The Compound Eye of the Dung Beetle, *Bolbocerosoma nigroplagiatum* (Coleoptera: Scarabaeidae)

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(Received 17 June 1996; Accepted 9 August 1996)

The compound eye of a nocturnal dung beetle, *Bolbocerosoma nigroplagiatum*, was observed by light and electron microscopy and compared with that of other scarabaeid beetles. The eye is completely divided into two parts by a well developed *canthus* into relatively narrow dorsal and comparatively wide ventral eyes. The inner surface of the cornea protrudes inwards and forms an exocone, while at the same time the crystalline cone is much reduced. Such a condition of a “duocone” dioptric apparatus seems to have been found so far only in *Omorgus* of the Trogidae. The retina is of the scotopic type, and the configuration of the rhabdom is different in dorsal and ventral eyes. The rhabdoms of several rows of the dorsalmost region in the dorsal eye are composed of three rhabdomeres in which parallel and long microvilli are orientated in three directions, and four V-shaped rhabdomeres in transverse section. In the remaining part of the dorsal eye as well as in the entire ventral eye, the rhabdoms exhibit a seven-lobed pattern. One of the rhabdomeres is formed by parallel and linear microvilli which are less electron-dense. A microvillar arrangement such as this suggests a special function e.g. polarization vision.

Key words: compound eye, *Bolbocerosoma nigroplagiatum*, Scarabaeidae, “duocone” dioptrics, ultrastructure

INTRODUCTION

The relationship among eye structure, activity and phylogeny in scarabaeid beetles and 52 species of leaf-chafers has previously been examined by light microscopy (GOKAN, 1973). Up to date several species of dung beetles, as well as leaf-chafers, also have been studied by electron microscopy e.g. *Lethrus apterus*: FRANTSEVICH et al. (1977); *Euoniticellus africanus*: MEYER-ROCHOW (1978); *Geotrupes auratus*: GOKAN (1989 a); *Aphodius haroldianus*: GOKAN (1989 b); *Ochodaeus maculatus*: GOKAN (1989 c); *Onthophagus lenzii*: GOKAN (1990); *O. posticus*: GOKAN and MEYER-ROCHOW (1990). However, with the exception of *O. lenzii* most of the sample species are diurnally active. Therefore, further observations were needed on nocturnal dung beetles. *Bolbocerosoma nigroplagiatum* is a nocturnal, relatively rare Japanese dung beetle approx. 10 mm long, and attracted to the light at night. Fortunately, one female of this beetle was collected at our farm (Atsugi, Kanagawa Pref.) last autumn. The compound eye of this material was observed by light (LM) and electron microscopy (EM) and considered in comparison with other dung beetles previously studied.

MATERIALS AND METHODS

The insect was decapitated in day time (light adapted state). The head was then split in

half along the sagittal plane. For fixation, the same KARNOVSKY's solution and osmium tetroxide were used as in previous papers (GOKAN,1990). After dehydration with ethanol and QY-1, the specimen was embedded in Epon 812. Gold-silvery sections were double stained with uranyl acetate (15 min) and lead citrate (10 min) and observed under a JEM-100CX EM operated at 80 kV. Semithin section (1–3 μm thick) were stained with Azur-B and observed under ordinary LM.

Fig. 1. A. Semidiagrammatic representation of an ommatidium of *Bolbocerosoma nigroplagiatum* in longitudinal section. Numerals alongside refer to the figure number of micrographs in this paper. B–D. Diagrammatic figures of the rhabdom pattern in transverse section at the mid level of rhabdom region. B. Distalmost region of dorsal eye. C. Central part of dorsal eye. D. Central part of ventral eye. bm, basement membrane; c, cornea; cc, crystalline cone; cz, clear zone; ec, exocone; n.rc, nucleus of retinula cell; pp, primary pigment cell; rc, retinula cell; rh, rhabdom; rt, retinular tract; sp, secondary pigment cell; tr, tracheole.

