Spectral Sensitivity of the Common
Prawn, *Palaemonetes vulgaris*

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**ABSTRACT** The vision of *Palaemonetes* is of particular interest in view of ex-
tensive studies of the responses of its chromatophore systems and eye pigments
to light. The spectral sensitivity is here examined under conditions of dark
adaptation and adaptation to bright colored lights. In each case the relative
number of photons per one-fiftieth sec flash needed to evoke a constant peak
amplitude (usually 25 or 50 μV) in the electroretinogram (ERG) was measured
at various wavelengths throughout the spectrum. The sensitivity is the reciprocal
of this number. In dark-adapted animals the spectral sensitivity curve consists
of a broad, almost symmetrical band, maximal at about 540 μmλ, with a shoulder
near 390 μmλ. Adaptation to bright red or blue light, left on continuously
throughout the measurements, depresses the 540 μmλ peak without notably
changing its shape or position, implying that only one visual pigment operates
in this region. Adaptation to red light, however, spares a violet-sensitive system,
so that a high, narrow peak at 390 μmλ now dominates the spectral sensitivity
function. The 540 and 390 μmλ peaks are apparently associated with different
visual pigments; and these seem to be segregated in different receptor systems,
since the associated ERG's have markedly different time constants. It is sug-
gested that these two sensitivity bands may represent the red- and violet-sensitive
components of an apparatus for color differentiation.

The common prawn, *Palaemonetes vulgaris*, has been a classic object for the
investigation of visual phenomena since Parker (1897) used this animal for
the first detailed study of pigment migration in a crustacean eye, and Perkins
(1928) discovered here the eye stalk hormone. There was also an early study
of the spectral sensitivity of *Palaemonetes larvae*, carried out by observing their
distribution in two beams of light at right angles to each other, one white,
the other restricted to broad bands of the spectrum (White, 1924); the maxi-
imum phototactic effectiveness was found in the blue-green, at 470–510 μmλ.

We report here electroretinographic measurements of the spectral sensitivity
of adult animals, dark-adapted and adapted to bright colored lights. Two
visual systems seem to be present, with maximal sensitivities at about 390
and 540 μmλ, segregated apparently in different receptors.
APPARATUS AND PROCEDURE

These experiments employed much the same procedures as those of Wald and Krainin (1963) and Wald (1968). The light of a 100 w zirconium arc was projected with quartz lenses through a Bausch and Lomb grating monochromator, which transmitted a wave band 5 μm wide throughout the spectrum. A photographic shutter at the exit slit controlled the exposure, and another quartz lens focused the beam upon a pair of quartz neutral wedges, rotating in opposite directions so as to compensate each other. The wedges together yielded a range of densities of about 3.4 which could be extended further with neutral filters. The light emerging from the wedges was focused on the eye.

![Figure 1](image-url)  
**Figure 1.** Spectral transmissions of the blue and red filters used to provide the colored adapting lights in these experiments.

The energy distribution of this system was calibrated with a Moll thermopile (Kipp and Zonen, Delft, Holland) and an electronic microvoltammeter (Keithley 150A). The wedge transmission was calibrated with a Welch Densichron (Welch Scientific Co., Skokie, Ill.) at 30-50 μm intervals from 330-680 μm. Since it varied somewhat from one end of the spectrum to the other, three calibration curves were used.

To adapt the eye to bright colored lights, the light from a tungsten filament projection lamp, after passing through color filters, was reflected from the surface of a thin glass cover slip held at an angle of 45° in the light path of the test flash, so that when the adapting light was on, the test flash coming through the cover slip was co-axial with it. The two ends of the spectrum were isolated with either the red Corning filter 2403, which transmits all wavelengths longer than 635 μm; or the blue Jena filter combination BG 3 + BG 18, which transmits a broad band of radiation shorter than 470 μm. The transmissions of these filters are shown in Fig. 1. No attempt was made to measure the brightnesses of the adapting lights, which were adjusted in each case to yield the desired depression of sensitivity.

Electroretinograms were measured with a Tektronix 502 dual beam oscilloscope and a Grass P5 AC preamplifier set for the frequency range 0.1 to 100 cps, run from
a Grass regulated power supply. The ERG was photographed with a Polaroid camera mounted on the oscilloscope.

