NCP.

Predicting the effect of UV radiation (UVR) on plankton metabolic balance is cumbersome because, in addition to its direct effects on the inhibition and reduction of photosynthetic

and community respiration rates, metabolic imbalances may derive from differential sensitivities to UVB among planktonic organisms within the community (De Lange et al. 2003; Yuan et al. 2011). Additionally, photochemical transformations of dissolved organic matter (DOM) mediated by UVR (Zepp et al. 2007) can influence the availability of DOM to bacteria (Tranvik and Bertilsson 2001), potentially increasing respiration rates. Hence, predicting how UV and increased UVB radiation from ozone reduction might influence planktonic NCP remains an elusive goal.

Here, we experimentally evaluate the effect of UVR (mostly UVB) on net community production and community respiration of planktonic communities from oceanic surface waters. We do so on the basis of two series of complementary experiments, each conducted in contrasting regions of the ocean (subtropical North Atlantic, Southern Ocean, and Mediterranean Sea): (1) a set of experiments evaluating the response of NCP to removal of ambient UVB and (2) a set of experiments evaluating the response of dark respiration following exposure of the community to different solar radiation fields: full solar spectrum, removal of UVB, and darkness. In the second set of experiments, we also evaluate the response of bacterial production to exposure to the different light environments tested.

### Methods

The experiments were performed during the Antarctic tourism opportunity spectrum (ATOS) Antarctica cruise (24 January 2009–02 March 2009, Southern Ocean) and RODA II (05 February 2007–25 February 2007, subtropical northeast Atlantic Ocean), oceanographic cruises on board the Spanish R/V *BIO Hesperides*, and in the

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Table 1. Locations, dates, and water characteristics of the sampling stations. Asterisks indicates the respiration experiment stations.

	Station	Date	Latitude	Longitude	Chl <i>a</i> ( $\mu\text{g L}^{-1}$ )	Temperature ( $^{\circ}\text{C}$ )
Atlantic Ocean	T2*	09 Feb 2007	26°24.99'N	18°08.34'W	0.21	20.08
	T6*	12 Feb 2007	21°49.14'N	20°55.60'W	0.44	20.45
	T9*	14 Feb 2007	20°10.55'N	22°01.74'W	0.90	21.07
Southern Ocean	4*	31 Jan 2009	62°23.65'S	51°15.50'W	0.44	-0.08
	7*	02 Feb 2009	65°00.70'S	55°27.44'W	25.0	-0.24
	8	03 Feb 2009	64°16.20'S	55°51.54'W	18.2	-0.17
	17*	13 Feb 2009	67°14.70'S	70°30.23'W	0.55	0.79
Mediterranean Sea	Faro1*	29 Jul 2009	39°15.52'N	3°31.55'E	0.12	25.4
	Faro2*	27 Aug 2009	39°15.52'N	3°31.55'E	0.28	28.0
	Faro3	05 Oct 2009	39°15.52'N	3°31.55'E	0.21	25.1

community of an oligotrophic coastal area off Majorca Island (Faro de Cap Ses Salines, northwest Mediterranean Sea; Table 1).

Surface water (1 m depth in the Southern Ocean and Mediterranean Sea, 5 m in the Atlantic Ocean) was sampled using 30 liter Niskin bottles or a 5 liter Niskin bottle in the Mediterranean Sea. Chlorophyll *a* (Chl *a*) concentration were determined by filtering 250 mL of samples through Whatmann GF/F filters, extracted for 24 h with 90% acetone. Chl *a* concentration was derived from the fluorescence of the extracts measured using a Shimadzu RF-5301 fluorometer (Yentsch and Menzel 1963).

For metabolism measurements, seawater was carefully siphoned into five 100 mL quartz bottles and into 21, 125 mL narrow-mouth glass Winkler bottles. Seven replicates of the glass Winkler bottles were used to determinate the initial oxygen concentration, and seven replicate bottles were used for incubation in the "dark" and in the "light," respectively. The five quartz bottles were also incubated in the light. Quartz Winkler bottles allow the entire solar spectrum to pass through, whereas glass Winkler bottles remove the UVB band and reduced the transmittance at the shorter UVA wavelengths (50% transmittance at 317 nm). The communities were incubated on deck for 24 h in a large 2000 liter tank with continuous circulation of surface seawater to maintain the temperature of the surface waters and natural solar radiation. Bottles were covered with neutral screens to reduce incident radiation by 20%, in addition to the 8% to 12% reduction in photosynthetically active radiation transmittance by the bottles themselves. The light conditions experienced by the incubated communities mimic those in the upper layer of the ocean but represent only an approximation, in that changes in the light environment due to mixing were not reproduced. Oxygen concentration was determined by Winkler titration using a potentiometric electrode and automated endpoint detection (DL28 titrator, Mettler Toledo; Oudot et al. 1988). The average coefficient of variation of O<sub>2</sub> concentration determinations was 0.17%. Community respiration (R) and NCP rates were calculated from changes in dissolved oxygen concentration after incubation of samples in dark and light conditions, respectively, relative to the initial oxygen concentration.

