Spectral and polarized light sensitivity of photoreceptors in the compound eye of the cricket (Gryllus bimaculatus)

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Summary. Retinula cells in the compound eye of the cricket (Gryllus bimaculatus) were recorded intracellularly and stained with Lucifer yellow. Two different methods were used to determine the spectral sensitivity of these cells: a) the spectral scanning method, and b) the conventional flash method. Three spectral types, with S(λ)-curves close to the rhodopsin-absorption functions, were found with $\lambda_{\text{max}}$ at 332 nm (UV), 445 nm (blue) and 515 nm (green), respectively.

Blue receptors were only recorded in the anatomically specialized dorsal rim area (DRA), and UV and green receptors in the dorsal region of the pigmented part of the eye, whereby green receptors were only found in the ventral eye. On the basis of these results, model calculations are presented for dichromatic and trichromatic colour vision in the cricket.

The fluorescence markings revealed green receptors whose axons project with short visual fibres to the lamina, and a UV receptor with a long visual fibre which projects through the lamina to the medulla. The blue receptors send their axons either to the lamina and medulla (long visual fibres) or only to the lamina (short visual fibres).

The temporal dynamics of the three receptor types were examined. The blue receptors lack a phasic component of the receptor potential, and the time from stimulus onset to peak potential is strongly increased compared to the UV and green receptors. Light adaptation reduces the latency to less than half of the dark adapted state.

Spectral adaptation experiments revealed an 'unidirectional coupling' between UV and green receptors, and it was found that polarization sensitivity (PS) in blue cells was much higher (PS = 6.5 ± 1.5) than that of UV (PS = 1.76 ± 0.05) and green (2.26 ± 0.57) receptors. The functional aspects of the three receptor types are discussed with respect to the presented physiological and morphological data.

Introduction

The cricket is a well studied model system for neuroethological research on acoustic communication, neural processing of sound perception, and sound production (Huber 1980). Visual orientation and neural processing in the visual system, however, has not attracted a great deal of attention until recently. For example, Burghause (1979) published a paper on the fine structure of the compound eye, and demonstrated that crickets use a specialized dorsal rim area (DRA) to orient to polarized light. These observations were confirmed and extended in a physiological study by Labhart et al. (1984) who found for Gryllus campestris in the DRA only blue receptors with a high polarization sensitivity and large visual fields. In the adjacent dorsal area (DA) of the eye only green and UV receptors were found, both with narrow visual fields. We have extended these studies with Gryllus bimaculatus with the aim of further characterizing the spectral receptor types physiologically and anatomically not only in the dorsal, but also in the ventral part of the eye. The new spectral scanning method (Menzel et al. 1986) allows the measurement of a more accurate spectral sensitivity function. Other major points of this work are the model calculation for potential dichromatic role
our vision, and the interpretations of coupling between receptor cells on the basis of spectral adaptation experiments.

Confirming Labhart et al. (1984) we find that the cricket eye is a good example of a design principle of many insect eyes, namely the regionalization of visual functions in the ommatidial matrix of the compound eye. The receptors of the dorsal rim area respond with a slow, highly sensitive receptor potential, are maximally sensitive to blue polarized light, and integrate light over large receptive fields. These characteristics may be the result of adaptations that optimize the role of the blue receptors as detectors of polarized skylight which also integrate stimuli of polarized light over an extended area and time in a narrow spectral range thus eliminating spatial, temporal, and spectral disturbances of the natural polarized light signal already at the receptor level. The dorsal to frontal eye region is potentially a di-variant colour coding system with UV and green receptor types, and the ventral eye is possibly a mono-variant green sensitive system. The reduced polarized light sensitivity in both UV and green receptors simplifies neural integration and this is in good agreement with similar observations from other insect eyes (Snyder et al. 1974; Labhart 1980; Meyer and Labhart 1981). Some of the results described here were part of the diploma thesis of one of us (Zufall 1984).

