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Photosynthetic Action Spectra and Adaptation to Spectral Light Distribution in a Benthic Cyanobacterial Mat

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We studied adaptation to spectral light distribution in undisturbed benthic communities of cyanobacterial mats growing in hypersaline ponds at Guerrero Negro, Baja California, Mexico. Microscale measurements of oxygen photosynthesis and action spectra were performed with microelectrodes; spectral radiance was measured with fiber-optic microprobes. The spatial resolution of all measurements was 0.1 mm, and the spectral resolution was 10 to 15 nm. Light attenuation spectra showed absorption predominantly by chlorophyll *a* (Chl *a*) (430 and 670 nm), phycocyanin (620 nm), and carotenoids (440 to 500 nm). Blue light (450 nm) was attenuated 10-fold more strongly than red light (600 nm). The action spectra of the surface film of diatoms accordingly showed activity over the whole spectrum, with maxima for Chl *a* and carotenoids. The underlying dense *Microcoleus* population showed almost exclusively activity dependent upon light harvesting by phycobilins at 550 to 660 nm. Maximum activity was at 580 and 650 nm, indicating absorption by phycoerythrin and phycocyanin as well as by allophycocyanin. Very little Chl *a*-dependent activity could be detected in the cyanobacterial action spectrum, even with additional 600-nm light to excite photosystem II. The depth distribution of photosynthesis showed detectable activity down to a depth of 0.8 to 2.5 mm, where the downwelling radiant flux at 600 nm was reduced to 0.2 to 0.6% of the surface flux.

The adaptation of microalgae to ambient light conditions in phytoplankton communities has been studied for many years. It is well known that these organisms are adapted to various degrees to the prevailing light intensities, as well as to the spectral quality of the light (4, 31). Organisms living deeper within the photic zone develop more antenna pigment per reaction center and become light saturated at lower intensities than those living near the surface (see reference 24 and references therein). These organisms also adapt to the prevailing blue-green light at depth by increasing the amount of accessory pigments such as carotenoids in algae (J. B. SooHoo, D. A. Kiefer, D. J. Collins, and I. S. McDermid, J. Plankton Res., in press) or phycoerythrin in cyanobacteria (2, 35), which can absorb quanta of these wavelengths.

Such adaptations in benthic phototrophs have been scarcely studied, mainly because appropriate techniques have not been available for these highly compressed communities. Adaptations to higher and lower radiant flux at the sediment surface have, however, been recognized, but only at a very general level (6). Furthermore, the motility of many benthic microalgae, which allows a vertical migration based on their phototactic response to the varying light conditions (3, 10), is an additional complicating factor which requires direct measurements of light penetration into the sediment. Such measurements have been done at spatial resolutions down to 1 mm (5, 9, 32). Since the euphotic zone in sediments is often less than 1 mm thick (26), it was necessary to develop a fiber-optic microprobe with 10-fold-higher resolution to analyze the light gradients in sufficient detail (15).

In the present study, we have combined these light microprobes with similar high-resolution techniques for the measurement of photosynthesis by oxygen microelectrodes (25) to study light adaptation in a benthic cyanobacterial mat. Although of limited distribution, such mats are ideally suited for ecological and physiological studies of microbenthic photosynthesis.

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MATERIALS AND METHODS

All cyanobacterial mats used for the present study were collected in April and June 1985 within a small area of a hypersaline pond situated at Guerrero Negro on the Pacific coast of Baja California, Mexico (11). A series of 0.5- to 1-m-deep artificial evaporating ponds have been constructed in this area for the production of sea salt. Cyanobacterial mats grow on the bottom of those ponds at salinities ranging from about 60 to 120‰, and they reach a thickness of 10 to 15 cm. The collected mats grew in pond 5 (number according to the local salt company, Exportadora de Sal) at 72 to 91‰ salinity and 18 to 25°C.

Microscopy and measurements of the vertical distribution of oxygen, photosynthesis, and light were done at Guerrero Negro within a few days of sampling. Cores of the mats were kept outdoors in the shade, submerged in pond water at near in situ temperature. Before the measurements, they were illuminated vertically on the surface for a few hours by a halogen lamp at 500 microeinsteins (μE) m^{-2} s^{-1}, which is close to the in situ light intensity around noon below 1 m of pond water. This allowed a stable distribution of organisms and chemical gradients to become established. Action spectra were analyzed in cores taken from 5-dm^2 blocks of intact mats which were brought back to Ames Research Center. The blocks were carefully cut out of the mats with a knife and were transported in plastic boxes under humid air. This procedure was found to give the least disturbance to the mat surface. The mats were maintained in a greenhouse at 30 to 50% daylight intensity in aquaria with aerated pond water at near in situ temperature.