Experiments to evaluate the effect of exposure to solar radiation on community respiration were conducted in

parallel with the incubations for community metabolism (Fig. 1) at most of the stations (Table 1), except for Sta. 7, where sampling could not be completed because of heavy sea conditions. Seawater was incubated on deck in 2 liter bottles, in a tank as indicated above, during day hours under different light treatments, including incubation in quartz (full solar radiation spectrum), polycarbonate (UVB and part of UVA removed), and black bottles (darkness). At sunset ( $t_0$ ; Fig. 1), the water was distributed in 12 replicate Winkler bottles; six of these bottles were used to measure the initial concentration of dissolved oxygen, and the other six were incubated in the dark for 15 h (including nighttime + 2–3 h) to measure dark community respiration after each light treatment. After incubation ( $t_1$ ; Fig. 1) dissolved oxygen was measured using micro-Winkler techniques, as indicated above, to calculate oxygen consumption by respiration.

A blank was run in both Antarctic and Mediterranean waters to determine the oxygen consumption attributable to photooxidation and other abiotic processes. A 2 liter water sample was filtrated through a 0.2  $\mu\text{m}$  pore filter to remove all organisms, and the filtrate was dispensed into a large carboy, which was shaken and allowed to sit for 1 h before distributing the water in five 100 mL quartz Winkler and five glass Winkler bottles. Initial oxygen concentration was determined from the five glass Winkler bottle replicates, and the five quartz bottles were exposed to incident sunlight during 7 h of incubation (at in situ temperature) during day hours, to examine possible changes in oxygen concentration. No detectable oxygen consumption was observed during the blank incubations, showing a small ( $0.55\% \pm 0.05\%$ ), nonsignificant ( $t$ -test;  $t = 0.0015$ , degrees of freedom (df) = 9,  $p = 0.19$ , and  $t = 0.002$ , df = 9,  $p = 0.42$ , for Antarctic and Mediterranean waters, respectively) increase of oxygen concentration after the incubation.

Bacterial production (BP) was measured at the beginning and end of the day and night periods of incubation in the different light treatments (Fig. 1), as we did for respiration incubation for Atlantic and Antarctic communities and in the Mediterranean communities sampled in August 2009. Bacterial production was estimated from the incorporation of <sup>3</sup>H-labeled leucine as described by Smith and Azam (1992). Briefly, five replicate subsamples (1.2 mL each) from the 2 liter bottles were collected into 2 mL centrifuge vials. Two of the replicates (blanks) were killed immediately