Material and methods

Preparation and recording. Conventional microelectrode techniques were used to record intracellularly from the compound eye of the adult cricket, Gryllus bimaculatus. A Kopf puller was used to pull microelectrodes from Hilgenberg glass capillaries containing filaments, and these microelectrodes were then filled with 2.5 M KCl (60–150 MΩ) or 3% Lucifer yellow and 0.1 M LiCl (150–400 MΩ) for the purpose of intracellular staining. Cells were dye injected by applying constant hyperpolarizing currents of 5–10 nA for 10–15 min, and the position of the electrode during this time was checked by light flashes. Current injection was immediately stopped if the depolarizing light response was no longer registered. The ground electrode was inserted into the thorax. To avoid movement, the test animal was waxed onto a holder after removing the legs, abdomen and antennae.

Stimulation. Two different sets of stimuli were applied: a) the spectral scanning method, and b) the conventional flash method. The spectral scanning method, which is described in detail in Menzel et al. (1986), allows for the measurement of spectral sensitivity with high spectral resolution (4 nm) in a short time (15 s). A grid monochromator (Schoeffel GM 252/1) was used to scan the spectrum from 300–700 nm (and vice versa) and the response of a cell was clamped to a preselected receptor potential (3–6 mV above resting potential) by continuously adjusting the quantal flux with a circular neutral density wedge (Melles Griot, 3 log units). A microprocessor was used to control both the feedback system (time course 20 ms/4 nm) and also to calculate on-line the spectral sensitivity. The set-up for the flash method was basically the same as that described by Menzel et al. (1986). Monochromatic light, at various intensities (3.8 log I units) and 13 different wavelengths (341–621 nm, Schott interference filters), was flashed to the eye (stimulus duration 300 ms, interstimulus interval 10 s), and the point light source (0.8° visual angle) was carefully centred on the axis to the recorded cell with the help of a perimeter. Polarization sensitivity was determined by the rotation of a polarizing filter in steps of 10° or 20°. For the adaptation experiments, a second monochromatic light beam (349, 441, 547, 591 nm), which could be independently controlled in intensity, duration and timing, was superimposed onto the first light beam.

Intensity, which was measured with a radiometer IL 700 and a PM 270 D detector (International Light Corp.), reached \(2.3 \times 10^{13}\) quanta/cm²·s with the 540 nm filter. The receptor potentials were stored on tape (Hewlett Packard 3964 A), and all data were evaluated from a paper trace by calculating the spectral sensitivities from the efficiency measurements and the corresponding intensity characteristics (V/log I functions). Only those recordings with a resting potential stable for more than 30 min and with receptor responses of \(I_{max} > 30\) mV were included in the analysis.

Histological procedures. After injection with Lucifer yellow, the brains and attached optic lobes were fixed for 2 h in 5% paraformaldehyde in phosphate buffer (freshly prepared) at room temperature. The tissue was then dehydrated with ethanol and cleared in methylsalicylate, before being photographed under epi-fluorescence on Kodak Ektachrome 400 ASA. In several cases frontal or horizontal sections were made with a vibratome (50 μm thick) after inspection and photographing of the whole mount. The cleared whole mounts were rehydrated in ethanol (50%), embedded in agar (2%) and sectioned at 4°C. We verified for all cases the pattern of stained axons seen in whole mounts. This is due to the very low autofluorescence in our preparations.

