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UV effects on invertebrate and diatom assemblages of Greece

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Abstract

The effects of solar radiation (PAR, UVA, UVB) on the productivity and structure of diatom and invertebrate assemblages were assessed during primary succession on artificial substrate near a rocky shore of the Saronikos Gulf, Greece. Three light treatments were performed (PAR, PAR+UVA, and PAR+UVA+UVB) at 0.5, 1.0 and 1.5 m of depth. Pennate diatoms were the major component of the developing periphytic communities during the study period. Exposure to solar UVB initially reduced the biomass and altered the structure of the diatom assemblages. The highest biomass of diatom assemblages was observed under PAR (49.2 g/m^2). This value was significantly higher than the biomass of assemblages growing under PAR+UVA+UVB, but not significantly different compared to the biomass of assemblages exposed to PAR+UVA. These differences, however, did not persist at later stages. The most abundant invertebrate groups present were Polychaetes and Crustaceans. Solar UVB did not have significant effects on invertebrate biomass. Analysis of the invertebrate assemblage structure revealed time-course differences but no clear trends among the different treatments.

Keywords: Algae; Grazers; Invertebrates; Solar UV radiation; Artificial substrate; Greece; Rocky shore; Mesocosm

1. Introduction

The rapid decline in stratospheric ozone concentrations results in increased levels of UV radiation, especially UVB (280–315 nm) incident on the Earth's surface [1,2]. Peak global losses of ozone were predicted for the year 1998–99 with a maximum loss of 6–7% at northern mid latitudes in winter/spring relative to the late 1960s, coinciding with peak stratospheric chlorine and bromine abundances [3]. Although ozone depletion was initially restricted over the Earth's poles, recent studies have shown a 5% decrease of the ozone layer over the Mediterranean [4]. Based on this, an increase of 2.7% per year or 27% per decade in solar irradiance at 305 nm has been predicted [5].

Short-term UV screening experiments confirm that UV radiation can reduce the growth rates of benthic diatoms in shallow freshwater during summer at northern mid latitudes [6]. Prolonged exposure to UV radiation increased diatom biomass and species diversity. This effect was attributed to the greater UVB susceptibility of grazers (larval chironomids) than their food, the algae.

Literature on the effects of UV radiation on periphyton is rather scarce [7–9]. Direct UVB effects on diatom assemblages grown on ceramic tiles in a natural marine habitat in Saronicos Gulf, Greece included shifts in species composition and temporary inhibition of biomass production [8].

The sensitivity and tolerance of marine invertebrates to UVB radiation may vary considerably [10]. Cleavage reduction has been observed in sea urchin eggs as a result of exposure to ultraviolet radiation [11]. The egg capsules of the gastropod *Nucella emarginata* provide embryos with substantial protection from UVR. These capsules allow less than 5% of the incident UVB at 300 nm and less than 55% of incident UVA at 360 nm to enter the capsule chamber [12]. The sensitivity of corals to UVB may depend on the depth at which they grow [13]. A correlation between depth and concentration of UV-absorbing pigments has been found in different organisms [14].

Earlier investigations assessed solar UVR effects on microalgae at the assemblage level [8]. These studies showed the existence of adverse short-term UVR effects on diatom assemblages, but did not address the effects of grazing and its possible interaction with UVR. Freshwater studies have reported that reduced grazing pressure due to

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enhanced UVR resulted in increased primary productivity [6]. The objectives of the present study are to assess (a) UVR effects on the development of associated microalgal and invertebrate assemblages, and (b) grazing/UVR interaction effects on diatom assemblage structure and productivity during the process of primary ecological succession in a typical marine habitat of Saronikos Gulf, Greece.

2. Materials and methods

The experiment was conducted at a distance of 50 m from an east-facing rocky shore of Saronikos Gulf, 5 km southeast of Korinthos, Greece (37°58′ N, 23°0′ E). Using UV-transmitting Plexiglas and plastic foil filters, nine treatment combinations (three UV regimes×three depths; Table 1) were performed in duplicate. The filters used were UV-transmitting Plexiglas (Plexiglas GS 2458) and plastic cut-off filters (295 Ultraphan, PR Montagefolie 320 nm Art. No. 10155099; and Ultraphan UV Opak, thickness, 0.3 mm). Each experimental unit consisted of eight 10×10-cm ceramic tiles placed on a polypropylene screen fixed onto a 3/4′, 45×90-cm PVC frame. The 18 units were suspended in random pairs from nine 2×1-m rafts at 0.5, 1.0 and 1.5 m. The rafts were constructed from 2.5′ PVC pipe filled with polyurethane for waterproofing and tied together in tiers of three. Each raft tier was anchored at one end only to allow free swinging of the apparatus with the current. The filters were cleaned regularly to prevent alteration of the transmittance properties due to biofouling. Light transmittance was periodically checked for replacement of defective filters.