Microelectrode measurements. Depth distributions of oxygen and photosynthesis were measured by oxygen microelectrode techniques as described by Revsbech and

Jørgensen (26). The mats were under constant illumination at 500 μE m^{-2} s^{-1}, and readings were taken at depth intervals of 70 to 100 μm with a Clark-type oxygen microelectrode (28) attached to a micromanipulator. Photosynthesis was measured by the light-dark shift technique (25). A datalogger (HP 3421A; Hewlett-Packard Co.) used for data collection was programmed with a calculator (HP 41CX; Hewlett-Packard) which calculated the slope of the oxygen curve from 0.5 to 2.5 s after a light-dark shift by linear regression analysis.

Action spectra for oxygen evolution were measured directly in the intact mats among the two dominant groups of oxygenic phototrophs, the diatom surface layer, and the *Microcoleus* layer, as described by Jørgensen et al. (16). Under a dissecting scope, the oxygen microelectrode was positioned a few tens of micrometers into a dense cluster of the organisms. The mat was illuminated by continuous monochromatic light of 12 to 14 nm bandwidth at about 20 μE m^{-2} s^{-1}. This light intensity was well within the range of linear photosynthetic response. The light intensity was standardized against a calibrated quantameter (LI-185A; Li-Cor). Duplicate measurements were taken at 10-nm intervals throughout the visible spectrum of 400 to 700 nm with 600 nm as a reference wavelength for every third reading to correct for possible changes in overall photosynthetic activity. At each wavelength, the mat was illuminated for a few minutes before measurements were made, to allow a steady-state oxygen profile to become established. A full action spectrum was recorded within 2 to 3 h. For one action spectrum of *Microcoleus* sp., the effect of simultaneously exciting both photosystems I and II was studied. A continuous background illumination of 20 μE m^{-2} s^{-1} at 600 nm was provided with a narrow band-pass filter in addition to the shifting light source.

Light measurements. The microscale spectral distribution of light in the mats was determined with fiber-optic microprobes as described by Jørgensen and Des Marais (15). The probe was constructed from a single optical fiber with a diameter of 80 μm. It was tapered and rounded at the tip to a diameter of 25 μm to provide optical axial symmetry and to facilitate penetration through the mats. The half-angle of the acceptance cone was about 20° in water. The detector was a simple but ultra-low noise hybrid photodiode-amplifier (EG & G; TCN 1000-93), and the sensitivity for white light was about 0.001 W m^{-2} or 0.01 μE m^{-2} s^{-1}. The probe was attached to a micromanipulator, and measurements were taken at 0.1-mm depth intervals through the surface layers of the mats. The undisturbed mat core with a thin layer of overlying pond water was illuminated on the surface by monochromatic light of half bandwidth 12 to 14 nm, which was shifted at 10-nm intervals through the spectrum of 400 to 700 nm. For most measurements, the fiber-optic microprobe approached the surface from below at a 0° angle between the fiber axis and the collimated light beam. Backscattered light was measured at a 145° fiber-light angle. The exact position of the fiber tip relative to the mat surface was observed under a dissecting scope and subsequently read on a micromanipulator. The light data were recalculated for each wavelength as relative values in the percentage of the collimated light beam at the mat surface.

RESULTS

Vertical zonation: three examples. The mat communities showed significant variation in the density of organisms within the gelatinous polysaccharide sheaths which functioned as a binding material in the mats. There were also large variations in the development of the tufted surface layer of diatoms, partly in response to seasonal changes of light and temperature, etc., in the ponds.

The laminated microbial communities were rather similar to those described for *Microcoleus chthonoplastes* mats of the Solar Lake, Sinai (17–19). The surface was populated by a thin film of diatoms, mostly *Nitzschia* and *Navicula* spp., often growing in small tufts, together with unicellular cyanobacteria, mostly *Synechococcus* spp. and more sparse filamentous cyanobacteria. Below was a dense layer, 300 to 800 μm thick, of filamentous cyanobacteria, mostly *M. chthonoplastes* growing in bundles with a common, thick sheath together with fewer *Oscillatoria* sp. The next layer was less densely populated, with mostly filamentous green bacteria, *Chloroflexus* spp., together with filamentous and unicellular cyanobacteria, some purple sulfur bacteria, and a distinct layer of colorless sulfur bacteria, *Beggiatoa* spp. Below this was an older *Microcoleus* layer, and the sequence was repeated. Although this general pattern of vertical zonation was consistent in the mats, these were quite heterogeneous when analyzed on a microscale level.

In Fig. 1, we show three examples of oxygen, photosynthesis, and light distribution in the *Microcoleus* mats. The vertical profiles of photosynthetic rates reflected the zonation of the dominant phototrophs. Fig. 1A shows a very dense *M. chthonoplastes* layer with a poorly developed diatom film on the surface; oxygenic photosynthesis reached a depth of 0.8 mm with only one maximum. Fig. 1B shows a well-developed diatom film causing a second photosynthesis maximum at the surface; oxygenic photosynthesis reached a depth of 1.4 mm. Fig. 1C shows a loose structure of diatom tufts, giving a high photosynthesis maximum at the surface and two lower maxima in consecutive *M. chthonoplastes* layers; oxygenic photosynthesis reached a depth of 2.5 mm.