Results

Spectral types

The recordings from over 150 receptor cells show that three different spectral types exist: UV (\(\lambda_{max} = 332\) nm), blue (\(\lambda_{max} = 445\) nm) and green (\(\lambda_{max} = 515\) nm). These results confirm those of Labhart et al. (1984) who measured the spectral sensitivities of photoreceptors in the dorsal rim area (DRA) and the adjacent dorsal area (DA) of the cricket eye. Figure 1A–C gives the average spectral sensitivities of these three receptor types as determined by the spectral scanning method (no data were excluded from the averaging). The three curves are very close to the rhodopsin absorption functions as calculated by the visual pigment nomogram of Ebrey and Honig (1977) or the spectral absorbance of the UV-photopigment in the Ascaphalus eye (Hamdorf and Gogala 1973). For example, the S(λ)-function of the green receptor has a secondary maximum at 350 nm, which on average reaches 35% of the value at 515 nm, and this may be a
Fig. 1. Average spectral sensitivity of UV (A) (11 cells, 53 spectral runs averaged), blue (B) (6 cells, 26 runs) and green receptors (C) (17 cells, 48 runs) as obtained by the spectral scanning method. The S(λ)-function of the green receptor is close to a rhodopsin resonance function ($\lambda_{\text{max}} = 515$ nm) with a slightly enhanced β-peak at 350 nm. The UV and blue cells closely follow a rhodopsin resonance function with peak absorption at 332 nm and 445 nm, respectively. D–F Camera lucida drawings from Lucifer yellow stainings of the corresponding three spectral receptor types as viewed in whole mounts from the frontal direction. CBL cell body layer, EPL external plexiform layer, ChI first optic chiasma, LA lamina, ME medulla, R retinula cell axon. D One marked UV cell with a long visual fibre projecting to the medulla and no apparent structure in the lamina. E Four examples of blue receptors – the three on the right side have short visual fibres projecting to the proximal layer of the lamina, while the receptor on the left side has a long visual fibre and obvious branches in the lamina. F Five examples of marked green receptors that terminate with thick branches in the proximal and medial lamina. G Drawings from Golgi preparations of visual cell axons (R1–R8). (Fig. 1 G – Courtesy of Dr. L. Williams, unpublished results)
consequence of the $\beta$-absorption of the respective rhodopsin pigment. The blue receptor shows an elevated sensitivity at about the same wavelength region and, once again, this may be indicative of $\beta$-absorption. For the UV receptor, the S(\lambda) resembles the absorption function of the photopigment in the UV receptors of the insect Ascalaphus (Gogala 1967; Hamdorf and Gogala 1973). Labhart et al. (1984) describe average S(\lambda)-functions calculated from preselected data with the assumption that high spectral side band sensitivities are indicative of faulty measurements, and this assumption is essentially valid since the fast spectral scanning method is not as sensitive to recording artifacts as the conventional flash method (see Menzel et al. 1986 for further discussion). Indeed, the S(\lambda)-functions are very close to the expected absorbance functions of the respective rhodopsin.

It should be noted that the spectral scanning method works with responses to continuous dim spectral light, and thus measures receptor sensitivity in the tonic response component below 10 mV. Therefore, when S(\lambda)-curves from the spectral scanning method are compared with those of the light flash method, one should calculate the sensitivity functions using the plateau potential rather than the peak potential. Indeed, the values of $\lambda_{\text{max}}$ of the three receptor types are the same if the plateau response to flashes of spectral light is used. However, if S(\lambda) is calculated using the peak-potential, then the UV receptor shows a prominent secondary maximum of about 30% sensitivity near 500 nm, and this is not apparent in the plateau-potential calculations. Furthermore, the green receptor shows a secondary peak of 40% (n = 39) in the UV, if sensitivity is calculated using the peak-potential. This is only a slightly elevated value compared to that obtained with the spectral scanning method. The S(\lambda)-function of the blue receptor type is identical for both methods, which is not surprising, since these cells respond only with a tonic receptor potential (see below).

The three spectral receptor types are not equally distributed over the compound eye of the cricket as was found already by Jud and Labhart (1985) for the upper part of the eye of G. campestris. Figure 2 gives the locations of most of the recorded cells in the eye, as obtained by the position of the perimeter and relative to the head axes (dorsal-ventral, anterior-posterior). Blue receptors were only found in the DRA, and this part of the eye is visually conspicuous because it does not contain a screening pigment and has flat lenses (Burghause 1979). No other spectral receptor type was found in the DRA. The UV receptors were recorded predominantly in the adjacent dorsal region (DA) within the pigmented part of the eye, whereas the green receptors were found over the whole eye excluding the DRA.