Attenuation of the PAR, UVA and UVB bands in the water column was measured with an Optronic 752 double monochromator spectroradiometer using a sensor in 4π geometry. Irradiance levels at 1.5 m were ca. 35% lower than immediately below the surface for PAR; 20% for UVA; and 6% for UVB.

Surface irradiance measurements were obtained using an ELDONET dosimeter equipped with three sharp band sensors (Gröbel, Ettlingen) for PAR, UVA and UVB calibrated against the 752 Optronic spectroradiometer [16]. The signals from the sensors were amplified, digitized and stored in a dedicated computer (Visual Basic program 'Windose 2000' developed by M. Lebert, University of Erlangen, Germany). Daily surface irradiance averages

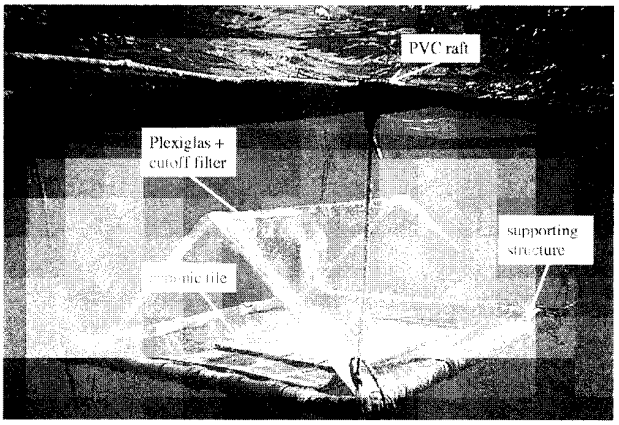

Fig. 1. Experimental partial enclosure. Eighteen experimental units were suspended at 0.5, 1.0 and 1.5 m from nine 2×1 m rafts. Sixteen 10×10-cm ceramic tiles were placed under each of the three light treatments.

were 10 027 kJ/m^2 (PAR), 1110 kJ/m^2 (UVA), and 28 kJ/m^2 (UVB).

The experimental enclosures were placed in the sea on June 17, 1995 (Fig. 1). The developing communities were sampled once every 2 weeks between July 15 and September 9, for a total of five times by removing one ceramic tile from each treatment. The collected biomass was strained free of salt water, and oven-dried to constant weight at 80°C. The samples were processed for microscope analysis of the diatom assemblage using mild digestion of the cell contents with H_2SO_4 [17].

Invertebrate collection was carried out by placing each tile in a plastic container and selective staining of living animal tissues with 20% Rose Bengal solution. Individual organisms were picked with forceps under a dissecting scope, sieved through a 0.5-mm mesh size metal sieve, and preserved in 4% neutralized formaldehyde solution. Invertebrates were identified and counted with a light microscope and/or a dissecting scope.

The metrics calculated for each sample were: number of species (S), abundance (N/0.01 m^2), species diversity H' [18], evenness of distribution j [19] and biomass (g/0.01 m^2). Multidimensional scaling (MDS) was performed with the Primer software (Plymouth Marine Laboratory) using the Bray–Curtis index as a measure of community similarity [20]. Using the sample similarity matrix MDS determines the two-dimensional map of points best representing this similarity as follows. A dimensional map is

Table 1
Experimental design ($n=2$): nine light×depth treatment combinations were performed

Cut-off filter depth (m)	286 nm	320 nm	390 nm
0.5	PAR+UVA+UVB @ 0.5 m	PAR+UVA @ 0.5 m	PAR @ 0.5 m
1.0	PAR+UVA+UVB @ 1.0 m	PAR+UVA @ 1.0 m	PAR @ 1.0 m
1.5	PAR+UVA+UVB @ 1.5 m	PAR+UVA @ 1.5 m	PAR @ 1.5 m

initially drawn at random. Regression is then performed among the ranked distances of points given by the similarity matrix. The regression's goodness-of-fit is measured by the stress value, calculated by the formula:

$$\text{Stress} = \frac{\sum_{j}^{n}\sum_{k>1}^{n}(d_{jk} - \hat{d}_{jk})^2}{\sum_{j}^{n}\sum_{j>k}^{n} d_{dk}^2}$$

where $\hat{d}_{jk}$ is the distance between sample points j and k that corresponds to the given dissimilarity d_{jk}. Stress values between 0 and 1 indicate excellent configurations; values between 1 and 2 are considered satisfactory, while values >2 show virtually random configurations. The software alters the position of the points on the map and the value of stress is calculated again. This procedure is repeated for as many times as necessary until stress reaches its minimum value.