The depth distribution of oxygen reflected the zonation of photosynthesis maxima, especially in Fig. 1C, for which an oxygen profile with unusual detail was recorded. A very dynamic balance between photosynthesis and respiration is required to establish such details in the oxygen curves despite the rapid diffusion in these dimensions. In accordance with this dynamic balance, a comparison of the oxygen concentrations and production rates at the highest photosynthesis maxima (depth of 0.3 mm in Fig. 1A and C) showed turnover of the whole oxygen pool once every minute.

Log plots of relative, downwelling radiant flux into the mats show that light decreased roughly exponentially at depth or with gradually increasing attenuation coefficients (Fig. 1). Blue light of 450 nm was attenuated more strongly than red light of 600 nm. Near-infrared light of 825 nm, which can potentially be utilized by bacteriochlorophyll *a*-containing phototrophic sulfur bacteria, may support anoxygenic photosynthesis in deeper layers (14). In some mat samples, a distinct band of purple sulfur bacteria occurred just about or below the depth of maximum O_2 penetration. Infrared light of 1,000 nm penetrated deep into the mats, since it is not absorbed by photosynthetic pigments. A comparison of the corresponding light and photosynthesis data shows that visible light, as measured by the optical fiber, had decreased to between 0.1 and 1% of the surface intensity at the lower boundary of the oxygenic photosynthesis zone.

Spectral light distribution. Analysis of the actual light limitation of photosynthesis requires a more detailed spectral study of light penetration into the mats. We measured the downwelling radiance in a fresh mat core at 0.1-mm