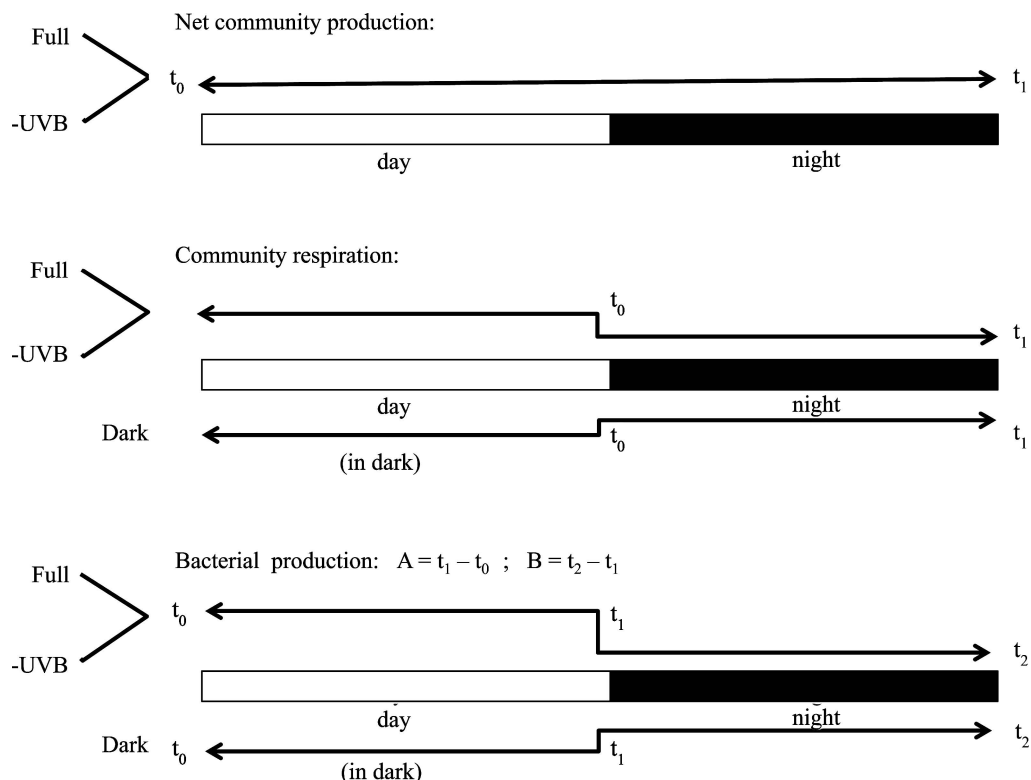


Fig. 1. Schematic representation of the experimental design, including treatments in which communities received full solar radiation (Full), UVB was removed (-UVB), and communities were kept in the dark (Dark). Arrows span the entire experimental period, and  $t_0$ ,  $t_1$ , and  $t_2$  represent the sequential sampling events in which measurements of oxygen concentration (for calculation of NCP and R between  $t_0$  and  $t_1$ ) and bacterial production (estimated at each of  $t_0$ ,  $t_1$ , and  $t_2$ ) were conducted.  $A = t_1 - t_0$ , and  $B = t_2 - t_1$ , represent the difference in BP for the day and night hours, respectively. Bars indicate the daytime (open) and nighttime (closed) periods.

by adding a concentrated solution of trichloroacetic acid (TCA) to a final concentration of 5%. Subsequently,  $^3\text{H}$ -leucine was added to all five subsamples to a final concentration of  $40 \text{ nmol L}^{-1}$  and incubated for 1–2 h at in situ temperature. To minimize isotope use, while maintaining the sensitivity of the assay, the commercial solution of  $^3\text{H}$ -leucine was diluted to a specific activity of  $0.7 \text{ TBq mol}^{-1}$  using unlabeled leucine. The incubation was terminated by adding TCA (5% final concentration) to the remaining three live subsamples. The tubes were then centrifuged ( $13,000 \times g$ , 10 min), and the supernatant containing the unincorporated tracer was discarded. The pellets, containing the microbial biomass, were washed twice by sequentially adding 1 mL of 5% TCA, centrifuging ( $13,000 \times g$ , 10 min), and aspirating the supernatant. The amount of radioactivity incorporated in the samples was measured in a liquid scintillation counter (Wallac 1414) after thoroughly mixing the samples with 1 mL of liquid scintillation cocktail. The rates of leucine incorporation were calculated by subtracting the average radioactivity measured in the killed blanks from that measured in the living samples and dividing the resulting number by the incubation time. The different intervals of sampling for the BP measurements are represented in Fig. 1. NCP was incubated for 24 h, including the day and night period of each sampling area. Dark respiration was incubated during the night hours, after the day period (Fig. 1). The

differences in BP were calculated for the day interval (interval  $A = t_1 - t_0$ ; Fig. 1) and for the night hours, similar to dark respiration incubations (interval  $B = t_2 - t_1$ ; Fig. 1). The net BP change calculated for the total incubation included the day and night periods (net bacterial production =  $\text{BP}_{t_2} - \text{BP}_{t_0}$ ; Fig. 1).

We used *t*-test, Pearson correlation, and ANOVA (JMP program) to analyze the significance of differences observed between treatments and measurements.

## Results

The NCP of seven of the nine communities tested, three at each location, was enhanced when UVB was removed by enclosing the communities in glass, instead of quartz, bottles (Fig. 2). The increase in NCP upon removal of UVR averaged  $0.57 \pm 0.23$  (19%),  $0.56 \pm 0.20$  (48%), and  $1.83 \pm 0.52$  (63%)  $\mu\text{mol O}_2 \text{ L}^{-1} \text{ d}^{-1}$  for the subtropical northeast Atlantic, the Mediterranean Sea, and the Southern Ocean communities, respectively (Fig. 2). In one of the stations in Antarctic waters and during both summer measurements in the Mediterranean Sea the NCP values measured were negative, indicative of net heterotrophic communities (Fig. 2). Incubation of these communities under full solar radiation (quartz bottles) resulted in more net heterotrophic NCP values than those observed when removing UVB using glass bottles (Fig. 2). Experiments