RESULTS AND DISCUSSION

General features

In common with other scarabaeid beetles e.g. *Euoniticellus* (MEYER-ROCHOW, 1977) and *Geotrupes* (GOKAN, 1989 a) the compound eye of *B. nigroplagiatum* is completely divided into a relatively narrow dorsal and a comparatively wide ventral part by a strongly developed *canthus* (Fig. 2). The dorsal eye is composed of nearly 20 rows of ommatidia and the ventral eye comprises nearly 40 rows counted from a longitudinal section through the centre of the eye.

The structure of the dioptric apparatus exhibits a number of conspicuous features when compared with that of the usual type of scarabaeid eye. Thus, the inner surface of the cornea protrudes strongly inward, forming a corneal cone in an exocone eye. The crystalline cone, on the other hand, is much reduced in size. Such a condition (Figs. 1, 3) has so far only been found in *Omorgus* (CAVENEY, 1986; CAVENEY and SCHOLTZ, 1993) of the Trogidae, so that it seems justified to use the term “duocone” to describe this condition (CAVENEY writes: “the duocone ommatidia has a long corneal exocone and short crystalline cone”). MEYER-ROCHOW (1988) has analyzed co-axial double-lens system in compound eyes and it is possible that the “duocone condition” now observed in the eye of *B. nigroplagiatum* helps the animal to see in both far and near fields. In the nocturnal *Onthophagus lenzii* (GOKAN, 1990), however, the cone is considerably thicker than the cornea and such a relationship has also been found in leaf-chafers (GOKAN, 1973); consequently, the presence of a “duocone condition” in the eye of a nocturnal dung beetle is surprising.

The retina of *B. nigroplagiatum* is of the typical scotopic type (Fig. 1).

Biometrical and morphological data of the eye are given in Table 1.

*Fine structure**a. Dioptric apparatus*

Cornea: The inner protrusion of the cornea (= CAVENEY's exocone) is approx. 54 μm thick and exhibits multiple equidistant lamellae in longitudinal section (Fig. 4). The surface of cornea is smooth and without corneal nipples or any other processes. Interommatidial hairs are not present.

Crystalline cone: In longitudinal sections through the centre, the cones are “heart-shaped” (Figs. 3, 4, 5). Each cone is formed by four Semper cells (= cone cells) which is the norm for the eucone eye generally in insects, and is surrounded by the ER (endoplasmic reticulum) of over 20 layers in the distal and one or two layers in the proximal part (Figs. 4,

Table 1. Morphological data of one ommatidium

		Dorsal eye (μm)	Ventral eye (μm)
Cornea	thickness*	95	same as dorsal
	diameter	28	same as dorsal
Cone	thickness	12.5	15.0
	diameter	13.0	15.0
Retinula cell		208	same as dorsal
Rhabdom region		95	same as dorsal
Rhabdom		84	same as dorsal

* Including the exocone.

Fig. 2. Scanning electron micrograph of the head of *B. nigroplagiatum*. The compound eye is completely divided into dorsal (d) and ventral (v) components by the *canthus* (ca). Frontal (f) direction is indicated. Scale: 0.5 mm.

Fig. 3. Light micrograph of the compound eye. The "clear-zone" is artifactually twisted during fixation. c, cornea; cc, crystalline cone; ec, exocone. Scale: 50 μ m.

Figs. 4–9. Electron micrographs.

Fig. 4. Longitudinal section of an ommatidium through the proximal end of the "exocone" and beginning the crystalline cone. Some particles (arrows) are visible. cc, crystalline cone; ec, exocone; n.sc, nucleus of Semper cell. Scale: 5 μ m.

Fig. 5. Longitudinal section of an ommatidium through the cone and distal region of the retinula cells. ct, cone tract; n.rc, nucleus of retinula cell. Scale: 5 μ m.

Fig. 6. Longitudinal section of several ommatidia at the level of the basement membrane, showing the nuclei of the basal cells (*) situated in or below the basement membrane. bm, basement membrane; tr, tracheole. Scale: 2.5 μ m.

Fig. 7. Transverse section of an ommatidium through the basement membrane, showing centrally situated nucleus of the basal retinula cell (*). bm, basement membrane; tr, tracheole. Scale: 2.5 μ m.

Fig. 8. Longitudinal section of an ommatidium through the distal part of the rhabdom region and retinula cells, crowded with numerous mitochondria. mi, mitochondria; rh, rhabdom; sp, secondary pigment cell. Scale: 5 μ m.