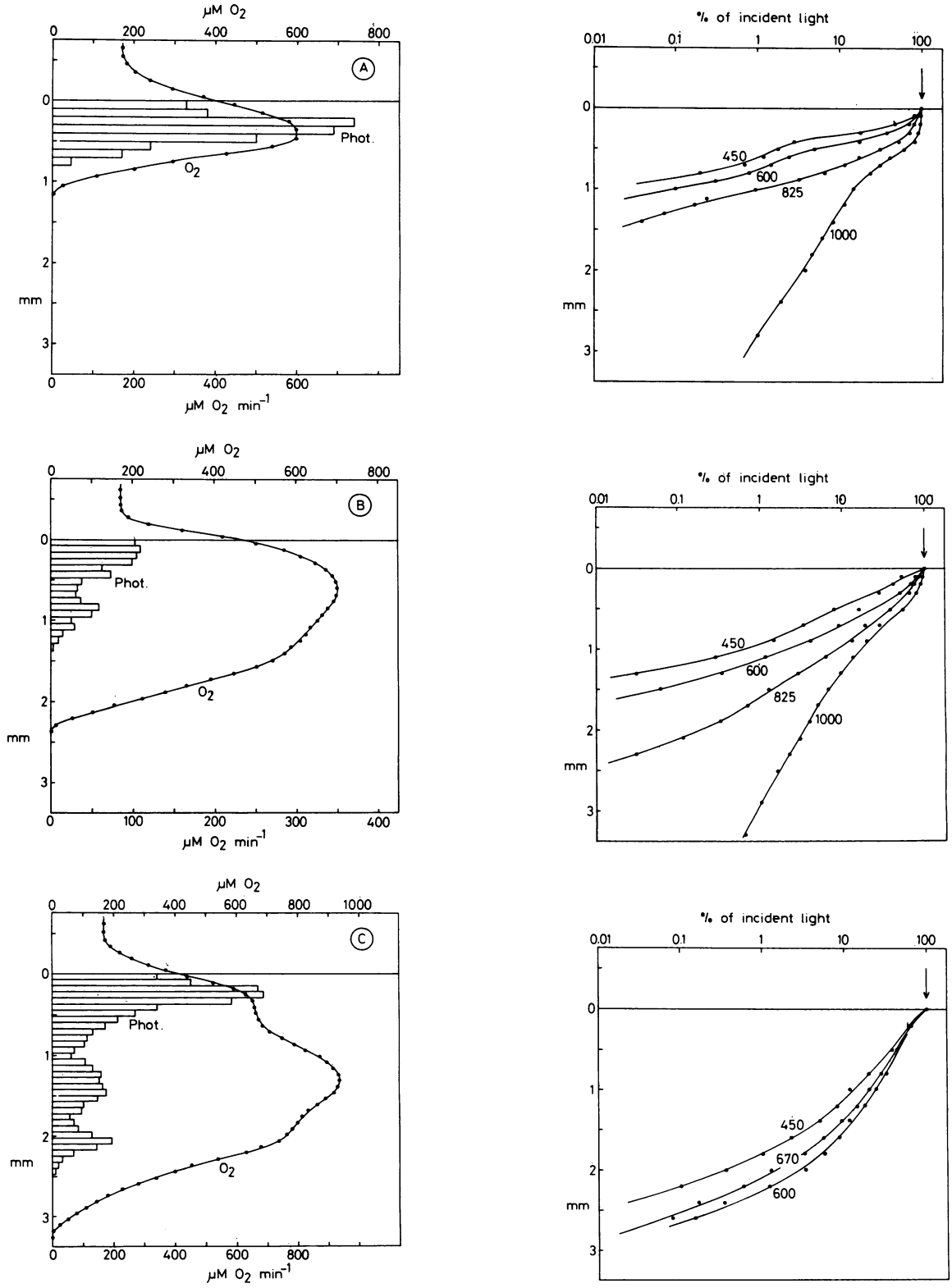

FIG. 1. Three examples of photosynthesis, oxygen, and light distribution in *Microcoleus* mats. One, two, and three bands, respectively, of dense phototrophic communities can be recognized. Measurements of downwelling radiance were done in the visible range at the wavelengths of maximum (450 nm) and minimum (600 nm) attenuation as well as in the near infrared (825 and 1,000 nm).

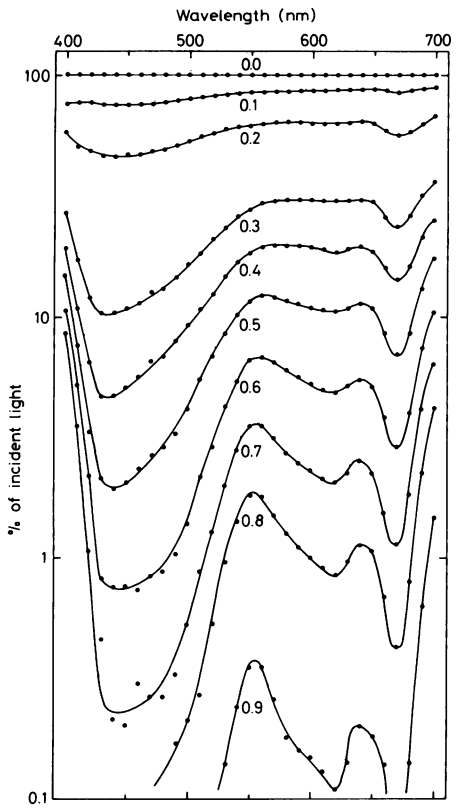

FIG. 2. Spectral distribution of downwelling radiance in a *Microcoleus* mat. For each depth and wavelength, light energy is calculated as a percentage of the surface value. Absorption maxima of Chl *a*, phycocyanin, and carotenoids are seen as minima in the logarithmic spectra. Increased data noise of low blue light was due to limited detector sensitivity. Numbers on curves indicate depths below the surface in millimeters.

depth intervals from the surface to 0.9 mm (Fig. 2). The spectral composition of the light changed rapidly through this thin mat layer relative to the collimated light beam illuminating the surface. A high concentration of chlorophyll *a* (Chl *a*) was evident from the absorption peaks at about 670 nm and, less pronounced, at 430 nm. The blue absorption band of Chl *a* fused with depth into a broad absorption maximum between 430 and 500 nm, probably caused by carotenoid pigments. A less pronounced absorption peak due to phycocyanin was present at 620 nm. Violet (400-nm) and far-red (700-nm) light penetrated deepest into the mat.

Action spectra. The spectral light quality changed strongly from the surface to the depth at which *M. chthonoplastes* and other filamentous cyanobacteria grew (Fig. 2). The action spectra for oxygen evolution were analyzed for the diatom community living on the mat surface exposed to full-spectrum light and for the dense *M. chthonoplastes* layer below the surface exposed to a more limited spectrum. Before measurements were taken in the *Microcoleus* layer, the overlying diatom film was carefully removed to obtain reproducible spectral light conditions at the measuring point.

The action spectrum of the diatom community showed maximum activity in the blue (430 nm) and red (670 nm) absorption bands of Chl *a* (Fig. 3A and B). A broad shoulder around 500 nm was probably due to photosynthetically active carotenoids in the diatoms, while a shoulder at around 590 to 620 nm was probably due to phycocyanin in unicellular cyanobacteria growing among the diatoms.