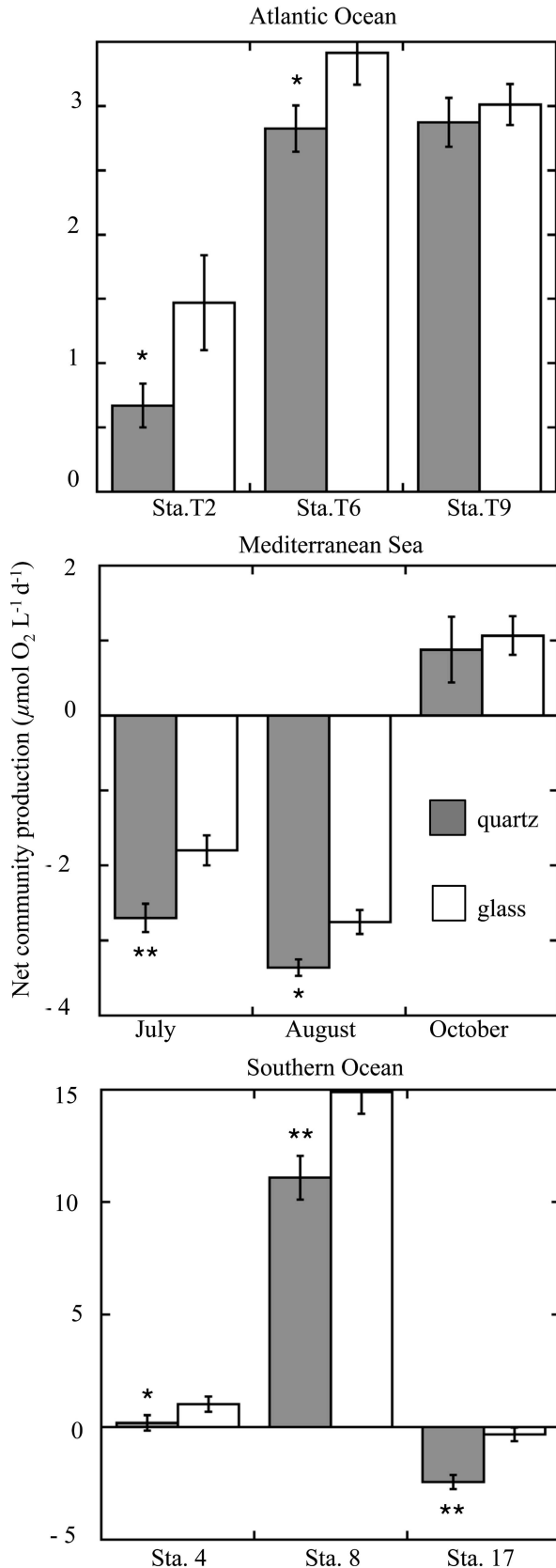


Fig. 2. Net community production rates observed for surface waters during incubation of planktonic communities under full solar radiation (grey columns, quartz bottles) and under solar

examining the responses of dark community respiration after exposure to different solar radiation conditions showed that community respiration rates differed in response to prior solar radiation regimes. In general, the highest dark respiration rates were observed when communities were previously exposed to full solar radiation (i.e., when plankton had been incubated in the light in quartz bottles; Fig. 3). Dark respiration of communities in the subtropical northeast Atlantic, the Mediterranean Sea, and in two of the three experiments run in the Southern Ocean was consistently higher after exposure to the full solar spectrum compared with communities incubated in the absence of UVB and part of UVA radiation (Fig. 3). Communities from the Southern Ocean showed more variability in dark respiration responses, with the community of the more productive Sta. 7 showing inhibition of dark respiration under the full solar spectrum, contrasting with the homogeneous responses in the respiration of communities from the subtropical northeast Atlantic (Fig. 3). In most of the experiments, dark respiration tended to show the lowest values when the community had been maintained previously in the dark for several hours (Fig. 3). The increase in dark respiration after exposure to the full solar radiation spectrum averaged 91%, 28%, and 49% for the subtropical northeast Atlantic, the Mediterranean Sea, and the Southern Ocean, respectively, when compared with dark respiration measured after removing UVB by glass borosilicate (Fig. 3).

BP was also highly dependent on the light history of the communities and declined when exposed to light ( $A = t_1 - t_0$ , as indicated in Fig. 1), showing the highest decline when exposed to the full solar spectrum (Fig. 4). Conversely, bacterial production increased after the dark period ( $B = t_2 - t_1$ , as indicated in Fig. 1), with the communities previously exposed to full solar radiation conditions showing the highest increases in BP (Fig. 4a–d,g), except for some of the Antarctic communities that showed higher variability (Fig. 4e,f). Changes in bacterial production were generally smaller in the communities that had not been previously exposed to light and remained in darkness for the whole experimental period (Fig. 4a,b,d,e,g). The enhancement in bacterial growth after recovery in the dark showed a compensatory response to the extent of inhibition in BP on exposure to UVB radiation, as indicated by a significant negative correlation (Pearson, log-transformed values,  $p < 0.05$ ) between the percent remaining BP after UVB exposure and the percentage of BP recovered after dark incubation. However, a significant net increase in BP was observed when integrating BP over the light and dark periods ( $BPT_2 - t_0$ ; Fig. 1) for the communities exposed to full solar radiation (Fig. 4) in the Mediterranean Sea ( $t$ -test;  $t = 0.052$ ,  $df = 9$ ,  $p < 0.05$ ) and for those in the northeast

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radiation with UVB excluded (white columns, glass borosilicate bottles). Columns with asterisks represent significantly lower NCP under full solar radiation ( $t$ -test; Sta. T2:  $t = 0.8$ , Sta. T6:  $t = 0.59$ ; August:  $t = 0.9$ , Sta. 4:  $t = 0.82$ ,  $df = 10$ , \*  $p < 0.01$ ; July:  $t = 2.11$ , Sta. 8:  $t = 3.5$ , Sta. 17:  $t = 2.5$ ,  $df = 10$ , \*\*  $p < 0.001$ ).