3. Results

3.1. Diatoms

One hundred and seventy-three diatom taxa were recorded under the nine treatment combinations. The clear separation of the three UV treatments shown by MDS ordination indicates a UV effect on species composition during the first 4 weeks of primary succession (Fig. 2; July 15, July 29). On the contrary, there was no distinct separation of the assemblages into depth-groups during the course of the experiment.

Species contributing to the dissimilarity among the different UV radiation treatments: (a) only under PAR+UVA+UVB: *Amphora biggiba*, *A. coffeaeformis*, *A. inariensis*, *A. spectabilis*, *Bacillaria paxillifer*, *Cocconeis fluminensis*, *C. pinnata*, *Climacosphenia elongata*, *Gyrosigma attenuatum*, *Mastogloia pumila*, *Opephora olsenii*, *Surirella comis*; (b) only under PAR (no UVA+UVB): *Amphora graeffi*, *Opephora martyi*; (c) under PAR+UVA+UVB and PAR+UVA: *Cocconeis costata*, *Cymbella affinis*, *Diploneis crabro*, *D. smithi*; (d) in PAR+UVA+UVB and PAR: *Navicula clementis*, *Nitzschia sigma*.

Species absent from the PAR treatments include *Amphora ostrearia*, *Cocconeis placentula*, *Mastogloia corsicana*, *M. paradoxa*, *Nitzschia dissipata*. In the PAR+UVA and PAR+UVA+UVB treatments their abundance generally tends to decrease as depth increases.

Solar radiation had significant effects on diatom biomass ($F=4.82$, df=2 and 36, $P<0.05$). More specifically, the highest biomass of diatom assemblages was observed under PAR (49.2 g/m^2, Fig. 3a). This value was significantly higher than the biomass of assemblages under PAR+UVA+UVB.

The effects of depth on diatom biomass depend on the sampling date ($F=2.75$, df=6 and 36, $P<0.05$). The highest value of diatom biomass was observed on August 26 for the diatom assemblage at 1.0 m depth (Fig. 3 b). This value was significantly higher than the biomass at 0.5 m on the same date, but not significantly different from the biomass at 1.5 m on the same date. On any date, except August 26, biomass at shallow depths (0.5 m) was not statistically different compared to biomass values at other depths.

3.2. Invertebrates

Forty-nine taxa of invertebrates were identified and the most abundant phyla were Polychaetes (36.7%) and Crustaceans (35.6%). The number of invertebrate species is plotted in Fig. 4 (left panel), while a list of dominant taxa is given in Table 2. The four metrics analyzed (number of species, number of individuals, species diversity, species evenness) correlated with time but not with either light or depth treatment. The total number of individuals was lower under the PAR+UVA+UVB treatment. However, no significant correlation was found ($P>0.05$).

Species diversity and evenness generally follow similar trends (Fig. 5). Species number (S), diversity (H'), abundance (N) and total biomass (g) increased during the first 8 weeks of the experiment. The sharpest increase in relative abundance in almost all treatments occurred between weeks 4 and 6. This increase was even more dramatic in the PAR+UVA+UVB treatment at 1.0 m. The highest number of individuals was also found in the same treatment on August 26 (8 weeks). However, this value dropped dramatically on September 9 (10 weeks). The highest value of diversity (H') was recorded on August 26 (8 weeks) in the PAR treatment at 0.5 m ($H'=2.46$).

Correlation analysis did not reveal strong associations of biotic parameters with either light or depth. Clustering of the invertebrate assemblages also revealed time-course differences but no clear trends among the different UV-depth treatment combinations. Differences were primarily observed in species number and number of individuals.

