

DELAYED LIGHT PRODUCTION BY BLUE-GREEN ALGAE, RED ALGAE, AND PURPLE BACTERIA*

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INTRODUCTION

Green plants have been shown to emit light for some seconds after they have been illuminated. The action spectrum for the production of the delayed light has been shown to be the same as the action spectrum for photosynthesis (1). The emission spectrum for the delayed light has been shown to be the same as the emission spectrum for the fluorescence of the living plant (2).

In the present paper, these observations have been extended to include the blue-green algae, the red algae, and the purple bacteria.

Materials and Methods

The determinations of the action spectra were made by the method described by Strehler and Arnold (1) and the emission spectra of the blue-green algae and the purple bacteria were made by the method described by Arnold and Davidson (2). The emission spectrum of the red algae was made by M. J. Cormier, who dispersed the light by means of the f-4 Farrand monochromator; a photomultiplier immersed in liquid nitrogen was used as a quantum counter. Since the sensitivity, as a function of wave length, was not known for the photomultiplier, a determination of the emission spectrum of *Chlorella* was made under the same conditions as for the red algae.

Three species of blue-green algae were used. *Synechococcus cedrorum* was obtained from Dr. M. B. Allen of the University of California and was grown in Allen's No. 4 medium (3). *Anacystis nidulans*, also obtained from Dr. Allen, was grown in the Gerloff *et al.* (4) modification of Chu No. 10. An unidentified species of *Lyngbya*, isolated by E. S. Meek in this laboratory, was grown in the Chu No. 10 modification, minus the organic components.

The red alga used, *Porphyridium cruentum*, was obtained from Dr. Richard C. Starr of the University of Indiana along with the growth requirements as set forth in the Punnett and Chalmers adaptation of R. H. Swain's medium for *Porphyridium cruentum*.

All the algae were grown at 25°C., bubbled with 5 per cent carbon dioxide in air, and were illuminated with the light from a sodium arc.

The three species of purple bacteria used were: *Rhodospirillum rubrum* and *Rho-*

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dopseudomonas palustris, obtained from Dr. C. B. van Niel of the Hopkins Marine Station, and *Rhodopseudomonas gelatinosa*, furnished by Dr. J. M. Siegel of the University of Arkansas. The purple bacteria were grown at 30°C. under anaerobic conditions in 1 per cent yeast extract, or in the enrichment medium of Siegel and Kamen (5) in 280 cc. glass-stoppered bottles, and were illuminated with incandescent lamps.

RESULTS

The action spectrum for the blue-green algae, *Synechococcus cedrorum*, is given by the top curve in Fig. 1, the ordinate being the relative effectiveness

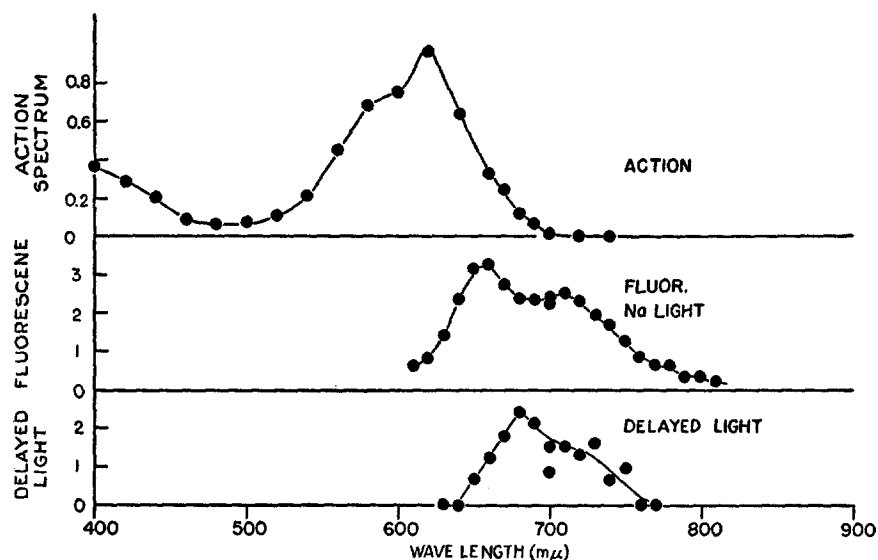


FIG. 1. For the blue-green algae, *Synechococcus cedrorum*, the action spectrum is given by the top curve, the fluorescent spectrum by the middle curve, and the emission spectrum for the delayed light production is given by the bottom curve.

