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Diurnal and Circadian Rhythm in Compound Eye of Cricket (*Gryllus bimaculatus*): Changes in Structure and Photon Capture Efficiency

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ABSTRACT—Day-night changes in rhabdom size of compound eyes were investigated in three groups of crickets (*Gryllus bimaculatus*): nymphs and adult males and females. In both adults and nymphs, the rhabdoms were larger at night than during a day. In adults, the mean rhabdom occupation ratios (RORs) of ommatidial retinulae at midnight were about two times greater than the values at midday. This change contributes to control of the photon capture efficiency (PCE) of the eye according to photic environment. The RORs of adult males at midnight were higher than those of both adult females and nymphs. This suggests that the PCE of the compound eye of adult males is the greatest of all groups. Under constant darkness, day-night changes in ROR were detected only in adult males, but neither in adult females nor in nymphs. On the other hand, no day-night changes were detected in any experimental group under constant light. These results suggest that the change in rhabdom size between day and night is an adaptation to the photic environment that is controlled mainly by the light-dark (day-night) cycle. However, the change in male adults is induced by an endogenous circadian clock.

Key words: retinular cell, rhabdom, circadian rhythm, compound eye, cricket

INTRODUCTION

Several comparative studies have reported differences in the structure and function of the compound eyes of diurnal and nocturnal arthropods (Land, 1981; Eguchi, 1982; Laughlin and Weckström, 1993; Land *et al.*, 1999). As an example of one of structural differences, the rhabdoms of nocturnal lepidopterans such as moths are much bigger than those of diurnal butterflies, presumably resulting in greater sensitivity to dim light at night (Eguchi, 1982). In fact, it has been demonstrated that the compound eyes of nocturnal species are much more sensitive to dim light than those of diurnal species (Eguchi and Horikoshi, 1984).

Circadian changes in the sensitivity of compound eyes were reported in several species, including adult crickets (Tomioka and Chiba, 1982b), cockroaches (Wills *et al.*, 1986),

sea slaters (Hariyama *et al.*, 1986, 2001), grapsid crabs (Arikawa *et al.*, 1987) and horseshoe crabs (Barlow *et al.*, 1987). A rhabdom containing visual substance is the initial site of visual excitation in retinular cell in the compound eye; it is characterized by tightly packed tubular membranes called microvilli. The increase in photon capture efficiency (PCE), primarily depends on morphological factors such as larger rhabdom size, development of palisades surrounding rhabdoms in the photoreceptor cell cytoplasm (for light reflection to rhabdoms), and migration of screening and/or reflecting pigment granules in compound eyes according to light and dark adaptation (Horridge and Barnard, 1965; Williams, 1982; Arikawa *et al.*, 1987; review by Meyer-Rochow, 1999). In particular, it can be said that the changes in rhabdom sizes influence PCE much greater than the effects of palisades and pigment granules in compound eyes. Because, a rhabdom, in general, has a property of wave-guide of light, namely the light entered in a rhabdom propagates inside the rhabdom by repeating internal reflection in the rhabdom, which means small quantity of light may leak from a rhabdom (for review see Stavenga, 1979).

The cricket *Gryllus bimaculatus* is diurnally active in the nymphal stage, however, in the adult stage it becomes nocturnally active. Tomioka and Chiba (1982a) reported that a

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Abbreviations: DRA, dorsal rim area; LB, lamellated body; PCE, photon capture efficiency; ROR, rhabdom occupation ratio