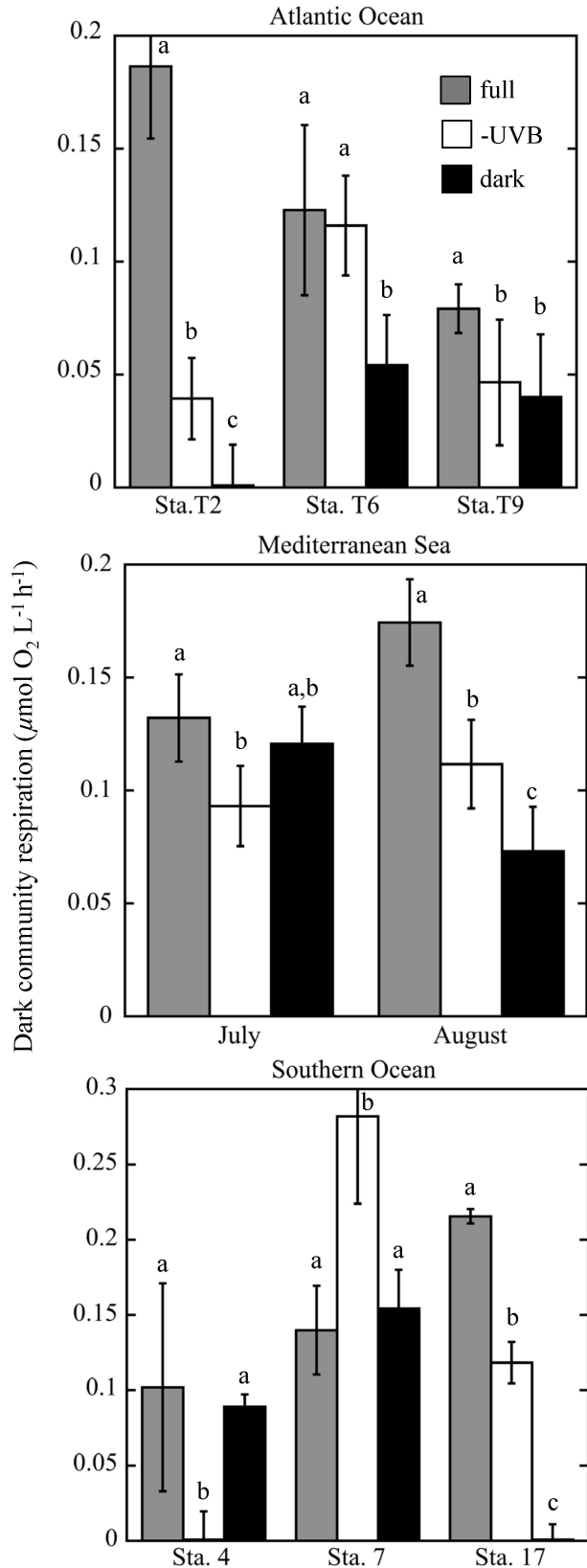


Fig. 3. Dark community respiration rates observed in communities previously exposed to different light treatments: Full solar radiation (grey columns), UVB removed (white columns), darkness (black columns). Columns connected by the

Atlantic Ocean (*t*-test; Sta. T2:  $t = 0.075$ , Sta. T6:  $t = 0.003$ , Sta. T9:  $t = 0.1$ ,  $df = 9$ ,  $p < 0.001$ ; Fig. 4a–d) that was parallel to the observed increase in respiration rates (Fig. 3). Hence, the recovery in BP after a dark period overcompensated for the preceding inhibition of BP when the community was exposed to full solar radiation. Responses for Antarctic communities were more variable, with a net decrease in BP under total solar radiation in the more productive Sta. 7 (Fig. 4f), consistent with the lack of increase in community respiration under full solar radiation at that station (Fig. 3).