per incident quantum in producing the delayed light. The middle curve shows the spectrum for the fluorescence of *Synechococcus* when excited by sodium light as the relative energy per unit of wave length. The bottom curve is the emission spectrum of the delayed light as relative energy per unit wave length.

The bottom curve in Fig. 2 is the action spectrum for the blue-green algae, *Anacystis nidulans*. The top curve gives the transmission of an aliquot of the suspension measured in a Beckman spectrophotometer.

The action spectrum for the production of delayed light for the red algae, *Porphyridium cruentum*, is given by the bottom curve of Fig. 3. The top curve is the transmission of an aliquot of the suspension. The emission spectrum of the delayed light emitted by this alga is given in Fig. 4, together with the

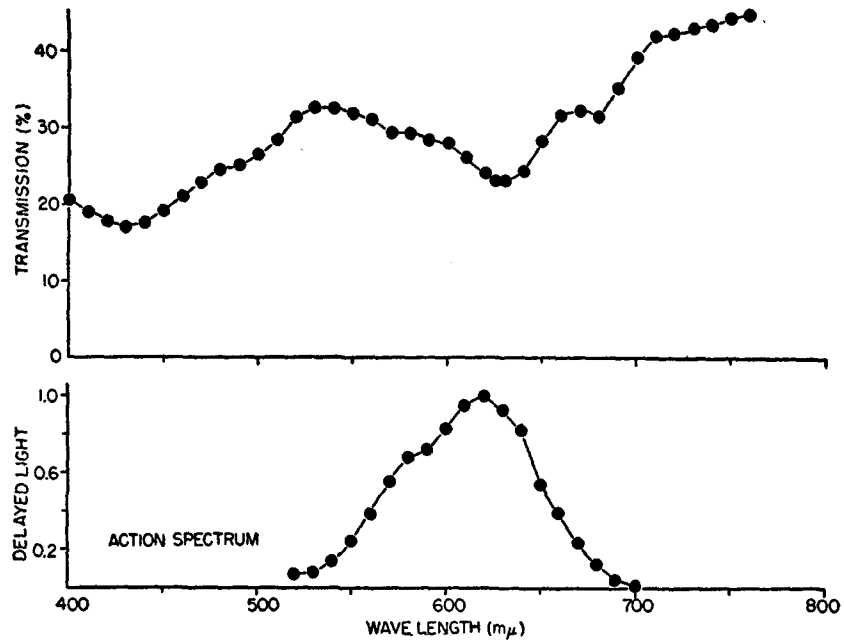


FIG. 2. The action spectrum for the blue-green algae, *Anacystis nidulans*, is given by the bottom curve, and the top curve is the transmission spectrum of the cell suspension.