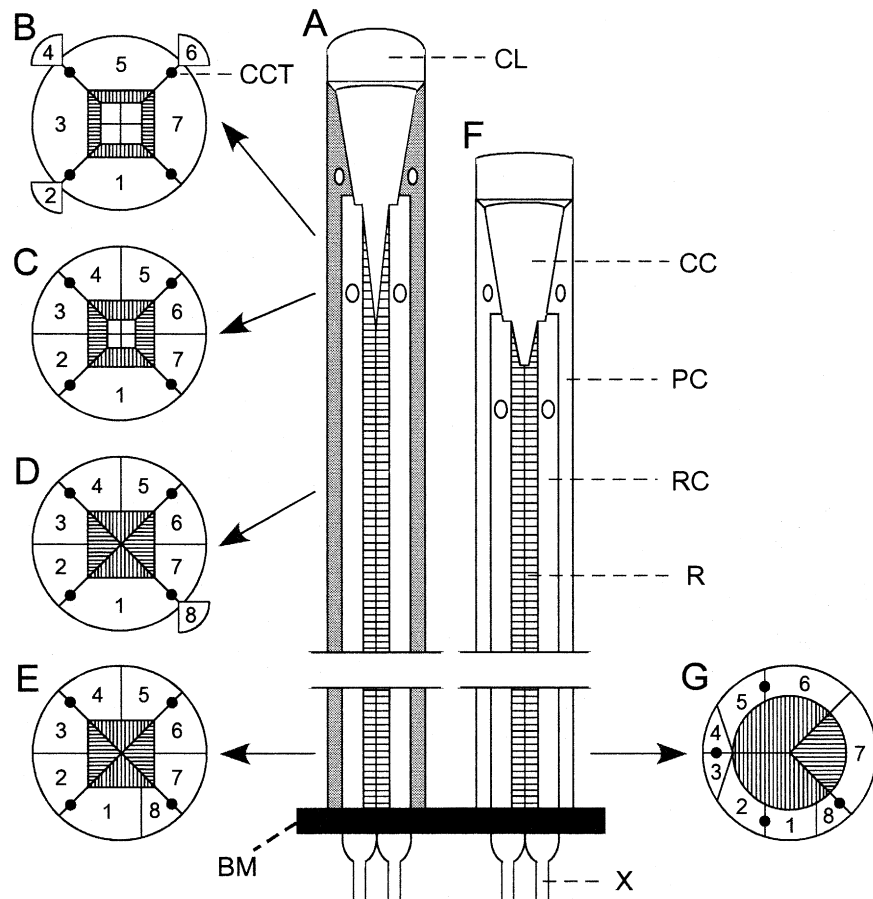


Fig. 1. Diagrammatic illustration of an ommatidium of a cricket eye in longitudinal (A, F) and transverse sections at different levels (B–E, G) with reticular cells numbered 1–8. (A–E) Ommatidia in the central part of a compound eye. (F–G) Ommatidia in the dorsal rim area (DRA) of a compound eye. CL, corneal lens; CC, crystalline cone; PC, pigment cell; RC, reticular cell; R, rhabdom; BM, basement membrane; X, axon. Filled black circles indicate the four crystalline cone tracts (CCT).

reversal of circadian rhythm occurs on 4 to 5 days after imaginal molt. It would be useful to know how the structure and function of compound eyes change before and after this reversal of circadian rhythm from diurnal nymph to nocturnal adult. First, the compound eyes of both the nymphal and adult stages are of apposition-type (Fig. 1), i.e., generally adapted to photopic vision despite a nocturnal life-style in the adult stage, as previously reported by Nilsson *et al.* (1987). Second, there are specialized ommatidia in the dorsal rim area (DRA) of the cricket eye (Fig. 1F, G). Functionally, the DRA is characterized by extremely high sensitivity to polarized light (Labhart *et al.*, 1984). Morphologically, the DRA is clearly distinguished from the other main region by its characteristic structures, i.e., its flat corneal surface, (Ukhanov *et al.*, 1996), larger rhabdoms, and absence of pigment granules in the cytoplasm of its photoreceptor cells (Burghause, 1979).

Hoff (1985) has reported that the cross-sectional areas of the rhabdoms of the compound eye increased from day to night by almost two-fold in the adult *Gryllus bimaculatus*, and therefore the capacity of photon absorption of the eyes also increased significantly at night. She concluded that this change in rhabdom size is suited to the cricket life-style, i.e.,

it lives in a wide range of light intensities.

In this study, we sought to clarify the changes in the structure of the compound eye between day and night in relation to the rhythm-reversal from nymph to adult. We focused on the rhabdom occupation ratios (RORs) of ommatidia as one of the major factors influencing eye PCE. RORs were measured separately in the DRA and in the central area of the compound eye, in both the nymphal and adult stages. We also compared PCE of compound eyes between day and night estimated by a theoretical equation based on morphological factors of the compound eyes (Land, 1981).

MATERIALS AND METHODS

Animals and light/dark regimes

Nymphs (in the last instar) and adult crickets (*Gryllus bimaculatus*) were used in the present study. They were obtained from a laboratory colony maintained under a 12:12 light/dark regime (light: 04:00–16:00, local time) at 27°C. Daylight fluorescent lamps were used for the light source, and light intensity was about 1000 lux. We put some tips of crumpled white paper in the cage as shelters for the crickets. The light intensity under these paper shelters was about 500 lux. The compound eyes were cut at midday (at 10:00)

or at midnight (at 22:00). In addition, to clarify whether the structural changes were induced by an intrinsic biological circadian rhythm or not, some animals were moved into a constant dark or light box, and the eyes were fixed at their subjective midday (at 10:00) or subjective midnight (at 22:00) on second day in their constant regimes. For the experiments at midnight and under constant darkness, dissections were performed under a dim red light to prevent light adaptation of the eyes.

Histology

Compound eyes were prefixed in 2% glutaraldehyde and 2% paraformaldehyde in 0.1 M cacodylate buffer at pH 7.3 for 12–15 h at 4°C. For the prefixation of the eyes at midnight and under constant darkness, vials containing the eyes were placed in a dark room. After washing with the same buffer, the tissues were post-fixed for 2–3 h in 2.0% OsO₄ in the same buffer, dehydrated in an acetone series and then embedded in Epon. For light microscopy, semi-thin sections were cut with glass knives, stained with toluidine blue, and examined with a light microscope (Olympus BH-2). For electron microscopy, ultra-thin sections were cut with a diamond knife, double-stained with uranyl acetate and lead citrate for 5 min each, and then examined under a JEM 1200EX electron microscope.

Measurement of rhabdom occupation ratio (ROR) of an ommatidium

In order to examine the day-night change in rhabdom size, RORs were measured in light micrographs. Pictures of cross-sections containing at least 20 hexagonally arranged ommatidia were examined from the central region and from the DRA, at the level of the nucleus of a retinular cell. Rhabdom area and inter-ommatidial distance (center-to-center) were measured by an image analyzing system connected to a computer. Generally, shapes and sizes of retinular and pigment cells are not constant among ommatidia. Thus, in order to reduce measurement errors, the mean value of all inter-ommatidial distances for an eye was taken as the mean ommatidial diameter at the level of the nuclei of retinular cells for calculation of the ROR. A Mann-Whitney *U*-test was used for statistical analysis of the RORs between day and night in the same developmental stage and sex of crickets. In addition, a Kruskal-Wallis test and a Scheffe's test were used for multiple comparison among different groups under same light condition.

Calculation of sensitivity

Photon capture efficiency (PCE) of an ommatidium was calculated from Land's equation (Land, 1981):

$$\text{PCE} = (\pi/4)(A/f)^2 S(1 - e^{-kx}).$$

Where, *A* = aperture (diameter of a corneal lens), *f* = focal length, *S* = area of receptor (rhabdom area in cross-section), *x* = receptor length (rhabdom length) and *k* = absorption coefficient (= 0.0067).