Time had significant effects on biomass of invertebrate assemblages ($F=10.28$, df=4 and 45, $P<0.05$). Invertebrate biomass values during the early stage of the study period (July 15 and 29) were significantly lower than during the second half of the study period (Fig. 6). However, these values stabilized and no significant differences were observed after August 12.

4. Discussion

Numerous studies have examined solar UV radiation effects on isolated aquatic photosynthetic organisms [21–24]. The understanding of ecosystem functions, however,

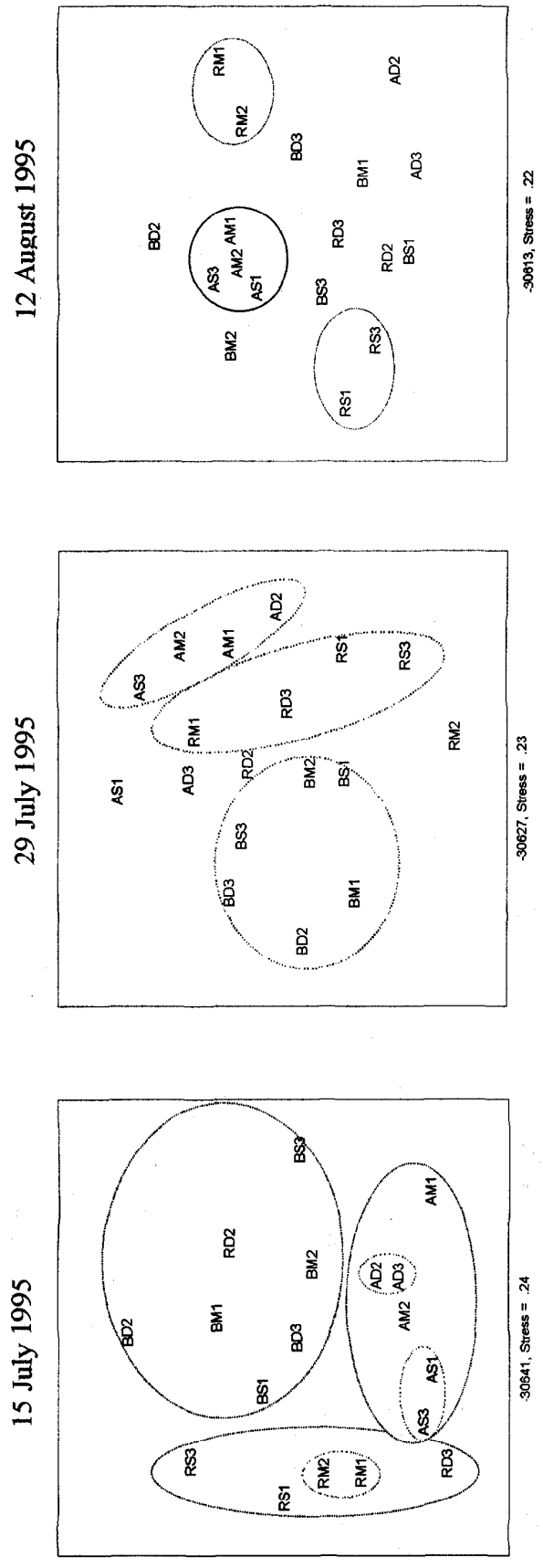

Fig. 2. MDS ordination of diatom assemblages. The clear separation of the three UV treatments indicates a UV effect on community structure on the first two sampling dates (July 15, 29). No clear separation into depth-groups is observed. Key: (A) PAR+UVA; (B) PAR+UVA+UVB; (R) PAR; (S) 0.5 m; (M) 1.0 m; (D) 1.5 m; (AM1, AM2) replicates.