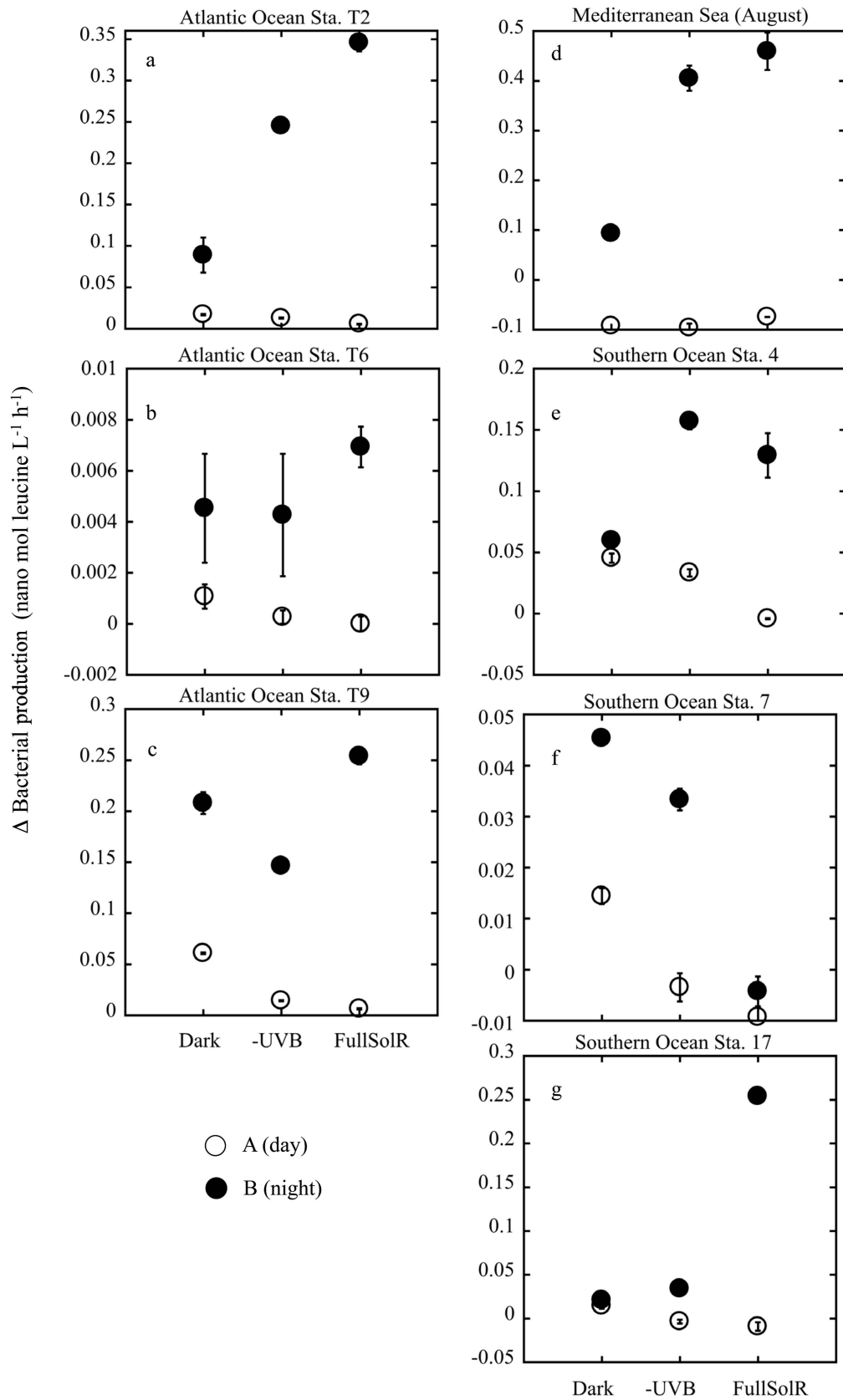
## Discussion

The results showed evidence of major influences of UVR on community respiration, bacterial production, and NCP. The net community production in surface waters tended to be 43% higher, on average, when UVR was excluded during the incubations than when communities were incubated under the full solar spectrum. Moreover, community respiration was significantly affected by light conditions, and communities exposed to the ambient light field (full solar spectra) subsequently supported a greatly enhanced dark respiration, 62% and 50% higher on average, compared with communities maintained in the dark and when UVR was excluded, respectively. The results showed that, as with community respiration, bacterial production was also greatly affected by the light history of the community.

There is still a paucity of studies addressing the effect of the full solar spectrum, including the UVB band, in the metabolic balance of planktonic communities. Yet, higher gross primary production estimates derived using  $^{18}\text{O}_2$  additions than estimated using bulk oxygen changes (González et al. 2008) is generally interpreted as resulting from enhanced respiration in the light, as the bulk oxygen method assumes respiration in the dark and the light to be equal (Bender et al. 1987; Grande et al. 1989). Godoy et al. (2012) showed recently that UVB radiation generally inhibited NCP in the southwest Pacific off Chile and found significantly different NCP rates between communities incubated under full ambient radiation and those incubated under reduced UVB (and partially reduced UVA) as observed here. Evaluation of the effects of UVR on plankton processes indicates that UVB inhibits photosynthesis (Cullen and Neale 1994), reducing oceanic primary production by about 15% (Cullen et al. 1992). UVB may affect respiration by damaging deoxyribonucleic acid (DNA), proteins, and cell membranes of heterotrophic plankton and has been shown to inhibit bacterial production (Herndl et al. 1993; Banaszak 2003). In contrast,

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same letter are not significantly different: *t*-test;  $df = 11$ ; Sta. T2: (a–b)  $t = 0.14$ ,  $p < 0.001$ ; (a–c)  $t = 0.18$ ,  $p < 0.001$ ; Sta. T6: (a–b)  $t = 0.11$ ,  $p < 0.05$ ; Sta. T9: (a–b)  $t = 0.03$ ,  $p < 0.05$ ; July: (a–b)  $t = 0.04$ ,  $p < 0.05$ ; August: (a–b)  $t = 0.06$ ,  $p < 0.05$ , (a–c)  $t = 0.1$ ,  $p < 0.001$ ; Sta. 4: (a–b)  $t = 0.11$ ,  $p < 0.01$ ; Sta. 7: (b–a)  $t = 0.24$ ,  $p < 0.05$ ; Sta. 17: (a–b)  $t = 0.09$ ,  $p < 0.001$ , (a–c)  $t = 0.17$ ,  $p < 0.001$ .



phytoplankton respiration appears to be relatively unaffected by UVR (Larkum and Wood 1993; Heraud and Beardall 2002). This is in agreement with our results showing that the effect of UVB on heterotrophic processes may be much larger than on primary production.