Diameter of a corneal lens, cross-sectional rhabdom area and rhabdom length were measured in light micrographs. As for the focal length of an ommatidium, the distance between the inner corneal surface and the backfocal plane was measured following Ukhonov *et al.*, (1996). Namely, the retinal tissue was removed from a freshly-cut eyecup by a thin strip of filter paper. The eyecup (composed of corneal lenses and crystalline cones) was rinsed with Ringer solution and the cleaned eyecup was placed on a coverslip so that the corneal surface was exposed to air. The preparation then was sealed in a moist chamber and placed on the stage of a light microscope (Olympus BH-2) with the outer surface of the corneal lens oriented toward the condenser lens of the microscope. The backfocal plane was found by a sharp image of a square shape drawn on a transparent sheet just above the light source of the microscope. The backfocal plane and morphological factors were measured in 10 ommatidia in the central region of 5 compound

eyes, taken from both nymphs and adults, and their averaged values were used for the PCE calculation. A Mann-Whitney *U*-test was used for the statistical analysis between day and night in the same developmental stage and sex of the groups of crickets.

RESULTS

The structure of a compound eye

The central region of the compound eye

The compound eyes of both nymphs and adults are of the apposition type, and their ommatidial structures are basically the same as those shown diagrammatically in Fig. 1. The small compound eyes of the early nymphal stage gradually become larger as nymphs become adults. For example, rhabdoms of approximately 50–60 μm in length in the early nymphal stage become 200–250 μm long in adults. Typically, a long proximal process of a crystalline cone elongates deeply in the center of an ommatidial rhabdom (Figs. 1A–C and 2). One ommatidial retinula consists of 8 retinular cells, numbered 1–8 by Burghause (1979). The No. 1, 3, 5 and 7 cells have crystalline cone tracts on both sides at the distal layer (Fig. 1B). Microvilli of an ommatidial rhabdom are, in principle, oriented in two directions perpendicular to each other (Figs. 1 and 3). In the most distal layer, a rhabdom is composed of 4 rhabdomeres, numbered R1, R3, R5 and R7, where the microvilli of R1 and R5 are perpendicular to those of R3 and R7. This pattern of rhabdom formation is seen throughout almost its entire length (Fig. 1B–E). In the central and proximal layers, the microvilli of rhabdomeres from pairs R1, R8 (R8 only in the limited proximal layer) and

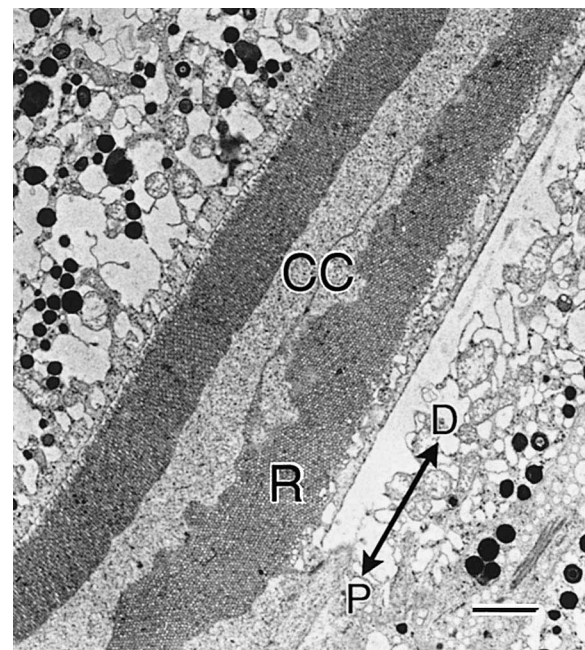


Fig. 2. Longitudinal section through the distal part of an ommatidium in the central region of the eye of an adult male cricket at midday. A long proximal process of a crystalline cone (CC) elongates deeply into the center of an ommatidial rhabdom (R). D, distal; P, proximal. Scale: 2 μm.

