Photopic Vision in Eels

Evidences of Color Discrimination

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ABSTRACT: Several classes of second-order retinal neurons have been studied electrophysiologically in European eel (*Anguilla anguilla*) from two different localities, Lake Seliger in Russia and the coastal waters of the Adriatic Sea in Montenegro. The majority of L-horizontal cells (68 explored) had both rod and cone inputs, an uncommon phenomenon among teleosts. Pronounced color-opponent properties, often taken as pointing to the capacity of color vision, were identified in one amacrine cell, apparently of the "blue/yellow" (or "blue/green") type. Microspectrophotometric measurements revealed two different spectral classes of cones with absorption maxima at about 525 and 434 nm. The existence of green-sensitive and blue-sensitive cone units was thus revealed by both electrophysiological and microspectrophotometric techniques.

KEYWORDS: amacrine cells; bipolar cells; horizontal cell; color vision; microspectrophotometry; European eel; spectral sensitivity

INTRODUCTION

The visual system of the eel changes dramatically during their migration from freshwater to a marine environment. In freshwater, yellow eels posses a duplex retina containing both cones and rods and their visual pigments are dominated by porphyropsins (A_2-based pigment). The rods of immature and early stages of silver eels contain a pigment-pair porphyropsin/rhodopsin ($P523_2/P501_1$) with λ_{max} close to 520 nm. During migration the ratio of the mixture changes and the λ_{max} shifts towards 500 nm. Thereafter, a transition occurs from the rod opsin of the yellow eel and early stages of the silver eel to a different rod opsin. The new opsin, forming an A_1-based visual pigment ($P482_1$), is inserted into retinal rods. The expression of new

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opsin during development was once thought to be unique to the eel, but Munz and McFarland[1] reported opsin change in the rods of cardinal fish (*Apogon brachygrammus*) from λ_{max} 482 nm in the larval stage to λ_{max} 494 nm in adults. There is also an opsin change in the rods of the deep-sea fish *Scopelarchus analis*.[2] Therefore, in addition to possessing paired visual pigments based on the same opsin, a feature not uncommon in teleosts,[3] the eel has the additional feature of a third pigment, another rhodopsin based on a different opsin that appears during the transition from the yellow to silver stage. The shift from long-wave to short-wave sensitive rhodopsin is generally interpreted as an adaptive change. It leads to sensitivity matching between visual pigment spectra and the spectral distribution of light in different water types: freshwater usually having a relative excess of long-wave photons, the deep sea having a preponderance of short-wave photons, and coastal waters being intermediate in spectral characteristics.[4]

In great contrast to the fairly accurately evaluated scotopic spectral sensitivity of eels[5] and the changes in rod pigments during metamorphosis, details of the pigment content of cones and how these change with maturation have not been determined. The photopic spectral sensitivity and the capacity of wavelength discrimination in the eel remain largely unknown.[6] In our electroretinogram (ERG) study of the European eel,[7] we did not succeed in determining the spectral characteristics of the photopic units, and the Purkinje shift in sensitivity could not be observed. In the American eel (*Anguilla rostrata*), however, a complete photopic, ERG-based spectral sensitivity curve was obtained by exposure to a bright white 20-Hz flickering light.[8] Working with an isolated and bleached retina and exploiting the fact that cone pigments regenerate after bleaching, while rod pigments do not, indications were obtained that in the absence of rod responses, two cone-like mechanisms were discernible: one with λ_{max} of 550 nm, and the other with λ_{max} below 450 nm. Unfortunately, more precise information about the cone spectral mechanisms was not offered.

Difficulties associated with the electroretinographic approach in cone spectral analysis[1,8] indicated that the elucidation of the photopic sensitivity mechanism in eels requires the application of the more precise techniques of intracellular exploration and microspectrophotometric measurements. We compared two groups of eels differing in their geographic origin (Lake Seliger in Russia, and coastal running waters along the South Adriatic in Montenegro). Several classes of second-order neurons were successfully explored and the functional characteristics of a number of photopic units defined, including color opponency.

MATERIALS AND METHODS

Animals

Two groups of European eels (*Anguilla anguilla*) were used for electrophysiology. One group consisted of 16 yellow-stage eels (4^+–5^+, $L < 40$cm) electrofished during winter months in the mouth regions of creeks inflowing into the Boka Kotorska Bay (Southern Adriatic, Montenegro; referred to as "Adriatic eels" in the text). They were transported to the Moscow laboratory and kept for at least 15 days at 10–15°C in cold-room freshwater aquaria. The second group consisted by nine considerably larger eels ($L > 50$ cm) captured with hoop nets, at the end of October, in the

freshwater Lake Seliger (Russia, Tver region; referred to as "Seliger eels" in the text). They were also kept for at least 7 days prior to the experiments in freshwater aquaria (water temperature 12–15°C) without feeding.

Histology

For histological analysis eyecups were prepared from eyeballs excised after rapid decapitation of the fish. Cornea, lens, and most of the vitreous were removed surgically. The eyecups were fixed in Bouin solution, embedded in paraffin, sectioned into 8 μm thick slices, and finally stained with hematoxylin. Radial sections of the retina were studied under a light microscope.