Animals, 3–4 cm long, were mounted on a pad of absorbent cotton on a 4 cm cork disc, and held in place with insect pins bent over them. The cork platform stood in a bath of oxygenated seawater so that the cotton pad was partly submerged while the animal lay above the surface. The eye was immobilized by bringing the eye stalk through a small stretched slit in a sheet of rubber dam, which also helped to insulate the eye electrically. The active electrode was a fine cotton wick touching the outermost pole of the eye. This was held in the capillary tip of a glass tube filled with seawater, connected through a silver–silver chloride wire with the recording system. Another such nonpolarizable wire dipping into the bath of seawater acted as indifferent electrode.

**MEASUREMENTS**

All the measurements involved determining at each wavelength the relative number of photons per 18 msec flash needed to evoke a constant peak amplitude in the ERG, usually 25 or 50 μv. The sensitivity is the reciprocal of this number.
Fig. 2 shows typical spectral sensitivity curves measured in a dark-adapted animal, one adapted to bright red light, and one adapted to bright blue light.

In dark-adapted animals this curve consists of a broad, almost symmetrical band, maximal at 535–540 m\(\mu\), with a shoulder in the far violet, owing to a small secondary maximum near 390 m\(\mu\). The absolute sensitivity of *Palaemonetes* at the maximum is only about one hundredth as great as those of the crayfishes earlier examined (cf. Wald, 1968).

Adaptation to bright red light, as shown in Fig. 2, while lowering the sensitivity in the 540 m\(\mu\) region by almost 2 log units, spares a violet-sensitive system, represented by a high narrow peak maximal at about 390 m\(\mu\), so that it now dominates the spectral sensitivity function.

Adaptation to bright blue light, though it also greatly depresses the 540 m\(\mu\) band, hardly alters its shape or position. In Fig. 2 the blue-adapted spectrum is shifted about 10 m\(\mu\) toward longer wavelengths than the dark-adapted curve, its \(\lambda_{\text{max}}\) coming at about 545 m\(\mu\); but this change hardly exceeds the ordinary variation in such curves, and is contributed to in this instance by the selective adaptation of the violet receptor, which had so declined in sensitivity owing to the blue adaptation that measurements could not be made below 450 m\(\mu\).

The differential effects of red and blue adaptation seem clearly to indicate the presence of at least two visual pigments, responsible for the sensitivity bands at 390 and 540 m\(\mu\). The small changes induced in the shape and position of the 540 m\(\mu\) band by red and blue adaptation seem to imply that only one visual pigment functions in that region.
To gain some indication as to whether the 390 and the 540 m\(\mu\) pigments operate in the same or different receptors, we have recorded the ERG under conditions in which each of these pigments functions virtually alone (Fig. 3). As Fig. 2 shows, at 410 m\(\mu\) in the red-adapted eye, the response is almost entirely due to the violet-sensitive pigment; whereas at 620 m\(\mu\) in the blue-adapted eye it must be entirely due to the red-sensitive pigment. In each case the intensity was adjusted so as to evoke about the same amplitude of ERG, about 100 \(\mu\)v. Under these conditions the responses displayed widely different time constants. That associated with the red-sensitive pigment rose, peaked, and declined markedly faster than that due to the violet-sensitive pigment. Each of these responses, recorded in triplicate, showed these differences consistently.

Though no certain indication, this makes it most probable that the violet- and red-sensitive pigments act in different receptor systems.

**DISCUSSION**

Almost simultaneously with a first report of these experiments (Wald and Seldin, 1967), Goldsmith, Dizon, and Fernández (1967) announced the results of microspectrophotometric measurements of light-sensitive pigments in single rhabdoms of *Palaemonetes*. Two pigments were found: a dominant pigment with \(\lambda_{\text{max}} = 555\ m\mu\), that bleached in the light to a long-lived intermediate (metarhodopsin) with \(\lambda_{\text{max}} = 496\ m\mu\); and a minor pigment with \(\lambda_{\text{max}} = 496\ m\mu\), that bleached without detectable intermediates. (All these \(\lambda_{\text{max}}\) are for the difference spectra.)