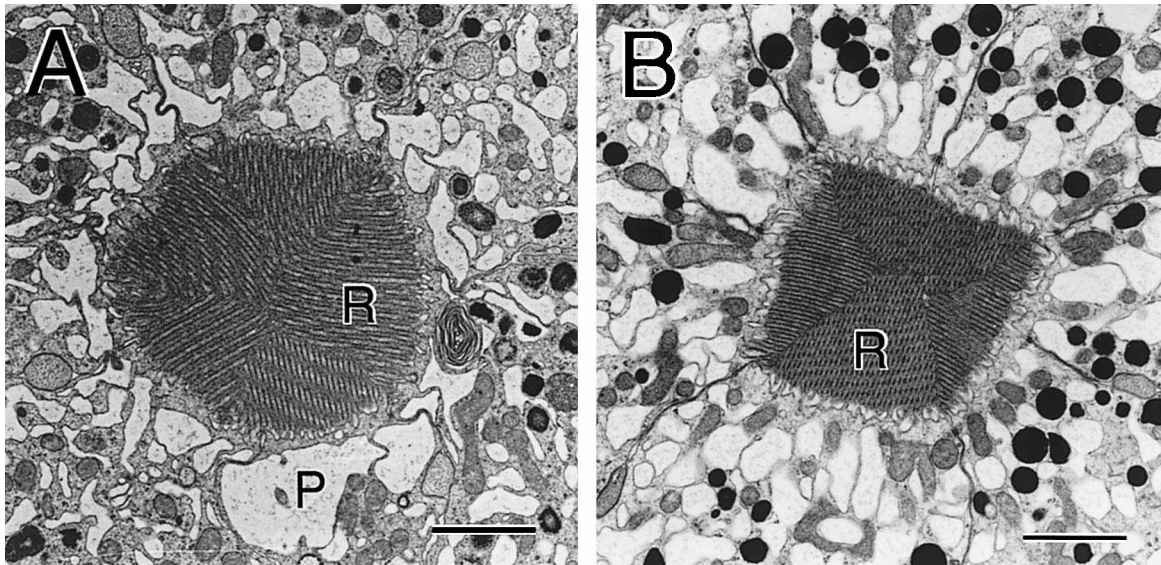


Fig. 3. Transverse sections through an ommatidium in the central region of a compound eye of an adult male cricket at midnight (A) and at midday (B). Microvilli of an ommatidial rhabdom are oriented in two directions perpendicular to each other. The rhabdom is larger at night than that during the day. There are palisades (P) around the rhabdom (R) at night. Scale: 2 μm .

R4, R5 are oriented in the same direction, while those from pairs R2, R3 and R6, R7 are oriented in a perpendicular direction (Fig. 1C-E).

The dorsal rim area of the compound eye

Ommatidial structure in the DRA is characterized by its flattened cornea; the rhabdom is 4–5 times bigger than that from the central region of the eye and there are no pigment granules in the reticular cells (Fig. 1F, G). An ommatidial retinula is composed of 8 reticular cells, same as in the central region of the eye. An ommatidial rhabdom is composed of 6 rhabdomeres (R1, R2, R5, R6, R7 and R8), but R3 and R4 do not contribute to the rhabdom formation (Figs. 1G and 4). As shown in Fig. 1G, microvilli from R1, R2, R5, R6 and R8 are arranged in the same direction, while those from R7 are perpendicular to those of the other cells. Functionally, it is well known that the DRA is characterized by a significantly high polarization sensitivity (Labhart *et al.*, 1984).

Structural changes between day and night

We examined day-night structural changes in the compound eyes of four groups, male and female nymphs and adult males and females, under the different light and dark regimes mentioned in “Materials and methods”. In all cases, male nymphs did not display any detectable differences from female nymphs (data not shown). In the following sections, the results from male and female nymphs are combined and simply described as nymphs.

The central region of the compound eye

Fig. 5A shows the RORs in the central region of the compound eyes between midday and midnight under a 12:12 light:dark regime. The RORs at midnight (nymph 2.5%, adult male 4.1%, adult female 3.0%) were significantly higher than those at midday (nymph 1.5%, male adult 2.0%, female adult 1.1%), in both nymphal and adult stages

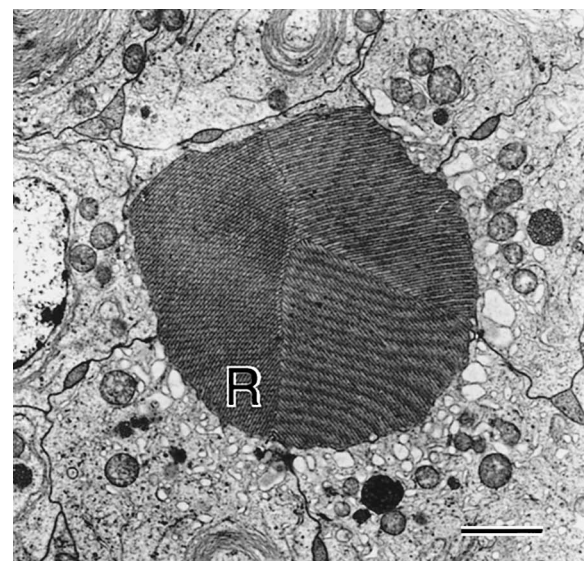


Fig. 4. Transverse sections through an ommatidium in the DRA of a compound eye of an adult male cricket at midday. Rhabdom (R) is larger than that of central region of compound eye. Microvilli of an ommatidial rhabdom are oriented in two directions perpendicular to each other, and rhabdom is formed by six reticular cells. Scale: 2 μm .

(nymph, $P < 0.01$; adult male, $P < 0.005$; adult female, $P < 0.05$). The rhabdom of a dark-adapted eye at midnight was round in cross-section, surrounded by palisades, and there were few pigment granules around the rhabdom (Fig. 3A). On the other hand, the rhabdom cross-sections of light-adapted eyes at midday were square in shape, and the pigment granules aggregated around the rhabdom and the palisades disappeared (Fig. 3B). Moreover, many lamellated bodies (LBs) and characteristic structures of regularly arranged and wavy endoplasmic reticulum were seen in the

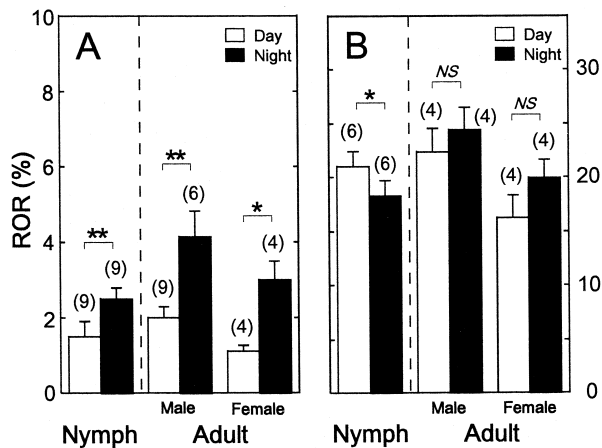


Fig. 5. The rhabdom occupation ratios (RORs) of an ommatidium in the central region (A) and in the DRAs (B) of compound eyes of nymphs and adult males and females under a normal LD cycle. Each data point represents a mean value with SD, and the number of crickets examined is shown above each column. Statistical significance of the difference between day and night levels was calculated with a Mann-Whitney *U*-test and is indicated by asterisks (NS, not significant; *, $P < 0.05$; **, $P < 0.01$).