The action spectrum of the *Microcoleus* layer was quite complementary to that of the diatoms and showed only photosynthetic activity of the phycobilins associated with photosystem II (Fig. 3C and D). The highest activity occurred between 550 and 650 nm, with maxima at 580 and 650 nm. The former could be due to phycoerythrin or phycocyanin or both (see Discussion), while the latter could be due to allophycocyanin. There was no significant photosystem I activity detectable in the absorption regions of Chl *a* or carotenoids. Thus, there was no peak or shoulder at 670 nm, and the activities below 500 nm were near the limit of detection. In action spectrum D, however, slightly increased rates were detected at 420 to 430 nm (data not shown).

To show whether the apparent lack of photosystem I activity was due to sparse or inefficient antenna pigments or to inefficient energy transfer between the two photosystems, another action spectrum of the *M. chthonoplastes* band was recorded with a continuous background illumination of 600-nm light. This wavelength, which excites mainly photosystem II, was chosen as an intermediate between the measured activity maximum at 580 nm and the absorption maximum of phycocyanin at 620 nm. The action spectrum (Fig. 3E) again showed predominant photosystem II activity, but a low activity based on Chl *a* absorption was now also detectable as a shoulder at 670 nm and as a maximum at 430 nm.

The action spectra in Fig. 3A to D were calculated relative to the quantum flux of monochromatic light incident on the mat surface. The action spectra were, however, measured a few tens of micrometers into the mats to reduce errors due to steep activity gradients at the mat-water interface (27). The incident light beam was therefore slightly attenuated at the point of photosynthesis measurement. Furthermore, the phototrophic organisms also received backscattered light from the mat. Backscattered light has been filtered through the surface layers of the mat and is therefore spectrally altered relative to the collimated beam. It is especially depleted in blue light, which is most strongly absorbed in the mats. The action spectra should therefore ideally be based on the total quantum flux from all directions, i.e., on the scalar irradiance. Scalar irradiance spectra were obtained for these mats during the subsequent year (B. B. Jørgensen and D. J. Des Marais, in press). To illustrate the effect of the spectrally altered scattered light just below the mat surface, the action spectrum in Fig. 3E was recalculated by using a later-obtained scalar irradiance spectrum at a depth of 0.1 mm. The result is shown in Fig. 3F. The scalar irradiance spectrum is expressed relative to the incident light at the mat surface. The action spectrum shows that the photosynthetic activity based on blue-light absorption by Chl *a* is particularly enhanced after this recalculation. The action spectrum reveals less spectral detail in the red range. This is because the maxima and minima of photosynthesis coincide with maxima and minima of scalar irradiance. A recalculation to scalar irradiance at a depth of 0.1 mm exaggerates the spectral effects of scattered light. The true action spectrum will be intermediate between the two shown in Fig. 3E and F and will probably be closer to the former.

Light attenuation. A comparison between the action spectrum of the *Microcoleus* community and the light spectrum available for their photosynthesis showed a remarkable agreement. Figure 4 shows the spectrum of both downwelling and upwelling (backscattered) radiant flux relative to the