Fig. 9. Transverse section of an ommatidium through the proximal region of the rhabdom, showing the basal cell (*) and four cone cell extension (arrows). Scale: 2.5 μ m.

Fig. 10. Light micrograph of the compound eye in transverse section through the rhabdom region of the dorsal eye, showing regional differences in rhabdom pattern between dorsalmost (right area bounded with dotted line) and other parts. The irregularity in the central part of this picture is artifactual. Scale: 50 μ m.

Figs. 11–13. Electron micrographs of the rhabdom in transverse section at the level indicated in Fig. 1.

Fig. 11. Dorsalmost part of dorsal eye, showing arrowhead-shaped rhabdomeres formed by long and linear

5). The nuclei of the cone cells are positioned on the distal surface of the cone close to the bottom of the exocone and some electron dense particles of approx. $0.2\ \mu\text{m}$ are scattered around them (Figs. 4, 5). Proximally, the four cone cell bodies taper off and enter the distal end of the retinula cells along the central intercellular space. Below the retinular tract, the proximal ends of the cone cells, the so-called "cone extensions," split into four and are traceable as far as the proximal end of the rhabdom (Fig. 9).

b. Retinula cell. Although the basic anatomy of the retina, the number and the arrangement of the retinula cells are similar to those of other scarabaeid beetles possessing scotopic type eye, some differences were noticed. Thus, usually, the nucleus of retinula cell 8 (= basal retinula cell which has no rhabdomere) lies near the proximal end of the rhabdom, whereas in this beetle, most nuclei of that cell are situated within or below the basement membrane (Figs. 6, 7). In spite of the light-adapted state in this sample the nuclei of the seven retinula cells are located distally and close to the cone. This fact suggests that the nuclei of this eye are incapable of movement in response to light- and dark-adaptation or exhibit a response pattern that is as yet unknown (Figs. 3, 5).

c. Rhabdom. The overall rhabdom organization is similar to that of the scotopic eyes of other scarabaeid beetles, but a distal rhabdomere known from the eyes of *Sericesthis* (MEYER-ROCHOW, 1977), *Anomala* (GOKAN, 1982 a), *Maladera* and *Paraserica* (GOKAN, 1982 b) and *Maladera* (MEYER-ROCHOW and GOKAN, 1987) was not seen. The distal part of the rhabdom region is filled with numerous mitochondria (Fig. 8). The rhabdom exhibits a seven-lobed pattern in transverse section at mid level but the detailed microvillar arrangement differs in dorsal and ventral eye parts as well as from one region of the eye to another.

Dorsal eye: Regional differences of microvillar arrangement of the rhabdomere are found (Fig. 10). Transverse sections through the mid level of the rhabdom show that in several dorsalmost rows of the dorsal eye, the rhabdomeres of cells 1, 3 and 6 are arrow-head-shaped. These rhabdomeres are composed of long and parallel microvilli which are restricted to three directions and lighter stained than other V-shaped rhabdomeres both in LM and EM sections (Figs. 1B, 11).

In the rest of the eye, transverse section at the same level of the rhabdoms as above exhibit a seven lobed pattern formed by seven V-shaped rhabdomeres. The rhabdomere of cell 1 is of the shape of a shallow V-shape consisting of longer microvilli and marked by lower electron density (Figs. 1C, 12).

In the transitional region between the dorsalmost and the remaining part, irregularly constructed rhabdoms are seen (Fig. 13).

The ratio of rhabdom area/whole cell area in transverse sections (rhabdom occupation ratio, ROR) at mid level of the rhabdom is approx. 53% in the dorsalmost part and 40% in the other regions of the eye.

Ventral eye: In the ventral eye, no obvious regional differences of rhabdomeres and microvillar arrangements were noticed at corresponding levels (Fig. 14). In transverse sec-

microvilli. Scale: $2.5\ \mu\text{m}$.

Fig. 12. Central part of dorsal eye. Scale: $2.5\ \mu\text{m}$.

Fig. 13. Transitional region between dorsalmost and remainder of dorsal eye. Scale: $2.5\ \mu\text{m}$.