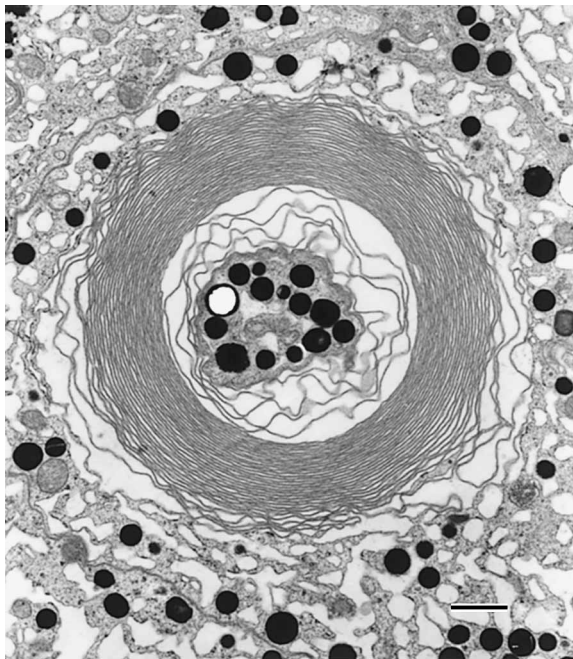


Fig. 6. A lamellated body (LB) in the cytoplasm of a light-adapted reticular cell of an adult male cricket. Scale: 1 μ m.

cytoplasm of light-adapted reticular cells (Figs. 6 and 7). Such wavy endoplasmic reticulum was frequently observed by the side of LBs in reticular cells, and we could confirm that some of them were directly continuous with each other (Fig. 7B). In contrast to the pigment granules in the reticular cells, pigment granules in the pigment cells did not show notable migrations between day and night (data not shown).

At midnight, the ROR of adult male crickets (4.1%) was larger than that of nymphs (2.5%) ($P < 0.05$), and the ROR

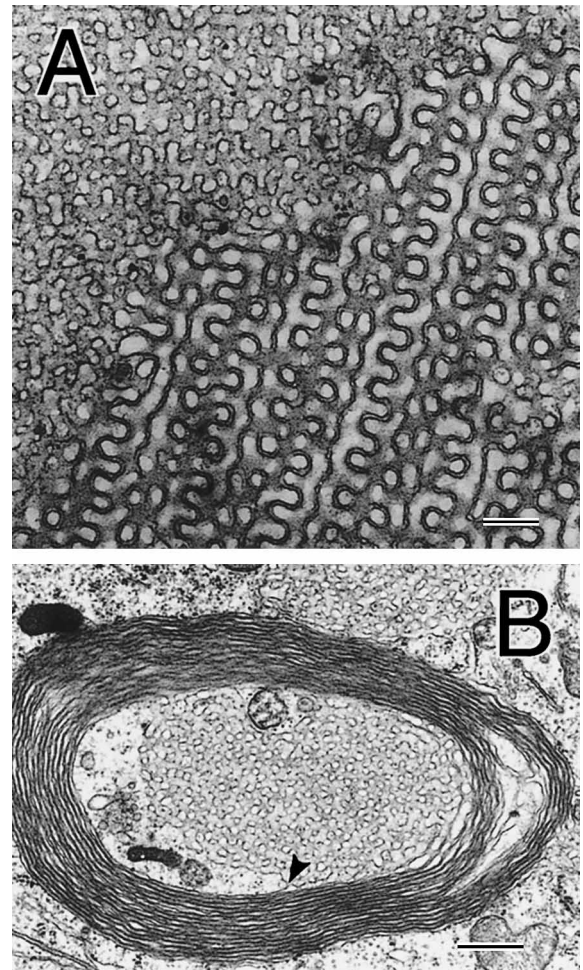


Fig. 7. (A) Wavy endoplasmic reticulum in the cytoplasm of a light-adapted reticular cell in a compound eye of an adult male cricket. Scale: 500 nm. (B) Wavy endoplasmic reticulum and LB are directly continuous with each other (arrowhead). Scale: 1 μ m.

of adult female crickets (3.0%) did not show significant difference to that of nymphs (by Kruskal-Wallis and Scheffe's tests). This result suggests that, at night, the reticular cells of adult males have higher sensitivity than those of adult females and nymphs, even under the same light conditions.

The dorsal rim area of the compound eye

Fig. 5B shows the RORs of the DRAs of the compound eyes at midday and midnight. In contrast to the central region of the eye, the RORs of the DRAs of adults did not exhibit any significant differences between midday and midnight (male adult, 22.3~24.4%; female adult, 16.2~20.0%). In nymphs, strangely, the RORs at midday (21.0%) were significantly higher (17.5%) than at midnight ($P < 0.05$).

Changes in photon capture efficiency (PCE) between day and night

To compare the PCE of ommatidia using Land's equation (1981), we measured the necessary morphological factors of ommatidia in adult males, females and nymphs sep-

Table 1. Ommatidial structures and calculated photon capture efficiencies (PCEs) (see Materials and methods). A, diameter of a corneal lens; S, rhabdom cross-sectional area; x, rhabdom length; f, focal length of a corneal lens. All factors are averages from 10 ommatidia in 5 compound eyes each, and the PCEs are calculated from averaged values. The results of statistical comparison in an identical group between day and night, using a Mann-Whitney *U*-test, are shown with the significance level (*P*). NS, not significant.