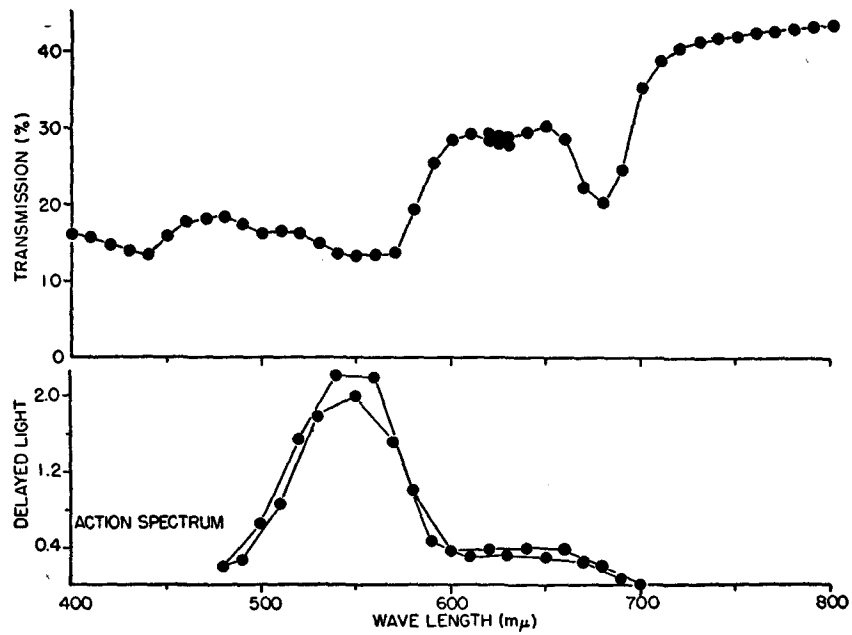


FIG. 3. The action spectrum for the red algae, *Porphyridium cruentum*, is given by the bottom curve, and the top curve is the transmission spectrum of the cell suspension.

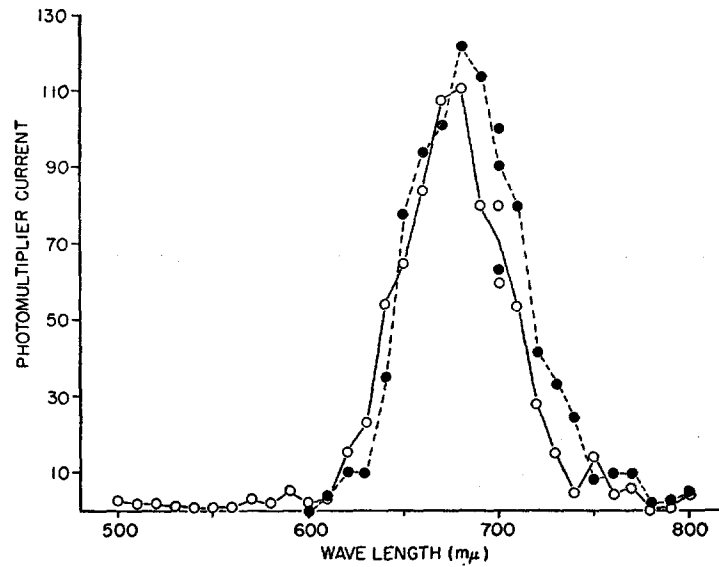


FIG. 4. The curve with the open circles gives the emission spectrum for the delayed light for the red algae, *Porphyridium cruentum*, and the curve with the closed circles gives the emission spectrum for the green algae, *Chlorella*. It should be noted that these curves are not corrected for the slit width of the monochromator or for the sensitivity of the photomultiplier.

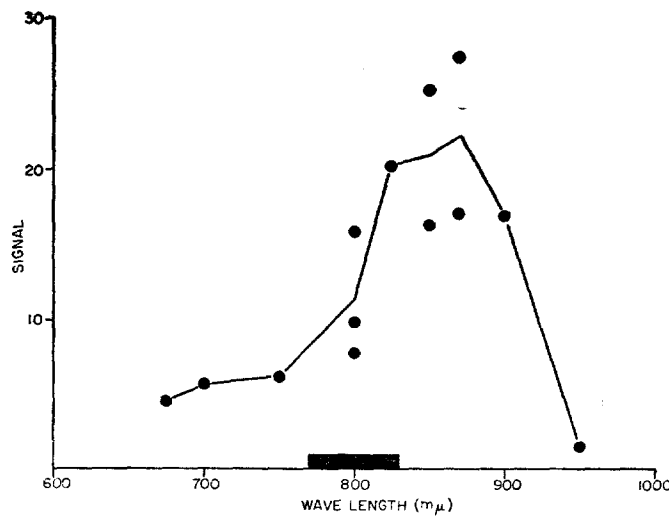


FIG. 5. The action spectrum for the purple bacterium, *Rhodospirillum rubrum*, is shown in Fig. 5.

curve for the delayed light emitted by *Chlorella*. It must be remembered that the ordinates here are the counts per second, minus the background, not corrected for slit width or the sensitivity of the photomultiplier.