The purpose of the Lucifer yellow stainings was to investigate possible correspondence between the physiological and morphological receptor types, and representative examples of these stainings are shown in Fig. 1D–F. Only one UV receptor was stained (Fig. 1D), and this cell has a long visual fibre (l.v.f.) which is typical for UV receptors in insects. The l.v.f. passes through the lamina to the medulla and terminates in a forked branching in the median part of the medulla. Blue receptors were stained most frequently (Fig. 1E). Although most cells terminate in the lamina (three are shown in Fig. 1E), some (e.g. Z111) terminate in the medulla and are characterized by a great number of branchings and spines in the external plexiform layer of the lamina. Z112 (two axons stained) and Z113 show a complicated pattern of branching in the lamina, whereas Z104 looks very similar to the green receptors. In spite of our search for physiological features which may explain the great morphological diversity of the blue receptor, we were unable to find any correlation with the particular S(\lambda)-function, the location within the DRA or the sensitivity to polarized light. For example, Z11 with a long visual fibre has a PS of 5.9 which is
in the middle range for the blue receptors (see below). However, it is possible that a correlation with preferred e-vector direction might occur, a question which we have not examined further.

All of the five stained green receptors (Fig. 1F) terminate in the intermediate plexus of the lamina, and have single (Z103) or forked terminals (Z92, Z91, Z89) with numerous protrusions or spines. The cell Z89 was stained immediately after recording of another green receptor within the same ommatidium. This double marking may either indicate a dye coupling between green receptors or a leakage of the dye during the initial recording. Labhart et al. (1984) speculate about the possibility of electrical coupling which, in effect, would reduce polarized light sensitivity in the green receptors. Dye coupling would support the possibility of coupling between green receptors that have different orientations of their microvilli, but conclusive experimental evidence is still lacking. Z110 is a peculiar marking, because the 3 thick axon terminals appear to project to 3 different lamina cartridges. Neighbouring cartridges are also reached by axon terminals from other green receptors (see Z91, Z92), and whilst this is also the case for several blue receptors, this was not observed for the marked UV cell.

Our findings do not completely coincide with those of Jud and Labhart (1985) for the upper part of the G. campestris eye. They found all stained green receptors to project to the lamina, and all UV- and blue receptors to terminate in the medulla, whereas in our case blue receptors project to the medulla or to the lamina. It is yet unknown whether this discrepancy reflects another aspect of eye regionalisation or a species difference (see Discussion).

L. Williams kindly provided us with unpublished results from Golgi stainings of the visual system of G. bimaculatus (Fig. 1G). Two of the 8 retinula cells in each ommatidium project through the lamina to the medulla (denoted with R7, 8), whereas 6 terminate either in the outer (R3, 4, 5, 6) or middle (R1, 2) plexiform layer. The numbering of the retinula axons R1–R8 cannot be related to the structure of the respective somata within the retina, because neither the Lucifer markings nor the Golgi-stainings of L. Williams have been examined in the retina. Other previous studies have also failed to close this gap in our knowledge. The markings Z103 and Z89 resemble those of R1 and R2, respectively, but the other Golgi-stained cells look very different from our markings. It is particularly obvious that the fine branches which are clearly visible in the Lucifer filled cells do not appear in the Golgi stains, although the thick terminals of R1 and R2 do not differ very much between intracellular stained cells and Golgi preparations. We are inclined to conclude, therefore, that retinula cell types R3, 4, 5, 6 are not completely stained by the Golgi technique. Another possibility is that the structure of the retinula axons depends on the region of the eye, and that L. Williams samples may come from a different eye region than ours.

Our current knowledge of the spectral input functions of photoreceptors permits reasonable assumptions to be made with respect to a potential colour coding system and the resulting colour vision of the animal. The results presented here, and those of Labhart et al. (1984), suggest that the cricket has a triparted eye: a mono-variant DRA (only blue receptors, specialized for polar sensitivity), a di-variant (UV, green) upper region of the eye (DA to about median frontal eye regions) and possibly a mono-variant (green) median to ventral eye region. In every case, it would appear that crickets possess the potential of at least a dichromatic colour vision system in the upper half of their eyes (excluding the DRA). Since it cannot be ruled out that some blue receptors at the border between the DRA and the dorso-frontal eye may contribute to a colour coding system, we have considered both a dichromatic and trichromatic colour vision system, and the respective chromatic diagrams are presented in Fig. 3.