Microspectrophotometry

An eel was purchased commercially in London, dark-adapted overnight, then sacrificed by cervical dislocation. Eyes were enucleated, hemisected, and the anterior portion discarded. The retina was then separated from the pigment epithelium and small samples were prepared for measurements. All preparations were carried out under a dim red safelight. The methods of tissue preparation, recording spectra, and the design of the microspectrophotometer, a modified Liebman dual beam instrument, are described elsewhere.[9] Spectra were recorded at 2-nm intervals from 750–370 nm, and from 371–749 nm on the return scan. The outward and return scans were averaged. A baseline spectrum was measured for each cell, with both beams in an unoccupied area close to the cell, and subtracted to form the final spectrum. Two (or more) baseline spectra were recorded for each cell and averaged. Cells were routinely bleached by exposure to a beam of white light from the monochromator and the post-bleach spectrum recorded from which difference spectra were calculated.

Electrophysiology

Eyecups were cut into two approximately equal fragments. While experimenting with one eyecup fragment, the remaining fragments from the two eyes were maintained in a refrigerator at 4°C. Four experiments were thus performed on one fish. In each case, the eyecup fragment was stretched, scleral side down, over a plastic ball, fixed in a small platform inside a lightproof Faraday cage. Since residues of the vitreous are known to prevent the oxygenation of the retina, the vitreous was expelled by means of forced perfusion. The perfusion was continued during the entire experiment using teleost Ringer at 10–12°C.

Microelectrodes with 100–400 MΩ resistance, filled with 2 M potassium acetate, were used for intracellular recording. The reference electrode was positioned on a strand of filter paper connected with the eyecup and soaked with teleost Ringer. Electrodes were connected to the input stage of a microelectrode preamplifier. The amplified signals were recorded using an *X-Y* plotter.

For photostimulation, a halogen lamp was used with a series of eight interference filters (427, 470, 490, 543, 587, 620, 648, and 664 nm), with 50% bandwidths of 10 to 12 nm. Quantum intensities of unattenuated stimuli of different wavelengths were made equal using a calibrated selenium photocell. In all our figures, light intensity is expressed in units of the neutral density filters (NDF) used for attenuation, log

I=0.0 corresponding to 3.2×10^3 quanta $\cdot \mu m^{-1} \cdot sec^{-1}$. The duration of the photostimuli was usually around 1.3 seconds.

Action Spectra

The relative quantum spectral sensitivity (S_q), varying within the range [0–1], was calculated for different stimulus wavelengths (λ), λ_{max} representing the stimulus wavelength at which the maximal S_q value is observed. S_q was defined as the reciprocal of quanta necessary to evoke the response of a given (standard) amplitude and was calculated from the spectral response curves using V-log I profiles for monochromatic or white light stimuli. It is well known that eel rods possess rhodopsin with λ_{max} close to 500 nm (r500) and porphyropsin with λ_{max} close to 523 nm (p523), the ratio between them changing during sexual maturation.[4,6,10] In all our figures in FIGURES 5a–c therefore, S_q values are accompanied with data on r500 or p523 absorption spectra, calculated according to Maksimov.[11]

Statistical differences between means of individual spectral sensitivity data, and differences between λ_{max} values, obtained by fitting spectral sensitivity data-points in individual fishes by the procedure of Maksimov, were evaluated by means of the nonparametric two-tailed Mann-Whitney test, considering $P>0.05$ as not significant.

The Ocular Index

Experimental eels were characterized by their eye index, *I*, according to Pankhurst[12]:

$$I = \frac{\left[\left(\frac{A+B}{4}\right)\right] \cdot \pi}{L} \cdot 100,$$

where *A* and *B* are the horizontal and the vertical diameter of the eye (in mm), while *L* is the total body length (in cm). All Adriatic eels, classified as yellow eels were characterized by $L < 40$cm and $I < 2$. Eels from Lake Seliger were longer than 50 cm, and had an eye index between 6.5 and 13. No attempt was made to further differentiate, on the basis of *I* and *L*, between males and females, and between migratory and non-migratory forms.

RESULTS

Retinal Morphology

Radial sections of the retina (FIG. 1) revealed a highly developed pigment epithelium. In the outer nuclear layer two types of nuclei were distinguished: the nuclei above the external limiting membrane (ELM) belonging to cones, and several sublayers of much smaller and optically denser rod nuclei below ELM. The rod/cone ratio in Seliger eels was 40:1, twice as high as in the much smaller Adriatic yellow eels (20:1), but still much lower than the ratio known to characterize eels of the silver stage (100:1 and 250:1, according to Braekvelt[13] and Pankhurst[14], respectively).