The effects of UVR on heterotrophic activity are complex, for bacterial production is suppressed in the presence of UVR (Herndl et al. 1993; Arrieta et al. 2000) but greatly enhanced in the dark after exposure to ambient UVR levels for most communities studied here and elsewhere (Kaiser and Herndl 1997; Arrieta et al. 2000). The enhanced community respiration and heterotrophic bacterial activity associated with exposure to UVR is most likely mediated by photochemical processes affecting the availability of DOM for bacteria.

DOM undergoes UV-induced transformations that may increase DOM bioavailability (Moran and Zepp 1997; Zepp et al. 2007), thereby enhancing heterotrophic bacterial activity (Kaiser and Herndl 1997; Tranvik and Bertilsson 2001; Anesio et al. 2005). The increased lability of DOM may overcompensate for the inhibitory effect of exposure to UVR on bacterial activity when UVR is reduced during the night, since photochemical transformations of organic matter have been reported to increase both bacterial cell numbers and activity (Lindell et al. 1995; Tranvik and Bertilsson 2001; Anesio et al. 2005). Our results agree with these observations, showing a correlation between the intensity of UV damage to BP and the magnitude of the subsequent dark recovery, suggesting that high radiation doses during the day may result in enhanced heterotrophic activity during the night. In addition to the photochemical effects of UVR on DOM, an effect of UVR on phytoplankton release of dissolved organic carbon, associated with phytoplankton cell mortality (Agustí and Duarte 2013), is also a possibility. High cell death rates of UVR-induced picophytoplankton mortality in oligotrophic waters of the Atlantic (Llabrés and Agustí 2006; Agustí and Llabrés 2007) and Mediterranean Sea (Llabrés et al. 2010) may be able to sustain enhanced bacterial heterotrophic activity when UV stress is relieved during the night hours, as observed in our results.

Our results provide an understanding of the processes involved in reducing NCP after exposure to UVB, as we demonstrate that respiration and BP are enhanced in the dark after exposure to UVB radiation, overcompensating for the inhibition of BP when communities are exposed to the light. Hence, exposure to solar radiation, particularly UVB radiation, suppresses heterotrophic activity during the day but enhances it to a larger extent during the night. Hence, the traditional interpretation of higher gross primary production (GPP) estimates derived from  $^{18}\text{O}_2$  compared with bulk oxygen—that dark respiration underestimates diel

respiration in plankton communities (Bender et al. 1987; Grande et al. 1989)—is correct; however, this does not result from higher respiration in the light, but from enhanced dark respiration in communities that cycle from light to dark with the photoperiod. Exposure to UVR resulted in a net increase of BP in most of the Atlantic and Mediterranean communities, since the increased BP in dark hours overcompensated for the inhibitory effect of UVR exposure during daylight hours. However, the results showed great variability in the Antarctic waters, with a net inhibitory effect on BP in the most productive community, parallel to a lack of increased respiration under full spectrum solar radiation exposure.

The results presented here have important practical consequences for the interpretation and reliability of estimates of net metabolism and respiration of plankton communities and bacterial production. Net community metabolism is typically assessed from oxygen changes in communities enclosed in glass Winkler bottles, which block UVB radiation. The results presented here suggest that net community production derived in this manner should yield overestimates of NCP for the upper photic layer, because net community production increases when UVR, which is part of the ambient light field in surface ocean waters, is excluded. Our findings explain the observations of a general suppression of NCP by UVB radiation recently reported by Godoy et al. (2012). Although community respiration is typically measured in the dark (Robinson and Williams 2005), our results demonstrate that community respiration in the dark is greatly enhanced after periods of exposure to the full solar spectrum in surface waters. Dark respiration is typically measured in communities kept in the dark for 24 h (Robinson and Williams 2005), thereby excluding periodic exposure to solar radiation along the 24 h cycle. Moreover, many investigators sample at dawn, so that communities sampled in this manner may be deprived of solar radiation for more than 30 h, likely leading to an even greater underestimation of dark respiration. Our results suggest that these procedures lead to a severe underestimation of dark respiration and, thereby, GPP, which is typically calculated as  $\text{NCP} + R_{\text{dark}}$ . Pringault et al. (2007) discussed the effect of light on respiration and how dark incubations must result in an underestimation of GPP, but their experiments excluded UVR and therefore did not show the full effect of exposure to solar radiation on respiration.