The calculation of the trichromatic diagram (Fig. 3a) is based on the assumption that the S(λ) functions of the 3 spectral receptor types (Fig. 1) represent the primary colours for a tri-stimulus space (Cornsweet 1970; Rushton 1972). The triangle is the area which cuts through this space at a unity distance from the origin, and the vector of any chromatic stimulus is thus condensed to a point (‘colour locus’). The colour mixture rules apply to such a chromatic diagram. The dichromatic case (Fig. 3b) is calculated in the same way by simply setting the contribution of the blue receptor to zero. Theoretical spectral discrimination functions (Fig. 3c) for both cases are calculated according to Backhaus and Menzel (1987). The noise properties of the photoreceptors are taken as the unit length in the tri- or di-stimulus space and interpreted as the perceptual line elements for spectral discrimination (just noticeable difference steps: jnd’s). For a first approximation, the noise properties of the receptors were assumed to be equal in all 3 receptor types and similar to those measured in the honey bee’s photoreceptors (Backhaus and Menzel 1987). These assumptions are
considered to be justified in a first approximation, but require more careful attention if the predictions of the model calculations are to be compared with behavioural results for future experiments. According to the model, spectral lights should be discriminated on the basis of hue differences, in the range of approximately 370 to 450 nm (with a neutral point in between) for the dichromatic case, and approximately 360 to 550 nm for the trichromatic case. Therefore, one would expect spectral discrimination to be optimal in the violet region and somewhat enhanced at around 510 nm for trichromatic colour vision. The small additional peaks of the model calculation that are observed for both cases at around 310–320 nm (Fig. 3c) are a consequence of the abrupt cut-off of all 3 receptors at very short wavelengths and are likely to be below threshold.

Temporal dynamics of the spectral receptor types

The dynamic properties of the three spectral receptor types were further analysed using the flash method. The phasic-tonic time course typical for fast insect photoreceptors has already been described by Lambin and Jeanrot (1982) for the green receptor of the cricket. The V/log I-function is independent of the wavelength of the stimulating light. According to the modified self-shunting formula (Lipetz 1971), an exponent \( n \) has been calculated, which is 0.65 ± 0.07 (from 40 cells). On average, around \( 7 \times 10^{10} \) quanta/cm²/s are needed (from 40 cells) to reach 50% of the peak response. The tonic response saturates at 65% of the amplitude of the phasic response independent of the wavelength of the stimulating light.

The relationship between the phasic and tonic response components is different for both the UV and blue receptors. Figure 4A shows the V/log I-function for an UV receptor and Fig. 4B gives examples of corresponding recordings. UV receptors display a very high proportion of tonic response, and this exceeds 90% of the phasic response if they are stimulated with UV light. However, if these cells are stimulated with blue or green light the tonic response saturates at much smaller response values (about 60%) even though the phasic response reaches the same high potentials as with UV light. For this reason, the S(\( \lambda \))-function of the UV receptor shows a secondary maximum near 500 nm if it is calculated from the peak-potential. The value \( n \) for the UV receptors is 0.72 ± 0.04 (from 8 cells), irrespective of the wavelength of the stimulating light, and this is not significantly different from that observed for the green receptors.

An example of a blue receptor is shown in Fig. 4C, D. These cells completely lack a phasic component and respond only with a tonic potential. This was verified not only for the stimulus duration of 300 ms but also for a stimulus of 1 s. The V/log I-function shows that the same high response values are reached as with the peak potentials of the other receptors. The number of quanta needed to obtain 50% of the maximum response
is roughly 10 times higher compared to the green and UV cells (see above and Fig. 4). This agrees with findings by Laughlin (1976). The time course of the response is slowed down for both light increments and decrements. Small voltage fluctuations, which are apparent in the UV receptors at low intensities (Fig. 4B), are very small or absent in the blue receptors. For Gryllus campestris, Labhart et al. (1984) also showed a slower response of the blue-cells compared to green-cells. Both, latency and duration of the on-transient are extended.