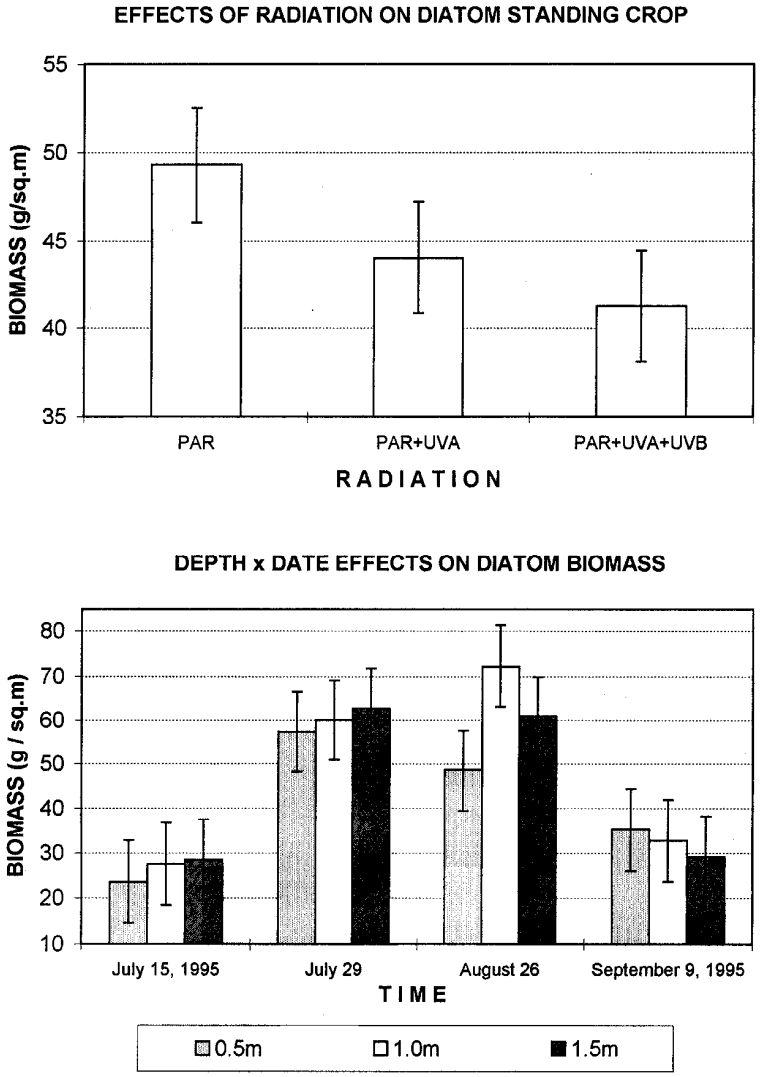

Fig. 3. Mean diatom standing crop after removal of invertebrates. (a, top) Effects of solar radiation: the standing crop under PAR (mean value of all depths) was significantly higher than under PAR+UVA+UVB. (b, bottom) Date by depth effects: time had a significant effect on standing crop (higher in intermediate dates). Error bars: 95% confidence intervals.

also requires analysis at the population, assemblage and community levels. The structural differences between the diatom assemblages were more pronounced during the early stages of the experiment (first three sampling dates), and became less clear in the second half. The same pattern was observed in a previous experiment [9]. The weakening of structural differences at later stages in both studies can be attributed partly to the inherent capacity of periphytic communities to adapt to increased solar UVB irradiance, and partly to the masking interaction effects of grazing and primary succession. Assemblage separation during the early stages is the result of (a) different relative abundances of species common in different treatments, and (b) the presence of several UV sensitive species in the UV protected assemblages (*Amphora ostrearia*, *Cocconeis placentula*, *Mastogloia corsicana*, *M. paradoxa*, *Nitzschia dissipata*). However, although the two studies were conducted in the same experimental site and with the same apparatus, and many of the diatoms were recorded in both studies, the UV-sensitive and UV-tolerant diatoms differed. This suggests that community structure is a safer criterion to use rather that individual species. The discrepancy between the two studies is probably due to the different years and seasons the two studies were conducted.

Exposure to UVB radiation resulted in a significant reduction of the diatom biomass. This agrees with the findings of previous studies [8]. Light-related differences were more pronounced than depth-related differences: the relative effect of the water column on light composition is much stronger than that of the cut-off filters at increased