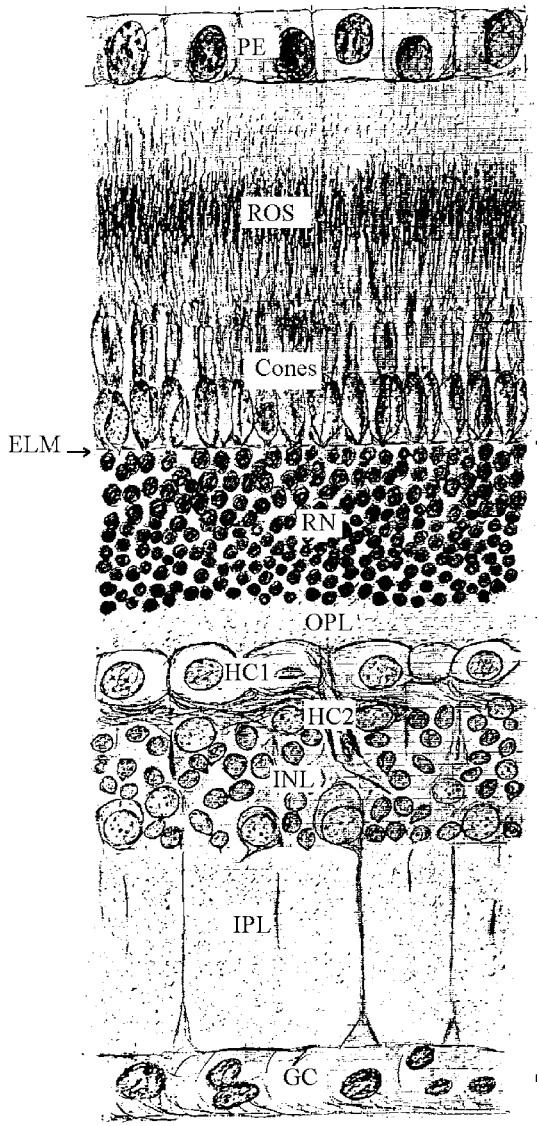

FIGURE 1. Retinal morphology in European eels (semischematic drawing of a radial section). ELM, external limiting membrane; GC, ganglion cells; HC1 and HC2, horizontal cells of the first and second layer, respectively; INL, inner nuclear layer; IPL, inner plexiform layer; OPI, outer plexiform layer; PE, pigment epithelium; RN, rod nuclei; ROS, rod outer segments.[25]

Only one compact layer of horizontal cells (HCs) was clearly distinguished, although HCs of the second layer could also be observed occasionally. In the inner nuclear layer (INL), containing bipolar and amacrine cells, the total number of cells was significantly smaller than in the outer nuclear layer (ONL). The rod/INL ratio in the yellow eel retinas was approximately 3:1, whereas in the larger Seliger eels it amounted to 6:1. The ganglion cell (GC) layer contained considerably fewer cells than the INL. The observed relation between retinal layers (numerous photoreceptors, much less abundant INL cells and relatively scarce GCs) points to a high degree of convergence of signals from photoreceptors in the European eel.

Electrophysiology

Horizontal Cell Responses

All explored horizontal cells belonged to the luminosity-type cells (L-HCs). Among the 33 such cells explored in Seliger eels, 6 were cone-driven (cone-HCs), 9 were rod-driven (rod-HCs), and 18 were driven by both rods and cones (mixed-HCs). In 35 HCs explored in yellow Adriatic eels, 5 were rod-HCs, 12 were cone-HCs, and 18 were mixed-HCs, and (FIG. 2A). In rod-HCs (FIG. 2B, upper panel), light offset was regularly followed, particularly at high stimulus intensities, by a sustained hyperpolarizing plateau (after potential) and the response decayed relatively slowly. At maximal intensity (white light, NDF = 0.0), the after potential plateau was sustained for up to 10–15 seconds. In the case of stimulation with monochromatic light of λ=490 nm, saturating levels were reached around NDF=−0.9. In cone-HCs (FIG. 2B, lower panel) responses decayed much faster than in rod-HCs. The after potential was not present even at maximal light intensities (white light, NDF = 0.0) and the saturation of the response was not reached. In the mixed-HCs, the stimulus offset was first followed by a fast cone component and then by a slow rod component (FIG. 2C). In the majority of the mixed-HCs from both experimental groups, cone inputs were dominant. In some cells, cone and rod components were of approximately equal amplitude, and in two cells from Adriatic eels, the rod component was dominant. When mixed-HCs were stimulated in the presence of different monochromatic backgrounds (red or blue), the shape of the response changed; only the cone component was observed at light offset (FIG. 2C, the second and third record on the right). Sometimes the cone component revealed a positive off-peak, increasing with background intensity. Similar but smaller and wider off-peaks were observed in some of the rod-HCs as well (FIG. 2B).

The spatial properties of HCs were not investigated in detail. In the case of the cone-HC response shown in FIGURE 2D (right), the light spot was 0.7 mm. The strong additional hyperpolarization, evoked by annulus stimulation, indicated that the receptive field of this HC was very large, larger by far than 1 mm. On the other hand, in the case of one mixed-HC (FIG. 2D, left), the light spot of a considerably smaller diameter (0.27 mm) evoked an almost maximal response; the additional hyperpolarization caused by the annulus was hardly noticeable. These two HCs, shown in FIGURE 2D, represent two extremes: HCs with the smallest and the largest receptive fields from among the presently explored horizontal cells.