Spectral adaptation

As mentioned above, the UV receptors exhibit a prominent secondary maximum near 500 nm if $S(\lambda)$ is calculated from the peak-potential. In addition, the green receptors have a secondary maximum in the UV range. We performed spectral adaptation experiments with both cell types in order to test whether the respective secondary peaks can be attributed to positive electrical interactions between the two spectral cell types, or whether intrinsic properties of each cell type, such as mixtures of photopigments, account for these effects. One possibility, the occurrence of a UV absorbing antennal pigment in green receptors as e.g. in fly R1–6 receptor cells (Kirschfeld et al. 1977; Hardie 1986) is unlikely, since the spectrally high resolving scanning method did not reveal any fine structure in the UV. This fine structure would have been indicative of an antennal pigment such as 3-hydroxy-retinol. If an antennal pigment would be retinol as in the case of the simulid fly (Kirschfeld and Vogt 1986), we would have not detected it by our spectral measurements. However, the
transfer of energy from an antennal pigment should not be restricted to the initial period of light absorption during the peak potential.

The procedure for the adaptation experiment is described in Fig. 5A for a UV receptor. After centring the point light source to the optical axis of the cell, and with a test light of 360 nm to monitor the intensity response function, a spectral run is performed with an ordered succession of wave-lengths (341–570 nm). When the adaptation light (547 nm) is switched on, the cell depolarizes to approximately a third of its maximal voltage range (12 mV), and during this steady adaptation a second spectral run is performed with the same succession of wavelengths as described above. After a re-dark adaptation of 5 min, the experiment is repeated. The result is given in Fig. 5B and C. Since the shortest wavelength was 341 nm the UV receptor was most sensitive to this wavelength (compare with Fig. 1A). If a UV receptor, which possesses a secondary maximum near 500 nm of 47% (as calculated from the peak-potential), is green adapted then the secondary maximum is totally eliminated (Fig. 5B) and, furthermore, the S(λ) becomes increasingly narrow at its long-wavelength cut-off. The selective adaptation effect is particularly obvious for the peak potential (compare Fig. 5B and C), and this observation supports the concept that the UV receptors are electrically coupled to green receptors through a high frequency pass filter. These effects are fully reversible (see Fig. 5A) and can be repeated several times. The green receptors behave quite differently in adaptation experiments. For example, if a green receptor is UV adapted (15 mV depolarization) then there is no change in S(λ) (not shown). However, the UV adaptation experiments were carried out with green receptors that have a moderate sensitivity in the UV, and this in fact is typical for most green receptors.

The selective adaptation effect of the UV receptors suggests that the secondary sensitivity of UV receptors in the green results from a unidirectional positive electrical coupling of UV to green receptors. In other words, the UV sensitivity of green receptors does not originate in the UV receptors.

In a further series of experiments, the effect of chromatic adaptation of the blue receptor was examined and compared with similar experiments on green receptors. Both green and blue receptors were green adapted (547 nm, 15 mV depolarization), and the time between on-set of the stimuli and maximal response was measured for a response height of the half maximal response (time-to-peak in Fig. 6). For the green receptor, the time-to-peak was 28 ms, and thus no difference between dark and green adaptation was obvious. For the blue receptor, however, the very long time of 245 ms during dark adaptation (dark column) was significantly reduced to 110 ms by adaptation (light column). The results indicate that green adaptation has a strong effect on the kinetics of the receptor potential of the blue receptor, but that the green receptors are not effected (see Fig. 6).
tor is equipped with just one photopigment ($\lambda_{\text{max}} = 445 \text{ nm}$) and there is no indication of interaction with the green or UV photopigments. (2) the blue receptors occur only in the dorsal rim region, and no other spectral receptor types have been found in this area.

**Polarization sensitivity**

The blue receptors show a high PS of $6.5 \pm 1.5$ ($n = 7$), and this confirms the measurements of Labhart et al. (1984). For the UV and green receptor, two different wavelengths were tested in order to examine the wavelength-dependence of the PS. The PS for the UV receptor is $1.76 \pm 0.05$ ($n = 3$) at 363 nm and $2.26 \pm 0.57$ ($n = 4$) at 540 nm. There is no apparent wavelength-dependent phase shift of the minima and maxima of the PS. Labhart et al. (1984) found PS = 2.9 and 4.2 for the two UV cells they recorded.