	Nymph		Adult			
			Male		Female	
	Day	Night	Day	Night	Day	Night
A	34.9± 2.3	34.7± 1.9	36.1± 2.2	36.7± 1.3	37.0± 0.2	37.4± 0.7
(μm)		(NS)		(NS)		(NS)
S	14.3± 4.5	23.8± 2.8	21.0± 3.0	43.0± 6.9	12.1± 1.4	32.8± 4.9
(μm ²)		(<i>P</i> < 0.05)		(<i>P</i> < 0.005)		(<i>P</i> < 0.05)
x	243.9±26.7	243.6±21.4	286.7±15.3	281.7±16.1	288.3±14.0	291.4±19.4
(μm)		(NS)		(NS)		(NS)
f	63.9± 5.4	64.5± 4.4	67.0± 4.9	66.3± 3.5	66.5± 3.3	69.4± 6.5
(μm)		(NS)		(NS)		(NS)
PCE	2.7	4.3	4.1	8.8	2.5	6.4

arately; the PCE of the compound eye was calculated for each group from the average values (Table 1). Cross-sectional rhabdom areas (S) increased from midday to midnight in all groups (nymphs, 14.3~23.8 μm², *P* < 0.005; adult males, 21.0~43.0 μm², *P* < 0.005; adult females, 12.1~32.8 μm², *P* < 0.05). However, the day-night differences were not statistically significant (*P* > 0.1) for other factors such as focal length (f) (63.9~69.4 μm), rhabdom length (x) (243.6~291.4 μm) and diameter of a corneal lens (A) (34.7~37.4 μm). As shown in Table 1, the PCEs of the compound eyes of all groups at night were approximately twice as those of the corresponding group during the day. Among the groups, the PCE of the compound eyes of adult males at midnight was the highest (8.8), while the daytime PCE of adult female compound eyes was the lowest value obtained (2.5).

Circadian structural changes

Under constant darkness, only the RORs of adult male crickets showed a significant difference between subjective day (4.4%) and subjective night (6.6%) (*P* < 0.05; Fig. 8A). This indicates that the changes in rhabdom size are controlled by an endogenous circadian clock, at least in adult male crickets. On the other hand, under constant light with 1000 lux, we could not detect any significant difference between subjective day (2.2~3.0%) and subjective night (2.2~3.0%) in any experimental group (Fig. 8B). The inhibitory effect of constant light on the increase of rhabdom size appears to be stronger than that of the endogenous circadian clock.

In contrast to the changes in rhabdom size, we could not detect any circadian rhythm in palisade formation around the rhabdom nor migration of pigment granules in the reticular cell. Regardless of time of the day, many palisades were observed around the rhabdom and pigment granules were diffused in dark-adapted eye. While, pali-

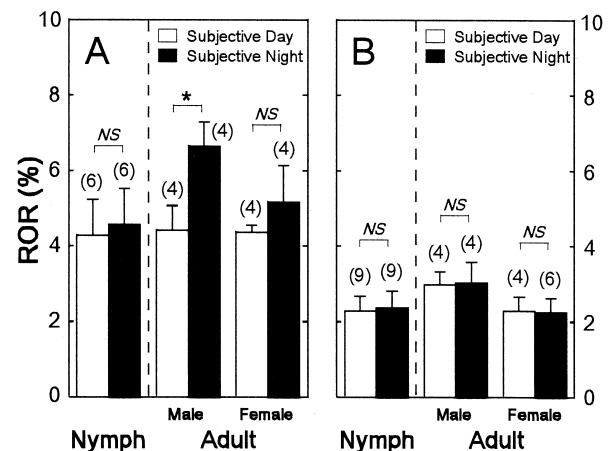


Fig. 8. The rhabdom occupation ratios (RORs) of the ommatidia in the central region of compound eyes of nymphs and adult males and females under constant darkness (A) and under constant lightness (B). Each data point represents the mean value with SD, and the number of crickets examined is shown above each column. Statistical significance of the difference between day and night was calculated with a Mann-Whitney *U*-test and is indicated by asterisks (NS, not significant; *, *P* < 0.05).

sades disappeared and pigment granules aggregated around the rhabdom in light-adapted eye (data not shown). These results indicate that the formation of palisades is solely caused by dark adaptation and aggregation of pigment granules by light adaptation in the cricket eye.

DISCUSSION

We have discovered several interesting diurnal features of cricket compound eye. (1) Under normal light/dark cyclic conditions, RORs increased significantly from day (light) to night (dark) in both nymphs and adult crickets, except for the DRAs of nymphs, which showed the reverse trend. (2) Other

morphological factors that influenced the sensitivity of the eye, such as length of rhabdom, lens diameter, etc., all did not show significant change between day and night. Thus ROR is the major factor which influenced PCE of the eye of crickets. (3) Under constant darkness, the RORs of nymphs and female adults exhibited no change; only the RORs of adult males independently displayed a free-running circadian rhythm. (4) Under conditions of constant light, the RORs of all groups remained consistently at a low level, and did not display any circadian rhythm. These results represent the first report of day-night morphological changes in the compound eye of the cricket *Gryllus bimaculatus* by developmental stage and sex. Hoff (1985) has already reported results similar to (1) in adult crickets.