These pigments resemble greatly in their properties similar pairs of pigments ("iodopsins" and "rhodopsins") recently extracted from two species of crayfish (Wald 1967). In each of the crayfish the long-wavelength pigment, which as in *Palaemonetes* bleached over a long-lived metarhodopsin, could be

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**Table I**

<table>
<thead>
<tr>
<th>Animal</th>
<th>Difference spectra</th>
<th>Spectral sensitivities</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(\lambda_{\text{max}}) m(\mu)</td>
<td>(\lambda_{\text{max}}) m(\mu)</td>
</tr>
<tr>
<td>Northern crayfish (<em>Orconectes virilis</em>)</td>
<td>510, 562</td>
<td>435, 570</td>
</tr>
<tr>
<td>Swamp crayfish (<em>Procambarus clarkii</em>)</td>
<td>525, 558</td>
<td>450, 575</td>
</tr>
<tr>
<td>Prawn (<em>Palaemonetes vulgaris</em>)</td>
<td>496, 555</td>
<td>390, 540</td>
</tr>
</tbody>
</table>
identified with the electroretinographically measured red-sensitive component of an apparently two-component system of color vision, containing also a second, blue-sensitive component for which no photosensitive pigment was found in extracts of the eyes. Each of the crayfish extracts, as here the *Palaemonetes* rhabdom, contained also a rapidly bleaching pigment with \( \lambda_{\text{max}} \) near 500 \( \mu \) for which no ERG sensitivity peak could be found.

The extraordinary parallelism between these observations on crayfish and

![Figure 4. Comparison of the major spectral sensitivity band in the dark-adapted prawn with the Dartnall nomogram for the absorption spectrum of a visual pigment absorbing maximally at 535 \( \mu \). The agreement is reasonably good, yet the spectral sensitivity band is slightly narrower than the Dartnall function.](image)

*Palaemonetes* is apparent from Table I. It seems likely that in *Palaemonetes* also the red-sensitive pigment found in the rhabdom is the source of the spectral sensitivity function peaking at 540 \( \mu \). That the \( \lambda_{\text{max}} \) of its difference spectrum lies somewhat farther toward the red is to be expected of a pigment that bleaches to a metarhodopsin that still absorbs strongly in the visible. We would suggest that in parallel with the crayfish results, the visual function associated with the 496 \( \mu \) pigment has escaped our electrophysiological measurements; and that conversely the visual pigment responsible for our sensitivity peak at 390 \( \mu \) escaped the microspectrophotometry of the rhabdom.

The alternative possibility, that our 540 \( \mu \) sensitivity band is made up of contributions from both the 496 and 555 \( \mu \) pigments found by Goldsmith et al. seems excluded by the following considerations: (a) In that case red and blue adaptations should shift the band widely back and forth in the spectrum, whereas they hardly affect its shape and position. (b) Two pigments so unlike in their kinetics of bleaching could hardly maintain the same relative relationship in light and dark adaptation, as these would have to do to yield the nearly invariant spectral sensitivity curves that we have measured. (c) Were the sensitivity band at 540 \( \mu \) compounded of two visual pigments, that should broaden it as compared with the absorption spectrum of either
pigment. The absorption spectra of visual pigments tend to approximate the curves summarized in Dartnall's nomogram (1953). As Fig. 4 shows, the spectral sensitivity of *Palaemonetes* in the region of 540 μm fits the Dartnall nomogram for a visual pigment with λ<sub>max</sub> 535 μm quite closely, yet if anything is a little narrower rather than broader than the nomogram.

All these considerations lead us to believe that the 540 μm band revealed by our ERG measurements is based upon a single visual pigment; and that the visual system that emerges in such measurements consists primarily of this and a minor spectral sensitivity band at about 390 μm. These together may constitute the red- and violet-sensitive components of an apparatus for color differentiation.

As in the crayfishes, that leaves a third light-sensitive pigment, at about 496 μm in *Palaemonetes*, that does not seem to give rise to an ERG-sensitivity curve under the conditions examined, and whose function is still to be explored. This could be a highly important function, and yet not loom large in the ERG. It is interesting in this regard that the phototactic sensitivity of *Palaemonetes* larvae measured many years ago by White (1924) seemed to center in this region of the spectrum (470–510 μm). This would be well worth reinvestigation; as would be other measures of visual response. The electroretinogram yields a very limited type of signal, that can readily miss not only small but slow changes of potential as ordinarily measured.

These experiments were performed during the summer of 1965, and the results were reported at the meeting of the Society of General Physiologists in Woods Hole in September, 1967 (Wald and Seldin, 1967). This investigation was supported in part with funds from Harvard University and the National Science Foundation.

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REFERENCES