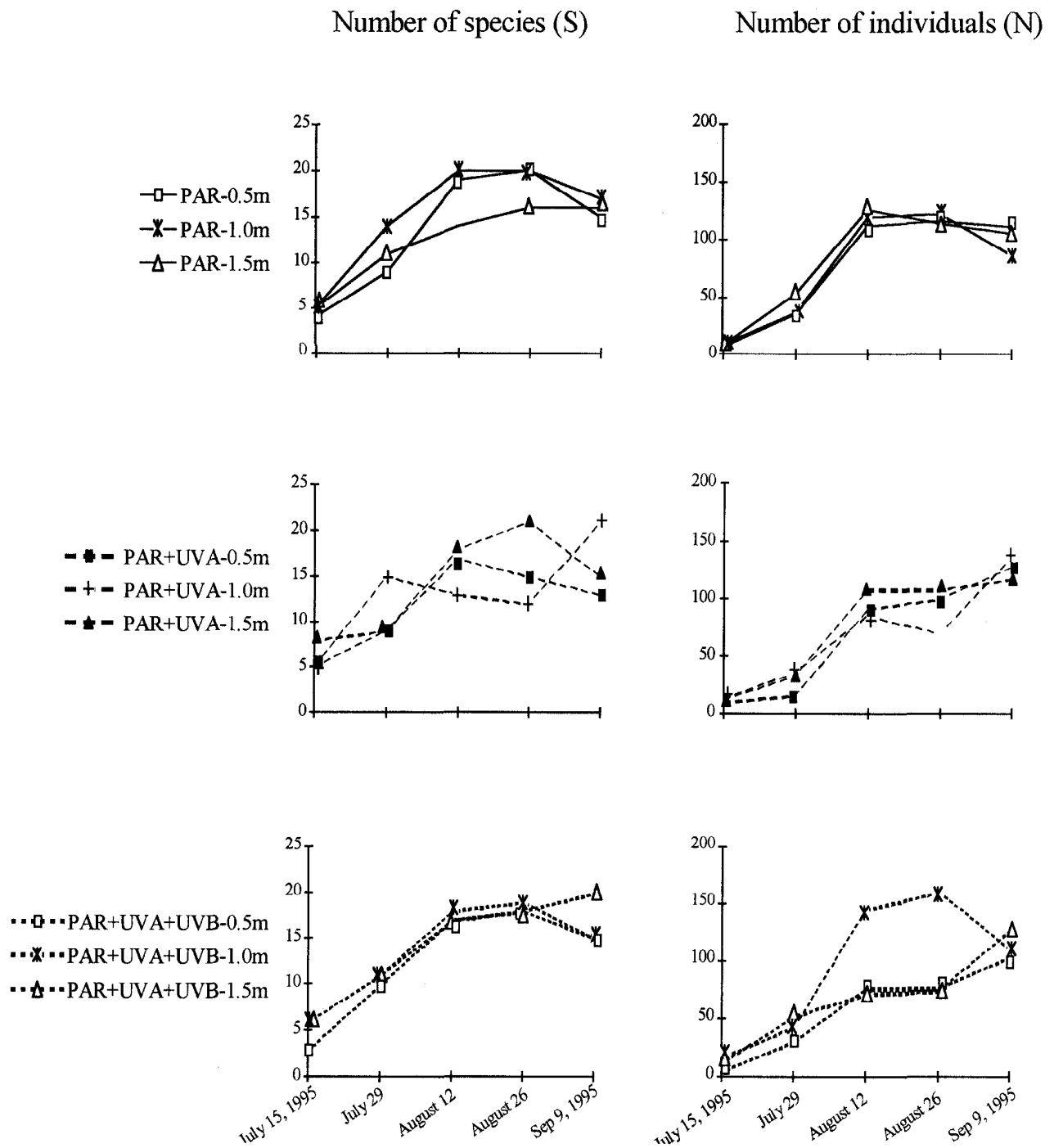

Fig. 4. Mean total number of invertebrate species (left panel) and mean total number of individuals (right panel) grouped by light and depth. The greatest number of species was recorded on 26 August 1995. A negative correlation was found between wavelength band and total number of individuals. This correlation was not significant ($P>0.05$).

depths. The only significant depth-related effect observed was between the diatom biomass at 0.5 and 1.0 m on August 26.

The structural differences in the diatom assemblages due to light treatment were not accompanied by similar changes in the invertebrate assemblages: the structure of the invertebrate assemblages correlated with time, but no clear trends were found among the different UV-depth treatment combinations. A possible explanation is that larger organisms are likely to be less susceptible to UV than smaller organisms with shorter life cycles such as bacteria and microalgae [25]. Many of the dominant invertebrate species were photophilic and/or characteristic of hard substrate. Their diet mainly consists of macroalgae,