For the green receptors, the average PS is $2.4 \pm 1$ ($n = 26$), but varied between 1.2 and 4.9 (Fig. 7), a result which confirms Labhart et al. (1984) for *G. campestris* (PS = 2.6 ± 0.8, $n = 14$). Once again, no significant wavelength-dependency was found, both with respect to the absolute values of PS and the phase. The inset of Fig. 7 gives an example for such a recording of a green receptor with (a) low and (b) high PS. Although we searched for a correlation between the secondary UV sensitivity of green receptors and the corresponding PS, we were unable to find a strong effect (Fig. 7). However, it is noteworthy that the high PS values are found only in green receptors with low $(\leq 40\%)$ sensitivity in the UV. These recordings may, therefore, come from cells which have

![Fig. 6. Comparison of the temporal dynamics of typical green and blue receptors during dark adaptation (dark columns) and after spectral adaptation with green light (547 nm) (light columns). In both cases, the tonic depolarization by adaptation was 15 mV. The time-to-peak (ordinate) is the time between onset of the stimulus and the maximal response reached by a light stimulus lasting for 300 ms of an intensity which results in half the optimal response height. In other words, time-to-peak is measured at half the maximal response of the receptor potential.

Unfortunately, this experiment was not repeated with different wavelengths of the adapting light. Therefore, one cannot postulate whether the shortening of the time course is due to a simple light adaptation effect or whether it is controlled by a wavelength dependent component. The latter is quite unlikely for two reasons: (1) the blue receptor is equipped with just one photopigment ($\lambda_{\text{max}} = 445 \text{ nm}$) and there is no indication of interaction with the green or UV photopigments. (2) the blue receptors occur only in the dorsal rim region, and no other spectral receptor types have been found in this area.

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![Fig. 7. Correlation between secondary UV sensitivity of green receptors (in %) and the corresponding PS. The inset gives an example of a green receptor with low (a) and high (b) PS. In both cases the wavelength dependence of the PS was tested with two different wavelengths. No differences in absolute value of the PS or in phase shift of the minima and maxima can be seen. The corresponding V/log I-function proceeds the PS-measurement.](image)
no electrical coupling to other receptors, irrespective of whether strong electrical coupling is physiological or artefactual. Thus, it appears possible that the cricket eye contains green receptors with considerably high PS, but no UV receptors of a comparably high PS. These green receptors are unlikely to look skywards because they are pre-dominantly positioned in the region of the eye ventral to the DRA (4 out of 5 cells), and it should thus be examined whether the cricket detects polarized light patterns from reflecting surfaces.

Discussion

Photoreceptors are signal transducers and filters for certain stimulus combinations (quantal flux, spectral range, e-vector orientation, spatial and temporal window). A necessary step in the neural integration of visual stimuli is the extraction of the relevant information for certain orientational subsystems, and retinal specialization is an important design principle in all eyes that optimizes and facilitates the extraction of such information (Snyder 1979; Snyder et al. 1977; Srinivasan et al. 1982). Indeed, regional specialization may be extreme, as is the case in the cricket eye where the monochromatic DRA extracts pol information from the sky, and where adjacent eye regions show a succession from (potentially) trivariant (UV, blue, green), to divariant (UV, green) to the monovariant ventral eye. Furthermore, high polarized light sensitivity (PS), large receptive fields, slow response times, a narrow spectral window in the blue and full occupation of a specialized part of the eye with one receptor type reduces the neural operations in the DRA necessary for the extraction of just one visual parameter, namely the e-vector information. Thus, small disturbances of the polarization pattern in space and time are already filtered out at the receptor level.