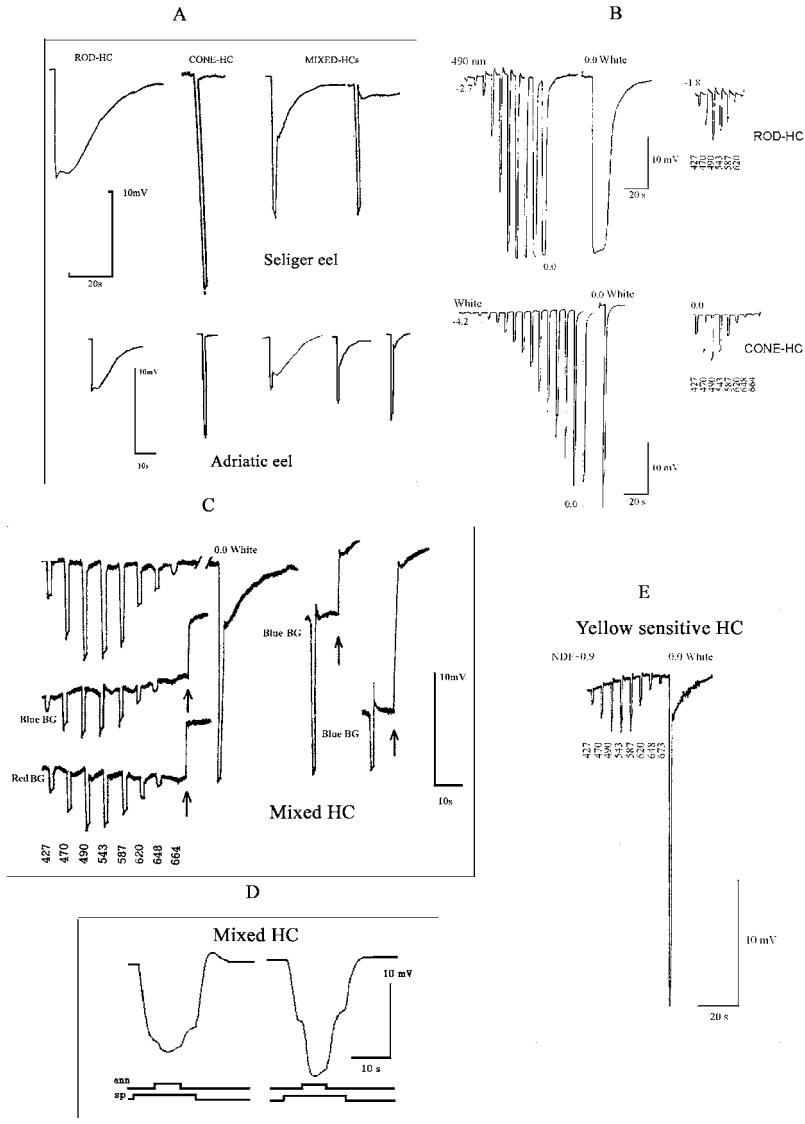

FIGURE 2. Responses of horizontal cells (HCs) to various modalities of photostimulation. (**A**) Examples of the three types (as indicated) of intracellular records of HCs responses to white light stimuli (NDF=0.0, 1.3 sec). Note that in Adriatic eels rod-HCs are lacking and that a substantially greater amplification had to be applied. (**B**) Responses of cone- and rod-driven HCs to incremental stimulation by 490 nm light flashes (0.3 log unit increments) and by a white light flash of maximal intensity. (**C**) Responses of a mixed-type HC (Seliger eel) to white light, as influenced by blue and red background illumination of increasing intensity (*right*: blue background two steps, second and third record). Note that in the presence of background illumination (ending at *arrows*) only the cone component is

Bipolar Cell (BC) Responses

Our experiments revealed the presence of both on-BCs and off-BCs in the retina of the European eel (FIG. 3). Surround illumination by an annulus light stimulus, applied after the onset of a light spot stimulus, evoked a response of an opposite sign: depolarization in off-BCs, and hyperpolarization in on-BCs. The recordings from on-BCs in Adriatic eels were particularly frequent and stable, enabling the elucidation of their spectral properties (see later).

Amacrine Cell Responses

Amacrine cell responses were usually recorded at a retinal depth of 20–40 μm (FIG. 4). The majority of responses were of the transient type (hyperpolarizing or depolarizing; FIG. 4 a and b), with prominent on- and off-peaks, sometimes equal to, or greater than 10 mV. Some of the recorded amacrine cells responses were of the transient/sustained type (FIG. 4c), with a large positive on-wave followed by a sustained depolarization. Sometimes a negative peak appeared at stimulus offset (FIG. 4c, lower trace). One of the amacrine cells showed increasing amplitude of response after2 min of dark adaptation (FIG. 4d, lower trace).

Spectral Properties of Retinal Neurons

In the majority of the HCs, maximal responses were observed in the same middle-wave region of the spectrum, under scotopic as well as under photopic conditions (exemplified by records in FIG. 2B). This strongly indicates that rods and middle-wave-sensitive (MWS) cones contain visual pigments with similar absorption spectra. Two cells with cone input and one mixed cell with dominant cone input from an Adriatic eel showed maximal responses at longer wavelengths. Records obtained from HCs of Seliger eels belonged to three types: with cone, rod, and mixed inputs. The latter were studied under both photopic (high light intensities, $-0.6 \leq NDF \leq 0.0$) and scotopic conditions (low light intensities, $NDF \leq -1.2$). FIGURE 5A shows averaged data from 6 photopic and 4 scotopic responses. In both cases, fitted spectral sensitivity maxima were in the same region, close to the maximum for the rod porphyropsin (523 nm), and without a statistically significant difference between their means ($P > 0.05$). In Adriatic eels (FIG. 5B) spectral sensitivity was analyzed under photopic conditions in 9 cells. The fitted λ_{max} values for 6 cells was about 530 nm, whereas the other three cells had λ_{max} values around 543 nm. The fitted λ_{max} value of one cell with a rod input was 513 nm.