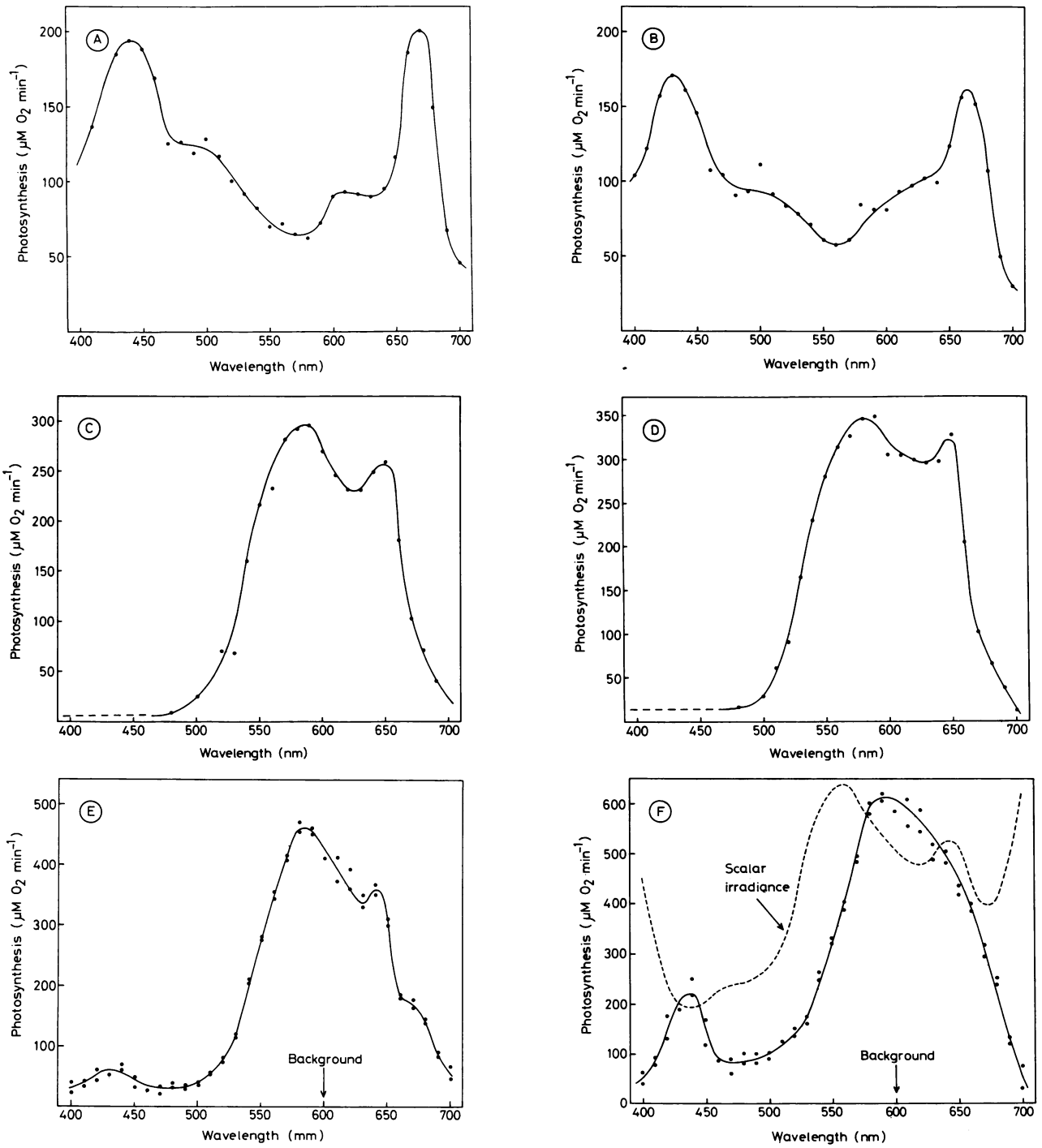

FIG. 3. In situ action spectra for oxygen evolution of two mat communities. (A and B) Diatom film on surface; (C to F) *Microcoleus* band at a depth of 0.3 to 0.8 mm. In panel E, a background illumination of 600 nm was provided to excite photosystem II. Photosynthesis rates were normalized for each wavelength to a quantum flux of 20 μE m^{-2} s^{-1} from the collimated light beam. In panel F, the action spectrum from panel E was recalculated from the shown scalar irradiance spectrum at a depth of 0.1 mm in a similar mat (Jørgensen and Des Marais, in press).

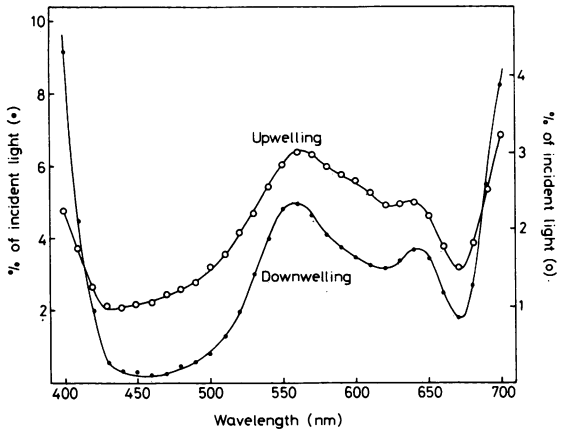

FIG. 4. Spectra of downwelling and upwelling radiant flux in mat at the lower and upper boundaries of the *Microcoleus* band.

surface flux. The downwelling light was measured at a depth of 0.65 mm in the lower, light-limited region of a *M. chthonoplastes* band. The upwelling light was measured at 0.2 mm, just above the *M. chthonoplastes* band, and had been partly backscattered through this layer. Although there was abundant light near 400 and 700 nm, this cannot be efficiently utilized by the light-harvesting pigments. The available light was thus largely restricted to the 550- to 650-nm region after it had been filtered through the thin film of phototrophs. Within this spectral region, maximum light was available at the absorption peaks of phycoerythrin (560 nm) and allophycocyanin (650 nm). A minimum at 620 nm showed, however, that the phycocyanin of the mat organisms absorbed light more efficiently than the other two phycobiliproteins did.

From the spectra of downwelling light at different depths in the mats, the corresponding attenuation spectra over the depth intervals can be calculated. An example is shown in Fig. 5, based on data from Fig. 2. Over the depth interval of the *Microcoleus* band of 0.3 to 0.7 mm, there was a strong attenuation of all blue light (430 to 500 nm) and smaller attenuation maxima at 620 and 670 nm.

DISCUSSION

The microscale distribution of photosynthesis within the microbial mats was closely related to the available light intensity and spectral composition. Artificial white light at 500 $\mu E\ m^{-2}\ s^{-1}$ was used for the experiments because this was estimated to be close to in situ irradiance at the pond bottom where the mats grew. A more accurate imitation of the actual light conditions to extrapolate the results to the natural mat situation would, however, require underwater spectral irradiance data, which are presently not available.