Changes in ommatidial structure and photon capture efficiency

The central region of the compound eye

The increase in rhabdom size from day to night has been reported previously in some other species, such as locusts *Locusta migratoria* (Williams, 1982), sea slaters *Ligia exotica* (Hariyama *et al.*, 1986) and crabs *Hemigrapsus sanguineus* (Arikawa *et al.*, 1987). In locusts (Williams, 1982) and crabs (Arikawa *et al.*, 1987), the formation of palisades around rhabdoms at night has also been observed as in cricket eyes; these contribute to the heightened sensitivity of the eye at night. The change in rhabdom size is determined by a reciprocal relationship between the synthesis and degradation of the rhabdom membrane (Eguchi, 1999). Thus, if the synthesis of rhabdom membrane is greater than its degradation, the rhabdom increases in size, while if degradation dominates synthesis, its size decreases. The LBs and wavy endoplasmic reticulum in the cytoplasm of the cell body of a reticular cell are probably associated with the processes of membrane turnover of rhabdoms. Similar lamellar structures were reported in the compound eyes of horseshoe crabs *Limulus* (Chamberlain and Barlow, 1984), where they appeared during the process of membrane shedding caused by the light-onset or endogenous circadian clocks. Since many LBs (Fig. 6) and wavy endoplasmic reticulum (Fig. 7A) appeared at midday, this suggests that the cricket rhabdom membrane is also shed under daylight conditions.

The change in PCE of the cricket compound eye from day to night depends solely on the change in rhabdom size, because the other morphological factors exhibit no significant changes according to day and night. The PCE of the compound eyes of all groups of crickets examined in the present experiments ranged from a low of 2.5 (adult females at day) to a high of 8.8 (adult males at night). The value 2.7 for nymph compound eyes at daytime is very high for diurnal insects; Land (1981), for example, reported 0.32 for *Apis* workers. On the other hand, the 8.8 of adult males at night is much lower than the 82.8 of the typical nocturnal moth *Ephestia* (Land, 1981). This may reflect a difference in life-style, that is, visual information is more important for flying insects, such as moths, than for field insects, such as crick-

ets. Nymphs shows the smallest difference in sensitivity between day and night because they are diurnally active, that is to say, they may not use their visual sense in dim light environments such as night.

The dorsal rim area of the compound eye

Hoff (1985) reported that rhabdom size in the DRA also changed according to day and night. Contrary to this observation, we could find no clear change in the RORs of ommatidia in the DRA of the adult eye. A possible cause for the lack of agreement could be attributed to differences in light intensity. Namely, in Hoff's experiments, crickets were put in a small cage so that they were directly exposed to light, while our crickets were put in a large cage with paper shelters (see Materials and methods). Under natural conditions in the field, it is known that both nymphs and adults avoid direct bright light and spend most of the day hidden under shaded shelters (Honegger and Campan, 1989). Therefore, it can be said that our experimental conditions were closer to the cricket's natural habitat. The fact that the change in ROR occurred in the central area but not in the DRA under the same conditions, suggests that stronger light may be necessary to induce shedding of rhabdom membrane in the DRA than is required in the central area of the eye. It is unclear why daytime rhabdom size in the DRA of nymphs was significantly larger than their nighttime rhabdom size.

The bigger rhabdom in the DRAs of the cricket's compound eye even in daytime could contribute to keeping relatively high PCE under light condition. Since the detection of polarized skylight in an ommatidium in DRA is attained through the polarization antagonism caused by e-vectors inputs from the mutually orthogonal oriented microvilli (Labhart and Mayer, 1999).

Effect of the circadian clock on rhabdom size

It is notable that only adult males showed a distinct circadian change in rhabdom size under conditions of constant darkness (Fig. 8A). The compound eyes of adult males were much more influenced by the endogenous circadian clock than the eyes of the other cricket groups in the present experiments. The fact that no clear change was observed in the ROR under constant light suggests that light reduced the rhabdom area regardless of the time of day. Such circadian changes in structure and function of compound eyes under constant dark or light conditions have been intensively studied in several other species. They have been studied morphologically in: horseshoe crabs, Chamberlain and Barlow (1984); crabs, Arikawa *et al.* (1987); and sea slaters, Hariyama *et al.* (1986). They also have been studied electrophysiologically in several species: including cockroaches, Wills *et al.* (1986); crickets, Tomioka and Chiba (1982b); and crabs, Arikawa *et al.* (1987).

In the cricket *Gryllus bimaculatus*, the optic lamina-medulla complex is thought to be the site of the circadian clock (Tomioka and Chiba, 1986, 1992). It also has been reported that the amplitude of an electroretinogram (ERG) from an adult male cricket compound eye showed circadian

rhythm under constant darkness; this rhythm was maintained even after the optic lamina-medulla complex and compound eye were neurally isolated from the central nervous system (Tomioka and Chiba, 1982b).

Reversal of circadian rhythm from nymph to adult stage

It is well known that crickets are diurnally active in the nymphal stage, but that they become nocturnal 4 to 5 days after the imaginal molt (Tomioka and Chiba, 1982a). Therefore, high sensitivity to light at night is not necessary for nymphs. We know that the ovipositional activity of mated females is significantly higher at night (Sugawara, 1997, 1998) and male calling song elicits strong locomotor activity in virgin females regardless of the time of a day (Loher, 1979). Virgin females walk around searching for a mate mainly by phonotaxis, but after mating, they devote themselves to laying eggs, with little locomotion. Thus, it is likely that visual information is less important for females. The present experiments revealed that the compound eyes of adult males exhibit higher PCE (8.8) at night than females' (6.4) and nymphs' (4.3) eyes at night. This fact may reflect that visual information is more important to males at night.