The longer-wave sensitivity of some of the HCs was strongly supported by our finding of maximal responses to yellow light (543–587 nm) in four on-BCs from Adriatic eels and in one off-BC from a Seliger eel (FIG. 4, right-hand records). In the majority of bipolar cells, responses were of a relatively small amplitude (less than

present at light stimulus offsets (signaled by conspicuous off-peaks, greatest in the last record). (**D**) Spatial properties of HCs from Seliger eels as evaluated by the spot-and-annulus test (sp, spot; ann, annulus). (*Left*) A mixed-type HC; spot diameter 0.27 mm. (*Right*) A cone-driven HC; spot diameter 0.7 mm. (**E**) Spectral responses of a yellow-sensitive HC (maximal response at 543 nm; Adriatic eel).

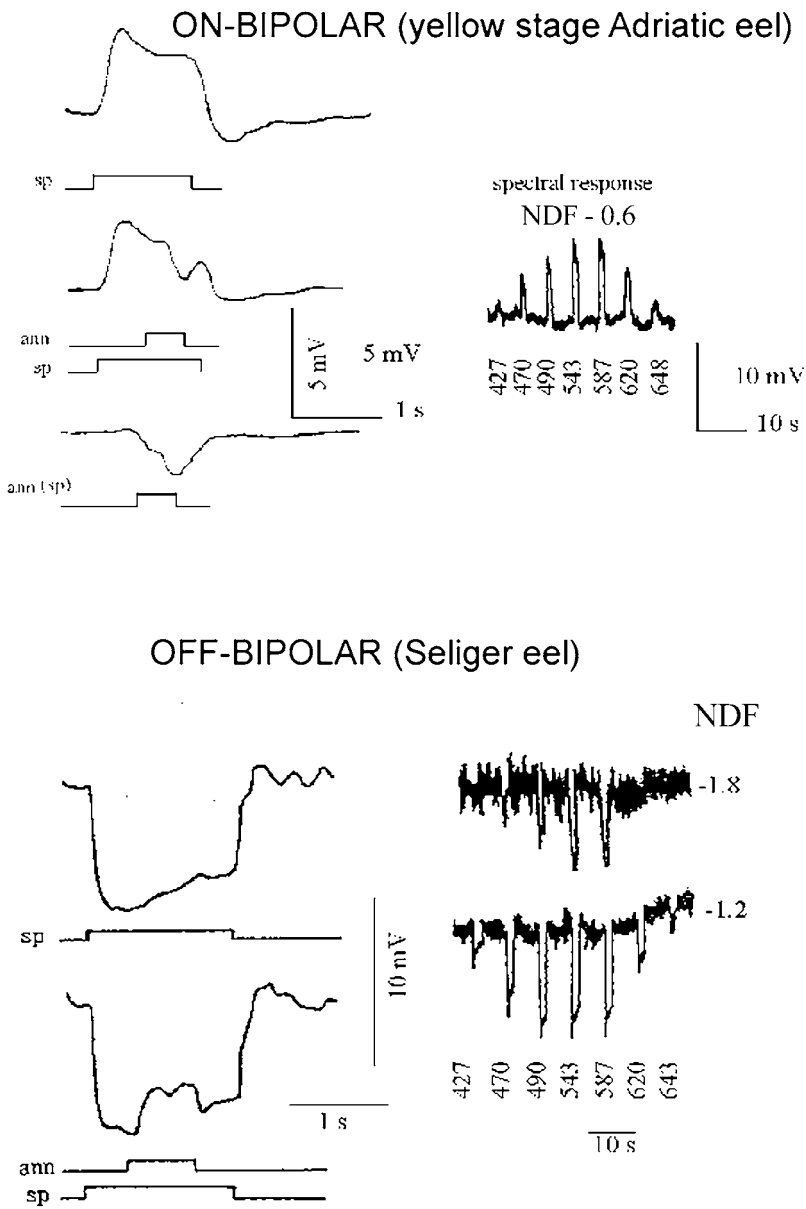

FIGURE 3. Responsiveness of bipolar cells. Responses to different stimulation patterns are shown on the left sides of the two panels (sp, spot; ann, annulus; ann(sp), annulus superimposed upon a continuous spot stimulus), and responses of the same cells to serial flashes of increasing wavelength (427–648 nm) are displayed on the right sides (calibration and NDF values as indicated). (*Upper panel*) On-bipolar cell from a yellow stage Adriatic eel. (*Lower panel*) Off-bipolar cell from a Seliger eel.