Figs. 5 and 6 show, respectively, the action spectrum and the emission spectrum for the purple bacteria, *Rhodospirillum rubrum*. The black bars at the

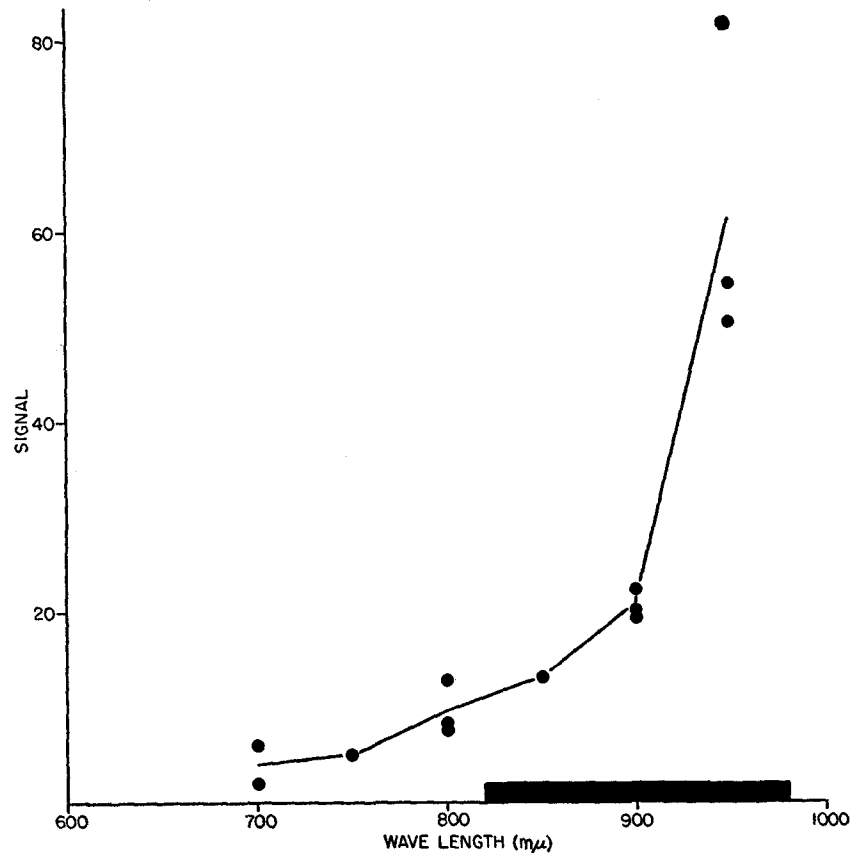


FIG. 6. The emission spectrum for the delayed light for the purple bacterium, *Rhodospirillum rubrum*.

bottom of the figures give the product of slit width in millimeters by the linear dispersion of the Farrand monochromator in millimicrons per millimeter.

DISCUSSION

That the emission spectra of the delayed light for the blue-green algae and for the red algae are the same as for *Chlorella* may be seen by comparing the bottom curve in Fig. 1 with the curve for *Chlorella*, published by Arnold and

Davidson (2), and by comparing the two curves given in Fig. 4 with one another. Thus it must be that chlorophyll emits the delayed light.