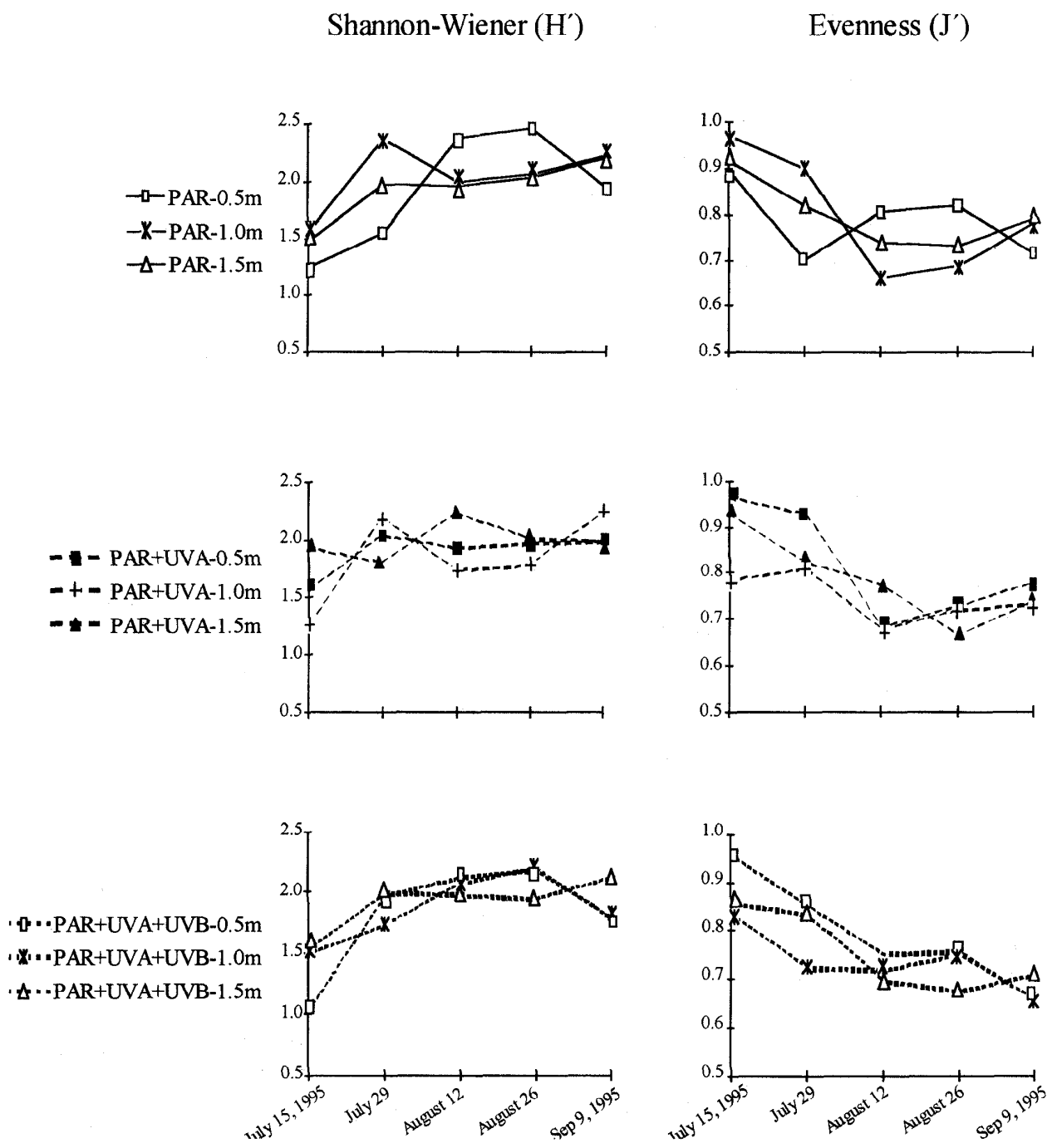

Fig. 5. Species diversity (H'; right panel) and evenness (J'; left panel) grouped by light and depth. Diversity increased during the first 8 weeks of the experiment. The highest value of diversity (H') was recorded on 26 August 1995 in the PAR treatment at 0.5 m.

diatoms and detritus, while some feed on hydroids, bryozoans and other invertebrates.

Sampling area size and homogeneity might have been two sources of error in identifying differences in the structure of the invertebrate assemblages. The 10×10-cm surface area of the ceramic tiles was adequate for analyzing diatom assemblages, but might have been small for motile animal organisms. Moreover, natural ecosystems are characterized by a large variety of microhabitats and niches as opposed to the flat configuration of the ceramic tiles used.