The rhabdoms in the DRA are approximately twice as long as those of the central eye with a circa 50% larger cross-section (Burghauser 1979; Hoff 1985), and almost double their volume at night in a circadian rhythm of membrane turnover. Also, ommatidia in the DRA lack screening pigments (Burghauser 1979) and the rhabdom does not change its architecture due to illumination, as is the case with the central rhabdoms (Hoff 1985). The sensitivity of the blue receptors can be estimated to be a factor of up to 60 times higher than the green receptors (compare Labhart et al. 1984: 5-20 times) if one takes into account that the visual field volume is up to 600 times larger (Zufall 1984), and the blue receptors need roughly 10 times more light to obtain the same response height. The existence of other mechanisms that enhance the absolute sensitivity of these polarized light receptors is also likely. The light response is very slow and has a long latency (Fig. 6. 240 ms, dark adapted; 100 ms, light adapted); voltage fluctuations at low light conditions are absent (Fig. 4D) possibly indicating an electrical coupling between the receptors, and the axons branch extensively with the consequence that several lamina and medulla cartridges are reached by one receptor cell. Particular patterns of electrical coupling between the receptors and synaptic connectivities to the second order neurons in the lamina and medulla would be quite feasible, especially since the PS is not destroyed in the receptors and is transmitted to higher order visual neurons (Labhart 1988). It would appear, therefore, that the DRA is designed to extract e-vector information under low light conditions e.g. dawn, dusk, night, and this is particularly relevant when one considers that crickets are active at low light and even fly at night. This would suggest that crickets use their ability to orient to the sky’s polarization pattern not only during walking (Burghauser 1979; Brunner and Labhart 1987), but also when flying (for G. campestris see Rost and Honegger 1987).

A specialized DRA exclusively equipped with blue receptors has also been demonstrated in Acheta domestica (Zufall, unpublished), contrary to the paper by Gribakin et al. (1980). Since Acheta is also active at night, the combination of blue sensitivity and pol-sensitivity in a specialized eye region may be a common adaptation of insects that are active at very low light intensities, as opposed to day active insects which predominantly use UV receptors as pol-detectors, e.g., the honey bee (von Helversen and Edrich 1974).

The S(λ) of the 3 receptor types, as determined by the highly resolving and fast spectral scan method, indicate rhodopsin pigments without any additional antenval or screening pigments. Deviations from the typical rhodopsin functions may partially be interpreted as artefacts (see discussion in Labhart et al. 1984), and partially as effects of functional electrical coupling. In addition, the spectral adaptation experiments reported here demonstrate that the increased sensitivity of UV receptors in the green originates in the green receptors, and is transferred to the UV receptors via unidirectional electrical coupling, whereas the UV sensitivity of the green receptors is due to their β-absorption. The unidirectional electrical coupling attenuates low frequencies more strongly than high frequencies, because only the transient potentials of the
green receptors are transferred to the UV receptors. These conditions rule out the possibility of an artefactual coupling along the shaft of the recording electrode.

It is still unknown whether the properties of inter-receptor coupling can be dynamically adjusted under natural conditions, as our adaptation experiment (Fig. 5) would suggest. If this is in fact the case, then the UV receptor would become uncoupled at higher light intensities and this would improve the spectral information. Similar mechanisms have already been suggested for other insect species (Menzel and Blakers 1976; Tsukahara and Horridge 1977), but conclusive experimental proof is lacking.

Likewise, the predictions of our model calculations for a dichromatic or trichromatic colour vision system (Fig. 3) still require further experimentation. For example, orthopteran insects have rarely been studied with respect to wavelength selective behaviours, spectral preferences, colour vision or colour learning. Wavelength discrimination has been claimed for the grasshopper Phalaenop spec. (Wasserman and Kong 1982), but the data do not support any other conclusion than intensity dependent phototactic responses. Nymphs of the grasshopper Melanoplus sanguinipes have been successfully trained to coloured (green, yellow) cardboards with food reward, but intensity effects of colour discrimination were not ruled out (Bennays and Wrubel 1985). In Gryllus, wavelength discrimination should be optimal around 390-420 nm for both possible cases of colour vision (di- and trichromatic), although a trichromatic colour vision would be favoured if discrimination above 460 nm was recorded. Furthermore, our model calculations predict a better discrimination of violet than bluish-green colours. If the UV receptor loses electrical coupling to the green receptor in the light adapted state, then wavelength discrimination should improve with increasing light conditions.

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References

Hoff R (1987) Diurnal ultrastructural changes in the compound eye of Gryllus bimaculatus (Orthoptera, Gryllidae) in particular the dorsal rim area. Zool Jb Physiol 75:87-102
Menzel R, Ventura DF, Herrel H, Souza JM de, Greggers U
Snyder AW (1979) The physics of vision in compound eyes.

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