The action spectrum for the delayed light production by the blue-green alga, shown in Figs. 1 and 2, consists of one broad band with a peak at 620 $m\mu$. The same curve was also found for *Lyngbya*. It should be noted that the action spectrum shows no trace of a peak at 680 $m\mu$. Using the Beckman spectrophotometer, some of the blue-green cultures exhibited an absorption at 680 $m\mu$ due to chlorophyll that was fully as strong as the phycocyanin absorption at 620 $m\mu$; however, the action spectrum remained the same. The curves in Figs. 1 and 2 agree with the action spectra for photosynthesis and for chlorophyll fluorescence in blue-green algae described by Duysens (6), and with the action spectrum for photosynthesis in blue-green algae given by Haxo and Blinks (7). However, they do not agree with the action spectrum for photosynthesis in the blue-green algae, *Chroococcus*, given by Emerson and Lewis (8) in which the chlorophyll was found to be fully active.

The action spectrum for the red algae (Fig. 3) has a peak at 550 $m\mu$ and a relatively flat shoulder between 600 and 660 $m\mu$ and falls off to zero at about 700 $m\mu$. This agrees fairly well with the action spectra for photosynthesis and chlorophyll fluorescence of the red algae given by Duysens (6) and with the action spectra for photosynthesis given by Haxo and Blinks (7). Again it should be noted that there is no trace of active chlorophyll absorption in the action spectrum although the transmission curve, as made on the Beckman, shows a strong band due to chlorophyll at 680 $m\mu$ that is considerably larger than the phycocyanin band at 620 $m\mu$.

In both the blue-green and red algae, certainly for delayed light production and chlorophyll fluorescence, and presumably for photosynthesis, energy is transferred from the "accessory pigments," phycocyanin and phycoerythrin, to a part of the chlorophyll; this is demonstrated by the fact that the action spectrum is that of the "accessory pigments," and the emission spectrum is that of chlorophyll. The larger part of the chlorophyll, however, must be so located that it does not absorb energy from the accessory pigments, and so that the energy that it does absorb is wasted as heat. At least, this energy does not appear as delayed light, chlorophyll fluorescence, or as photosynthesis.

Dr. Conrad Yocum, in a private communication, stated that he had placed red algae under deep red light for several days and then found a peak in the action spectrum at 680 $m\mu$, corresponding to an active absorption by chlorophyll. This experiment has been repeated in this laboratory without finding any change in the action spectrum. However, Yocum's experiment, and that of Emerson and Lewis (8) on *Chroococcus*, show that there are conditions, not yet well defined or understood, in which the chlorophyll in blue-green and red algae can all be active.

Most of the delayed light emitted by the purple bacteria has a wave length longer than 900 $m\mu$ (Fig. 6), and thus falls in a region in which the sensitivity of our photomultiplier is quite small (see Table I, of reference 2). It was necessary to use heavy suspensions along with wide slits on the monochromator in order to make the signal large enough for measuring.

The action spectrum given in Fig. 5 was made with a suspension transmitting about 50 per cent of the light in the 800 to 900 $m\mu$ region. This was much heavier than the suspensions, that transmit 85 to 95 per cent of the light, that are generally used for making an action spectrum. The wide slits (shown by the black bars in Figs. 5 and 6) completely hide the finer details in the spectrum that might be due to the different bacteriochlorophylls, B800, B850, and B890, that have been described by Duysens (6).

Essentially the same results were obtained with all the species available. Description of the details of the action and emission spectra of the delayed light from purple bacteria must wait for an instrument of far greater light-gathering power than the f-4 monochromator in this laboratory, or for some means of detecting infrared light much more sensitive than the present photomultipliers.

SUMMARY

1. Blue-green algae, red algae, and purple bacteria all show the emission of delayed light.
2. The action spectra for the production of delayed light by three species of blue-green algae have one broad band with a peak at 620 $m\mu$.
3. The action spectrum for production of delayed light by the red algae has one peak at 550 $m\mu$ with a shoulder from 600 to 660 $m\mu$.
4. The emission spectra of the delayed light from both the blue-green and red algae were the same as from the green algae, *Chlorella*.
5. The action spectra for the production of delayed light by the different species of purple bacteria tested consisted of one or more bands not resolved between 800 and 900 $m\mu$.
6. The emission spectrum of the delayed light from the purple bacteria was largely at wave lengths longer than 900 $m\mu$.

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