Photosynthetically available light. The spectrum of incident light at the mat surface was strongly modified over just a few tenths of a millimeter, leaving mostly yellow to red light of 550 to 650 nm for the subsurface mat-building organisms. At the deepest photosynthesis maximum in mat C (at 2.2 mm; Fig. 1), 1% of 600-nm light but only 0.1% of 450-nm light remained, i.e., a 10-fold difference. Although these relative spectral changes were quite similar among the three mats investigated, there were significant differences in the rate of light attenuation. The relative attenuation of blue light of 450 nm over the top 0 to 1 mm of the mats ranged from 10,000-fold in mat A to only 10-fold in mat C (Fig. 1). It is not yet clear which pigments were responsible for efficient absorption of blue and green light. The diatoms at the surface contain fucoxanthin as the dominant carotenoid, while the cyanobacteria generally contain myxoxanthophyll or other xanthophylls (22). The lack of carotenoid-based photosynthetic activity in blue and green light of 450 to 520 nm in the cyanobacterial layers indicates that much of the pigment may not be associated with the thylakoid membranes but rather with the cell membranes or cell walls. Furthermore, strong yellow and brown pigmentation of the sheaths is known for several mat-forming cyanobacteria, e.g., *Lyngbya* sp.

The action spectra in combination with the light spectra at different depths in the mat (Fig. 2 and 3) show that the 600-nm wavelength is a useful average for estimating the distribution of photosynthetically usable light for the cyanobacteria. With a radiant flux of 500 $\mu E\ m^{-2}\ s^{-1}$ at the mat surface, the lower boundary of detectable photosynthesis in mats A, B, and C occurred where the downward radiance of 600-nm light had been reduced to 0.3, 0.6, and 0.2%, respectively. If calculated for the full spectrum of white light, the fraction remaining would be somewhat less than this, up to 50% lower (cf. Fig. 4). Although this indicates that the cyanobacteria are able to utilize quite low radiant energy fluxes, it should be noted that the microelectrode technique measures gross and not net photosynthesis. The rates measured at the lower boundary of the photic zone may therefore not be sufficient to balance the respiration of the organisms at this depth. Scattered light, on the other hand, which we did not measure, may have contributed significantly to the total light available in the lower photic zone.

The spectral distribution of light in the benthic mats was quite different from that of light in planktonic systems, where absorption by ambient water plays a relatively important role (12). In mats, absorption by pigments in combination with scattering by organisms and mineral grains are completely dominant. Discrimination between the effects of absorption and scattering requires more-detailed light measurements from which the downward and upward irradiance can be calculated. Such measurements were recently done and were used to calculate the total scalar irradiance in the different layers, i.e., the total radiant flux which the organisms receive from all directions (Jørgensen and Des Marais, in press). Similar measurements have been done for plant leaf tissue (33). The results have shown that the scalar

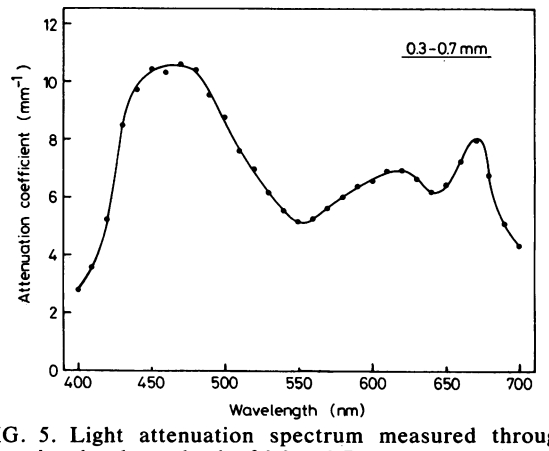

FIG. 5. Light attenuation spectrum measured through a *Microcoleus* band at a depth of 0.3 to 0.7 mm.

irradiance near the mat or leaf surface may be significantly higher than the radiant flux of the illuminating light source owing to high scattering in the tissue.

The use of a fiber-optic microprobe has important advantages over earlier approaches for measuring light in benthic environments. A spatial resolution of at least 100 μm is required to resolve many of the important features of benthic photosynthesis and light distribution (15, 26). The present data show that a spectral resolution of about 10 nm is necessary to resolve the strong qualitative changes in the photosynthetically available light. The present technique, however, still has serious limitations, mostly due to the lack of spectral analysis by the detector. Since many of the benthic phototrophic organisms migrate vertically in response to light, the use of a spectrally varying monochromatic light source is not ideal and precludes a number of interesting light adaptation studies.