The increased grazing pressure resulting from the sharp

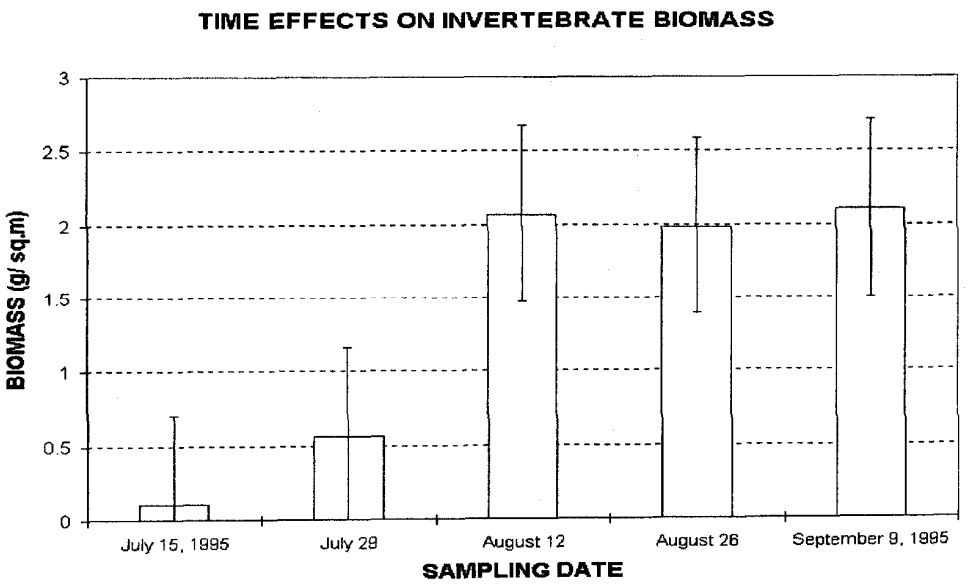

Fig. 6. Mean values of invertebrate biomass. Invertebrate biomass was significantly lower during the early stage of the study period (July 15 and 29). No significant differences were found after August 12. Error bars: 95% confidence intervals.

increase in the total number of invertebrate individuals on August 12 in all treatment combinations (Fig. 4) caused a decrease in the diatom biomass on August 26. The total number of invertebrate individuals generally remained constant thereafter with the exception of the last sampling event.

Bothwell et al. [6] reported that enhanced UVB reduced grazer abundance, which, in turn, resulted in increased primary producer biomass. These findings, however, are not supported by the results of the present study. The differences between the two studies can be attributed to: (a) the different experimental conditions (open enclosures placed in natural marine environment under naturally occurring solar UVB as opposed to artificial freshwater flumes excluding higher level predators exposed to enhanced UVB); and (b) the larger number of grazer species of the present study versus the dominance of a single group (chironomids).

Increased UVB radiation is considered as a stress for marine organisms. Since primary producers are a fundamental component of most ecosystems, UVB may play an important role in niche separation, trophic chain and community structure. However, factors such as substrate hardness and porosity, food type, seasonality and turbulence may cloud the effects of solar ultraviolet radiation.

5. Conclusions

The results of the present study do not show any significant UVR effects on invertebrate assemblages and/or interaction between the effects of grazing and UVR on periphytic diatom assemblages.

Previous studies [8,9,26] have shown that diatom assemblage structure can be used in predicting short-term ecosystem responses to the increase in solar UVB. For longer time periods, however, UV effects cannot be easily separated from the effects of many other environmental factors in the natural ecosystem. Therefore, the understanding of large-scale UV effects requires long-term field investigations on the interaction of such factors on biological systems.

A comparison between previous investigations and the present one suggests that community metrics are more reliable criteria than isolated 'indicator' species.

Table 2
Dominant invertebrate taxa (all treatments)[a]

July 15, $S=15$	July 29, $S=30$	August 12, $S=39$	August 26, $S=41$	September 9, $S=39$
Caprella acanthifera (a)	*Leptochelia savignyi* (t)	*Leptochelia savignyi* (t)	Nereidae (p)	Nereidae (p)
Copepoda	*Platynereis dumerilii*	*Platynereis dumerilii* (p)	Syllidae (p)	Syllidae (p)
	Perinereis cultifera (p)	*Perinereis cultifera* (p)		*Polyophthalmus pictus* (p)
	Syllis hyalina (p)	*Syllis hyalina* (p)		*Leptochelia savignyi* (t)
	Syllis cornuta (p)	*Syllis cornuta* (p)		Copepoda

[a] (a) amphipod; (t) tanaid; (p) polychaete.

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