Fig. 14. Light micrograph of the compound eye in transverse section through the rhabdom region of the ventral eye, showing almost identical patterns irrespective of direction in the eye. Scale: $50\ \mu\text{m}$.

Fig. 15. Electron micrograph of the rhabdom of the ventral eye in transverse section at the level indicated in Fig. 1. The retinula cell is largely made up of voluminous rhabdomeres, numerous mitochondria are seen in the cytoplasm. Scale: $5\ \mu\text{m}$.

tion, the rhabdomere of cell 1 exhibits a somewhat shallower V-shape and somewhat lower in electron density than the others (Figs. 1D, 15). The ROR is approx. 70%.

In general, rhabdoms are more massively developed in nocturnal than diurnal species (GOKAN and HOSOBUCHI, 1979; GOKAN et al., 1987). In *Onthophagus lenzii* (GOKAN, 1990), a nocturnal dung beetle, for example, RORs of approx. 77% (dorsalmost) and 80% (ventral eye) were recorded. These values are comparable to those of *Bolbocerosoma*.

In the eye of *Geotrupes auratus*, a diurnal dung beetle, the distal tip of the rhabdom is composed of short and parallel microvilli indicative of the photopic eye (GOKAN, 1989 a), but in *Bolbocerosoma* a rhabdom pattern like that is not seen in either the dorsal or the ventral eyes.

As a rule, seven retinula cells contribute to the formation of the rhabdom in scarabaeid scotopic eyes. In these species, rhabdoms exhibit multi-lobed patterns in transverse section, and amongst them six- and seven-lobed pattern are most common. In the former, the eyes have a distal rhabdom and the rhabdom is made up by six rhabdomeres other than the distal one. In contrast, eyes with the seven-lobed rhabdomere arrangement are lacking the distal rhabdomere and all seven rhabdomeres of the seven retinula cells contribute to the formation of the rhabdom at the same level. Such a situation, in addition to *Bolbocerosoma*, has been found in *Repsinus* (HORRIDGE and GIDDINGS, 1971), *Anoplognathus* (MEYER-ROCHOW and HORRIDGE, 1975), *Geotrupes* (GOKAN, 1989 a) and *Melolontha* (LABHART et al., 1992).

The microvilli of one rhabdomere, however, show a markedly less electron density than the rest of the rhabdom. In relation to this unusual rhabdomere, LABHART (1992) has written "This special cell is possibly homologous to the distal receptor of other species termed R1 and is, therefore, also given the number 1 in *Melolontha*." Therefore, in this paper that cell is also given the number 1.

d. Pigment cells. In common with most other insect eyes, two primary and some ten secondary pigment cells are surrounding each ommatidium. Pigment grains are approx. 0.9–1.0 μm in diameter, and are distributed from the distal tip of both pigment cells to the level of the nuclei of the retinula cells in the distal region of the eye. More proximally, grains of the secondary pigment cell are included from the basement membrane 60 μm inward, but they were never seen to invade the "clear-zone" (Figs. 1A, 3, 8).

e. Tracheoles. A few tracheoles enter the retinular zone together with an axon bundle of one ommatidium and pass through the basement membrane (Figs. 6, 7). These tracheoles divide into about 50 and surround an ommatidium as far as halfway up the rhabdom forming a thick tracheal tapetum (Fig. 7).

CONCLUSION

The most unusual feature of this eye is the structure of its dioptric apparatus. The "duocone condition" has so far only been reported from the Trogid *Omorgus* (CAVENEY, 1986; CAVENEY and SCHOLTZ, 1993). It is surprising that similar types of lens system are developed in so distantly allied groups. Regional differences of rhabdom patterns in the eye are found. In particular the dorsalmost rhabdoms are equipped with long linear and parallel microvilli. A rhabdom arrangement of this kind is likely to present an adaptation for polarization vision.

ACKNOWLEDGEMENTS

I wish to thank Dr. V.B. MEYER-ROCHOW of the University of Oulu, Finland, for his critical reading of

manuscript and kind correction of English. Thanks are also due to Miss S. NAKANO of the Tokyo University of Agriculture for her friendly offer of material and Mr. S. NIITSU of the same University for his technical assistance.

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