A more specific problem was the very efficient penetration by violet light, which was unexpected owing to the generally stronger attenuation of shorter wavelengths in sediments (5). The observation was checked by absorption measurements on thin layers of mat material by the scattering technique (30) in a scanning spectrophotometer (Cary model 14). The results showed a distinct absorption minimum at 380 to 390 nm. We also checked that light of second-harmonic wavelengths from the monochromator did not affect measurements at the short wavelengths. The possibility still remains, however, that the violet light caused significant far-red fluorescence by Chl *a* in the mat, which would then penetrate more efficiently and erroneously be recorded as violet light.

Action spectra. Action spectra similar to those presented in Fig. 3A to D have been recorded for diatoms and cyanobacteria in *M. chthonoplastes* mats of the Solar Lake, Sinai (Y. Cohen and B. B. Jørgensen, unpublished results). All the action spectra of the *Microcoleus* community have shown very little Chl *a*-dependent activity. Phycobiliproteins were the dominant antenna pigments active in photosynthetic oxygen evolution. The maximum activity at 580 nm fell between the typical absorption maxima of phycoerythrin and phycoerythrocyanin (560 to 570 nm) and phycocyanin (610 to 630 nm) (23). On the basis of absorption and action spectra of pure cultures of cyanobacteria (7, 8, 21), a maximum at 580 nm would most probably imply both phycocyanin and phycoerythrin absorption. Phycoerythrin occurs at high concentration in some filamentous cyanobacteria of the LPP (29) group, especially in *Phormidium* spp., which is present in the mat, but it has not been reported for *M. chthonoplastes* (29). It also occurs in some *Oscillatoria*, *Aphanothece*, and *Aphanocapsa* strains, which are members of the mat community. Phycoerythrocyanin has been isolated from several strains of cyanobacteria but not from *M. chthonoplastes*. A closer evaluation of the active pigments requires concurrent measurements of photosynthesis and scalar irradiance at exactly the same point in a mat as well as pigment analyses.

Oxygenic photosynthesis requires simultaneous excitation and electron transport through both photosystems I and II. The light-harvesting pigments of photosystem II in cyanobacteria are allophycocyanin, phycocyanin, and sometimes phycoerythrin and phycoerythrocyanin, while reaction center Chl *a* is a very minor constituent (8). Since the action spectrum of the *Microcoleus* community in continuous, monochromatic light showed very predominant activity of the phycobilins, these pigments can evidently pass energy efficiently to both photosystems II and I. This has also been found in several pure-culture studies of cyanobacteria (20, 34). Antenna Chl *a* is associated with photosystem I in cyanobacteria. The light attenuation spectra showed abundant Chl *a* in the mats, but Chl *a* activity was rather insignificant in the cyanobacterial layers. In monochromatic light, this could be due to inefficient energy transfer from photosystem I to photosystem II, which is known to cause both blue and red drop in the quantum yield of cyanobacteria and red algae (1). However, addition of 600-nm background light to excite photosystem II still gave a low, although detectable, photosystem I activity. This indicated that Chl *a*, although abundant, is very inefficient as a light-harvesting pigment in the mat-forming cyanobacteria.

Allophycocyanin showed the opposite trend. Its concentration in the mat organisms was relatively low as judged from the attenuation minimum at 650 nm, yet it caused a maximum in the action spectrum. This is in accordance with the results of Lemasson et al. (21), who found that the relative efficiencies of the antenna pigments in cyanobacteria decreased as allophycocyanin > phycocyanin ≫ Chl *a*.

Previous studies on the action spectra of cyanobacteria have been done for planktonic species. In the mixed photic zone where they live in the sea or in lakes, they are frequently exposed to the full spectrum of daylight composition. These planktonic species generally show significant Chl *a* activity (13). In nitrogen-starved cells with a low phycobilin level, Chl *a* even becomes the dominant antenna pigment (21). In low-Chl *a* mutants of *Anacystis* sp., however, action spectra similar to those presented here, with only a slight Chl *a* shoulder, have been recorded (34). The possibility that the spectral light field in benthic communities, which is totally different from that of the water column, selected against Chl *a*-based photosynthesis is interesting. Such an adaptation would be an additional flexibility of cyanobacterial photosynthesis, different from the well-known chromatic adaptation caused by variations in the phycocyanin-to-phycoerythrin ratio (2).

By a combination of techniques based on microelectrodes and fiber optics, we have now reached a spatial resolution of about 100 μm in benthic photosynthesis studies. As shown here, it is possible to analyze the adaptation of the phototrophic organisms to different light intensities and spectral qualities without removing the organisms from their natural surroundings and without destroying the community structure. Concurrent studies of the microbial mats have also demonstrated the important role of light scattering as well as the complementary utilization of visible and near-infrared light between the oxygenic phototrophs and photosynthetic bacteria (Jørgensen and Des Marais, in press).

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