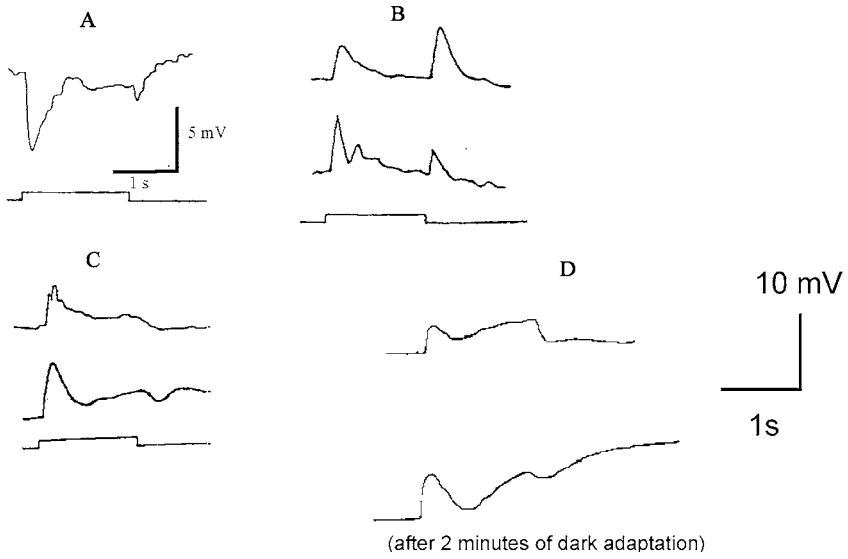

FIGURE 4. Types of amacrine cell responses to white light stimuli (Seliger eels). (**A**) one hyperpolarizing response; (**B**) two depolarizing responses; (**C**) two transient/sustained-type responses. (**D**) Adriatic eel: amacrine cell response to white stimuli before (*upper trace*) and after (*lower trace*) 2 min of dark adaptation.

10 mV) and unstable, not allowing precise spectral sensitivity measurements. However, in one on-BC from an Adriatic eel we succeeded in constructing a complete spectral sensitivity curve that had a maximum about 560 nm (NDF=–0.6; FIG. 5C).

One amacrine cell from a Seliger eel exhibited pronounced color-opponent properties (FIG. 7). At two intensities (NDF values of –0.3 and 0.0), the maximal response was obtained at λ=543 nm. However, the shape of the response was strongly dependent on stimulus wavelength. The response to λ= 490 nm and longer wavelength stimuli (543 nm in FIG. 7) consisted of an initial transient depolarization followed by hyperpolarization (FIG. 7, the second trace from the bottom). In contrast, stimulation at short wavelengths with λ=427 and 470 nm evoked depolarizing responses (FIG. 7, last trace), indicating that the amacrine cell was of the blue/yellow (B/Y) or blue/green (B/G) type.

Photoreceptors

Microspectrophotometry of photoreceptors from an eel caught off shore, when the retina was dominated by rhodopsin, identified at least two classes of single cone (FIG. 6). The majority of cones were MWS with λ_{max} values close to 525 nm, but there was also a sparse population of short-wave-sensitive (SWS) cones with λ_{max} at about 434 nm. Rods contain visual pigment with λ_{max} at 498 nm. The data are best fitted by a mixed visual pigment templates based on an 80%/20% mixture of A_1 and A_2 pigments. This would suggest λ_{max} for the pigment pairs of the MWS cones at about 520_1 and 545_2 nm and for the SWS cones of about 433_1 and 438_2 nm.[15]

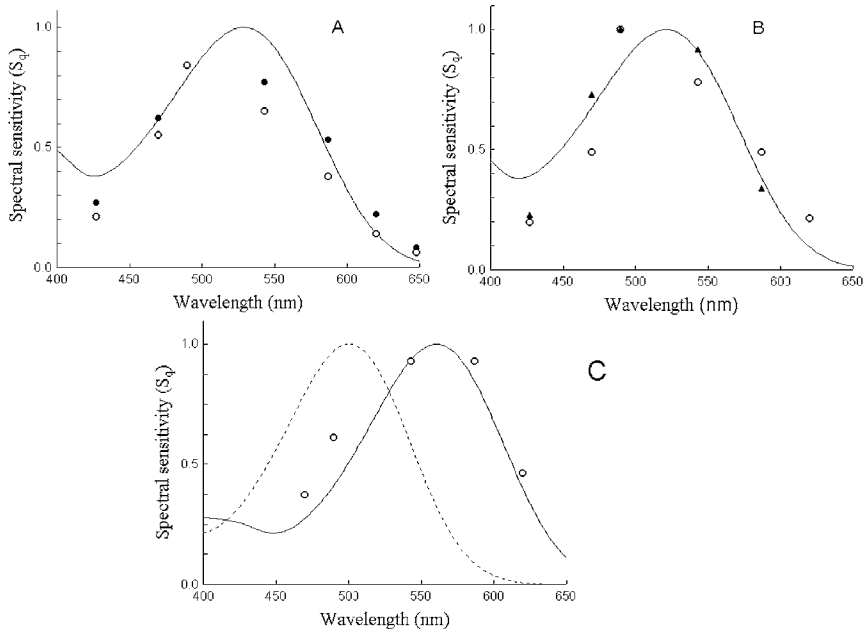

FIGURE 5. Action spectra of second-order neurons. (**A**) Comparison between responses of HCs under photopic (*open circles*, $N=6$) and scotopic conditions (*closed circles*, $N=4$); means ± SE; Seliger eels. (**B**) Comparison between HCs under photopic (*open circles*, $N=9$) and scotopic conditions (*closed circles*, $N=1$) Adriatic eels. Continuous curves in **A** and **B**: absorption spectrum of porphyropsin-523 (derived according to Maksimov[11]; see *Materials and Methods*). (**C**) Responses of an on-bipolar cell (Adriatic eel). Serial stimulation by light flashes of increasing wavelength under photopic conditions (NDF=–0.6). *Continuous and dotted curves*: porphyropsin (p550 nm) and rhodopsin, respectively.