Our experiments also suggest that bacterial production can vary by up to fourfold, depending on the time of sampling, with the lowest estimates obtained at the end of the light cycle after exposure to the full solar spectrum and the highest estimates obtained at dawn. These results are consistent with reports of sizeable diel variability in bacterial production in surface Mediterranean waters,

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Fig. 4. (a–g) Changes in bacterial production (BP,  $\text{nmol leucine L}^{-1} \text{h}^{-1}$ ) in the different light treatments (Dark = dark control, –UVR = UVR removed, and FullSolR = full solar radiation) for each sampling station. Open circles represent the change in BP during the light hours (BP measured at sunset minus BP measured at the preceding sunrise; i.e., day period A in Fig. 1), and solid circles represent the change in BP during the night hours (the difference in BP between the end and beginning of the night period; i.e., night period B in Fig. 1).

involving changes comparable to the fourfold differences derived experimentally here (Gasol et al. 2008). Our results provide an explanation for the observed changes in natural communities in surface waters through enhancement of bacterial activity in the dark period after exposure to UVR. However, our incubations do not include the effect of mixing that may result in changes in the light environment experienced during the daytime, which might reduce the inhibitory effect of UVR on BP (Bertoni et al. 2011). Mixing is difficult to reproduce in incubations, but a special device was used by Bertoni et al. (2011) to examine the effect of vertical mixing on UVR inhibition of BP in the Mediterranean Sea. As observed elsewhere (Herndl et al. 1993), UVR inhibited BP during the daytime, but simulation of mixing resulted in lower inhibition than incubations at a fixed depth (Bertoni et al. 2011). These results imply that reliable extrapolation of measurements of bacteria production in surface waters to daily rates should be based on multiple samples targeting the critical periods (dawn, noon, dusk) encompassing the variability in the exposure of the communities to solar radiation.

Although our study is limited to surface (1–5 m) waters, the effect of UVB on community respiration and NCP in oceanic waters could affect layers deeper than the upper surface considered here. Recent studies demonstrated UVB penetration in the oligotrophic ocean to be much higher than previously thought (Morel et al. 2007; Tedetti et al. 2007), with the description of the most transparent oceanic waters to UVB located at the South Pacific (Morel et al. 2007; Tedetti et al. 2007), where UVB and UVA penetrate down to 28 and 59 m at 305 and 325 nm, respectively (10% irradiance depths; Tedetti et al. 2007). Therefore, the effects of UVB on NCP described here are likely to have broader effect on the oligotrophic waters in the subtropical Southern Hemisphere, which combines high UVB penetration with high incident doses. A recent study by Bertoni et al. (2011) indicates that mixing within the mixed layer modulates the exposure of natural planktonic communities to UVR, subjecting planktonic organisms and DOM to cyclic exposure to surface levels of UVB and increasing UVA:UVB ratios in deeper waters, thereby reducing the effect of UVR on BP and the transformation of DOM. Thus, our results, which do not consider mixing, would represent the conditions of a calm day with low vertical mixing or those in polar water with shallow pycnoclines from melting ice, where mixing would be reduced, and may overestimate these effects under windy conditions, during which rapid vertical mixing occurs. Nevertheless, large differences were observed between incubations exposed to the full spectrum of solar radiation and those in which UVB was excluded.

Godoy et al. (2012) showed similar results, but for one of the Pacific communities studied in the area, NCP increased, instead of decreasing, under full solar radiation incubations. Indeed, UVR affects community metabolism through a suite of complex interactions, including photochemical reactions affecting the lability of DOM and increasing bacterial respiration (Obernosterer et al. 1999; Tranvik and Bertilsson 2001; Herndl et al. 2008), increased phytoplankton cell mortality and lysis (Agustí and Llabrés 2007), inhibitory effects on primary production (Cullen et al.

1992), and different sensitivities and complex interactions of the species and groups forming the communities (Boelen et al. 2002; De Lange et al. 2003). This complexity may challenge the prediction of net metabolism at the ocean's surface and, therefore, the associated exchange of CO<sub>2</sub> with the atmosphere. Whereas our results clearly demonstrate that bacterial production and respiration are enhanced in the dark after exposure to solar radiation, we cannot separate the possible roles of changes in the lability of DOM and bacterial physiology during exposure to solar radiation in accounting for these responses. In any case, our results show the effects of UVR on relevant processes, such as NCP, community respiration, and bacteria production, indicating that further research assessing these processes in the surface ocean through incubations should use UVR-transparent materials.

The evidence that UVR plays an important role in controlling net community metabolism and respiration in surface ocean waters suggests that the elevated UVR radiation derived from the erosion of the stratospheric ozone layer by chlorofluorocarbon (CFC) compounds three decades ago (Molina and Rowland 1974) must have a substantial effect on the surface ocean ecosystem, albeit difficult to evaluate because of a lack of predisturbance baseline measurements. Since the recovery of stratospheric ozone values like those experienced before the accumulation of CFCs in the atmosphere are expected to return only between 2050 and 2065 (Weatherhead and Andersen 2006), the effect of UVB on ocean metabolism is likely to be a pervasive one.

In summary, UVR reaching the ocean's surface consistently affects the metabolic balance of plankton communities. Ultraviolet radiation increased heterotrophic activity in our study by increasing BP and community respiration. We therefore conclude that changes in incident UVB radiation may have affected the metabolic balance of the ocean and must be taken into account in biogeochemical studies and methodologies because UVR is a key driver of the functioning of planktonic communities in the surface layers of the ocean.

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