REFERENCES

- Arikawa K, Kawamata K, Suzuki T, Eguchi E (1987) Daily changes of structure, function and rhodopsin content in the compound eye of the crab *Hemigrapsus sanguineus*. *J Comp Physiol A* 161: 161–174
- Barlow RB Jr., Kaplan E, Renninger GH, Saito T (1987) Circadian rhythms in *Limulus* photoreceptors I. Intracellular studies. *J Gen Physiol* 89: 353–378
- Burghause FMHR (1979) Die strukturelle Spezialisierung des dorsalen Augenteils der Grillen (Orthoptera, Grylloidea). *Zool Jb Physiol Bd* 83: S502–525
- Chamberlain SC, Barlow RB Jr. (1984) Transient membrane shedding in *Limulus* photoreceptors: control mechanisms under natural lighting. *J Neurosci* 4: 2792–2810
- Eguchi E (1982) Retinular fine structure in compound eyes of diurnal and nocturnal sphingid moths. *Cell Tissue Res* 223: 29–42
- Eguchi E (1999) Turnover of photoreceptive membrane. In "Atlas of arthropod sensory receptors – Dynamic morphology in relation to function" Ed by E Eguchi and Y Tominaga, Springer, Tokyo, pp 87–96
- Eguchi E, Horikoshi T (1984) Comparison of stimulus-response (V-log I) functions in five types of lepidopteran compound eyes (46 species). *J Comp Physiol A* 154: 3–12
- Hariyama T, Meyer-Rochow VB, Eguchi E (1986) Diurnal changes in structure and function of the compound eye of *Ligia exotica* (Crustacea, Isopoda). *J Exp Biol* 123: 1–26
- Hariyama T, Meyer-Rochow VB, Kawachi T, Takaku Y, Tsukahara Y (2001) Diurnal changes in retinula cell sensitivities and receptive fields (two-dimensional angular sensitivity functions) in the apposition eyes of *Ligia exotica* (Crustacea, Isopoda). *J Exp Biol* 204: 239–248
- Hoff R (1985) Diurnal ultrastructural changes in the compound eye of *Gryllus bimaculatus* (Orthoptera Grylloidea): in particular the dorsal rim area. *Zool Beitr N F* 29: 87–102
- Honegger H-W, Campan R (1989) Vision and visually guided behavior. In "Cricket behavior and neurobiology" Ed by F Huber, TE Moore, W Loher, Cornell University Press, New York, pp 147–177
- Horridge GA, Barnard PBI (1965) Movement of palisade in locust retinula cells when illuminated. *Q J Microsc Sci* 106: 131–135
- Labhart T, Hodel B, Valenzuela I (1984) The physiology of the cricket's compound eye with particular reference to the anatomically specialized dorsal rim area. *J Comp physiol A* 155: 289–296
- Labhart T, Mayer EP (1999) Detectors for polarized skylight in insects: a survey of ommatidial specializations in the dorsal rim area of the compound eye. *Microsc Res Tech* 47: 368–379
- Land MF (1981) Optics and vision in invertebrates. In "Handbook of sensory physiology Vol VII/6B" Ed by H Autrum, Springer, Berlin, pp 471–592
- Land MF, Gibson G, Horwood J, Zeil J (1999) Fundamental differences in the optical structure of the eyes of nocturnal and diurnal mosquitoes. *J Comp Physiol A* 185: 91–103
- Laughlin SB, Weckström M (1993) Fast and slow photoreceptors – a comparative study of the functional diversity of coding and conductances in the Diptera. *J Comp Physiol A* 172: 593–609
- Loher W (1979) The effect of male-calling on female locomotor activity of *Teleogryllus commodus*. *Behav Ecol Sociobiol* 5: 383–390
- Meyer-Rochow VB (1999) Compound eye: circadian rhythmicity, illumination and obscurity. In "Atlas of arthropod sensory receptors – Dynamic morphology in relation to function" Ed by E Eguchi, Y Tominaga, Springer, Tokyo, pp 97–125
- Nilsson D-E, Labhart T, Meyer E (1987) Photoreceptor design and optical properties affecting polarization sensitivity in ants and crickets. *J Comp Physiol A* 161: 645–658
- Stavenga DG (1979) Pseudopupils of compound eyes. In "Handbook of sensory physiology Vol VII/6A" Ed by H Autrum, Springer, Berlin, pp 357–439
- Sugawara T (1997) Diel rhythm of oviposition behavior in the cricket, *Teleogryllus commodus*. *Zool Sci* 14 (Suppl): 115
- Sugawara T (1998) Switch-over of the locomotory and ovipositional activity patterns by mating in the female cricket. *Zool Sci* 15 (Suppl): 102
- Tomioka K, Chiba Y (1982a) Post-embryonic development of circadian rhythm in the cricket, *Gryllus bimaculatus*: a rhythm reversal. *J Comp Physiol A* 147: 299–304
- Tomioka K, Chiba Y (1982b) Persistence of circadian ERG rhythms in the cricket with optic tract severed. *Naturwissenschaften* 69: 395–396
- Tomioka K, Chiba Y (1986) Circadian rhythm in the neurally isolated lamina-medulla-complex of the cricket, *Gryllus bimaculatus*. *J Insect Physiol* 32: 747–755
- Tomioka K, Chiba Y (1992) Characterization of an optic lobe circadian pacemaker by *in situ* and *in vitro* recording of neural activity in the cricket, *Gryllus bimaculatus*. *J Comp Physiol A* 171: 1–7
- Ukhanov KY, Leertouwer HL, Gribakin FG, Stavenga DG (1996) Dioptics of the facet lenses in the dorsal rim area of the cricket *Gryllus bimaculatus*. *J Comp Physiol A* 179: 545–552
- Williams DS (1982) Ommatidial structure in relation to turnover of photoreceptor membrane in the locust. *Cell Tissue Res* 225: 595–617
- Wills SA, Page TL, Colwell CS (1986) Circadian rhythms in the electroretinogram of the cockroach. *J Biol Rhythms* 1: 25–37

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