DISCUSSION

Microspectrophotometric measurements of retinal photopigments show that in adult eels at least three classes of photoreceptor exist that contain mixtures of rhodopsins and porphyropsins. Middle-wave sensitivity cones probably have a mixture of 520_1 and 545_2 and short-wave-sensitive cones a mixture of 433_1 and 438_2 nm pigments. The rods of eels possess a "freshwater" opsin that is expressed as a pigment pair with λ_{max} at 501_1 and 523_2 nm[16] and a deep-sea opsin that is expressed as a pigment pair with λ_{max} at about 482_1 and 493_2 nm.[16,17]

Cone Inputs to Second-Order Neurons

Both photopic (cone) and scotopic (rod) inputs to horizontal cells have been identified in the present study of the European eel. Rod-HCs and cone-HCs differed greatly as to their respective saturation levels, in full agreement with earlier findings that in some fish species rod responses saturate at considerably lower stimulus inten-

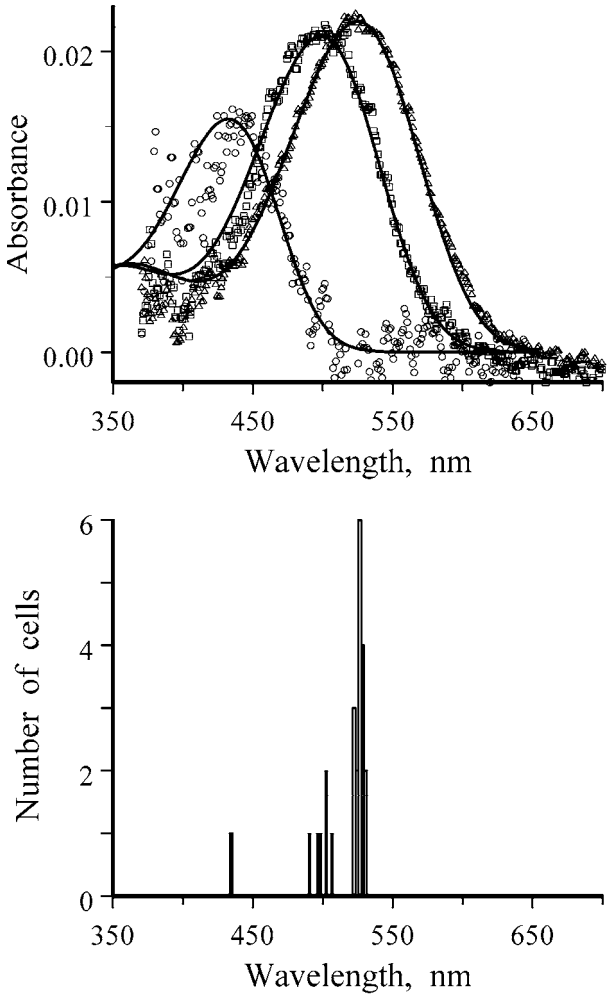

FIGURE 6. (*Top panel*) Mean absorbance spectra for the three classes of photoreceptor. Triangles, MWS cones; squares, rods; circles, SWS cone. The full curves are visual pigment templates[26] based on an 80%/20% mixture of A_1 and A_2 pigments with λ_{max} at 525, 498, and 434 nm. (*Lower panel*) Distribution of the λ_{max} of individual rods and cones. Bin size of 2 nm.

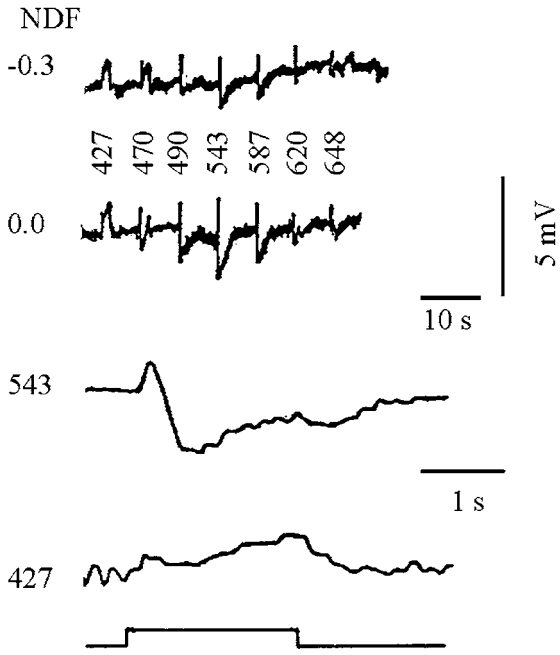

FIGURE 7. Color-opponency in an amacrine cell from a Seliger eel. (*Top*) Responses to serial stimulation by light flashes of increasing wavelength (427–648 nm), and of two stimulus intensities (NDF = –0.3 and 0.0); maximal response amplitude at 543 nm. (*Bottom*) Different type responses to different wavelength flashes (depolarization followed by hyperpolarization in response to a 543 nm light flash; depolarizing response to 427 nm).

sities than cone responses.[18] In rod-HCs of the eel, saturation was reached at relatively low intensities of monochromatic stimuli, while in cone-HCs there was no saturation even when white light flashes of maximal intensity were applied. Furthermore, in some of the mixed-HCs, saturation of the scotopic component occurred in the presence of background illumination of a relatively low intensity, and the rod component disappeared completely under photopic conditions.

The majority of cone driven L-horizontal cells had a maximum sensitivity around 520 nm, which strongly implies input from MWS cones dominated by the 520_1 nm pigment. However, 3 of the 55 cone-HCs were maximally sensitive at longer wavelengths around 545 nm. This probably indicates input from a small population of MWS cones still retaining the porphyropsin MWS cone pigment with λ_{max} at about 543 nm. However, the longer-wave input could suggest a long-wave-sensitive (LWS) class of cone, not identified by MSP, in addition to the MWS and SWS cones. still retaining the porphyropsin MWS cone pigment with λ_{max} at about 543 nm. Because of the random sampling used in MSP, a sparse of regionally distributed population of cones may be missed. It is also possible that LWS cone class is present in freshwater eels, but had already been lost in the maturing silver eel used in the present MSP study.

Rod-HCs, like the majority of cone-HCs, also showed maximum sensitivity around 520 nm and the difference between photopic and scotopic maxima was not statistically significant. This would imply that the rods contained primarily porphyropsin, $P523_2$ and this is supported by the observation that the scotopic spectral sensitivity curve appears somewhat broader than the photopic curve (the differences between scotopic and photopic data-points were not statistically significant, although for 7 of 8 wavelengths tested the average sensitivity values were higher under scotopic conditions). The conclusion that different classes of photoreceptor within the same retina can contain different ratios of rhodopsin and porphyropsin has been shown to occur in other teleost, notably in some deep-sea fish[19,20] and in the cavefish, *Astyanax fasciatus*.[21] The eels used in present study, both for electrophysiology and MSP, were from very different locations and their respective developmental stages in the metamorphosis between freshwater and marine forms were not determined so that levels of A_1 and A_2 pigments may have been very different.

Only three of the 55 horizontal cells presently explored were clearly more LWS. In this connection, it should be mentioned that in the Japanese eel (*Anguilla japonica*) the photopic L-response of a few horizontal cells exhibited a spectral maximum around 548 nm[22] similar to our records. However, the strongest indication that there are two spectrally distinct cone units in the European eel has been obtained in the present study from the five longer-wave-sensitive bipolar cells. Why the longer-wave of the two cone units are mainly found in bipolar cells, remains to be elucidated. Similar longer-wave responses with maximum sensitivity around 550 nm have also been identified by electroretinographic analyses in the congener American eel (*Anguilla rostrata*).[8]

One amacrine cell with color-coding properties has been identified in our experiments, presumably of the B/Y (or B/G) type. The presence of such cells is usually considered as pointing to the possibility of wavelength discrimination and color vision. The existence of green-sensitive and blue-sensitive cone units was thus revealed by both electrophysiological and MSP.

Eels from Lake Seliger differed from the considerably smaller yellow eels caught in Adriatic coastal waters by a number of features characterizing the structure of their retina.The rod/cone ratio was 40:1 in Seliger eels, as compared to the ratio of 20:1 in the yellow Adriatic eels, and the rod/INL ratio amounted to 6:1 as compared to the ratio of approximately 3:1 in the Adriatic eel retina. These differences were considerably less important than those known to exist between silver- and yellow-stage eels. In the silver stage, for instance, eyes enlarge dramatically,[12] new rods are added to the retina, and the rod/cone ratio increases to 100:1[13] and even 250:1,[14] becoming thus much higher than in yellow-stage eels (35:1 and 79:1, as reported by Braekvelt[13] and Pankhurst,[14] respectively).

Our electrophysiological and spectral sensitivity data also testify against large differences in the developmental stage between our two experimental groups of eels. The fitted scotopic spectral sensitivity maxima of individual Seliger eels, ranging from 524.9 to 533.1 nm (mean 530.2 ± 3.1nm), are distinctly different from the 500 nm maximum found in silver eels. Therefore, according to all the enumerated criteria our Seliger eels were closer to the yellow (immature) than to the silver stage, although according to the eye index[12] they should be classified as mature silver eels. The peculiarities of eels from Lake Seliger as compared to eels from the Adriatic region may simply be a consequence of the differences in local conditions for growth.

It was emphasized, for instance, in connection with the ichthyological features of Lake Seliger, that the so-called virgin lake provides conditions for an unusually rapid growth of introduced fish.[23] Conversely, it was found that contrary to expectations eels from the relatively warm southern regions, the Adriatic in particular, do not show an exceptionally rapid growth.[24]

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